DRY MATTER PARTITIONING IN *Zantedeschia* K. Spreng, AS INFLUENCED BY TEMPERATURE AND PHOTOSYNTHETIC PHOTON FLUX

Keith Allen Funnell
1993

A thesis presented in partial
fulfilment of the requirements
for the degree
of Doctorate of Philosophy in
Horticultural Science at
Massey University

ABSTRACT

The in vivo and in vitro dry matter accumulation and partitioning in plants of the *Zantedeschia pentlandii*-like (Watson) Wittm. selection 'Best Gold' were described under a range of either temperature and photosynthetic photon flux (PPF) regimes, or sucrose concentrations, using plant growth analysis.

The initiation of tuber growth, as denoted by increases in both structural and starch dry weights, did not require an obligative environmental trigger.

Relative rates of dry matter accumulation (RGR_W) increased linearly with increasing temperature up to a maximum of 28 C, with maximum final total and tuber dry weight occurring between 21 and 26 C both in vivo and in vitro. The linear relationship between the relative rate of dry matter accumulation of the tuber (RGR_T) and temperature, indicated a PPF dependent base temperature for tuber growth between 4.8 and 6.1 C.

By principally altering dry matter partitioning, total dry matter accumulation was highly adaptive to PPF regime. The ability to alter the photosynthetic rate and the partitioning of the daily increment of dry matter into leaf area (LWP), resulted in greater values of the estimated final total plant dry matter under the low PPF regime (348 $\mu mol \cdot m^{-2} \cdot s^{-1}$), at temperatures less than 22 C. At temperatures greater than 19 C the estimated maximum total plant dry weight was either not influenced by PPF or was slightly greater under the high PPF regime (694 $\mu mol \cdot m^{-2} \cdot s^{-1}$). This ability to effectively utilize a low PPF regime indicates that this selection is shade tolerant. The optimum PPF for growth was found to be temperature dependent: estimated maximum total plant dry weight occurred under high PPF at 25 C, whereas the estimated maximum tuber dry weight occurred at 24.5 C under low PPF.

RGR_W was highly correlated with LWP. In contrast, only a poor correlation was determined between RGR_W, and either the efficiency of these leaves to produce additional dry matter, i.e., net assimilation rate (NAR), or starch concentration or soluble carbohydrate concentration. Photosynthetic rate was correlated with RGR_W, but not with RGR_T. While the photosynthetic process must be involved in contributing photoassimilates for tuber growth, it was suggested that the plant's response to dry matter partitioning into the leaf, i.e., LWP, and the tuber, i.e., TWP, had a greater influence in determining tuber growth than could be accounted for by the photosynthetic rate.

Mechanisms of acclimation under both PPF regime suggested that tuber growth was principally source limited. Source limitation was expressed either in terms of:

1) enhanced intersink competition for assimilates, as occurred under the low PPF regime, where enhanced leaf area development (LWP) was in direct competition with enhanced tuber growth (RGR_T). This was also confirmed in vitro where dry matter partitioning to the tuber was reduced under limited source strength.

2) efficiency of dry matter accumulation of leaf area present, as occurred under the high PPF regime, where large increases in RGR_T were correlated with increased NAR. This was also confirmed in vitro where increased source strength increased tuber dry weight.

However, in vitro experiments where source strength was controlled, illustrated that tuber growth was also potentially sink limited at temperatures both lower and higher than the optimum. At 31 C the sink limitation of tuber growth arose from more than the temperature-induced limitation on growth and respiration found at other sink limiting temperatures. At this temperature an additional form of sink limitation was evident where partitioning of dry matter towards the tuber was also restricted. It was suggested that this additional form of sink limitation may have arisen from high temperature inactivation of starch metabolising or sucrose unloading enzymes.

Application of the dry matter partitioning term TWP, provided a more sensitive measure of short term changes in partitioning than the conventionally used term, harvest index.

The optimum temperature range for growth was close to the average daily air temperature during the season for the sites of natural habitat of the suggested parent specie, *Zantedeschia pentlandii*. Similarly the shade tolerance status of this selection was paralleled by the diversity of PPF habitats it naturally occupies, as created by open grassland and forest margins. It was therefore suggested that *Zantedeschia* 'Best Gold' is well adapted to optimise growth under the temperature and PPF regimes of its natural habitat.

This study suggests that improvements in commercial yield of *Zantedeschia* tubers can be achieved in all regions of New Zealand through the use of protected cultivation with supplemental heating. However, unless using protected cultivation, the potential improvements in commercial tuber yields, through the application of shading, are only likely to be evident in warmer regions of New Zealand where growers utilize extended periods of cultivation and optimise leaf area duration.

ACKNOWLEDGEMENTS

The completion of this thesis is a piece of independent research, but so many individuals have contributed either directly or indirectly. In particular, I would like to the thank the following:

my supervisors Dr I.J. Warrington, Dr J.A. Plummer and Dr E.W. Hewett for their challenges, guidance and constructive criticism;

Dr D.J. Chalmers for his leadership and foresight as Head of Department of the then Department of Horticultural Science, to initiate the unique opportunity for me to study towards my PhD;

the management and staff of HortResearch (formally DSIR Plant Physiology Division, and DSIR Fruit and Trees) especially Dr D. Cohen, and the Department of Plant Science (formally the Department of Horticultural Science) for the provision of facilities and technical advice;

Massey University Research Fund (MURF), Massey University Agricultural Research Fund (MUARF), and The C. Alma Baker Trust who all provided various forms of financial contribution;

Dr J.M. Wilson for an endless supply of tubers and seed, as well as thought provoking discussion;

my family and friends who either provided moral support or without knowing it, suitable distractions that created some semblance of normality during a demanding few years of my life.

Parts of Sections 1, 3, 4 and 5 of this thesis have been published in the following;

Funnell, K.A. 1993. Zantedeschia, p. 683-739. In: A. De Hertogh and M. Le Nard (eds.). The physiology of flower bulbs. A comprehensive treatise on the physiology and utilization of ornamental flowering bulbous and tuberous plants. Elsevier Science Publishers, Amsterdam.

Funnell, K.A., J.A. Plummer and I.J. Warrington. 1990. Temperature and light effects on tuber growth in calla lilies. Abstracts XXIII Intl. Hort. Congr., Firenze, Italy, 1990, no. 3261.

CONTENTS PAGE

LIST OF ABBREVIATIONS

α . . . . . apparent photosynthetic quantum yield
$\alpha_{W,A,L,Ls,T \text{ or } Ts}$ . . . . . upper asymptote of factor under investigation
A . . . . . leaf area
$\beta_{W,A,L,Ls,T \text{ or } Ts}$ . . . . . a measure of the starting size of the factor under investigation
BA . . . . . benzyl (1*H*-purin-6-yl) amine
C . . . . . Celsius
cm . . . . . centimetre
cm^2 . . . . . square centimetre
CE . . . . . controlled environment
CO_2 . . . . . carbon dioxide
DTEMP . . . . . day temperature
g . . . . . gram
GA_3 . . . . . gibberellic acid
GA_{4+7} . . . . . gibberellin 4 and 7
h . . . . . hour
ha . . . . . hectare
HPLC . . . . . high performance liquid chromatograph
i.e. . . . . *(id est)* that is
$\kappa_{W,A,L,Ls,T \text{ or } Ts}$ . . . . . rate constant of factor under investigation as a function of size
kg . . . . . kilogram
L . . . . . leaf weight
LAP . . . . . leaf area partitioning
LAR . . . . . leaf area ratio
LWP . . . . . leaf weight partitioning
LWR . . . . . leaf weight ratio
$\log_e$ . . . . . natural logarithm
Ls . . . . . leaf starch dry weight
LS . . . . . Linsmaier and Skoog organic additives
m . . . . . metre
m^2 . . . . . square metre
m^3 . . . . . cubic metre
mg . . . . . milligram
min . . . . . minute
ml . . . . . millilitre
M . . . . . molar
mm . . . . . millimetre
MS . . . . . Murashige and Skoog medium

n . . . . . number of observations in a sample
ng . . . . . nanogram
nm . . . . . nanometre
NAR . . . . . net assimilation rate
N.B. . . . . (*nota bene*) note well
NTEMP . . . . . night temperature
P . . . . . probability
Pa . . . . . Pascal
pH . . . . . measure of acidity or alkalinity
Pmax . . . . . maximum photosynthetic rate at saturating PPF
Pn . . . . . net photosynthetic rate
PPF . . . . . photosynthetic photon flux
ppm . . . . . parts per million
r . . . . . partial correlation
R . . . . . respiration rate
r^2 . . . . . coefficient of determination
RGR . . . . . relative growth rate
RLAER . . . . . relative leaf area expansion rate
RLSWR . . . . . relative leaf starch weight rate
RLWR . . . . . relative leaf weight rate
RWP . . . . . root weight partitioning
s . . . . . second
SAS . . . . . Statistical Analysis System (statistical software)
s.e. . . . . standard error of the mean
SLA . . . . . specific leaf area
str . . . . . dry weight of structural material (i.e., minus soluble sugars and starch)
t . . . . . time
t_c . . . . . time to commencement of tuber growth
T . . . . . tuber dry weight
$T_{\%}$ . . . . . percentage tuber weight loss at the commencement of tuber growth
tanh . . . . . hyperbolic tangent
Ts . . . . . tuber starch dry weight
Tstr . . . . dry weight of tuber structural material (i.e., minus soluble sugars and starch)
TWP . . . . . tuber weight partitioning
µl . . . . . microlitre
µm . . . . . micrometre
µmol . . . . . micromole
viz. . . . . . *(videlicet)* namely
v/v . . . . . volume (mix ratio)

W . . . . . total plant dry weight
%LA . . . . . .percentage maximum leaf area
_°_'S . . . . angular distance on its meridian South of equator in degrees and minutes
δ . . . . . .mathematical notation for an interval
ΔP . . . . . difference between photosynthetic rate under saturating PPF and photosynthetic rate under the growth PPF
ns,*,**,*** . . . . . unless otherwise stated, probability of a significant F value; nonsignificant or significant at P = 0.10, 0.05, or 0.01, respectively

NOTES ON CITATION FORMAT

With a view to publishing this thesis as a series of scientific papers in journals such as those produced by the American Society for Horticultural Science (ASHS), the style of literature citation follows that recommended by ASHS. The citation system used therefore follows the Harvard system, and abbreviations for periodical titles are as suggested by ASHS.

LIST OF TABLES

PAGE

LIST OF FIGURES

PAGE

LIST OF PLATES

PAGES

1 BOTANICAL, ECOLOGICAL, PHYSIOLOGICAL, AND HORTICULTURAL BACKGROUND OF THE GENUS *Zantedeschia*.

1.1 Introduction and overview

Zantedeschia K. Spreng. species, and their hybrids, may be grown outdoors as garden plants and commercial cut flowers in temperate to sub-tropical climates. Forcing programmes are under development to provide cut flower and pot plant production year round. The range of available flower colours, foliage spotting (maculation), and plant heights has created interest in their use as bedding plants.

1.2 World production areas and volumes

Currently New Zealand produces the widest range of *Zantedeschia*, with more than 3 million flower stems of over 120 cultivars exported during 1990/91 (N.Z. Trade Development Board, personal communication). Tuber exports from New Zealand were estimated to be in excess of 1.4 million during 1988/89 (Kepner et al., 1990). While no data are available on the total area used for *Zantedeschia* production in New Zealand, it is estimated to be in excess of 100 ha (unpublished observations). Other countries with significant areas of production are The Netherlands (10 ha; 1987/88) and Israel (10 ha; 1987/88) (De Hertogh, personal communication). Japan also produces cut flowers (30 ha; 1991), predominantly *Z. aethiopica* (L.) Spreng. 'Childsiana' and cultivates a small area of pot plants (Hayashi, personal communication). However, Japan is a net importer of *Zantedeschia* flowers and tubers. While the U.S.A. also produces a significant quantity, no production data have been published since the 1940's (3.5 million plants; Butterfield, 1948; Hayward, 1948). Emerging cut flower production industries are also located in Italy, South Africa, Kenya, Mexico, Columbia and Costa Rica.

International production of true species is limited to *Z. aethiopica*, *Z. albomaculata* (Hook.) Baill., *Z. elliottiana* (Watson) Engl., and *Z. rehmannii* Engl. With ongoing breeding and selection programmes, New Zealand currently produces two species and over 120 cultivars. However, planting material of only 56 species/cultivars is currently available in commercial quantities. Together with supplies from The Netherlands and U.S.A. planting material of 61 species/cultivars are commercially available internationally. To avoid the continued use of the phrase species/cultivars, the term "Selection(s)" will be used hereafter.

1.3 Botanical classification, morphology, distribution and habitat

1.3.1 Botanical classification and morphological description

The name *Zantedeschia* was first established by Sprengel (1826), but other genus names such as *Arodes, Aroides, Calla* and *Richardia* have also been used subsequent to the first recording in 1687 (Letty, 1973). The genus has been revised on a number of occasions (Engler, 1915; Traub, 1948; Letty, 1973). It is the most recent revision by Letty (1973) which is now generally followed, in which the genus is presented as containing six species and two subspecies (Table 1.1). A seventh species, *Z. odorata* (Perry), was proposed subsequent to that revision (Perry, 1989).

Horticulturally, two important groups have been recognised.

(1) The first group is typified by *Z. aethiopica*, in which the foliage does not die down in winter in the native habitat (Letty, 1973). It flowers from late winter to late spring with male and female flowers interspersed in the lower part of the spadix (Plate 1.1). The fruits turn an orange colour and become soft and mucilaginous with maturity. For completeness *Z. odorata* is included in this group, but as discussed later in this Section differences do exist.

(2) The second group contains the five acknowledged remaining species that typically exhibit complete foliage senescence in winter and flower during the summer months (Letty, 1973). The male and female flowers are not interspersed on the spadix, being physically separated with female flowers at its base (Plate 1.1). The fruits remain firm and green with maturity.

Z. aethiopica plants grow to 120 cm tall, and comprise ovate-cordate or hastate shaped leaves up to 60 cm in length (Letty, 1973). The leaves do not commonly exhibit the maculation present in some of the other species (Table 1.1), and are, therefore, referred to as being immaculate. However, maculate forms of this species have been noted (Letty, 1973). The other species are smaller in stature, not exceeding 80 cm in height, especially *Z. rehmannii* which does not exceed 60 cm. While leaf shape of these latter species is generally similar to that of *Z. aethiopica*, plants of *Z. rehmannii* have lanceolate leaves. The degree of leaf maculation also varies between these species in group 2 (Table 1.1).

The perennating storage organ of *Z. aethiopica* is a rhizome, whereas species in group 2 have what is best described as a compact stem. While both forms may be branched, the latter is globular in shape. Classic horticultural texts describe all species of *Zantedeschia* as possessing a rhizome (Bailey, 1930), but a satisfactory answer as to whether botanically the latter of the two structures is a corm, tuber, or rhizome, has not been resolved. It is widely accepted that the storage organ of other members of the *Araceae* family, e.g.,

Colocasia, is a corm (Coursey, 1968; Okonkwo, 1987). While Traub (1948) described the storage organs of *Zantedeschia* as rhizomes or corms, Letty (1973) referred to rhizomes or tubers in her revision of the genus. While such debates are often referred to as being primarily of academic interest, the acknowledgement of the existence of the two forms is important when growing members of the two groups (refer Sections 1.5 and 1.6). In an effort to avoid confusion, at least during this thesis, the term rhizome will be used to refer to the storage organs of group 1, while the term tuber will be used for those in group 2.

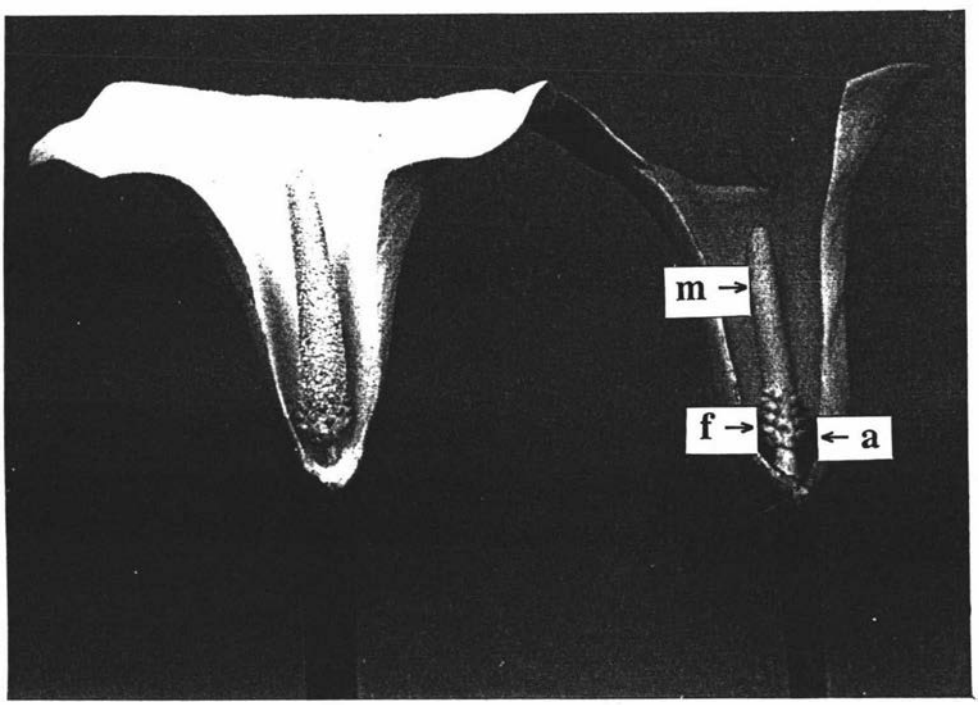

Plate 1.1 Dissection of spathe to reveal complete separation of (m) male and (f) female flowers on spadix of the group 2 selection 'Best Gold' (right), compared with being interspersed on lower part of spadix in the group 1 specie *Z. aethiopica* (left). (a) region of dark pigmentation at the base of the spathe of 'Best Gold'.

The preceding discussion of the two forms of storage organs provides strong evidence for supporting the recently reported seventh species (Perry, 1989). While leaves and flowers of *Z. odorata* appear similar to those of *Z. aethiopica*, plants grow and flower later in the season, and they do not possess the typical rhizomatous storage organ (Perry, 1989). The storage organ is similar to that of group 2. It is compact and has a pyriform shape. Unlike *Z. aethiopica*, the fruits of *Z. odorata* do not change colour with maturity but remain firm and green like those of group 2.

Table 1.1

Descriptive features of the species and subspecies of *Zantedeschia* Spreng.

Species	Group	Foliage			Flower		
		Duration	Maculation	Shape	Duration	Colour	Dark throat
Z. *aethiopica*	1	Evergreen, late winter to late spring	Rare	Ovate-cordate or hastate	Late winter to late spring	Milk-white, pink	No
Z. *rehmannii*	2	Deciduous, spring to late autumn	No	Lanceolate	Summer	White, pink, dark maroon	No
Z. *jucunda*	2	Deciduous, spring to late autumn	Yes, densely	Triangular-hastate	Summer	Golden yellow	Yes
Z. *elliottiana*	2	Deciduous, spring to late autumn	Yes	Orbicular-ovate	Summer	Golden yellow	No
Z. *pentlandii*	2	Deciduous, spring to late autumn	Seldom	Oblong-elliptic to oblong-lanceolate	Summer	Lemon-chrome yellow	Yes
Z. *albomaculata* sub. *albomaculata*	2	Deciduous, spring to late autumn	Rare	Oblong-hastate	Summer	White, ivory, pale yellow, coral pink	Yes
Z. *albomaculata* sub. *macrocarpa*	2	Deciduous, spring to late autumn	Yes, sparse	Triangular-hastate	Summer	Straw yellow	Yes
Z. *albomaculata* sub. *valida*	2	Deciduous, spring to late autumn	No	Ovate-cordate to ovate-orbicular-cordate	Summer	Ivory to cream	Yes
Z. *odorata*	1	Deciduous, late winter to late spring	No	Ovate to cordate	Late winter	Milk white	No

Reference: Letty, 1973; Perry, 1989.

Studies of the root systems have not been previously reported. In all species they are predominantly contractile and not extensively branched (unpublished observations).

The inflorescence comprises a spadix, carrying the true male and female flowers, subtended by a coloured bract known as the spathe (Plate 1.1). The inflorescence and spathe are typically presented at or above foliage height on a fleshy peduncle. The spathe may vary in shape from regularly funnel or trumpet-shaped with minimal convolutions, e.g., *Z. pentlandii* (Watson) Wittm., to a tightly folded tube with a tapering tip, e.g., *Z. rehmannii*, (Letty, 1973). Spathe colour varies from the milk-white of *Z. aethiopica*, through yellow, orange, pink and dark maroon, with the latter colours being primarily derived from the other species (Table 1.1). The inside of the spathe may also exhibit a region of dark pigmentation at its base, i.e., dark throat (Plate 1.1). The presence of this additional pigmentation is also dependent on species (Table 1.1). At initial emergence, the spathe is pigmented with chlorophyll. With subsequent unfurling the spathe develops its full colour at or near the commencement of pollen shed. In this thesis, the term "Flower" will refer to the combination of the spadix and spathe.

1.3.2 Distribution and climate of origin

While other members of the *Araceae* are endemic to South America, Asia, and Africa, the genus *Zantedeschia* is confined to the African continent. It is most prevalent in the south (i.e., Cape Province, Orange Free State, Natal, Lesotho, Swaziland, Transvaal), but also extends into Zimbabwe, Malawi, Zambia, Angola, and Nigeria (Letty, 1973). *Z. aethiopica* is almost completely confined to the south and eastern coastal belt of southern Africa, but is also found in the south-eastern mountainous regions at altitudes up to 1000 m (Figure 1.1) (Letty, 1973; Anon., 1989). While *Z. albomaculata* is widespread in its distribution across the coastal and mountainous regions of south-eastern Africa, *Z. jucunda* (Letty), *Z. pentlandii*, and *Z. rehmannii* are restricted to the eastern mountainous regions (Figure 1.1), at altitudes of 1200 to over 2000 m (Letty, 1973; Anon., 1989). The distribution of *Z. elliottiana* has not been documented, since it has not been found in the wild (Letty, 1973).

While frequently found at *"forest margins"*, the natural habitat of *Zantedeschia* species has generally been associated with open grasslands (Letty, 1973).

While exceptions are always evident, the natural distribution of perennial species has been shown to be critically dependent on the temperature minima during both the growing season and/or annual period (Korner and Larcher, 1988; Woodward, 1988). In addition, the heat sum during the season has also been shown to be influential. If like other genera,

the natural distribution of *Zantedeschia* species has been influenced by such temperature parameters, it would be expected that climatological data of the sites of origin would provide some indication of the possible temperature tolerance and growth response ranges.

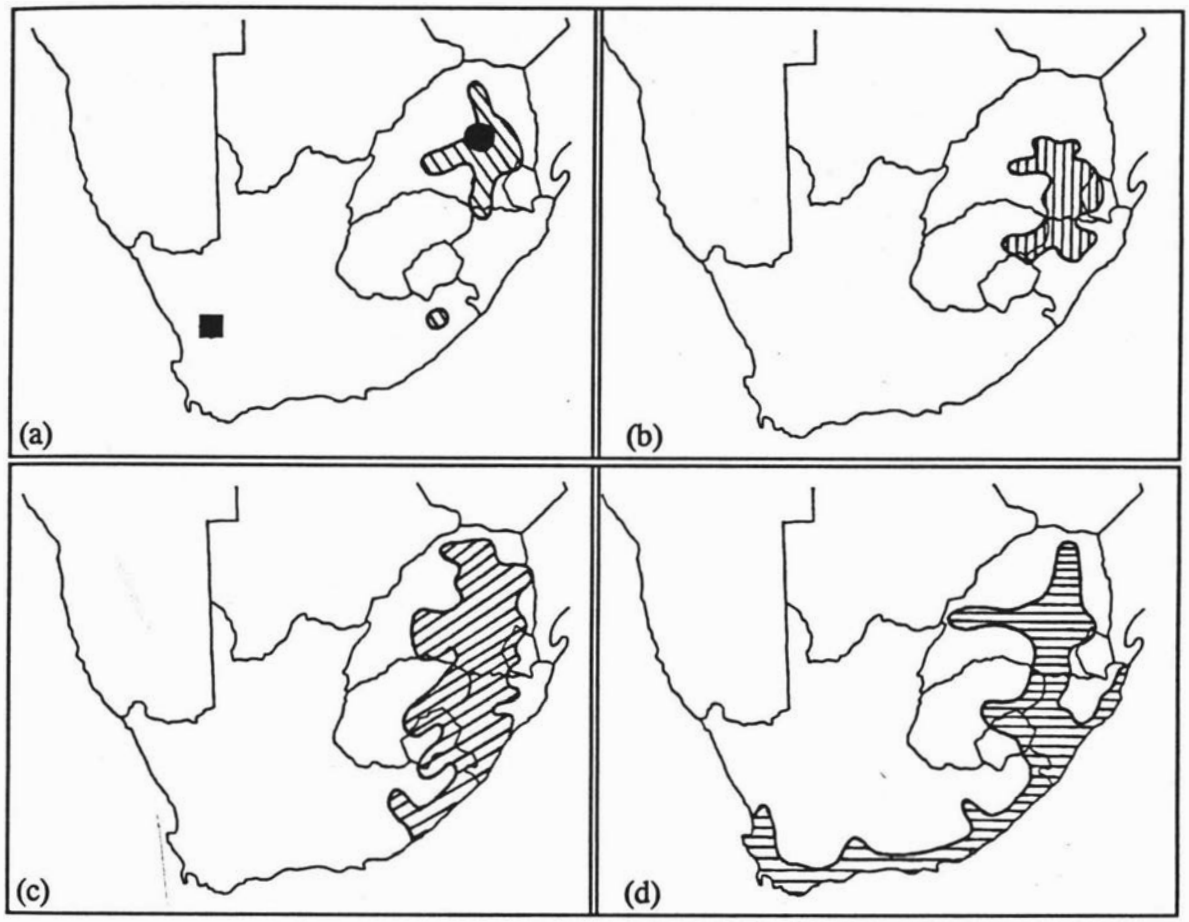

Figure 1.1 Distribution of species in the genus *Zantedeschia* across the southern regions of Africa. (a) *Z. odorata* ■, *Z. jucunda* ●, and *Z. pentlandii* ◱, (b) *Z. rehmannii* ◫, (c) *Z. albomaculata* ◰, and (d) *Z. aethiopica* ⊟. Adapted from Letty (1973); Anon (1989); Perry (1989).

Three species are discussed in an endeavour to more closely describe the climates of origin of those currently important to horticulture;

(1) *Z. aethiopica* (an example of a group 1 species).
(2) *Z. pentlandii* (an example of a group 2 species and presumed to be dominant in the parentage of the selection 'Best Gold').
(3) *Z. rehmannii* (an example of a group 2 species).

Climatological data for 24 sites of natural habitat of *Z. aethiopica*, 10 sites of *Z. pentlandii*, and 14 sites of *Z. rehmannii*, as outlined by Letty (1973) and Anon. (1989), were analysed

from 20-year average climate statistics (Anon., 1954). Due to the wide geographic distribution of *Z. aethiopica* (Figure 1.1), climatological data for this species were also analysed as two subgroups consisting of;

(1) those sites located along the south and south-eastern coast (warm).

(2) those sites located in inland mountainous regions (cool), typically also occupied by *Z. pentlandii* and *Z. rehmannii*.

This analysis by subdivision was not intended to infer the existence of ecotypes of *Z. aethiopica*, but to facilitate an appropriate focus on the climatological data of the regions where this species is most abundant.

Climatological data from the natural habitats of the three species examined varied primarily in temperature during the winter (i.e., June to July), and in the seasonal distribution of rainfall. During winter, the average mean daily air temperature of all sites of *Z. aethiopica* (12.2 C) was between 1 and 2 C warmer than that of *Z. pentlandii* (11.1 C) and *Z. rehmannii* (10.6 C, Table 1.2). The average mean daily temperature in winter, for those sites of *Z. aethiopica* identified as being cool (10.8 C), only varied from that of *Z. pentlandii* and *Z. rehmannii* by less than half a degree Celsius. However, the winter average for those sites of *Z. aethiopica* identified as being warm (14.3 C), was between 3 and 4 C warmer than that of *Z. pentlandii* and *Z. rehmannii*.

Table 1.2

Average daily maximum, minimum, mean air, and minimum soil (8.00 am, at 10 cm), temperatures (C) during the winter (June to July), for sites of natural habitat of *Zantedeschia aethiopica*, *Zantedeschia pentlandii* and *Zantedeschia rehmannii*.

	Z. aethiopica			*Z. pentlandii*		*Z. rehmannii*	
	Air			Air	Soil[w]	Air	Soil[v]
	warm[z]	cool[y]	all[x]				
max.	20.0	19.2	19.5	20.2	-	19.9	-
min.	8.5	2.4	4.9	2.9	8.1	2.7	6.1
mean	14.3	10.8	12.2	11.1	-	10.6	-

[z] data from 10 sites from coastal region.
[y] data from 14 sites from inland mountainous regions
[x] data from all 24 sites.
[w] average from 2 sites.
[v] data from 1 site.

Average daily mean air temperature during summer (i.e., October to February) for sites where each of the three species grow varied by only half a degree Celsius (Table 1.3). While mean summer temperatures varied little, the diurnal range in temperature was greater for sites where *Z. pentlandii*, *Z. rehmannii*, and those of *Z. aethiopica* identified as being cool, were located (i.e., up to 14 C). In contrast, those sites of *Z. aethiopica* identified as being warm exhibited a diurnal range in temperature of only 9.4 C in summer. Similarly, the diurnal range in temperature was also greater for these same sites in winter, with approximately a 17 C range for sites of *Z. pentlandii*, *Z. rehmannii*, and those of *Z. aethiopica* identified as being cool (Table 1.2). This was in contrast to the 11.5 C range for those sites of *Z. aethiopica* identified as being warm.

Table 1.3

Average daily maximum, minimum, mean air, and minimum soil (8.00 am, at 10 cm), temperatures (C) during the summer (October to February) for sites of natural habitat of *Zantedeschia aethiopica*, *Zantedeschia pentlandii* and *Zantedeschia rehmannii*.

	Z. aethiopica			*Z. pentlandii*		*Z. rehmannii*	
	Air			Air	Soil[w]	Air	Soil[v]
	warm[z]	cool[y]	all[x]				
max.	25.0	27.8	26.8	25.4	-	27.1	-
min.	15.6	13.8	14.5	14.3	21	14.8	17.9
mean	20.5	20.8	20.6	20.0	-	20.4	-

[z] data from 10 sites from coastal region.
[y] data from 14 sites from inland mountainous region.
[x] data from all 24 sites.
[w] average from 2 sites.
[v] data from 1 site.

Differences between species in daily minimum temperature were most evident during winter with sites of *Z. pentlandii*, *Z. rehmannii*, and those of *Z. aethiopica* identified as being cool, averaging 2.7 C (Table 1.2). This was in comparison to 8.5 C for those sites of *Z. aethiopica* identified as being warm. Even the average of all sites of *Z. aethiopica* had a winter daily minimum air temperature at least 2 C warmer than the average for the sites of *Z. pentlandii* or *Z. rehmannii*.

While data available on soil temperatures was limited to two sites of *Z. pentlandii* and one of *Z. rehmannii*, the results at least indicate the temperatures experienced by apical

meristems of these species during growth (20 C), and winter quiescence (7.1 C) (Table 1.2 and Table 1.3).

Annual rainfall was predominantly distributed during the summer for sites of *Z. pentlandii*, *Z. rehmannii*, and those of *Z. aethiopica* identified as being cool, compared with being distributed either relatively evenly or predominantly during winter for those sites of *Z. aethiopica* identified as being warm (data not presented).

In summary, *Zantedeschia* species in group 1 originate from a warm-temperate climatic zone, but also extend into cool-temperate zones. This warm-temperate zone is typified by rainfall in the winter months. In those cases where *Z. aethiopica* is endemic to regions with summer rainfall, it occupies the habitat of marshy depressions (Letty, 1973; Anon., 1989). With relatively warm temperatures all year round, plus winter rainfall, it is not surprising that foliage of *Z. aethiopica* does not die down in winter. In fact, the main period of growth and flowering is during late winter through spring, with no flowering occurring during summer (Letty, 1973). It is suggested that the lack of any pronounced differences in summer air temperatures, between sites of origin of the three species examined, indicates *Z. aethiopica* exhibits an upper temperature limit in summer, for growth and flowering, which is lower than that of group 2 species.

In contrast, species in group 2, such as *Z. pentlandii* and *Z. rehmannii*, are more restricted in their natural distribution to cool-temperate climates with associated summer rainfall. As with other temperate geophytes (Rees, 1972) it is not surprising that species/cultivars in group 2 exhibit a seasonal periodicity which requires a period of endodormancy before growth can recommence (Corr and Widmer, 1988). Thus winter is concomitant with complete foliage senescence, while growth and flowering occur during the late spring through summer (Letty, 1973).

1.4 Breeding: goals and specific problems

The incidence of bacterial soft-rot (*Erwinia carotovora*) is one of the most important factors limiting the future commercial viability of this crop. While disease avoidance production programmes have proven successful, tolerance or resistance to bacterial soft-rot needs to be developed. *Z. aethiopica* is more tolerant to *E. carotovora* than selections from group 2 (Long et al., 1988). Interspecific hybrids between the two groups may, therefore, result in transference of this tolerance to the summer flowering group (group 2).

In addition to breeding for flower colour, high floral productivity must be a primary objective of breeders. Those selections that exhibit minimal apical dominance, and

therefore tend to carry a greater number of dominant buds, will be multibranching and produce a greater number of flowers (refer Section 1.6.2). This multibranching growth habit is readily seen in selections with *Z. rehmannii* or *Z. aethiopica* as a parent. The potted plant requirement for "Fullness" is also facilitated by this multibranching growth habit, since those selections with increased shoot number result in an increased density of foliage. High floral productivity is also dependent on the proportion of shoots producing flowers (refer Section 1.6.2). With regard to this factor, it is uncertain whether or not genetic differences exist, but breeding for a growth habit that ensured that a high proportion of shoots flowered, especially after prolonged storage (refer Section 1.6.4.1) would be beneficial. In addition to high floral productivity, the ability to schedule year-round flowering is desirable. With selections classified as group 1 flowering naturally in winter/spring, and those in group 2 flowering in summer, breeders are endeavouring to achieve interspecific hybrids that will produce a greater range of flowering times and durations.

With reduced international acceptance of the use of agricultural chemicals, the development of selections with a natural dwarf growing habit has been given a high priority by breeders. In contrast, the current availability of a wide range of peduncle lengths from different selections does not present a problem for breeders focusing on cut flower or bedding plant production.

Spathe regreening is a primary determinant limiting the display life of selections in group 2 (Funnell and Downs, 1987). Selections vary in the rate of regreening (Funnell and Downs, 1987), and breeders are now utilizing this knowledge in developing improved selections with slow rates of spathe regreening. While the spathe also regreens in selections of *Z. aethiopica* (Pais and Chaves das Neves, 1982/83), the decline in quality in this specie is first seen as a wilting of the spathe (Tjia and Funnell, 1986; Plummer et al., 1990).

Little is known about market preferences for leaf and flower shape, or leaf maculation, but wide variations in these morphological features are possible (Shibuya, 1956; Harrison, 1972; Letty, 1973). Fragrance is another characteristic that, while not considered of high priority, may offer a new dimension to consumer acceptance of the product. *Z. odorata* (Perry, 1989) and selections of *Z. aethiopica* (Letty, 1973) are fragrant. The fragrance is somewhat like that of freesias, but subtle. Research is required to determine market preference for these characteristics before breeders can set new goals.

Interspecific hybrids are readily achieved between species within group 2. However, because of incompatibility, no successful crosses have been achieved between species in

group 1 and 2 (Traub, 1948; Letty, 1973; Chi, 1990). While embryo culture techniques have enabled the fertilized embryos of these incompatible crosses to be grown in vitro, the resulting plants were albino (Chi, 1990; Jialong and Cohen, 1991). The techniques of embryo culture have also been utilized to produce triploid and tetraploid selections (Jialong and Cohen, 1991). The normal chromosome number of species of *Zantedeschia* is $n = 16$ (Earl, 1957).

1.5 Vegetative growth and development

1.5.1 General overview

Following tuber planting, tuber dry weight initially declines while leaf development commences (Kobayashi et al., 1978; Funnell and MacKay, 1987; Warrington and Southward, 1989). As further leaf development continues, tuber dry weight commences to increase following a sigmoidal pattern of growth. While it is uncertain what control mechanisms are exerted over vegetative growth in *Zantedeschia*, preliminary studies point to the importance of leaf area development and its duration (Funnell and MacKay, 1987; Warrington and Southward, 1989).

1.5.2 Influence of internal factors

1.5.2.1 Dormancy

While a popular viewpoint is that selections in group 1 do not exhibit endodormancy, no published reports are evident to substantiate this. Leaf production of the group 1 selection 'Childsiana' does cease in summer (Sakanishi, 1955), but buds on harvested rhizomes readily resume growth under controlled conditions (Welsh et al., 1988; Plummer, 1990).

Bud endodormancy during winter has been reported for selections in group 2 (Corr and Widmer, 1988). Just when bud endodormancy commences is unknown, but the cessation of leaf development occurred chronologically earlier as the temperature of growth increased up to 24 C (unpublished observations). The cessation of further leaf development was not as a result of the exhaustion of a limited number of preformed leaves present at the time of planting. Dissection of primary shoots of the group 2 selections 'Galaxy' and 'Pink Petticoat' once endodormant, revealed an average of 10 ± 1 primordial structures, with the most developed primordium carrying senesced leaf lamina.

Few investigations into the influence of tuber storage temperatures have studied the phenomenon of bud endodormancy. Nearly all experiments have utilized tubers with buds

in a non-dormant state, and have, therefore, often unknowingly investigated the amount of growth and development that has occurred while tubers were held dry. Changes in endogenous growth regulator concentration and activity, which might be expected to be associated with bud endodormancy, have not been reported.

Endodormant buds on tubers of *Z. rehmannii* and *Z. elliottiana* have been induced to develop by either withholding water for 45 days from plants in leaf, or by lifting and storing tubers for 42 days at 22 C (Corr and Widmer, 1988). Plants replanted after having their foliage mechanically removed did not emerge until they also were dry stored, confirming that the buds were endodormant at the commencement of the treatment. However, the induction of bud ecodormancy by withholding water for 56 days, commencing when shoots of the group 2 selection 'Chromatella' were still in rapid growth, did not result in rapid and uniform bud growth (Funnell and MacKay, 1989b). When subsequently stored at 10 C for 84 days bud growth was found to be rapid and uniform. In both of the aforementioned studies no developing shoots had flowered after 100 days following replanting. Clearly the topic of bud eco- and endodormancy requires further investigation to assist with the development of rapid programming of tubers for forcing.

1.5.3 Influence of external factors

1.5.3.1 Temperature

While detailed analyses of growth response to temperature have not been reported for *Zantedeschia*, growth of plants of the group 1 selection 'Childsiana' was severely restricted when grown at 12 C (Halligan and Warrington, personal communication). While leaf differentiation continued at 28 C, leaf area expansion was restricted and the duration of leaf area was reduced, when compared to plants grown at 20 C. To extend the flowering season, cooling is typically required in summer (Sakanishi, 1955).

Similarly for group 2 selections, a complete temperature response curve for growth has not previously been reported. Research carried out on group 2 selections primarily pertains to flowering and will therefore be dealt with in Section 1.6.4.1. However preliminary studies indicated that leaf area development and subsequent tuber growth commenced earlier and at greater rates with increasing temperature up to 25 C (Warrington and Southward, 1989). The time interval between planting and commencement of net tuber growth was approximately 60 days at 25 C compared with 140 days at 13 C.

1.5.3.2 Light

Tuber growth occurs year-round and is, therefore, presumed to be independent of day length (personal observations).

Detailed analyses of vegetative growth and development of selections of *Zantedeschia* in response to photosynthetic photon flux (PPF) regimes have not been reported. However a preliminary study using the group 1 selection 'Childsiana' indicated that a 12 h day at a PPF regime higher than 450 $\mu mol \cdot m^{-2} \cdot s^{-1}$ resulted in chlorotic foliage regardless of temperature (White and Halligan, personal communication). In contrast, tuber growth commenced earlier and tuber size was greater when group 2 selections were grown under a 12 h day at higher (700 $\mu mol \cdot m^{-2} \cdot s^{-1}$) compared with lower (350 $\mu mol \cdot m^{-2} \cdot s^{-1}$) PPF (Warrington and Southward, 1989). In addition, leaf area expansion and duration were greater under the low PPF regime.

1.5.3.3 Chemical growth regulators

While the preplant application of gibberellic acid (50 $mg \cdot litre^{-1}$ GA_3) to tubers resulted in an increased proportion of buds emerging as shoots, total plant leaf area, leaf number and tuber size were reduced (Funnell and MacKay, 1987). The subsequent removal of flowers did not result in any difference in final tuber size, indicating that flowers were not a significant competitive sink for photoassimilates. The preplant application of GA_{4+7} between 2 and 8 $mg \cdot litre^{-1}$ also resulted in reduced tuber size (Funnell and MacKay, 1988a).

Neither the preplant application of benzyl (1*H*-purin-6-yl) amine (BA) between 0 and 7.2 $mg \cdot litre^{-1}$, nor ethylene (500 $\mu l \cdot litre^{-1}$ at 20 C for 24 h), resulted in any alteration of vegetative growth or development (Funnell and MacKay, 1988a and 1988b).

Since the primary influence of chemical growth retardants is on height control, their influence will be discussed in Section 1.6.4.4.

1.5.3.4 Growing medium, irrigation, nutrition and weed control

Growth and development of *Zantedeschia* is believed to be optimal in a free draining growing medium with pH near 6.0. While tolerance ranges for growing medium moisture content and aeration have not been quantified, it is popularly believed that *Z. aethiopica* is tolerant of wet, poorly aerated soils. However, while being tolerant, growth is not necessarily optimal under such conditions. Inadequate irrigation regimes can result in reduced leaf area development (Sakanishi, 1955). With leaf area development being a

potentially important determinant of plant and tuber growth (refer Section 1.5.3), water stress must be avoided if growth is to be maximized.

Nutrient requirements are closely correlated with the pattern of growth, with the greatest rates of uptake occurring between six to 12 weeks after field planting in summer (Clark and Boldingh, 1991). In anticipation of this growth, initial recommendations for broadcast application rates are 300 $kg{\cdot}ha^{-1}$ N, 45 $kg{\cdot}ha^{-1}$ P and 400 $kg{\cdot}ha^{-1}$ K (Clark and Boldingh, 1991).

Applications of the following residual herbicides, prior to crop emergence, have given the most satisfactory weed control while being tolerated by *Zantedeschia*: terbumeton/terbuthylazine (3 $kg{\cdot}ha^{-1}$), simazine (2 $kg{\cdot}ha^{-1}$), and oxadiazon (1.5 $kg{\cdot}ha^{-1}$) (Ingle and Bussell, 1991). Methabenzthiazuron (2 $kg{\cdot}ha^{-1}$), metribuzin (0.6 $kg{\cdot}ha^{-1}$), and prometryn (1.6 $kg{\cdot}ha^{-1}$) have been successfully applied after emergence without causing plant damage.

1.5.4 Commercial rhizome and tuber production

1.5.4.1 Goals and techniques

The primary goal of rhizome and tuber growers is to produce, in as short a period of time as practical, rhizomes or tubers that will provide multiple flowered, quality plants, that can be scheduled for production for given market periods.

Commercially, multiplication is achieved using three main systems, i.e., seed, offsets/division, and tissue culture. Only five selections are known to be commercially produced from seed, *viz.* the species *Z. rehmannii*, *Z. elliottiana*, *Z. albomaculata*, and the group 2 selections 'Chromatella' and 'Best Gold'. However only 'Best Gold' grows true-to-type from seed. Annual multiplication through offsets and division of flowering sized tubers (refer Section 1.6.3.1) can increase the number of flowering sized tubers, and total tuber number by over 100% and 300%, respectively. While this may be adequate once stock numbers are high, tissue culture techniques have been developed to provide large scale bulk-up rates, especially of virus tested lines (Cohen, 1981; Rong et al., 1989; Anon., 1990). Multiplication may also be achieved by scooping dominant buds with some tuber material attached, and subsequently treating this as a cutting (Clark et al., 1987; Funnell and MacKay, 1989a). When non-flowering sized material is replanted for annual replacement, increases in tuber dry weight are frequently in excess of 400% per annum (Funnell and MacKay, 1987). Selections of *Z. aethiopica* normally produce between 25 to 30 rhizomatous offsets after 6 months' growth (Zizzo, 1987; Welsh et al., 1988).

Under optimum growth conditions in controlled environment facilities, natural flowering plants of *Z. aethiopica* can be obtained in 9 months from seed (Cohen, personal communication), but under normal field conditions the production of flowering sized tubers of all selections takes two to three years. Depending on selection, 90% of 1 to 2 cm diameter tubers, replanted for annual tuber replacement under optimum New Zealand field conditions, will reach a flowering size by the end of one growing season (Funnell and MacKay, 1987). With tuber producers endeavouring to supply flowering sized tubers of new selections to the market place as quickly as possible, this two to three year period of cultivation is seen as being undesirably long. The need for repeated periods of cultivation, tuber lifting and storage are associated with increased costs of production which must be passed onto the market place. Hence investigations into means of reducing this time interval may provide tuber producers with the potential to reduce the costs of production.

Environmental control of cultural factors such as temperature (refer Section 1.5.3.1), light intensity (refer Section 1.5.3.2), nutrition, and irrigation regimes (refer Section 1.5.3.4) are used to maximise tuber yield. While the majority of tuber production utilizes natural mineral soils, the need to have greater control over principle environmental factors and disease (refer Section 1.7) has resulted in increasing production utilizing soilless growing media. However, as noted in the aforementioned sections, detailed analyses of vegetative growth and development in response to these environmental parameters have not been reported.

1.5.4.2 Planting to harvest requirements

Seed, offsets/divisions, and tubers for annual replacement are sown in spring in an insect, disease and weed free area. With growth potential varying among selections and environments a median spacing for seed is 5 cm square. Small non-flowering tubers, which will be lifted after one year, are spaced at 10 cm square. Tissue cultured plants are ex-flasked into containers of soilless growing media at a 3.5 cm square spacing. Under controlled environmental conditions (refer Section 1.5.3), ex-flasking can be carried out year-round. Flowering sized tubers, which will be lifted after two or more years, are spaced at 30 cm square.

When replanting for annual replacement, tubers of *Z. elliottiana* inverted at planting produced tubers of a distorted shape (Clark et al., 1987). Thus, if tuber shape is considered to be an important quality criterion, tubers should not be inverted at planting.

Without any evident period of endodormancy (refer Section 1.5.2.1), it is believed that lifting and division of plants in group 1 can be carried out at any time of the year.

However, since the natural season of growth and flowering is during the winter and spring months, this activity is generally carried out during late summer/autumn. While plants from this group may be lifted and divided for immediate replanting, plants may also be dried for a month to facilitate handling as dry rhizomes (Welsh et al., 1988). To achieve maximum tuber size, harvesting of tubers from selections in group 2 is not carried out until the foliage has senesced. Except where plants have been artificially dried down (refer Section 1.5.2.1), tubers at this stage of development will have had their endodormancy requirement satisfied. Under New Zealand conditions, buds on tubers remaining in the ground after endodormancy produce minimal growth because of the cool autumn/winter soil temperatures that prevail naturally.

1.5.4.3 Postharvest storage and transport requirements

It is popularly believed that actively growing offsets/divisions of selections in group 1 do not require curing or dormancy-breakage treatments. Replanting can be carried out once foliage and roots have been trimmed and plants treated with fungicide. If not immediately replanted plants may be coolstored in a slightly moist, free-draining growing medium until planted. Alternatively, rhizomes lifted and divided from plants which have been dried down, can be graded, treated in a fungicide solution, and cured at 18 C until remaining roots and leaf bases senesce (Welsh et al., 1988). Rhizomes are stored dry at 10 C until replanted (Sakanishi, 1955).

Once lifted and treated in a fungicide/bactericide solution, tubers of selections in group 2 are cured at 20 to 30 C and high relative humidity (i.e., 80% or greater), for a minimum of seven days, to enhance the rate of suberin and wound periderm deposition (Funnell et al., 1987). The extent of tuber weight loss, and the weight of tissue lost due to "chalking", was reduced under high (80%) compared with low (40%) humidity (Funnell and MacKay, 1988c). This "chalking", i.e., the formation of a layer of white coloured dead tissue on cut tuber surfaces, was the result of desiccation and death of cells before an adequate barrier of suberin and wound periderm could be formed.

To suppress shoot growth, non-dormant tubers of group 2 selections must be stored at 7 ± 3 C (Funnell and MacKay, 1988b). Exposure to ethylene (500 $\mu l \cdot litre^{-1}$ at 20 C for 24 h) prior to storage or planting did not alter growth (Funnell and MacKay, 1988b), hence during transport mixed container consignments with ethylene-producing materials should not cause any problems.

1.6 Control of flowering

1.6.1 General overview

With control over tuber availability, and over both the storage and forcing environments, *Zantedeschia* may be flowered year-round. Floral differentiation occurs under conditions suitable for vegetative development (refer Section 1.6.2). Therefore, the potential for flowering exists as long as development continues. Providing that a minimum growth temperature (refer Section 1.6.4.1) is met by the use of protected cultivation, the primary limitation to successfully achieving year-round flowering of selections in group 1 appears to be the cessation of flowering during the heat of summer (refer Section 1.3.2). The growth and flowering of selections in group 2 are not limited by exposure to these same summer temperatures, and, therefore, are readily programmed for forcing.

1.6.2 Flowering process and terminology

Flowering sized tubers (refer Section 1.6.3.1) have buds with a range of flowering potentials. Dominant buds are those that flower when placed under "normal" cultural conditions (refer Section 1.6.4). These dominant buds are physically swollen and encircled by a number of axillary buds arranged spirally on the tuber (Plate 1.2).

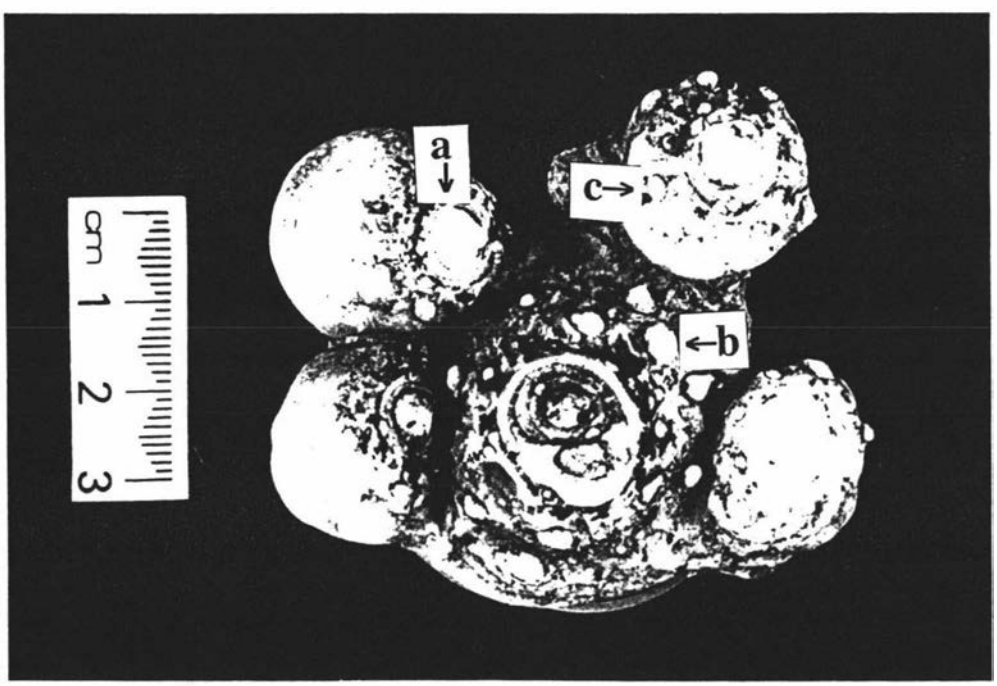

Plate 1.2 Flowering sized tuber of a group 2 selection indicating examples of (a) dominant bud, (b) developed axillary bud, and (c) undeveloped axillary bud.

Axillary buds are smaller in size than dominant buds, and are not encircled by smaller buds. The growth and flowering potential of axillary buds may be further classified as;

(1) Developed - Those axillary buds that are physically swollen and are readily induced to flower with the application of gibberellins.

(2) Undeveloped - Those axillary buds that are not swollen and are not readily induced to grow and flower, even with application of gibberellins.

The growth habit of *Zantedeschia* is sympodial, with dominant buds on tubers initially emerging to produce primary shoots (Figure 1.2) (Funnell and MacKay, 1988a). Above ground, each of these primary shoots typically present two to three protective sheath leaves followed by two leaves below the flower (Kobayashi et al., 1977; Funnell and MacKay, 1988a; Funnell et al., 1992). Additional flowers may also develop from apical meristems of secondary shoots, stimulated into growth from primordia located in the leaf axils of the primary shoot (Figure 1.3). This readily discernable developmental pattern, of secondary shoots arising from primary, may continue with tertiary shoots arising from the leaf axils of secondary shoots *ad infinitum* until endodormancy ensues. Secondary and tertiary shoots carry one bract and between zero and two leaves below the flower.

Floral differentiation commences once vegetative growth has commenced (Takahashi et al., 1957; Kobayashi et al., 1978). Therefore, unless planting is delayed, flowers are typically not differentiated at planting. The sequential differentiation of the components of the flower has been described for selections in group 1 (Takahashi et al., 1957) and group 2 (Kobayashi et al., 1978), with no differences being evident. Corr (1988) confirmed that the spathe is the first component of the flower to differentiate.

Therefore, total flower productivity per tuber is a function of:

(1) the number of buds that are stimulated to grow;

(2) the number of primary shoots that subsequently flower;

(3) the number of secondary and/or tertiary shoots that flower (refer Sections 1.6.4.1 and 1.6.4.4).

Hence, flowering sized tubers of selections derived from *Z. elliottiana*, for example, typically carrying only one dominant bud, will have an inherently lower flowering potential than tubers of similar diameter derived from *Z. rehmannii*, which typically carry four or more dominant buds (Funnell et al., 1988).

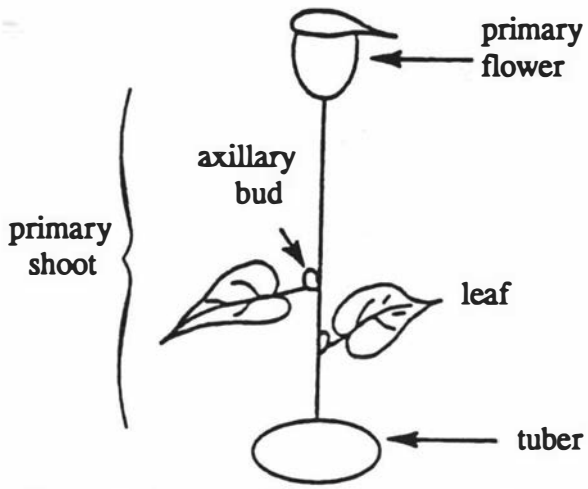

Figure 1.2 Diagrammatic illustration of sympodial growth habit of a single primary shoot of *Zantedeschia.*

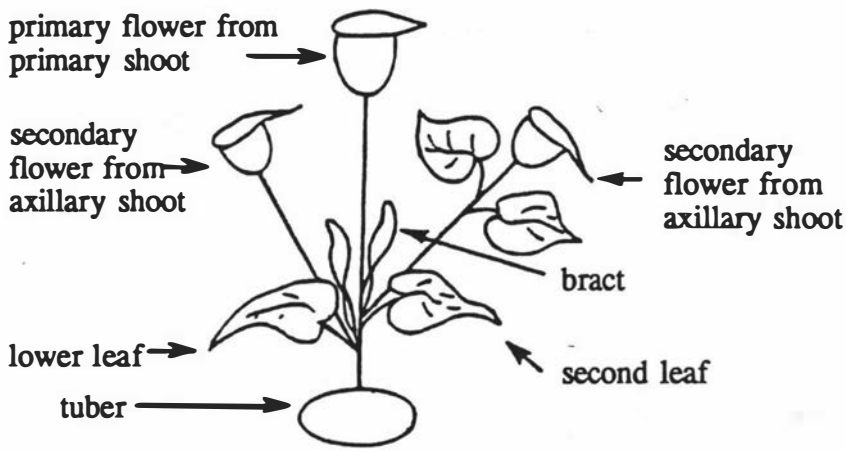

Figure 1.3 Diagrammatic illustration of primary shoot of *Zantedeschia*, with secondary shoots arising from leaf axils.

1.6.3 Influence of internal factors

1.6.3.1 Rhizome and tuber size

Rhizome size of the group 1 selection 'Childsiana' influences the growth of flowers and foliage, with smaller rhizomes producing plants with reduced height, reduced number and size of leaves, and reduced flower size (Welsh et al., 1988). While a minimum rhizome size to achieve flowering for group 1 selections has not been reported, the commercial use of the saleable product dictates the size of rhizome selected. A maximum production of 18 flowers per plant was recorded for rhizomes of *Z. aethiopica* weighing between 201 to 250 g fresh weight (Zizzo, 1987). Rhizomes of both smaller (50 to 200 g), and larger (251 to 300 g) weight, produced fewer flowers.

While varying between selections, a 2.5 cm tuber diameter is necessary to ensure 100% natural flowering of group 2 selections not exposed to extended periods of storage (Corr and Widmer, 1991). Tubers of a smaller size are readily induced to flower by the application of gibberellins (Cohen, 1981; Funnell and Go, 1993), but foliage and flower size are smaller than that achieved with larger sized tubers. Again, the commercial use of the saleable product dictates the tuber size selected.

1.6.3.2 Dormancy and floral induction

Dormant buds of group 2 selections induced to grow, either by withholding water (45 days) or manipulating temperature in storage (42 days at 22 C or 84 days at 10 C), did not flower (refer Section 1.5.2.1) (Corr and Widmer, 1988; Funnell and MacKay, 1989b; Corr and Widmer, 1991). In addition, buds on tubers of *Z. rehmannii* which were lifted from the field in California, U.S.A. during early September, emerged within 41 days, but did not flower over a subsequent 150 day growing period (Corr, 1988). However storage of these tubers at 15 C for a minimum of 21 days resulted in flowering, with the number of flowers per plant increasing with longer periods of storage. While the emergence of shoots from non-stored tubers indicated that the buds were not dormant, the inability of plants from these tubers to flower unless tubers were stored for 21 days, indicates that something other than the presence of vegetative growth is needed to achieve flowering. It is possible that the level of bud ecodormancy induced by artificially drying the plants may be very different from the endodormancy encountered under normal cultural conditions. Clearly the topics of bud eco- and endodormancy, and their association with floral induction, requires further investigation to assist with the development of programming tubers for forcing.

1.6.4 Influence of external factors

1.6.4.1 Temperature

As noted in Section 1.6.2, differentiation of flower parts does not commence until shoot growth has begun. Shoot growth occurring during tuber storage or forcing, at a range of temperatures, did not result in any alteration of the sequence of floral differentiation (Funnell and MacKay, 1988b; Corr, 1988). Therefore, it is assumed that as long as the requirements for minimum rhizome or tuber size (refer Section 1.6.3.1) and endodormancy (refer Section 1.6.3.2) have been satisfied, temperatures conducive to vegetative growth are also conducive to floral differentiation. However, while the growing temperature does not result in differences in flower productivity of selections in group 2 (Warrington and Southward, 1989), flowering of the group 1 selection 'Childsiana' was inhibited at 28 C (Halligan and Warrington, personal communication).

Plant growth is faster at increasing temperatures, and flower development is also accelerated (Post, 1936). Plants of the group 2 selection 'Galaxy' grown in a controlled environment temperature regime of 28/22 C day/night, reached first flowering 57 $\pm$ 1 days after planting, but this increased to 80 $\pm$ 2 days when grown at 22/16 C, and to 140 $\pm$ 3 days at 16/10 C (Warrington and Southward, 1989). When grown under controlled conditions in the greenhouse (minimum air temperature of 15 C and ventilation at 20 C), comparable results were obtained with an average 70 days to flower for four selections in group 2 (MacKay et al., 1991). Selections in group 1 responded similarly, with plants of 'Childsiana' taking 88 days to flower at 20 C (White and Halligan, personal communication). Basal heating at temperatures above ambient air temperatures can reduce the time to flowering (Corr and Widmer, 1990).

In addition to reducing time to flower, increasing temperatures increased total plant height and peduncle length. Warrington and Southward (1989) reported a 13% increase in peduncle length when the daily mean temperature was increased from 19 to 25 C. The use of night temperatures which were higher than day temperatures did not result in significant differences in total plant height or peduncle length compared with use of the reversed day/night regime, or with constant mean temperatures (Reiser and Langhans, 1993). Hence the application of the technology of programmed diurnal temperature control, i.e., DIF (Karlsson et al., 1989), may not provide a satisfactory means to reduce plant height.

The effects of storage temperature on non-dormant rhizomes of selections in group 1 have not been extensively studied. Sakanishi (1955) determined that if storage was required, subsequent growth and flowering of *Z. aethiopica* 'Childsiana' was optimal after storage

of plants in leaf at 10 C. Welsh et al. (1988) reported increased leaf production of 'Childsiana' if rhizomes were dried, and cured at 18 C compared with immediate replanting of offsets/divisions in leaf. However, no resultant increase in flowering was reported. With selections in group 2 a storage temperature of 7 ± 3 C is recommended to inhibit shoot growth (refer Section 1.5.4.3). Inhibition of shoot growth is important if physical damage during transport is to be minimized. However, a reduced time to flower can be achieved by storage at warmer temperatures and/or with increasing duration of storage (Tjia, 1987a; Funnell and MacKay, 1988b). The earlier flowering was attributed to shoot growth and subsequent floral differentiation in storage. Further research is required to develop combinations of cool and warm storage into commercial scheduling programmes.

In addition to influencing the time to flower, short-term storage of non-endodormant tubers at fixed temperatures ranging between 8 and 15 C, increased the proportion of buds emerging as shoots (Corr and Widmer, 1988; Funnell and MacKay, 1988b). In contrast, tubers stored at the higher temperatures of 22 or 25 C did not respond similarly. The rapid establishment of apical dominance (i.e., paradormancy) at the high temperatures resulted in the reduced number of buds developing into shoots. The resultant proportion of buds at planting that subsequently flowered was also increased after storage at these cooler temperatures with the optimum proportion occurring after 10 weeks at 8 C or 4 weeks at 15 C. After consideration of the unknown comparative endodormancy status, this result is not too dissimilar to that reported by Corr (1988), where total flower number was greatest after 12 weeks storage at 15 C.

While storage of non-endodormant tubers permits scheduled planting programs, the flowering potential of these tubers is reduced compared with non-endodormant tubers planted without any period of storage (Funnell et al., 1988). The proportion of tubers of *Z. elliottiana* which flowered was progressively reduced with increased storage temperature up to 24 C, but not with the *Z. rehmannii*-like selection 'Pink Satin'. The maintenance of the proportion of tubers of 'Pink Satin' flowering, with increased storage temperature, reflects the greater number of dominant buds per tuber, and therefore greater potential floral initiation sites (refer Section 1.6.2). Tubers of *Z. elliottiana* used in this experiment typically carried only one dominant bud compared with the four or five dominant buds on tubers of 'Pink Satin'. It is unlikely that this decline in flowering resulted from chilling injury or abortion of the apex, since the reduction in flowering occurred regardless of storage temperature, *viz.* 5, 12, 18, or 24 C (Funnell et al., 1988). Also, dissection of buds did not reveal abortion of apices (Funnell and Go, 1993). While the actual mechanisms involved in this reduction in flowering potential have not been determined, a preplant application of gibberellin (GA_3 or GA_{4+7}) is able to compensate almost totally for

the reduction in flowering potential (refer Section 1.6.4.4) (Funnell et al., 1988; Funnell and Go, 1993). In addition, storage of tubers in moist media partially alleviated the reduction in flowering potential associated with storage, but was not as effective as the application of gibberellin (Funnell et al., 1988).

Not only did increased temperatures during tuber storage result in a reduction in flowering potential, but increasing duration of storage also resulted in a reduction in flower number (Funnell et al., 1988; Funnell and MacKay, 1990). While the preplant application of gibberellins was able to maintain commercially acceptable floral productivity for up to 9 months storage, the rapid increase in plant death with storage beyond 5 months, identifies the need for further research into using extended periods of tuber storage to schedule flowering (Funnell and MacKay, 1990).

1.6.4.2 Light

Flowering of *Zantedeschia* is not dependent on photoperiod (Greene et al., 1932; Corr and Widmer, 1990). However, under an 8 h photoperiod, the addition of a 4 h night interruption of non photosynthetically active radiation, increased plant height of *Z. elliottiana* by 40% and flower peduncle length by 23% (Corr and Widmer, 1990). The influence of photoperiod on peduncle length was highly dependent on selection, since the same treatment resulted in a 95% increase with *Z. rehmannii*.

Warrington and Southward (1989) reported a 26% increase in peduncle length from plants of the *Z. rehmannii*-like selection 'Galaxy' with a 50% reduction in PPF (700 to 350 $\mu mol \cdot m^{-2} \cdot s^{-1}$). These environments resulted in total daily integrals of light of 30 and 15 $mol \cdot m^{-2}$, and are similar to that intercepted during clear skies in summer and winter (Latitude 45°S), respectively. Other researchers have reported increases in peduncle length and total plant height, as a result of reduced PPF, ranging between 13% and 87% depending on growing environment and selection (MacKay et al., 1987b; Corr and Widmer, 1990). While differential responses of selections may account for the preceding variation in results, a more pronounced increase in peduncle length with reduced PPF resulted when plants were grown under cooler temperatures (MacKay et al., 1987b; Warrington and Southward, 1989).

While Corr and Widmer (1990) reported no reduction in total flower production, as a result of a PPF reduction by 45% or 15% of normal summer conditions, Warrington and Southward (1989) reported a 40% reduction in flower number with a reduction in PPF from 700 to 350 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Although Corr and Widmer (1990) did not report total daily integrals of light under each treatment environment, their treatments would have

encompassed the range reported by Warrington and Southward (1989). Clearly, further research is required to clarify this apparent contradiction in response to PPF and/or daily light integral.

The time to flowering was not influenced by PPF regime (MacKay et al., 1987b; Warrington and Southward, 1989; Corr and Widmer, 1990).

As noted in Section 1.5.3.2, foliage of the group 1 selection 'Childsiana' became chlorotic when grown under a 12 h PPF regime greater than 450 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (White and Halligan, personal communication).

1.6.4.3 Nutrition

The proportion of plants flowering was optimal when liquid fertilizer (15N-7P-14K) was applied twice weekly at 100 or 200 ppm N, but was reduced by 40% when increased to a rate of 400 ppm N (MacKay et al., 1987a). As a result of salinity damage, the incidence of plant death was increased at the higher rate. Using a slow release fertilizer (19N-2.6P-10K) at rates of 2 $g \cdot litre^{-1}$ and 4 $g \cdot litre^{-1}$, provided optimal flowering in peat and bark-based growing mediums, respectively (Dennis, 1986). In view of this preceding information, crop nutrition during forcing does not appear to influence flowering unless fertilizing at excessively high rates, or if moisture is inadequate.

1.6.4.4 Chemical growth regulators

Application of gibberellin (GA_3 or GA_{4+7}) promotes flowering in *Zantedeschia* selections of both groups (Corr and Widmer, 1987; Plummer, 1990), and *Z. odorata* (unpublished observations). Corr and Widmer (1987) reported optimal flowering after applying 500 $mg \cdot litre^{-1}$ GA_3 as a preplant tuber immersion. However Funnell and Go (1993) illustrated that maintenance of floral productivity after increasing periods of tuber storage required an increased concentration of gibberellin. This requirement reflects the decline in flowering potential with tuber storage duration (refer Section 1.6.4.1). Further research is required to provide recommended rates of gibberellin application after removal from storage at progressively increasing periods. Treatment with gibberellins prior to storage at 9 C did not increase flower number beyond one months' storage (Funnell and MacKay, 1990). It is important, therefore, if cool-storage is to be used, that tubers are treated after removal from storage. The duration of tuber immersion in gibberellin, ranging from 10 seconds to 30 minutes, did not result in differences in flower number (Funnell et al., 1992). Therefore, it is not surprising that spray applications of gibberellins to the tuber have also been effective (Funnell et al., 1988). Kobayashi (1980) showed that the

temperature of the gibberellin solution in which the tubers are immersed, *viz.* 5, 15, 25 and 35 C, did not influence flowering.

Both GA_3 and GA_{4+7} are effective, but GA_{4+7} was found to be more effective when applied at the same concentration as GA_3 (Funnell and MacKay, 1988a). Both sources of gibberellin increase flower production through an increase in the number of buds emerging as primary shoots, in addition to an increase in the proportion of shoots flowering. In New Zealand, a label recommendation exists for the pre-plant tuber immersion in GA_{4+7} (i.e., Promalin: Abbott Lab., N. Chicago, IL 60064) at a rate of 8 mg·litre^{-1} for 1 minute. However, while 100% of dominant buds flowered when following current label recommendations, only 30% of all buds (i.e., dominant + axillary), and 60% of shoots flowered (Funnell and MacKay, 1988a). Clearly, this indicates that further potential flowering sites exist. The BA component of Promalin did not influence flower production (Funnell and MacKay, 1988a).

Foliar application of GA_3 at rates of 50 and 150 mg·litre^{-1} increased flower production by three- and five-fold, respectively (Kobayashi, 1977a). The combination of a preplant tuber immersion (100 ppm GA_3) and subsequent foliar application (100 mg·litre^{-1} GA_3) resulted in a seven-fold increase in total flower number. In contrast, when only a preplant tuber immersion was used a five-fold increase was obtained (Kobayashi, 1977b & 1977c).

Most selections of *Zantedeschia* suitable for potted plant production require growth retardation to control plant height. Tjia (1987c) reported that the application of the chemical growth retardant paclobutrazol, as a growing medium drench at 4 mg/pot, resulted in plants with aesthetic proportionality. Ancymidol was not found to be effective (Tjia, 1987c), but uniconazole at 4 mg/pot resulted in similar results to that of paclobutrazol (Reiser, 1990). The application of paclobutrazol as a preplant tuber immersion of 80 mg·litre^{-1} for 24 h was more effective in reducing height than that achieved with the 4 mg/pot growing medium drench (MacKay et al., 1986). When paclobutrazol was applied as a preplant tuber immersion, foliage height, flower peduncle and spathe length were reduced by 40%, 56% and 10%, respectively. Regardless of which growth retardant is used, the rate of application to achieve aesthetic proportionality will be dependent both on selection and on the forcing conditions (refer Sections 1.6.4.1 and 1.6.4.2). The application of paclobutrazol or uniconazole also reduces the number of flowers produced, with the application as a preplant tuber immersion reducing flowers more than that if applied as a growing medium drench (Corr, 1988; Reiser, 1990). While the application of gibberellic acid partially alleviated the reduction in flowering, higher concentrations of gibberellic acid were required to achieve the same floral productivity when a growth retardant was applied (Corr, 1988).

1.6.4.5 Air pollutants

In contrast to bulbous crops such as tulip and iris (Kamerbeek and De Munk, 1976), growth and flowering of *Zantedeschia* was not affected by exposure of non-dormant tubers to ethylene (500 $\mu l \cdot litre^{-1}$ at 20 C for 24 h) either prior to storage or planting (Funnell and MacKay, 1988b).

1.6.5 Commercial forcing for pot and cut flower production

Information presented in this Section covers aspects of commercial pot and cut flower forcing not already covered above.

1.6.5.1 Rhizome and tuber storage

Optimum flower productivity is obtained when non-endodormant rhizomes or tubers are planted without storage (refer Section 1.6.4.1). While lifting of rhizomes of selections in group 1 may be carried out at any time of year, rhizomes are normally available from the southern hemisphere in December through February, and the northern hemisphere in June through August. Application of current technology allows tubers of selections in group 2 to be stored for up to 5 months after breakage of endodormancy and lifting (refer Section 1.6.4.1). In New Zealand, tubers are lifted in May through July and processed prior to shipping in June through December. In the northern hemisphere tubers are lifted in October through November and processed prior to shipping in January through April. Therefore, potted plant and cut flower forcers can expect to store tubers for a further one to four months, as dictated by lifting date.

1.6.5.2 Pre-plant treatments

Between arrival and planting, tubers should be dipped in a fungicide/bactericide solution. Tubers should be allowed to air dry before further handling. The use of disease-free planting material and proper sanitation practices are of prime importance in disease control (refer Section 1.7).

Flowering of *Zantedeschia* may be improved by over 400% by the application of gibberellins (refer Section 1.6.4.4). While a label recommendation exists in New Zealand for a pre-plant tuber immersion in GA_{4+7} (i.e., Promalin: Abbott Lab., N. Chicago, IL 60064) at a rate of 8 $mg \cdot litre^{-1}$ for 1 minute, application of higher concentrations of both GA_3 and Promalin have increased flowering, and may be a requirement to maintain satisfactory floral productivity with increased periods of tuber storage (refer Section 1.6.4.1), or if chemical growth retardants are used (refer 1.6.4.4).

1.6.5.3 Planting and nutrition

Satisfactory rhizome or tuber size for flowering varies with selection and desired use (refer Section 1.6.3.1). For pot plant usage of group 1 selections, generally only a single rhizome of a minimum diameter of 1.5 cm and several offsets of 0.5 to 1 cm in diameter are placed in a 0.7 litre pot. For selections in group 2 suitable for pot production, a single tuber measuring 4 to 5 cm in diameter with at least three dominant buds (refer Section 1.6.2) is considered suitable for a 15 cm pot (approximately 1.5 litre volume). For a 10 cm pot, a single tuber measuring 3 to 4 cm in diameter with three dominant buds is considered suitable. Since flower size varies with rhizome or tuber size (refer Section 1.6.3.1), the selection of rhizome or tuber size for cut flower production is dictated by market preferences. However, for group 2 selections, a minimum tuber diameter of 4 cm is generally preferred. Cut flower producers using tubers for forcing should also seek a high number of dominant buds per tuber (refer Section 1.6.2).

Unlike most other bulbous crops, roots of *Zantedeschia* emerge from around the emerging shoots at the top of the tuber. Therefore, it is important when planting that the top of the tuber is covered with 2 to 3 cm of medium to avoid roots drying out. Tubers should be planted upright with minimal damage to buds. Planting tubers upside down delays flowering and results in poor quality plants (Clark et al., 1987).

Media and nutrition requirements have been discussed above in Sections 1.5.3.4 and 1.6.4.3.

1.6.5.4 Height control

Some form of height control is often required in the production of *Zantedeschia* as pot plants. Paclobutrazol has been found to be an effective growth retardant. It can be applied as a growing medium drench at a rate of 2 to 4 mg/pot at shoot emergence. The precise rate depends on the selection and the forcing environment conditions (1.7.4.1 and 1.7.4.2). For crops in 15 and 20 cm pots, the recommended volume of diluted paclobutrazol to apply is 120 and 180 ml, respectively.

Root confinement appears to provide adequate height control of the group 1 selection 'Childsiana' obviating the need to use chemical growth retardants for pot production (unpublished observations).

1.6.5.5 Forcing environment

A day/night temperature regime of 20/15 C to 25/18 C is suitable for growth (refer Section 1.6.4.1). An 88-day production period should be used when initially planning crop schedules for group 1 selections, while 70 days should be used for group 2 selections. Forcers may find that under their particular cultural conditions (refer Sections 1.6.4.1 and 1.6.4.2) and depending on prior storage history of the tubers (1.6.4.1), modifications may be required to these schedules.

While under controlled environment conditions plants of group 1 selection 'Childsiana' produce a higher quality plant if grown under a PPF near 450 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (daily light integral of 19 $mol \cdot m^{-2}$), group 2 selections produce a higher quality pot plant if grown under a minimum PPF of 700 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (daily light integral of 30 $mol \cdot m^{-2}$) (refer Section 1.6.4.2). Increases in peduncle length as a result of manipulation of the environment (refer Sections 1.6.4.1 and 1.6.4.2) may be desirable for cut flower production, but pot forcers must give consideration to minimum light intensities, photoperiod and temperature if excessive plant height is to be avoided. The use of chemical growth retardants to assist control of plant height was discussed above (refer Sections 1.6.4.4 and 1.6.5.4).

Both temperature and PPF can have an important effect on flower colour. Cool temperatures and high PPF enhance colour expression of some pink through dark maroon selections (MacKay et al., 1987b; Corr, 1988). Lowering greenhouse night temperatures to less than 15 C or moving potted plants to an outdoor environment during spathe opening enhances colour expression without greatly increasing production time (personal observations). Flower colour of those selections in the cream through yellow to gold range is generally not affected by growing environment. While with some selections the expression of colour may change with environment, some selections are naturally dynamic in their expression as the flower develops.

1.6.5.6 Physiological disorders

Some incidents of malformed flowers after gibberellin application have been reported (Corr and Widmer, 1987; Tjia, 1987b; Funnell et al., 1992). These malformations typically consist of fluted spathes, double spathes, and coloured leaves. While the number of malformed flowers increased with increasing rates of gibberellin, the proportion of the total number of flowers that were malformed was not increased (Funnell et al., 1992). In addition, the classification of malformation is highly subjective. In some cut flower markets, premium prices have been paid for malformed flowers such as those with double spathes.

The expression of flower colour (refer Section 1.6.5.5), and the occurrence of "chalking" on cut surfaces of tubers (refer Section 1.5.4.3), have been discussed previously.

1.6.5.7 Post-greenhouse handling and marketing

Potted *Zantedeschia* in group 1 should be marketed when the first flowers are unfurling (Plummer et al., 1990). However, as a result of the need to enhance flower colour, potted group 2 selections are often not marketed until the first flowers are showing colour. Standard procedures for shipping flowering pot plants should ensure maintenance of quality, i.e., keep cool, at 10 C, and well ventilated.

Harvesting of cut flowers is carried out one to two days prior to pollen shed. This coincides with the development of full spathe opening and colour. Flowers of *Z. aethiopica* may also be bud-harvested when the spathe is still cigar-shaped (unpublished results). Harvest at this stage of development can be used to facilitate packaging and anticipate long market periods, without reducing vaselife. Investigations into bud harvesting of flowers from group 2 selections have not been reported.

Pulsing cut stems in a solution of 0.1 M sucrose and 0.5 g·litre^{-1} iprodione successfully minimized the incidence of stem splitting and postharvest spathe spotting (refer Section 1.7) (Tjia and Funnell, 1985). The use of commercial preservatives commonly available on the market, or solutions containing 3 g·litre^{-1} sucrose, resulted in rapid dehydration and necrosis of the spathes of *Z. aethiopica* (Bakker and Stephan, 1961). The vaselife of cut flowers of selections in group 2 was not influenced by exposure to ethylene (10 μl·litre^{-1} at 20 C for up to 48 h) subsequent to harvest (Funnell and Downs, 1987).

1.7 Diseases and insects

The major diseases affecting *Zantedeschia* are:

(1) *Alternaria* species (Tjia and Funnell, 1985). Symptoms are chlorotic/necrotic leaf margins, and black spots on the spathe.

(2) *Erwinia carotovora* (Dosdall, 1955; Forsberg, 1963; Long et al., 1988). Bacterial soft-rot; symptoms are erratic shoot emergence, shoot collapse, and tuber decay. This pathogen has also been associated with postharvest collapse of the peduncle of cut flowers. A pungent odour is associated with tissue collapse.

(3) *Penicillium* species (unpublished observations). Symptoms are blue-green mould on surface of tubers during storage. Considered more unsightly than detrimental to growth.

(4) *Phytophthora* species (Dosdall, 1955; Forsberg, 1963). Tuber rot, root rot, leaf blight; symptoms are yellowing/necrotic leaf and spathe margins, shoot collapse, and/or tuber and root decay.

(5) *Pythium* species (Forsberg, 1963; Long, 1988). Symptoms are translucent or pink coloured tissue on infected tubers and roots. Translucent lesions on tuber surface increase in size, but remain odourless.

(6) *Rhizoctonia* species (unpublished results). Symptoms are shoot collapse, and root and tuber rot.

(7) *Xanthomonas campestris* (Joubert and Truter, 1972). Symptoms are water-soaked lesions on leaves that result in leaf collapse under moist conditions. Under dry conditions lesions become chlorotic and necrotic.

(8) Virus diseases reported (Rana et al., 1983; Long, 1988; Anon., 1990) include:

(a) alfalfa mosaic virus;

(b) arabis mosaic virus;

(c) cucumber mosaic virus (CMV), with symptoms of mosaic foliage patterning;

(d) dasheen mosaic virus (DMV), causing leaf distortion, ring-spotting of foliage, and colour break in flowers;

(e) potato virus x;

(f) tomato spotted wilt virus (TSW), causing white or yellow foliage spots, or spots on leaves and flowers.

Generally *Zantedeschia* are remarkably tolerant of most diseases. Apart from tuber soft-rot, disease outbreaks are rare. Latent tuber infection was found to be a major source of inoculum of the soft-rot causing bacterium *Erwinia carotovora* (Long et al., 1988). While the pathogen did not survive in soil for more than five months, cross-contamination from plant material and equipment can provide a ready source of inoculum. The use of disease-free planting material in addition to sanitation practices that avoid disease should ensure minimal occurrence of diseases. Since dry tubers infected with soft-rot often appear firm and healthy, it is important for growers to ensure a clean health status at the time of tuber purchase. While laboratory tests have indicated streptomycin as the most effective chemical inhibiting growth of *E. carotovora* (Long et al., 1988), use of this chemical in the field has not been satisfactory.

Insects of significance in field or greenhouse production of *Zantedeschia* include thrips and aphids. These can be readily controlled by standard insecticides. Root-knot nematode has been reported as a problem in the U.S.A. (Hayward, 1948) resulting in the need for soil sterilization/nematocide treatments.

1.8 Miscellaneous physiological and biochemical studies

1.8.1 Spathe regreening

During the formation of fruit on *Z. aethiopica*, the cytokinins 6-(O-hydroxybenzylamino)-9-β-D-ribofuranosylpurine and 6-(O-Hydroxybenzylamino)-2-methylthio-9-β-D-glucofuranosylpurine have been isolated (Chaves das Neves and Pais, 1980a and 1980b). These naturally occurring cytokinins, as well as the synthetic cytokinin 6-(O-hydroxybenzylamino)-purine, have been associated with initiating the onset of regreening in the spathe (Pais and Chaves das Neves, 1982/83). The regreening of the spathe has been associated with a 341% increase in chlorophyll content (Pais and Chaves das Neves, 1982/83) as amyloplasts, in *Z. aethiopica*, or chromoplasts, in group 2 selections, convert to chloroplasts (Gronegress, 1974). While removal of the spadix prevented regreening in *Z. aethiopica* (Pais and Chaves das Neves, 1982/83), no significant response was evident in group 2 selections (Tjia, 1986). However, the possibility of different mechanisms initiating regreening in selections of the two groups has not been reported.

1.8.2 Tuber respiration

Non-endodormant tubers of the group 2 selection 'Pink Petticoat' respired at a rate of 20 $mg \cdot kg^{-1} \cdot hr^{-1}$ CO_2 at a temperature of 20 C (Funnell and MacKay, 1990). For the first three weeks of storage, respiration rates were higher for tubers removed from 25 C storage than in those removed from either 15 C or 8 C. The rates of respiration were not influenced by previous storage temperatures subsequent to this initial three week period. Exposure of tubers to ethylene (500 $\mu l \cdot litre^{-1}$ at 20 C for 24 h) prior to storage at 25 C resulted in respiration rates being reduced by 50% for the first two weeks of storage, compared with non-ethylene treated tubers (Funnell and MacKay, 1990). Subsequent to this initial two week period, respiration rates of ethylene treated tubers returned to that of non-ethylene treated. No ethylene related responses were obtained in subsequent growth and flowering.

1.9 Concluding remarks

The species and hybrids of *Zantedeschia* offer a wide diversity of uses for the ornamental industry. While a comparatively new crop, in recent years the amount of research and commercial attention has expanded rapidly. With distinct physiological differences being evident between the two horticultural groups, it is evident that research must recognise these differences when developing industry orientated production programmes.

While the goals outlined under the section on breeding (refer Section 1.4) form part of the future research requirements for this crop, more generic areas for future research are:

(1) improved resistance and/or control of bacterial soft-rot (*E. carotovora*).

(2) determining the factors controlling vegetative growth and tuber development.

(3) determining the factors controlling bud eco- and endodormancy.

(4) more precisely determining the factors that control floral initiation and development.

(5) development of post-harvest treatments to enhance flower longevity, especially aimed at delaying spathe regreening of group 2 selections.

1.10 References

Anon. 1954. Climate of South Africa. Part 1 - Climate statistics. Climatology Branch, Weather Bureau, Dept. Transport. Govt. Printer, Union of Sth. Africa, Pretoria.

Anon. 1989. *Zantedeschia.* In: Pretoria National Herbarium Computerized Information System (PRECIS). Amalgamated Botanical Research Institute/National Botanic Gardens, Pretoria, South Africa.

Anon. 1990. Virus-free callas - world first for NZ. Commercial Horticulture, N.Z. May, 1990, p. 16-19.

Bailey, L.H. 1930. The Standard Cyclopedia of Horticulture, Vol III, The Macmillan Company, London.

Bakker, J. and H. Stephan. 1961. Experiments on the vase life of cut flowers. Jversl. Proefst. Bloem. Aalsmeer 1:133-135.

Butterfield, H.M. 1948. The *Zantedeschia* (Calla) industry in California, p. 34-35. In: H.P. Traub and H.N. Moldenke (eds.). Plant Life, Vol. 4, Aroid Lily Edition. Amer. Plant Life Soc., Stanford.

Chaves das Neves, H.J. and M.S.S. Pais. 1980a. A new cytokinin from the fruits of *Zantedeschia aethiopica.* Tetrahedron Letters 21:4387-4390.

Chaves das Neves, H.J. and M.S.S. Pais. 1980b. Identification of a spathe regreening factor in *Zantedeschia aethiopica*. Biochem. Biophys. Res. Comm. 95:1387-1392.

Chi, Y. 1990. A study of the incompatibility within the genus *Zantedeschia.* M.Hort.Sc. dissertation, Massey Univ., Palmerston North, N.Z.

Clark, C.J. and H.L. Boldingh. 1991. Biomass and mineral nutrient partitioning in relation to seasonal growth of *Zantedeschia*. Scientia Hort. 47:125-135.

Clark, H.K., K.A. Funnell and B.R. MacKay. 1987. Influence of scooping and tuber orientation on shoot growth and multiplication of *Zantedeschia.* Tech. Rpt. 87/12, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Cohen, D. 1981. Micropropagation of *Zantedeschia* hybrids. Proc. Int. Plant Prop. Soc. 31:312-317.

Corr, B.E. 1988. Factors influencing growth and flowering of *Zantedeschia elliottiana* and *Z. rehmannii*. PhD thesis, Univ. Minnesota, U.S.A.

Corr, B.E. and R.E. Widmer. 1987. Gibberellic acid increases flower number in *Zantedeschia elliottiana* and *Z. rehmannii*. HortScience 22:605-607.

Corr, B.E. and R.E. Widmer. 1988. Rhizome storage increases growth of *Zantedeschia elliottiana* and *Z. rehmannii*. HortScience 23:1001-1002.

Corr, B.E. and R.E. Widmer. 1990. Growth and flowering of *Zantedeschia elliottiana* and *Z. rehmannii* in response to environmental factors. HortScience 25:925-927.

Corr, B.E. and R.E. Widmer. 1991. Paclobutrazol, gibberellic acid, and rhizome size affect growth and flowering of *Zantedeschia*. HortScience 26:133-135.

Coursey, D.G. 1968. The edible aroids. World Crops 20:25-30.

Dennis, D.J. 1986. Pilot study on nutrition of *Zantedeschia* hybrids in two container media. Final Report, Levin Horticultural Research Centre, Ministry of Agriculture and Fisheries, New Zealand.

Dosdall, L.T. 1955. Calla rhizome treatments. Plant Dis. Rptr. 39:779-780.

Earl, G. 1957. Chromosome numbers and speciation in the genus *Zantedeschia*. Plant Life 13:140-146.

Engler, A. 1915. Araceae. In: Das Pflanzenreich IV, 23 Dc:61. Leipzig: Engelmann. (Cited by Letty, 1973).

Forsberg, J.L. 1963. Diseases of ornamental plants. Univ. Illinois, College Agr., Special Publ. 3:31-34.

Funnell, K.A. and C.G. Downs. 1987. Effect of ethylene on spathe regreening in *Zantedeschia* hybrids. HortScience 22:1333.

Funnell, K.A. and A.R. Go. 1993. Tuber storage, floral induction, and gibberellin in *Zantedeschia*. ISHS, 2nd Intl. Symp. Development New Floricultural Crops, Baltimore, U.S.A., 1991. (in press).

Funnell, K.A. and B.R. MacKay. 1987. Effect of GA_3 on growth of *Zantedeschia*. Tech. Rpt. 87/14, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Palmerston North, Massey Univ., N.Z.

Funnell, K.A. and B.R. MacKay. 1988a. Identification of the active constituents of Promalin in optimising flowering in *Zantedeschia*. Tech. Rpt. 88/2, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Funnell, K.A. and B.R. MacKay. 1988b. Effect of temperature and ethylene during storage on growth and flowering of *Zantedeschia*. Tech. Rpt. 88/3, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Funnell, K.A. and B.R. MacKay. 1988c. Verification of in vitro curing regimes for *Zantedeschia*, using whole tubers. Tech. Rpt. 88/4, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Funnell, K.A. and B.R. MacKay. 1989a. Scooping of *Zantedeschia* tubers as a means of vegetative propagation. Tech.. Rpt. 88/5, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Funnell, K.A. and B.R. MacKay. 1989b. Investigations into the practice of early lifting of *Zantedeschia* tubers. Tech. Rpt. 88/6, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Funnell, K.A. and B.R. MacKay. 1990. Alleviation of storage induced flowering decline, in *Zantedeschia*, with Promalin. Tech. Rpt. 89/1, Dept. Hort. Sci., Massey Univ., Palmerston North, N.Z.

Funnell, K.A., B.R. MacKay and N.W. Comber. 1987. The effect of curing temperature, humidity and duration on the development of protective barriers in *Zantedeschia* tubers. Tech. Rpt. 87/7, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

Funnell, K.A., B.R. MacKay and C.R.O. Lawoko. 1992. Comparative effects of Promalin and GA_3 on flowering and development of *Zantedeschia* 'Galaxy'. ISHS, Hort. Sci. in the Tropics, Malaysia, 1990. Acta Hort. 292:173-179.

Funnell, K.A., B.O. Tjia, C.J. Stanley, D. Cohen and J.R. Sedcole. 1988. Effect of storage temperature, duration, and gibberellic acid on the flowering of *Zantedeschia elliottiana* and *Z.* 'Pink Satin'. J. Amer. Soc. Hort. Sci. 113(6):860-863.

Greene, L., R.B. Withrow and M.W. Richman. 1932. The response of greenhouse crops to electric light supplementing daylight. Purdue Univ. Agr. Expt. Sta. Bul. 366:1-20.

Gronegress, P. 1974. The structure of chromoplasts and their conversion to chloroplasts. J. Microscopie 19:183-192.

Harrison, R.E. 1972. *Zantedeschia* hybrids. J. Royal Hort. Soc. 97:131-132.

Hayward, W. 1948. The *Zantedeschia* (Calla) industry in Florida, p. 36-39. In: H.P. Traub and H.N. Moldenke (eds.). Plant Life, Vol. 4, Aroid Lily Edition. Amer. Plant Life Soc., Stanford.

Ingle, A. and W.T. Bussell. 1991. Tolerance of calla lilies to herbicides. Proceedings of the 44th Weed and Pest Control Conference, New Zealand, 44:138-142.

Jialong, Y. and D. Cohen. 1991. *Zantedeschia* breeding project. Ann. Rpt. 1990, N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z., p. 22-23.

Joubert, J.J. and S.J. Truter. 1972. A variety of *Xanthomonas campestris* pathogenic to *Zantedeschia aethiopica*. Neth. J. Plant Pathol. 78:212-217.

Kamerbeek, G.A. and W.J. De Munk. 1976. A review of ethylene effects in bulbous plants. Scientia Hort. 4:101-115.

Karlsson, M.G., R.D. Heins, J.E. Erwin, R.D. Berghage, W.H. Carlson and J. A. Biernbaum. 1989. Temperature and photosynthetic photon flux influence Chrysanthemum shoot development and flower initiation under short-day conditions. J. Amer. Soc. Hort. Sci. 114:158-163.

Kepner, K.W., T.E. Welsh and G.D. Steven. 1990. The future export potential for New Zealand calla flowers. Rpt., N.Z. Trade Dev. Board, Wellington, N.Z.

Kobayashi, T. 1977a. An examination of spraying gibberellic acid solution of differing strengths onto the stems and leaves of calla lily. Nagano Agr. Lab. Rpt. no. 21, Japan. (Japanese).

Kobayashi, T. 1977b. An examination of spraying gibberellic acid solution onto stems and leaves of calla lily. Nagano Agr. Lab. Rpt. no. 20, Japan. (Japanese).

Kobayashi, T. 1977c. A study of spraying gibberellic acid on to stems and leaves of calla lily at several times. Nagano Agr. Lab. Rpt. no. 23, Japan. (Japanese).

Kobayashi, T. 1980. Examination of temperature of calla tubers in gibberellic acid solution. Nagano Agr. Lab. Rpt. no. 95, Japan. (Japanese).

Kobayashi, T., T. Tsukaka and F. Otsuka. 1977. A study of the flower bud formation and the progression of growth in the golden calla lily (*Zantedeschia elliottiana* Engl.). Bull. Nagano Veg. Ornamental Crops Expt. Sta., Japan, 2:63-68. (Japanese).

Kobayashi, T., A. Tsukada, F. Otsuka and F. Yamanishi. 1978. Yellow calla (*Zantedeschia elliottiana* Engl.) Sun Light. Rpt. Hort. Assn., Nagano Prefecture, Japan, p. 57-58. (Japanese).

Korner, Ch. and W. Larcher. 1988. Plant life in cold climates, p. 25-58. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Letty, C. 1973. The genus *Zantedeschia*. Bothalia 11(1&2):5-26.

Long, P.G. 1988. New records of *Pythium* species in New Zealand. N.Z. J. Expt. Agr. 16:165-166.

Long, P.G., S. Miller, K. Everett and L. Fisher. 1988. Bacterial softrot diseases of *Zantedeschia*. Rpt. to Int. Calla Prod. Assn., Dept. Plant Health, Massey Univ., Palmerston North, N.Z.

MacKay, B.R., K.A. Funnell and H.K. Clark. 1987a. Nutrition of potted *Zantedeschia* 'Red Emperor'. Tech. Rpt. 87/3, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

MacKay, B.R., K.A. Funnell and N.W. Comber. 1987b. The effect of shade and growing environment temperature on the growth and flower pigmentation development of *Zantedeschia* hybrids. Tech. Rpt. 87/4, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

MacKay, B.R., K.A. Funnell, N.W. Comber and K.A. Bowers. 1986. The effects of PP333 concentrations and application methods on clones of *Zantedeschia* hybrids. Tech. Rpt. 87/2, Dept. Hort. and Plant Health, & N.Z. Nursery Research Centre, Massey Univ., Palmerston North, N.Z.

MacKay, B.R., K.A. Funnell and T.C.H. Tan. 1991. Evaluation of the performance of export grade *Zantedeschia* cultivars. Tech. Rpt. 91/1, Dept. Hort. Sci., Massey Univ., Palmerston North, N.Z.

Okonkwo, C.A.C. 1987. Polymorphism in taro (*Colocasia esculenta* (L.) Schott) and its implication in taro breeding and production in Nigeria. J. Hort. Sci. 62:395-400.

Pais, M.S. and H.J. Chaves das Neves. 1982/83. Regreening of *Zantedeschia aethiopica* Spreng. spathe induced by reapplied cytokinins. Plant Growth Regulat. 1:233-242.

Perry, P.L. 1989. A new species of *Zantedeschia* (Araceae) from the western Cape. South African J. Bot. 55:447-451.

Plummer, J.A. 1990. The influence of rhizome storage and growth regulators on flower production in potted *Zantedeschia aethiopica* 'Childsiana'. Tech. Rpt. Dept. Hort. Sci., Massey Univ., Palmerston North, N.Z.

Plummer, J.A., T.E. Welsh and A.M. Armitage. 1990. Stages of flower development and postproduction longevity of potted *Zantedeschia aethiopica* 'Childsiana'. HortScience 25:675-676.

Post, K. 1936. Further responses of miscellaneous plants to temperature. Proc. Amer. Soc. Hort. Sci. 34:627-629.

Rana, G.L., C. Vovlas and F.W. Zettler. 1983. Manual transmission of dasheen mosaic virus from Richardia to non-araceous hosts. Plant Dis. Rptr. 67:1121-1122.

Rees, A.R. 1972. The Growth of Bulbs. Academic Press, London.

Reiser, R.A. 1990. Cultivation of *Zantedeschia* species for potted plant production. M.S. thesis, Cornell, U.S.A.

Reiser, R.A. and R.W. Langhans. 1993. Cultivation of *Zantedeschia* species for potted plant production. ISHS, 2nd Intl. Symp. Development New Floricultural Crops, Baltimore, U.S.A., 1991. (in press).

Rong, L., Wang, X. Qin, Wang, R. Zhen and Y. Jun. 1989. Tissue culture and rapid propagation of *Zantedeschia* hybrid. Guihaia 9:97-102.

Sakanishi, Y. 1955. Effects of various treatments during the summer months on growth and flower production of common calla (*Zantedeschia aethiopica* var. Minor). Inst. Floriculture, Dept. Agr., Univ. Osaka Prefecture 24(3):63-68.

Sprengel, C. 1826. Systema Vegetabilium 3:765. (Cited by Letty, 1973).

Shibuya, R. 1956. Intercrossing among pink calla, white-spotted calla and yellow calla. Monograph. Kasai Publ., Tokyo.

Takahashi, J., M. Kosugi and M. Yokoi. 1957. Flower habit and flower bud progression of calla (*Zantedeschia aethiopica* var. Childsiana). Hort. Assn. Japan 14:288-289. (Japanese).

Tjia, B. 1986. Keeping quality of *Zantedeschia*. Commercial Horticulture, N.Z., Feb. 1986, p. 20-21.

Tjia, B. 1987a. Calla lily tubers thrive in higher temperatures. Greenhouse Manager, July 1987, p. 106,109,110,112.

Tjia, B. 1987b. The acid test. Greenhouse Manager, March 1987, p. 128-134.

Tjia, B. 1987c. Growth regulator effect on growth and flowering of *Zantedeschia rehmannii* hyb. HortScience 22:507-508.

Tjia, B. and K.A. Funnell. 1985. Control of stem split and alternaria flower spot following harvest. Commercial Horticulture, N.Z., April 1985, p. 28-29.

Tjia, B.O. and K.A. Funnell. 1986. Postharvest studies of cut *Zantedeschia* inflorescences. Acta Hort. 181:451-458.

Traub, H.P. 1948. The genus *Zantedeschia.* Plant Life 4:9-32.

Warrington, I.J. and R.C. Southward. 1989. Influence of temperature and light intensity on *Zantedeschia* growth and development. Commercial Rpt. No. 46, Plant Physiol. Div., DSIR, Palmerston North, N.Z.

Welsh, T.E., J.A. Plummer and A.M. Armitage. 1988. Preliminary evaluation of the dwarf white calla lily as a potted plant. Proc. Int. Plant Prop. Soc. 38:384-388.

Woodward, F.I. 1988. Temperature and the distribution of plant species, p. 59-75. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Zizzo, G.V. 1987. Influenza del peso dei rizomi di calla (*Zantedeschia aethiopica* Spreng) sulla produzione dei fiori, rizomi e tubercoli. [Influence of the weight of rhizomes of arum lily (*Zantedeschia aethiopica* Spreng.) on production of flowers, rhizomes and nodules]. Annali dell'Istituto Sperimentale per la Floricoltura 18:117-125. (Italian).

2 INTRODUCTORY OVERVIEW AND AIM OF THE CURRENT STUDY

2.1 Overview of study

With 90% or more of plant dry matter comprising carbon (Irvine, 1975), it is self-evident that the processes of carbon acquisition (i.e., photosynthesis) and utilization (i.e., dry matter accumulation and respiration) should be a principal focus of any investigation into plant growth. Temperature and photosynthetic photon flux (PPF) are just two of the environmental parameters influencing these processes of acquisition and utilization.

The responses of plants to temperature are considered to be governed by both the direct effects of temperature on metabolic (i.e., enzyme and membrane carrier) activity and indirect physical (i.e., diffusion and viscosity) effects (Farrar and Williams, 1991). In addition to any indirect effects of PPF on photosynthesis through stomatal aperture (Dwelle et al., 1981; Valenzuela et al., 1990), as a source of energy used in the acquisition and utilization of carbon, the interception of PPF is an integral component of dry matter accumulation (Warrington and Norton, 1991).

The importance of photosynthesis as a determinant of dry matter accumulation and how it is influenced by temperature and PPF, will be presented in Chapter 5. Consideration of total plant dry matter can provide a quantitative description of the plants' response to temperature and PPF (refer Chapter 3). Since leaves are the primary site of carbon acquisition and the tuber is the organ associated with harvestable yield, the partitioning of dry matter between organs (refer Chapters 3, 4 and 7) not only provides quantitative description, but environment induced changes in this partitioning may be used to suggest possible control mechanisms (Konings, 1990; Poorter, 1990). However development of a greater understanding of the potential mechanisms of control, requires investigation into the various biochemical components of the entire plant and its various organs (Warren Wilson, 1972; Obata-Sasamoto and Suzuki, 1979). Being the primary products of photosynthesis, carbohydrates potentially provide a major biochemical component of dry weight with which to begin examining the mechanisms of partitioning (refer Chapter 6).

Commercial production of *Zantedeschia* in New Zealand primarily utilizes group 2 selections (refer Section 1.2). Flower and tuber producers in New Zealand were therefore more likely to benefit from adopting the research findings resulting from investigations into selections within this group. The *Zantedeschia pentlandii*-like selection 'Best Gold' was chosen as the subject of this study for a number of reasons.

(1) Its classification as a group 2 selection.
(2) Its current use as a commercial cut flower crop.
(3) Local availability of large quantities of disease free plant material of an even grade and at no financial cost.

If, like other genera, the natural distribution of *Zantedeschia* species has been influenced by the temperature minima during both the growing season and/or annual period (Korner and Larcher, 1988; Woodward, 1988), it would be expected that climatological data of the sites of origin (refer Section 1.3.2) would provide some indication of the possible temperature tolerance range, and therefore those temperatures worthy of investigation. Similarly, the natural habitat of *Zantedeschia* species in open grassland or forest margins (refer Section 1.3.2) might be indicative of an ability to tolerate shade. Using the data collated in Section 1.3.2, treatments in the current study were selected to reflect such a range of possible temperatures and PPF.

2.2 Aim of this study

International demand exists for *Zantedeschia* tubers year-round. However, at present the natural supply of quality tubers from New Zealand is restricted to a few months of the year. Although it is known that tubers can be produced out of season, the optimal environmental conditions to ensure the induction and maintenance of rapid tuber growth have not been defined. While preliminary studies have been conducted on the environmental response of vegetative growth and development of *Zantedeschia* (refer Section 1.5.3), no detailed analysis has been conducted that would enable possible control mechanisms to be elucidated. Further, gaining an understanding of how the plant responds to these environmental parameters is the foundation of information required to subsequently develop crop management strategies, crop models, and future breeding strategies. The aim of this study therefore, was to describe and interpret the vegetative growth and development of a group 2 selection in response to the environmental parameters of temperature and PPF.

2.3 References

Dwelle, R.B., G.E. Kleinkopf and J.J. Pavek. 1981. Stomatal conductance and gross photosynthesis of potato (*Solanum tuberosum* L.) as influenced by irradiance, temperature, and growth stage. Potato Res. 24:49-59.

Farrar, J.F. and M.L. Williams. 1991. The effects of increased atmospheric carbon dioxide and temperature on carbon partitioning, source-sink relations and respiration. Plant, Cell and Environ. 14:819-830.

Irvine, J.E. 1975. Relations of photosynthetic rates and leaf and canopy characteristics to sugar cane yield. Crop Sci. 15:671-676.

Konings, H. 1990. Physiological and morphological differences between plants with a high NAR or a high LAR as related to environmental conditions, p.101-124. In: H. Lambers, M.L. Cambridge, H. Konings, and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing bv., The Hague.

Korner, C. and W. Larcher. 1988. Plant life in cold climates, p. 25-58. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Obata-Sasamoto, H. and H. Suzuki. 1979. Activities of enzymes relating to starch synthesis and endogenous levels of growth regulators in potato stolon tips during tuberization. Physiol. Plant. 45:320-324.

Poorter, H. 1990. Interspecific variation in relative growth rate: on ecological causes and physiological consequences, p.45-68. In: H. Lambers, M.L. Cambridge, H. Konings, and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing bv., The Hague.

Valenzuela, H.R., B. Schaffer and S.K. O'Hair. 1990. Shade and nitrogen influence gas exchange and growth of Cocoyam (*Xanthosoma sagittifolium*). J. Amer. Soc. Hort. Sci. 115:1014-1018.

Warren Wilson, J. 1972. Control of crop processes, p. 7-30. In: A.R. Rees, K.E. Cockshull, D.W. Hand, and R.G. Hurd (eds.). Crop processes in controlled environments. Academic Press, London.

Warrington, I.J. and R.A. Norton. 1991. An evaluation of plant growth and development under various daily quantum integrals. J. Amer. Soc. Hort. Sci. 116:544-551.

Woodward, F.I. 1988. Temperature and the distribution of plant species, p. 59-75. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

3 DRY MATTER ACCUMULATION AND LEAF LAMINA DEVELOPMENT OF *Zantedeschia* 'Best Gold' IN RESPONSE TO TEMPERATURE AND PHOTOSYNTHETIC PHOTON FLUX

3.1 Introduction

Zantedeschia is a relatively new horticultural crop with an expanding world-wide market for both cut flowers and tubers (refer Section 1.2). While complications with endodormancy have restricted the commercial development of all year round cultivation of *Zantedeschia*, it is known that under protected cultivation non-endodormant tubers of *Zantedeschia* can be forced into growth at any time of the year (Funnell and MacKay, 1990). However, the response of this genus to manipulation of environmental parameters such as temperature and photosynthetic photon flux (PPF) during the growing season have not been previously described. For the commercial horticulturist the relatively large diurnal and seasonal variation of these parameters results in the need to investigate what impact they will have on growth. Further, gaining an understanding of how the plant responds to these environmental parameters is the foundation of information required to subsequently develop crop management strategies, crop models, and future breeding strategies.

Traditional plant growth analysis provides an empirical description of growth and development of the plant through the derivation of *"derived quantities"* from *"measured quantities"* (Warren Wilson et al., 1986). Equations relating these derived quantities present the relative growth rate (RGR) as a product of two components (e.g., equation (3.1); West et al., 1920).

$$RGR = LAR \times NAR \tag{3.1}$$

where;

RGR = relative growth rate
LAR = leaf area ratio
NAR = net assimilation rate

The first component is a measure of size, and the second a measure of the efficiency of dry-matter production in terms of a resource of assimilates (Warren Wilson et al., 1986). Hunt (1982) summarised this simplistic but fundamental equation describing the growth of plants as *"the growth rate of the plant depends simultaneously upon the efficiency of its leaves as producers of new material and upon the leafiness of the plant itself"*. Therefore, this empirical description also has the beginnings of being a mechanistic model, as the relative importance of the various components may be assessed.

While early research utilizing growth analysis endeavoured to illustrate a causal relationship between RGR and NAR, because of a strong negative correlation between NAR and LAR (Konings, 1990; Poorter, 1990), many examples have been reported where this correlation is weak (Heath and Gregory, 1938; Watson, 1952; Lambers, 1987; Tollenaar, 1989b; Poorter, 1990). Hence, while NAR is recognised as an important component of RGR, its direct contribution is frequently masked by concomitant changes in LAR. Such changes in LAR may result from changes in leaf thickness (i.e., specific leaf area) and/or the proportion of total plant dry matter within the leaf (i.e., leaf weight ratio) (Konings, 1990).

In contrast to the poor correlation between plant growth and NAR, for a large number of species strong correlations between RGR and the various forms of expressing the components of LAR (equation (3.2)) have been reported (Poorter, 1990).

$$LAR = SLA \times LWR \tag{3.2}$$

where;

LAR = leaf area ratio
SLA = specific leaf area
LWR = leaf weight ratio

For a large number of species, grown under a range of environments, this strong correlation between RGR and the partitioning of dry matter to leaf area has also been expressed in terms of relative rates of leaf area expansion (RLAER) (Watson, 1947a & 1947b; El-Sharkawy et al., 1965; Muramoto et al., 1965; Duncan and Hesketh, 1968; Slatyer, 1970; Hanson, 1971; Delaney and Dobrenz, 1974; Potter and Jones, 1977; Collins and Jones, 1988). However, it should be appreciated that this correlation is not necessarily causal. Even though Jackson (1963) illustrated a clear correlation between RGR and the instantaneous ratio in the rates of change with time of leaf area and total plant dry weight (LAP, equation (3.3)), it was not until Potter and Jones (1977) highlighted this relationship, that the greater potentially-causal relationship was appreciated.

$$LAP = \frac{\delta A/\delta t}{\delta W/\delta t} \tag{3.3}$$

where;

LAP = leaf area partitioning
W = total plant dry weight
A = total plant leaf area
t = time
δ = mathematical notation for an interval

Potter and Jones (1977) suggested that since the daily partitioning of growth into leaf area will determine the area on the next day, the daily growth on the next day will depend to a large extent on the value of LAP. Patterson et al. (1978) extended our understanding of LAP by illustrating that as the ratio of RGR and RLAER approaches unity, then LAP approaches LAR. The value of LAP as a component of growth analysis is that it takes into account differences in RGR and RLAER when they occur. The strong correlation between RGR and LAP has been illustrated using a number of species grown under a range of temperatures and PPF regimes (Potter and Jones, 1977; Patterson et al., 1978; Hunt and Halligan, 1981; Tollenaar, 1989a).

Over the range of annual temperatures prevailing over much of the earth's inhabited surface (0 to 30 C), and for a diversity of plant species, the rate of development increases linearly with increasing temperature (Grace, 1988). In contrast, the response of RGR and final yield to this temperature range has generally been shown to be parabolic (Rajan et al., 1973; Porter and Delecolle, 1988), with the optimum temperature of this parabolic response being species dependent (Rajan et al., 1973). While increased temperatures have been associated with increased partitioning of dry matter towards leaf growth and therefore increasing the LAR and LAP (Farrar, 1988) over the naturally occurring range of PPF (0 to 2000 $\mu mol \cdot m^{-2} \cdot s^{-1}$), increased PPF typically is associated with increased dry matter accumulation, but with reduced LAR and LAP (Konings, 1990; Poorter, 1990). In addition, both increased temperature and PPF have been associated with increased NAR (Konings, 1990; Poorter, 1990).

The objective of this study was to use the technique of plant growth analysis to gain an understanding of how growth of *Zantedeschia* 'Best Gold' responds to variable temperature and PPF during development, and, in doing so, define horticulturally-relevant environmental parameters and management strategies.

3.2 Materials and Methods

3.2.1 Cultural

Non-endodormant tubers of the *Zantedeschia pentlandii*-like selection 'Best Gold', were lifted from the field (Palmerston North, New Zealand; 40°20'S) immediately prior to two plantings; one in early August 1988 and the other in late July 1989. On both occasions tubers were graded for evenness of size (approx. 1 cm diameter, 0.4 g dry weight), and for the presence of a single dominant bud. Tubers were planted in 60 ml cell trays, containing a 50 peat : 30 pumice (v/v) growing medium to which 3 $kg \cdot m^{-3}$ dolomite lime was added. Plants were subsequently grown for 18 days on a heated (20 C), closed, mist bench, under greenhouse conditions (minimum air temperature 15 C, ventilation at 24 C). A uniform grade of emerged plants were selected, on the basis of development of protective sheath leaves (1 cm in length), potted into 1.2 litre pots containing the same growing medium, and transferred to treatment environments. The experiment was carried out in controlled environment (CE) facilities in the National Climate Laboratory at the Horticulture and Food Research Institute of N.Z. Ltd, at Palmerston North, New Zealand. Over the two year period, plants were randomly allocated to one of 12 temperature-PPF treatment combinations, with two of the 12 combinations being employed at both plantings. Each environmental treatment comprised 96 plants with each pot supplied with a complete nutrient solution (5 × 100 ml daily, half-strength Hoagland's A modified with the use of chelated iron, type ONC; Brooking (1976)). The amounts used ensured drainage at each application and avoided any outward symptoms of plant moisture stress. The avoidance of moisture stress was also validated through the measurement of water potential of leaves during destructive harvests using a pressure bomb apparatus. Throughout the experiment an open plant canopy was maintained by the regular sampling of individual plants, eliminating interplant shading effects.

3.2.2 Environmental

The day/night temperature treatments were: 16/10, 22/10, 22/16, 28/16, 28/22 and 28/28 C ± 0.5 C. These day/night regimes resulted in the following daily mean temperatures: 13, 16, 19, 22, 25 and 28 C. In all temperature treatments, the day/night vapour pressure deficit was maintained at 1.0/0.4 kPa. Photoperiod was 12 h and day/night and night/day temperature and vapour pressure changeovers were each of 2 h duration, with the lights switching off and on at the midpoint of each respective changeover.

Lighting was provided by four 1000 Watt high-pressure multivapour-lamps (Sylvania "Metalarc") and four 1000 Watt Philips quartz halogen lamps, separated from the plant

growth room by a plate glass, and water, thermal barrier. The PPF at pot surface height was 694 ± 20 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (high PPF). Within the same CE room a PPF of 348 ± 10 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (low PPF) was achieved for half the plants, by covering four of the eight trolleys per room with a spectrally-neutral woven polypropylene shade cloth of nominal 50% density. At the same time as relocating the position of trolleys within each room at weekly intervals, trolleys were rotated 180° about their own axis. This procedure was carried out to minimise any influence of environmental gradients within each room. Additional information on room and lighting design has been described (Warrington et al., 1978).

3.2.3 Experimental

To monitor growth, 6 or 12 plants were destructively harvested from the time of planting in cell trays until growth rate declined. Once in the CE rooms harvests occurred at the appearance of each successive leaf. This was taken to be when a minimum of 75% expansion of that leaf had occurred on 90% of plants within a treatment. Once increases in tuber dry weight were detected, sampling occurred more frequently, until the rate of total plant dry weight accumulation declined.

At each harvest the following were recorded: number of sheath leaves (i.e., protective structures which envelop the shoot, but do not produce leaf blades), number and area of exposed leaves (i.e., leaf blades not enveloped by subtending petioles), and number of leaves commencing senescence (i.e., when a minimum of 5 to 10% of the leaf had senesced). Senescence was defined as that stage when leaf colour had progressed to the Judd-Hunter colorimetric $L^*a^*b^*$ values (Francis and Clydesdale, 1975) of $L^* = 75 \pm 4$, $a^* = 5 \pm 1$, and $b^* = 50 \pm 5$. Leaf area data (i.e., leaf blade excluding petiole) were recorded using a LICOR model 3100 leaf area meter. All measurements were carried out within 4 h of removal from treatment, after which plants were vacuum dried at 0.3 kPa and 40 C for a minimum of 48 h and the following dry-weights recorded: exposed leaves, shoot (i.e., petioles of exposed leaves, together with the apex, non-exposed leaves, and sheath leaves), petioles of senescing exposed leaves, senescing exposed leaf blades, tuber, and roots (Plate 3.1).

Within each temperature-PPF treatment, plants were arranged in a completely randomized design. The number of treatments included in this study, and the time required to complete each treatment, precluded any possibility of totally replicating the experiment. To partially evaluate the variation due to time and to between CE room effects, two temperature-PPF combinations (22/16 C high PPF and 22/16 C low PPF) were employed in different CE rooms at the two planting times.

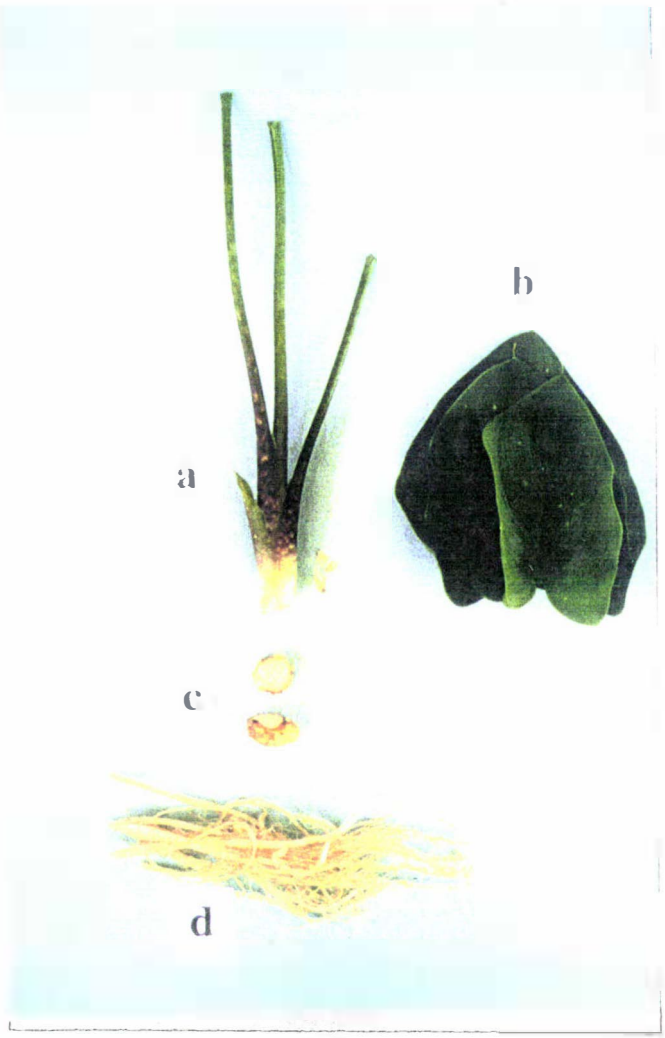

Plate 3.1 Harvested plant of *Zantedeschia* 'Best Gold' illustrating components measured. (a) shoot (sheath leaves, petioles and apex), (b) exposed leaves, (c) tuber, (d) roots. (N.B., scale half actual size)

A $\log_e$ transformation of the dry weight data was used to meet the statistical assumption of homogeneity of variance throughout the period of study (Causton and Venus, 1981). Log_e transformed total plant dry weight data, collected subsequent to placement in the CE rooms, were fitted to the Gompertz function, equation (3.4) (Causton and Venus, 1981), utilizing the non-linear regression parameter estimation procedure of the Statistical Analysis System (SAS; SAS Institute, Inc., Cary, N.C.). A constant was added to all transformed data to eliminate negative values from the fitting process. Because the Gompertz function is asymptotic, the magnitude of the added constant influences the calculated value of β_W. Hence the addition of a constant to all values prior to curve fitting eliminated any value of subsequent interpretation of differences in the value of β_W (i.e., a measure of the starting size of total plant dry weight). The Gompertz function was chosen over other sigmoidal growth functions due to the derivation of more biologically relevant parameters, its wide acceptability by other researchers (Causton and Venus, 1981; Hunt, 1982), and its ability to fit the current data-set with small mean square error values. In addition, with data from

some treatments, preliminary attempts to apply the more flexible Richard's function indicated over-parameterization of the model (Causton and Venus, 1981). This over-parameterization may have resulted from an inadequate number of harvest dates near critical points of the curve, such as near the upper asymptote.

$$\log_e W = \alpha_W \cdot e^{(-e^{(\beta_W - \kappa_W \cdot t)})} \qquad (3.4)$$

where;

$\log_e W$ = $\log_e$ transformed value of total plant dry weight ($\log_e$ g)
α_W = upper asymptote of total plant dry weight ($\log_e$ g)
β_W = a measure of the starting size of total plant dry weight ($\log_e$ g)
κ_W = rate constant of total dry weight as a function of size ($g \cdot g^{-1} \cdot g^{-1}$)
t = time (days)
e = the base of natural logarithms

Log_e transformed total plant leaf area, and leaf dry weight data, collected subsequent to placement in the CE rooms, were fitted to the logarithmic form of the Gompertz function, equations (3.5) and (3.6) respectively (Causton and Venus, 1981), utilizing the non-linear regression parameter estimation procedure of SAS.

$$\log_e A = \alpha_A - e^{(\beta_A - \kappa_A \cdot t)} \qquad (3.5)$$

$$\log_e L = \alpha_L - e^{(\beta_L - \kappa_L \cdot t)} \qquad (3.6)$$

where;

$\log_e A$, $\log_e L$ = $\log_e$ transformed value of total plant leaf area and leaf weight, respectively ($\log_e$ cm^2 and $\log_e$ g, respectively)
α_A, α_L = asymptote of attribute under investigation ($\log_e$ cm^2 and $\log_e$ g, respectively)
β_A, β_L = a measure of the starting size of attribute under investigation ($\log_e$ cm^2 and $\log_e$ g, respectively)
κ_A, κ_L = rate constant of attribute as a function of size ($cm^2 \cdot cm^{-2} \cdot cm^{-2}$ and $g \cdot g^{-1} \cdot g^{-1}$, respectively)
t = time (days)
e = the base of natural logarithms

During this time period total plant relative growth rate (RGR_W), as a function of time, was calculated using equation (3.7), being the first differential of equation (3.4) (Causton and Venus, 1981).

$$RGR_W = \alpha_W \cdot \kappa_W \cdot e^{(\beta_W - \kappa_W \cdot t)} \cdot e^{(-e^{(\beta_W - \kappa_W \cdot t)})} \qquad (3.7)$$

Similarly, total plant relative leaf area expansion rate (RLAER), and relative leaf weight rate (RLWR), as functions of time, were calculated using equations (3.8) and (3.9), being the first differentials of equations (3.5) and (3.6), respectively (Causton and Venus, 1981).

$$RLAER = \kappa_A \cdot e^{(\beta_A - \kappa_A \cdot t)} \qquad (3.8)$$

$$RLWR = \kappa_L \cdot e^{(\beta_L - \kappa_L \cdot t)} \qquad (3.9)$$

Net assimilation rates (NAR), as defined by West et al. (1920), equation (3.10), were calculated using equation (3.11), utilizing derived formulae and data from equations (3.4), (3.5) and (3.7).

$$NAR = \frac{\delta W}{\delta t} \times \frac{1}{A} \qquad (3.10)$$

$$NAR = \frac{RGR_{W(t)} \cdot e^{(\log_e W_{(t)})}}{e^{(\log_e A_{(t)})}} \qquad (3.11)$$

Leaf area partitioning (LAP) and leaf weight partitioning (LWP), as defined by Jackson (1963), equations (3.3) and (3.12), were calculated using equations (3.13) and (3.14), utilizing derived formulae and data from equations (3.4) to (3.9), respectively.

$$LWP = \frac{\delta L / \delta t}{\delta W / \delta t} \qquad (3.12)$$

$$LAP = \frac{RLAER_{(t)} \cdot e^{(\log_e A_{(t)})}}{RGR_{W(t)} \cdot e^{(\log_e W_{(t)})}} \qquad (3.13)$$

$$LWP = \frac{RLWR_{(t)} \cdot e^{(\log_e L_{(t)})}}{RGR_{W(t)} \cdot e^{(\log_e W_{(t)})}} \qquad (3.14)$$

The value of t used in the calculation of NAR, LAP, and LWP was the time (days from planting) at the midpoint of exponential total plant dry weight accumulation.

All dry weight data were expressed on the basis of daily mean temperature. When linear regression of parameters derived from the above equations, (3.4) to (3.14), against temperature, was considered inappropriate, relationships were derived by fitting the hyperbolic tangent function, equation (3.15), utilizing the non-linear regression parameter estimation procedure of SAS. This function has been illustrated to be more biologically meaningful than the frequently used rectangular hyperbola (Jassby and Platt, 1976).

$$y = y_{max} \cdot \tanh\left(\frac{\alpha \cdot C}{y_{max}}\right) - y_{int} \qquad (3.15)$$

where;

y_{max} = maximum value of attribute under investigation
tanh = hyperbolic tangent
α = initial slope of line
C = mean temperature in degrees Celsius
y_{int} = value of y at intercept of y axis

For each PPF regime determination of the relationship between RGR_W and temperature was established by regression analysis using the REG procedure of SAS. Comparisons of the derived slopes and intercepts between PPF treatments for homogeneity were conducted as outlined by Zar (1984).

3.3 Results

3.3.1 Overview and initial establishment

During the period that plants were in cell trays, and prior to transfer to treatments (i.e., first 18 days), total plant dry weight declined (Figure 3.1). This period coincided with the establishment of roots, and development of the shoot to approximately 1 cm in length, but with no leaf lamina exposed.

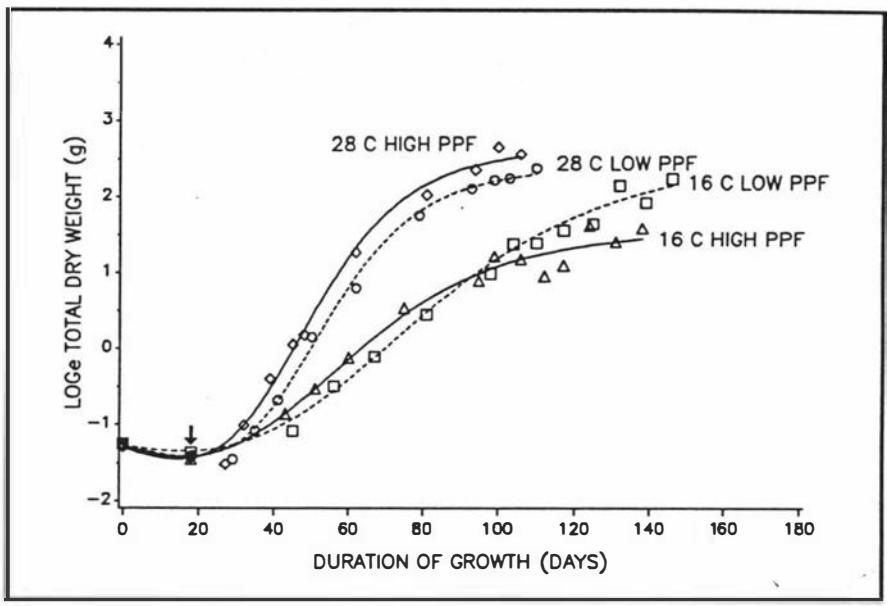

Figure 3.1 Examples of total plant dry weight ($\log_e$) as a function of time, for *Zantedeschia* 'Best Gold'. ↓ indicates day of transfer to treatments.

However, at the time of the first harvest after placement within the treatments, total plant dry weight had either stabilized or begun to increase. This period coincided with the first leaf attaining 75% or more of its complete expansion. Increases in temperature reduced the time to attainment of this stage of development, but the harvest frequency was not able to detect any significant differences between PPF regimes (Table 3.1, Figure 3.1). At this time of establishment of the first leaf, a curvilinear relationship existed between total plant dry weight (W) and increasing temperature (C) ($P \leq 0.05$, equation (3.16)).

$$W = -0.002 \cdot C^2 + 0.08 \cdot C - 0.02 \qquad (r^2 = 0.49,^*) \qquad (3.16)$$

Solving this equation, total plant dry weight was greater at 19 C than at both higher and lower temperatures. This difference in total plant dry weight, at first leaf, was most noticeable between 19 C and 28 C, between which total plant dry weight declined linearly at a rate of 0.021 g ($\pm$ 0.007) for each degree Celsius increase in temperature ($P \leq 0.05$).

This resulted in total plant dry weight at 19 C (0.55 g ± 0.04) being approximately 1.5 times that attained at 28 C (0.36 g ± 0.02). At the time of first leaf, there were no differences in total plant dry weight between PPF regimes, except at 25 C. Leaf area and leaf dry weight of this first leaf responded similarly to temperature and PPF regime as did total plant dry weight (data not presented).

Table 3.1

Duration of growth (days) until attainment of 75% or more expansion of the first leaf of *Zantedeschia* 'Best Gold' at a range of daily temperatures, and high and low PPF regimes.

	Mean temperature (C)					
PPF	13	16	19	22	25	28
			(days)			
High	58	43	35	30	30	25
Low	58	45	37	33	32	27

During the initial stages of growth, the relative rate of dry weight accumulation (RGR_w) was higher with higher temperature and PPF (Figure 3.2). However, after the point of inflection of the original Gompertz function, while higher temperatures resulted in higher values of RGR_w, values were higher under the low PPF regimes than under the high PPF regimes.

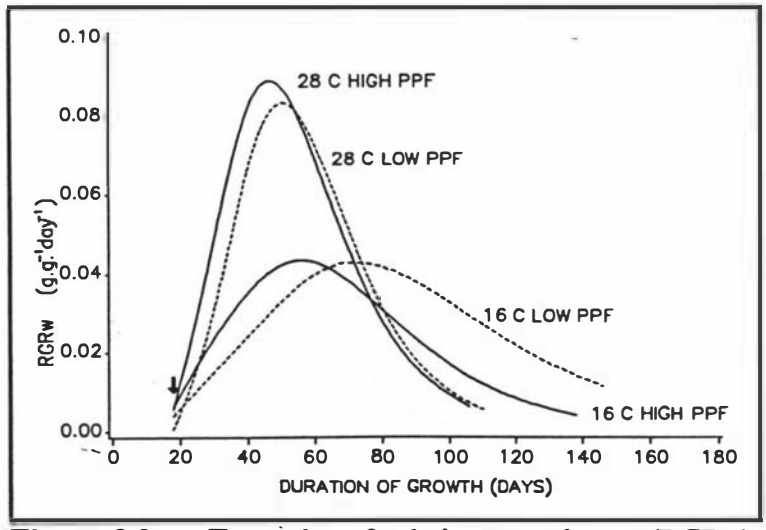

Figure 3.2 Examples of relative growth rate (RGR_w) as a function of time, for *Zantedeschia* 'Best Gold'.
↓ indicates day of transfer to treatments.

At the time RGR_w was maximum, i.e., at the inflection point of the fitted total plant dry weight Gompertz curve, a positive linear relationship between RGR_w and temperature was determined under both high ($P \leq 0.001$, equation (3.17)) and low ($P \leq 0.001$, equation (3.18)) PPF regimes (Figure 3.3). Comparison of the slopes for homogeneity did not detect differences between PPF regimes ($P \leq 0.05$), but RGR_w was consistently greater under the high PPF regime than under the low PPF regime, as indicated by differences between the values of the intercepts ($P \leq 0.05$). Hence at this stage of development an approximate doubling of the temperature from 13 C to 28 C resulted in an approximate doubling in the value of RGR_w, regardless of PPF regime. The linear response of RGR_w with temperature allowed extrapolation of the linear regression to predict a base temperature for growth of 0.8 $\pm$ 4 C under the high PPF regime, and 3.4 $\pm$ 4.2 C under the low PPF regime. Pooling the data from both PPF regimes resulted in an estimated base temperature for total plant growth of 2.1 $\pm$ 2.7 C.

$$\text{(High PPF)} \quad RGR_w = 0.0033 \cdot C - 0.003 \qquad (r^2 = 0.96,^{***}) \quad \textbf{(3.17)}$$

$$\text{(Low PPF)} \quad RGR_w = 0.0034 \cdot C - 0.01 \qquad (r^2 = 0.90,^{***}) \quad \textbf{(3.18)}$$

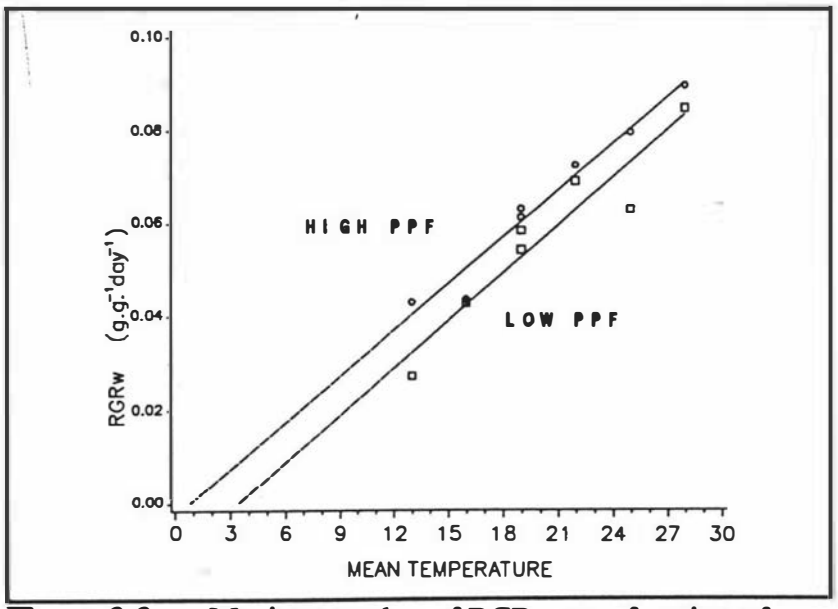

Figure 3.3 Maximum value of RGR_w as a function of temperature, for plants of *Zantedeschia* 'Best Gold' grown under high -○- and low -□- PPF regimes.

3.3.2 Repetition of treatments over years

Comparison of relative growth rates (RGR_w) is recognised as providing *"a more informative comparison of the plants' relative performances"* (Hunt, 1982). For the purpose of comparison of treatments over the two years in two different CE rooms (i.e., 22/16 C day/night, at both high and low PPF), values of RGR_w were calculated as the slope of the line of $\log_e$ transformed data (Hunt, 1982), during the period of exponential growth (i.e., between 40 to 90 days from planting, Table 3.2). Examination of the slopes of the linear regressions for homogeneity did not detect differences between years ($P \leq 0.05$). In light of this finding it was assumed that CE room, and time, effects were small. Subsequent interpretation of data therefore utilized the mean value of data collected from both years for these treatments.

Table 3.2

Total plant relative growth rates ($RGR_w \pm$ standard error), and associated r^2, for plants of *Zantedeschia* 'Best Gold' grown at day/night temperatures of 22/16 C, at high and low PPF, in two CE rooms over two years.

	PPF regime			
	High		Low	
Year	$RGR_w \pm$ s.e.	r^2	$RGR_w \pm$ s.e.	r^2
1st	0.047 ± 0.002	0.89	0.050 ± 0.002	0.93
2nd	0.050 ± 0.002	0.89	0.053 ± 0.003	0.89
signif.#	n.s.		n.s.	

n.s. = years not significantly different ($P < 0.05$) (Zar, 1984).

3.3.3 Curve fitting of total plant dry weight

Under all treatment regimes total plant dry weight followed a sigmoidal pattern of growth, with a family of Gompertz functions adequately describing the progression of total plant dry weight ($\log_e$ transformed) subsequent to placement in the treatments (Figure 3.4 and Figure 3.5, Table 3.3).

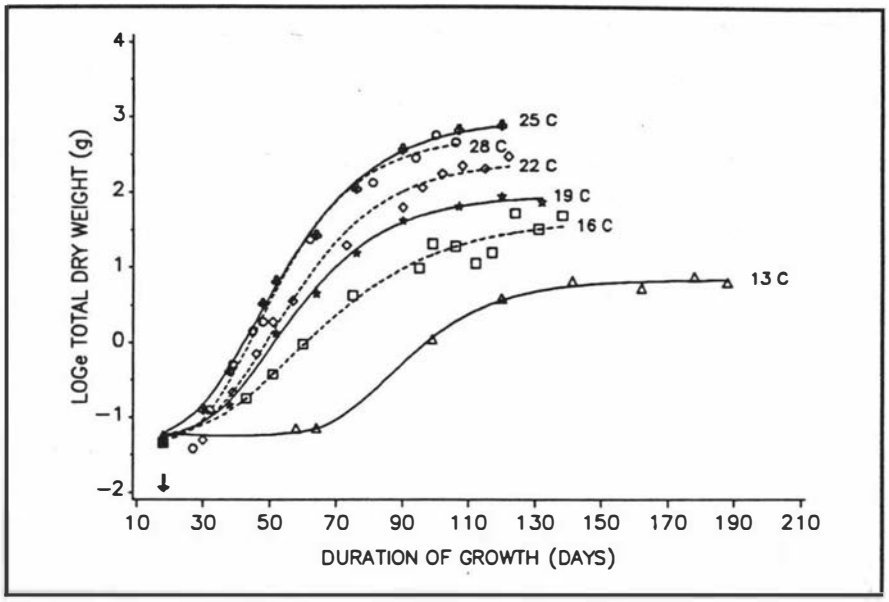

Figure 3.4 Total plant dry weight (log$_e$ fitted Gompertz curves) for *Zantedeschia* 'Best Gold' at a range of temperatures, under a high PPF regime. ↓ indicates day of transfer to treatments.

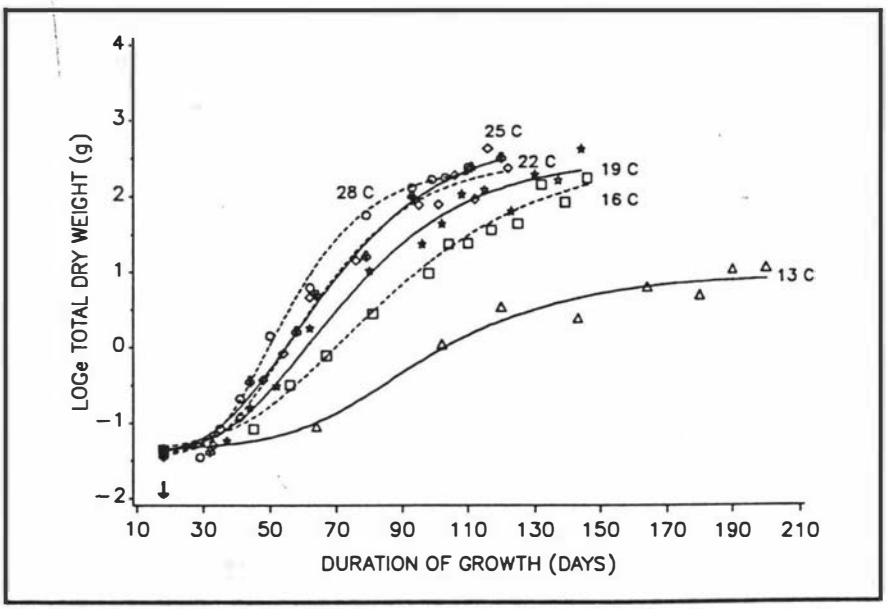

Figure 3.5 Total plant dry weight (log$_e$ fitted Gompertz curves) for *Zantedeschia* 'Best Gold' at a range of temperatures, under a low PPF regime. ↓ indicates day of transfer to treatments.

Table 3.3

Nonlinear least-squares parameter estimates, associated asymptotic standard error (s.e.), and mean square error values, from fitting the Gompertz function to $\log_e$ transformed total plant dry weight data for *Zantedeschia* 'Best Gold' grown at a range of temperatures, and under high and low PPF regimes. The PPF treatments at 19 C were repeated in two plantings, i.e., (1st) and (2nd).

Mean temperature (C)	PPF	α_w'	s.e.	β_w	s.e.	κ_w	s.e.	mean square error
13	High	2.06	0.08	4.88	1.33	0.057	0.014	0.20
	Low	2.33	0.13	2.69	0.69	0.032	0.008	0.37
16	High	2.98	0.11	2.22	0.24	0.040	0.005	0.10
	Low	3.90	0.24	2.15	0.21	0.030	0.004	0.15
19 (1st)	High	3.25	0.07	2.64	0.22	0.053	0.004	0.09
	Low	3.80	0.09	2.40	0.17	0.042	0.003	0.08
19 (2nd)	High	2.88	0.07	3.05	0.31	0.058	0.007	0.12
	Low	3.90	0.13	2.34	0.17	0.038	0.003	0.12
22	High	3.66	0.08	2.72	0.18	0.054	0.004	0.07
	Low	3.92	0.10	2.65	0.19	0.048	0.004	0.09
25	High	4.26	0.07	2.34	0.10	0.051	0.002	0.05
	Low	4.19	0.12	2.34	0.15	0.041	0.003	0.08
28	High	4.07	0.09	2.75	0.17	0.060	0.004	0.07
	Low	3.79	0.08	3.03	0.18	0.061	0.004	0.06

' Values of α_w presented are those resulting from the addition of a constant to avoid negative values of $\log_e$ transformed data.

Values of the estimated maximum total plant dry weight (α_w) were highly dependent on treatment. Under the high PPF regime α_w increased linearly with increasing temperature (C), up to 25 C ($P \leq 0.001$, equation (3.19), Figure 3.6). However, in contrast to the high PPF regime, values of α_w under the low PPF regime were not influenced by temperatures above 13 C. Above 13 C, under the low PPF regime, α_w averaged 3.92 (i.e., 13.64 g) $\pm$ 0.13, while at 13 C α_w was depressed by 45% to 2.33 (i.e., 3.28 g) $\pm$ 0.13.

$$\alpha_w = 0.17 \cdot C - 1.2 \qquad (r^2 = 0.97,^{***}) \qquad (3.19)$$

This interaction between temperature and PPF on the estimated maximum total plant dry weight, resulted in greater values of α_w under the low PPF regime than under the high PPF

regime, at temperatures less than 22 C (Figure 3.6). At temperatures greater than 19 C the estimated maximum total plant dry weight was either not influenced by PPF (e.g. 22 C) or was greater under the high PPF regime. The maximum value of α_w was attained at 25 C, reaching an estimated 4.26 (i.e., 24.0 g) $\pm$ 0.07 under the high PPF regime, and 4.19 (i.e., 19.9 g) $\pm$ 0.12 under low PPF. A depression in the value of α_w was noted at temperatures above this, i.e., at 28 C, under both PPF regimes (Figure 3.6).

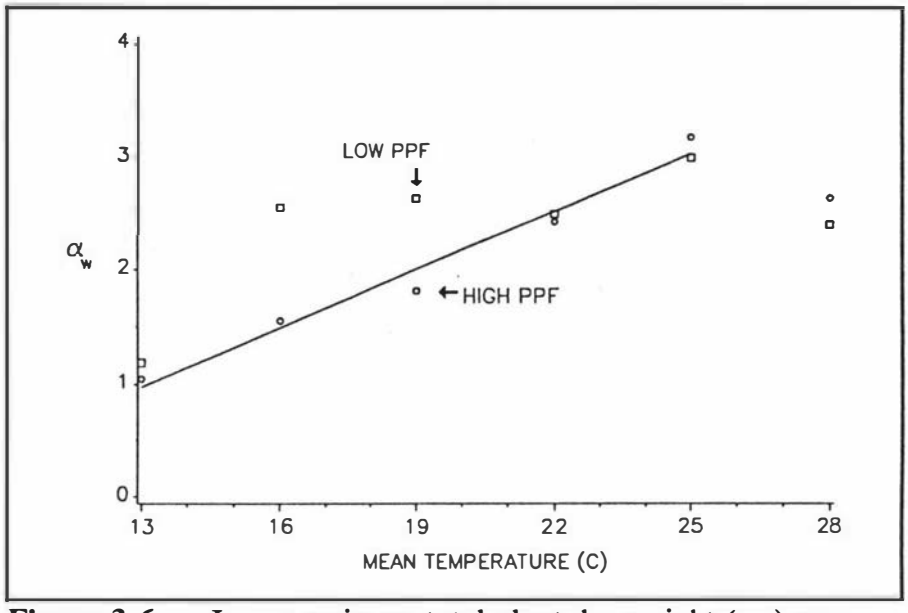

Figure 3.6 Log$_e$ maximum total plant dry weight (α_w) as a function of temperature, for *Zantedeschia* 'Best Gold' under high and low PPF regimes. Fitted line for high PPF regime only.

The value of parameter β_w of the fitted Gompertz curves, was not influenced by temperature nor PPF regime ($P \leq 0.10$). Across all treatments β_w averaged 2.7 $\pm$ 0.2.

In contrast to parameter β_w, the rate at which RGR_w declined as a function of size, i.e., parameter κ_w, was generally greater under the high PPF regime than under the low PPF regime ($P \leq 0.1$). Only at a temperature of 28 C was no difference in the value of κ_w detected between PPF regimes. Under the low PPF regime κ_w increased linearly with increasing temperature (C) ($P \leq 0.05$, equation (3.20), Figure 3.7). However, in contrast to the low PPF regime, values of κ_w under the high PPF regime were not influenced by temperature. Across all temperatures under the high PPF regime, κ_w averaged 0.053 $\pm$ 0.003.

$$\kappa_w = 0.018 \cdot C + 0.006 \qquad (r^2 = 0.77,^{**}) \qquad (3.20)$$

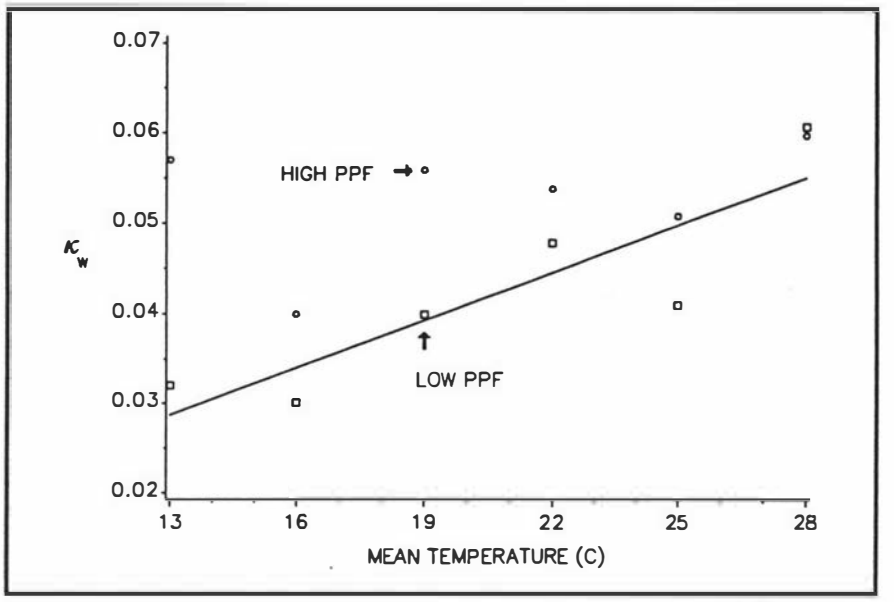

Figure 3.7 Interrelation between the rate of decline of RGR_w as a function of plant size (κ_w), and temperature, for *Zantedeschia* 'Best Gold' under high and low PPF regimes. Fitted line for low PPF only.

3.3.4 Curve fitting of leaf area and dry weight

Under all treatments total plant leaf area followed a sigmoidal pattern of growth subsequent to expansion of the first leaf and until the onset of declining total leaf area. A family of logarithmic Gompertz functions adequately described this progression of total plant leaf area ($\log_e$ transformed) (Figure 3.8, Figure 3.9, Table 3.4). The progression of total plant leaf dry weight ($\log_e$ transformed) with time was similarly fitted to logarithmic Gompertz functions (Table 3.5). Due to the similarity of the fitted curves for leaf area and dry weight, only figures depicting leaf area are presented.

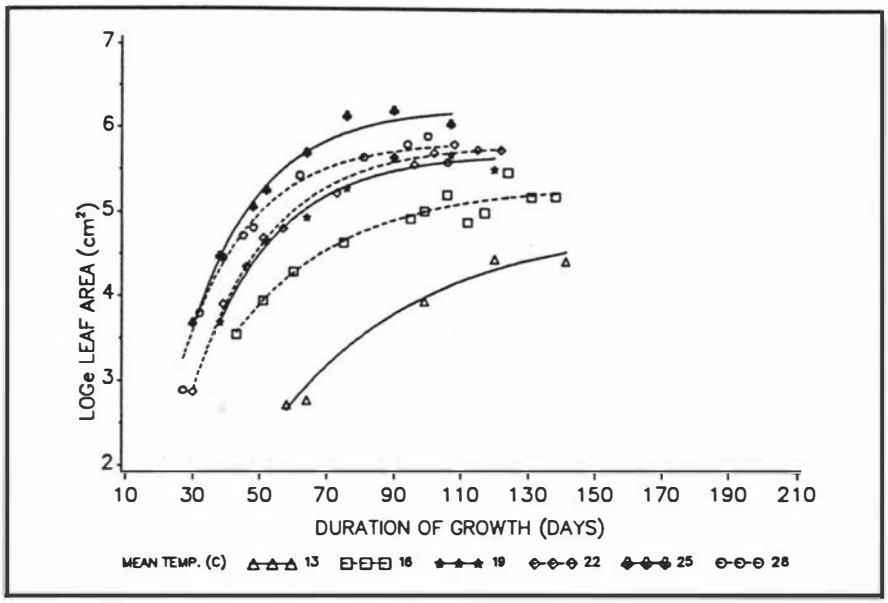

Figure 3.8 Fitted logarithmic Gompertz curves and mean data of total plant leaf area ($\log_e$) as a function of time, for *Zantedeschia* 'Best Gold' at a range of temperatures, under a high PPF regime.

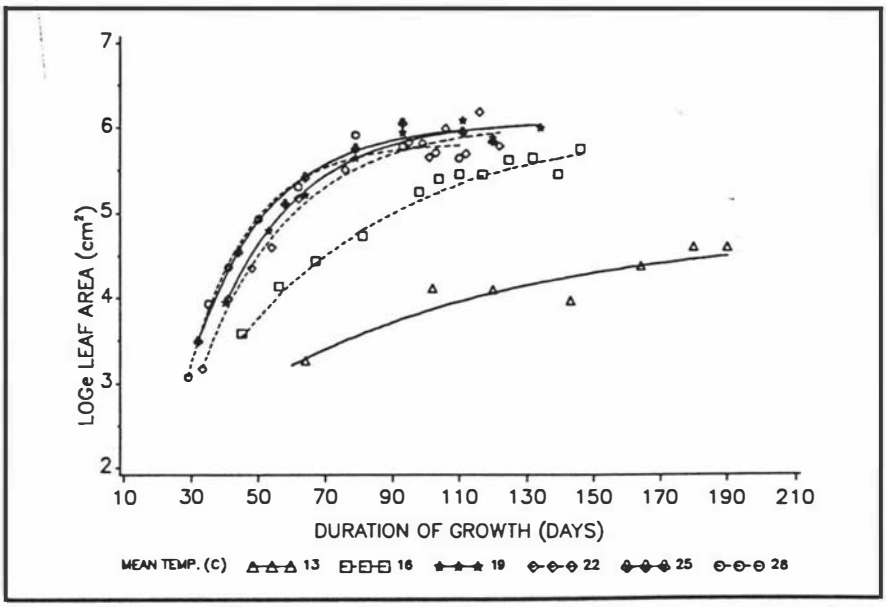

Figure 3.9 Fitted logarithmic Gompertz curves and mean data of total plant leaf area ($\log_e$) as a function of time, for *Zantedeschia* 'Best Gold' at a range of temperatures, under a low PPF regime.

Table 3.4

Nonlinear least-squares parameter estimates, associated asymptotic standard error (s.e.), and mean square error values, from fitting the logarithmic Gompertz function to $\log_e$ transformed total plant leaf area data for *Zantedeschia* 'Best Gold' grown at a range of temperatures, and under high and low PPF regimes. The PPF treatments at 19 C were repeated in two plantings, i.e., (1st) and (2nd).

Mean temperature (C)	PPF	α_A	s.e.	β_A	s.e.	κ_A	s.e.	mean square error
13	High	4.87	0.62	2.07	0.53	0.022	0.013	0.35
	Low	4.87	0.88	1.22	0.47	0.012	0.014	0.72
16	High	5.31	0.12	1.84	0.23	0.030	0.006	0.10
	Low	6.07	0.28	1.78	0.15	0.019	0.005	0.17
19 (1st)	High	5.68	0.09	2.34	0.25	0.004	0.007	0.11
	Low	6.09	0.08	2.37	0.20	0.040	0.005	0.10
19 (2nd)	High	5.12	0.13	2.68	0.34	0.054	0.010	0.16
	Low	5.74	0.08	2.29	0.18	0.038	0.005	0.11
22	High	5.78	0.05	2.37	0.08	0.044	0.003	0.05
	Low	6.06	0.08	2.25	0.11	0.036	0.003	0.08
25	High	6.22	0.06	2.36	0.11	0.047	0.004	0.06
	Low	6.05	0.06	2.34	0.12	0.044	0.004	0.07
28	High	5.82	0.09	2.26	0.21	0.049	0.007	0.09
	Low	5.83	0.06	2.59	0.13	0.055	0.004	0.07

As with total plant dry weight, the estimated maximum leaf area of plants grown under the high PPF regime, i.e., α_A of the fitted Gompertz curves, increased linearly with increasing temperature (C) up to 25 C ($P \leq 0.05$, equation (3.21), Figure 3.10). However, in contrast to the high PPF regime, values of α_A under the low PPF regime were not influenced by temperatures above 13 C ($P \leq 0.10$). Above 13 C, under the low PPF regime, α_A averaged 5.98 (i.e., 395.4 cm^2) $\pm$ 0.06, while at 13 C α_A was depressed by 67% to 4.87 (i.e., 130.3 cm^2) $\pm$ 0.88. (N.B. cm^2 as units of comparison).

$$\alpha_A = 0.11 \cdot C + 3.5 \qquad (r^2 = 0.97,^{**}) \qquad (3.21)$$

The estimated maximum leaf dry weight (α_L) responded similarly to α_A, with maximum leaf dry weight increasing linearly with temperature (C) up to 25 C, under the high PPF regime ($P \leq 0.01$, equation (3.22)). As occurred with α_A under the low PPF regime, temperatures above 13 C did not influence the value of α_L ($P \leq 0.10$), averaging 0.73 (2.08 g) $\pm$ 0.07. At 13 C α_L was depressed

by 78% to -0.77 (i.e., 0.46 g) ± 0.27. (N.B. g as units of comparison).

$$\alpha_L = 0.14 \cdot C - 2.3 \qquad (r^2 = 0.94,^{**}) \qquad (3.22)$$

Table 3.5

Nonlinear least-squares parameter estimates, associated asymptotic standard error (s.e.), and mean square error values, from fitting the logarithmic Gompertz function to $\log_e$ transformed total plant leaf dry weight data for *Zantedeschia* 'Best Gold' grown at a range of temperatures, and under high and low PPF regimes.

Mean temperature (C)	PPF	α_L	s.e.	β_L	s.e.	κ_L	s.e.	mean square error
13	High	-0.43	0.49	2.53	0.66	0.027	0.014	0.46
	Low	-0.77	0.27	2.28	0.97	0.028	0.016	0.83
16	High	-0.09	0.10	2.39	0.28	0.039	0.007	0.14
	Low	0.77	0.30	2.04	0.16	0.021	0.005	0.24
19 (1st)	High	0.28	0.08	2.99	0.33	0.058	0.009	0.15
	Low	0.74	0.08	2.62	0.20	0.042	0.005	0.14
19 (2nd)	High	-0.28	0.12	3.18	0.31	0.062	0.009	0.18
	Low	0.37	0.09	2.45	0.17	0.037	0.005	0.15
22	High	0.64	0.06	2.62	0.08	0.045	0.003	0.07
	Low	0.92	0.13	2.32	0.10	0.031	0.003	0.14
25	High	1.23	0.08	2.45	0.09	0.043	0.003	0.07
	Low	0.93	0.09	2.33	0.09	0.035	0.003	0.09
28	High	0.78	0.11	2.44	0.16	0.044	0.005	0.11
	Low	0.69	0.09	2.57	0.09	0.044	0.003	0.09

The greatest values of α_A, i.e., 6.22 (502.7 cm^2) ± 0.06, and of α_L, i.e., 1.23 (3.42 g) ± 0.08, were attained at a temperature of 25 C under high PPF. Both maximum leaf area (Fig. 4.9) and leaf dry weight were reduced at 28 C.

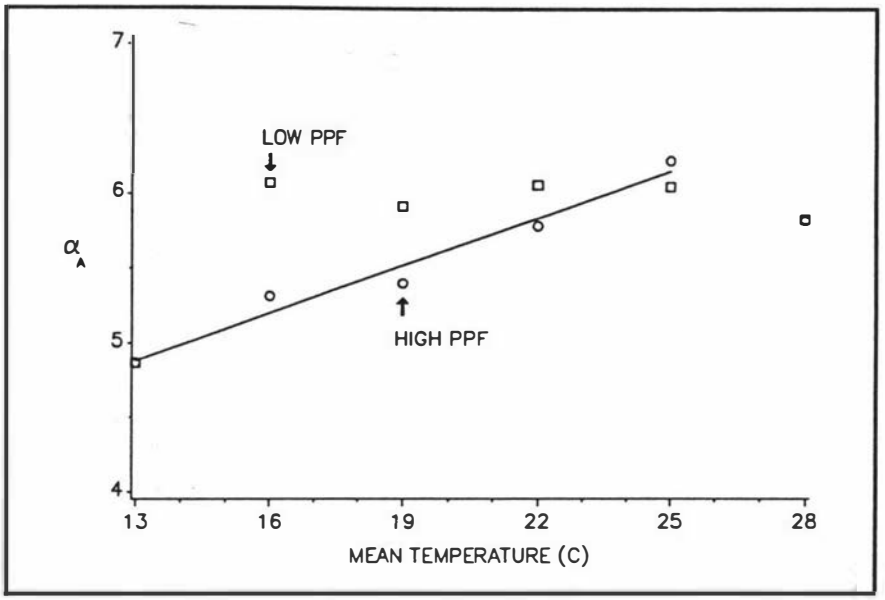

Figure 3.10 Log_e maximum total plant leaf area (α_A) as a function of temperature, for *Zantedeschia* 'Best Gold' under high and low PPF regimes. Fitted line for high PPF regime only.

Treatment differences in the development of leaf area and dry weight were evident throughout the period of study. At the time RGR_W was maximum, i.e., inflection point of the fitted total plant dry weight Gompertz curve, total plant leaf area was greater under the low PPF regime compared with high PPF, at temperatures less than 22 C (Table 3.6). In contrast, at this stage of development at temperatures greater than 19 C, leaf area was greater under the high PPF regime.

Table 3.6

Leaf area (cm^2) at inflection point of the fitted total plant dry weight Gompertz curve of *Zantedeschia* 'Best Gold', grown at a range of temperatures, and high and low PPF regimes.

	Mean temperature (C)					
PPF	13	16	19	22	25	28
			(cm^2)			
High	33.3	70.1	82.5	73.9	99.9	87.4
Low	38.8	77.1	97.6	65.5	71.1	77.7

The value of β_A of the fitted Gompertz curves was influenced by an interaction between temperature and PPF regime ($P \leq 0.08$). Under the low PPF regime there was a hyperbolic increase of β_A with increasing temperature (C) (equation (3.23), Figure 3.11).

$$\beta_A = 12.5 \cdot \tanh\left(\frac{1.4 \cdot C}{12.5}\right) - 9.9 \qquad \begin{pmatrix} mean \\ square = 0.02 \\ error \end{pmatrix} \qquad (3.23)$$

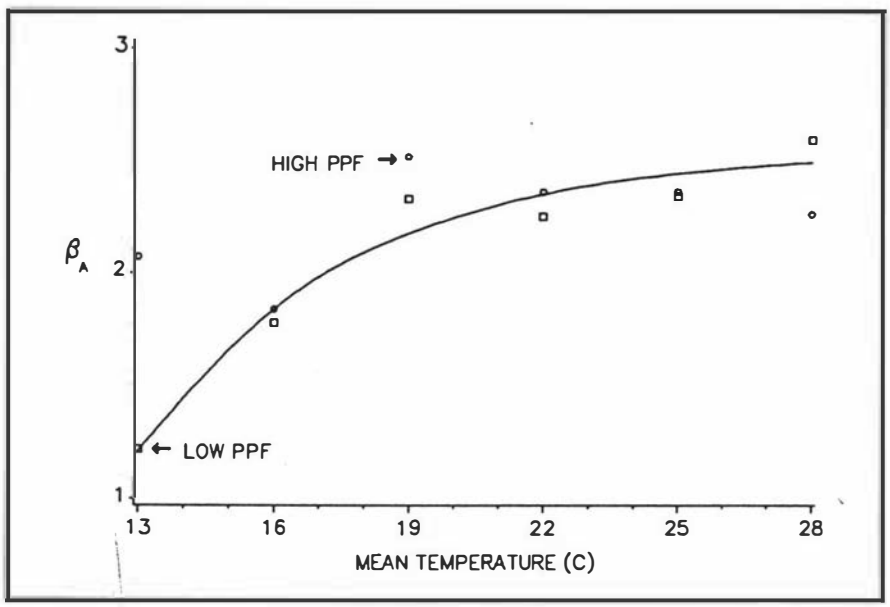

Figure 3.11 Parameter β_A as a function of temperature, for *Zantedeschia* 'Best Gold' under high and low PPF regimes. Fitted line for low PPF only.

There was no effect of temperature on β_A under the high PPF regime, the mean value being 2.27 ± 0.10, across all temperatures.

In contrast to β_A, the value of β_L was not influenced by temperature. However, across all temperature treatments the value of β_L was greater under the high PPF regime (i.e., 2.66 ± 0.12) than under the low PPF regime (i.e., 2.40 ± 0.07; $P \leq 0.07$).

The rate of decline of the relative leaf area expansion rate, as a function of size ($RLAER_s$), i.e., parameter κ_A, of the fitted Gompertz curves, increased with increasing temperature ($P \leq 0.001$). Under the low PPF regime this increase with temperature (C) was linear (equation (3.24), Figure 3.12). However, under the high PPF regime the increase in rate of decline of $RLAER_s$, with increasing temperature, was hyperbolic (equation (3.25), Figure 3.12).

$$\kappa_A = 0.0027 \cdot C - 0.022 \qquad (r^2 = 0.92,^{**}) \qquad (3.24)$$

$$\kappa_A = 0.3 \cdot \tanh\left(\frac{0.03 \cdot C}{0.3}\right) - 0.24 \qquad \begin{pmatrix} mean \\ square = 0.00003 \\ error \end{pmatrix} \qquad (3.25)$$

At each temperature the rate of decline of $RLAER_s$ was generally greater under the high PPF regime than the low PPF regime, except at a mean temperature of 28 C (Figure 3.12).

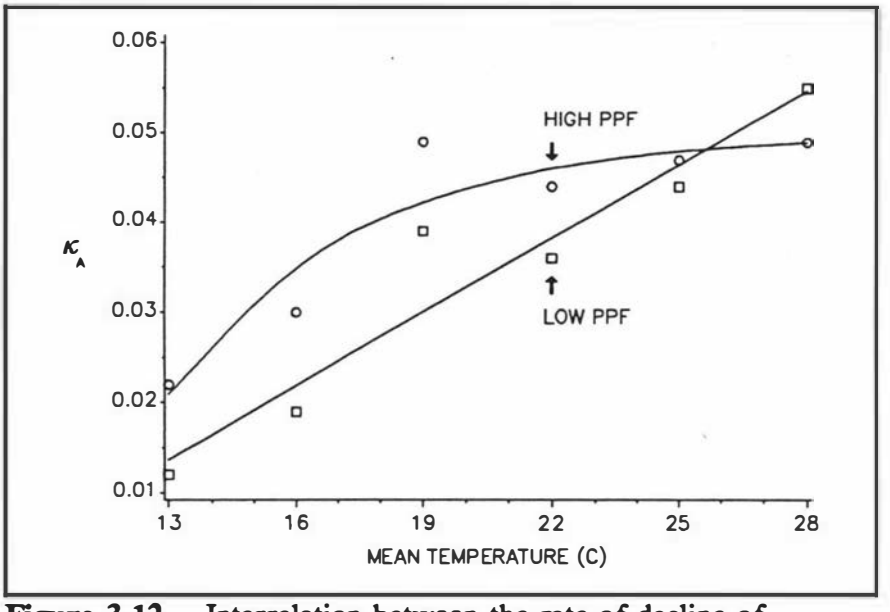

Figure 3.12 Interrelation between the rate of decline of $RLAER_s$ (κ_A) and temperature, for *Zantedeschia* 'Best Gold' under high and low PPF regimes.

As found with the parameter κ_A, for plants grown under the low PPF regime the rate of decline of relative leaf weight expansion rate, as a function of size ($RLWR_s$) i.e., parameter κ_L of the fitted Gompertz curves, increased linearly with increasing temperature (equation (3.26)).

$$\kappa_L = 0.0012 \cdot C + 0.01 \qquad (r^2 = 0.62,^{**}) \qquad (3.26)$$

In contrast to the rate of decline of $RLAER_s$ under high PPF, no response of the rate of decline of $RLWR_s$ to temperature was evident under the high PPF regime. Across all temperatures under the high PPF regime, the mean value of the rate of decline of $RLWR_s$ was 0.045 ± 0.004. However, as with the rate of decline of $RLAER_s$, the rate of decline of $RLWR_s$ was generally greater under

the high PPF regime than the low PPF regime ($P \leq 0.07$).

3.3.5 Relationships between derived parameters

During the exponential phase of total plant dry weight accumulation, a positive linear relationship existed between total plant relative growth rate (RGR_w) and both relative leaf area expansion rate (RLAER) and relative leaf weight rate (RLWR), regardless of PPF regime and temperature ($P \leq 0.0001$) (equations (3.27) and (3.28), Figure 3.13).

$$RGR_w = 0.82 \cdot RLAER + 0.015 \qquad (r^2 = 0.88,^{***}) \qquad (3.27)$$

$$RGR_w = 0.75 \cdot RLWR + 0.01 \qquad (r^2 = 0.92,^{***}) \qquad (3.28)$$

In contrast, no relationship between RGR_w and net assimilation rate (NAR) was detected for the entire data set, nor for separate PPF regimes ($P \leq 0.10$, Figure 3.14). While NAR was typically greater under the high PPF regime compared with the low PPF regime ($P \leq 0.01$), no influence of temperature was detected ($P \leq 0.10$).

Independent, positive, linear relationships were determined between RGR_w and leaf area partitioning (LAP) under the high and low PPF regimes (equations (3.29) and (3.30) respectively, Figure 3.15). The slope of the line describing the relationship between RGR_w and LAP under the high PPF regime was greater than that from the low PPF regime ($P \leq 0.05$).

$$\text{(High PPF)} \quad RGR_w = 0.0008 \cdot LAP + 0.003 \qquad (r^2 = 0.72,^{**}) \qquad (3.29)$$

$$\text{(Low PPF)} \quad RGR_w = 0.0003 \cdot LAP + 0.023 \qquad (r^2 = 0.93,^{***}) \qquad (3.30)$$

In contrast, a single, positive, linear relationship between RGR_w and the leaf weight partitioning (LWP) encompassed the response regardless of PPF regime and temperature (equation (3.31), Figure 3.16).

$$RGR_w = 0.156 \cdot LWP \qquad (r^2 = 0.98,^{***}) \qquad (3.31)$$

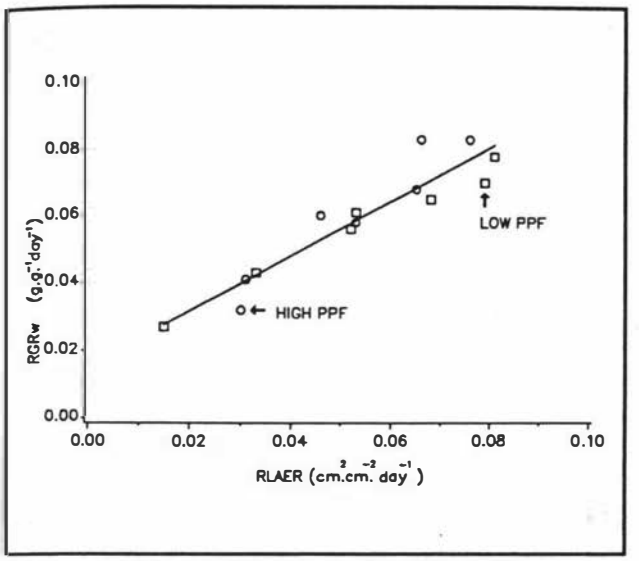

Figure 3.13 RGR_w as a function of RLAER, for *Zantedeschia* 'Best Gold' grown under two PPF regimes and six temperatures.

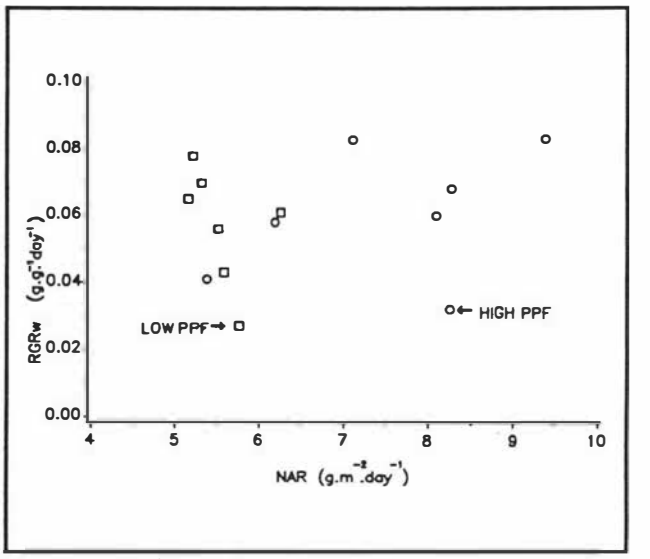

Figure 3.14 RGR_w as a function of NAR, for *Zantedeschia* 'Best Gold' grown under two PPF regimes and six temperatures.

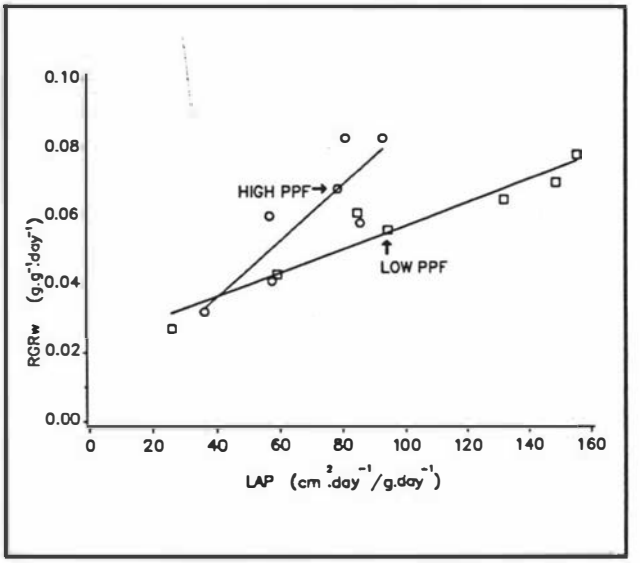

Figure 3.15 RGR_w as a function of LAP, for *Zantedeschia* 'Best Gold' grown under two PPF regimes and six temperatures.

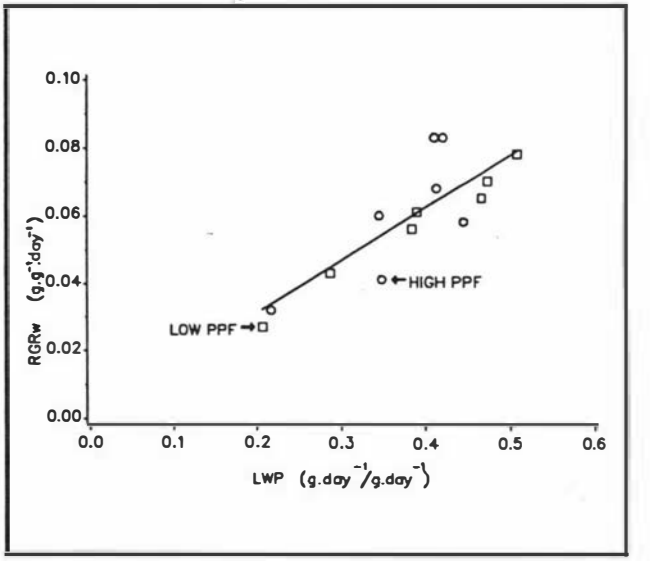

Figure 3.16 RGR_w as a function of LWP, for *Zantedeschia* 'Best Gold' grown under two PPF regimes and six temperatures.

3.4 Discussion

The strong correlation between relative rate of leaf area expansion and growth during the exponential phase (Figure 3.13) has been illustrated with a number of plant species, grown under a range of environments (Watson, 1947a & 1947b; El-Sharkawy et al., 1965; Muramoto et al., 1965; Duncan and Hesketh, 1968; Slatyer, 1970; Hanson, 1971; Delaney and Dobrenz, 1974; Potter and Jones, 1977). It should be appreciated however, that this correlation is not necessarily causal. In fact, extrapolation of this equation (equation (3.27)) infers that at a RLAER of 0 $cm^2 \cdot cm^{-2} \cdot day^{-1}$, i.e., no expansion of leaf area, a RGR_w of 0.015 $\pm$ 0.005 $g \cdot g^{-1} \cdot day^{-1}$ would be expected. Therefore clearly RLAER can not be considered solely causal of growth. However, since the daily partitioning of growth into leaf area will determine the area on the next day, the daily growth on the next day will depend to a large extent on the value of the LAP (Potter and Jones, 1977). While a strong correlation between RGR_w and LAP was shown to exist for plants of *Zantedeschia* 'Best Gold' grown at a range of temperatures, the magnitude of the relationship, i.e., slope of line, was more than doubled when plants were grown under high PPF compared with under low PPF (Figure 3.15). In contrast a single, positive, linear correlation between RGR_w and LWP was determined, which accounted for the differing temperature and PPF induced responses (Figure 3.16). In developing the potentially more causal relationship between RGR_w and LAP, Potter and Jones (1977) utilized data derived from a number of species grown at a range of temperatures without variation in PPF regime. If the current experiment had used only one PPF regime the same conclusion would have been reached as that of Potter and Jones (1977). It is therefore evident that the LAP may be limited in its application in inferring a more direct relationship with RGR_w to situations where a single PPF regime is utilized. The improvement in the ability to predict RGR_w via a single relationship using LWP rather than two independent relationships using LAP, was as a result of LWP accounting for differences in specific leaf weight. Changes in daily quantum integral of light have been shown to account for differences in specific leaf weight of a number of plant species (Warrington and Norton, 1991). In addition, differences in specific leaf weight have accounted for variation in LAP in perennial ryegrass and maize (Hunt and Halligan, 1981; Tollenaar, 1989a), thereby supporting the logic of utilizing LWP under variable PPF regimes.

The theory that LWP may be more robust in its use for determining plant growth in *Zantedeschia* 'Best Gold' is further strengthened when consideration is given to extrapolation of equation (3.31) to where a LWP of 0 $g \cdot day^{-1}/g \cdot day^{-1}$ would result in a RGR_w of 0 $g \cdot g^{-1} \cdot day^{-1}$. This outcome is more logical than that achieved when using RLAER as a determinant of plant growth, as discussed above. Since in the natural environment PPF levels are not constant (i.e., seasonal changes as well as changes with latitude) it is suggested that LWP will provide a more robust determinant of growth of

Zantedeschia as well as other plant genera.

The poor correlation between NAR and growth (Figure 3.14) has been noted with other species (Heath and Gregory, 1938; Watson, 1952; Hunt and Parsons, 1974; Lambers, 1987; Poorter, 1990). Equally however, examples of increased RGR_w as a result of increased NAR have been reported (Rajan et al., 1973; Patterson et al., 1978; Lambers, 1987; Tollenaar, 1989b; Poorter, 1990). When one considers that a primary component of NAR is derived from the photosynthetic rate per unit area, coupled with the frequently poor success of correlating unit photosynthetic rates with growth (Lambers, 1987), it is not surprising that no relationship between RGR_w and NAR was determined in the current experiment. However, it should not be overlooked that Poorter (1990) indicated that the poor correlation was primarily attributable to strong negative correlation between NAR and LAR. Therefore, the poor correlation determined here does not necessarily negate the possible dominance of NAR determining growth when the growing environment more directly influences photosynthesis. This has been clearly illustrated with gaseous pollutants whose presence resulted in a direct influence on photosynthesis, and thereby resulted in a strong correlation between growth and NAR (Jensen, 1981). Further discussion of any possible relationship between photosynthetic rate and plant growth in *Zantedeschia* 'Best Gold' will be presented in Chapter 5.

In addition to the aforementioned variation between species in the degree of correlation between growth and NAR, variation in both the presence and strength of any correlation between growth and LWP and/or LAP, between species, has also been reported (Potter and Jones, 1977; Patterson et al., 1978; Ibrahim and Buxton, 1981; Sage and Pearcy, 1987). It is evident that species vary in their mechanisms of acclimating to their growing environment. While exceptions exist for any attempt to classify plants into definitive categories of sun and shade species/ecotypes, shade-tolerant species/ecotypes often exhibit an ability to acclimate to alteration in PPF through an enhanced alteration of LAR and thereby LWP and LAP (Bjorkman and Holmgren, 1966; Gauhl, 1976; Boardman, 1977; Pons, 1977; Bjorkman, 1981; Kuiper and Smid, 1985). In contrast sun-species/ecotypes frequently possess a greater capacity to alter NAR under conditions of increased PPF. While consideration of other factors contributing to the classification of *Zantedeschia* 'Best Gold' as a typical shade-tolerant or sun species will be discussed in Chapter 5, i.e., adaptability of photosynthetic rate, from the data presented here it appears that *Zantedeschia* 'Best Gold' does exhibit the phenotypic plasticity of a shade-tolerant species.

In terms of Hunt's (1982) simplistic description of growth, differences in *Zantedeschia* plant dry weight that occur during the exponential phase of total plant dry weight accumulation, appear to be primarily attributable to changes in *"leafiness"* of plants. This

change in leafiness is clearly illustrated by the fact that increasing temperature resulted in increased leaf development (Figure 3.8, Figure 3.9, Table 3.4, and Table 3.5). By way of an example of this relationship, at the mid point of the exponential phase of total plant dry weight accumulation, at intermediate temperatures, leaf areas under the low PPF regime, were greater than under high PPF (Table 3.6). This increased leaf development under low PPF was able to almost fully compensate for any reduction in NAR, and maintain RGR_w at rates similar to those achieved under high PPF (Figure 3.3). Similarly, at intermediate temperatures the prediction that maximum total plant dry weight would be greater under the low PPF regime than under high PPF (Figure 3.6), even though photosynthetic rate was found to be greater under the latter (refer Chapter 5), can primarily be explained by greater leaf area development under the low PPF regime (Figure 3.8 to Figure 3.10).

As reported for other plant species, leaf area duration may also have contributed to the prediction of greater maximum total plant dry weight under the low PPF regime than under the high PPF regime (Radley, 1963; Rees, 1972; Allen and Scott, 1980). However, the duration of the current study did not permit collection of data to the cessation of leaf development to investigate this possibility. Certainly the greater rate of decline of RLAER and RLWR, as a function of size, under the high PPF regime (Figure 3.12), in conjunction with the earlier onset of leaf senescence under the high PPF regime, are indicative of a reduced leaf area duration.

Differences in estimated maximum total plant dry weight between PPF regimes were greatest at intermediate temperatures, with little or no difference being evident between PPF regimes at extreme temperatures, i.e., mean temperatures of 13 C and 28 C (Figure 3.6). This variation in response of dry matter accumulation to PPF regime, with temperature, is equally explained by variation in LAP and LWP (Figure 3.15 and Figure 3.16). Therefore it is apparent, that *Zantedeschia* 'Best Gold' is readily able to acclimate and compensate for changes in PPF, via alteration in LAP and LWP, at temperatures greater than 13 C and less than 28 C. A growth temperature of 28 C, while initially stimulating very rapid growth (Figure 3.3), resulted in a reduction of maximum total plant dry weight compared with that attained at 25 C (Figure 3.6). With reference to the potentially causal relationship between RGR_w and LAP and LWP, values of all these parameters at 28 C were similarly reduced relative to those at 25 C, hence supporting the theory of the dominance of Hunt's (1982) *"leafiness of the plant"* correlating to total plant growth.

While in theory parameter β_w of the fitted Gompertz curves is an estimate of the total plant dry weight at day 0, this interpretation is based on the assumption that substrate supply is non-limiting (Thornley and Johnson, 1990). Clearly during the initial establishment stage,

i.e., until functional leaf area was present, this was not the case as total plant dry weight declined (Figure 3.1). Under normal circumstances the magnitude of differences in the value of β_W between treatments, might be indicative of the relative magnitude of weight loss that occurred until the establishment of functional leaf area, i.e., higher values of β_W inferring less loss of total plant dry weight during this phase. However, due to the need to add a constant to all transformed data to eliminate negative values being submitted for curve fitting, no treatment differences between the values of β_W were evident. Hence in this experiment the magnitude of values of β_W do not carry any biological significance. As would be expected from the increase in specific leaf weight at increased PPF, the value of β_L was generally greater under the high PPF regime than the low PPF regime. Being estimates of the leaf area and leaf dry weight at day 0, β_A and β_L should be considered as being merely important parameters for curve fitting, as their subsequent influence on growth is better explained in terms of LAP and LWP, as discussed above.

The pooled predicted base temperature for growth of 2.1 $\pm$ 2.7 C is within the range predicted for germination of many plant species utilized as winter crops, such as wheat and turnips, i.e., generally between 2 and 5 C. In contrast, warm season crops originating from temperate/warm-temperate climes, such as cotton, sorghum, rice, and corn, have base temperatures between 8 and 10 C (Angus et al., 1981; Singh and Dhaliwal, 1972; Warrington and Kanemasu, 1983). Just how accurate such estimations of base temperature are is open to debate, as base temperatures have been noted to change with development in crops such as wheat (Porter and Delecolle, 1988). A positive linear response of growth rate with temperature has generally been found between 5 and 20 C (Grace, 1988). While this linear response has been used to predict the base temperature (Figure 3.3), without actual examination of growth at such low temperatures, the predicted base temperature may have to be taken as merely an estimation for mathematical crop modelling purposes only. The low base temperature for *Zantedeschia* 'Best Gold' is also somewhat surprising considering climatological data for the sites of natural habitat indicate a warm-temperate climate for this crop (refer Section 1.3.2). While exceptions are always evident, the natural distribution of perennial species has been shown to be critically dependent on the temperature minima during both the growing season and/or annual period (Korner and Larcher, 1988; Woodward, 1988). With a predicted base temperature of 2.1 C compared with minimum soil temperatures in its natural habitat reaching 8.1 C (Table 1.2), it is suggested that either ecodormancy, resulting from soil moisture stress, and/or endodormancy, prevent growth during the winter period of its natural habitat. The minimal rainfall during winter was outlined in Section 1.3.2, as was the existence of endodormancy in summer flowering *Zantedeschia* (Corr and Widmer, 1988). Since derivation of the base temperature used data collected when plant leaf area was well established, an alternative explanation for the low predicted base temperature could be that

rates of photoassimilate accumulation decline less, with decreases in temperature, than do rates at which assimilates are used (Warren-Wilson, 1966; Verkleij and Challa, 1988; Acock et al., 1990). Hence, while growth and development of a recently planted tuber may be restricted at temperatures below 7 ± 3 C (refer Section 1.5.4.3), dry matter accumulation would be able to continue on a plant with developed leaf area due to continued photosynthesis at these temperatures.

Maximum RGR_w values of 0.080 $g \cdot g^{-1} \cdot day^{-1}$ at a temperature of 25 C and at high PPF (Figure 3.3) is well within the range of 0.02 to 0.4 $g \cdot g^{-1} \cdot day^{-1}$ reported for other plant species, but is between typical values for sun species (e.g., 0.1 to 0.4 $g \cdot g^{-1} \cdot day^{-1}$) on the one hand, and shade species (e.g., 0.02 to 0.04 $g \cdot g^{-1} \cdot day^{-1}$) on the other (Briggs et al., 1920; Blackman and Black, 1959; Warren Wilson, 1972). In reviewing the linear temperature response of RGR_w in C_3 crop species, Grace (1988) indicated an average 7% increase in RGR_w per degree Celsius. In the current experiment RGR_w also increased by 7% per degree Celsius (Figure 3.3).

In addition to the distribution of perennial species being determined by the temperature minima of the natural habitat, the heat sum during the growing season has also been shown to be critical (Woodward, 1988). While no attempt has been made to determine the equivalent heat sum for the natural habitat of *Zantedeschia pentlandii*, the mean temperature for maximal growth of the *Zantedeschia pentlandii*-like selection 'Best Gold', i.e., between 22 and 25 C, was greater than the average daily mean air temperature during the growing season for the sites of natural habitat, i.e., 20 C (refer Section 1.3.2). In addition, with daily maximum air temperatures during the growing season averaging 25.4 C, it is apparent that *Zantedeschia* 'Best Gold' is well adapted to take advantage of the temperature regimes of its natural habitat.

The linear relationship between RGR_w and temperature (Figure 3.3) indicates that horticulturally relevant temperatures can be identified, i.e., base temperature (2.1 ± 2.7 C), the linear range of mean temperature response (2.1 to 28 C), and mean temperatures resulting in maximum growth (i.e., 25 C).

During the initial stages of plant development no leaves were present, and therefore it is acknowledged that the *"resource"* of assimilates referred to by Warren Wilson et al. (1986), refers to the tuber at this stage of development. The change with development in role of organs as net assimilate source or sink, has previously been noted in geophytic crops (Rees, 1972). During this initial establishment stage no importation of recently-produced assimilates is possible, thus respiratory losses associated with the development of leaf and root tissue results in a net loss in total plant dry weight (Figure 3.1). Since

leaves of plants on other species are found to be net exporters of assimilates once they have attained 50% or more of their full expansion (Giaquinta, 1978), the increments in total plant dry weight at the time of the first harvest within treatments, i.e., when 75% or more expansion of the first leaf had occurred, was not unexpected (Figure 3.1). The rate and extent of recovery of this lost dry weight was subsequently dependent on those factors associated with equation (3.1), in particular partitioning of assimilates into leaf growth.

Plant responses to temperature are considered to be governed by both the direct effects of temperature on metabolic activity (i.e., enzyme and membrane carrier) and indirect physical effects (i.e., diffusion and viscosity) (Farrar and Williams, 1991). While PPF can indirectly affect photosynthesis through stomatal aperature (Dwelle et al., 1981; Valenzuela et al., 1990), as a source of energy utilized to drive the acquisition and utilization of carbon, the interception of PPF is an integral component of plant growth (Warrington and Norton, 1991).

While in the current study the focus of the environmental influences on plant growth has been presented in terms of mean temperature and PPF regimes within a controlled environment, it is appreciated that the concepts of heat units (Warrington and Kanemasu, 1983) and daily quantum integrals of light (Warrington and Norton, 1991) will need to be developed before a suitably robust crop model can be developed. In addition, between plant competition did not exist within the current study, eliminating the need to consider the concept of leaf area index. Considering the ability of *Zantedeschia* 'Best Gold' to compensate for reduced PPF via increased partitioning to the development of leaf, the attainment of increased total plant dry weight under low PPF may not result in the field once competition between plants exists.

3.4.1 Conclusions

In response to variable temperature and PPF, dry matter accumulation of *Zantedeschia* 'Best Gold' was highly correlated with the partitioning of dry matter into leaf development. In contrast, the correlation between dry matter accumulation and net assimilation rate was poor.

Dry matter accumulation of *Zantedeschia* 'Best Gold' was highly adaptive to the PPF regime. While the optimum PPF under which to grow *Zantedeschia* 'Best Gold' was dependent on temperature, maximum dry matter accumulation occurred under the high PPF regime at a mean temperature of 25 C. Hence the horticultural management consequences of these findings are that the establishment and maintenance of an effective leaf area will be critical if growth is to be maximised.

3.5 References

Acock, B., M.C. Acock and D. Pasternak. 1990. Interactions of CO_2 enrichment and temperature on carbohydrate production and accumulation in muskmelon leaves. J. Amer. Soc. Hort. Sci. 115:525-529.

Allen, E.J. and R.K. Scott. 1980. An analysis of growth of the potato crop. J. Agr. Sci. (Cambridge) 94:583-606.

Angus, J.F., R.B. Cunningham, M.W. Moncur and D.H. MacKenzie. 1981. Phasic development in field crops. I. Thermal response in the seedling phase. Field Crops Res. 3:365-378.

Bjorkman, O. 1981. Responses to different quantum flux densities, p. 57-107. In: O.L. Lange, P.S. Nobel, C.B. Osmond, and H. Ziegler (eds.). Encyclopedia of Plant Physiology, vol. 12 A, Physiological plant ecology. Springer Verlag, Berlin.

Bjorkman, O. and P. Holmgren. 1966. Photosynthetic adaptation to light intensity in plants native to shaded and exposed habitats. Physiol. Plant. 19:854-859.

Blackman, G.E. and J.N. Black. 1959. Physiological and ecological studies in the analysis of plant environment. XI. A further assessment of the influence of shading on the growth of different species in the vegetative phase. Ann. Bot. 23:51-63.

Boardman, N.K. 1977. Comparative photosynthesis of sun and shade plants. Annu. Rev. Plant Physiol. 28:355-377.

Briggs, G.E., F. Kidd and C. West. 1920. A quantitative analysis of plant growth. I. Ann Applied Biol. 7:103-123.

Brooking, I.R. 1976. Soilless potting media for controlled-environment facilities. N.Z. J. Expt. Agr. 4:203-208.

Causton, D.R. and J.C. Venus. 1981. Single leaf growth and the Richards function: methodology, p. 87-143. In: The biometry of plant growth. Edward Arnold, London.

Collins, R.P. and M.B. Jones. 1988. The effects of temperature on leaf growth in *Cyperus longus*, a temperate C_4 species. Ann. Bot. 61:355-362.

Corr, B.E. and R.E. Widmer. 1988. Rhizome storage increases growth of *Zantedeschia elliottiana* and *Z. rehmannii*. HortScience 23:1001-1002.

Delaney, R.H. and A.K. Dobrenz. 1974. Yield of alfalfa as related to carbon exchange. Agron. J. 66:498-500.

Duncan, W.G. and J.D. Hesketh. 1968. Net photosynthetic rates, relative leaf growth rates, and leaf numbers of 22 races of maize grown at eight temperatures. Crop Sci. 8:670-674.

Dwelle, R.B., G.E. Kleinkopf and J.J. Pavek. 1981. Stomatal conductance and gross photosynthesis of potato (*Solanum tuberosum* L.) as influenced by irradiance, temperature, and growth stage. Potato Res. 24:49-59.

El-Sharkawy, M., J. Hesketh and H. Muramoto. 1965. Leaf photosynthetic rates and other growth characteristics among 26 species of *Gossypium*. Crop Sci. 5:173-175.

Farrar, J.F. 1988. Temperature and the partitioning and translocation of carbon, p. 203-236. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Farrar, J.F. and M.L. Williams. 1991. The effects of increased atmospheric carbon dioxide and temperature on carbon partitioning, source-sink relations and respiration. Plant, Cell and Environ. 14:819-830.

Francis, F.J. and F.M. Clydesdale. 1975. Food colorimetry: theory and applications. AVI, Westport, Conn.

Funnell, K.A. and B.R. MacKay. 1990. Alleviation of storage induced flowering decline, in *Zantedeschia*, with Promalin. Tech. Rpt. 89/1, Dept. Hort. Sci., Massey Univ., Palmerston North, N.Z.

Gauhl, E. 1976. Photosynthetic response to varying light intensity in ecotypes of *Solanum dulcamara* L. from shaded and exposed habitats. Oecologia 22:275-286.

Giaquinta, R. 1978. Source and sink leaf metabolism in relation to phloem translocation. Carbon partitioning and enzymology. Plant Physiol. 61:380-385.

Grace, J. 1988. Temperature as a determinant of plant productivity, p. 91-107. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Hanson, W.D. 1971. Selection for differential productivity among juvenile maize plants: associated net photosynthetic rate and leaf area changes. Crop Sci. 11:334-339.

Heath, O.V.S. and F.G. Gregory. 1938. The constancy of the mean net assimilation rate and its ecological importance. Ann. Bot. 2:811-818.

Hunt, R. 1982. Plant growth curves. The functional approach to plant growth analysis. Edward Arnold, London.

Hunt, R. and I.T. Parsons. 1974. A computer program for deriving growth-functions in plant growth-analysis. J. Applied Ecol. 11:297-306.

Hunt, W.F. and G. Halligan. 1981. Growth and development responses of perennial ryegrass grown at constant temperature. I. Influence of light and temperature on growth and net assimilation. Australian J. Plant Physiol. 8:181-190.

Ibrahim, M.E. and D.R. Buxton. 1981. Early vegetative growth of cotton as influenced by leaf type. Crop Sci. 21:639-642.

Jackson, J.E. 1963. Relationship of relative leaf growth rate to net assimilation rate and its relevance to the physiological analysis of plant yield. Nature 200:909.

Jassby A.D. and T. Platt. 1976. Mathematical formulation of the relationship between photosynthesis and light for phytoplankton. Limnology and Oceanography 21:540-547.

Jensen, K.F. 1981. Growth analysis of hybrid poplar cuttings fumigated with ozone and sulphur dioxide. Environ. Pollution 26:243-250.

Konings, H. 1990. Physiological and morphological differences between plants with a high NAR or a high LAR as related to environmental conditions, p. 101-124. In: H. Lambers, M.L. Cambridge, H. Konings, and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague.

Korner, C. and W. Larcher. 1988. Plant life in cold climates, p. 25-58. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Kuiper, D. and A. Smid. 1985. Genetic differentiation and phenotypic plasticity in *Plantago major ssp major*: I. The effect of differences in level of irradiance on growth, photosynthesis, respiration and chlorophyll content. Physiol. Plant. 65:520-528.

Lambers, H. 1987. Does variation in photosynthetic rate explain variation in growth rate and yield ? Neth. J. Agr. Sci. 35:505-519.

Muramoto, H., J. Hesketh and M. El-Sharkawy. 1965. Relationships among rate of leaf area development, photosynthetic rate, and rate of dry matter production among American cultivated cottons and other species. Crop Sci. 5:163-166.

Patterson, D.T., C.R. Meyer and P.C. Quimby. 1978. Effects of irradiance on relative growth rates, net assimilation rates, and leaf area partitioning in cotton and three associated weeds. Plant Physiol. 62:14-17.

Pons, T.L. 1977. An ecophysiological study in the field layer of ash coppice. II Experiments with *Geum urbanum* and *Cirsium palustre* in different light intensities. Acta Bot. Neerl. 26:29-42.

Poorter, H. 1990. Interspecific variation in relative growth rate: on ecological causes and physiological consequences, p. 45-68. In: H. Lambers, M.L. Cambridge, H. Konings, and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague.

Porter, J.R. and R. Delecolle. 1988. Interaction of temperature with other environmental factors in controlling the development of plants, p. 133-156. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Potter, J.R. and J.W. Jones. 1977. Leaf area partitioning as an important factor in growth. Plant Physiol. 59:10-14.

Radley, R.W. 1963. The effect of season on growth and development of the potato, p. 211-220. In: J.D. Ivins and F.L. Milthorpe (eds.). The growth of the potato. Butterworths, London.

Rajan, A.K., B. Betteridge and G.E. Blackman. 1973. Differences in the interacting effects of light and temperature on growth of four species in the vegetative phase. Ann. Bot. 37:287-313.

Rees, A.R. 1972. The Growth of Bulbs. Academic Press, London.

Sage, R.F. and R.W. Pearcy. 1987. The nitrogen use efficiency of C_3 and C_4 plants. I. Leaf nitrogen, growth, and biomass partitioning in *Chenopodium album* (L.) and *Amaranthus retroflexus* (L.). Plant Physiol. 84:954-958.

Singh, N.T. and G.S. Dhaliwal. 1972. Effect of soil temperature on seedling emergence in different crops. Plant and Soil 37:441-444.

Slatyer, R.O. 1970. Comparative photosynthesis, growth and transpiration of two species of *Atriplex*. Planta 93:175-189.

Thornley, J.H.M. and I.R. Johnson. 1990. Plant growth functions, p. 74-89. In: Plant and crop modelling - a mathematical approach to plant and crop physiology. Clarendon Press, Oxford.

Tollenaar, M. 1989a. Response of dry matter accumulation in maize to temperature: I. Dry matter partitioning. Crop Sci. 29:1239-1246.

Tollenaar, M. 1989b. Response of dry matter accumulation in maize to temperature: II. Leaf photosynthesis. Crop Sci. 29:1275-1279.

Valenzuela, H.R., B. Schaffer and S.K. O'Hair. 1990. Shade and nitrogen influence gas exchange and growth of cocoyam (*Xanthosoma sagittifolium*). J. Amer. Soc. Hort. Sci. 115:1014-1018.

Verkleij, F.N. and H. Challa. 1988. Diurnal export and carbon economy in an expanding source leaf of cucumber at contrasting source and sink temperature. Physiol. Plant. 74:284-293.

Warren Wilson, J. 1966. An analysis of plant growth and its control in arctic environments. Ann. Bot. 30:383-402.

Warren Wilson, J. 1972. Control of crop processes, p. 7-30. In: A.R. Rees, K.E. Cockshull, D.W. Hand, and R.G. Hurd (eds.). Crop processes in controlled environments. Academic Press, London.

Warren Wilson, J., R. Hunt and D.W. Hand. 1986. Philosophical aspects of measurements, equations and inferences in plant growth studies. Ann. Bot. 58:73-80.

Warrington, I.J., T. Dixon, R.W. Robotham and D.A. Rook. 1978. Lighting systems in major New Zealand controlled environment facilities. J. Agr. Eng. Res. 23:23-26.

Warrington, I.J. and E.T. Kanemasu. 1983. Corn growth response to temperature and photoperiod. I. Seedling emergence, tassel initiation, and anthesis. Agron. J. 75:749-754.

Warrington, I.J. and R.A. Norton. 1991. An evaluation of plant growth and development under various daily quantum integrals. J. Amer. Soc. Hort. Sci. 116:544-551.

Watson, D.J. 1947a. Comparative physiological studies on the growth of field crops. I. Variation in net assimilation rate and leaf area between species and varieties, and within and between years. Ann. Bot. 11:41-76.

Watson, D.J. 1947b. Comparative physiological studies on the growth of field crops. II. The effect of varying nutrient supply on net assimilation rate and leaf area. Ann. Bot. 11:375-407.

Watson, D.J. 1952. Physiological basis of variation in yield. Adv. Agron. 4:101-145.

West, C., G.E. Briggs and F. Kidd. 1920. Methods and significant relations in the quantitative analysis of plant growth. New Phytol. 19:200-207.

Woodward, F.I. 1988. Temperature and the distribution of plant species, p. 59-75. In: S.P. Long and F.I. Woodward (eds.). Plants and Temperature. Symp. of the Soc. for Expt. Biol. XXXXII. The Company of Biologists, Cambridge.

Zar, J.H. 1984. Biostatistical analysis. 2nd ed. Prentice Hall, New Jersey.

4 TUBER DRY MATTER ACCUMULATION OF *Zantedeschia* 'Best Gold' IN RESPONSE TO TEMPERATURE AND PHOTOSYNTHETIC PHOTON FLUX

4.1 Introduction

Zantedeschia is a relatively new horticultural crop with a rapidly expanding world-wide market for tubers (refer Section 1.2). Although *Zantedeschia* may be grown at any time of the year under protected cultivation (refer Sections 1.5 and 1.6), responses of this genus to manipulation of environmental parameters, such as temperature and photosynthetic photon flux (PPF), have not been described in detail. Since the above environmental parameters are frequently under the control of the horticulturist, gaining an understanding of how the plant responds to their modification would provide information required to develop assimilate partitioning models, crop management and plant breeding strategies.

Improvements in the partitioning of photoassimilates between the economic yield component and the remainder of the plant have generally been responsible for the improved yields obtained from many field crops over the last century (Gifford et al., 1984). When this partitioning is expressed at a single point in time, a measure of the cumulative partitioning to that date is derived (i.e., harvest index). The harvest index therefore provides a simple but coarse measure of assimilate partitioning. With partitioning frequently changing with plant development, the use of harvest indices is generally restricted to crops with a single, readily determined point of harvest, e.g. grain crops, and therefore is less applicable to crops such as *Zantedeschia*, cassava (*Manihot esculenta* Crantz.; Boerboom, 1978), potato (*Solanum tuberosum* L.; Menzel, 1985), taro (*Colocasia esculenta* (L.) Schott) and tannia (*Xanthosoma sagittifolium* (L.) Schott; Caesar, 1980), which possess geophytic storage organs with indeterminate storage organ capacity.
In situations where a number of harvests are carried out over time, the proportion of the daily increment in total dry weight gain partitioned into tuber growth, i.e., tuber weight partitioning (TWP, equation (4.1)) may be calculated. While known as different terms by different authors, such as "*efficiency of storage root production*", and "*distribution ratio*", this method of calculation of partitioning provides a more sensitive measure of short term changes in assimilate partitioning than does the harvest index (Boerboom, 1978; Keating et al., 1982). The importance of TWP is that it can be used as an improved indicator of the mechanisms of assimilate partitioning and to account for the way in which partitioning might be manipulated by environmental influences.

$$TWP = \frac{\delta T/\delta t}{\delta W/\delta t} \qquad (4.1)$$

where;

TWP = tuber weight partitioning
T = tuber dry weight
W = total plant dry weight
t = time
δ = mathematical notation for an interval

In its simplest form, two possible situations can be envisaged in the control of assimilate accumulation within a plant: either the system is source limited, or the system is sink limited (Wareing and Patrick, 1975). Which one is limiting under any particular circumstance will be dependent on both genotype and environment, and their interaction (Gifford et al., 1973; Farrar, 1992). In proposing this model where either source or sink limitation can exist, it is assumed that assimilate movement between source and sink is non-limiting. While in most cases examined this assumption is valid (Wareing and Patrick, 1975; Minchin and Thorpe, 1992), it was not the intention of the current experiment to investigate this aspect.

If sinks control partitioning of dry weight, then the corollary is that it is those factors which determine the initiation and relative activity of sinks which determine the pattern of assimilate distribution. The initiation of the storage organ in commercial selections of onion (*Allium cepa* L.), potato, and tulip (*Tulipa gesneriana* L.) has an obligative requirement for specific photoperiods and/or temperatures (Garner and Allard, 1923; Niimi, 1978; Ewing, 1987; Taeb and Alderson, 1990). In contrast, the initiation of the storage organ of other genera (e.g., *Beta*), does not appear to be under obligative environmental control. Thickening of the storage organ in sugar beet (*Beta vulgaris* L.), comprised of the swollen hypocotyl and tap root, commenced soon after germination, and resulted from the successive initiation of cambia and associated production of parenchymatous tissues (Rapoport and Loomis, 1986). Although partitioning of assimilates between shoot and storage root of crops such as sugar beet followed an allometric relationship for the majority of the growth cycle, this was not applicable during the early stages of growth (McLaren, 1984). Milford et al. (1988) illustrated that, during early stages of growth, partitioning of assimilate gradually changed towards the storage root, and was not a sudden change as proposed by Green et al. (1986). Therefore the data of Milford et al. (1988) provided further support for the theory that initiation of storage organ growth in sugar beet was not under obligative environmental control.

Initiation and activity of sinks is also dependent on the supply of photoassimilates at an earlier ontogenetic stage (Gifford and Evans, 1981). Increased sink initiation and activity through increased supply of photoassimilates can result from environmental manipulation

of photosynthetic activity (Gifford, 1977), or removal of competitive sinks for assimilates (Biran et al., 1974). While tuber initiation in potato is primarily dependent on photoperiod, this response was ameliorated by both the temperature and irradiance (Borah and Milthorpe, 1963; Menzel, 1985). The inhibitory effects of long days or high temperatures were exaggerated by the reduced assimilate supply at low irradiance, resulting in a greater proportion of dry weight being allocated to the shoot.

During active growth, partitioning of assimilates to storage organs may be influenced by environmental parameters such as PPF and temperature. Reduced partitioning of assimilates to storage organs under low irradiance (presumably PPF) has been reported with cassava (Boerboom, 1978), potato (Menzel, 1985), taro and tannia (Caesar, 1980). However, this partitioning was not influenced by irradiance where the storage organ consisted of a tap root (e.g., sugar beet; Terry, 1968), but appeared to be primarily a function of a time-dependent allometric relationship between the shoot and storage root (Barnes, 1979). For most of the growing season an allometric relationship was also determined between total plant growth and storage root growth in cassava (Boerboom, 1978). While in most cases the reduced partitioning to the storage organ under low PPF regimes resulted in reduced final yield (Boerboom, 1978; Menzel, 1985; Caesar, 1980), members of the *Araceae* have been reported to be shade tolerant, achieving greater storage organ yield under low PPF regimes. From field experiments Valenzuela et al. (1991) reported increased corm yield in tannia when the daily integrated PPF was reduced from 35 $mol \cdot m^{-2} \cdot day^{-1}$ (i.e., full sun) to 17.5 or 10.5 $mol \cdot m^{-2} \cdot day^{-1}$ (i.e., 50% and 70% shading, respectively). Under the lower PPF regimes the increased total plant and corm yield was attributed to more light interception through increased leaf canopy development, and hence increased leaf area index. This occurred even though net photosynthetic rate was less under the lower PPF regimes than under the high PPF regime (Valenzuela et al., 1991). In contrast Caesar (1980) reported increased total plant dry weight but reduced corm yield when plants of taro and tannia were grown under 80% shade (daily integrated PPF $\approx$ 12 $mol \cdot m^{-2} \cdot day^{-1}$) compared with full sun. Since total plant dry weight was greater under the low PPF regime, it is unlikely that this PPF regime seriously limited growth, as postulated by Valenzuela et al. (1991). However, as Valenzuela et al. (1991) did not detect differences in corm weight until 210 days after planting, it is possible the contradicting result of Caesar (1980), reported after 146 days from planting, merely reflects some temporal function, i.e., delayed corm growth but subsequent increased leaf area duration under low PPF.

While the objective of using varying PPF regimes has been to manipulate source strength, low irradiance, which has been shown to inhibit tuber growth in potato, also greatly increased the gibberellin-like substances in leaves (Woolley and Wareing, 1972). Since application of gibberellic acid to potato plants inhibited tuber growth (Menzel, 1985), and

application of the chemical growth regulators gibberellic acid, indoleacetic acid and cytokinin enhanced storage root growth in sugar beet (Das Gupta, 1972), the assumption that alteration of PPF only influences source strength may not be valid.

While total plant dry weight generally increases with increasing temperature up to some optima, partitioning of assimilates towards the storage organ declines over that same temperature range (Ulrich, 1952 & 1955; Terry, 1968; Boerboom, 1978; Keating et al., 1982). However, as a result of increased total plant dry weight at higher temperature, final storage organ yield was maximum between 17 and 24 C for sugar beet and at 24 C for cassava.

Without any evidence to support the existence of an obligative environmental trigger for initiation of tuber growth in *Zantedeschia* (refer Section 1.5.3), it is suggested that the growth and development of sugar beet may provide an adequate preliminary model for comparison. However, in contrast to the tap root of sugar beet, the tuber in *Zantedeschia* is a stem morphologically (refer Section 1.3.1). Since a functional equilibrium of assimilate partitioning exists between the morphological shoot and root of plants (Richards and Rowe, 1977; Barnes, 1979), the application of a dry matter partitioning simulation model for sugar beet, such as that developed and tested by Fick et al. (1973), would therefore have to take cognizance of this potential difference in assimilate partitioning. With this in mind it is pertinent to summarise the hypotheses upon which the model of Fick et al. (1973) is based, i.e.;

(1) that storage beet growth and sugar accumulation requires an excess of photoassimilates beyond the needs for respiration and for growth of tops and fibrous roots (Ulrich, 1952 & 1955).

(2) that the relative effects of reserves on sink activity differs for various sinks as a function of reserve level, i.e., intersink competition increases as the reserve supply decreases, and the development of the storage structure, as well as the filling of the structure with sugars, all receive a lower priority than foliage development (Terry, 1968).

(3) that a functional equilibrium exists between partitioning of assimilates and relative growth rates of the shoot and root (Barnes, 1979).

(4) that respiration rates, growth and activity of sinks are functions of the amount of tissue capable of growth and of solar radiation (presumably PPF and temperature; Terry, 1968).

Preliminary studies on *Zantedeschia* suggested that leaf area development and duration are important in determining tuber growth (Funnell and MacKay, 1987; Warrington and Southward, 1989), but no detailed analysis of dry matter partitioning and how it might be influenced by temperature and PPF have been made. Therefore the objective of this study was to examine the influence of temperature and PPF on growth of tubers of *Zantedeschia* 'Best Gold' in terms of timing and extent of assimilate partitioning.

4.2 Materials and Methods

Data presented in this chapter were derived from the experiment described previously (refer Section 3.2). Cultural procedures (refer Section 3.2.1) and environmental conditions (refer Section 3.2.2) were the same. Tubers from each treatment were harvested throughout development and dried to constant weight. A $\log_e$ transformation of the dry weight data was used to meet the statistical assumption of homogeneity of variance throughout the period of study (Causton and Venus, 1981). Transformed data, collected during the period of increasing tuber dry weight, were fitted to the Gompertz function, equation (4.2) (Causton and Venus, 1981), utilizing the non-linear regression parameter estimation procedure of the Statistical Analysis System, i.e., SAS (SAS Institute Inc., Cary, N.C.). The Gompertz function was chosen over other sigmoidal growth functions due to the derivation of more biologically relevant parameters, its wide acceptability by other researchers (Causton and Venus, 1981; Hunt, 1982), and its ability to fit the current data-set with small mean square error values. A constant was added to all transformed data prior to curve fitting to eliminate negative values. Because the Gompertz function is asymptotic, the magnitude of the added constant influences the calculated value of β_T. Hence the addition of a constant to all values prior to curve fitting eliminated any value of subsequent interpretation of differences in the value of β_T.

$$\log_e T = \alpha_T \cdot e^{(-e^{(\beta_T - \kappa_T \cdot t)})} \qquad (4.2)$$

where;

$\log_e T$ = $\log_e$ transformed value of tuber dry weight ($\log_e$ g)
α_T = upper asymptote of tuber dry weight ($\log_e$ g)
β_T = a measure of the starting size of tuber dry weight ($\log_e$ g)
κ_T = rate constant of tuber dry weight as a function of size ($g \cdot g^{-1} \cdot g^{-1}$)
t = time (days)
e = the base of natural logarithms

During the period of increasing tuber dry weight, the tuber relative growth rate (RGR_T), as a function of time, was calculated using the first differential of equation (4.2) i.e., equation (4.3) (Causton and Venus, 1981).

$$RGR_T = \alpha_T \cdot \kappa_T \cdot e^{(\beta_T - \kappa_T \cdot t)} \cdot e^{(-e^{(\beta_T - \kappa_T \cdot t)})} \qquad (4.3)$$

Tuber weight partitioning (TWP; equation (4.1)), i.e., the proportion of the daily increment in total plant dry weight partitioned into tuber growth, was calculated using equation (4.4) utilizing derived formulae from equations (3.4), (3.7), (4.2) and (4.3).

$$TWP = \frac{RGR_{T_{(t)}} \times e^{(\log_e T_{(t)})}}{RGR_{W_{(t)}} \times e^{(\log_e W_{(t)})}} \qquad (4.4)$$

where;

W = total plant dry weight (g)

The value of t used in the calculation of TWP was the time (days from planting) of maximum RGR_T, which corresponds to the inflection point of the Gompertz curve, i.e., equation (4.5) (Causton and Venus, 1981).

$$t = \frac{\beta_T}{\kappa_T} \qquad (4.5)$$

The time of commencement of tuber growth was estimated as the minimum of the fitted quadratic function describing changes in $\log_e$ transformed tuber dry weight data from the time of placement in the treatments and the initial increase in tuber dry weight.

Partial correlations between parameters (Table 4.1) were examined in the formulation of possible multiple regression functions, which might offer a mechanistic explanation of the results. Model formulation used the Stepwise model selection method of SAS.

Table 4.1

Parameters examined in the development of a mechanistic multiple regression model.

RGR_T	Tuber relative growth rate
TWP	Tuber weight partitioning
LWP	Leaf weight partitioning
NAR	Net assimilation rate
RWP	Root weight partitioning
RLAER	Relative leaf area expansion rate
RLWR	Relative leaf weight rate

4.3 Results

4.3.1 Overview

Changes in the dry weights of various components of the plant followed a sigmoidal pattern, with the commencement of net tuber growth occurring later than that of the other plant components (Figure 4.1). Higher temperatures and higher PPF resulted in the chronologically earlier commencement of tuber growth, but the extent of the initial decline in tuber weight was greater at higher temperatures and at low PPF (Figure 4.2). In contrast, commencement of tuber growth was delayed morphologically at higher temperature; tuber growth commenced when only one fully expanded leaf was present on plants grown at 13 C compared with four expanded leaves on plants grown at 28 C (data not presented). Different PPF regimes had no effect on the morphological timing of commencement of tuber growth. While maximum tuber weight was not attained during the period of study, the estimated maximum tuber weight was greater under low PPF than high PPF regimes irrespective of temperature.

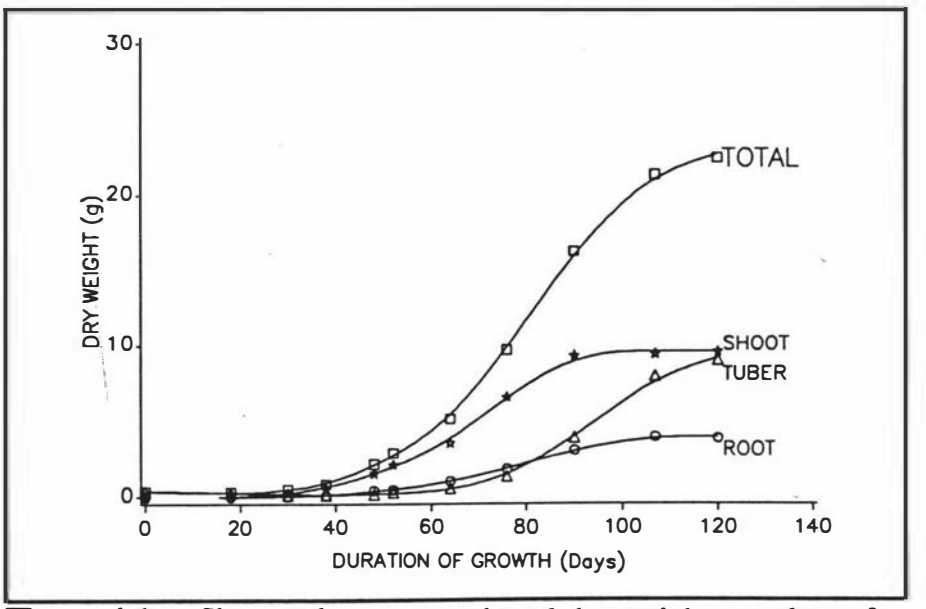

Figure 4.1 Shoot, tuber, root and total dry weight per plant of *Zantedeschia* 'Best Gold' plants grown at 25 C under high PPF. n=6 or 12.

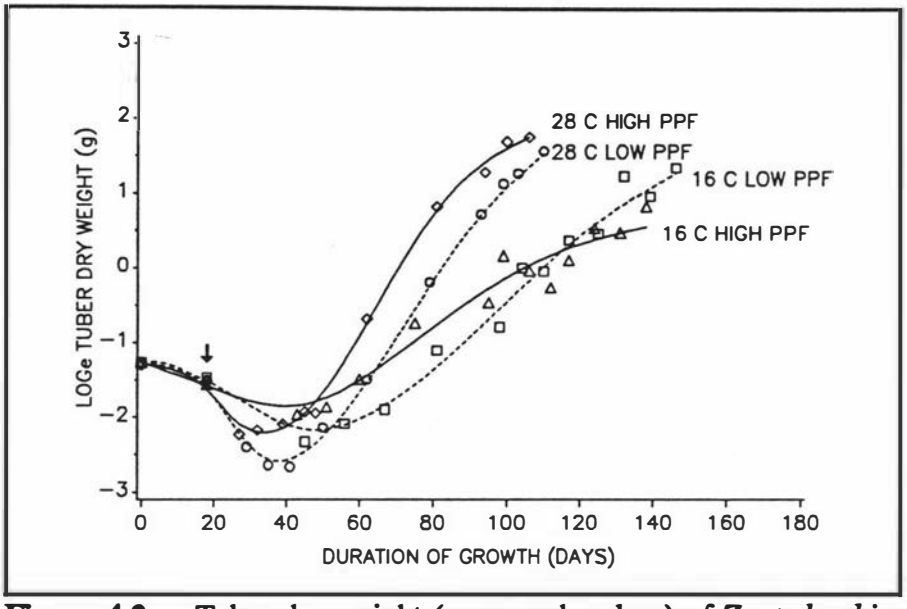

Figure 4.2 Tuber dry weight (expressed as $\log_e$) of *Zantedeschia* 'Best Gold' plants grown under high or low PPF at 16 or 28 C. ↓ indicates day of transfer to treatment environments.

4.3.2 Commencement of tuber growth

Tuber weight declined after transfer to treatment environments. Duration of this decline was longer under low PPF than under high PPF for all temperatures ($P \leq 0.05$, Figure 4.3). A negative curvilinear relationship existed between time to commencement of tuber growth (t_c) and increasing mean temperature (C) under the low PPF regime ($P \leq 0.01$, equation (4.6)). While a negative curvilinear relationship also existed under the high PPF regime between 13 and 19 C, no difference in the time to commencement of tuber growth was evident between 19 and 28 C ($P \leq 0.0001$, Figure 4.3). Plants grown under high PPF at temperatures of 19 C or greater commenced tuber growth 36 days from planting. In contrast, plants at 13 C and low PPF commenced tuber growth 52 days after planting (Figure 4.3).

$$t_c = 0.05 \cdot C^2 - 2.7 \cdot C + 79 \qquad (r^2 = 0.98,^{**}) \qquad (4.6)$$

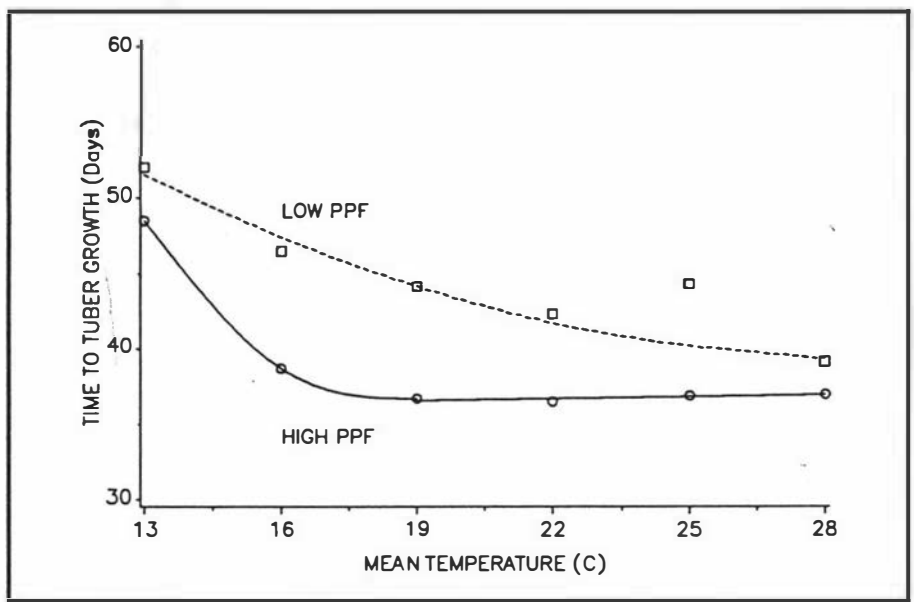

Figure 4.3 Time to commencement of tuber growth (t_c) as a function of temperature for *Zantedeschia* 'Best Gold', grown under high and low PPF regimes.

There was a greater loss in tuber weight prior to the commencement of tuber growth in plants grown under low PPF than under high PPF at all temperatures ($P \leq 0.05$, Figure 4.4). A positive curvilinear relationship existed between the proportion (%) tuber weight loss at the commencement of tuber growth ($T_{\%}$) and increasing temperature (C) under low ($P \leq 0.10$, equation (4.7)) and high ($P \leq 0.01$, equation (4.8)) PPF regimes.

$$\text{(Low PPF)} \qquad T_{\%} = -0.09 \cdot C^2 + 5 \cdot C + 9 \qquad (r^2 = 0.79,^{*}) \qquad (4.7)$$

$$\text{(High PPF)} \qquad T_{\%} = -0.03 \cdot C^2 + 3 \cdot C + 15 \qquad (r^2 = 0.95,^{**}) \qquad (4.8)$$

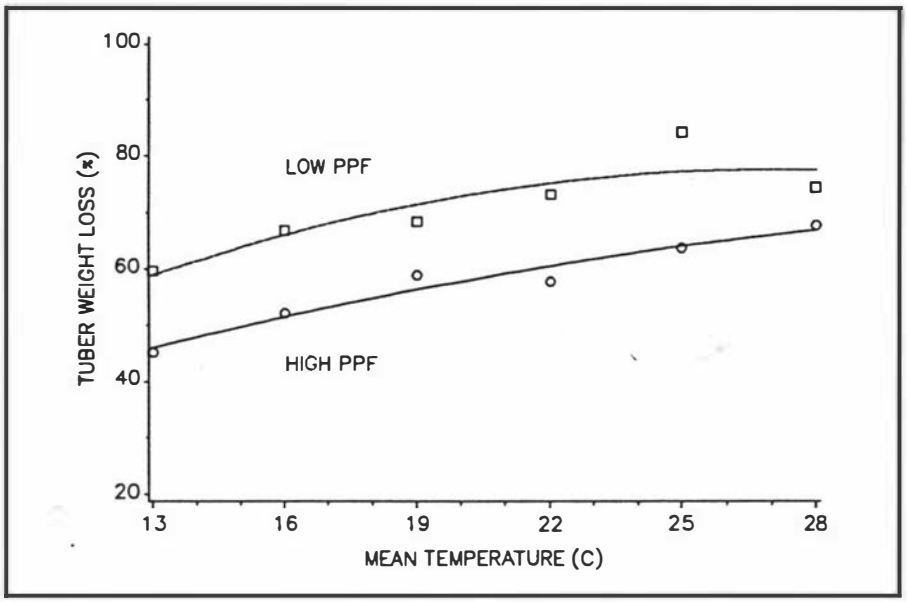

Figure 4.4 Extent of tuber weight loss ($T_{\%}$) at the time of commencement of tuber growth, as a function of temperature, for *Zantedeschia* 'Best Gold', grown under high and low PPF regimes.

4.3.3 Curve fitting of tuber growth

Under all treatment regimes $\log_e$ tuber weight followed a sigmoidal pattern of growth, with a family of Gompertz functions adequately describing the progression of tuber weight ($\log_e$ transformed) during the period of its increase (Figure 4.5 and Figure 4.6, Table 4.2).

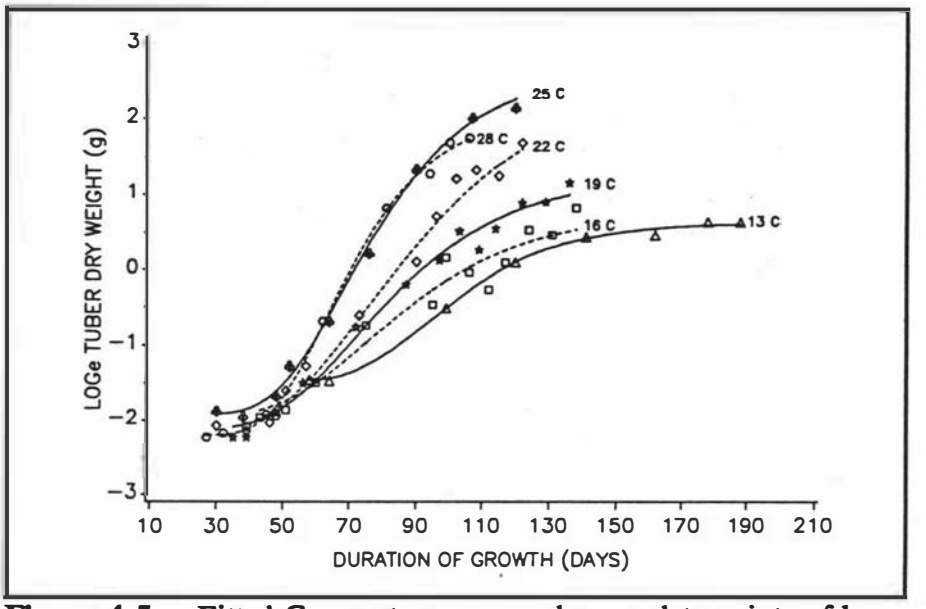

Figure 4.5 Fitted Gompertz curves and mean data points of $\log_e$ tuber dry weight (minus constant) as a function of time, for *Zantedeschia* 'Best Gold' at a range of temperatures, under high PPF regime.

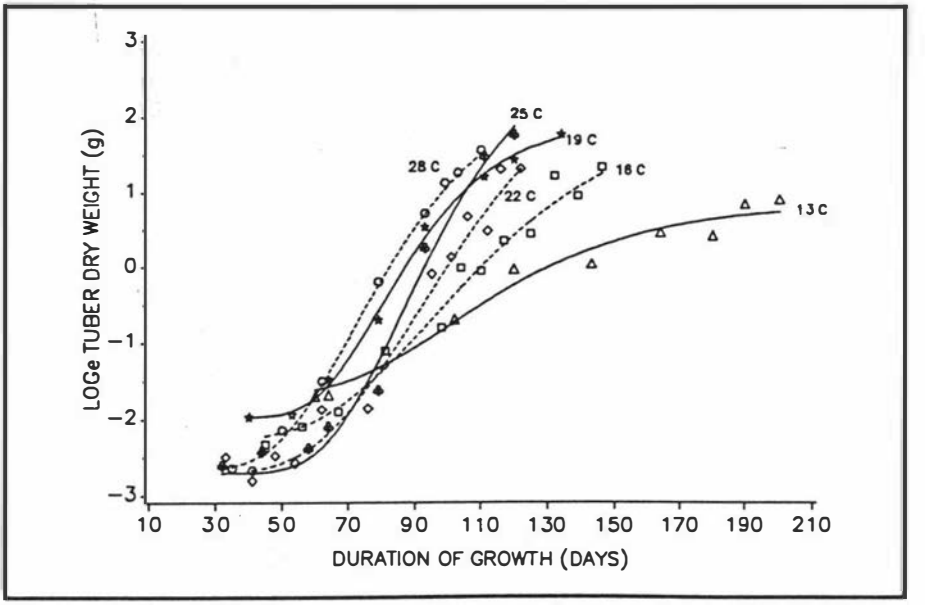

Figure 4.6 Fitted Gompertz curves and mean data points of $\log_e$ tuber dry weight (minus constant) as a function of time, for *Zantedeschia* 'Best Gold' at a range of temperatures, under a low PPF regime.

Table 4.2

Nonlinear least-squares parameter estimates, associated asymptotic standard error (s.e.), and mean square error values, from fitting the Gompertz function to $\log_e$ transformed tuber dry weight data for *Zantedeschia* 'Best Gold' grown at a range of temperatures, under high and low PPF regimes. The PPF treatments at 19 C were repeated in two plantings, i.e., (1st) and (2nd).

Mean temperature (C)	PPF	α_T #	s.e.	β_T	s.e.	κ_T	s.e.	mean square error
13	High	2.12	0.09	4.52	1.18	0.048	0.012	0.17
	Low	2.55	0.17	3.13	0.70	0.031	0.007	0.29
16	High	2.79	0.25	2.80	0.41	0.037	0.007	0.19
	Low	4.49	0.61	2.79	0.40	0.029	0.006	0.29
19 (1st)	High	3.24	0.09	3.23	0.28	0.059	0.005	0.09
	Low	3.97	0.16	4.07	0.42	0.051	0.006	0.13
19 (2nd)	High	3.40	0.20	2.89	0.32	0.040	0.005	0.15
	Low	4.54	0.21	3.32	0.34	0.041	0.005	0.18
22	High	4.64	0.37	2.66	0.22	0.035	0.004	0.13
	Low	5.89	1.05	2.93	0.40	0.032	0.007	0.25
25	High	4.60	0.20	3.28	0.24	0.048	0.004	0.11
	Low	5.82	0.47	3.72	0.38	0.043	0.005	0.16
28	High	4.30	0.20	3.66	0.34	0.058	0.006	0.10
	Low	5.14	0.43	3.03	0.30	0.042	0.006	0.12

Values of α_T presented are those resulting from addition of a constant to avoid negative values of $\log_e$ transformed data.

Differences between the fitted curves of tuber weight were principally the result of differences in the estimated maximum tuber dry weight attained (α_T). There was a cubic increase of estimated maximum tuber weight (α_T) with increasing temperature (C) for both PPF regimes ($P \leq 0.001$). Under the high PPF regime this relationship with temperature was highly significant ($P \leq 0.001$, equation (4.9), Figure 4.7), but the relationship was significant under the low PPF regime only after exclusion of the value estimated for 16 C ($P \leq 0.05$, equation (4.10), Figure 4.7).

(High PPF)

$$\alpha_T = -0.0035 \cdot C^3 + 0.02 \cdot C^2 - 3.7 \cdot C + 22 \qquad (r^2 = 0.98,^{***}) \qquad (4.9)$$

(Low PPF)

$$\alpha_T = -0.003 \cdot C^3 + 0.19 \cdot C^2 - 3 \cdot C + 19 \qquad (r^2 = 0.98,^{**}) \qquad (4.10)$$

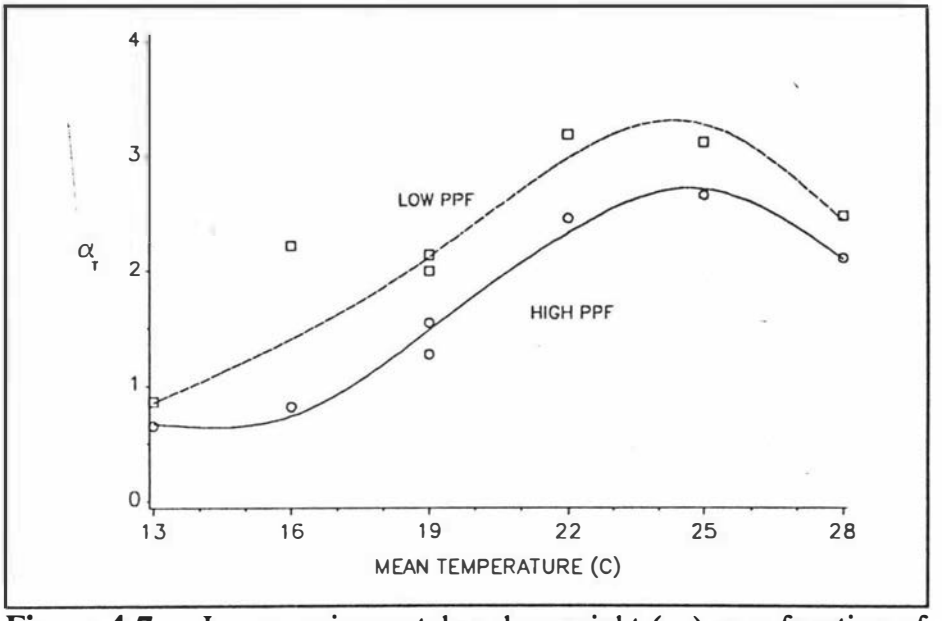

Figure 4.7 Log_e maximum tuber dry weight (α_T) as a function of temperature, for *Zantedeschia* 'Best Gold' under high and low PPF regimes. (nb., value of added constant removed)

The value of the parameters β_T (an estimate of tuber dry weight at day 0) and κ_T (rate at which RGR_T declined as a function of tuber size) were not influenced by temperature nor PPF regime. Mean values for β_T and κ_T across all environments were 3.3 ± 0.1 and 0.042 ± 0.002, respectively. However, as noted in Section 3.2, the need to add a

constant to all values prior to curve fitting eliminated any value of subsequent interpretation of differences in the value of β_T.

4.3.4 Maximum tuber weight

Estimated maximum tuber weight was greater under the low PPF regime than under the high PPF regime at all temperatures ($P \leq 0.001$, Figure 4.7). From equation (4.10) maximum α_T [3.29 (i.e., 26.83 g) $\pm$ 0.1] was attained under the low PPF regime at 24.5 $\pm$ 0.1 C. This was more than 1.7 times that attained under the high PPF regime [2.72 (i.e., 15.17 g) $\pm$ 0.08] at the same temperature. The optimal temperature under the high PPF regime was estimated to be 24.9 $\pm$ 0.1 C (equation (4.9)). While the optimal temperatures under each PPF regime were significantly different, the magnitude of the differences can not be considered to be of biological significance. α_T decreased at 28 C under both PPF regimes (Figure 4.7).

4.3.5 Base temperature for tuber growth

At the time tuber relative growth rate (RGR_T) was maximal, i.e., inflection point of the fitted tuber dry weight Gompertz curve, a positive linear relationship existed between RGR_T and temperature up to 28 C under the high PPF regime, and up to 25 C under the low PPF regime (Figure 4.8). A reduction in the value of RGR_T was noted at 28 C under the low PPF regime and this data was therefore not included in the determination of the linear regressions. Examination of the slopes of the linear regressions for homogeneity (Zar, 1984) did not detect differences between PPF regimes ($P \leq 0.05$), but the point of interception of the Y-axis of the two functions were different ($P \leq 0.05$). Extrapolation of the two linear functions allowed an estimation of the base temperature for tuber growth of 4.8 $\pm$ 2.7 C under high PPF ($P \leq 0.001$, equation (4.11)), and 6.1 $\pm$ 2.6 C under low PPF ($P \leq 0.01$, equation (4.12)). Pooling data from both PPF regimes resulted in an estimated base temperature for tuber growth of 4.6 $\pm$ 1.1 C.

$$\text{(High PPF)} \quad RGR_T = 0.0039 \cdot C - 0.018 \quad (r^2 = 0.93,^{***}) \qquad (4.11)$$

$$\text{(Low PPF)} \quad RGR_T = 0.0049 \cdot C - 0.03 \quad (r^2 = 0.90,^{**}) \qquad (4.12)$$

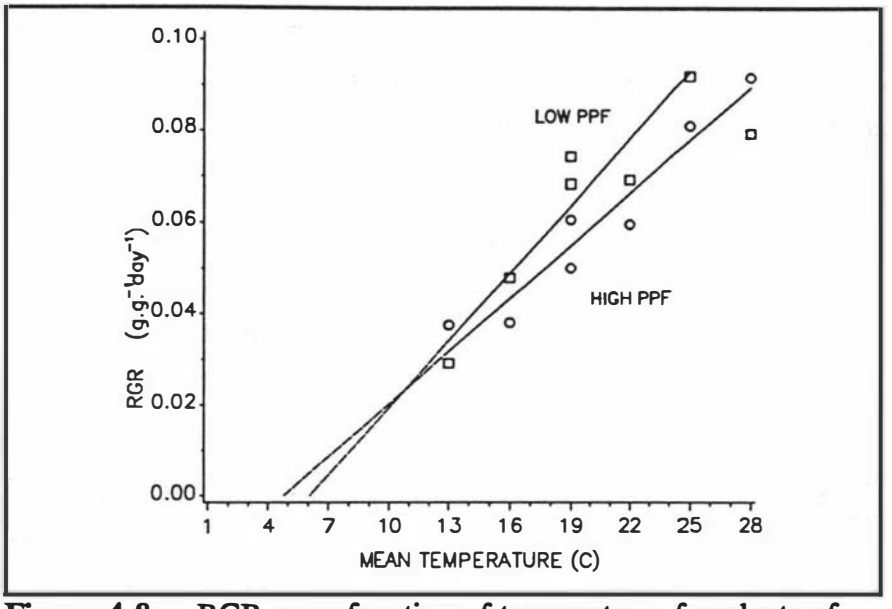

Figure 4.8 RGR_T as a function of temperature, for plants of *Zantedeschia* 'Best Gold', grown under high and low PPF regimes. (N.B., line for low PPF excludes 28 C)

4.3.6 Tuber and leaf weight partitioning

From the time of commencement of tuber growth, the proportion of the daily increment in total plant weight that was partitioned to the tuber, i.e., tuber weight partitioning (TWP), increased throughout the period of observation (Figure 4.9 and Figure 4.10). Use of TWP proved more sensitive in detecting the early stages of partitioning dry weight to the tuber than the frequently used harvest index. Harvest index, i.e. tuber dry weight as a fraction of total plant dry weight, continued to decline until well after the commencement of tuber growth, e.g. at 28 C under high PPF the harvest index did not increase until more than 45 days after planting (data not presented). In contrast, under these environmental conditions an increase in tuber dry weight was clearly detected after 36 days from planting (Figure 4.3).

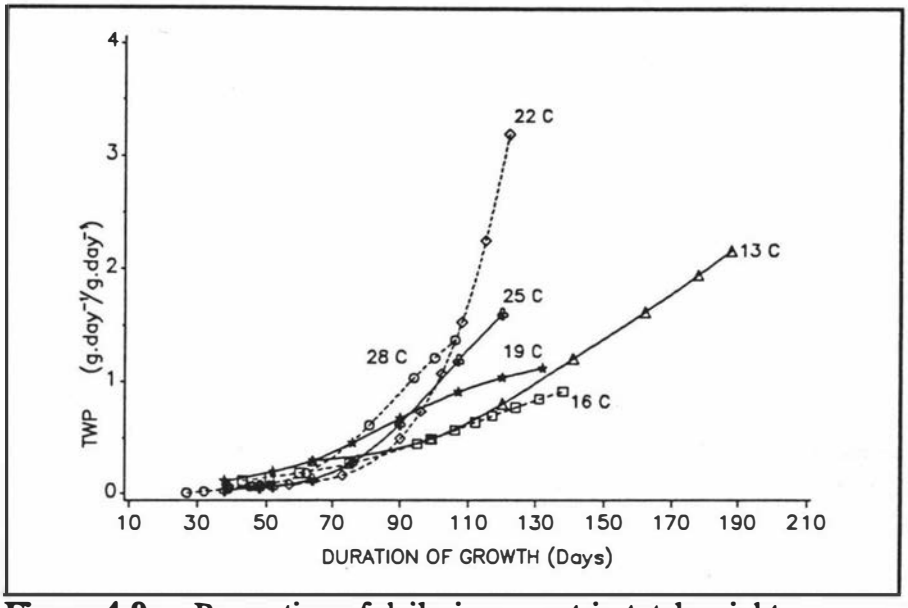

Figure 4.9 Proportion of daily increment in total weight partitioned to the tuber (TWP) as a function of time, for *Zantedeschia* 'Best Gold', at a range of temperatures, under a high PPF regime.

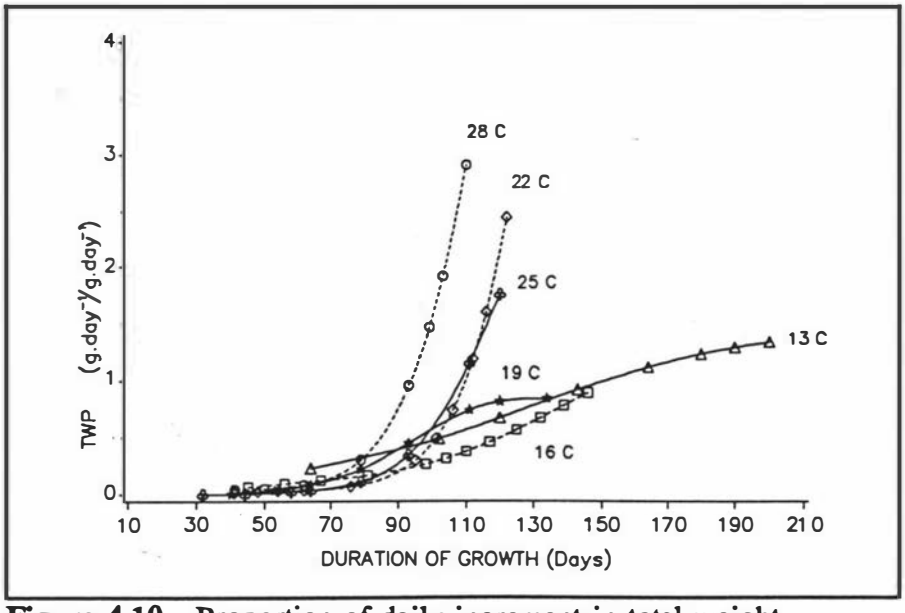

Figure 4.10 Proportion of daily increment in total weight partitioned to the tuber (TWP) as a function of time, for *Zantedeschia* 'Best Gold', at a range of temperatures, under a low PPF regime.

The magnitude of TWP increased earlier, both chronologically and morphologically (i.e., leaf number; data not presented), under the high PPF regime than under the low PPF regime at all temperatures (Figure 4.11). However, at both 25 C and 28 C this earlier onset of increased TWP under high PPF was subsequently surpassed, with the TWP under the low PPF regime exceeding that achieved under high PPF after approximately 90 and 110 days from planting, respectively.

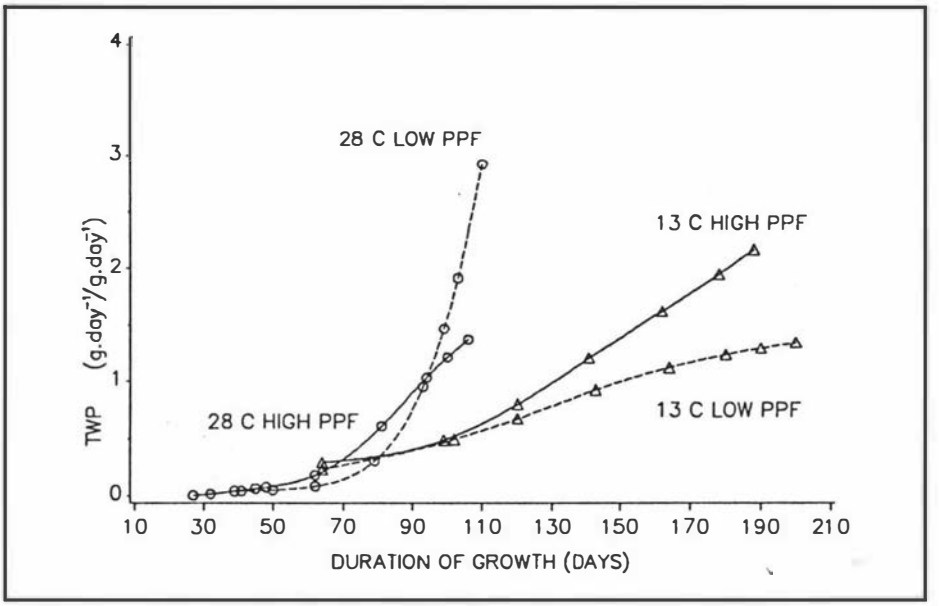

Figure 4.11 Proportion of daily increment in total weight partitioned to tuber (TWP), as a function of time, for *Zantedeschia* 'Best Gold' grown under high and low PPF regimes at 13 or 28 C.

At the time tuber relative growth rate (RGR_T) was maximal, i.e., inflection point of the fitted tuber dry weight Gompertz curve, a curvilinear decline occurred in the value of the TWP with increasing temperature ($P \leq 0.0001$, equation (4.13), Figure 4.12), with no difference being detected between PPF regimes. Similarly, a curvilinear decline in the proportion of the daily increment in total plant weight that was partitioned to leaf area, i.e., leaf weight partitioning (LWP), with increasing temperatures of 16 C and above was determined under the high PPF regime ($P \leq 0.1$, equation (4.14), Figure 4.13). A reduction in the value of LWP was noted at 13 C under the high PPF regime. In contrast to the high PPF regime, under the low PPF regime there was a linear decline in the value of the LWP with increasing temperature ($P \leq 0.1$, equation (4.15), Figure 4.13).

$$TWP = 0.0016 \cdot C^2 - 0.08 \cdot C + 1.2 \qquad (r^2 = 0.84,^{***}) \qquad (4.13)$$

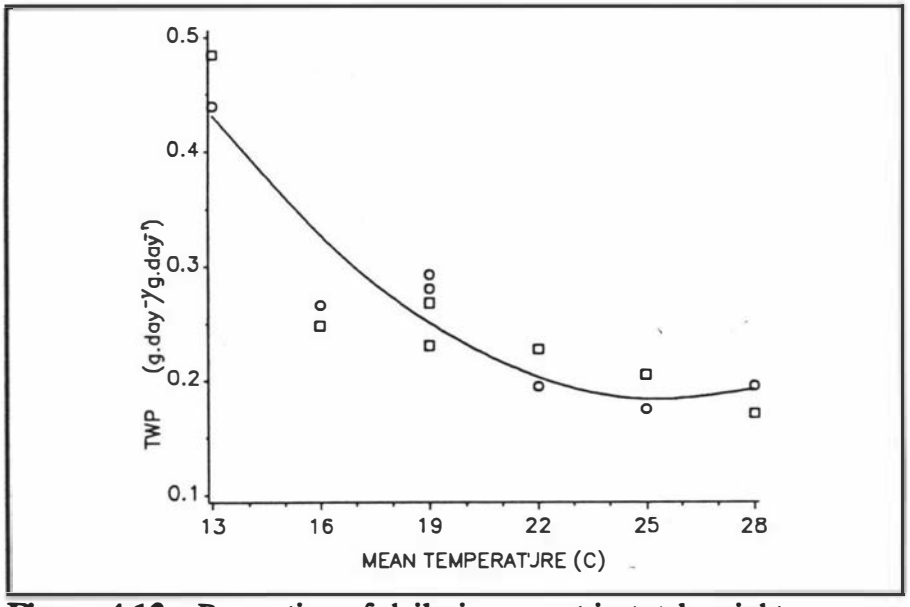

Figure 4.12 Proportion of daily increment in total weight partitioned to the tuber (TWP), as a function of temperature, for *Zantedeschia* 'Best Gold' under high (-○-) and low (-□-) PPF regimes.

$$\text{(High PPF)} \quad LWP = 0.001 \cdot C^2 - 0.06 \cdot C + 0.9 \qquad (r^2 = 0.78,^{*}) \qquad (4.14)$$

$$\text{(Low PPF)} \quad LWP = -0.002 \cdot C + 0.19 \qquad (r^2 = 0.47,^{*}) \qquad (4.15)$$

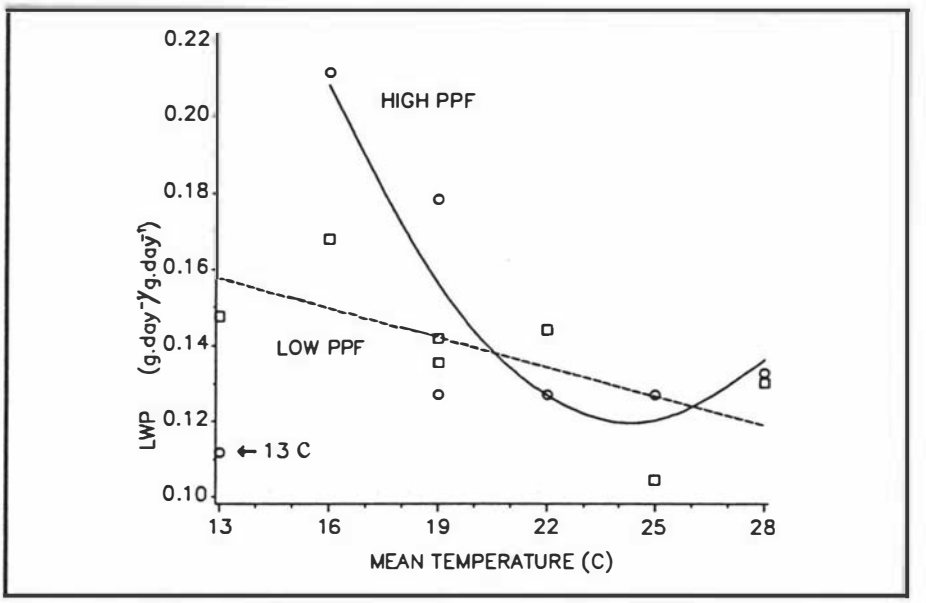

Figure 4.13 Proportion of daily increment in weight partitioned to leaf (LWP) as a function of temperature, for *Zantedeschia* 'Best Gold' under high (-○-) and low (-□-) PPF regimes. (N.B., line for high PPF excludes 13 C)

4.3.7 Net assimilation rate

A positive linear relationship existed between NAR and increasing temperature at both PPF regimes, but the magnitude of the slope under high PPF was more than four times greater ($P \leq 0.01$, equation (4.16), Figure 4.14) than that under low PPF ($P \leq 0.05$, equation (4.17), Figure 4.14). The relationship between NAR and temperature under high PPF was only linear at temperatures of 16 C and greater.

$$\text{(High PPF)} \quad NAR = 0.6 \cdot C - 4 \qquad (r^2 = 0.88,^{**}) \qquad (4.16)$$

$$\text{(Low PPF)} \quad NAR = 0.14 \cdot C + 3.8 \qquad (r^2 = 0.65,^{*}) \qquad (4.17)$$

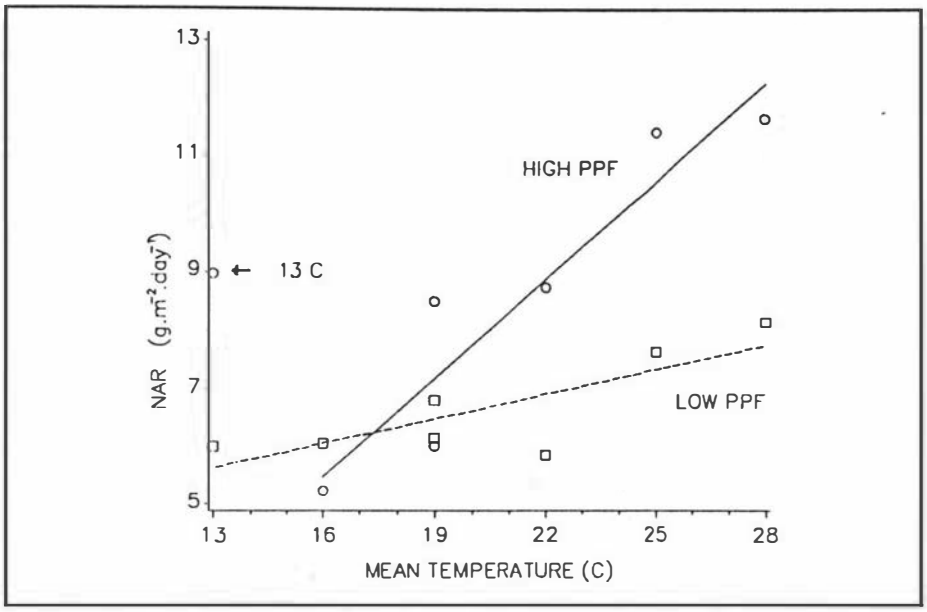

Figure 4.14 Net assimilation rate (NAR) as a function of temperature, for plants of *Zantedeschia* 'Best Gold', grown under high (-○-) and low (-□-) PPF regimes. (N.B., line for high PPF regime excludes 13 C)

4.3.8 Relationships between derived parameters

Examination of the partial correlations between the parameters (Table 4.1), at the time tuber relative growth rate (RGR_T) was maximal, i.e., inflection point of the fitted tuber dry weight Gompertz curve, indicated multicolinearity between parameters. A reduced range of parameters (Table 4.3) were subsequently used for development of a possible model. Across all treatments RGR_T was negatively correlated with TWP and LWP, and positively correlated with NAR (Table 4.3). Standardized multiple regression coefficients (beta coefficients) for each of the three parameters indicated the relative contribution made by each in determining RGR_T, independent of the others and unitless, i.e., TWP = -0.70, LWP = -0.31, and NAR = 0.19. However, NAR did not meet the 0.15 significance level for entry into the final model (equation (4.18)).

$$RGR_T = -0.16 \cdot TWP - 0.3 \cdot LWP + 0.14 \qquad (r^2 = 0.76,^{***}) \qquad (4.18)$$

Table 4.3

Partial correlation matrix between TWP, LWP, NAR and RGR_T at the inflection point of the Gompertz fit of tuber dry weight curves of *Zantedeschia* 'Best Gold' grown under a range of environments.

	RGR_T	TWP	LWP	NAR
RGR_T	1.00	-0.82	-0.52	0.33
TWP	-0.82	1.00	-0.45	0.11
LWP	-0.52	-0.45	1.00	-0.17
NAR	0.33	0.11	-0.17	1.00

A negative curvilinear relationship existed between RGR_T and TWP across both PPF regimes ($P \leq 0.01$, equation (4.19), Figure 4.15). However, under the low PPF regime there was a high partial correlation between TWP and LWP ($r = -0.96$). In light of this, two possible models were subsequently developed under the low PPF regime. Firstly, at temperatures of 16 C and greater a negative linear relationship was determined between RGR_T and LWP ($P \leq 0.001$, equation (4.20), Figure 4.16). The further addition of NAR to this function did not improve the model.

$$RGR_T = 0.5 \cdot TWP^2 - 0.5 \cdot TWP + 0.16 \qquad (r^2 = 0.66,^{**}) \qquad (4.19)$$

$$RGR_T = -0.69 \cdot LWP + 0.167 \qquad (r^2 = 0.97,^{***}) \qquad (4.20)$$

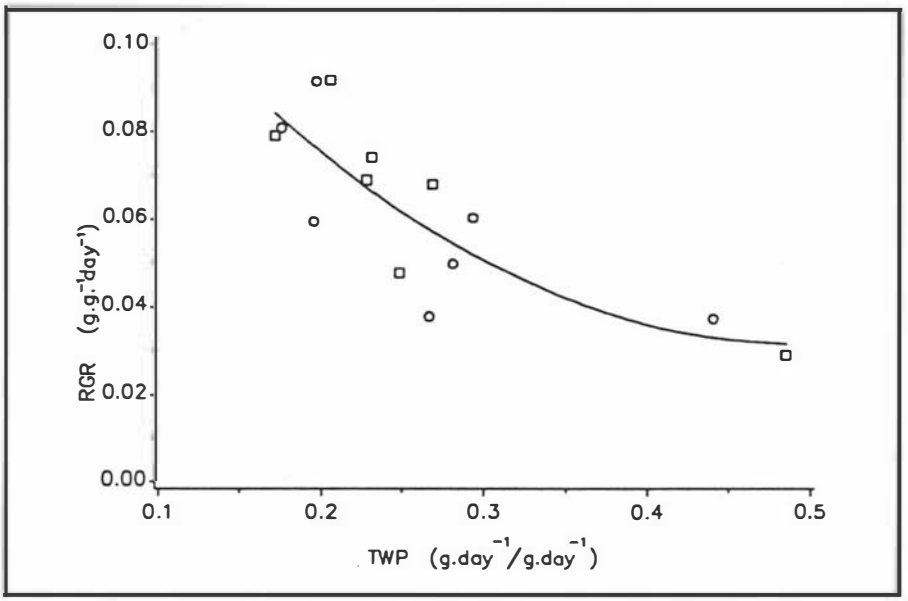

Figure 4.15 Relative growth rate of the tuber (RGR_T) as a function of daily partitioning to the tuber (TWP) for *Zantedeschia* 'Best Gold' grown under high (-○-) and low (-□-) PPF regimes at all temperatures.

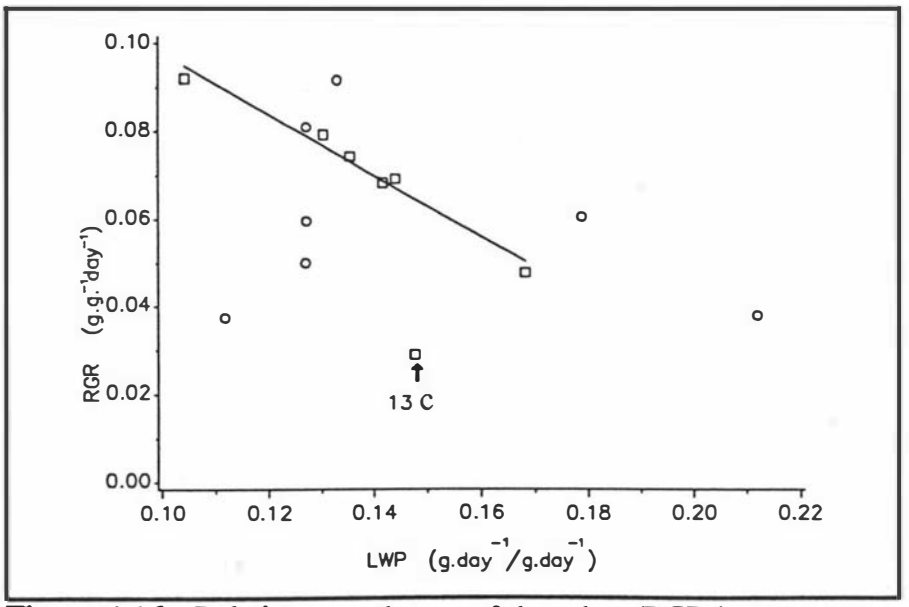

Figure 4.16 Relative growth rate of the tuber (RGR_T) as a function of daily partitioning to the leaf (LWP) under high (-○-) and low (-□-) PPF regimes, at all temperatures. Line is for low PPF regime at temperatures ≥ 16 C.

Secondly as an alternative model a negative linear relationship was determined between RGR_T and TWP under the low PPF regime at temperatures of 16 C and greater ($P \leq 0.01$, equation (4.21)). As with the previous model the addition of NAR did not significantly improve the model.

$$RGR_T = -0.17 \cdot TWP + 0.11 \qquad (r^2 = 0.73,^{**}) \qquad (4.21)$$

In contrast, under the high PPF regime, there was no significant correlation between RGR_T and LWP (Figure 4.16), but a positive linear relationship was determined between RGR_T and NAR at temperatures of 16 C and greater ($P \leq 0.001$, equation (4.22), Figure 4.17). No relationship was detected between RGR_T and NAR under the low PPF regime treatments.

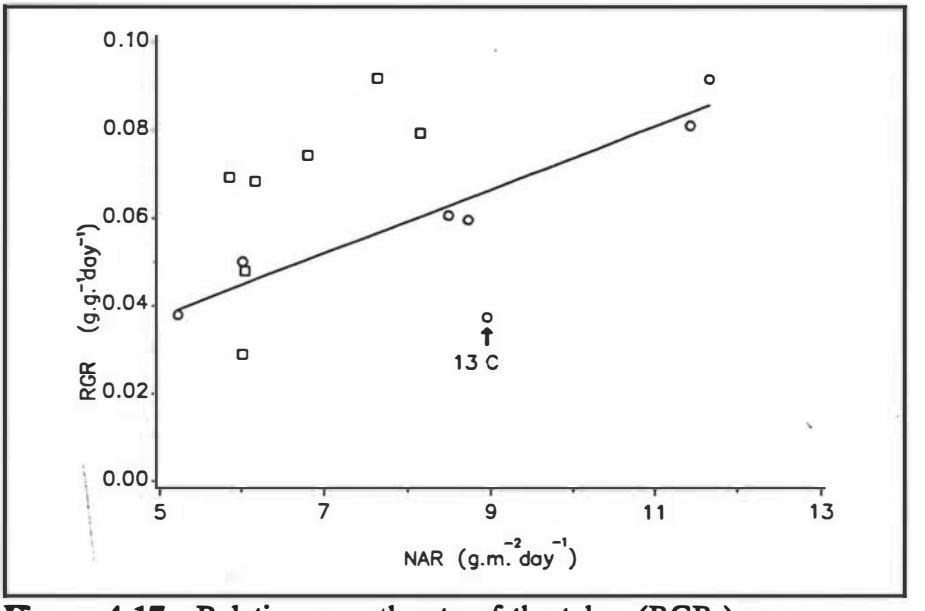

Figure 4.17 Relative growth rate of the tuber (RGR_T) as a function of net assimilation rate (NAR) under high (-○-) and low (-□-) PPF regimes, at all temperatures. Line is for high PPF regime at temperatures ≥ 16 C.

$$RGR_T = 0.0073 \cdot NAR + 0.001 \qquad (r^2 = 0.95,^{***}) \qquad (4.22)$$

The further addition of TWP to the model improved the fit and accounted for the additional variability of RGR_T at 13 C under high PPF ($P \leq 0.01$, equation (4.23)). However, the addition of LWP or RLWR did not meet the 0.15 significance level for entry into the model.

$$RGR_T = 0.006 \cdot NAR - 0.11 \cdot TWP + 0.04 \qquad (r^2 = 0.91,^{**}) \qquad (4.23)$$

Under the high PPF regime the beta coefficients for each of the parameters in the model indicated the relative contribution made by each in determining RGR_T, independent of the other, i.e., NAR = 0.56 and TWP = -0.53.

4.4 Discussion

Tuber growth occurred under all environmental regimes tested (Figure 4.5 and Figure 4.6). Even though at planting all plants possessed tubers, and not all possible environmental parameters were examined, the data presented here did not support the hypothesis of an obligative environmental trigger for the commencement of tuber growth. The proportion of the daily increase in total plant dry weight partitioned to the tuber (TWP) followed a gradual increase with time (Figure 4.9 and Figure 4.10). In contrast, an abrupt change would have been expected if environmentally determined induction was involved (Milford et al., 1988). With increasing temperature the onset of tuber growth was chronologically earlier (equation (4.6), Figure 4.3) but delayed morphologically. The additional delay and extent of tuber weight loss prior to tuber growth under the low PPF regime, suggested intersink competition rather than the existence of an obligative environmental trigger for the commencement of tuber growth. With some crops, where evidence exists for environmental triggers, e.g. short photoperiod promotion of tuberization in dahlia, tuberization occurred regardless of photoperiod once competing sinks were removed (Biran et al., 1974). In *Zantedeschia*, respiration, root growth and leaf canopy development, can be viewed as potential competing sinks for assimilates. This scenario of a non-obligative environmental trigger for the commencement of tuber growth in *Zantedeschia* is, therefore, similar to that found in storage organ development of sugar beet (Fick et al., 1973; Rapoport and Loomis, 1986), and is in contrast to the obligative environmental requirements that exist with crops such as potato, onion, and tulip (Garner and Allard, 1923; Niimi, 1978; Ewing, 1987).

The strong negative relationship between RGR_T and TWP under both PPF regimes at the time of maximum RGR_T (equation (4.19), Figure 4.15) is indicative of the tuber, as a sink, receiving a higher priority for assimilates than other organs. This interpretation is illustrated by the increased partitioning of the daily increment in total dry weight to the tuber, as conditions for growth became less optimal (e.g. cooler temperature) (Figure 4.12). This change in partitioning to the storage organ with temperature is similar to that reported for cassava, being a negative curvilinear decline with increasing temperature, to the maximum tested, i.e., 24 C (Keating et al., 1982). Assimilate partitioning toward the storage organ in sugar beet was also increased as temperatures departed from the optimum of 24 C (Ulrich, 1952 & 1955; Terry, 1968). While at this stage of development a negative relationship also existed between LWP and increasing temperature (equations (4.14) and (4.15), Figure 4.13), the magnitude of the TWP was always greater than that of the LWP, and therefore indicates greater sink strength of the tuber over that of the leaf canopy. This increase in TWP with declining temperature was not a direct result of increased source nor sink activity as both photosynthetic rate (refer Chapter 5) and RGR_T (Figure 4.8) also declined with declining temperature. Clearly this priority of tuber growth over leaf development, at the time of maximum RGR_T, was not

evident in the early stages of tuber growth. Just as the magnitude of the TWP increased over time, so the magnitude of the LWP decreased over time. At the commencement of tuber growth the magnitude of the TWP was small, while LWP was at least 5 times greater. The time dependent nature of the TWP exhibited here with *Zantedeschia* is different from the allometric relationship found between growth rates of the shoot and root, in root crops such as sugar beet and carrot (Barnes, 1979), and total plant growth and storage root growth in cassava (Boerboom, 1978). In light of this difference, any attempts to create a predictive, deterministic model of tuber growth in *Zantedeschia* will have to include this dynamic aspect of assimilate partitioning between organs.

The strong correlation between RGR_T and LWP under the low PPF regime (equation (4.20), Figure 4.16) is indicative of the mechanism of acclimation to the low PPF regime and resultant intersink competition for limited assimilates. The mechanism of acclimation, i.e., enhanced leaf canopy development, has been introduced previously (refer Chapter 3), and is a response recorded for other crops (Boerboom, 1978; Caesar, 1980; Menzel, 1985; Valenzuela et al., 1991). Any enhancement in the relative partitioning of dry matter to one organ, e.g. leaves, will be at the expense of other organs within the same system. Hence under this low PPF regime temperatures conducive to greater tuber growth were associated with reduced daily partitioning to leaves, with an increased proportion of assimilates having already been allocated to leaf canopy development, (Figure 4.16). Interpretation of this relationship and its effect on actual tuber dry weight must also take into account the dynamic nature of the partitioning of assimilate distribution. At the time of maximum RGR_T plants grown under the low PPF regime had already established a greater leaf area than under the high PPF regime (refer Chapter 3). Therefore, while temperatures conducive to greater tuber growth were associated with reduced partitioning to leaves, the photosynthetic activity of the enhanced leaf area was able to more than compensate for the low PPF conditions. As a net result greater values of RGR_T were attained under the low PPF rather than high PPF regimes at most temperatures (equations (4.11) and (4.12), Figure 4.8).

The reduced RGR_T at 28 C under the low PPF regime (Figure 4.8) was associated with an increase in the LWP from that obtained at 25 C. Since a reduction in RGR_T at 28 C did not occur under the high PPF regime, it is suggested that source limitation was the primary determinant of this reduction in RGR_T under the low PPF regime. This source limitation may have resulted from respiration having a greater priority for available assimilates than either tuber or leaf growth (Ulrich, 1952 & 1955). The higher priority of respiration has been suggested and utilized in assimilate partitioning simulation models (Fick et al., 1973). The failure of the 13 C environment to illustrate this correlation between RGR_T and LWP was presumably not as a result of reduced photosynthetic capacity or leaf canopy development, as neither the addition of NAR nor RLAER significantly improved the

model. At 13 C the value of the TWP diverged from the negative linear relationship between RGR_T and TWP that existed at temperatures of 16 C and greater (i.e., equation (4.19) compared with equation (4.21)). At the same time the value of the LWP attained was less than that predicted from equation (4.20). While the presence of multicolinearity between RGR_T, TWP and LWP, under the low PPF regime, negates the value of interpreting a model comprising all three parameters (Zar, 1984), it is suggested that a low temperature-mediated response in assimilate partitioning may explain the aberrant result. Such an enhancement in the value of TWP might be expected at low temperatures as a survival mechanism in warm season perennial crops originating from temperate/warm-temperate climes, as it supports the theories of evolution of the geophytic storage organ as a means of avoiding periods of adverse climate (Aoba, 1976; Rees, 1984).

The strong correlation between RGR_T and NAR (equation (4.22), Figure 4.17) and the poor correlation between RGR_T and LWP (Figure 4.16), under the high PPF regime, is in contrast to the complete lack of correlation between these parameters under the low PPF regime. This contrast indicates differences in the primary mechanism of acclimation to the two PPF regimes. Under the low PPF regime tuber growth was primarily determined by increased intersink competition for assimilates resulting from the enhanced leaf canopy development. However, under the high PPF regime increased tuber growth was primarily determined by the enhanced assimilatory capacity of the leaf area present, i.e., NAR. Unit increases in NAR with increases in temperature were four times greater under the high PPF regime than under the low PPF regime, reflecting the ability of the leaf area to more effectively utilize the incoming PPF (refer Chapter 5). In addition to NAR, differences in TWP contributed a similar, albeit negative, level of determination in the final model under the high PPF regime (equation (4.23)). The poor correlation of RGR_T with LWP under the high PPF regime should not be interpreted as suggesting that the partitioning of dry matter into leaf canopy development is not important, but rather the greater response of the unit increase of NAR outweighs any significant contribution of LWP in determining tuber growth under these conditions.

The frequent breakdown of the above correlations at 13 C does not justify extrapolation of the proposed models to explain the lower predicted base temperature for tuber growth of 4.8 ± 2.7 C under the high PPF regime than the 6.1 ± 2.6 C under the low PPF regime. The predicted base temperature from pooling data from both PPF regimes did not result in significant differences between that estimated from RGR_W against temperature (refer Chapter 3). The similarity of the optimal temperatures for growth rates of the storage organ and total plant growth is in contrast to sugar beet where the optimal temperature for growth rates of the storage root was slightly lower than that for total plant growth (Terry, 1968).

While at the inflection point of the fitted dry weight curve increasing temperature was generally associated with a positive linear increase in RGR_T (Figure 4.8), final tuber weight followed a cubic function with temperature (Figure 4.7). As reported for other species, leaf area duration may also have contributed to the prediction of greater maximum tuber dry weight under the low PPF regime (Radley, 1963; Rees, 1972; Allen and Scott, 1980). However, the contribution of leaf area duration was not investigated as the duration of the current study did not permit collection of data to the cessation of leaf development. As noted in Chapter 3 under the high PPF regime, the greater rate of decline of the leaf area and leaf weight expansion rates, as a function of size, in conjunction with the earlier onset of leaf senescence, are indicative of a reduced leaf area duration compared with under the low PPF regime. The maximum leaf area attained also influences leaf area duration. Under the high PPF regime the reduced maximum leaf area estimated at 28 C from that at 25 C (refer Chapter 3, Figure 3.10) may therefore explain the reduced maximum tuber weight at 28 C (Figure 4.7). Leaf canopy development and storage organ growth were also restricted in sugar beet at a temperature of 31 C compared with 24 C (Terry, 1968). Although the response at temperatures intermediate to these have not been reported for sugar beet, it is suggested that the results presented in the current study indicate the existence of an optimal temperature for tuber growth similar to that for sugar beet storage root growth.

The greater final tuber weight under the low PPF regime may not be of relevance under all commercial horticultural situations. With financial pressure growers seek crops and/or production systems that require a short cropping period. Hence while tubers may have attained a larger size if grown at 25 C under low PPF, tuber weight under the high PPF regime would be greater than that under the low PPF regime until day 140 (i.e., 5 months of growth). Hence growers wishing to maximise tuber weight, but requiring a shorter production period, would be more likely to meet these goals by utilizing a high PPF regime. This time dependent aspect of tuber weight being greater under low PPF regimes may also explain the difference in results reported for other members of the *Araceae* e.g., taro and tannia (Caesar, 1980; Valenzuela et al., 1991). When taro was cultivated under a comparable temperature regime (22 to 32 C day) and PPF (shaded to 350 $\mu mol \cdot m^{-2} \cdot s^{-1}$) Caesar (1980) reported reduced storage organ weight under the low PPF regime after 146 days from planting. Hence, if the final harvest had been carried out later, storage organ yield may have been greater under the low PPF regime, as presented here and as also reported by Valenzuela et al. (1991).

4.4.1 Conclusions

Tuber growth in *Zantedeschia* appears to have been source limited, at least to some extent. Source limitation was expressed either in terms of:

1) enhanced intersink competition for assimilates, as occurred under the low PPF regime through enhanced leaf canopy development,

2) assimilatory capacity of the leaf canopy, i.e., NAR, as occurred under the high PPF regime.

However, at the same time, the correlation between RGR_T and TWP cannot be ignored. Whether the calculation of TWP merely provides an empirical description of dry matter distribution under source control, as is one interpretation of the correlation between TWP and LWP under low PPF regimes, or whether situations where true sink limitation exist, cannot be determined from the data presented here. Certainly with storage root crops such as sugar beet, carrot and cassava, a constant functional equilibrium exists between source and sink (Boerboom, 1978; Barnes, 1979). While such an equilibrium does not exist in *Zantedeschia*, the priority of the tuber as a sink, over other organs, indicates that situations of sink limitation may also exist. Examples of sink limited systems have been reported for other crops, e.g. ovule growth in peas where pod warming increased ovule growth and assimilate transport to the developing sinks (Williams and Williams, 1978). Similarly warming of the tap root of sugar beet increased the importation of assimilates (Geiger and Fondy, 1985). In addition, the possibility of both source and sink limitation within the same crop, but under differing environmental situations, has previously been acknowledged (Gifford et al., 1973).

Since the manipulation of both temperature and PPF may potentially alter both source and sink activity, further experiments will be required to elucidate which is more limiting.

4.5 References

Allen, E.J. and R.K. Scott. 1980. An analysis of growth of the potato crop. J. Agr. Sci. (Cambridge) 94:583-606.

Aoba, T. 1976. Effect of temperature on the seasonal periodicity of bulb formation in plants. Environ. Control in Biol. 14:81-90.

Barnes, A. 1979. Vegetable plant part relationships. II. A quantitative hypothesis for shoot/storage root development. Ann. Bot. 43:487-499.

Biran, I., B. Leshem, I. Gur and A.H. Halevy. 1974. Further studies on the relationship between growth regulators and tuberization of Dahlias. Physiol. Plant. 31:23-28.

Boerboom, B.W.J. 1978. A model of dry matter distribution in cassava (*Manihot esculenta* Crantz). Neth. J. Agr. Sci. 26:267-277.

Borah, M.N. and F.L. Milthorpe. 1963. Growth of the potato as influenced by temperature. Indian J. Plant Physiol. 5:53-72.

Caesar, K. 1980. Growth and development of *Xanthosoma* and *Colocasia* under different light and water supply conditions. Field Crops Res. 3:235-244.

Causton, D.R. and J.C. Venus. 1981. Single leaf growth and the Richards function: methodology, p. 87-143. In: The biometry of plant growth. Edward Arnold, London.

Das Gupta, D.K. 1972. Developmental physiology of sugar-beet. IV. Effects of growth substances and differential root and shoot temperatures on subsequent growth. J. Expt. Bot. 23:103-113.

Ewing, E.E. 1987. The role of hormones in potato (*Solanum tuberosum* L.) tuberization, p. 515-538. In. P.J. Davies (ed.). Plant hormones and their role in plant growth and development. Martinus Nijhoff Publishers, Dordrecht.

Farrar, J.F. 1992. The whole plant: carbon partitioning during development. p. 163-179. In: C.J. Pollock, J.F. Farrar and A.J. Gordon (eds.). Carbon partitioning within and between organisms. Bios Scientific Publishers, Oxford.

Funnell, K.A. and B.R. MacKay. 1987. Effect of GA_3 on growth of *Zantedeschia*. Tech. Rpt. 87/14, Dept. Hort. Sci. and N.Z. Nursery Res. Centre, Massey Univ., Palmerston North, N.Z.

Fick, G.W., R.S. Loomis and W.A. Williams. 1973. Computer simulation of dry matter distribution during sugar beet growth. Crop Sci. 13:413-417.

Garner, W.W. and H.A. Allard. 1923. Further studies in photoperiodism. The response of the plant to relative length of day and night. J. Agr. Res. 23:871-920.

Geiger, D.R. and B.R. Fondy. 1985. Responses of export and partitioning to internal and environmental factors. British Plant Growth Regulator Group, Monograph 12:177-194.

Gifford, R.M. 1977. Growth pattern, carbon dioxide exchange and dry weight distribution in wheat growing under differing photosynthetic environments. Australian J. Plant Physiol. 4:99-110.

Gifford, R.M., P.M. Bremner and D.B. Jones. 1973. Assessing photosynthetic limitation to grain yield in a field crop. Australian J. Agr. Res. 24:297-307.

Gifford, R.M. and L.T. Evans. 1981. Photosynthesis, carbon partitioning, and yield. Annu. Rev. Plant Physiol. 32:485-509.

Gifford, R.M., J.H. Thorne, W.D. Hitz and R.T. Giaquinta. 1984. Crop productivity and photoassimilate partitioning. Science 225:801-808.

Green, C.F., L.V. Vaidyanathan and J.D. Ivins. 1986. Growth of sugar-beet crops including the influence of synthetic plant growth regulators. J. Agr. Sci. 107:285-297.

Hunt, R. 1982. Plant growth curves. The functional approach to plant growth analysis. Edward Arnold, London.

Keating, B.A., J.P. Evenson and S. Fukai. 1982. Environmental effects on growth and development of cassava (*Manihot esculenta* Crantz.) III. Assimilate distribution and storage organ yield. Field Crops Res. 5:293-303.

McLaren, J.S. 1984. Investigation of quantitative relationships in dry matter distribution between tops and storage roots in sugarbeet. Ann. Bot. 54:383-390.

Menzel, B.M. 1985. Tuberization in potato at high temperatures: Interaction between temperature and irradiance. Ann. Bot. 55:35-39.

Milford, G.F.J., K.Z. Travis, T.O. Pocock, K.W. Jaggard and W. Day. 1988. Growth and dry-matter partitioning in sugar beet. J. Agr. Sci. 110:301-308.

Minchin, P.E.H. and M.R. Thorpe. 1992. Carbon-11 in the study of phloem translocation. p. 225-247. In: C.J. Pollock, J.F. Farrar and A.J. Gordon (eds.). Carbon partitioning within and between organisms. Bios Scientific Publishers, Oxford.

Niimi, Y. 1978. Influence of low and high temperatures on the initiation and the development of a bulb primordium in isolated tulip embryos. Scientia Hort. 9:61-69.

Radley, R.W. 1963. The effect of season on growth and development of the potato, p. 211-220. In: J.D. Ivins and F.L. Milthorpe (eds.). The growth of the potato. Butterworths, London.

Rapoport, H.F. and R.S. Loomis. 1986. Structural aspects of root thickening of *Beta vulgaris* L.: Comparative thickening in sugarbeet and chard. Bot. Gaz. 147:270-277.

Rees, A.R. 1972. The Growth of Bulbs. Academic Press, London.

Rees, A.R. 1984. Dormancy, flowering and periodicity in ornamental bulbs. The Plantsman 6(1):33-44.

Richards, D. and R.N. Rowe. 1977. Effects of root restriction, root pruning and 6-benzylaminopurine on the growth of peach seedlings. Ann. Bot. 41:729-740.

Taeb, A.G. and P.G. Alderson. 1990. Effect of low temperature and sucrose on bulb development and on the carbohydrate status of bulbing shoots of tulip in vitro. J. Hort. Sci. 65:193-197.

Terry, N. 1968. Developmental physiology of sugar beet. I. The influence of light and temperature on growth. J. Expt. Bot. 19:795-811.

Ulrich, A. 1952. The influence of temperature and light factors on the growth and development of sugar beets in controlled climatic environments. Agron. J. 44:66-73.

Ulrich, A. 1955. Influence of night temperature and nitrogen nutrition on the growth, sucrose accumulation and leaf minerals of sugar beet plants. Plant Physiol. 30:250-257.

Valenzuela, H.R., S.K. O'Hair and B. Schaffer. 1991. Shading, growth, and dry-matter partitioning of cocoyam [*Xanthosoma sagittifolium* (L.) Schott]. J. Amer. Soc. Hort. Sci. 116:1117-1121.

Wareing, P.F. and J. Patrick. 1975. Source-sink relations and the partition of assimilates in the plant, p. 481-499. In. J.P. Cooper (ed.). Photosynthesis and productivity in different environments. Cambridge Univ. Press, Cambridge, U.K.

Warrington, I.J. and R.C. Southward. 1989. Influence of temperature and light intensity on *Zantedeschia* growth and development. Commercial Rpt. No. 46, Plant Physiol. Div., DSIR, Palmerston North, N.Z.

Williams, A.M. and K.R. Williams. 1978. Regulation of movement of assimilate into ovules of *Pisum sativum* cv. Greenfeast. Australian J. Plant Physiol. 5:295-300.

Woolley, D.J. and P.F. Wareing. 1972. Environmental effects on endogenous cytokinins and gibberellin levels in *Solanum tuberosum*. New Phytol. 71:1015-1025.

Zar, J.H. 1984. Biostatistical analysis. 2nd ed. Prentice Hall, New Jersey.

5 PHOTOSYNTHETIC ACTIVITY OF *Zantedeschia* 'Best Gold' IN RESPONSE TO TEMPERATURE AND PHOTOSYNTHETIC PHOTON FLUX

5.1 Introduction

The importance of photosynthesis to plant growth is self-evident since 90-95% of the dry weight of plants is derived from photosynthesis (e.g. Irvine, 1975). However, while photosynthesis is accepted as being of fundamental importance, the relationship between harvestable yield and net photosynthesis is not always clear, with variations in growth rate and yield not always being shown to relate to variation in net photosynthesis (Lambers, 1987). Assuming the importance of photosynthesis in determining plant growth and harvestable yield in *Zantedeschia*, modelling of such processes will require description of photosynthetic characteristics and how these might change with environment and development.

Use of the term photosynthetic rate has generally referred to rates obtained on a sub-sample of leaves on a plant, and often fails to take account of leaf age, arrangement or angle. In many cases estimates of photosynthetic rate are limited to defining rate per unit area on a recently expanded leaf. However, a close relationship exists between the increasing rate of photosynthesis as the leaf expands, and the change of leaf activity from a net importer to a net exporter of assimilates as the leaf develops and expands. During this period of development the photosynthetic rate increased, reaching a maximum at between 70% and 80% of final leaf area with sugar beet (*Beta vulgaris* L.; Giaquinta, 1978), 95% with soybean (*Glycine max* L. Merrill; Woodward, 1976), and closer to 100% final leaf area with taro (*Colocasia esculenta* Schott; Sato et al., 1978) and cocoyam (*Xanthosoma sagittifolium* Schott; Schaffer and O'Hair, 1987). Following the attainment of maximum photosynthetic rate, the rate declines with age. Any experiments to define the photosynthetic characteristics of *Zantedeschia* would therefore need to be conducted with consideration of the influence of leaf expansion and maturation.

Leaves within the upper canopy typically contribute a proportionately larger quantity of the total plant photoassimilates that are accumulated than those within the lower canopy (Acock et al., 1978; Hodanova, 1979). While the potential photosynthetic activity of leaves is reduced with age, mutual shading of leaves within the canopy is also a contributing factor to this disproportionate contribution (Hodanova, 1979). The photosynthetic photon flux (PPF) at which photosynthetic rate becomes light saturated varies between species and growing environment, with photosynthetic activity acclimating to the environment in which the leaves have grown (Bowes et al., 1972; Pallas and Samish, 1974; Bjorkman, 1981). Shade tolerance, where the photosynthetic rate is greater when leaves have been grown under shade compared with full sunlight, is a feature of a number of members of the *Araceae* (O'Hair and Asokan, 1986; Schaffer and O'Hair, 1987). Bjorkman (1981)

suggested that obligate shade plants have an intrinsically low potential for PPF acclimation and particularly a low ability to increase their capacity for effective utilization of high PPF. However, at the same time obligate shade plants are more efficient at utilizing PPF when PPF is low than plants adapted to sun habitats. Even within the same species clones which have adapted to shade habitats exhibit differing acclimation ability than clones from sun habitats, as might normally be expected between sun and shade species (Bjorkman and Holmgren, 1966). With several tropical tree seedlings grown at a range of PPF, the increase in photosynthetic rate at increased PPF was associated with increased leaf thickness (Kwesiga et al., 1986). The effect of low PPF on many species is to cause a reduction in respiratory loss and to decrease specific leaf weight (Boardman, 1977, Kwesiga et al., 1986).

Photosynthetic rate is dependent on both PPF and temperature. Photosynthetic rate increases with rising temperature up to an optimum which depends on growing environment and species (Warren-Wilson, 1966a). Both short-term and long-term preconditioning effects of temperature on photosynthetic rate of plants have been noted in many species (Bennett et al., 1982; Bunce, 1985; Laing, 1985; Caulfield and Bunce, 1988). However, while some minor differences exist between plant species in the relationship between photosynthetic rate and day and night temperatures, the temperature at the time of measurement had the greatest single effect on photosynthetic rate (Bennett et al., 1982). With most species, leaf thickness and therefore specific leaf weight values decrease with an increase in temperature (Laing, 1985). Such changes usually result in changes in photosynthetic rate per unit leaf area.

The photosynthetic rate can be altered significantly by manipulating sink strength. In a number of crop species, once new sinks have been initiated or sink strength increased, photosynthetic rate has increased several fold (Moorby, 1968; Chatterton et al., 1972; Habeshaw, 1973; Thorne and Koller, 1974). In addition, such increases in photosynthetic rate have been correlated with increased assimilate export from the leaf to the developing sink. Conversely the removal of major sinks or reduction in sink activity has resulted in a lowered demand for assimilate, and a reduction in net photosynthesis (Habeshaw, 1973; Thorne and Koller, 1974; Wang and Breen, 1986). In contrast Geiger (1976) found that manipulation of sink demand, or of export rate, in beans (*Phaseolus vulgaris* L.) resulted in no observed adjustment in the rate of net photosynthesis. Obviously a simple, negative feed-back mechanism does not seem to be adequate to bring about the control between sink demand and net photosynthesis in all cases. If changes in sink activity of *Zantedeschia* were to result in changes in photosynthetic rate, then it would be most likely that those leaves with the greatest potential photosynthetic rate (i.e., at or near full expansion and without mutual shading) would exhibit the greatest measurable change in this parameter.

Under controlled environmental conditions several plant species show a decrease in photosynthetic rate during the day (Pallas, 1973; Warrington et al., 1977). In contrast constant photosynthetic rates during the entire photoperiod have been reported for other species (Pallas, 1973; Massimino et al., 1980; Mortensen and Moe, 1983). A negative correlation between assimilate level and photosynthetic rate during the photoperiod has been determined in a number of the cases describing decreased photosynthetic rates, being suggestive of feed-back inhibition (Chatterton et al., 1972; Warrington et al., 1977). However this correlation has not been found to be universally applicable, varying between species and with leaf maturity (Potter and Breen, 1980; Correia et al., 1990). Since it is generally agreed that differences in PPF result in differences in photosynthetic rate, and therefore production of photoassimilates, the ability to detect differences between treatments in the experiments outlined in the current study, may be dependant on the time within the photoperiod at which photosynthetic rate is measured.

The rates of those processes in which assimilates are utilized (i.e., respiration and new growth) fall more rapidly with decrease in temperature than do the rates at which assimilates are produced (Warren-Wilson, 1966b; Verkleij and Challa, 1988; Acock et al., 1990). At lower temperatures, therefore, assimilates such as starch will tend to accumulate. However, this may not happen with all species, especially those adapted to cooler temperatures (Paul et al., 1990). Hence, if changes in PPF result in changes in photoassimilate production, and differing temperature regimes influence assimilate partitioning, then measurement of assimilate concentrations and photosynthetic rates would be potential tools to determine the mechanisms of assimilate acquisition and partitioning in *Zantedeschia*.

There are no previously reported studies of the photosynthetic characteristics of the genus *Zantedeschia*. Assuming the potential importance of photosynthesis in determining growth in *Zantedeschia*, such basic information and its possible correlation with total growth and harvestable yield will be required for determination of optimum growing environments, and for future development of crop simulation models.

5.2 Materials and Methods

Data presented in this chapter were primarily derived from the experiment described previously (refer Section 3.2). Cultural procedures (refer Section 3.2.1) and environmental conditions (refer Section 3.2.2) were as described in those sections.

5.2.1 Experimental

5.2.1.1 Photosynthesis as a function of leaf expansion

Net photosynthetic rate and leaf area were monitored during expansion of the first leaf produced from plants grown under three treatments. Treatments used consisted of a day/night temperature regime of 22/16 C under both high and low PPF regimes, and 28/22 C under the high PPF regime. Photosynthetic rates were measured using an open system infra-red gas analyser (LCA-2, ADC Ltd., Hoddesdon, England), with air obtained from outside the building for the inlet supply (350 $\pm$ 10 μmol CO_2 per mol), calibrated against Wiscoff pumps. All photosynthetic measurements were carried out at the relevant treatment day temperature and PPF. The air supply was dried to a constant 20% relative humidity and pumped into the assimilation cuvette (Parkinson leaf chamber) at a rate of 0.625 litre$\cdot$min^{-1}. Net photosynthetic rates were calculated from the difference between inlet and outlet CO_2 concentrations as outlined in the LCA-2 user manual. Leaf area expansion over time was determined nondestructively by tracing the outline of selected leaves on paper following each photosynthetic measurement, and subsequently measuring the area of each tracing using a LICOR model 3100 leaf area meter. Within each temperature $\times$ light treatment, plants were arranged in a completely randomized design. Net photosynthesis data were recorded from a random sample of 6 leaves at two day intervals, 8 h after the commencement of daily lighting, beginning from when the leaf lamina was free from enveloping protective sheath leaves, and continuing until further increases in leaf expansion were minimal.

Partial correlations between; net photosynthetic rate, day temperature, night temperature, PPF, and proportion (%) maximum leaf area, were examined in the formulation of predictive models. Models were determined by regression analysis using the stepwise selection method of the REG procedure within the Statistical Analysis System (SAS; SAS Institute, Inc., Cary, N.C.)

5.2.1.2 Photosynthetic rate as a function of duration from commencement of daily lighting

Photosynthetic rates of recently-mature leaves were monitored during a daily period of lighting, from plants grown under four treatments. Recently-mature was defined as the

stage when a leaf had attained a minimum of 75% expansion, and the experiment was carried out on the first such leaf on selected plants. Treatments consisted of day/night temperature regimes 16/10, 22/16 and 28/22 C under the high PPF regime, and 22/16 C under the low PPF regime. Photosynthetic rates were measured as outlined above except air flow rates were sometimes reduced to 0.356 litre·min^{-1} to account for negative or minimal photosynthetic rates encountered. Data were recorded from a random sample of 6 plants, initially at 30 min intervals commencing 15 min prior to the commencement of lighting, and subsequently at more infrequent intervals across the 12 h period of lighting.

Within each temperature × light treatment combination, plants were arranged in a completely randomized design. To avoid potential inherent problems arising from repeated measures, a different sample of 6 leaves was used from each treatment population at each time of measurement. Rates of change in net photosynthetic rate with time were determined by regression analysis using the REG procedure of SAS.

5.2.1.3 Photosynthetic rate as a function of photosynthetic photon flux

At the time when the first leaf had reached maturity (as defined above) response curves for photosynthesis as a function of PPF were derived from a sample of 6 plants per treatment. Photosynthetic rates were measured 8 h after the commencement of daily lighting. PPF was measured using the quantum sensor on the LCA-2 apparatus. Measured leaves were sealed in the assimilation cuvette at the day temperature at which the plants were grown. The PPF was then adjusted stepwise using spectrally neutral mesh screens to reduce PPF, and raising the height of the plant closer to the light source to increase PPF. Respiration was measured in the dark using black cloth to eliminate light from the cuvette, with reduced air flow rates as previously discussed. The photosynthetic rate and PPF data were fitted to a hyperbolic tangent function of the form:

$$Pn = P_{max} \cdot Tanh\left(\frac{\alpha}{P_{max}} \cdot PPF\right) - R \qquad (5.1)$$

where;

- Pn = net photosynthetic rate (μmol·m^{-2}·s^{-1} or μg·g^{-1}·s^{-1})
- P_{max} = maximum photosynthetic rate at saturating PPF (μmol·m^{-2}·s^{-1} or μg·g^{-1}·s^{-1})
- $Tanh$ = hyperbolic tangent
- α = apparent quantum yield (i.e., the initial slope, mol CO_2 per mol PPF or g CO_2 per mol PPF)
- PPF = PPF (μmol·m^{-2}·s^{-1})
- R = respiration rate (μmol·m^{-2}·s^{-1})

This function was chosen above others reported in frequent use, such as the rectangular hyperbola, as the former offers parameters which are both mutually independent and biologically meaningful (Jassby and Platt, 1976).

Specific leaf weights were calculated from destructive harvests carried out subsequent to recording photosynthetic rates. The data, both on a leaf area and leaf weight basis, were fitted by nonlinear regression analysis using the NLIN procedure of SAS. All 6 replicates from a given treatment were fitted individually. Derived parameters from the curve fitting procedure were subjected to analysis of variance using the general linear models procedure of SAS.

In addition to measuring actual rates of dark respiration as part of determining the PPF response curves, rates of dark respiration were also measured commencing 2 h after the termination of the lighting period. Plants used were derived from treatments consisting of day/night temperature regimes 22/10, 22/16, 28/16 and 28/22 C, under both high and low PPF. Respiration rates were measured as outlined above for photosynthetic rates, during the established night temperature and using the reduced air flow rate. Data were recorded from a random sample of 6 mature leaves. Rates of change in respiration rate as a function of actual leaf temperature, were determined by regression analysis using the REG procedure of SAS. In addition, rates were subjected to analysis of variance using the general linear models procedure of SAS.

5.2.1.4 Photosynthesis during plant development

Prior to the destructive harvesting of 6 or 12 plants used to monitor growth (refer Section 3.2.3), the photosynthetic activity of the most recently-mature leaf was measured. Initial measurements were recorded at the time of recent-maturation of successive leaves. Once increases in tuber dry weight were detected, sampling occurred more frequently, but photosynthetic rate was always monitored on the most recently-mature leaf of plants sampled, 8 h after the commencement of daily lighting. Monitoring continued until the rate of total plant dry weight accumulation declined (refer Section 3.2.3). At the time total plant relative growth rate (RGR_W) and tuber relative growth rate (RGR_T) were maximum, coefficients of correlation between net photosynthetic rates and both RGR_W and RGR_T were determined by correlation analysis using the CORR procedure of SAS.

5.3 Results

5.3.1 Photosynthesis as a function of leaf expansion

Net photosynthetic rate (*Pn*) increased with expansion of the leaf under all treatment environments (Figure 5.1). Maximum photosynthetic rates did not occur until leaves had reached approximately 90% expansion regardless of temperature and PPF regime. At the stage of expansion subsequently defined as mature (i.e., 75% expansion), leaves had achieved 81% of the maximum photosynthetic rate. While described in more detail under Section 5.3.3, in brief, at maximum leaf expansion photosynthetic rates were greater at higher temperatures and under high PPF.

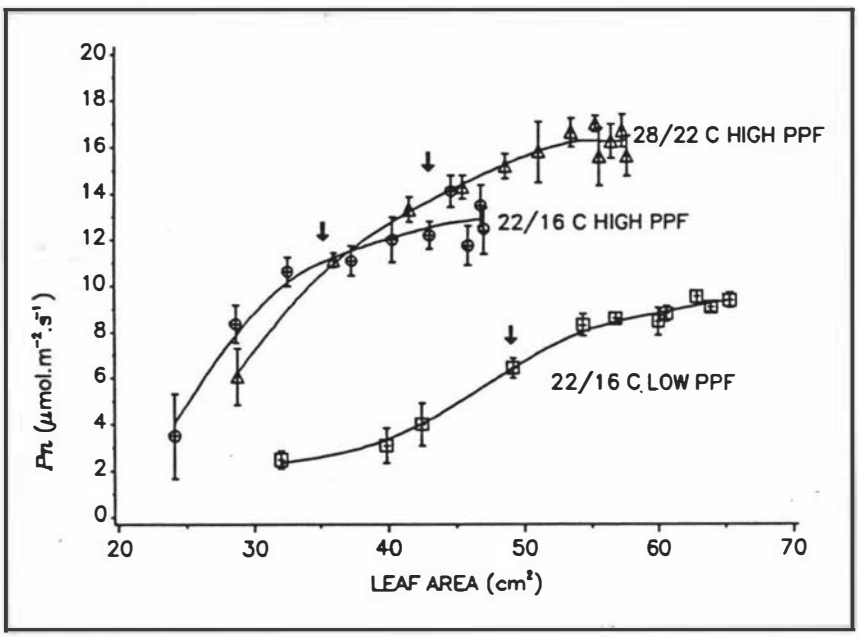

Figure 5.1 Net photosynthetic rate (*Pn*) as a function of individual leaf area expansion, at selected temperature and PPF regimes. Mean values ± se., n=6, cubic spline fit.
↓ represent 75% maximum leaf area.

Across all treatments *Pn* was positively correlated with: day temperature (DTEMP), night temperature (NTEMP), PPF, and percentage maximum leaf area (%LA). The magnitude of the standardized multiple regression coefficients (beta coefficients) were indicative of the relative contribution made by each in determining *Pn* over the entire period of expansion, i.e., %LA = 0.56, PPF = 0.37, and DTEMP = 0.34. However, the addition of NTEMP did not meet the 0.15 significance level for entry into the model. A single predictive model including the above parameters was developed (equation (5.2)) which accounted for 78% of the variation in *Pn*, under the environments examined and throughout the entire period of expansion.

$$Pn = 0.122 \cdot \%LA + 0.011 \cdot PPF + 0.53 \cdot DTEMP - 18 \qquad (r^2 = 0.78,^{***}) \quad (5.2)$$

Over the entire period of leaf expansion a strong partial correlation existed between *Pn* and %LA ($r = 0.76$). However, once leaves had reached 75% or more of their maximum leaf area this partial correlation was reduced to 0.46. In contrast, during this later stage of expansion the partial correlation between *Pn* and PPF was 0.72, and with DTEMP was 0.66. As a result, beta coefficients for the prediction of *Pn* were: %LA = 0.22, PPF = 0.55, and DTEMP = 0.45. As noted earlier, the addition of NTEMP to this model did not meet the 0.15 significance level for entry. A single predictive model for this later stage of expansion was developed (equation (5.3)) which accounted for 81% of the variation in *Pn* under the environments examined.

$$Pn = 0.06 \cdot \%LA + 0.013 \cdot PPF + 0.55 \cdot DTEMP - 14 \qquad (r^2 = 0.81,^{***}) \quad (5.3)$$

5.3.2 Photosynthesis as a function of duration from commencement of daily lighting

Net photosynthetic rate increased rapidly during the first hour of lighting (Figure 5.2; low PPF regime data not presented). Subsequent changes in net photosynthetic rates were minor for the remaining period of stable, temperature, vapour pressure deficit, and PPF conditions. Over this time period, the magnitude of the greatest difference in photosynthetic rate, i.e., 1.0 $\mu mol \cdot m^{-2} \cdot s^{-1}$, was only slightly greater than the estimated accuracy of the equipment, i.e., 0.7 $\mu mol \cdot m^{-2} \cdot s^{-1}$. Considering the potential for a decline in net photosynthetic rate purely as a result of repeated measures on the same leaf (Biddington, 1986), it was felt that further interpretation of these data was not warranted.

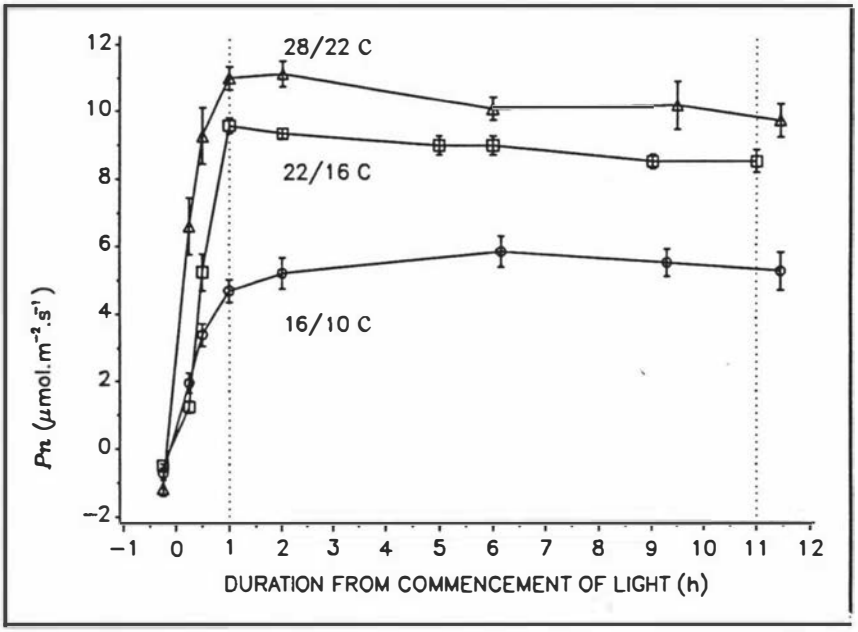

Figure 5.2 Photosynthetic rate (*Pn*) as a function of duration from commencement of daily lighting, at selected temperature regimes and high PPF. Mean values ± se., n=6. Broken lines indicate limits of diurnal temperature changeovers.

5.3.3 Photosynthetic rate as a function of photosynthetic photon flux

Under all treatments the increasing rate of net photosynthesis with increasing PPF was adequately described by a family of Tanh functions on a basis of both leaf area (Figure 5.3a and Figure 5.3b) and leaf weight (Figure 5.3c and Figure 5.3d). Data from plants grown at a common PPF and day temperature were pooled, as differences in the magnitude of the fitted parameters (as per equation (5.1)), between treatments with the same day temperature were not significant ($P \leq 0.05$).

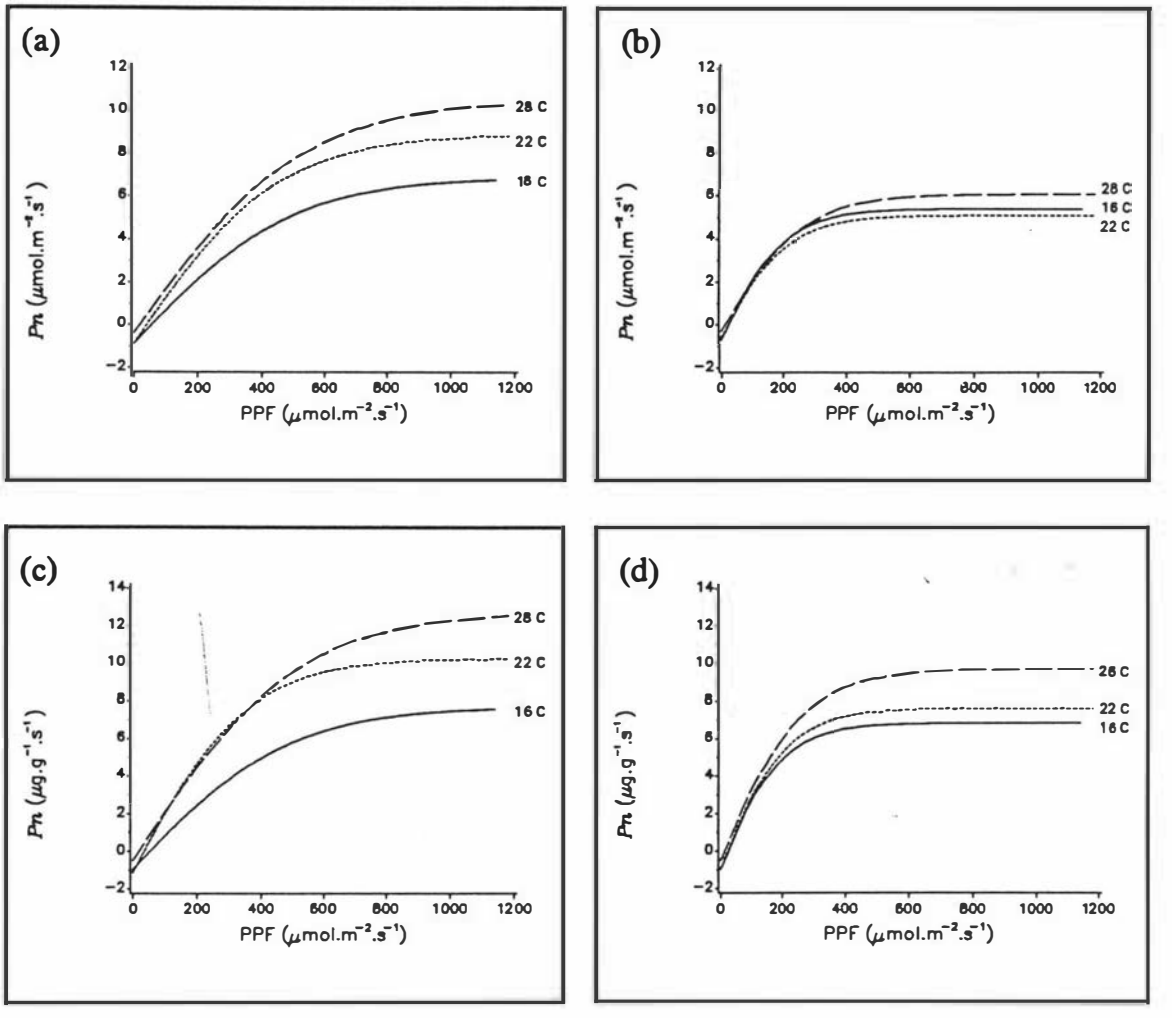

Figure 5.3 Photosynthetic rate (*Pn*) per unit leaf area (a) and (b), and per unit leaf weight (c) and (d), as a function of photosynthetic photon flux (PPF), for plants of *Zantedeschia* 'Best Gold' grown at day temperatures of 16, 22 and 28 C, under high (a) and (c), or low (b) and (d), PPF regimes. n=6 or 18, function = equation 5.1.

Across all temperatures maximum photosynthetic rate (P_{max}) was greater under the high PPF regime than under the low PPF regime ($P \leq 0.001$, Figure 5.4, Figure 5.5). Under the high PPF regime P_{max} per unit leaf area increased linearly with increasing temperature, resulting in a 40% increase across the temperature range examined ($P \leq 0.001$). In contrast, under the low PPF regime, differences among temperatures in P_{max} per unit leaf area were minimal, amounting to less than 1 $\mu mol \cdot m^{-2} \cdot s^{-1}$, i.e., only 12%. When expressed on the basis of leaf weight, P_{max} increased linearly with increasing temperature under both PPF regimes ($P \leq 0.001$, Figure 5.5). This response of P_{max} with temperature resulted in a maximum 53% and 31% increase across the temperature range, under the high and low PPF regimes respectively.

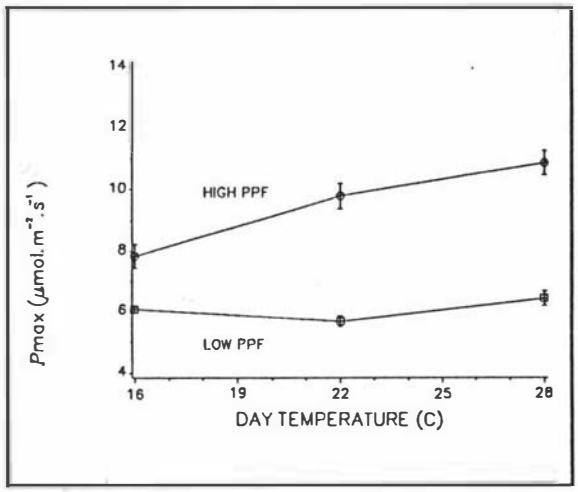

Figure 5.4 Maximum photosynthetic rate (*Pmax*) per unit leaf area as a function of day temperature, for plants of *Zantedeschia* 'Best Gold' grown under high and low PPF. Vertical bars = 2 × standard error.

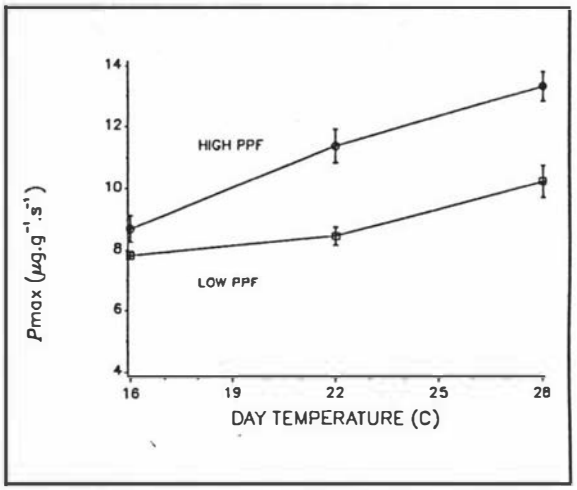

Figure 5.5 Maximum photosynthetic rate (*Pmax*) per unit leaf weight as a function of day temperature, for plants of *Zantedeschia* 'Best Gold' grown under high and low PPF. Vertical bars = 2 × standard error.

When expressed on the basis of both leaf area and weight, apparent quantum yield (α) was greater under the low PPF regime than under the high PPF regime ($P \leq 0.001$, Figure 5.6, Figure 5.7). Under the high PPF regime, the reduced value of α per unit leaf area at 16 C resulted in a quadratic relationship between apparent quantum yield and increasing day temperature ($P \leq 0.01$). However, under the low PPF regime, α declined linearly with increasing day temperature ($P \leq 0.05$). On a leaf weight basis under the high PPF regime the quadratic relationship between α and temperature was even more pronounced, while under the low PPF regime α was independent of temperature (Figure 5.7).

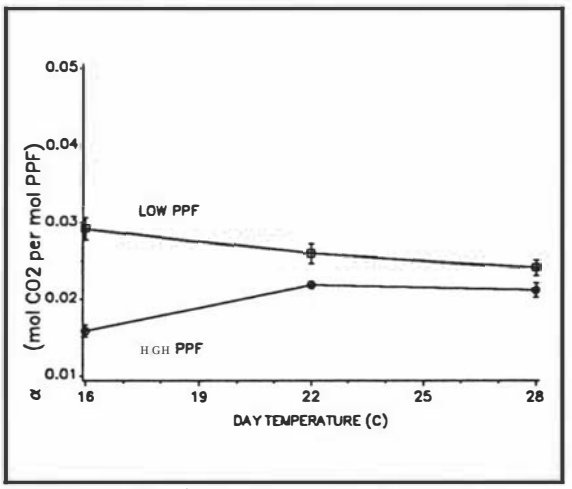

Figure 5.6 Quantum yield (α) per unit leaf area as a function of day temperature, for plants of *Zantedeschia* 'Best Gold' grown under high and low PPF. Vertical bars = 2 × standard error.

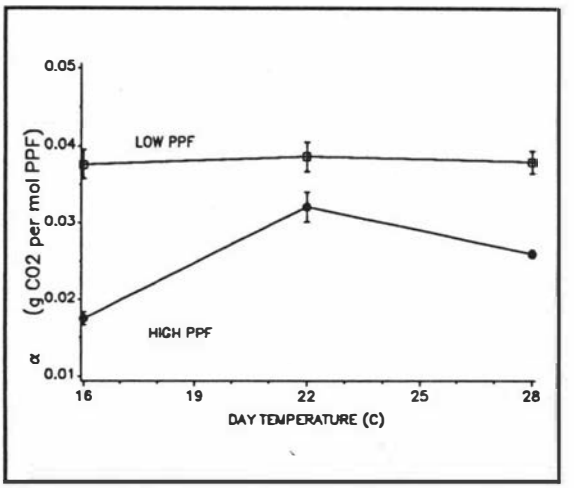

Figure 5.7 Quantum yield (α) per unit leaf weight as a function of day temperature, for plants of *Zantedeschia* 'Best Gold' grown under high and low PPF. Vertical bars = 2 × standard error.

The difference between photosynthetic rate under saturating PPF (P_{max}) and the photosynthetic rate under the growth PPF (P_{grow}), i.e. ΔP, indicates the ability of the leaf to adjust photosynthetic rate to sudden changes in PPF. Positive values of ΔP occurred in all treatments, with the magnitude of ΔP under the high PPF regime being at least twice that determined under the low PPF regime (Figure 5.8, Figure 5.9). When expressed either on the basis of leaf area or leaf weight the magnitude of ΔP under the high PPF regime was not influenced by temperature. Under the low PPF regime ΔP was lower at 28 C than at either 16 or 22 C ($P \leq 0.05$). However, the magnitude of this reduction (0.3 ± 0.1 $\mu mol \cdot m^{-2} \cdot s^{-1}$) was smaller than the accuracy of measurement of the equipment ($\pm$ 0.7 $\mu mol \cdot m^{-2} \cdot s^{-1}$). Given the magnitude of measurement error, the biological significance of the statistically determined difference cannot be assessed.

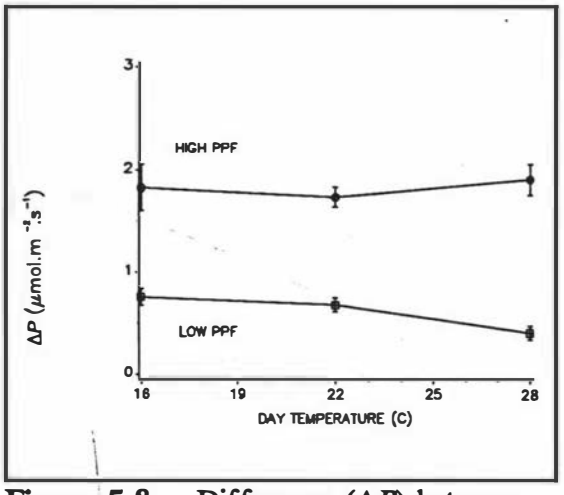

Figure 5.8 Difference (ΔP) between P_{max} and P_{grow} per unit leaf area, for plants of *Zantedeschia* 'Best Gold' grown at a range of temperatures, under high and low PPF.

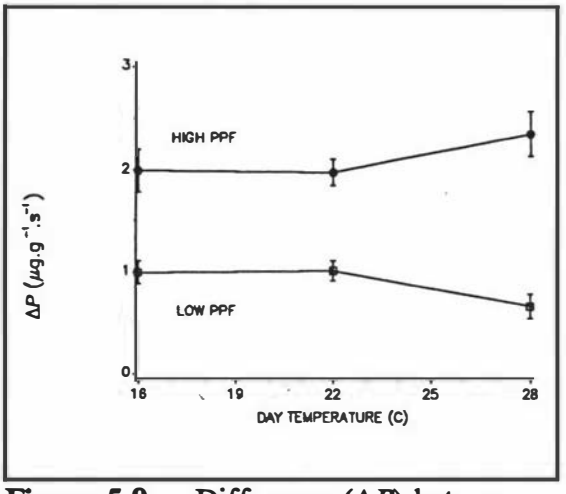

Figure 5.9 Difference (ΔP) between P_{max} and P_{grow} per unit leaf weight, for plants of *Zantedeschia* 'Best Gold' grown at a range of temperatures, under high and low PPF.

On a leaf area basis, rates of dark respiration measured during the day period, were greater for leaves grown under high PPF (0.7 ± 0.1 $\mu mol \cdot m^{-2} \cdot s^{-1}$) than low PPF ($0.5 \pm 0.1$ $\mu mol \cdot m^{-2} \cdot s^{-1}$; $P \leq 0.05$). Both the measured dark respiration rates and those determined from fitting equation (5.1) appeared to be reduced at day temperatures of 28 C compared with either 22 or 16 C (data not presented). However, as discussed with reference to ΔP, the magnitude of this reduction in respiration rate (0.4 ± 0.1 $\mu mol \cdot m^{-2} \cdot s^{-1}$) was only marginally greater than the accuracy of measurement of the equipment ($\pm$ 0.3 $\mu mol \cdot m^{-2} \cdot s^{-1}$) even with a reduced air flow rate.

Across all temperatures the rate of dark respiration, when measured during the night period, was greater for leaves grown under high PPF (0.7 ± 0.1 $\mu mol \cdot m^{-2} \cdot s^{-1}$) than under

low PPF (0.5 ± 0.1 $\mu mol \cdot m^{-2} \cdot s^{-1}$; $P \leq 0.05$). Leaf temperature under the measurement conditions varied between 13 and 28 C, but differences between temperatures in rates of actual dark respiration were not detected ($P = 0.10$).

5.3.4 Photosynthesis during plant development

With the exception of the temperature extremes of 16/10 C and 28/28 C, net photosynthetic rate per unit leaf area, of the most recently matured leaf, changed considerably both during plant development and with environment (Figure 5.10). Photosynthetic rate of the first two to three successive leaves were as described in the PPF response curves (Figure 5.3). However, rates of leaves maturing later were successively lower, under the high PPF regime. With the exclusion of the 16/10 C and 28/28 C treatments, rates of photosynthesis in later emerging leaves were approximately half of those earlier emerging leaves at the termination of the temperature treatments under the high PPF regime. In contrast, with the exclusion of the same treatments under the low PPF regime, photosynthetic rates of later developing leaves were successively greater than those recorded initially. Under these low PPF treatments photosynthetic rates were approximately 2 fold greater at the termination of the treatments compared with at the beginning of the treatments. With the succession of recently matured leaves at 16/10 C, the change in photosynthetic rates followed a similar trend of decline under both high and low PPF regimes, with rates tending to be greater under the low PPF regime for the later stages of growth than under the high PPF regime (Figure 5.10a). At 28/28 C photosynthetic rates of the succession of recently matured leaves did not alter, being maintained at a similar value throughout the period of study (Figure 5.10f).

When expressed on the basis of unit leaf weight, the progressive reversal in magnitude of photosynthetic rate with ontogeny, between PPF regimes, was only evident (to a lesser extent) for plants grown at 22/10 and 22/16 C (Figure 5.11b and c). Concurrent with the decline in net photosynthetic rate at these temperatures, recently mature leaves showed signs of chlorosis under the high PPF regime. In contrast, older leaves under the high PPF regime and all leaves under the low PPF regime showed no signs of chlorosis. While photosynthetic rates per unit weight were progressively lower with ontogeny under both PPF regimes, the rate initially declined more quickly under the high PPF regime than under low PPF. As a result, photosynthetic rates per unit leaf weight were similar under both PPF regimes for most of the period of study.

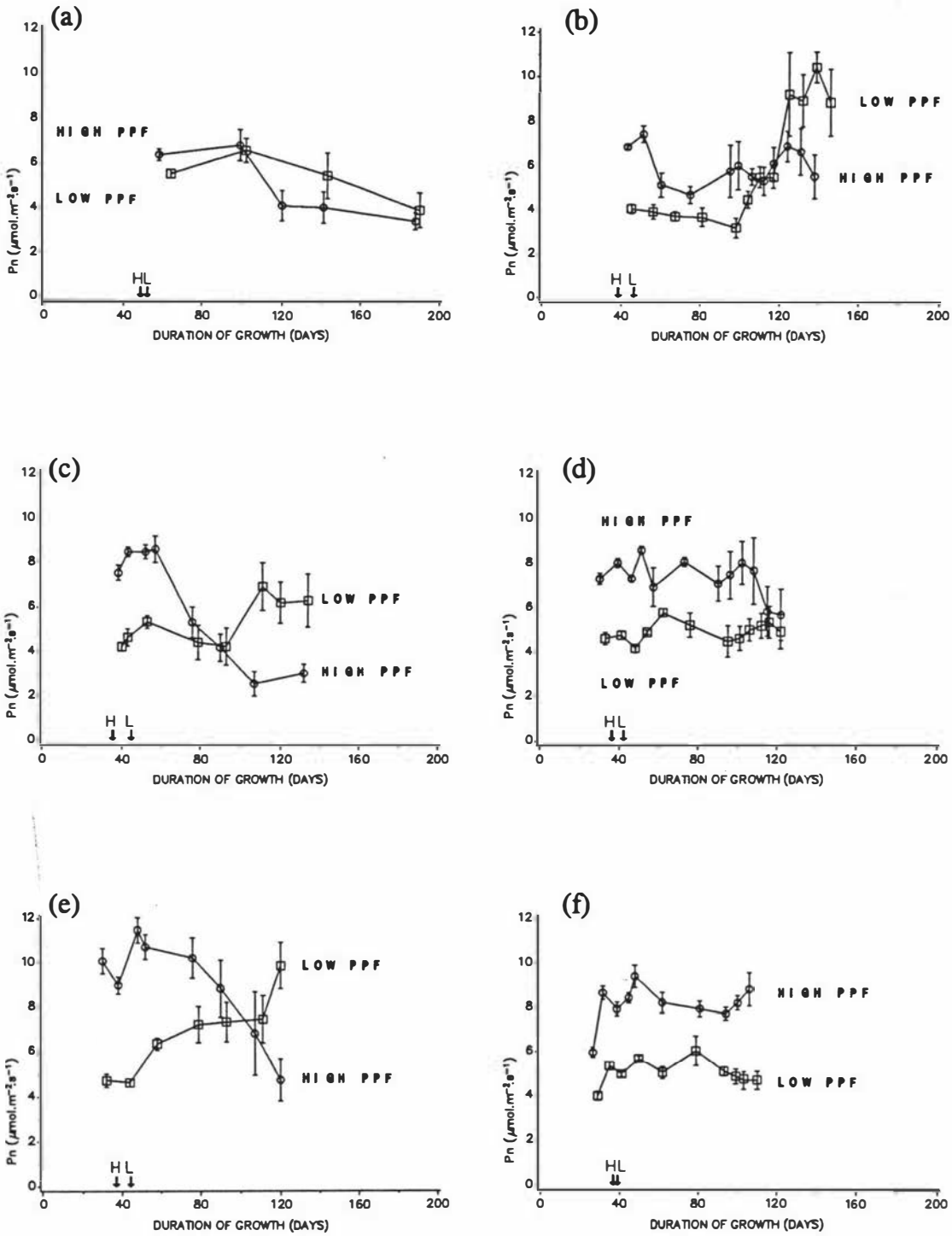

Figure 5.10 Photosynthetic rate (Pn), per unit leaf area, of the most recently expanded leaf as a function of time, for *Zantedeschia* 'Best Gold' grown at a range of day/night temperatures under high and low PPF regimes. (a) 16/10 C (b) 22/10 C (c) 22/16 C (d) 28/16 C (e) 28/22 C (f) 28/28 C. Vertical bars = 2 × standard error, arrows indicate commencement of tuber growth under high (H) and low (L) PPF.

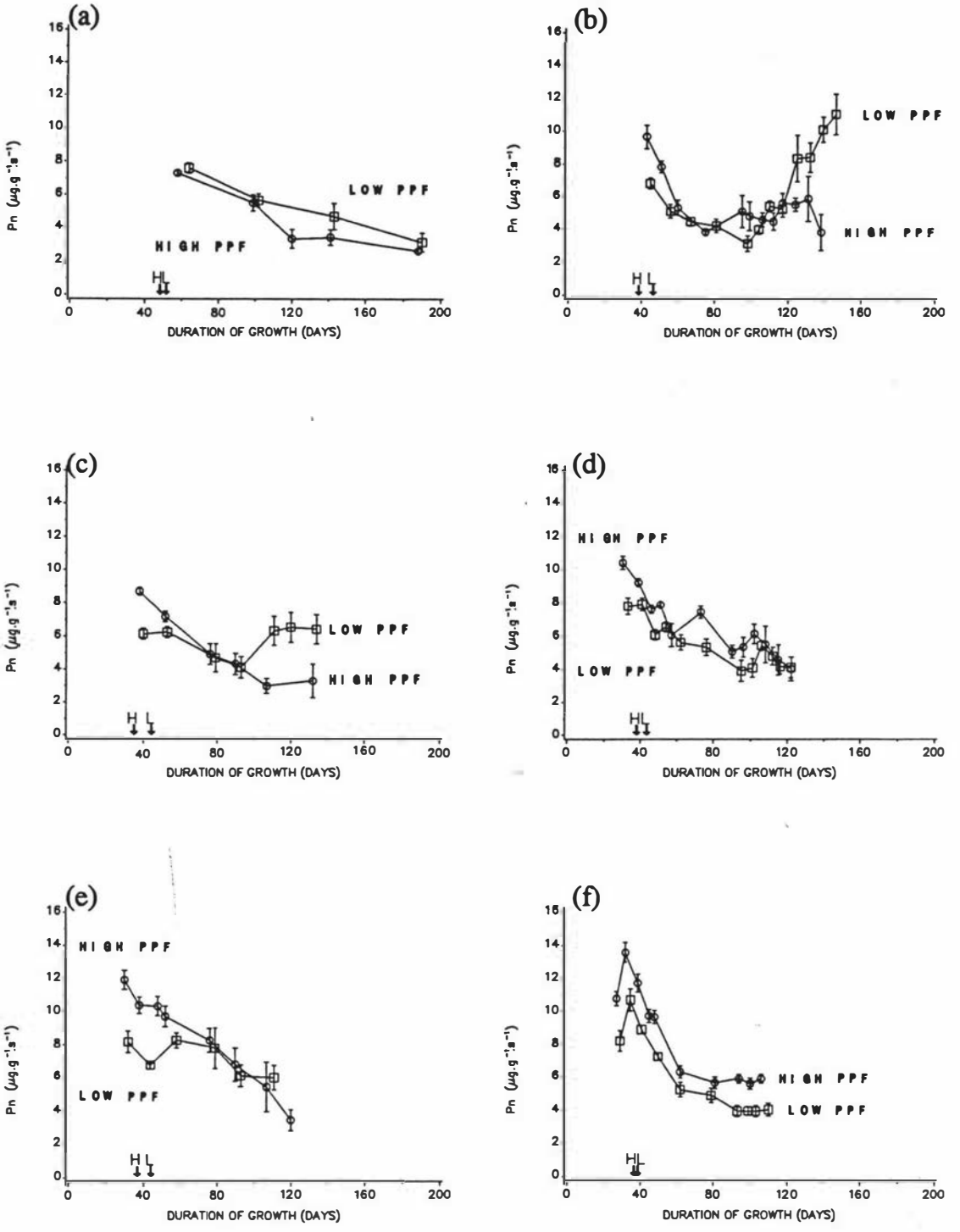

Figure 5.11 Photosynthetic rate (Pn), per unit leaf weight, of the most recently expanded leaf as a function of time, for *Zantedeschia* 'Best Gold' grown at a range of day/night temperatures under high and low PPF regimes. (a) 16/10 C (b) 22/10 C (c) 22/16 (d) 28/16 C (e) 28/22 C (f) 28/28 C. Vertical bars = 2 × standard error, arrows indicate commencement of tuber growth under high (H) and low (L) PPF.

Under the low PPF regime the increase in photosynthetic rate per unit leaf weight of successive leaves, commencing after 90 to 100 days, was only evident under the day/night temperature regimes of 22/10 and 22/16 C (Figure 5.11b and c). With the onset of tuber growth occurring between 35 and 52 days after planting (refer Chapter 4), such changes in photosynthetic rate could not be readily attributed to tuber growth under either PPF regime.

5.3.5 Photosynthesis as a predictor of growth and yield

Across all treatments photosynthetic rate of the most recently mature leaf was positively correlated with total plant relative growth rate (RGR_W), at the time RGR_W was maximum, i.e., the inflection point of the fitted total dry weight curve. Expression of the photosynthetic rate on a per unit leaf weight basis improved the strength of this correlation, compared with that expressed on a leaf area basis, from $r = 0.53$ to $r = 0.87$ (Table 5.1). The correlation between photosynthetic rate per unit leaf area and RGR_W was maintained with later stages of development, but no significant correlation could be detected when expressed on a leaf weight basis.

Correlations between photosynthetic rate per unit leaf area and tuber relative growth rate (RGR_T) were not significant at either stages of development examined (Table 5.1). At the time of maximum RGR_W, a weak correlation between photosynthetic rate per unit leaf weight and RGR_T was determined, but there was no significant correlation at the time of maximum RGR_T.

Table 5.1

Correlation between photosynthetic rate (*P*n) and relative growth rate of total plant weight (RGR_W) and tuber dry weight (RGR_T) of *Zantedeschia* 'Best Gold' grown at six day/night temperatures and two PPF regimes, during two stages of development ($n = 14$).

	RGR_W		RGR_T	
Developmental stage	CO_2 assimilation ($\mu mol \cdot m^{-2} \cdot s^{-1}$)	CO_2 assimilation ($\mu g \cdot g^{-1} \cdot s^{-1}$)	CO_2 assimilation ($\mu mol \cdot m^{-2} \cdot s^{-1}$)	CO_2 assimilation ($\mu g \cdot g^{-1} \cdot s^{-1}$)
	r			
Mid-exponential total dry-weight	0.53^{**}	0.87^{***}	0.36^{ns}	0.47^{*}
Mid-exponential tuber dry-weight	0.54^{**}	0.39^{ns}	0.37^{ns}	0.38^{ns}

*, **, *** Significant at $P = 0.10$, 0.05, and 0.001, respectively.
^{ns} not significant at $P = 0.10$.

5.4 Discussion

Maximum photosynthetic rate did not occur until individual leaves had reached approximately 90% expansion. However, leaves had attained 81% of the maximum rate at 75% expansion. This synchrony of photosynthetic rate and leaf area expansion was similar to that described for numerous crops such as cucumber (*Cucumis sativus* L.), sugar beet and soybean (Woodward, 1976; Giaquinta, 1978; Ho et al., 1984) as well as other members of the *Araceae* such as taro and cocoyam (Sato et al., 1978; Schaffer and O'Hair, 1987). Since subsequent experiments presented in this thesis use the 75% level of expansion as a minimum, interpolation of results should be tempered by the knowledge that photosynthetic rates may be up to 20% less in magnitude than their maximum. Since leaves of other plant species have been reported as being net exporters of photoassimilates when between 30 and 50% of maximum leaf expansion (Wardlaw, 1968; Suzuki and Mortimer, 1973; Ho et al., 1984), it is assumed that leaves monitored in subsequent experiments were also able to export assimilates.

There are no known published models relating nondestructive measurements of *Zantedeschia* leaf photosynthesis to leaf area expansion. Hence the ability to accurately estimate leaf area expansion under the current experiment was difficult. Percentage completion of leaf area expansion was a primary determinant of photosynthetic rate during initial expansion. However, at and beyond the 75% expansion stage, the inclusion of percentage completion of expansion in the predictive model only accounted for an additional 5% of the variation in photosynthetic rate not already accounted for by day temperature and PPF. Since this stage of expansion was used for subsequent investigations, it can be assumed that photosynthetic rates reported were primarily a function of growing environment.

Changes in photosynthetic rate during the daily period of lighting were small enough to be within the margin for measurement error, and were therefore not considered large enough to warrant further interpretation. Since the data were recorded after 8 h of the commencement of the photoperiod, it can be assumed that photosynthetic rates were at their maximum for the treatment conditions. A diurnal decline of photosynthetic rate has been reported for some plant species (Pallas, 1973; Warrington et al., 1977), but the rates of other species have been shown to be constant (Pallas, 1973; Massimino et al., 1980; Mortensen and Moe, 1983). While the result reported here does not preclude the accumulation of photoassimilates within the leaves, it does not support the existence of a simple feed-back inhibition mechanism controlling the photosynthetic rate in *Zantedeschia* 'Best Gold', as proposed for some species (Chatterton et al., 1972; Warrington et al., 1977).

Photosynthetic rates of leaves of *Zantedeschia* 'Best Gold' are similar to those reported for

other members of the *Araceae*, e.g. taro and cocoyam (Schaffer and O'Hair, 1987; Valenzuela, 1990). In comparison with other plant species these photosynthetic rates can be considered to be in the low to intermediate range (Bjorkman, 1981; Connor and Sadras, 1992). In addition, the photosynthetic rates of leaves illustrated a ready acclimation to the mean daily temperature and PPF conditions of growth. Across all treatments this acclimation to temperature and PPF resulted in photosynthetic rates under each environment (i.e., P_{grow}) reaching an average 85% of the maximum rate possible (i.e., P_{max}). While accurate quantification of this acclimation ability would require experiments using the appropriate exchanges in PPF and temperature treatments, the data reported here indicates an ability for short-term acclimation. Hence a substantial ability for acclimation was seen at the temperature optima of 28 C, where doubling the PPF during growth resulted in a 70% increase in the maximum photosynthetic rate per unit leaf area (Figure 5.4). The ability of leaves of *Zantedeschia* 'Best Gold' to acclimate to PPF can be considered to be substantial compared with that reported for a number of other plant species. The doubling of the PPF during growth of *Lolium multiflorum* L. and *Solidago virgaurea* L. resulted in increases in maximum photosynthetic rate of 25% or less (Prioul and Bourdu, 1973). In contrast, a similar increase in PPF with lettuce (*Lactuca sativa* L.) resulted in a 100% increase in maximum assimilation rate (Prioul and Bourdu, 1973).

With increased PPF the greater magnitude of ΔP (Figure 5.8 and Figure 5.9), has been reported as indicating an enhanced ability to adjust to sudden increases in PPF when grown under high compared with low PPF (Prioul and Bourdu, 1973). Validation of this point would require quantification through the appropriate exchanges in PPF treatments, but the calculated extent of this ability to adjust in the current experiment was small compared with other more adapted plant species. Therefore, these data cannot be interpreted as indicating *Zantedeschia* 'Best Gold' is obligatively adapted to sun or shade habitats. Rather *Zantedeschia* 'Best Gold' has a restricted level of adjustment of photosynthetic rate at both high and low PPF regimes. Light saturation at PPF regimes close to that of growth is common for many plant species, but not all species respond in the same manner (Bowes et al., 1972; Pallas and Samish, 1974). Shade grown plants of other members of the *Araceae* attained higher net photosynthetic rates than those under full sunlight (O'Hair and Asokan, 1986; Schaffer and O'Hair, 1987). However, PPF levels of full sunlight reported by these authors were at least twice those used in the experiment reported here. It is, therefore, possible that the high PPF regime used in the current experiment was not high enough to result in a reduction in assimilation rate during determination of PPF response curves. The low and high PPF treatments used in the current experiment were designed to equate to total daily integrals of PPF experienced outdoors in Palmerston North during winter and summer, respectively. However, the daily variation in extremes in PPF, experienced outdoors, were not tested. Since leaves of *Zantedeschia* 'Best Gold'

were readily able to acclimate to the PPF regime under which they were grown, data derived from treatments comprising PPF regimes with similar daily variation would be required before more direct comparisons to reports by O'Hair and Asokan (1986) or Schaffer and O'Hair (1987) could be made.

The increased apparent quantum yield under the low PPF regimes (Figure 5.6 and Figure 5.7) allows for a high rate of CO_2 fixation at low PPF, as is typical of plant species with an ability to acclimate to shade habitats. While acclimation to low PPF through an enhancement in apparent quantum yield was found for *Zantedeschia* 'Best Gold', the actual values obtained were low compared with those for other plant species (McCree, 1972; Bjorkman and Demmig, 1987). Accurate measurements of quantum yield need to be carried out under conditions of saturating CO_2 and presented in terms of PPF actually absorbed. McCree (1972) illustrated a 33% increase in quantum yield of sugar beet leaves as a result of increasing the CO_2 concentration from 350 to 600 ppm. In addition, quantum yield for leaves of taro and *Alocasia macrorrhiza* Schott were reduced by 22% when presented in terms of incident rather than absorbed PPF (Bjorkman and Demmig, 1987). Since the data reported here were derived under conditions of limiting CO_2 concentration and reported in terms of incident PPF, the low value for apparent quantum yield might be expected. Under a low PPF regime the decline in apparent quantum yield with increased temperature (Figure 5.6) has been reported for other C_3 plant species, and this may have been associated with an increase in oxygen inhibition of photosynthesis (McCree, 1972; Jolliffe and Tregunna, 1973; Ehleringer and Bjorkman, 1977).

The numerical product of quantum yield and specific leaf weight is a useful overall measure of the capacity of the leaf to intercept photons at low PPF (Kwesiga et al., 1986). Across all temperatures this capacity to intercept photons was, on average, 60% greater under the low PPF regime than under high PPF. This greater capacity to intercept photons indicates that the ability to acclimate to low PPF regimes is not only through changes in specific leaf weight. As found in other plant species differences in specific leaf weight may not totally explain differences in assimilation capacity under low PPF, with differences in chlorophyll and protein content, chloroplast size, thylakoid to grana ratio, and stomatal resistance, being other potential causes of this enhancement (Boardman, 1977; Bjorkman, 1981; Bennett et al., 1982; Inaba, 1984; Givnish, 1988).

The reduction in apparent quantum yield under the combination of high PPF and low temperatures is indicative of photoinhibition. The depression in photosynthetic rate at cooler temperatures resulting in an excess of quanta and excitation energy within the photosynthetic reaction centres and/or production of membrane damaging free radicals, has previously been reported (Bjorkman, 1981). An alternative mechanism of photoinhibition has also been suggested, where long periods of exposure to high PPF result in an alteration

of abscisic acid levels (Correia et al., 1990). As a result, photosynthetic rate is reduced without any evidence of increased concentration of leaf assimilates.

Temperature optima for photosynthetic rates reported here were similar to those reported for potato (*Solanum tuberosum* L.) and taro (Dwelle et al., 1981; Sato et al., 1978). Under high PPF the optimum temperature for photosynthesis per unit leaf area was 28 C (Figure 5.4). However, under the low PPF regime the inability to detect differences in photosynthetic rate per unit leaf area with increasing temperature was a result of increased specific leaf weight. When expressed on a leaf weight basis the optimum temperature for photosynthesis was 28 C regardless of PPF (Figure 5.5). This is in contrast with previous reports that PPF and temperature interact with photosynthetic rate so that the optimal temperature for assimilation increases with increased PPF. With plants of *Solidago virgaurea* L. the optimal temperature for photosynthesis increased by approximately 5 C for plants grown at a PPF of approximately 160 $\mu mol \cdot m^{-2} \cdot s^{-1}$ compared with 40 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (Bjorkman and Holmgren, 1966).

Rates of dark respiration were within the bounds reported for other species (Zelitch, 1971). In addition, the greater rate of dark respiration under the high PPF compared with the low PPF regime was similar to that reported for other species (Bjorkman, 1981). However, the expected increase in respiration rate with increased temperature did not occur. Increasing rates of dark respiration with increasing temperature have been reported for other members of the *Araceae* between 20 and 37 C (Sato et al., 1978; Miura and Osada, 1981). The estimation of dark respiration by quantification of the post illumination burst of CO_2 has been reported to be inaccurate (Sharkey, 1988). Such inaccuracies result from post illumination assimilation of CO_2, as well as continued mitochondrial respiration. While this methodology may have resulted in less accuracy when conducted during the daily photoperiod, the inability to detect temperature differences when conducted 2 h after the termination of the photoperiod can not be attributed to this. The previously discussed measurement error (refer Section 5.3.3) may, therefore, have resulted in this discrepancy.

Over the daily 10 h period where temperature and PPF regimes were constant, changes in photosynthetic rate were minimal (Figure 5.2). In other crops reduction in the rate of net photosynthesis during the lighting period have ranged from 0% to 73%, being highly dependent on species (Pallas, 1973). In addition, Potter and Breen (1980) reported that while the assimilation rate declined by 30% for sunflower (*Helianthus annuus* L.) leaves that were nearly fully expanded, the rate declined by only 10% for less mature leaves. Since the experiment reported here used leaves that had attained only 75% or more of their maximum expansion, the minimal change may have resulted from this relative leaf immaturity in addition to being species related.

Evidence supporting the possibility of a direct photoinhibition effect of extended periods of exposure to high PPF, resulting in reduced photosynthetic rates are well known (Anderson and Osmond, 1987). Other members of the *Araceae* have shown an obligative requirement for shade, and consequent photoinhibition at high PPF (Sato et al., 1978; Schaffer and O'Hair, 1987). However, such reports were based on leaves at or beyond full maturity. Using another member of the *Araceae*, e.g. Konjak (*Amorphophallus konjac* K. Koch), Miura and Osada (1981) did not detect photoinhibition under high PPF until after leaves had developed beyond the stage of maximum leaf area. Hence, the less than 100% expansion stage used in the current experiment may explain why photoinhibition under the high PPF regime was not detected during the determination of the PPF response curves (Figure 5.3). In addition, the change in photosynthetic rate with PPF during ontogeny (Figure 5.10 and Figure 5.11) was interpreted as indicating that response curves of photosynthetic rate as a function of PPF may have been quite different if conducted at a later stage of ontogeny.

Changes in photosynthetic rate with plant ontogeny are well known, with rates typically declining with later leaf positions (Ticha et al., 1985). In the current experiment, photosynthetic rates under the high PPF regime followed a similar progression, but under the low PPF regime the photosynthetic rate increased with plant ontogeny (Figure 5.10 and Figure 5.11). However, neither of these changes in rate consistently coincided with changes in tuber growth. This is in contrast to examples with other plant species, where changes in source activity coincided with changes in sink demand (Moorby, 1968; Chatterton et al., 1972; Habeshaw, 1973; Thorne and Koller, 1974). RGR_T generally increased with increasing temperature (refer Chapter 4), but photosynthetic rate per unit leaf area did not increase with increased temperature under the low PPF regime (Figure 5.4). Clearly then, a change in sink strength during ontogeny is not the only factor influencing photosynthetic rate in *Zantedeschia*. The improved correlation between RGR_W and photosynthetic rate per unit leaf weight over that for per unit leaf area (Table 5.1) corroborates the findings in Chapter 3 where the correlation between RGR_W and LWP was superior to that of NAR. Examples of increased growth rates correlating to increased photosynthetic activity have been reported (Lambers, 1987). However the lack of any significant correlation between RGR_T and photosynthetic rate suggests that while the photosynthetic rate forms an integral part of supplying photoassimilates, the partitioning of these assimilates and, therefore, harvestable yield may be determined by other factors (e.g. TWP, refer Chapter 4).

In summary the photosynthetic rate of leaves of *Zantedeschia* 'Best Gold' has the ability to acclimate to increased cultivation PPF and temperature. However, the acclimation to any one environment is less than complete, i.e.,

1) any gain from the ability to acclimate to high PPF, through greater P_{max}, must be contrasted with a simultaneous reduction in quantum yield,

2) any gain from the ability to acclimate to low PPF, through increased quantum yield, must be contrasted with a simultaneous reduction in P_{max}.

From this it is concluded that *Zantedeschia* 'Best Gold' is a shade tolerant selection, being neither obligatively adapted to sun nor shade habitats. In addition, while net photosynthetic rate was correlated with the relative growth rate of the total plant, this parameter did not correlate with tuber growth, which is the harvestable yield.

5.5 References

Acock, B., D.A. Charles-Edwards, D.J. Fitter, D.W. Hand, L.J. Ludwig, J.W. Wilson and A.C. Withers. 1978. The contribution of leaves from different levels within a tomato crop to canopy net photosynthesis: An experimental examination of two canopy models. J. Expt. Bot. 29:815-827.

Acock, B., M.C. Acock and D. Pasternak. 1990. Interactions of CO_2 enrichment and temperature on carbohydrate production and accumulation in muskmelon leaves. J. Amer. Soc. Hort. Sci. 115:525-529.

Anderson, J.M. and C.B. Osmond. 1987. Shade-sun responses: compromises between acclimation and photoinhibition, p. 1-38. In: D.J. Kyle, C.B. Osmond and C.J. Arntzen (eds.). Photoinhibition, Vol 9. In: J. Barber (ed.). Topics in Photosynthesis. Elsevier, Amsterdam.

Bennett, K.J., H.G. McPherson and I.J. Warrington. 1982. Effect of pretreatment temperature on response of photosynthesis rate in maize to current temperature. Australian J. Plant Physiol. 9:773-781.

Biddington, N.L. 1986. The effects of mechanically-induced stress in plants - a review. Plant Growth Regulat. 4:103-123.

Bjorkman, O. 1981. Responses to different quantum flux densities, p. 57-107. In: O.L. Lange, P.S. Nobel, C.B. Osmond and H. Ziegler (eds). Encyclopedia of Plant Physiology, New Series vol. 12A, Physiological Plant Ecology. Springer Verlag, Berlin.

Bjorkman, O. and B. Demmig. 1987. Photon yield of O_2 evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. Planta 170:489-504.

Bjorkman, O. and P. Holmgren. 1966. Photosynthetic adaptation to light intensity in plants native to shaded and exposed habitats. Physiol. Plant. 19:854-859.

Boardman, N.K. 1977. Comparative photosynthesis of sun and shade plants. Annu. Rev. Plant Physiol. 28:355-377.

Bowes, G., W.L. Ogren and R.H. Hageman. 1972. Light saturation, photosynthesis rate, RuDP carboxylase activity, and specific leaf weight in soybeans grown under different light intensities. Crop Sci. 12:77-79.

Bunce, J.A. 1985. Effects of day and night temperature and temperature variation on photosynthetic characteristics. Photosyn. Res. 6:175-181.

Caulfield, F. and J.A. Bunce. 1988. Comparative responses of photosynthesis to growth temperature in soybean (*Glycine max* L. Merrill) cultivars. Can. J. Plant Sci. 68:419-425.

Chatterton, N.J., G.E. Carlson, W.E. Hungerford and D.R. Lee. 1972. Effect of tillering and cool nights on photosynthesis and chloroplast starch in pangola. Crop Sci. 12:206-208.

Connor, D.J. and V.O. Sadras. 1992. Physiology of yield expression in sunflower. Field Crops Res. 30:333-389.

Correia, M.J., M.M.C. Chaves and J.S. Pereira. 1990. Afternoon depression in photosynthesis in grapevine leaves - evidence for a high light stress effect. J. Expt. Bot. 41:417-426.

Dwelle, R.B., G.E. Kleinkopf and J.J. Pavek. 1981. Stomatal conductance and gross photosynthesis of potato (*Solanum tuberosum* L.) as influenced by irradiance, temperature, and growth stage. Potato Res. 24:49-59.

Ehleringer J. and O. Bjorkman. 1977. Quantum yields for CO_2 uptake in C_3 and C_4 plants: Dependence on temperature, CO_2, and O_2 concentration. Plant Physiol. 59:86-90.

Geiger, D.R. 1976. Effects of translocation and assimilate demand on photosynthesis. Can. J. Bot. 54:2337-2345.

Giaquinta, R. 1978. Source and sink leaf metabolism in relation to phloem translocation. Carbon partitioning and enzymology. Plant Physiol. 61:380-385.

Givnish, T.J. 1988. Adaptation to sun and shade: a whole-plant perspective. Australian J. Plant Physiol. 15:63-92.

Habeshaw, D. 1973. Translocation and the control of photosynthesis in sugar beet. Planta 110:213-226.

Ho, L.C., R.G. Hurd, L.J. Ludwig, A.F. Shaw, J.H.M. Thornley and A.C. Withers. 1984. Changes in photosynthesis, carbon budget and mineral content during the growth of the first leaf of cucumber. Ann. Bot. 54:87-101.

Hodanova, D. 1979. Sugar beet canopy photosynthesis as limited by leaf age and irradiance. Estimation by models. Photosynthetica 13:376-385.

Inaba, K. 1984. Effect of shading on leaf anatomy in Konjak plant. Jpn. J. Crop Sci. 53:243-248.

Irvine, J.E. 1975. Relations of photosynthetic rates and leaf and canopy characteristics to sugarcane yield. Crop Sci. 15:671-676.

Jassby, A.D. and T. Platt. 1976. Mathematical formulation of the relationship between photosynthesis and light for phytoplankton. Limnology and Oceanography 21:540-547.

Jolliffe, P.A. and E.B. Tregunna. 1973. Environmental regulation of the oxygen effect on apparent photosynthesis in wheat. Can. J. Bot. 51:841-853.

Kwesiga, F.R., J. Grace and A.P. Sandford. 1986. Some photosynthetic characteristics of tropical timber trees as affected by the light regime during growth. Ann. Bot. 58:23-32.

Laing, W.A. 1985. Temperature and light response curves for photosynthesis in kiwifruit (*Actinidia chinensis*) cv. Hayward. N.Z. J. Agr. Res. 28:117-124.

Lambers, H. 1987. Does variation in photosynthetic rate explain variation in growth rate and yield ? Neth. J. Agr. Sci. 35:505-519.

Massimino, D. M. Andre, C. Richaud, A. Daguenet, J. Massimino and J. Vivoli. 1980. Evolution horaire au cours d'une journee normale de la photosynthese, de la transpiration, de la respiration foliaire et racinaire et de la nutrition N.P.K. chez *Zea mays*. Physiol. Plant. 48:512-518.

McCree, K.J. 1972. The action spectrum, absorbance and quantum yield of photosynthesis in crop plants. Agr. Meterol. 9:191-216.

Miura, K. and A. Osada. 1981. Effect of shading on photosynthesis, respiration, leaf area and corm weight in Konjak plants (*Amorphophallus konjac* K. Koch.). Jpn. J. Crop Sci. 50:553-559.

Moorby, J. 1968. The influence of carbohydrates and mineral nutrient supply on the growth of potato tubers. Ann. Bot. 32:57-68.

Mortensen, L.M. and R. Moe. 1983. Growth responses of some greenhouse plants to environment. V. Effect of CO_2, O_2 and light on net photosynthetic rate in *Chrysanthemum morifolium* Ramat. Scientia Hort. 19:133-140.

O'Hair, S.K. and M.P. Asokan. 1986. Edible aroids: taxonomy and horticulture. Hort. Rev. 9:43-99.

Pallas, J.E. 1973. Diurnal changes in transpiration and daily photosynthetic rate of several crop plants. Crop Sci. 13:82-88.

Pallas, J.E. and Y.B. Samish. 1974. Photosynthetic response of peanut. Crop Sci. 14:478-482.

Paul, M.J., D.W. Lawlor and S.P. Driscoll. 1990. The effect of temperature on photosynthesis and carbon fluxes in sunflower and rape. J. Expt. Bot. 41:547-555.

Potter, J.R. and P.J. Breen. 1980. Maintenance of high photosynthetic rates during the accumulation of high leaf starch levels in sunflower and soybean. Plant Physiol. 66:528-531.

Prioul, J.L. and R. Bourdu. 1973. Graphical display of photosynthetic adaptability to irradiance. Photosynthetica 7:405-407.

Sato, T., M. Kawai and T. Fukuyama. 1978. Studies on matter production of taro plant (*Colocasia esculenta* Schott). I. Changes with growth in photosynthetic rate of single leaf. Jpn. J. Crop Sci. 47(3):425-430.

Schaffer, B. and S.K. O'Hair. 1987. Net CO_2 assimilation of taro and cocoyam as affected by shading and leaf age. Photosyn. Res. 11:245-251.

Sharkey, T.D. 1988. Estimating the rate of photorespiration in leaves. Physiol. Plant. 73:147-152.

Suzuki, M. and D. Mortimer. 1973. Sugar concentration gradients of the sugar beet plant in relation to translocation. Can. J. Bot. 51:1733-1739.

Thorne, J.H. and H.R. Koller. 1974. Influence of assimilate demand on photosynthesis, diffusive resistances, translocation, and carbohydrate levels of soybean leaves. Plant Physiol. 54:201-207.

Ticha, I., F. Catsky, D. Hodanova, F. Pospisilova, M. Kase and S. Sestak. 1985. Gas exchange and dry matter accumulation during leaf development, p. 157-216. In: Z. Sestak (ed.). Tasks for vegetation science 11: Photosynthesis during leaf development. Dr W. Junk Publishers, Dordrecht.

Valenzuela, H.R. 1990. Shade tolerance and photosynthesis of cocoyam [*Xanthosoma sagittifolium* (L.) Schott]. Doctor of Philosophy Thesis, University of Florida, 91 pp.

Verkleij, F.N. and H. Challa. 1988. Diurnal export and carbon economy in an expanding source leaf of cucumber at contrasting source and sink temperature. Physiol. Plant. 74:284-293.

Wang, Y. and P.J. Breen. 1986. Growth and photosynthesis of easter lily in response to flower bud removal. J. Amer. Soc. Hort. Sci. 111:442-446.

Wardlaw, I.F. 1968. The control and pattern of movement of carbohydrates in plants. Bot. Rev. 34:79-105.

Warren-Wilson, J. 1966a. Effect of temperature on net assimilation rate. Ann. Bot. 30:753-761.

Warren-Wilson, J. 1966b. An analysis of plant growth and its control in arctic environments. Ann. Bot. 30:383-402.

Warrington, I.J., M. Peet, D.T. Patterson, J. Bunce, R.M. Haslemore and H. Hellmers. 1977. Growth and physiological responses of soybean under various thermoperiods. Australian J. Plant Physiol. 4:371-380.

Woodward, R.G. 1976. Photosynthesis and expansion of leaves of soybean in two environments. Photosynthetica 10:274-279.

Zelitch, I. 1971. Dark respiration and photorespiration. p. 127-171. In: Photosynthesis, photorespiration, and plant productivity. Academic Press, New York.

6 CARBOHYDRATE CONCENTRATION OF *Zantedeschia* 'Best Gold' IN RESPONSE TO TEMPERATURE AND PHOTOSYNTHETIC PHOTON FLUX

6.1 Introduction

Improvements in the partitioning of photoassimilates between those organs which contribute to economic yield and the rest of the plant, have generally been responsible for the improved yields obtained from many crops over the last century (Gifford et al., 1984; Ho, 1988). The preceding chapters have endeavoured to provide the foundation of information required to subsequently develop crop growth and dry matter partitioning models for *Zantedeschia* 'Best Gold' which would be able to account for environmentally-induced differences in partitioning to tubers; those organs which contribute to economic yield. However, to develop a greater understanding of the potential mechanisms controlling this partitioning, an investigation into the various biochemical components of the entire plant and its various organs is required.

Some 90 - 95% of the dry weight of the plant is derived via the photosynthetic process (e.g. Irvine, 1975) and hence carbohydrates, the primary products of photosynthesis, provide a major component of dry weight with which to examine the mechanisms of partitioning. Those carbohydrates not used by the metabolic source in respiration and growth may be exported to other organs, i.e., other metabolic sinks, or temporarily stored before being subsequently exported. Sucrose and starch are primary photosynthetic products, and sucrose is the major translocated carbohydrate in many genera, including many bulb forming crops, e.g. tulip (*Tulipa gesneriana* L.; Ho and Rees, 1975) and narcissus (*Narcissus sp.*; Chen, 1969). Depending on the plant genera, sucrose and/or starch also serve as the temporary foliar storage carbohydrates that accumulate in the light and are remobilized in the dark (Gordon et al., 1980; Fondy and Geiger, 1982). Monosaccharides such as glucose and fructose are frequently found in plant tissue, being intermediaries in the metabolism of sucrose and starch.

While the primary form of carbohydrate in the storage organ of root crops such as sugar beet (*Beta vulgaris* L.) and carrot (*Daucus carota* L.) is soluble carbohydrate (Milford, 1973; Lester et al., 1982), the primary form in numerous bulb, tuber and corm forming crops is starch (Hashad et al., 1956; Rees, 1972; Brouk, 1975; Onwueme, 1978; van Es and Hartmans, 1987). The composition of carbohydrates in *Zantedeschia* tubers is currently not known.

Concomitant with seasonal regrowth from storage organs, starch stored from the previous season was found to be remobilized into soluble carbohydrates and utilised in the formation

of new structures (Ho and Rees, 1975 & 1976; Ross and Davies, 1985). In addition to this decline in stored starch, the quantity of non-starch ethanol-insoluble dry matter within the storage organ also declined. The rate of translocation of carbohydrates from the storage organ progressively declined as leaf area became established, indicating the ability of the recently expanded leaf area to supply carbohydrates as newly synthesized photoassimilates.

In this study it has been shown that photosynthetic rate generally increased with increasing temperature and PPF (refer Chapter 5). Such increases in photosynthetic rate will be associated with increased synthesis of photoassimilates. Apart from extrapolating from information on other members of the *Araceae* (Hashad et al., 1956; Brouk, 1975; Onwueme, 1978), just how these newly synthesised photoassimilates are utilized within the leaves and other organs of *Zantedeschia* is not known.

As discussed previously (refer Chapter 5) a number of crop species have shown changes in photosynthetic rate upon initiation of new sinks or manipulation of sink strength (Werner; 1935; Moorby, 1968; Chatterton et al., 1972; Habeshaw, 1973; Thorne and Koller, 1974; Rapoport and Loomis, 1985; Wang and Breen, 1986). Such changes in photosynthetic activity have been correlated with changes in synthesis and translocation of carbohydrates. However, not all investigations of this nature have reported such illustrations of the existence of positive and/or negative feed-back mechanisms (Geiger, 1976; Farrar and Farrar, 1987). Since differences in photosynthetic rate were noted between treatments, and with development (refer Chapter 5), it was considered possible that such changes could be associated with changes in carbohydrate concentration.

While the diurnal decline in photosynthetic rate was small (refer Chapter 5), such events have in some cases been correlated with the accumulation of photoassimilates (Chatterton et al., 1972; Warrington et al., 1977). This prior knowledge coupled with previous reports of diurnal differences in accumulation and translocation of foliar carbohydrates (Gordon et al., 1980; Fondy and Geiger, 1982; Forney and Austin, 1988), inferred that the time within the photoperiod at which photosynthetic rate and carbohydrate concentration were determined, would be important in detecting differences between treatments and interpreting results.

Regardless of whether the primary form of storage carbohydrate was soluble or starch, increases in storage organ growth have been correlated with increases in storage carbohydrate (Bradbury, 1953; Hashad et al., 1956; Lovell and Booth, 1967; Terry, 1968; Das Gupta, 1969; Obata-Sasamoto and Suzuki, 1979). In the storage root of sugar beet this correlation resulted in the concentration of storage carbohydrates remaining constant

throughout most of the season, regardless of growing environment (Terry, 1968; Milford and Thorne, 1973; Milford et al., 1988). In contrast, starch concentrations in the storage organs of taro (*Colocasia esculenta* Schott), potato (*Solanum tuberosum* L.), and tulip increased throughout growth (Hashad et al., 1956; Plaisted, 1957; Ching, 1970; Aung et al., 1973).

In traditional growth analysis frequent attempts are made to relate net assimilation rate (NAR) and total plant relative growth rate (RGR_w) to source and sink activities of the whole plant. However, opinion is divided as to whether 'sink strength' should apply only to the utilization of assimilates for structural material or whether it should also include storage materials (Wareing and Patrick, 1975). Warren Wilson (1972) suggested that justification for the inclusion of storage materials is simplest in the case of a plant that is conceived as containing a negligible proportion of storage materials. In such cases, the source strength (assimilate produced) is equal to the sink strength (assimilates utilized in structural growth), and the source and sink activities are obtained by dividing the change in dry matter by the size of the productive unit, represented by leaf area for the source and plant dry weight for the sink. In this situation the leaf area ratio, i.e. leaf area/plant dry weight, can be regarded as the ratio of source size to sink size. In reality, however, all plants contain stored assimilates. Accordingly, whereas source activity can still be represented by NAR, Warren Wilson (1972) suggested sink activity should be represented by the relative growth rate of structural material (RGR_{str}), and source-sink ratio by leaf area/plant structural dry weight. Hence use of RGR_w or RGR_T as an estimate of sink activity, as used in Chapters 3 and 4, is considered only appropriate under steady-state conditions in which growth is not accompanied by changes in the proportion of structural and storage material. Using impatiens (*Impatiens parviflora* DC.), Warren Wilson (1972) proposed that RGR_{str} was largely controlled by temperature, whilst photosynthesis depended on temperature, PPF and CO_2 concentration. In adopting this concept of separating structural and storage dry weight, it was hoped that any evidence of source and/or sink activity in *Zantedeschia* would be more clearly reflected.

With no relevant information available on the actual carbohydrate components and their dynamics within *Zantedeschia*, a primary objective of the current experiment was to describe these. In addition it was hoped that quantification of any environmentally-induced differences in these parameters would provide greater understanding of the mechanisms controlling growth and development of this plant.

6.2 Materials and Methods

6.2.1 Cultural and environmental

With the exception of the determination of the concentration of specific soluble carbohydrates and diurnal changes in carbohydrate concentration, data presented in this chapter were derived from the experiment described previously (refer Section 3.2). Cultural procedures (refer Section 3.2.1) and environmental conditions (refer Section 3.2.2) were the same.

For the determination of the concentration of specific soluble carbohydrates, plants of the *Zantedeschia pentlandii*-like selection 'Best Gold' were initially grown as described in Section 3.2.1. At the time of plant selection and potting, rather than being placed into controlled environment rooms (CE) rooms, plants were retained under greenhouse conditions (minimum air temperature 15 C, ventilation at 24 C), and were watered by capillary matting on drained benches.

Cultural procedures and environmental conditions used for determining diurnal changes in carbohydrate concentration were the same as those described in Sections 3.2.1 and 3.2.2, except placement in the CE treatments commenced 4 weeks later and treatments were limited to day/night temperature 22/16 C (low PPF regime) and 28/22 C (high PPF regime).

6.2.2 Experimental

6.2.2.1 Determination of concentration of specific soluble carbohydrates

Plants were harvested once they had attained 1 - 2 mature leaves (50 days' growth). Leaf (1 to 2 g fresh weight) and tuber tissue (4 to 5 g fresh weight) were extracted for 24 h in 10 ml 80% aqueous ethanol. Preliminary investigations determined an average 82% recovery of soluble sugars using this extraction procedure. Aliquots (10 μl) were filtered through a 0.45 μm filter and injected into a high performance liquid chromatograph (HPLC) equipped with an Aminex (Bio-Rad) carbohydrate column (HPX-87C, 300 mm $\times$ 7.8 mm), deash guard, and refractive index detector. The solvent was water, at a flow rate of 0.6 ml$\cdot$min^{-1} at 75 C and 3.45 MPa pressure. To calibrate and monitor efficiency, every tenth injection comprised a sample composite standard of 106, 108, and 107 ng$\cdot\mu$l^{-1} sucrose, glucose and fructose, respectively.

Changes in the concentration of soluble carbohydrates following vacuum drying were determined using a further sample of plants. Sampled leaf and tuber tissue were vacuum

dried at 0.3 kPa for 48 h at 40 C. Dried plant material was ground to pass a 0.5 mm screen and stored in tightly-capped vials in an air-conditioned room (22 C and 50% relative humidity) until being analyzed using the HPLC as described above.

Carbohydrate concentration was expressed on a dry weight basis using data obtained from paired samples of tissue following vacuum drying. Sucrose, glucose and fructose concentrations of samples were compared with those from tissue extracted and analyzed immediately after destructive harvest.

During cultivation, plants were arranged in a completely randomized design. At the time of sampling a total of nine plants were harvested. Three individual plant replicates were used for each determination of; soluble carbohydrates (fresh), soluble carbohydrates (dry), and percentage dry weight. Data were subjected to analysis of variance using the general linear models procedure of the Statistical Analysis System (SAS; SAS Institute, Inc., Cary, N.C.).

6.2.2.2 Starch and soluble carbohydrate concentration as a function of duration from commencement of daily lighting

Starch and soluble carbohydrate concentrations of samples of the leaf blades and tuber of the entire plant were determined during a daily period of lighting. At the same time, photosynthetic rate of recently-mature leaves was also determined. Leaves were defined as recently-mature when they had reached a minimum of 75% expansion, and the analysis was carried out on the fifth such leaf of selected plants.

Photosynthetic rates were measured as previously outlined (refer Section 5.2.3) except air flow rates were sometimes reduced to 0.356 litre·min^{-1} to account for those instances where minimal differences between inlet and outlet CO_2 concentrations were encountered.

During cultivation plants were arranged in a completely randomized design. Data were recorded from a random sample of 3 plants at approximately 2 h intervals, commencing 15 min prior to the commencement of lighting. The final recording was taken after 11.5 h of the 12 h period of lighting. Samples were harvested, organs separated, and placed at -4 C within 30 min of removal from treatments. All samples were subsequently vacuum dried at 0.3 kPa for 48 h at 40 C. Dried material was ground to pass a 0.5 mm screen and stored in tightly-capped vials in an air-conditioned room (22 C and 50% relative humidity) until analyzed using a method based on that of Haslemore and Roughan (1976), as outlined below. The resulting carbohydrate concentration data were expressed on a dry weight basis. Rates of change with time of photosynthetic rate, soluble carbohydrate concentration, and starch concentration, were determined by regression analysis using the

REG procedure of SAS. Comparison of slopes between treatments for homogeneity were conducted as outlined by Zar (1984).

Determination of soluble carbohydrate concentration

Dried plant material (100 mg) was extracted with 10 ml 62.5% (v/v) methanol for 30 min at 55 C using screw-capped culture tubes (16 mm × 125 mm) with teflon-lined caps. The samples were centrifuged and 4 ml aliquots were transferred to a second series of capped culture tubes, each containing 0.1 ml lead acetate (0.5 M). Standards were prepared by diluting 0, 2, 4, 6, 8, and 10 ml of a 2 $mg \cdot ml^{-1}$ sucrose stock solution, to 10 ml with 62.5% methanol. Aliquots (4 ml) were removed and treated in the same manner as the unknowns to give standards equivalent to 0, 4, 8, 12, 16, and 20% soluble carbohydrates on a dry weight basis, respectively. After 10 min. standing with occasional shaking, 5 ml chloroform was added, and the tubes capped and vortex mixed. Tubes were then briefly centrifuged to aid phase separation. Aliquots (50 μl) were removed from the upper, aqueous phase and added to 1 ml of 5% (w/v) phenol and 4 ml 98% sulphuric acid (specific gravity 1.84). Samples were stood to cool for 60 min. and absorbance read at 490 nm using a Hitachi, model 101, spectrophotometer.

Determination of starch concentration

The residual plant material following soluble carbohydrate extraction, and solution aspiration, was treated with 3 ml 100% methanol at 100 C for 5 min. This was repeated and the washings discarded after centrifugation. Starch standards were prepared by suspending soluble starch (100 mg, AR; British Drug Houses; corrected for moisture) in 10 ml water and placed in a boiling water bath until the suspension became translucent. This was diluted to 50 ml with water. Aliquots (0, 2 and 4 ml) of this stock solution were diluted to 4 ml with water to give standards equivalent to 0, 4 and 8 mg starch. The culture tubes of starch standards and 4 ml aliquots of unknowns were capped firmly and heated for 60 min at 100 C to gelatinise the starch. After cooling, 2 ml sodium acetate buffer (0.25 M, pH 4.5) and 0.1 ml amyloglucosidase (Boehringer Mannheim NZ Ltd; 2.5 $mg \cdot ml^{-1}$ protein in 25 mM sodium citrate, pH 6.0) were added and the samples vortex mixed and incubated at 55 C for 60 min. Samples were finally diluted to 10 ml with water, vortex mixed and centrifuged.

Glucose concentration was determined by incubating 0.2 ml aliquots of the diluted hydrolysate in a final volume of 1 ml water with 2 ml glucose oxidase reagent at 37 C for 60 min. A blank of water together with standards containing 25, 50, 75 and 100 $\mu g \cdot ml^{-1}$ glucose were treated similarly. If the colour of the unknown samples was darker than the glucose standard of highest concentration, a 1 in 10 dilution was carried out. Hydrochloric acid (5 ml, 5 M) was added, samples vortex mixed and absorbances read at

540 nm using a Hitachi, model 101, spectrophotometer.

Glucose oxidase reagent was prepared by dissolving o-dianisidine hydrochloride (120 mg) in 600 ml tris-glycerol buffer, and dissolving in this 180 mg glucose oxidase (Sigma Chemical Company) and 18 mg peroxidase (Horseradish; Sigma Chemical Company). Tris-glycerol buffer was prepared by dissolving 30.2 g Tris in 500 ml water (pH 7.0) and adding 330 ml glycerol (AR).

6.2.2.3 Starch and soluble carbohydrate concentration during plant development

During plant development, total soluble carbohydrate and starch concentration of leaf and tuber tissue were determined using plants sampled from CE rooms (refer Chapters 3 and 4). Samples were taken following grinding of the entire non-senesced leaf or tuber. Dried tissue was ground so as to pass a 0.5 mm screen.

Due to restrictions on time, soluble carbohydrate and starch concentration data could only be collected from three of the day/night temperature treatments (16/10, 22/16, and 28/22 C) under both high and low PPF regimes. In addition, not all harvest dates were able to be analyzed for carbohydrates. Four individual plant replicates from each destructive harvest were analyzed for both soluble carbohydrates and starch using the methods outlined above.

A $\log_e$ transformation of the dry weight data was used to meet the statistical assumption of homogeneity of variance throughout the period of study (Causton and Venus, 1981). Transformed tuber starch data, collected during the period of increasing tuber dry weight, were fitted to the Gompertz function, equation (6.1) (Causton and Venus, 1981), utilizing the non-linear regression parameter estimation procedure of SAS. Justification for the use of this type of sigmoidal curve was as discussed previously (refer Section 3.2.3). A constant was added to all transformed data prior to curve fitting to eliminate negative values. Because the Gompertz function is asymptotic, the magnitude of the added constant influences the calculated value of β_{T_a}. Hence the addition of a constant to all values prior to curve fitting eliminated any value of subsequent interpretation of differences in the value of β_{T_a}.

$$\log_e Ts = \alpha_{Ts} \cdot e^{(-e^{(\beta_{Ts} - \kappa_{Ts} \cdot t)})} \quad (6.1)$$

where;

$\log_e Ts$ = $\log_e$ transformed value of tuber starch dry weight
α_{Ts} = upper asymptote of tuber starch dry weight
β_{Ts} = a measure of the starting size of tuber starch dry weight
κ_{Ts} = rate constant of tuber starch dry weight as a function of size
t = time
e = the base of natural logarithms

During this time period tuber starch relative growth rate (RGR_{Ts}), as a function of time, was calculated using equation (6.2), being the first differential of equation (6.1) (Causton and Venus, 1981).

$$RGR_{Ts} = \alpha_{Ts} \cdot \kappa_{Ts} \cdot e^{(\beta_{Ts} - \kappa_{Ts} \cdot t)} \cdot e^{(-e^{(\beta_{Ts} - \kappa_{Ts} \cdot t)})} \quad (6.2)$$

Having determined tuber starch and soluble carbohydrate concentration, the non-carbohydrate residual, or tuber structural dry weight (T_{str}) was calculated using equation (6.3). Log_e transformed tuber structural dry weight data over the same time period were subsequently fitted to the Gompertz equation as described for tuber starch dry weight above.

$$\log_e Ts = \alpha_{Ts} \cdot e^{(-e^{(\beta_{Ts} - \kappa_{Ts} \cdot t)})} \qquad (6.1)$$

where;

$\log_e Ts$ = $\log_e$ transformed value of tuber starch dry weight ($\log_e$ g)
α_{Ts} = upper asymptote of tuber starch dry weight ($\log_e$ g)
β_{Ts} = a measure of the starting size of tuber starch dry weight ($\log_e$ g)
κ_{Ts} = rate constant of tuber starch dry weight as a function of size ($g \cdot g^{-1} \cdot g^{-1}$)
t = time (days)
e = the base of natural logarithms

During this time period tuber starch relative growth rate (RGR_{Ts}), as a function of time, was calculated using equation (6.2), being the first differential of equation (6.1) (Causton and Venus, 1981).

$$RGR_{Ts} = \alpha_{Ts} \cdot \kappa_{Ts} \cdot e^{(\beta_{Ts} - \kappa_{Ts} \cdot t)} \cdot e^{(-e^{(\beta_{Ts} - \kappa_{Ts} \cdot t)})} \qquad (6.2)$$

Having determined tuber starch and soluble carbohydrate concentration, the non-carbohydrate residual, or tuber structural dry weight (T_{str}) was calculated using equation (6.3). Log_e transformed tuber structural dry weight data over the same time period were subsequently fitted to the Gompertz equation as described for tuber starch dry weight above.

$$Tstr = T - Ts - Tsol \qquad (6.3)$$

where;

Tstr = tuber structural dry weight (g)
T = total tuber dry weight (g)
Ts = tuber starch dry weight (g)
Tsol = tuber soluble carbohydrate dry weight (g)

Log_e transformed leaf starch dry weight data were fitted to the logarithmic form of the Gompertz function, i.e., equation (6.4) (Causton and Venus, 1981), utilizing the non-linear regression parameter estimation procedure of SAS. During this time period, relative leaf starch weight rate (RLSWR), as a function of time, was calculated as outlined by Causton and Venus (1981), using equation (6.5).

$$\log_e Ls = \alpha_{Ls} - e^{(\beta_{Ls} - \kappa_{Ls} \cdot t)} \qquad (6.4)$$

where;

$\log_e Ls$ = $\log_e$ transformed value of leaf starch dry weight ($\log_e$ g)
α_{Ls} = upper asymptote of leaf starch dry weight ($\log_e$ g)
β_{Ls} = a measure of the starting size of leaf starch dry weight ($\log_e$ g)
κ_{Ls} = rate constant of leaf starch dry weight as a function of size ($g \cdot g^{-1} \cdot g^{-1}$)
t = time (days)
e = the base of natural logarithms

$$RLSWR = \kappa_{Ls} \cdot e^{(\beta_{Ls} - \kappa_{Ls} \cdot t)} \qquad (6.5)$$

6.3 Results

6.3.1 Leaf and tuber soluble carbohydrate composition

Both leaf and tuber material contained measurable quantities of sucrose, glucose and fructose (Figure 6.1 and Figure 6.2). When determined immediately after harvest, foliar sucrose and fructose concentrations were similar, and were more than three-fold greater than that of glucose. In contrast, sucrose was the most abundant soluble carbohydrate within the tuber. While tuber glucose and fructose concentrations were similar, averaging 6.3 $mg \cdot g^{-1}$ dry weight, tuber sucrose concentrations were more than four-fold greater.

Differences between the method of sample preparation, i.e., either immediate extraction or after vacuum drying, were not evident for tuber samples ($P \leq 0.05$, Figure 6.2). Similarly differences between the method of sample preparation were not evident for concentrations of foliar glucose and fructose ($P \leq 0.05$), but vacuum drying resulted in more than a three-fold reduction in foliar sucrose ($P \leq 0.01$, Figure 6.1).

For vacuum dried plant material, as used for the rest of the data presented in this chapter, total soluble carbohydrate concentration per unit dry weight was 4.1 ± 0.2% for tubers, and 7.2 ± 0.3% for leaves.

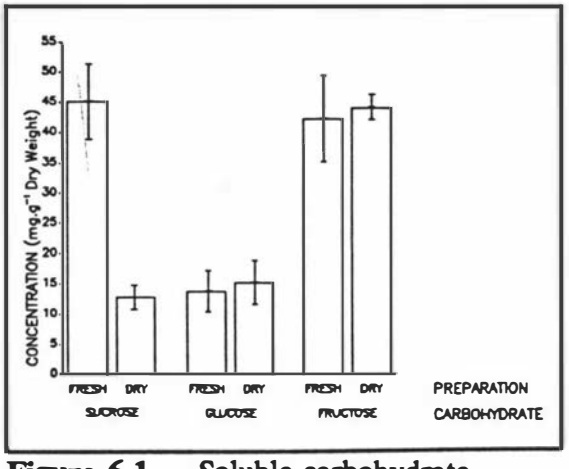

Figure 6.1 Soluble carbohydrate concentration of mature leaves of *Zantedeschia* 'Best Gold' determined either immediately after harvest (fresh) or after vacuum drying (dry). Mean values ± se., n=3.

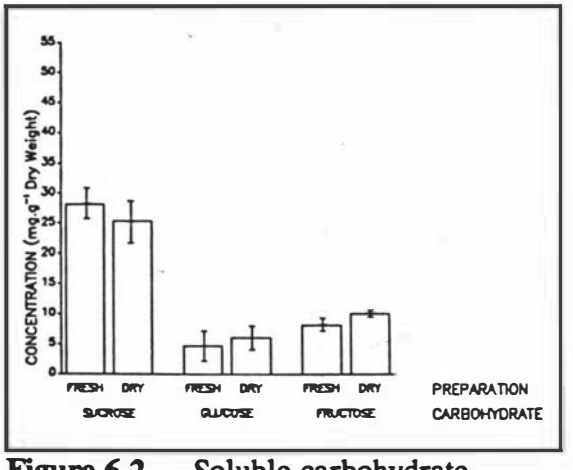

Figure 6.2 Soluble carbohydrate concentration of tubers of *Zantedeschia* 'Best Gold' determined either immediately after harvest (fresh) or following vacuum drying (dry). Mean values ± se., n=3.

6.3.2 Starch and soluble carbohydrate concentration as a function of duration from commencement of daily lighting

During the period of stable environment (i.e., constant; temperature, PPF, and vapour pressure deficit conditions), leaf starch concentrations increased linearly with time ($P \leq 0.001$, Figure 6.3). Between 1 and 9 h of lighting at 28 C, this linear increase with time resulted in a 65% increase in leaf starch concentration. While time did not permit complete carbohydrate analysis of plant material from the 22 C regime, foliar concentrations of starch also increased with time, with no indication of differences in the rate of increase compared with 28 C (data not presented). However, at 1 h after the commencement of lighting leaf starch concentrations were greater at 22 C (9.8 % dry weight) compared with 28 C (4.8 % dry weight; $P \leq 0.01$). During the period of constant environment no marked change in photosynthetic rate (*Pn*) nor foliar soluble carbohydrate were detected ($P \leq 0.10$).

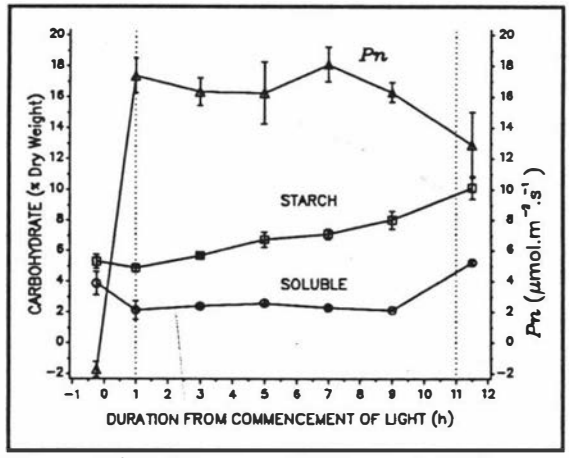

Figure 6.3 Photosynthetic rate (*Pn*) and foliar carbohydrate concentration, as a function of duration of daily lighting, at 28/22 C under high PPF. Mean values ± se., n=3. Broken lines indicate limits of diurnal environmental changeovers.

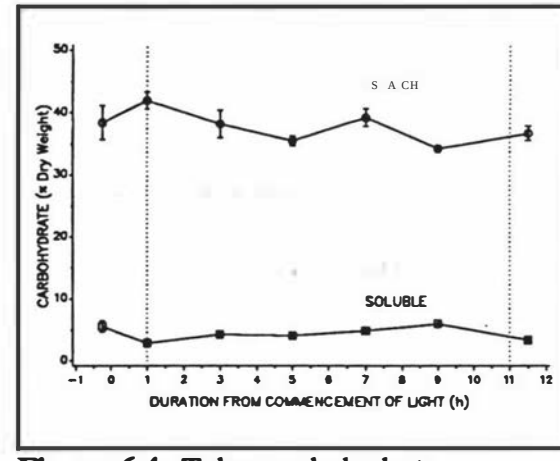

Figure 6.4 Tuber carbohydrate concentration as a function of duration of daily lighting at 28/22 C under high PPF. Mean values ± se., n=3. Broken lines indicate limits of diurnal environmental changeovers.

Tuber starch concentrations declined linearly during the period of stable environment, resulting in a 19% reduction at 28 C between 1 and 9 h ($P \leq 0.05$, Figure 6.4). While actual tuber starch concentration varied between treatments, a linear decline with time was also detected at 22 C (data not presented). Comparison of the slopes for homogeneity did not detect differences between treatments ($P \leq 0.10$). Concomitant with this decline in tuber starch concentration, the soluble carbohydrate concentration of tubers increased linearly over the period of lighting ($P \leq 0.10$, Figure 6.4). While actual concentrations of soluble carbohydrate varied between the two treatment environments, differences in slope were not detected ($P \leq 0.10$). Between 1 and 9 h of lighting at 28 C, soluble

carbohydrate concentration of tubers had increased by 100% compared with 78% at 22 C.

6.3.3 Starch and soluble carbohydrate concentration during plant development

6.3.3.1 Overview

As growth commenced, tuber starch and structural (i.e., non-carbohydrate residual) dry weight declined before commencing a sigmoidal pattern of increase (Figure 6.5). The initial decline in tuber structural dry weight was small relative to the decline in starch, but once tuber growth commenced, the relative rate of starch accumulation was greater than that of structural dry weight. Starch continued to accumulate within the tuber over the period examined.

Under all treatment regimes $\log_e$ tuber starch and structural dry weight followed a sigmoidal pattern of growth, with a family of Gompertz functions adequately describing the progression of tuber starch and structural dry weight ($\log_e$ transformed) during the period of its increase (data not presented). Similarly a family of logarithmic Gompertz functions adequately described the progression of the cumulative quantity of starch and structural dry weight contained within the leaf blades of the total plant (data not presented).

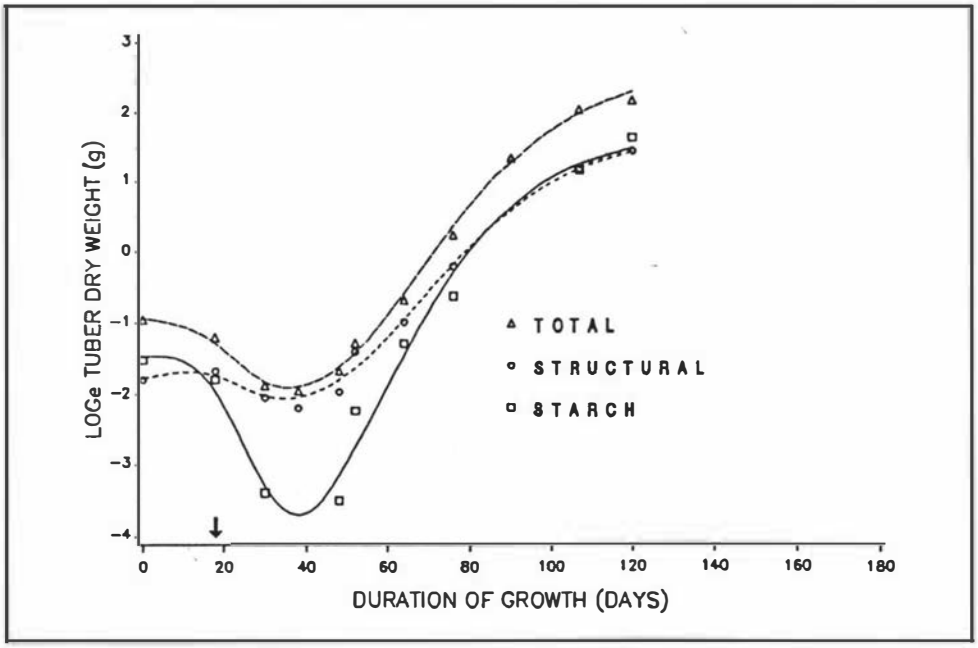

Figure 6.5 Total, structural and starch tuber dry weight ($\log_e$), as a function of time, of *Zantedeschia* 'Best Gold' plants grown at 25 C under high PPF. ↓ indicates day of transfer to treatment environment.

6.3.3.2 Tuber starch concentration

During plant development, changes in tuber starch dry weight (e.g. Figure 6.5), and tuber starch concentration (Figure 6.7), followed a pattern similar to that of total tuber dry weight (refer Chapter 4). As with tuber dry weight, tuber starch concentration initially declined and subsequently followed a sigmoidal pattern of increase.

The extent of the initial decline in tuber starch concentration was greater at higher temperatures and under low PPF (Figure 6.6). Under the high PPF regime the minimum tuber starch concentration declined linearly with increasing temperature ($P \leq 0.001$), compared with a quadratic decline under the low PPF regime ($P \leq 0.10$). Under the high PPF regime an approximate doubling of the temperature of cultivation, i.e., 13 C compared with 25 C, resulted in more than a two-fold reduction in the minimum tuber starch concentration at the higher temperature. In contrast, under the low PPF regime, this same increment in cultivation temperature resulted in more than a six-fold reduction in minimum tuber starch concentration. While cultivation under the low PPF regime compared with under the high PPF regime resulted in less than a one-fold reduction in the minimum tuber starch concentration at 13 C, this reduction was increased to two-fold at 19 C, and three-fold at 25 C.

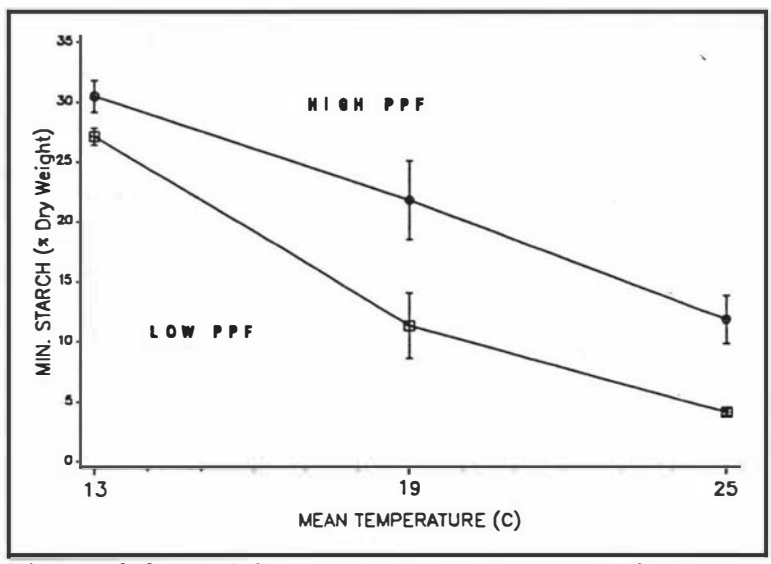

Figure 6.6 Minimum tuber starch concentration as a function of temperature, for plants of *Zantedeschia* 'Best Gold' grown under high (-○-) and low (-□-) PPF regimes. Mean values ± se., n=4.

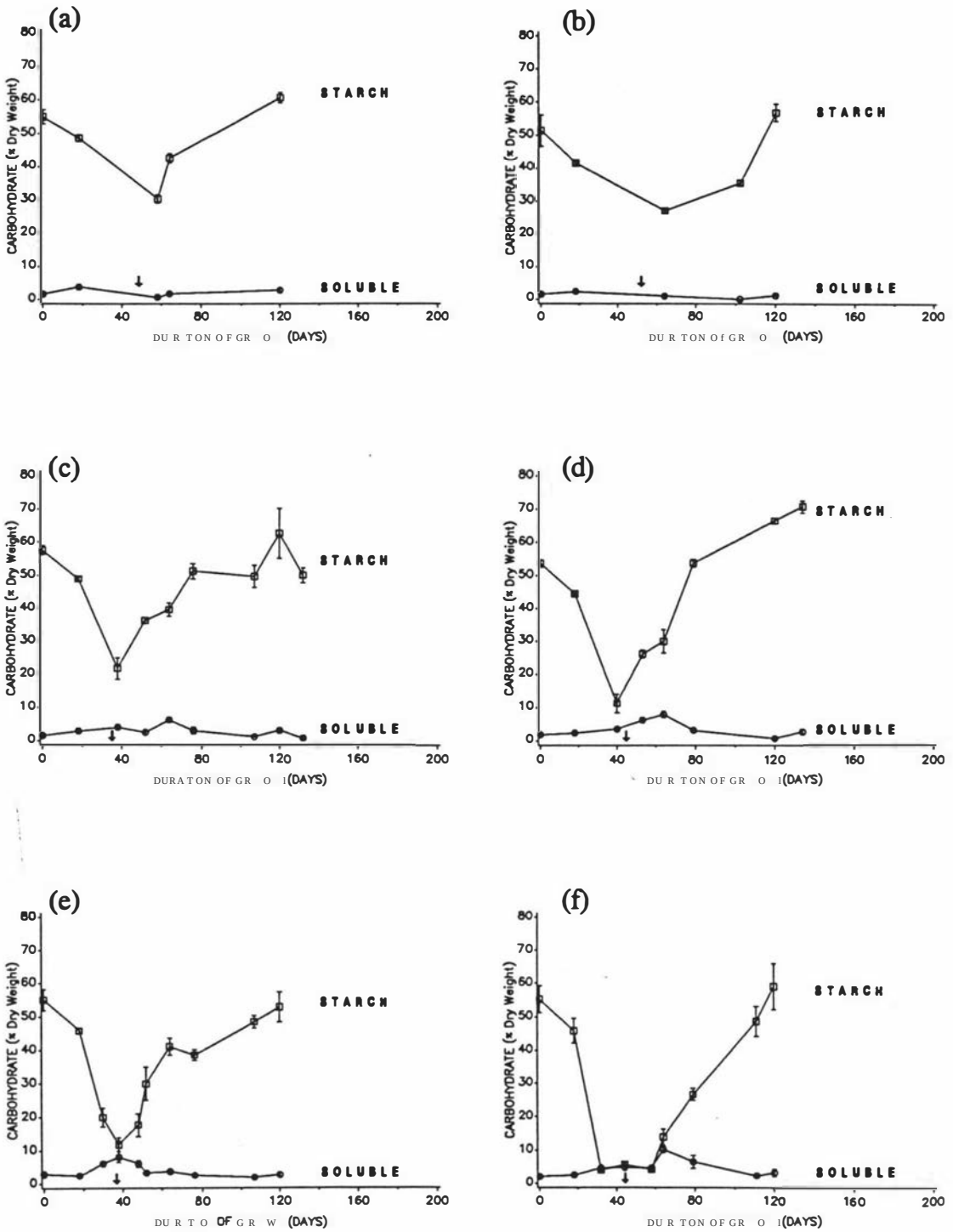

Figure 6.7 Carbohydrate concentration of the tuber as a function of time, for *Zantedeschia* 'Best Gold' grown at three temperatures under high and low PPF regimes. (a) 13 C, high PPF (b) 13 C, low PPF (c) 19 C, high PPF (d) 19 C, low PPF (e) 25 C, high PPF (f) 25 C, low PPF. Mean values ± se., n=4, ↓ indicates commencement of tuber growth.

Under all temperature and PPF regimes changes in tuber starch dry weight were correlated with changes in tuber structural dry weight. Under the high PPF regime this correlation was seen as a positive, linear relationship between $\log_e$ tuber starch dry weight and $\log_e$ tuber structural dry weight (equation (6.6), $P \leq 0.001$, Figure 6.8). Under the low PPF regime, once tuber growth had commenced, the same functional relationship was evident ($P \leq 0.05$). However, under the low PPF regime prior to the commencement of any increase in tuber dry weight, the relationship indicated proportionately larger reductions in tuber starch relative to structural dry weight (equation (6.7), $P \leq 0.001$). Within each of the two phases of development, examination of the slopes for homogeneity did not detect differences between temperature regimes ($P \leq 0.05$). Similarly, differences between temperature and PPF regimes were not evident once tuber growth had recommenced ($P \leq 0.05$).

$$\log_e Y = \log_e X^{1.36} + 0.06 \qquad (r^2 = 0.86,^{***}) \qquad (6.6)$$

$$\log_e Y = \log_e X^{3.5} + 3.9 \qquad (r^2 = 0.74,^{***}) \qquad (6.7)$$

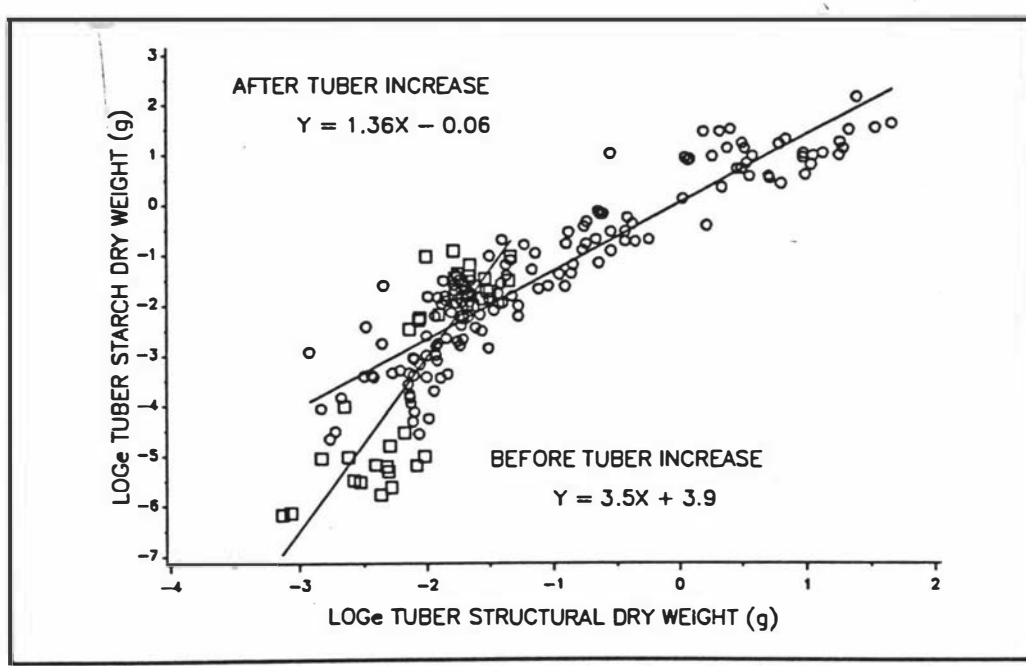

Figure 6.8 Tuber starch dry weight as a function of tuber structural dry weight. Low PPF before (-□-), and both high and low PPF after (-○-), the commencement of tuber growth, for plants grown at three temperatures and two PPF regimes.

The timing of initial increases in tuber starch weight and concentration coincided with the commencement of total tuber dry weight increments, with both weight and concentration subsequently increasing with duration (Figure 6.7). While both tuber starch weight and concentration were greater under the high PPF regime at the commencement of tuber growth, by the termination of the experiment they were greater under the low PPF regime at both 19 and 25 C. This difference between PPF regimes, in final tuber starch weight and concentration, primarily resulted from a slower decline in the relative growth rate of starch accumulation relative to structural dry weight (Figure 6.9). Since carbohydrate determinations of tubers grown at 13 C were not carried out beyond 120 days, it can not be definitely stated that the same result would have occurred at this temperature regime at the final harvest after 200 days. However, if it is assumed that the linear relationship between tuber starch concentration and tuber dry weight was still applicable over this time period, this greater tuber starch weight and concentration under the low PPF regime would also have occurred at this temperature.

6.3.3.3 Tuber soluble carbohydrate concentration

In contrast to tuber starch concentration, tuber soluble carbohydrate concentration increased slightly over the first few weeks from planting. However, over the entire period of growth tuber soluble carbohydrate concentrations remained relatively constant (Figure 6.7). At mean temperatures of 19 and 25 C, brief periods of marked increases in concentration of soluble carbohydrate were detected. These increases in tuber soluble carbohydrate concentration occurred at or soon after the commencement of increased tuber growth (Figure 6.7).

Tuber soluble carbohydrate concentration was not influenced by PPF regime ($P \leq 0.05$), but tubers grown at 13 C contained less than half the concentration of soluble carbohydrates than those at 19 or 25 C ($P \leq 0.05$). Tubers derived from 13 C contained an average $1.7 \pm 0.5\%$ soluble carbohydrate. In contrast tubers derived from 19 or 25 C contained an average $3.9 \pm 0.4\%$.

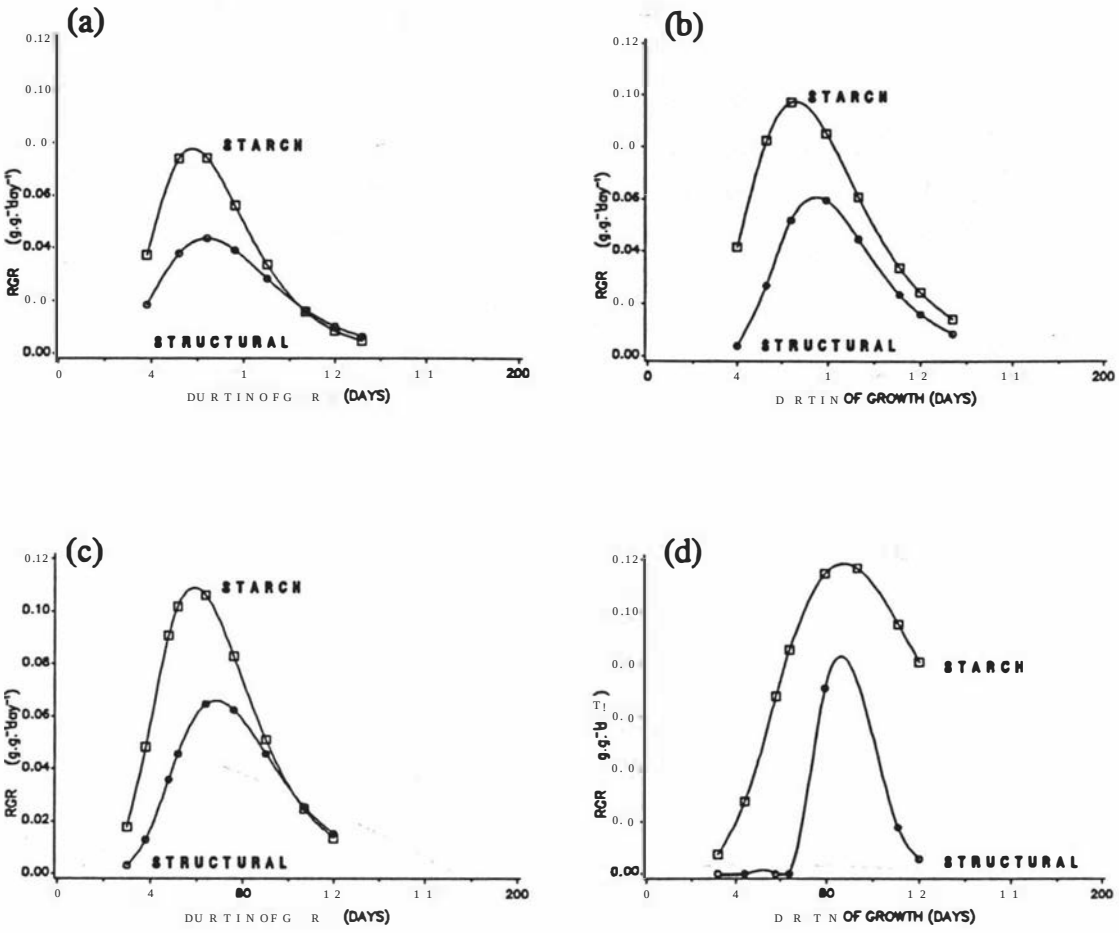

Figure 6.9 Relative growth rate (RGR) of tuber starch and tuber structural dry weight as a function of time, for *Zantedeschia* 'Best Gold' grown at three temperatures under high and low PPF regimes. (a) 19 C, high PPF (b) 19 C, low PPF (c) 25 C, high PPF (d) 25 C, low PPF.

6.3.3.4 Leaf starch and soluble carbohydrate concentration

Since minimal determinations of foliar starch concentrations were carried out on plant material grown at 13 C, description of the results will primarily pertain to data collected from the 19 and 25 C environments. As with total plant leaf area and leaf dry weight (refer Chapter 3), the total quantity of leaf starch followed a logarithmic Gompertz pattern of increase with duration of growth. In later stages of growth, once leaf senescence commenced, total plant foliar starch declined.

To avoid potential problems arising from mutual shading and ontogenic variation, but to enable some comparison between all treatments, leaf starch and soluble carbohydrate concentrations were compared at the first mature leaf development stage. Under both high and low PPF regimes a negative quadratic trend was determined between leaf starch concentration and increasing temperature (Figure 6.10; $P \leq 0.05$ and $P \leq 0.01$, respectively), but the magnitude of these differences under the low PPF regime were small compared with under the high PPF regime. As a result of this interaction between temperature and PPF, leaf starch concentration tended to be greater under the high PPF regime, and this difference increased with increasing temperature. In contrast, while under the high PPF regime temperature did not influence leaf soluble carbohydrate concentration ($P \leq 0.10$), under the low PPF regime soluble carbohydrate concentration declined linearly with increasing temperature (Figure 6.11, $P \leq 0.01$).

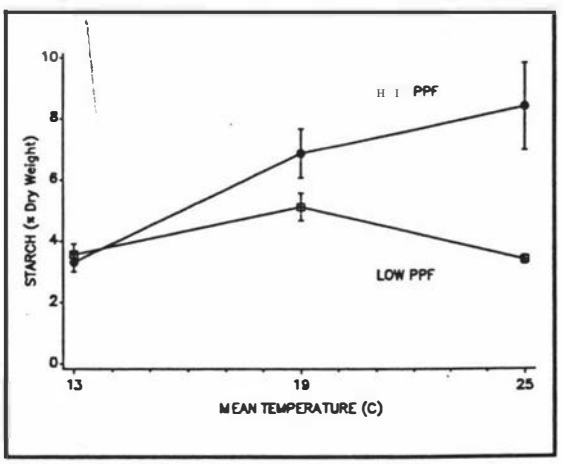

Figure 6.10 Starch concentration of first leaf, as a function of temperature, for plants of *Zantedeschia* 'Best Gold' grown under high and low PPF regimes. Mean values ± se., n=4.

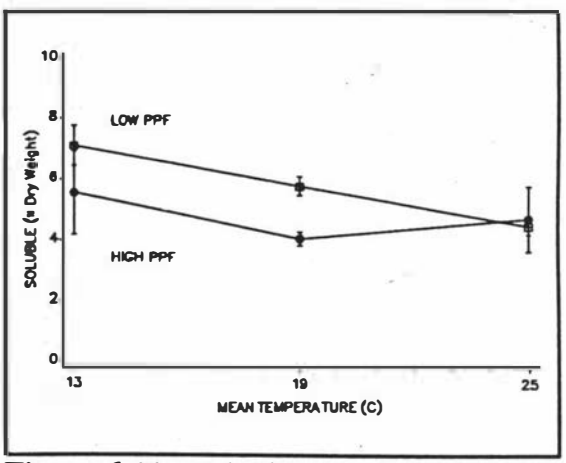

Figure 6.11 Soluble carbohydrate concentration of first leaf, as a function of temperature, for plants of *Zantedeschia* 'Best Gold' grown under high and low PPF regimes. Mean values ± se., n=4.

Under the high PPF regime no trend was detected between total leaf carbohydrate concentration, i.e., starch + soluble, and increasing temperature ($P \leq 0.10$), whereas a negative quadratic trend was detected under the low PPF regime ($P \leq 0.05$). The

existence of this trend resulted from the reduced total carbohydrate concentration at 25 C compared with either 13 or 19 C ($P \leq 0.05$).

With the exception of the incomplete data at 13 C, leaf starch concentration changed dramatically during plant development and with environment (Figure 6.12). Regardless of treatment environment the foliar starch concentration declined with duration of growth. With the exception of plants grown at 25 C under the low PPF regime, by the termination of the experiment foliar starch concentrations declined by more than 70% from that recorded at their maximum. For plants grown at 25 C under the low PPF regime foliar starch concentration had declined by only 33% over that same period.

With the exception of plants grown at 19 C under low PPF, soluble carbohydrate concentration in the leaf followed no consistent pattern of change (Figure 6.12). In contrast, for plants grown at 19 C under low PPF, the foliar concentration of soluble carbohydrate declined abruptly after 80 days by approximately 50%. Considering the highly variable nature of the soluble carbohydrate data with duration of growth, it was felt that further interpretation of these data was not warranted.

6.3.4 Carbohydrate and structural dry weight concentration as predictors of growth and yield

At the time of the first mature leaf, leaf starch concentration was positively correlated to increased photosynthetic rate per unit weight. While being significant, the magnitude of this correlation was small ($r = 0.56$), and when examined over the entire period of study, the correlation reduced even further ($r = 0.22$).

Under all treatment regimes the relative growth rate of total tuber dry weight (RGR_T) was positively correlated with the relative growth rate of both tuber starch (RGR_{Ts}) and tuber structural weight (RGR_{Tstr}). At the time RGR_T was maximum, i.e., the inflection point of the fitted total tuber dry weight curve, correlations attained were $r = 0.97$ and $r = 0.78$, respectively.

At the time RGR_T was maximum, no significant correlations were detected between RGR_T and neither the relative leaf starch weight rate (RLSWR), nor RGR_{Ts}, RGR_{Tstr}, or NAR. Similarly no significant correlations were detected between total plant relative growth rate (RGR_W) and these growth parameters.

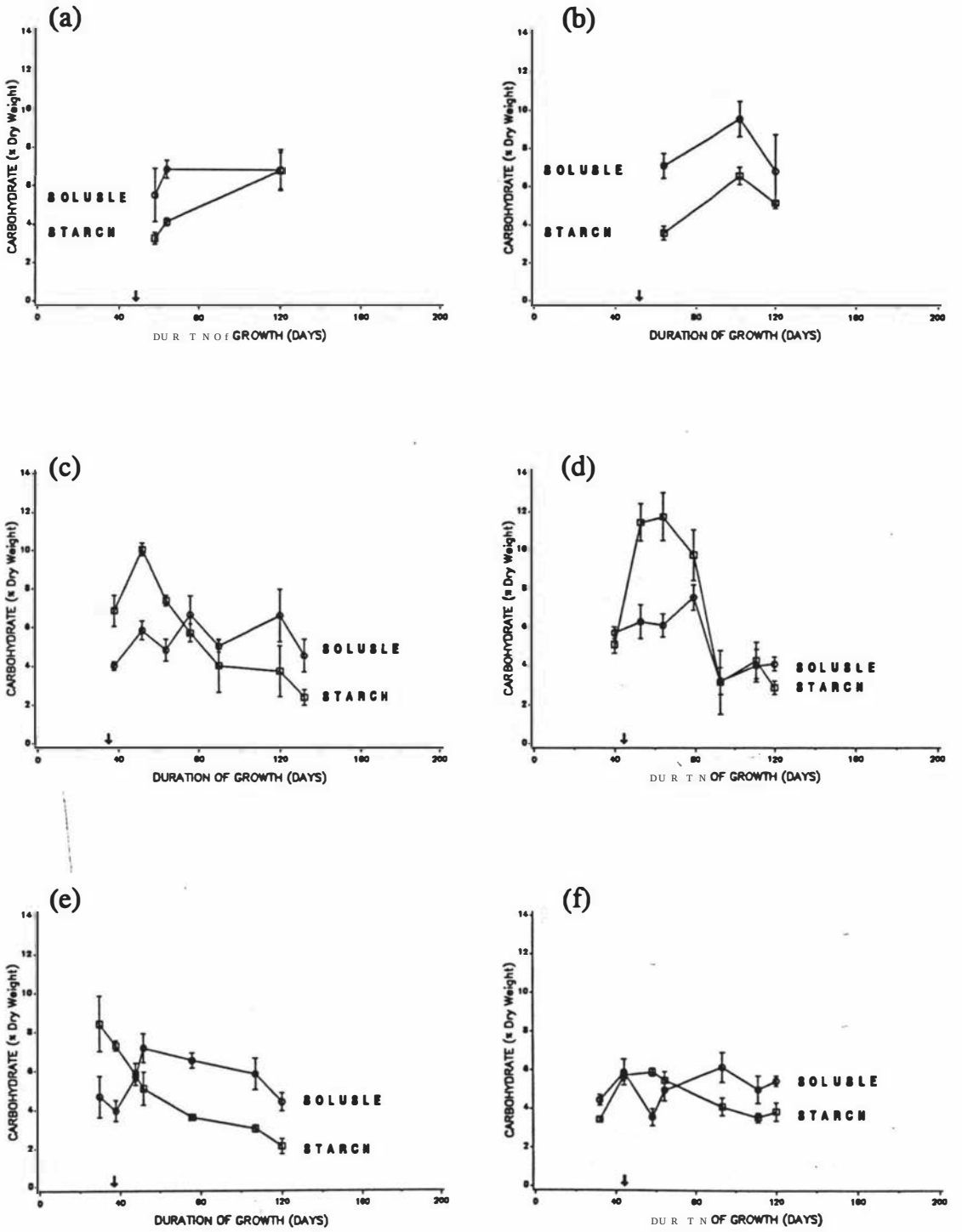

Figure 6.12 Carbohydrate concentration of mature leaves as a function of time, for *Zantedeschia* 'Best Gold' grown at three temperatures under high and low PPF regimes. (a) 13 C, high PPF (b) 13 C, low PPF (c) 19 C, high PPF (d) 19 C, low PPF (e) 25 C, high PPF (f) 25 C, low PPF. Mean values ± se., n=4, ↓ indicates commencement of tuber growth.

6.4 Discussion

6.4.1 Specific soluble carbohydrates and sample preparation

The primary products of photosynthesis in higher plants are partitioned between two major metabolic pathways, one leading to starch synthesis within the chloroplast, and the other, to cytoplasmic sucrose synthesis and extracellular translocation (Wardlaw, 1968; Chen, 1969; Ho and Rees, 1975; Giaquinta, 1978). Monosaccharides such as glucose and fructose are frequently found in plant tissue, being intermediaries in the metabolism of sucrose and starch. Sucrose, glucose and fructose were found to be present in both leaf and tuber tissue of *Zantedeschia* 'Best Gold' (Figure 6.1 and Figure 6.2). No attempt was made to determine the existence of additional soluble carbohydrates, following vacuum drying of tuber material, but the sum of the three determined by HPLC (4.1 $\pm$ 0.2%) was not significantly different from that determined by non-specific chemical analysis (3.9 $\pm$ 0.4%). This result is similar to that reported for two cultivars of taro, i.e., 3.5 - 3.9% carbohydrate, using other biochemical assays (Hashad et al., 1956). It is therefore concluded that sucrose, glucose and fructose comprise the major forms of soluble carbohydrate in *Zantedeschia* 'Best Gold'.

Reduction in leaf sucrose concentration, as a result of vacuum drying, was associated with smaller than expected increases in glucose and fructose concentrations (Figure 6.1). Therefore total soluble carbohydrate concentrations of the leaf can be expected to be 29% lower than if determined using fresh material. In contrast, no significant changes in soluble carbohydrate concentration were detected with tuber tissue following vacuum drying (Figure 6.2). The method of sample preparation can influence the resultant concentration of soluble carbohydrates and starch, the extent being dependent on the method used and plant genera (Haslemore et al., 1980). Haslemore et al. (1980) noted that vacuum drying generally resulted in only marginally inferior recoveries of soluble sugars and starch, compared with immediate determination. As vacuum drying of samples was used throughout this study, being consistent across all treatments and harvests, it is assumed that any reductions in soluble carbohydrate concentration should not affect interpretation of results.

6.4.2 Diurnal and developmental changes in starch and soluble carbohydrate concentration

Changes in starch and soluble carbohydrate concentrations, both during the daily lighting period and with plant development, indicate that carbohydrate concentrations within *Zantedeschia* 'Best Gold' result from highly controlled processes. As an indication of this control, soluble carbohydrates are some of the first derivatives of photosynthesis, but leaf

soluble carbohydrate concentration remained relatively unchanged, both during the daily period of lighting (Figure 6.3) and with duration of growth and development (Figure 6.12). Similarly, diurnal changes in soluble carbohydrate concentration occurred in tuber tissue, but the relative constancy of concentration within tubers during growth and development (Figure 6.7), is also indicative of concentrations being controlled. Regulation of both foliar and storage organ soluble carbohydrate concentrations has been associated with the activity of various enzymes (Giaquinta, 1978; Silvius et al., 1979; Preiss, 1982; Morrell and ap Rees, 1986; Rao, et al., 1990), and it is suggested that similar mechanisms of control may be operative within *Zantedeschia* 'Best Gold'.

Accumulation of foliar starch during the daily lighting period (Figure 6.3), while concentrations declined with development (Figure 6.12), is indicative of the foliage being used as a temporary store of photoassimilates during the light period. Similar cases of temporary storage of photoassimilates have been noted with taro (Hashad et al., 1956), barley (*Hordeum vulgare* L.; Gordon et al., 1980), sugar beet (Fondy and Geiger, 1982), lettuce (*Lactuca sativa* L.; Forney and Austin, 1988) and celery (*Apium graveolens* L.; Davis and Loescher, 1991). While accumulating during the daily lighting period, export of stored photoassimilates toward the developing sinks recommenced during the night (Gordon et al., 1980; Fondy and Geiger, 1982). Without the use of techniques such as radioactive labelling of photoassimilates, it can not be determined whether the increase in tuber soluble carbohydrate concentration during the light period (Figure 6.4) resulted from the continued export of soluble carbohydrates from leaves and/or transitory interconversion of starch to soluble carbohydrate within the tuber itself. The decline in tuber starch concentration, concomitant with the increase in concentration of soluble carbohydrate (Figure 6.4) is supportive of the latter hypothesis.

Accumulation of foliar carbohydrates during the light period also suggests that the processes involved in the utilization of photoassimilates were proceeding at a rate less than the rate of production of photoassimilate, i.e., sink limitation. In potato (Werner, 1935) and sugar beet (Rapoport and Loomis, 1985) conditions which restricted sink activity also resulted in the accumulation of foliar carbohydrates. Manipulation of source:sink ratios have not always resulted in accumulation of carbohydrate (Farrar and Farrar, 1987); in *Zantedeschia* 'Best Gold' the decline in concentration of foliar starch with growth, indicates non-limiting sink activity. However leaf age and mutual shading influence the ability of individual leaves to accumulate carbohydrates in other crops (Davis and Loescher, 1991), and therefore treating all leaves as having equal potential, as done in the current experiment, may invalidate this interpretation.

Foliar starch concentration increased by 65% over the light period with constant

environment, but no change in photosynthetic rate was detected (Figure 6.3). At the completion of this time starch accounted for only 8% of the leaf dry weight. Warrington et al. (1977) reported a progressive reduction in photosynthetic rate (21% maximum reduction) with increasing foliar starch (maximum concentration 11% of dry weight). However, others have either found no correlation, or have reported starch concentrations above 20 to 30% before reductions in photosynthetic rate were detectable (Forde et al., 1975; Geiger, 1976; Nafziger and Koller, 1976; Potter and Breen, 1980; Cao and Tibbitts, 1991). In addition to variation in response between species, variation in response also occurred with leaf maturity (Potter and Breen, 1980). Hence the failure to detect a diurnal reduction in photosynthetic rate with increased foliar starch concentration in *Zantedeschia* 'Best Gold', may be attributed to the comparatively low starch concentrations, the unresponsive nature of *Zantedeschia* 'Best Gold' itself, and/or the stage of leaf maturity chosen.

While it has been established that under any single treatment, diurnal increases in leaf starch concentrations were not associated with changes in photosynthetic rate, when treatments were pooled a positive correlation was found between these two parameters. This correlation was highest at the time of the first mature leaf, presumably due to the elimination of the confounding influence of variable leaf age (Davis and Loescher, 1991). In addition, increased photosynthetic rates, resulting from increased PPF, CO_2, or temperature, have been associated with a proportionate increase in translocation of carbohydrates out of the leaf (Williams and Williams, 1978; Ho, 1979; Borchers-Zampini et al., 1980; Geiger and Fondy, 1985; Farrar and Farrar, 1987). Hence the increased photosynthetic rates, as a result of increased temperature and PPF, presumably resulted in both increased accumulation of photoassimilates within the leaf and increased translocation rates. In light of this, the relatively low correlation should not be interpreted as indicating any lack of association between photosynthetic rate and production of carbohydrates in the leaf, but rather the inability to quantify parameters such as the flux of carbohydrates using the current data.

At a daily mean temperature of 13 C compared with higher temperatures, the low leaf starch and high soluble carbohydrate concentrations (Figure 6.10 and Figure 6.11) are supportive of the hypothesis that the rates of those processes in which assimilates are utilized (i.e., respiration and growth) fall more rapidly with decrease in temperature than does the rate of photosynthesis (Warren-Wilson, 1966; Verkleij and Challa, 1988; Acock et al., 1990). However, with regard to the concentration of total leaf carbohydrate, only plants grown at 25 C under the low PPF regime had a significantly lower carbohydrate concentration. Hence in addition to increased translocation rates at higher temperatures (Williams and Williams, 1978; Geiger and Fondy, 1985), the primary influence of

temperature on carbohydrate composition was the partitioning between forms, i.e., soluble carbohydrates and starch (Wardlaw, 1968; Chatterton et al., 1972; Yelenosky and Guy, 1977). Processes mediating the partitioning between soluble carbohydrate and starch are unknown, but enzymes such as fructose 2,6-bisphosphate have been implicated (Cseke et al., 1984; Stitt et al., 1984). Under most of the temperatures studied the insignificant differences between PPF regimes in total foliar carbohydrate concentration is similar to that reported for soybean (*Glycine max* L.; Silvius et al. 1979), but different from that reported for other genera, where higher concentrations are generally associated with increased PPF (Graper and Healy, 1992). Differences between genera may be the result of differential ability for translocation of assimilates out of the leaf. The low leaf starch concentration at 25 C under the low PPF regime may reflect a low concentration of photoassimilates remaining after utilization for new growth and respiration (Grange, 1985). Under this treatment the extended period before the commencement of tuber growth (Figure 6.7) also supports the hypothesis of that photoassimilate supply was limiting at this stage of growth.

6.4.3 Tuber starch and structural dry weight changes with development

The fact that starch accounted for approximately 55% of tuber dry weight at planting (Figure 6.7), and the correlation of subsequent changes with growth of the tuber, indicates that starch is the primary form of storage carbohydrate in *Zantedeschia* 'Best Gold'. This result was similar to that reported for other members of the *Araceae*, i.e., taro and tannia (*Xanthosoma sagittifolium* Schott) (Hashad et al., 1956; Brouk, 1975; Onwueme, 1978).

Once tuber growth had recommenced, the existence of a constant relationship between tuber starch and structural dry weights, under all treatment regimes (equation (6.6)), indicates that starch deposition within the tuber was a controlled process. However, prior to tuber growth the alteration of this relationship under the low PPF regime (equation (6.7)) indicates that while this process was controlled, in situations where the availability of newly-produced photoassimilate is unable to meet sink demand, the rate of remobilization of stored starch can be increased. A similar situation has been reported in soybean where only under conditions of limited assimilate availability was starch from the pod wall remobilized to enable continued seed growth (Fader and Koller, 1985). Also Davies (1984) was able to manipulate starch concentration of sprouting potato tubers by manipulating sink activity by varying shoot number. Both storage organ starch degradation and synthesis have been correlated with enzyme activity and orthophosphate concentration (Mares et al., 1981; Preiss, 1982; Ou-Lee and Setter, 1985). Future examination of changes in these components may provide new insights to the mechanisms controlling growth and development in *Zantedeschia*, in particular changes in the relationship between starch concentration and tuber growth.

Under the low PPF regime the bi-phaseal relationship between tuber starch concentration and tuber structural dry weight (Figure 6.8), is indicative of the tuber's transition from a source to a sink. However, there is no biological relevance to determining the point of intersection of the bi-phaseal relationship as a region of overlap of data points exists. Clearly, while separating the data into two phases of development has allowed the determination of two independent relationships, the extent of overlap of data points coupled with the inability to detect the bi-phaseal relationship under the high PPF regime, indicates that it would be inappropriate to infer the existence of any abrupt switch in the onset of tuber growth being related to the attainment of a specific starch concentration and/or vice versa. Rather than looking to the tuber for the point of control, it is more likely that the onset of tuber growth, reflected by increases in both structural and starch dry weights, was dictated by the establishment of a photosynthetic leaf area large enough to provide an adequate supply/flux of photoassimilates to meet the demands for continued leaf and root development, and respiration. The sink-source transition of leaves commences at 40% to 50% leaf expansion (Wardlaw, 1968; Giaquinta, 1978). However, tuber growth in *Zantedeschia* 'Best Gold' did not commence until one or more leaves had reached maturity. In addition, plants grown at low PPF and increasing temperature carried a greater leaf number and associated leaf area, at the time of onset of tuber growth, than plants grown at high PPF and cooler temperatures. This illustrates the greater demand for assimilates by organs other than the tuber. In addition the negative relationship between RGR_T and LWP (refer Chapter 4) also supports the hypothesis that tuber growth receives a lower priority for photoassimilates than foliage growth at this early stage (Loomis et al., 1979; Ho, 1988).

During tuber growth the increase in the proportion of starch dry weight relative to structural dry weight and/or total tuber dry weight, was similar to that reported for potato tubers (Plaisted, 1957), tulip bulbs (Aung et al., 1973) and corms of other members of the *Araceae* (Hashad et al., 1956; Ching, 1970). However, this change in partitioning with development was different from that reported for sugar beet where no change in the distribution of assimilates between storage root growth and storage carbohydrate (sucrose) occurred (Das Gupta, 1969; Watson et al., 1972; Milford and Thorne, 1973; Milford et al., 1988). Watson et al. (1972) suggested this constancy in distribution disputed the earlier proposed hypothesis (Ulrich, 1952, 1955) that carbohydrate stored in the tap root is photosynthate in excess of that which can be used for the growth of the rest of the plant. This constancy in distribution was not found with *Zantedeschia* 'Best Gold', and while not necessarily supporting Ulrich's hypothesis, the data presented here also does not dispute the validity of this hypothesis. The additional finding that the relationship can be manipulated under periods of high competitive sink demand (Figure 6.8), adds some validity to the application of Ulrich's hypothesis to *Zantedeschia*.

In reviewing the literature on starch storing organs, Jenner (1982) confirmed the intimate linkage between growth of storage organs and their capacity for storing starch. As found in the current experiment, Jenner concluded "that for any given genotype the proportion of dry matter contributed by starch does not vary with the size of the organ as much as does the size of the organ itself", i.e., large organs contain more starch than small ones. If correct, this would suggest that cell number will be a major determinant of tuber growth and the amount of starch stored in *Zantedeschia* 'Best Gold'. With phytohormones being implicated in the control of cell division in storage organs such as potato tubers (Lovell and Booth, 1967; Mares and Marschner, 1980), future investigations may develop a greater understanding of the mechanisms controlling tuber growth in *Zantedeschia* if consideration is also given to rates of cell division and hormonal status.

6.4.4 Carbohydrate and structural dry weight concentration as predictors of growth and yield

The inability to detect significant correlations between source activity, as quantified by NAR and RLSWR, and the relative rates of growth of the entire plant and/or its subcomponents (RGR_W, RGR_T, $RGR_{T_{str}}$, RGR_{T_s}) was not surprising. Warren Wilson (1972) suggested that more accurate determination of sink activity was gained by examining the relative growth rate of structural material, but with *Zantedeschia* 'Best Gold' no correlation was evident between the structural material of the tuber and the various measures of source activity. This inability to detect a correlation may have arisen from two sources. Firstly, time did not permit the structural dry weights of all organs of the plants in the current experiment to be quantified. Since roots, petioles and unexpanded leaves are likely to contribute towards the total sink activity, it is possible that their inclusion as the measure of sink activity was critical. Secondly, while leaves were the primary sites of synthesis of new carbohydrates, quantification of carbohydrate concentrations within the leaf and other organs, fails to account for respiratory losses and the rates of assimilate flux. Both have been shown to be important determinants of growth of the total plant and/or organs, and not just the actual concentration of carbohydrates and/or their rates of change (Borchers-Zampini et al., 1980; Fader and Koller, 1985; Ho, 1988, Farrar, 1990). Hence while not negating the importance of photosynthetic rate, photosynthetic area, and total photoassimilate supply, the methods of quantification/description of assimilate used here do not describe the rate of flux, nor respiratory losses once initially fixed.

The future development of mechanistic metabolic models to explain growth and development of *Zantedeschia* will therefore require the additional definition of the structural/carbohydrate concentrations of all organs, in addition to carbohydrate flux data

and respiratory losses.

6.4.5 Conclusions

As with many plant genera, sucrose, fructose, and glucose, accounted for the majority of soluble carbohydrates in *Zantedeschia* 'Best Gold'. The primary form of storage carbohydrate was starch, and changes in storage organ growth occurred together with changes in starch.

It is suggested that the inability to correlate growth with carbohydrate concentration is as a result of the inability to quantify carbohydrate fluxes. The maintenance of concentrations of both soluble carbohydrate and starch was highly controlled, presumably involving enzymatic activity associated with storage and flux. While Morrell and ap Rees (1986) suggested that control of the carbohydrate concentration of developing potato tubers was achieved via fine control, i.e., enzymatic synthesis and activity, Farrar (1990) suggested a modified form of coarse control, i.e., regulation of enzymatic synthesis and activity via supply of carbohydrate. Future examination of changes in these components may provide new insights to the mechanisms controlling growth and development in *Zantedeschia*.

In the absence of evidence suggesting the existence of a trigger for tuberization, rather than looking to the tuber for the point of control it is more likely that the onset of tuber growth, as increases in both structural and starch dry weights, is dictated by the establishment of a photosynthetic leaf area large enough to provide an adequate supply/flux of photoassimilates beyond the demands for continued leaf and root development, and respiration.

6.5 References

Acock, B., M.C. Acock and D. Pasternak. 1990. Interactions of CO_2 enrichment and temperature on carbohydrate production and accumulation in muskmelon leaves. J. Amer. Soc. Hort. Sci. 115:525-529.

Aung, L.H., F. Tognoni and A.A. De Hertogh. 1973. Changes in the carbohydrates of tulip bulbs during development. HortScience 8:207-208.

Borchers-Zampini, C., A.B. Glam, J. Hoddinott and C.A. Swanson. 1980. Alterations in source-sink patterns by modifications of source strength. Plant Physiol. 65:1116-1120.

Bradbury, D. 1953. Division of starch-containing cells. Amer. J. Bot. 40:286-288.

Brouk, B. 1975. Plants consumed by man. Academic Press, London.

Cao, W. and T.W. Tibbitts. 1991. Physiological responses in potato plants under continuous irradiation. J. Amer. Soc. Hort. Sci. 116:525-527.

Causton, D.R. and J.C. Venus. 1981. Single leaf growth and the Richards function: methodology, p. 87-143. In: The biometry of plant growth. Edward Arnold, London.

Chatterton, N.J., G.E. Carlson, W.E. Hungerford and D.R. Lee. 1972. Effect of tillering and cool nights on photosynthesis and chloroplast starch in pangola. Crop Sci. 12:206-208.

Chen, S. 1969. Carbohydrate metabolism in the narcissus leaf. J. Expt. Bot. 20:302.

Ching, K.W. 1970. Development of starch, protein, leaf and corm in *Colocasia esculenta*, p. 143-146. In: D.L. Pluckett (ed.). Proceedings 2nd Intl. Symp. on Trop. Root and Tuber Crops: Tropical root and tuber crops tomorrow. Univ. Hawaii.

Cseke, C., A. Balogh, J.H. Wong, B.B. Buchanan, M. Stitt, B. Herzog and H.W. Heldt. 1984. Fructose 2,6-bisphosphate: a regulator of carbon processing in leaves. Trends Biochem. Sci. 9:533-535.

Das Gupta, D.K. 1969. The effect of decapitation on growth of the sugar beet storage root, p. 247-255. In: W.J. Whittington (ed.). Proceedings of the Fifteenth Easter School in Agricultural Science, Univ. Nottingham. Butterworths, London.

Davis, J.M. and W.H. Loescher. 1991. Diurnal pattern of carbohydrates in celery leaves of various ages. HortScience 26:1404-1406.

Davies, H.V. 1984. Mother tuber reserves as factors limiting potato sprout growth. Potato Res. 27:209-218.

Fader, G.M. and H.R. Koller. 1985. Seed growth rate and carbohydrate pool sizes of the soybean fruit. Plant Physiol. 79:663-666.

Farrar, J.F. 1990. The carbon balance of fast-growing and slow-growing species, p. 241-256. In: H. Lambers, M.L. Cambridge, H. Konings, and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague, The Netherlands.

Farrar, S. and J. Farrar. 1987. Effects of photon fluence rate on carbon partitioning in barley source leaves. Plant Physiol. Biochem. 25:541-548.

Fondy, B.R. and D.R. Geiger. 1982. Diurnal pattern of translocation and carbohydrate metabolism in source leaves of *Beta vulgaris* L. Plant Physiol. 70:671-676.

Forde, B.J., H.C.M. Whitehead and J.A. Rowley. 1975. Effect of light intensity and temperature on photosynthetic rate, leaf starch content and ultrastructure of *Paspalum dilatatum*. Australian J. Plant Physiol. 2:185-195.

Forney, C.F. and R.K. Austin. 1988. Time of day at harvest influences carbohydrate concentration in crisphead lettuce and its sensitivity to high CO_2 levels after harvest. J. Amer. Soc. Hort. Sci. 113:581-583.

Geiger, D.R. 1976. Effects of translocation and assimilate demand on photosynthesis. Can. J. Bot. 54:2337-2345.

Geiger, D.R. and B.R. Fondy. 1985. Responses of export and partitioning to internal and environmental factors, p. 177-194. In: B. Jeffcoat, A.F. Hawkins and A.D. Stead (eds.). Regulation of sources and sinks in crop plants. British Plant Growth Regulator Group, Monograph 12.

Giaquinta, R. 1978. Source and sink leaf metabolism in relation to phloem translocation: carbon partitioning and enzymology. Plant Physiol. 61:380-385.

Gifford, R.M., J.H. Thorne, W.D. Hitz and R.T. Giaquinta. 1984. Crop productivity and photoassimilate partitioning. Science 225:801-808.

Gordon, A.J., G.J.A. Ryle, C.E. Powell and D. Mitchell. 1980. Export, mobilization, and respiration of assimilates in uniculm barley during light and darkness. J. Expt. Bot. 31:461-473.

Grange, R.I. 1985. Carbon partitioning and export in mature leaves of pepper (*Capsicum annuum*). J. Expt. Bot. 36:734-744.

Graper, D.F. and W. Healy. 1992. Modification of petunia seedling carbohydrate partitioning by irradiance. J. Amer. Soc. Hort. Sci. 117:477-480.

Habeshaw, D. 1973. Translocation and the control of photosynthesis in sugar beet. Planta 110:213-226.

Hashad, M.N., K.R. Stino and S.I. El Hinnawy. 1956. Transformation and translocation of carbohydrates in taro plants during growth. Ann. Agr. Sci. 1(1):261-267.

Haslemore, R.M. and P.G. Roughan. 1976. Rapid chemical analysis of some plant constituents. J. Sci. Food Agr. 27:1171-1178.

Haslemore, R.M., I.J. Warrington and P.G. Roughan. 1980. Influence of drying method and post-harvest conditions on total nitrogen, soluble sugar, and starch levels in plant tissue. N.Z. J. Agr. Res. 23:355-359.

Ho, L.C. 1979. Regulation of assimilate translocation between leaves and fruits in the tomato. Ann. Bot. 43:437-448.

Ho, L.C. 1988. Metabolism and compartmentation of imported sugars in sink organs in relation to sink strength. Annu. Rev. Plant Physiol. 39:355-378.

Ho, L.C. and A.R. Rees. 1975. Aspects of translocation of carbon in the tulip. New Phytol. 74:421-428.

Ho, L.C. and A.R. Rees. 1976. Re-mobilization and redistribution of reserves in the tulip bulb in relation to new growth until anthesis. New Phytol. 76:59-68.

Irvine, J.E. 1975. Relations of photosynthetic rates and leaf and canopy characteristics to sugarcane yield. Crop Sci. 15:671-676.

Jenner, C.F. 1982. Plant carbohydrates I: Intracellular carbohydrates, p. 700-747. In: F.A. Loewus and W. Tanner (eds.). Encyclopedia of Plant Physiology, New Series vol. 13A. Springer Verlag, Berlin.

Lester, G.E., L.R. Baker and J.F. Kelly. 1982. Physiology of sugar accumulation in carrot breeding lines and cultivars. J. Amer. Soc. Hort. Sci. 107:381-387.

Loomis, R.S., R. Rabbinge and E. Ng. 1979. Explanatory models in crop physiology. Annu. Rev. Plant Physiol. 30:339-367.

Lovell, P.H. and A. Booth. 1967. Effects of gibberellic acid on growth, tuber formation and carbohydrate distribution in *Solanum tuberosum*. New Phytol. 66:525-537.

Mares, D.J. and H. Marschner. 1980. Assimilate conversion in potato tubers in relation to starch deposition and cell growth. Ber. Dtsch. Bot. Ges. 93:299-313.

Mares, D.J., H. Marschner and A. Krauss. 1981. Effect of gibberellic acid on growth and carbohydrate metabolism of developing tubers of potato (*Solanum tuberosum*). Physiol. Plant. 52:267-274.

Milford, C.F.J. 1973. The growth and development of the storage root of sugar beet. Ann. Appl. Biol. 75:427-438.

Milford, C.F.J. and G.N. Thorne. 1973. The effect of light and temperature late in the season on the growth of sugar beet. Ann. Bot. 35:287-300.

Milford, G.F.J., K.Z. Travis, T.O. Pocock, K.W. Jaggard and W. Day. 1988. Growth and dry-matter partitioning in sugar beet. J. Agr. Sci. 110:301-308.

Moorby, J. 1968. The influence of carbohydrates and mineral nutrient supply on the growth of potato tubers. Ann. Bot. 32:57-68.

Morrell, S. and T. ap Rees. 1986. Sugar metabolism in developing tubers of *Solanum tuberosum*. Phytochem. 25:1579-1585.

Nafziger, E.D. and H.R. Koller. 1976. Influence of leaf starch concentration on CO_2 assimilation in soybean. Plant Physiol. 57:560-563.

Obata-Sasamoto H. and H. Suzuki. 1979. Activities of enzymes relating to starch synthesis and endogenous levels of growth regulators in potato stolon tips during tuberization. Physiol. Plant. 45:320-324.

Onwueme, I.C. 1978. The tropical tuber crops. John Wiley and Sons, Chichester.

Ou-Lee, T. and T.L. Setter. 1985. Enzyme activities of starch and sucrose pathways and growth of apical and basal maize kernels. Plant Physiol. 79:848-851.

Plaisted, P.H. 1957. Growth of the potato tuber. Plant Physiol. 32:445-453.

Potter, J.R. and P.J. Breen. 1980. Maintenance of high photosynthetic rates during the accumulation of high starch levels in sunflower and soybean. Plant Physiol. 66:528-531.

Preiss, J. 1982. Regulation of the biosynthesis and degradation of starch. Annu. Rev. Plant Physiol. 33:431-454.

Rao, M., A.L. Fredeen and N. Terry. 1990. Leaf phosphate status, photosynthesis, and carbon partitioning in sugar beet. Plant Physiol. 92:29-36.

Rapoport, H.F. and R.S. Loomis. 1985. Interaction of storage root and shoot in grafted sugarbeet and chard. Crop Sci. 1079-1084.

Rees, A.R. 1972. The growth of bulbs. Academic Press, London.

Ross, H.A. and H.V. Davies. 1985. A microscopic examination of starch depletion in tubers of cv. Maris Piper during sprouting. Potato Res. 28:113-118.

Silvius, J.E., N.J. Chatterton and D.F. Kremer. 1979. Photosynthate partitioning in soybean leaves at two irradiance levels. Plant Physiol. 64:872-875.

Stitt, M., B. Kurzel, and H.W. Heldt. 1984. Control of photosynthetic sucrose synthesis by fructose 2,6-bisphosphate. II. Partitioning between sucrose and starch. Plant Physiol. 75:554-560.

Terry, N. 1968. Developmental physiology of sugar beet. I. The influence of light and temperature on growth. J. Expt. Bot. 19:795-811.

Thorne, J.H. and H.R. Koller. 1974. Influence of assimilate demand on photosynthesis, diffusive resistances, translocation, and carbohydrate levels of soybean leaves. Plant Physiol. 54:201-207.

Ulrich, A. 1952. The influence of temperature and light factors on the growth and development of sugar beets in controlled climatic environments. Agron. J. 44:66-73.

Ulrich, A. 1955. Influence of night temperature and nutrition on the growth, sucrose accumulation and leaf minerals of sugar beet plants. Plant Physiol. 30:250-257.

van Es, A. and K.J. Hartmans. 1987. Starch and sugars during tuberization, storage and sprouting, p. 79-113. In: A. Rastovski, and A. van Es et al. (eds.). Storage of Potatoes. Puduc, Wageningen.

Verkleij, F.N. and H. Challa. 1988. Diurnal export and carbon economy in an expanding source leaf of cucumber at contrasting source and sink temperature. Physiol. Plant. 74:284-293.

Wang, Y. and P.J. Breen. 1986. Growth and photosynthesis of easter lily in response to flower bud removal. J. Amer. Soc. Hort. Sci. 111:442-446.

Wardlaw, I.F. 1968. The control and pattern of movement of carbohydrates in plants. Bot. Rev. 34:79-105.

Wareing, P.F. and J. Patrick. 1975. Source-sink relations and the partition of assimilates in the plant, p. 481-499. In: J.P Cooper (ed.) Photosynthesis and productivity in different environments. Cambridge Univ. Press, Cambridge.

Warren Wilson, J. 1966. An analysis of plant growth and its control in arctic environments. Ann. Bot. 30:383-402.

Warren Wilson, J. 1972. Control of crop processes, p. 7-30. In: A.R. Rees, K.E. Cockshull, D.W. Hand, and R.G. Hurd (eds.). Crop processes in controlled environments. Academic Press, London.

Warrington, I.J., M. Peet, D.T. Patterson, J. Bunce, R.M. Haslemore, and H. Hellmers. 1977. Growth and physiological responses of soybean under various thermoperiods. Australian J. Plant Physiol. 4:371-380.

Watson, D.J., T. Motomatsu, K. Loach, and G.F.J. Milford. 1972. Effects of shading and of seasonal differences in weather on the growth, sugar content and sugar yield of sugar beet crops, Ann. Applied Biol. 71:159-185.

Werner, H.O. 1935. The effect of temperature, photoperiod and nitrogen level upon tuberization in the potato. Amer. Potato J. 12:274-280.

Williams, A.M. and K.R. Williams. 1978. Regulation of movement of assimilate into ovules of *Pisum sativum* cv. Greenfeast. Australian J. Plant Physiol. 5:295-300.

Yelenosky, G. and C.L. Guy. 1977. Carbohydrate accumulation in leaves and stems of 'Valencia' orange at progressively colder temperatures. Bot. Gaz. 138:13-17.

Zar, J.H. 1984. Biostatistical analysis. Prentice Hall, Englewood Cliffs, New Jersey.

7 MANIPULATION OF IN VITRO SOURCE AND SINK STRENGTH, AND DRY MATTER PARTITIONING IN *Zantedeschia* 'Best Gold'

7.1 Introduction

In the preceding experiments classic growth analysis was used to describe the growth and tuber development of *Zantedeschia* 'Best Gold' under a range of environmental conditions (refer Chapters 3 and 4). The growth analysis results, together with measurements of photoassimilate capacity (refer Chapter 5) and tissue carbohydrate status (refer Chapter 6), were used to hypothesize the possible mechanisms controlling dry matter accumulation and its partitioning towards the tuber. In its simplest form two possible situations can be envisaged in the control of dry matter accumulation within the plant, i.e., either the system is source limited, or the system is sink limited (Wareing and Patrick, 1975). In the experiments described earlier, the range of environmental conditions used were chosen to offer a range of potential source and sink strengths, and thereby allow determination of controlling mechanisms. However, it was evident from these experiments that source and sink strengths may not have been manipulated independently when temperature and/or photosynthetic photon flux (PPF) regimes were altered. As an example, while cultivation under increased temperatures generally increased photosynthetic rate (refer Chapter 5), and therefore source activity, it was uncertain how much of the concomitant increase in tuber growth (refer Chapter 4) resulted from the increased source activity, compared with the direct effect of temperature on sink activity itself. In addition, source size also increased with increased temperature (refer Chapter 3), and together with the resultant intersink competition between leaves and tuber, accurate description of source and sink strengths were confounded. Similarly, while cultivation under low PPF conditions reduced the photosynthetic rate (refer Chapter 5) and therefore potential source activity, leaf area and therefore source size increased (refer Chapter 3), again preventing any clear interpretation of results. It was therefore evident that if a greater understanding of possible mechanisms of dry matter accumulation was to be gained, some means of independently manipulating the source and sink strengths was required.

In vitro systems have previously been used to examine the specific requirements for storage organ initiation, and/or investigation of source and sink limitations of growth and dry matter accumulation by storage organs (Gregory, 1956; Loomis and Torrey, 1964; Heath and Hollies, 1965; Palmer and Smith, 1970; Peterson, 1973; Gifford and Evans, 1981; Jones et al., 1981; Cobb and Hannah, 1986; Tovar et al., 1987; Cobb et al., 1988; Taeb and Alderson, 1990; Chow et al., 1992). In general the experimental unit utilized in such research has been the isolated storage organ, such as maize (*Zea mays* L.) kernels and sugar cane (*Saccharum officinarum* L.) stem segments (Jones et al., 1981; Gifford and Evans, 1981), or an in vitro cutting, as with potato (*Solanum tuberosum* L.) and yam

(*Dioscorea rotundata* Poir.) (Gregory, 1956; Ng, 1988). Only infrequently have established, whole plants been utilized, such as seedlings of sugar beet (*Beta vulgaris* L.), radish (*Raphanus sativus* L.) and turnip (*Brassica rapa* L.) (Peterson, 1973; Ting and Wren, 1980; Vreugdenhil and Bouwmeester, 1989), or vegetative explants as with tulip (*Tulipa gesneriana* L.) and narcissus (*Narcissus* sp. L.) (Taeb and Alderson, 1990; Chow et al., 1992). Although use of isolated organs has enabled examination of the requirements of dry matter accumulation, use of whole plants is likely to permit a more accurate investigation of dry matter partitioning between competing organs. In this way an integrated appraisal of the physiological mechanisms involved in source and sink relationships would be permitted, in contrast to the potentially unrestrained development of isolated organs. Since the great majority of information pertains therefore to isolated organs and/or cuttings, this information is presented and discussed with the understanding that the responses of whole plants may be different.

Zantedeschia plants have been successfully propagated in vitro from vegetative bud explants, using a Murashige and Skoog (1962) medium supplemented with various phytohormones to stimulate differentiation, and with between 2 and 3% sucrose as a source of carbohydrates (Cohen, 1981; Rong et al., 1989). Although in vitro germination of seeds of other members of the *Araceae* has been achieved on non-amended agar, seedlings were not reported as having been grown until the onset of tuber growth (Kikuta et al., 1937), as was proposed in these experiments.

Within the diversity of plant organs studied in vitro, generally sucrose has been found to be the preferred respirable substrate for uptake and short-term growth or storage, correlating well with the most common form of carbohydrate translocated (Lawrence and Barker, 1963; Heath and Hollies, 1965; Gifford and Evans, 1981; Cobb and Hannah, 1986). The development of storage organs in vitro has been found to increase with increasing sucrose concentration, i.e. with increasing source strength, and generally has been found to be optimal in the range of 8 to 12%, with higher concentrations reducing storage organ growth (Gregory, 1956; Lawrence and Barker, 1963; Heath and Hollies, 1965; Ting and Wren, 1980; Koda and Okazawa, 1983; Cobb and Hannah, 1986; Tovar et al., 1987; Mielke and Anderson, 1989; Taeb and Alderson, 1990; Chow et al., 1992). However, lower optimal concentrations between 3 and 6% sucrose for storage organ growth have been reported for some species (van Aartrijk and Blom-Barnhoorn, 1980; Rice et al., 1983; Ng, 1988).

Although the exogenous supply of carbohydrates should theoretically bypass the need for photosynthesis, and therefore the need to supply light in vitro, the presence of light may be required to stimulate morphogenic development (Kato, 1978; Hughes, 1981). The light

induced accumulation of starch in vitro has been implicated as a factor in morphogenesis of tobacco (*Nicotiana tabacum* L.; Thorpe and Murashige, 1970). In addition, the assimilation of nitrogen and, therefore, the consequent synthesis of amino acids and enzymes are associated with the photosynthetic process and therefore light (Gibbs and Latzko, 1979).

In in vitro systems, the utilization of very low light fluxes (e.g., 20 - 50 $\mu mol \cdot m^{-2} \cdot s^{-1}$ PPF), limited gas exchange, and the supply of exogenous sugars, were shown to be sub-optimal for any significant photosynthetic development (Grout and Aston, 1978; Hughes, 1981; Donnelly and Vidaver, 1984). Although photosynthesis does occur in vitro, limited gas exchange between the container and the outside environment results in significant recycling of respired CO_2 (Abbott and Belcher, 1982) and presumably, therefore, minimal net gain in photoassimilates. In fact, efforts to achieve in vitro photoautotrophism by the omission of a respirable carbohydrate source, so that plants might sustain their own growth, typically have not been successful unless supplied with high light intensities coupled with CO_2 enrichment (Lakso et al., 1986; Langford and Wainwright, 1987; Kozai, 1989). Hence it can be concluded that should light be required to optimise tuber growth in vitro, the use of conventional low PPF coupled with the limited gas exchange achieved under normal cultural conditions, should eliminate any significant contribution of photosynthesis to source strength.

The control of assimilate allocation among competing sinks is not well understood, but current research suggests factors regulating sink metabolism play a dominant role (Gifford and Evans, 1981; Wardlaw, 1990; Farrar and Williams, 1991). Relevant sink metabolism involves phloem unloading, cell wall invertase activity, plasma membrane and tonoplast membrane transport, and conversion of assimilates to storage products. These metabolic activities all influence the steepness of the osmotic gradient within the phloem which has been suggested as a factor controlling assimilate flux from source to sink. For any given genotype, the proportion of dry matter in storage organs that is contributed by carbohydrates does not vary with the size of the organ as much as does the size of the organ itself (Jenner, 1982). In general, large specimens contain more storage carbohydrates than small ones. Thus the growth of storage organs and their capacity for storing carbohydrates are intimately linked. The total quantity of storage carbohydrate accommodated by any given organ is a product of the amount of storage carbohydrate per cell and the total number of cells. Although there may be substantial regional differences in storage carbohydrate content per cell within the organ, it follows nevertheless that cell number is a major determinant of the amount stored within that organ (Reeve et al., 1973; Rapoport and Loomis, 1986).

Using tomato (*Lycopersicon esculentum* L.), Ho (1979) demonstrated that although assimilate export in vivo is primarily determined by the rate of assimilation in leaves, the utilization of assimilate in the sink organ (tomato fruit) can determine the rate of carbon import. This concept of sink activity being integral to growth of the storage organ was also exemplified through the reciprocal grafting of the large storage root forming sugar beet and the small storage root forming chard (*Beta vulgaris* L.; Rapoport and Loomis, 1985). This reciprocal grafting clearly illustrated that the hypocotyl-root determines cell division within the storage organ and therefore sink strength, i.e., it is a genetic characteristic.

Increases in temperature have been shown to increase metabolic activity and thereby sink activity (Wardlaw, 1968; Ho, 1979; Jones et al., 1985), with differences in storage organ growth at any temperature being dependent on the in vitro source strength (Dinar and Stevens, 1982; Koda and Okazawa, 1983). In vivo data to date have been interpreted as indicating that both total plant and tuber dry weights, and growth rates (i.e., sink activity) of *Zantedeschia* 'Best Gold' increase with increasing temperature to an optimum near 25 C (refer Chapters 3 and 4). The ability to manipulate source strength in vitro, via sucrose concentration, independent of manipulating sink activity, via temperature, would hopefully permit determination of the relative importance of source and sink limitation under similar temperatures in vivo.

Tissue water relations may play an important role in regulating solute transport processes within the plant. In particular, turgor pressure emerges as a potential regulatory component of sucrose fluxes in developing sinks. The lowering of cell turgor enhanced active sucrose uptake in discs of sugar beet root and potato tuber (Wyse et al., 1986; Oparka and Wright, 1988). In addition, the subsequent partitioning of sucrose to starch in discs of potato tuber was enhanced by reduced turgor pressure (Oparka and Wright, 1988). Although some response was evident as a result of increased osmotic potential, reduced turgor pressure was primarily responsible (Oparka and Wright, 1988). However, tuber formation of potato stolons in vitro, at concentrations of sucrose up to 12% was not a result of the osmotic concentration, as similar osmotic concentrations of the non-respirable carbohydrate mannitol, did not result in tuber formation (Lo et al., 1972). In light of these findings investigations examining a range of in vitro source strengths must recognise the potential influence of changes in turgor and osmotic potential. Although exceptions exist, plants which are able to metabolize mannitol naturally contain mannitol (Trip et al., 1964). With sucrose, fructose and glucose comprising the primary forms of soluble carbohydrate in *Zantedeschia* (refer Chapter 6) it is assumed that mannitol would not be metabolized by *Zantedeschia*, and could therefore be used to regulate osmotic potential and turgor in vitro, without altering source strength.

The ready uptake of sugars from the bathing media by organs in vitro suggests that in vivo they may take up assimilate by the phloem from the free space, and that this apoplastic route is most commonly favoured. In roots, where water flow through the free space occurs in the opposite direction, symplastic transport of sucrose from stele to cortex and apical cells seems more likely and is supported experimentally (Hampson et al., 1978; Gifford and Evans, 1981). Just how these hypotheses of assimilate transport apply to entire plants of *Zantedeschia* in vitro is uncertain. However, considering the aforementioned apoplastic movement of assimilates in in vitro systems, and the close proximity of the source to the sink in vitro, it is assumed that the significance of the assimilate transport pathway will be minimal.

The objectives of this research were two-fold;

1) To develop in vitro techniques applicable to the growth of seedlings of *Zantedeschia*, which would permit tuber formation.

2) To use these techniques to independently manipulate source and sink strength, so as to facilitate the formation of a hypothesis on the control mechanisms of dry matter accumulation and partitioning in *Zantedeschia* in vivo.

7.2 Materials and Methods

7.2.1 Germination media, media transfer and sucrose concentrations

Seeds of *Zantedeschia* 'Chromatella' had been harvested at the end of the previous season and stored at 20 C for approximately 12 months. Seeds were surface sterilized in a solution of 20% sodium hypochlorite and 0.5 ml·litre^{-1} Multifilm X-77 (Ivon Watkins-Dow Ltd., N.Z.) wetting agent for 30 min, and subsequently rinsed in sterile distilled water. To determine the influence of sucrose during germination the use of two media was examined. These media consisted of either 5 g·litre^{-1} agar (Davis) and half-strength Murashige and Skoog's (1962) medium (MS), or the same medium plus 2% sucrose. The pH was adjusted to 5.7 $\pm$ 0.1 prior to adding agar and sterilization. Sterilization was carried out at 121 C and a pressure of 0.1 MPa for 15 min. Each 200 ml jar contained 50 ml of medium with 20 seeds, and in total 120 seeds were sown onto each of the two germination media (i.e., 12 jars total). Jars with translucent polypropylene lids containing the seeds were incubated in the dark at 20 C for a period of 3 to 4 weeks.

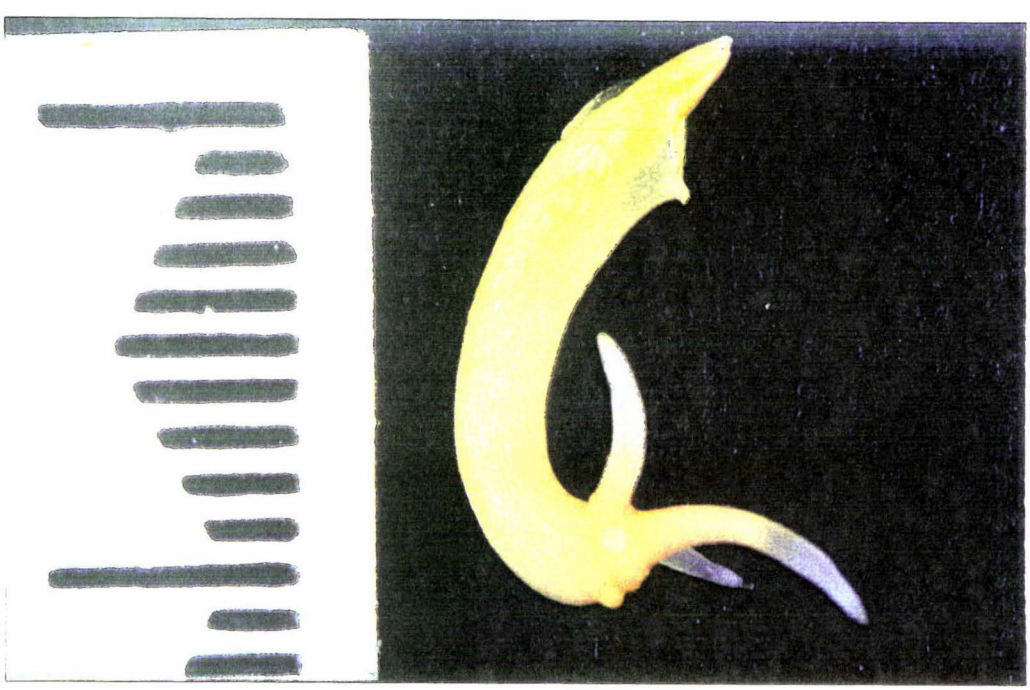

Plate 7.1 Germinated seedling of *Zantedeschia* 'Chromatella' after being excised from the cotyledon. Scale divisions = 1 mm.

Once seedlings had germinated to the stage where the point of emergence of the plumule was visible, seedlings were excised from the cotyledons (Plate 7.1) and transferred to the treatment media. At the time of transfer, roots were trimmed to 5 mm length and plants graded for even size. Seedlings were soaked in a solution of 0.25% sodium hypochlorite for 15 min and subsequently rinsed in a solution of 0.05% sodium hypochlorite prior to

transfer to the treatment media. From each of the two germination media 10 seedlings, in individual jars, were allocated to each of 3 treatment media. Treatment media consisted of 20 ml of 5 g·litre^{-1} agar and MS medium, plus either 2, 4, or 6% sucrose. A further 30 seedlings were transferred to an intermediary medium containing 2% sucrose until a second subsequent transfer after a three week period of establishment. At the time of the second transfer, 10 individual seedlings were transferred from each of the germination media prehistory groups, onto media containing 20 ml of 5 g·litre^{-1} agar and MS medium, plus either 2, 4, or 6% sucrose. Subsequent to germination all treatments were also divided between being incubated in the dark or light (45 μmol·m^{-2}·s^{-1} PPF). Figure 7.1 diagrammatically summarizes treatments and relevant treatment transfers. Individual jars were covered with a 5 μm, high density, translucent, polyethylene film, attached by rubber bands, and incubated at 25 C.

During germination, development was monitored daily, but once transferred to subsequent media, development was monitored weekly. After 29 weeks, leaf number per plant was recorded prior to the entire plant being vacuum dried at 0.3 KPa for 48 h at 40 C. Measurements of shoot (leaf + petiole), tuber, and root dry weights were subsequently recorded.

Seedlings were arranged in a completely randomized design within each treatment environment. Leaf number, dry weight and dry weight partitioning data were subjected to analysis of variance using the general linear models procedure of SAS. Plants with fungal or bacterial contamination were excluded from the analysis.

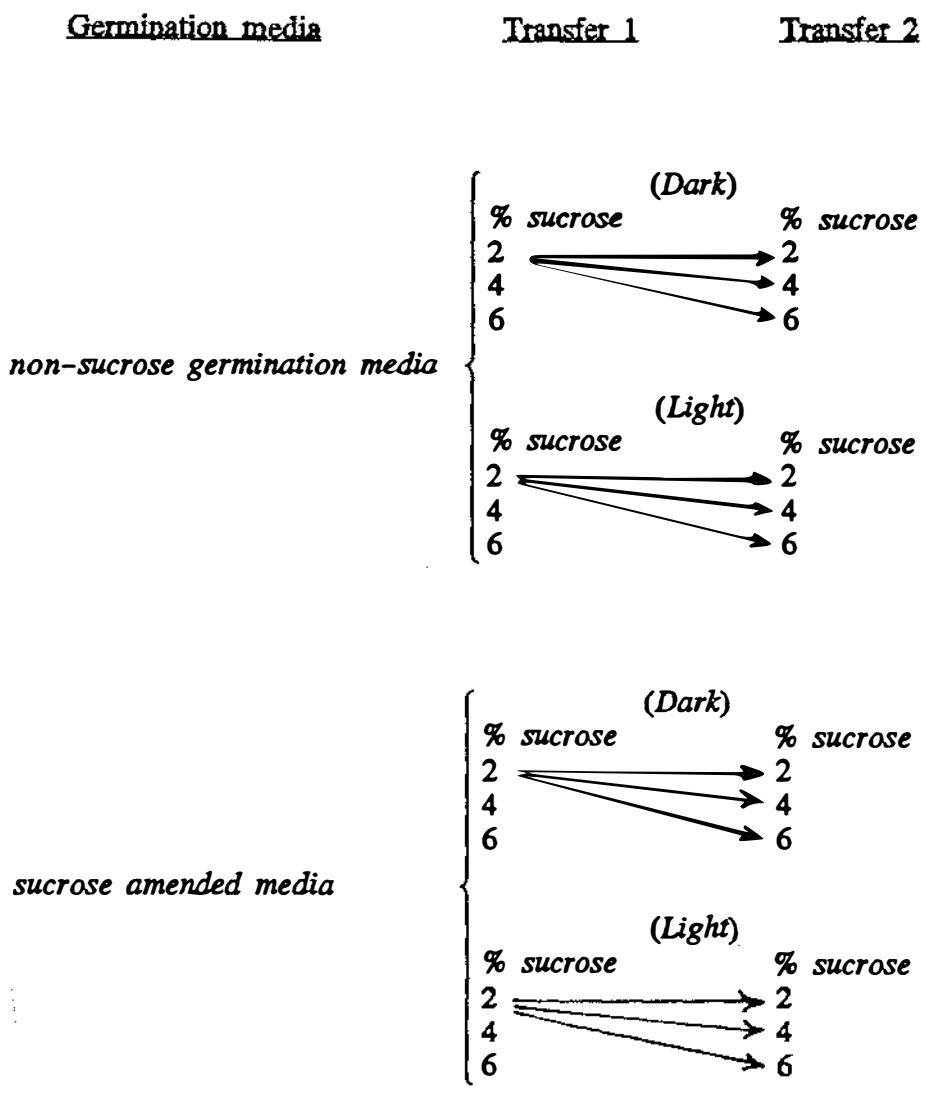

Figure 7.1 Diagrammatic summary of germination and transfer treatment media used for seedlings of *Zantedeschia* 'Chromatella'.

7.2.2 Manipulation of in vitro source and sink strengths

Seeds of the *Zantedeschia pentlandii*-like selection 'Best Gold' used in this experiment were harvested at the end of the previous season and stored at 25 C for approximately 12 months. Seed were surface sterilized and germinated as outlined in Section 7.2.1, using a germination medium consisting of 5 g·litre^{-1} agar amended with MS. The pH was adjusted to 5.7 $\pm$ 0.1 prior to adding agar and sterilization. During germination, 35 200 ml jars, each containing 50 ml of medium and 20 seeds (700 seeds in total) were incubated in the dark at 20 C.

Four weeks after sowing, germinated seedlings were excised from the cotyledon and transferred to treatment media. Excised seedlings were handled and sterilized as outlined in Section 7.2.1. Twenty seedlings in individual jars were allocated to 20 ml each of 4 treatment media. Treatment media comprised the germination medium amended with Linsmaier and Skoog's (1965) organic additives (LS) plus either 1, 2, 4, or 6% sucrose. Mannitol was added to each treatment medium to result in an osmotic potential equivalent to that of 6% sucrose as outlined below.

1% sucrose + 2.7% (0.15 M) mannitol
2% sucrose + 2.2% (0.12 M) mannitol
4% sucrose + 1.1% (0.06 M) mannitol
6% sucrose + 0.0% (0.00 M) mannitol

Final osmotic potential of each treatment medium was approximately 441.7 kPa. Individual jars were covered with a 5 μm, high density, translucent, polyethylene film, attached by rubber bands, and placed under a 12 h diurnal light period of 45 $\mu mol \cdot m^{-2} \cdot s^{-1}$ PPF, at one of 4 temperatures, i.e., 19, 25, 28, or 31 $\pm$ 1 C.

In addition to the above treatments, the ability of *Zantedeschia* 'Best Gold' to metabolize mannitol was evaluated by the inclusion of additional treatments consisting of 20 ml of 5 $g \cdot litre^{-1}$ agar, MS, LS, plus either 0, 1.1, 2.2, or 2.7% mannitol. The pH was adjusted to 5.7 $\pm$ 0.1 prior to adding agar and sterilization. Plants were treated as outlined above, but due to limited plant material, seedlings were grown at 25 C only.

Seedlings were evaluated after 24 weeks. Shoot and leaf number per plant were recorded prior to the entire plant being vacuum dried at a pressure of 0.3 kPa for 48 h at 40 C. Measurements of shoot (leaf + petiole), tuber, and root dry weights were subsequently recorded.

Seedlings were arranged in a completely randomized design within each treatment environment. Shoot and leaf number, dry weight and dry weight partitioning data were subjected to analysis of variance using the general linear models procedure of SAS. Plants with fungal or bacterial contamination were excluded from the analysis.

7.3 Results

7.3.1 Germination media, media transfer and sucrose concentrations

7.3.1.1 Germination

Within two weeks of sowing, radicles had developed to between 2 and 3 mm in length, and by three weeks seedlings on the non-sucrose amended media had germinated to the stage where the point of emergence of the plumule was visible. Seedlings on the sucrose amended media did not reach this stage of development until 4 weeks after sowing.

7.3.1.2 Dry matter accumulation and partitioning

In the absence of any significant influence of the germination medium ($P \leq 0.10$) or transfer onto an intermediary sucrose medium ($P \leq 0.10$), data presented here refer to those pooled across the aforementioned treatments.

The most influential factor on growth and dry matter accumulation was the presence or absence of light ($P \leq 0.001$, Table 7.1, Plate 7.2). Total plant dry weight was between two and six fold greater from seedlings grown under light compared with in the dark. Similarly tuber and shoot dry weights, and leaf production, were greater when seedlings were incubated under light compared with dark.

When incubated under light, a positive linear trend of increasing total plant dry weight with increasing sucrose concentration was determined ($P \leq 0.05$, Table 7.1). Total plant dry weight of seedlings grown on the 6% sucrose medium was more than 50% greater than that achieved on the 2% sucrose medium. However, no relationship between total plant dry weight and sucrose concentration, could be detected for those seedlings incubated in the dark.

Positive linear trends were evident between sucrose concentration and tuber dry weight under both dark ($P \leq 0.05$) and light ($P \leq 0.001$) regimes (Table 7.1). At the time of final harvest, tuber dry weight of seedlings grown on the 6% sucrose medium was more than eight fold greater than that achieved on the 2% sucrose medium when incubated in the dark, and approximately four fold greater when incubated in the light. While under the dark regime a significant positive linear trend was determined between sucrose concentration and the proportion of the total plant dry weight in the tuber ($P \leq 0.001$), under the light regime this relationship was quadratic ($P \leq 0.001$, Table 7.1).

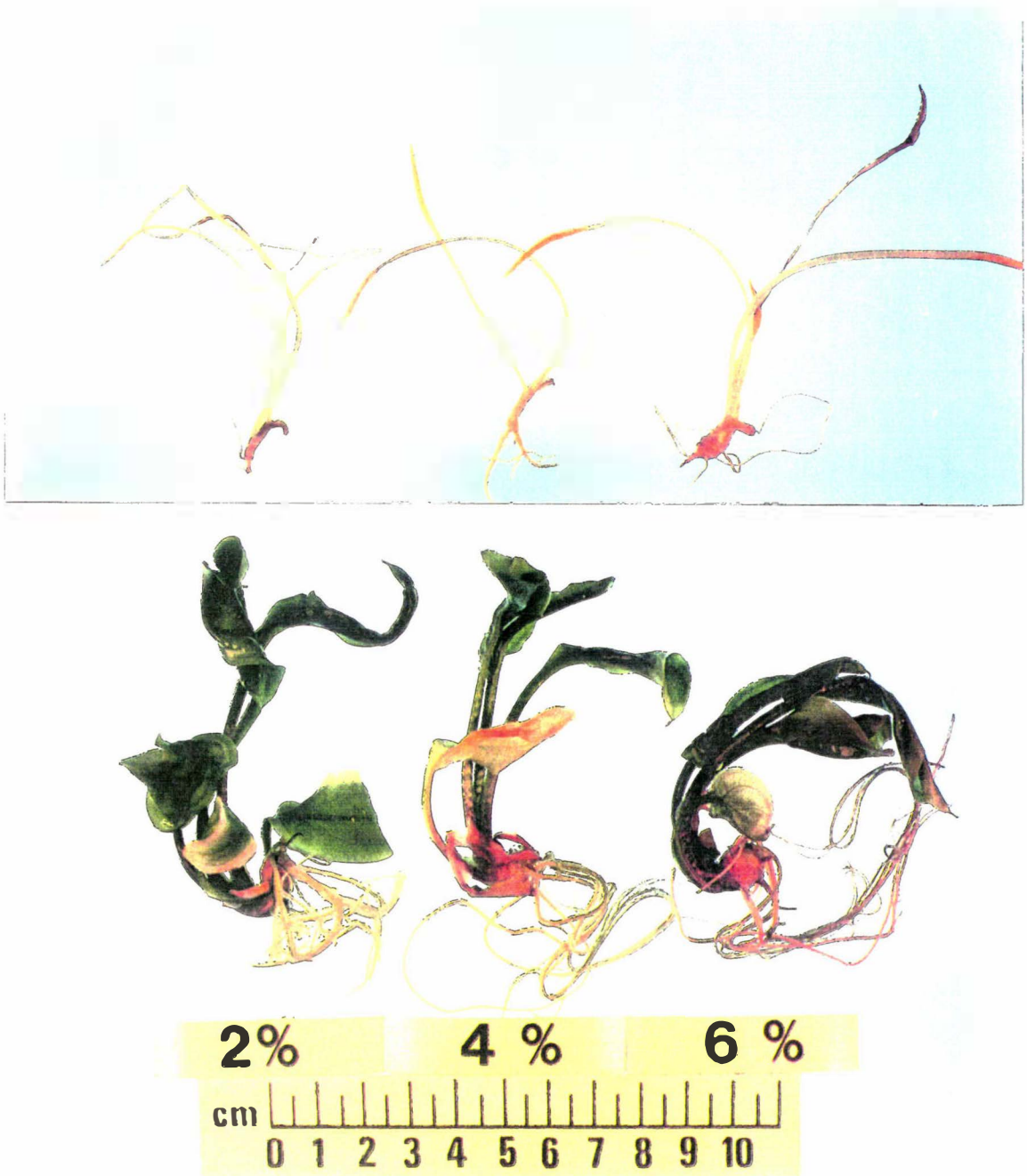

Plate 7.2 Seedlings of *Zantedeschia* 'Chromatella' after 29 weeks of cultivation in vitro at a range of sucrose concentrations in either the dark (upper) or light (45 μmol·m^{-2}·s^{-1} PPF) (lower).

Under both dark and light regimes increasing sucrose concentration resulted in a reduced number of leaves at the time of final harvest ($P \leq 0.001$, Table 7.1). While under the light regime this negative trend was linear ($P \leq 0.001$), under the dark regime the trend was quadratic ($P \leq 0.05$). In contrast, while shoot dry weight was between two and three fold greater under the light regime than under the dark, no relationship between sucrose concentration and shoot dry weight was detected ($P \leq 0.10$).

Table 7.1

Dry matter accumulation and partitioning in seedlings of *Zantedeschia* 'Chromatella,' as influenced by the presence of light or dark, and sucrose concentration in the growing medium.

Light /Dark	Sucrose conc. (%)	Total dry weight		Tuber dry weight		Proportion in tuber		Shoot dry weight		Leaf number	
		(mg)	se[z]	(mg)	se	(%)	se	(mg)	se		se
Dark	2	41.4	42.1	2.1	23.3	5.2	4.3	37.9	21.2	5.2	0.5
	4	31.4	42.1	5.7	23.3	18.1	4.3	21.9	21.2	2.2	0.5
	6	57.6	38.4	19.3	21.2	29.5	4.0	33.8	19.4	2.7	0.5
Light											
	2	144.5	23.5	27.6	13.0	22.0	2.4	97.2	11.9	6.8	0.3
	4	208.4	27.2	106.9	15.0	49.5	2.8	75.1	13.7	5.5	0.3
	6	224.8	24.3	128.9	13.5	54.3	2.5	71.2	12.2	5.1	0.3
Source[y]											
Light/Dark		***		***		***		***		***	
Sucrose		*		***		***		ns		***	
Light x Sucrose		ns		*		ns		ns		ns	
Contrast (Dark)											
Linear		ns		**		***		ns		**	
Quadratic		ns		ns		ns		ns		**	
Contrast (Light)											
Linear		**		***		***		ns		***	
Quadratic		ns		ns		***		ns		ns	

[z]Standard error of least squares means, i.e., adjusted for variable replication resulting from contamination.

[y]Probability of a significant F value; ns,*,**,*** = nonsignificant or significant at P = 0.10, 0.05, or 0.001, respectively.

7.3.2 Manipulation of source and sink strengths

At final harvest, all seedlings incubated in media amended with mannitol alone, were dead and showed little indication of previous growth. In contrast, plant loss in treatment media containing sucrose and mannitol was minimal, and attributable to fungal and/or bacterial contamination. In the absence of the plant's ability to assimilate mannitol, data presented here refer to those derived from media consisting of sucrose treatments amended with mannitol, excluding those exhibiting evidence of fungal and/or bacterial contamination.

With the exception of shoot dry weight and shoot number, significant interactions were determined between temperature and sucrose concentration with all parameters measured (Table 7.2 and Table 7.3). While the data in Table 7.2 through Table 7.4 are presented to allow determination of statistical validity, Figure 7.2(a) to (d) and Figure 7.3(a) and (b) present a more readily interpreted graphical display of the responses to increasing temperature and sucrose concentration.

As determined earlier (refer Section 7.3.1), a positive linear trend of increasing total plant dry weight with increasing sucrose concentration was determined at intermediate temperatures ($P \leq 0.001$, Table 7.4), but the magnitude of this response was minimal at the temperature extremes of 16 and 31 C (Figure 7.2a). At these temperature extremes increases in total plant dry weight were restricted compared with those at intermediate temperatures, but only at 16 C was a significant quadratic trend detected. Maximum total plant dry weight (0.4 g) occurred at a sucrose concentration of 6%, and between 21 and 26 C. These temperatures also resulted in maximum total plant dry weight at a sucrose concentration of 4%, but no differences in total plant dry weight were detected between temperatures at either 1 or 2% sucrose where growth was minimal ($P \leq 0.10$).

As with total plant dry weight, under all temperatures a positive linear trend was determined between tuber dry weight and sucrose concentration (Table 7.4, Figure 7.2b, Plate 7.3). Maximum differences between sucrose concentrations occurred between 21 and 26 C, with tuber weight being more than two hundred fold greater at 6% compared with 1% sucrose. As found for total plant dry weight, a quadratic trend of tuber dry weight, with increasing temperature, was detected at both 4 and 6% sucrose concentrations ($P \leq 0.001$, Table 7.4). However at both 1 and 2% sucrose, where growth was restricted, no trend of tuber weight with increasing temperature was detected ($P \leq 0.10$).

Table 7.2

Dry matter accumulation and partitioning in seedlings of *Zantedeschia* 'Best Gold', as influenced by temperature and sucrose concentration.

Temp. (C)	Sucrose conc. (%)	Total dry weight		Tuber dry weight		Proportion in tuber	
		(mg)	se[x]	(mg)	se	(%)	se
16	1	11.0	24.6	2.4	16.9	19.7	3.8
	2	99.8	31.8	13.6	21.9	15.4	4.9
	4	96.1	31.2	57.3	21.9	57.9	4.9
	6	169.7	34.8	130.7	23.9	77.8	5.4
21	1	10.6	29.4	1.5	20.2	14.4	4.6
	2	146.7	31.8	33.5	21.9	22.4	4.9
	4	233.6	29.4	147.9	20.2	63.0	4.6
	6	384.2	38.9	304.3	26.8	77.9	6.1
26	1	13.9	29.4	3.3	20.2	30.0	4.6
	2	142.7	27.3	20.9	18.9	15.6	4.3
	4	266.0	31.8	148.8	21.9	56.0	4.9
	6	357.4	27.5	259.5	18.9	71.2	4.3
31	1	11.8	24.6	3.0	16.9	25.1	3.8
	2	88.8	26.0	18.9	17.8	31.7	4.0
	4	114.6	24.6	54.3	16.9	40.2	3.8
	6	80.6	29.4	38.0	20.2	41.2	4.6
Source[y]							
Temperature		***		***		**	
Sucrose		***		***		***	
Temp x Sucrose		***		***		***	

[x]Standard error of least squares mean, i.e., adjusted for variable replication resulting from contamination.

[y]Probability of a significant F value; ns,*,**,*** = nonsignificant or significant at P = 0.10, 0.05, or 0.001, respectively.

Table 7.3

Dry matter accumulation within the shoot of seedlings of *Zantedeschia* 'Best Gold', as influenced by temperature and sucrose concentration.

Temp. (C)	Sucrose conc. (%)	Shoot dry weight		Shoot number		Leaf number	
		(mg)	se		se		se
16	1	7.8	10.0	1.1	0.8	2.5	1.8
	2	63.5	12.9	6.2	1.0	23.3	2.4
	4	30.3	12.9	1.7	1.0	5.5	2.4
	6	28.2	14.1	1.4	1.0	4.6	2.6
21	1	8.6	12.0	1.6	0.9	2.4	2.2
	2	84.7	12.9	3.7	1.0	16.5	2.4
	4	64.8	12.0	1.3	0.9	6.4	2.2
	6	61.9	15.8	1.0	1.2	4.3	2.9
26	1	9.8	12.0	1.1	0.9	3.1	2.2
	2	96.3	11.2	5.5	0.9	16.8	2.0
	4	84.2	12.9	1.5	1.0	6.7	2.4
	6	74.7	11.2	1.6	0.9	5.6	2.0
31	1	8.3	10.0	1.0	0.8	2.0	1.8
	2	66.0	10.6	4.0	0.8	5.6	1.9
	4	56.1	10.0	3.7	0.8	5.8	1.8
	6	36.5	12.0	1.4	0.9	5.4	2.2
Source[y]							
Temperature		***		ns		**	
Sucrose		***		***		***	
Temp x Sucrose		ns		ns		**	

[z]Standard error of least squares mean, i.e., adjusted for variable replication resulting from contamination.

[y]Probability of a significant F value; ns,*,**,*** = nonsignificant or significant at P = 0.10, 0.05, or 0.001, respectively.

Under all temperatures shoot dry weight was maximal at 2% sucrose, resulting in a significant quadratic trend of shoot dry weight with increasing sucrose concentration (Table 7.4, Figure 7.2c). With minimal shoot dry weight occurring at 1% and a maximum at 2%, under all temperatures the maximum difference in shoot dry weight, between these sucrose concentrations, averaged ten fold.

Table 7.4

Significance of trend analyses of increasing temperature and sucrose concentration on dry matter accumulation and partitioning in seedlings of *Zantedeschia* 'Best Gold'.

Source	Total dry weight		Tuber dry weight		Proportion in tuber		Shoot dry weight		Shoot number		Leaf number	
	L[z]	Q	L	Q	L	Q	L	Q	L	Q	L	Q
					Increasing temperature							
Sucrose conc. (%)												
1	ns[y]	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
2	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
4	**	*	**	*	ns	ns	ns	ns	ns	ns	ns	ns
6	**	*	**	*	ns	ns	ns	ns	ns	ns	ns	ns
					Increasing sucrose concentration							
Temperature (C)												
16	***	**	***	ns	***	***	**	***	ns	***	ns	***
21	***	ns	***	ns	***	***	***	***	ns	***	***	***
26	***	ns	***	*	***	***	***	***	ns	***	ns	***
31	***	ns	**	ns	**	ns	**	*	*	ns	**	ns

[z]Linear (L) and Quadratic (Q) contrasts/trends.
[y]Probability of a significant F value; ns,*,**,*** = nonsignificant or significant at P = 0.10, 0.05, or 0.001, respectively.

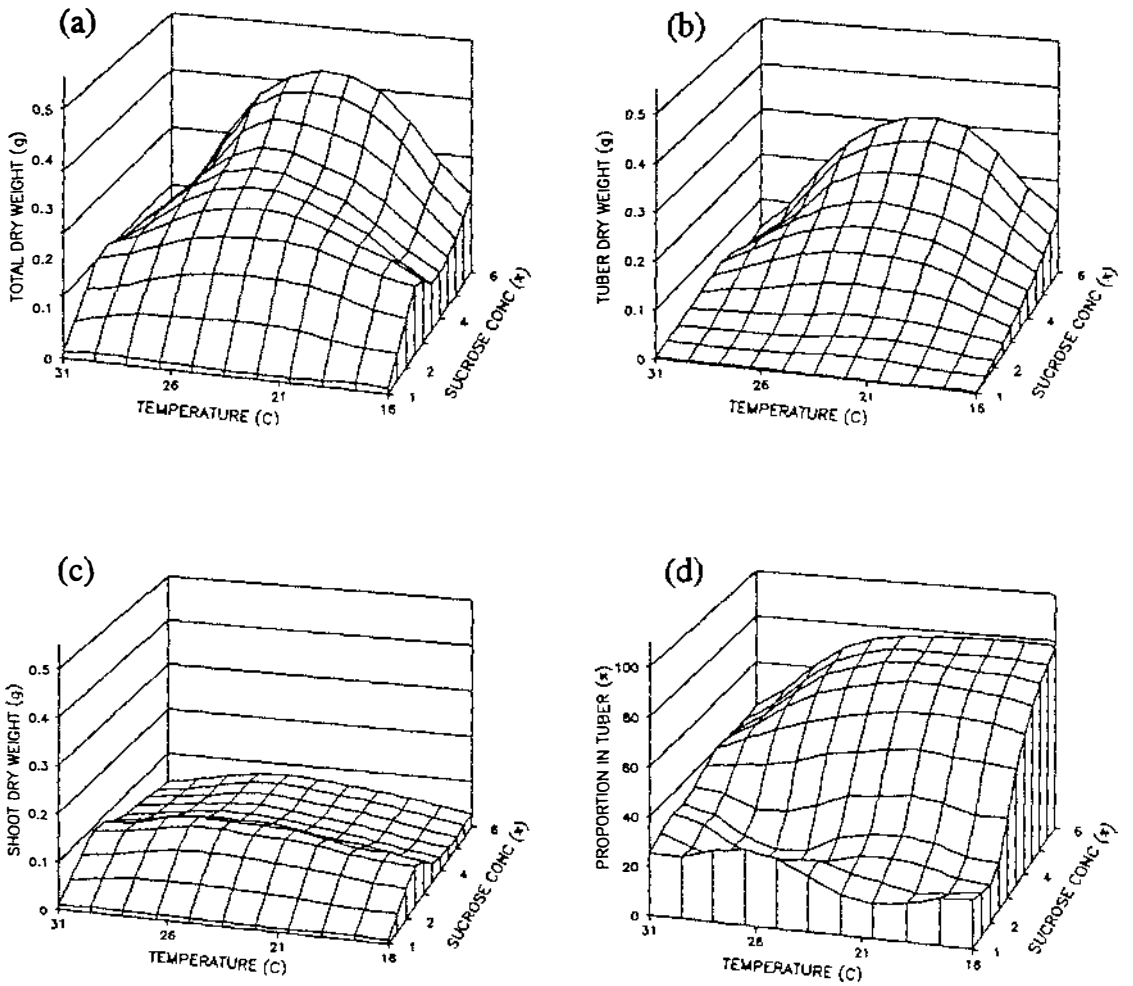

Figure 7.2 Surface response curves illustrating the influence of increasing temperature and sucrose concentration on the dry matter accumulation and partitioning in seedlings of *Zantedeschia* 'Best Gold'. (a) Total dry weight, (b) Tuber dry weight, (c) Shoot dry weight, and (d) Proportion of total dry weight in the tuber. N.B. reversal of temperature scale.

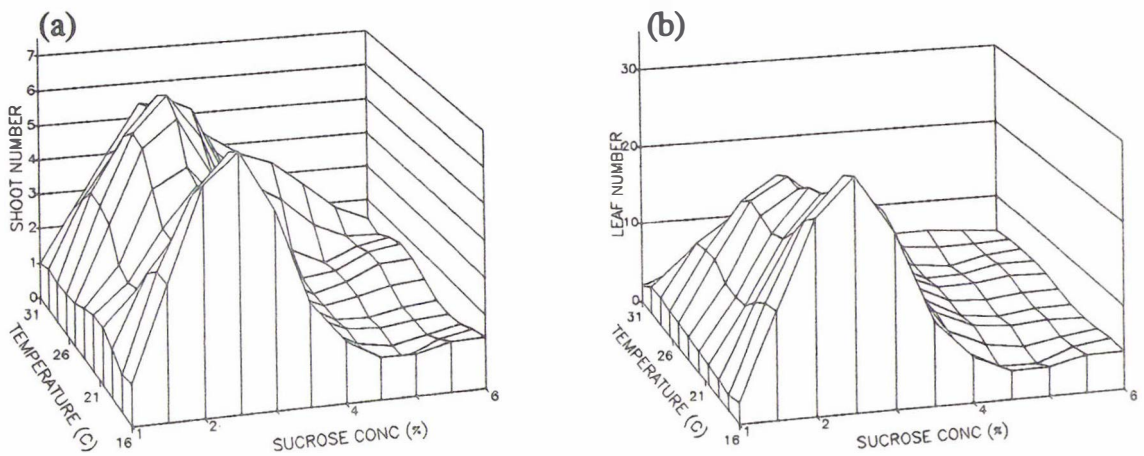

Figure 7.3 Surface response curves illustrating the influence of increasing temperatur and sucrose concentration on the number of (a) shoots and (b) leaves in seedlings o *Zantedeschia* 'Best Gold'. N.B. reversal of temperature scale.

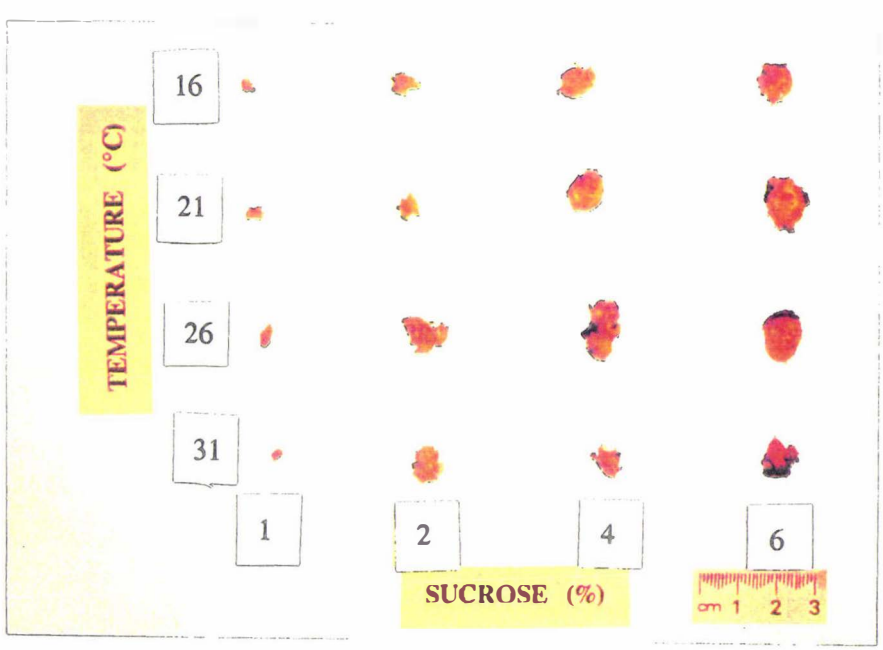

Plate 7.3 Tubers from seedlings of *Zantedeschia* 'Best Gold' after 24 weeks of growth in vitro, at a range of temperatures and sucrose concentrations. N.B. shoots and roots removed.

Treatment induced differences in shoot dry weight were paralleled by similar trends with shoot number (Figure 7.3a) and leaf number (Table 7.4, Figure 7.3b). With the exception of 31 C maximum shoot and leaf number were attained at 2% sucrose regardless of temperature. At 31 C the quadratic trend of shoot and leaf numbers with increasing sucrose concentration was not detected as at lower temperatures. Leaf number was only slightly greater at 2% sucrose than at higher concentrations at 31 C (Table 7.3), resulting in the detection of a positive linear trend at this temperature ($P \leq 0.05$).

With the exception of 6% sucrose, the proportion of total plant dry weight partitioned to the tuber differed little between temperatures ($P \leq 0.10$, Table 7.2, Figure 7.2d). At 6% sucrose this proportion was reduced by 34% at 31 C compared with all other temperatures ($P \leq 0.01$). A quadratic trend of an increased proportion of the dry weight partitioned to the tuber with increased sucrose concentration was detected at all temperatures below 31 C ($P \leq 0.001$, Table 7.4, Figure 7.2d). At 31 C a positive linear trend of increased partitioning to the tuber with sucrose concentration was detected ($P \leq 0.05$), but the greatest difference in magnitude between sucrose concentrations was always less than two fold. At 31 C the maximum proportion of dry weight partitioned to the tuber was 41.2% compared with an average of 75.6% across all other temperatures and at a sucrose concentration of 6%.

7.4 Discussion

Initiation of tuber growth in *Zantedeschia* 'Best Gold' appears to require no obligative environmental trigger. Whether incubation took place in the light or dark, and at a range of temperatures, tuber growth was noted in all treatments that sustained growth.

To discuss the current results in terms of source and sink, a number of assumptions are required. Firstly, it is assumed that sucrose concentration within the growing medium in vitro was equivalent to source strength in vivo. This seems a reasonable assumption since the primary derivatives of source activity are carbohydrates, and sucrose was found to be the principal soluble carbohydrate in tubers of *Zantedeschia* (refer Chapter 6). Secondly, it is assumed that manipulation of the incubation temperature was equivalent to manipulation of sink activity (Ho, 1979; Jones et al., 1985). With the exclusion of any influence of temperature on source strength, and a third assumption of the relative un-importance of the assimilate transport pathway (Minchin and Thorpe, 1992), the assumption that manipulation of temperature in vitro would principally alter sink activity and thereby sink strength, appears valid. Interpretation of in vitro dry weight data is also potentially confounded by respiratory losses and intersink competition (Wareing and Patrick, 1975). As the respiratory losses of imported assimilates in the sink organ can be substantial (Farrar, 1985), it should be acknowledged that in the current experiments only a measure of 'net sink strength' was possible. Similarly, the potential for inter-sink competition to alter the calculated sink strength was illustrated by Ho (1978), and therefore it must be inferred that only a measure of 'apparent' or 'net sink strength' was obtained in the current experiment. Finally, it must be assumed that growth and development was not abnormal from that achieved in vivo, with no other factors limiting growth and development. While this last assumption is less readily proven without further research, the use of in vitro techniques to repeatably produce plants of *Zantedeschia* for commercial use (Cohen, 1981) suggests that this too is a reasonable assumption. The possibility that after 24 or 29 weeks in vitro, plant growth may have become limited by the depletion of essential compounds (Morel and Wetmore, 1950; Loomis and Torrey, 1964) was not investigated. In vitro storage organ growth of other species has continued over similar time periods, and while repeated media renewal was not able to increase the duration of grain filling in maize kernels, it did increase the rate and extent of grain filling (Cobb et al., 1988; Mielke and Anderson, 1989). While it can not be stated that depletion of essential compounds did not occur, any occurrence of such an event should not have influenced the relative order of the treatment effects.

While assimilate partitioning in *Zantedeschia* seedlings must to some extent be determined genetically (Brooking and Kirby, 1981; Rapoport and Loomis, 1985; Ho, 1988; Chow et al., 1992), within the bounds of the temperature regimes used in the current experiment,

source strength appears most limiting to the partitioning of assimilates towards tuber growth (Figure 7.2d). However, at the same time, under conditions of controlled source strength it was evident that sink strength can limit final tuber yield (Figure 7.2b). The response of both total and tuber dry weights to increases in sink strength, via increased temperatures, were either not evident (e.g. 1 and 2% sucrose) or were quadratic in nature (e.g. 4 and 6% sucrose, Table 7.4). This quadratic response indicates a progressive decline in response to incremental sink strength, and hence minimal sink strength limitation. Similarly, therefore, the lack of any detectable response to increased sink strength at 1 and 2% sucrose, suggests strong source strength limitation at these concentrations. In contrast, the generally linear response of both total and tuber dry weights to increased source strength, via sucrose concentration, suggests that further potential exists for incremental source strength to increase both total and tuber weights. In addition, the lack of any detectable trend of dry matter partitioning to the tuber to increased sink strength, compared with the generally quadratic response to increased source strength (Table 7.4), also supports the hypothesis of source strength being most limiting.

Under conditions of minimal source limitation, e.g. a sucrose concentration of 6%, limitation of sink strength at the temperature extremes of 16 and 31 C, may have resulted from differing causes. As found in the in vivo experiments (refer Chapters 3 and 4), it is readily accepted that cooler temperatures, such as 16 C, might limit sink strength through limitations on growth and respiration (Wardlaw, 1968; Ho, 1979; Dinar and Stevens, 1982; Jones et al., 1985; Farrar and Williams, 1991). Such limitation is most likely to involve regulation of enzyme activity (Walker et al., 1978; Ou-Lee and Setter, 1985) and possibly either growth regulators (Melis and van Staden, 1984) or the plants' sensitivity to growth regulators (Trewavas, 1982; Firn, 1986). In contrast, at 31 C, not only was total dry matter accumulation restricted (Figure 7.2a), reflecting the same broad-spectrum limitations on growth, but also partitioning to the tuber was reduced (Figure 7.2d). This reduction in partitioning of assimilates to the tuber did not occur at 16 C, and infers the involvement of additional limitation(s) associated with assimilate utilization and/or partitioning at 31 C. Since the tuber of *Zantedeschia* is a starch accumulating sink (refer Chapter 6) those processes involved in starch synthesis are likely to be integral in determining sink strength. While not inhibiting the accumulation of soluble carbohydrates, temperatures of 30 C or higher inhibited starch synthesis in potato tubers, maize kernels, and endosperm tissue of wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) (Krauss and Marschner, 1984; Hanft and Jones, 1986; Rijven, 1986; MacLeod and Duffus, 1988). These examples of high temperature inhibition of starch synthesis were all associated with a reduction in the activity of starch metabolising enzymes, and it is suggested that the same situation may apply at 31 C in the current experiment. Since the shoot and root of the *Zantedeschia* plant can be considered to be

utilization sinks, i.e., sinks where assimilates are principally used for growth and respiration (Ho, 1988; Ho et al., 1989), high temperature inhibition of starch synthesis would not be expected to inhibit their respective sink strengths as much. Hence, while growth of all organs was restricted at 31 C (Figure 7.2a, b and c), tuber growth was restricted to a greater degree (Figure 7.2d). In addition to changes in enzyme activity, the inhibition of tuberization at high temperatures has also been associated with increased concentration of gibberellic acid (Krauss, 1978). Given the ability of exogenously applied phytohormones to influence storage organ development (Loomis and Torrey, 1964; Palmer and Smith, 1970; Peterson, 1973; Ting and Wren, 1980; Beruter, 1983; Koda and Okazawa, 1983; Rice et al., 1983; Mielke and Anderson, 1989; Vreugdenhil and Bouwmeester, 1989; Chow et al., 1992), phytohormones might also be involved in the interaction between temperature and the carbohydrate metabolism of *Zantedeschia*. A change in the plant's sensitivity to phytohormones under such temperature extremes (Trewavas, 1982; Firn, 1986) could also be involved in this interaction. The reduction in tuber growth in *Zantedeschia* following the application of GA_3 (Funnell and MacKay, 1987), has previously been discussed in Section 1.5.3.3, highlighting the relevance of considering the involvement of phytohormones.

The removal of the tuber as a competitive sink at 31 C resulted in the consequent increased partitioning to other organs. However, shoot dry weight at 31 C was no greater than at other temperatures. In addition, even with an increased source strength, this removal of the tuber as a competitive sink did not result in increased shoot dry weight accumulation. Hence in its simplest form it could be concluded that the peak in shoot dry weight accumulation at 2% sucrose (Figure 7.2c) reflected the upper limit of sink strength for this organ. Such an interpretation may be too simple however. The fact that a source strength above 2% sucrose resulted in reduced shoot growth and leaf number does not comply with such a simple hypothesis. Farrar (1990) and Farrar and Williams (1991) suggested that assimilate concentration can control growth and respiration rates. While not controlled directly through mass-action on pre-existing enzyme systems, they suggested that the concentration of enzymes is regulated by the supply of assimilate. It is therefore possible that in the current experiments, increased source strength above 2% sucrose, resulted in manipulation of the phytohormone and/or enzymatically regulated assimilate partitioning, with preference to tuber growth. If examined in terms of growth analysis such preferential partitioning of dry matter towards the tuber would be seen as an ever increasing enhancement of the magnitude of the TWP (refer Chapter 4) for as long as the availability of assimilates were in excess of the demands of competing sinks. Such a resultant increase in tuber sink strength would therefore be manifest as a reduction in shoot growth at higher concentrations as reported here. The enhanced partitioning towards tuber growth may be related to enhanced sink activity within the tuber over that of the shoot, or

simply that when grown in vitro, sucrose must pass through the tuber to gain access to the shoot. Hence in the latter case, proximity of competing sinks to the source and/or the probable use of symplastic assimilate transport to the shoot may be important (Wardlaw, 1990).

While the current experiment does not eliminate the potential involvement of osmotic potential and turgor pressure as a contributor to the control of assimilate accumulation, the use of mannitol to achieve similar osmotic potentials within all treatments indicated that the results obtained here, were independent of their influence. Although it is recognised that the osmotic potential of any one treatment medium would increase with increasing temperature, a maximum differential in osmotic potential of 3.5% would have resulted from a 2% sucrose medium held at 16 versus 31 C. This is in contrast to a 50% difference in osmotic potential as a result of increasing the main sucrose treatment concentration from 4 to 6%. After amendment with mannitol, differences in plant response were therefore more readily attributed to changes in sucrose concentration than to minor temperature induced osmotic changes. The lack of any ability to metabolise mannitol for growth and development in *Zantedeschia* was similar to that found for onions, potatoes and tulip (Heath and Hollies, 1965; Lo et al., 1972; Tovar et al., 1987; Taeb and Alderson, 1990).

The presence of light clearly has a role in stimulating growth and development in *Zantedeschia* 'Best Gold'. Although the exogenous supply of carbohydrates should theoretically have bypassed any need for development of photosynthesis, and therefore any need to supply light in vitro, the presence of light clearly enhanced dry matter accumulation and partitioning to the tuber (Table 7.1). As discussed in the introduction, the use of very low fluxes (45 $\mu mol \cdot m^{-2} \cdot s^{-1}$ PPF), together with limited gas exchange and the supply of exogenous sugars, should have been sub-optimal for any significant photosynthetic development in vitro (Grout and Aston, 1978; Hughes, 1981; Donnelly and Vidaver, 1984). At best, without increased gas exchange with the outside environment, recycling of CO_2 would only achieve compensation for respiratory losses (Abbott and Belcher, 1982). Hence it is unlikely that the presence of light in the current experiment was contributing to enhanced growth through photosynthetic carbohydrate production. Examples of the requirement of light to stimulate morphogenic development have been noted (Thorpe and Murashige, 1970; Kato, 1978; Hughes, 1981), but equally examples of nil or enhanced storage organ development in the dark have also been reported for hyacinth (*Hyacinthus orientalis* L.), lilium (*Lilium longiflorum* Hort.) and iris (Pierik and Ruibing, 1973; Stimart and Asher, 1981; Mielke and Anderson, 1989). Low irradiance, which tends to inhibit tuberization in potatoes, greatly increased the gibberellin-like substances in leaves (Woolley and Wareing, 1972). In addition, the joint application of sucrose and phytohormones stimulated growth in dark grown storage roots of turnip, radish and sugar

beet more than either sucrose or phytohormones alone (Loomis and Torrey, 1964; Peterson, 1973; Saftner and Wyse, 1984). Given the potential involvement of phytohormones in tuberization, it is possible that they, or changes in the plant's sensitivity to them, may be involved in the photomorphogenic response noted in the current experiment. In addition, it can not be ignored that synthesis of carbohydrates is not the only assimilatory process closely associated with photosynthesis e.g., assimilation of nitrogen, and the consequent synthesis of amino acids (Gibbs and Latzko, 1979) and subsequently of enzymes, also occurs. Given the involvement of enzymes in the active process of assimilate uptake and utilization (Oparka et al., 1992), limited synthesis of enzymes may also have contributed to the reduced dry matter accumulation and partitioning noted in plants incubated in the dark.

In the in vivo experiments, while diurnal changes were noted in the concentration of total soluble carbohydrates in the tuber, concentrations fluctuated little with development (refer Chapter 6). Without the ability to quantify rates of assimilate translocation, it is difficult to determine where the rate limiting steps for tuber growth exist. The constancy of tuber soluble carbohydrate concentrations could infer that their utilization for respiration, structural growth, and starch synthesis was not limited by enzyme activity within the tuber. Under such circumstances source strength and/or the assimilate translocation pathway would be limiting (Mares and Marschner, 1980; Jenner, 1982). However, if assimilate translocation occurs via a simple concentration gradient (Gifford and Evans, 1981; Jenner, 1982) this could also infer that the rate of utilization within the tuber is a rate limiting step (Ho, 1979; Dinar and Stevens, 1982). Clearly the increase of source strength in vitro increased tuber growth, indicating the likelihood of source limitation. However, due to the close proximity of the source to the sink, and the probable use of an apoplastic pathway of assimilate movement to the tuber (Gifford and Evans, 1981), any possible influence of the assimilate translocation pathway was virtually eliminated in the current in vitro experiments. Hence any direct comparison to the in vivo situation is hampered.

Results from the current in vitro experiments compare well with those of the previous in vivo experiments. Using similar temperature ranges, in both the in vitro and in vivo experiments the temperature response for final total and tuber dry weights was curvilinear. In the in vitro experiment the inability to detect a difference between the optimum temperatures of 21 and 26 C (Table 7.2), was similar to no difference being detected between 22 and 25 C in vivo (refer Chapters 3 and 4). Direct comparison between in vitro and in vivo experiments on the basis of source strength is more difficult as in vivo source size and activity was a function of PPF, temperature, ontogeny (refer Chapters 3 and 5), and carbohydrate flux. However, it can be concluded that the in vitro system described here provides a useful research tool to independently manipulate both source and sink strength for in vivo comparison.

7.4.1 Conclusions

Initiation of tuber growth in *Zantedeschia* does not require an obligate environmental trigger. The optimal conditions for in vitro tuber growth of *Zantedeschia* 'Best Gold' were between 21 and 26 C, and at a sucrose concentration of 6%. The similarity of the plant's response to temperature both in vivo, and in vitro, provides a useful research tool to independently manipulate both source and sink strength for in vivo comparison.

Both source strength and sink strength may limit tuber growth in *Zantedeschia* 'Best Gold'. At temperatures commonly encountered during the growing season, in warm-temperate climes, source strength would appear most limiting to tuber growth. While sink strength was found to be most limiting at temperatures above and below this optimum, this was under conditions of controlled source strength. Since in vivo such temperatures would potentially inhibit both source and sink strength, it is possible that both may be limiting at the same time.

Further research will be required to quantify the actual sink activity and rates of translocation to determine exactly where the rate limiting step for tuber growth in *Zantedeschia* is located. It is still uncertain whether increases in source strength beyond supra-optimal directly inhibit shoot growth and leaf development, or whether there is a direct effect of source strength on the sink activity which subsequently becomes a greater competitor for assimilates. Future investigations into concentrations and activity of carbohydrate metabolizing enzymes and phytohormones, and/or changes in the plant's sensitivity to them, should help identify and explain the biochemical basis of such rate limiting steps.

7.5 References

Abbott A.J. and A. Belcher. 1982. Analysis of gases in culture flasks. Long Ashton Rpt., 1980, p. 79.

Beruter, J. 1983. Effect of abscisic acid on sorbitol uptake in growing apple fruits. J. Expt. Bot. 34:737-743.

Brooking, I.R. and E.J.M. Kirby. 1981. Interrelationships between stem and ear development in winter wheat: the effects of a Norin 10 dwarfing gene, Gai/Rht2. J. Agr. Sci., Cambridge 97:373-381.

Chow, Y.N., C. Selby and B.M.R. Harvey. 1992. Stimulation by sucrose of *Narcissus* bulbil formation *in vitro*. J. Hort. Sci. 67:289-293.

Cobb, B.G. and L.C. Hannah. 1986. Sugar utilization by developing wild type and shrunken-2 maize kernels. Plant Physiol. 80:609-611.

Cobb, B.G., D.J. Hole, J.D. Smith and M.W. Kent. 1988. The effects of modifying sucrose concentration on the development of maize kernels grown in vitro. Ann. Bot. 62:265-270.

Cohen, D. 1981. Micropropagation of Zantedeschia hybrids. Proc. Intl. Plant Prop. Soc. 31:312-316.

Dinar, M. and M.A. Stevens. 1982. The effect of temperature and carbon metabolism on sucrose uptake by detached tomato fruits. Ann. Bot. 49:477-483.

Donnelly, D.J. and W.E. Vidaver. 1984. Pigment content and gas exchange of red raspberry in vitro and ex vitro. J. Amer. Soc. Hort. Sci. 109:177-181.

Farrar, J.F. 1985. Fluxes of carbon in roots of barley plants. New Phytologist 99:57-69.

Farrar, J.F. 1990. The carbon balance of slow-growing and fast-growing species, p. 241-256. In: H. Lambers, M.L. Cambridge, H. Konings and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague, The Netherlands.

Farrar, J.F. and M.L. Williams. 1991. The effects of increased atmospheric carbon dioxide and temperature on carbon partitioning, source-sink relations and respiration. Plant, Cell and Environ. 14:819-830.

Firn, R.D. 1986. Growth substance sensitivity: The need for clearer ideas, precise terms and purposeful experiments. Physiol. Plant. 67:267-272.

Funnell, K.A. and B.R. MacKay. 1987. Effect of GA_3 on growth of *Zantedeschia*. Tech. Rpt. 87/14, Dept. Hort. Sci. & N.Z. Nursery Research Centre, Palmerston North, Massey Univ., N.Z.

Gibbs, M. and E. Latzko. 1979. Introduction, p. 1-5. In: M. Gibbs and E. Latzko (eds.). Photosynthesis II, Photosynthetic carbon metabolism and related processes, Springer-Verlag, Berlin.

Gifford, R.M. and L.T. Evans. 1981. Photosynthesis, carbon partitioning, and yield. Annu. Rev. Plant Physiol. 32:485-509.

Gregory, L.E. 1956. Some factors for tuberization in potato. Ann. Bot. 41:281-288.

Grout, B.W.W. and M.J. Aston. 1978. Transplanting of cauliflower plants regenerated from meristem culture II. Carbon dioxide fixation and the development of photosynthetic ability. Hort. Res. 17:65-71.

Hampson, S.E., R.S. Loomis and D.W. Rains. 1978. Regulation of sugar uptake in hypocotyls of cotton. Plant Physiol. 62:851-855.

Hanft, J.M. and R.J. Jones. 1986. Kernel abortion in maize. I. Carbohydrate concentration patterns and acid invertase activity of maize kernels induced to abort in vitro. Plant Physiol. 81:503-510.

Heath, O.V.S. and M.A. Hollies. 1965. Studies in the physiology of the onion plant. VI. A sensitive morphological test for bulbing and its use in detecting bulb development in sterile culture. J. Expt. Bot. 16:128-144.

Ho, L.C. 1978. Translocation of assimilates in the tomato plant, p. 159-166. In: Opportunities for chemical plant growth regulation. BCPC Monograph 21. Boots, U.K.

Ho, L.C. 1979. Regulation of assimilate translocation between leaves and fruits in the tomato. Ann. Bot. 43:437-448.

Ho, L.C. 1988. Metabolism and compartmentation of imported sugars in sink organs in relation to sink strength. Annu. Rev. Plant Physiol. 39:355-378.

Ho, L.C., R.I. Grange and A.F. Shaw. 1989. Source/sink regulation, p. 306-343. In: D.A. Baker and J.A. Milburn (eds.). Transport of photoassimilates. Longman Scientific & Technical, Essex.

Hughes, K.W. 1981. In vitro ecology: exogenous factors affecting growth and morphogenesis in plant culture systems. Environ. Expt. Bot. 21:281-288.

Jenner, C.F. 1982. Plant carbohydrates I: Intracellular carbohydrates: Storage of starch, p. 700-747. In: F.A. Loewus and W. Tanner (eds.). Encyclopedia of Plant Physiology, New Series vol. 13A. Springer Verlag, Berlin.

Jones R.J., B.G. Gengenbach and V.B. Cardwell. 1981. Temperature effects on in vitro kernel development of maize. Crop Sci. 21:761-766.

Jones, R.J., J. Roessler and S. Ouattar. 1985. Thermal environment during endosperm cell division in maize: effects on number of endosperm cells and starch granules. Crop Sci. 25:830-834.

Kato, Y. 1978. The involvement of photosynthesis in inducing bud formation on excised leaf segments of *Heloniopsis orientalis* (Liliaceae). Plant & Cell Physiol. 19:791-799.

Kikuta, K., L. Whitney and G.K. Parris. 1937. Seeds and seedlings of the taro, *Colocasia esculenta*. Amer. J. Bot. 25:186-188.

Koda, Y. and Y. Okazawa. 1983. Influences of environmental, hormonal and nutritional factors on potato tuberization in vitro. Jpn. J. Crop Sci. 52:582-591.

Kozai, T. 1989. Photoautotrophic micropropagation of some horticultural crops. HortScience, 1989 ASHS Annual Meeting/Program and abstracts p.121 (Abstr.).

Krauss, A. 1978. Endogenous regulation mechanisms in tuberization of potato plants in relation to environmental conditions, p. 47-48. Proceedings 7th Triennial Conference European Assoc. Potato Res., Warsaw, Poland.

Krauss, A. and H. Marschner. 1984. Growth rate and carbohydrate metabolism of potato tubers exposed to high temperatures. Potato Res. 27:297-303.

Lakso, A.N., B.I. Reisch, J. Mortensen and M.H. Roberts. 1986. Carbon dioxide enrichment for stimulation of growth of in vitro-propagated grapevines after transfer from culture. J. Amer. Soc. Hort. Sci. 111:634-638.

Langford, P.J. and H. Wainwright. 1987. Effects of sucrose concentration on the photosynthetic ability of rose shoots in vitro. Ann. Bot. 60:633-640.

Lawrence, C.H. and W.G. Barker. 1963. A study of tuberization in the potato, *Solanum tuberosum*. Amer. Potato J. 40:349-356.

Linsmaier, E.M. and F. Skoog. 1965. Organic growth factor requirements of tobacco tissue cultures. Physiol. Plant. 18:100-127.

Lo, F.M., B.R. Irvine and W.G. Barker. 1972. In vitro tuberization of the common potato (*Solanum tuberosum*) is not a response to the osmotic concentration of the medium. Can. J. Bot. 50:603-605.

Loomis, R.S. and J.G. Torrey. 1964. Chemical control of vascular cambium initiation in isolated radish roots. Proc. Natl. Acad. Sci. 52:3-11.

MacLeod, L.C. and C.M. Duffus. 1988. Reduced starch content and sucrose synthase activity in developing endosperm of barley plants grown at elevated temperatures. Australian J. Plant Physiol. 15:367-375.

Mares, D.J. and H. Marschner. 1980. Assimilate conversion in potato tubers in relation to starch deposition and cell growth. Ber. Deutsch. Bot. Ges. 93:299-313.

Melis, R.J.M. and J. van Staden. 1984. Tuberization and hormones. Zeit fur Pflanzenphysiologie 113:271-283.

Mielke, K.A. and W.C. Anderson. 1989. In vitro bulblet formation in Dutch iris. HortScience 24:1028-1031.

Minchin, P.E.H. and M.R. Thorpe. 1992. Carbon-11 in the study of phloem translocation. p. 225-247. In: C.J. Pollock, J.F. Farrar and A.J. Gordon (eds.). Carbon partitioning within and between organisms. Bios Scientific Publishers, Oxford.

Morel, G. and R.H. Wetmore. 1950. Tissue culture of monocotyledons. Amer. J. Bot. 38:138-140.

Murashige, T. and F. Skoog. 1962. A revised medium for rapid growth and bioassays with tobacco tissue callus. Physiol. Plant. 15:473-497.

Ng, S.Y.C. 1988. In vitro tuberization in white yam (*Dioscorea rotundata* Poir). Plant Cell Tissue Organ Cult. 14:121-128.

Oparka, K.J. and K.M. Wright. 1988. Influence of cell turgor on sucrose partitioning in potato tuber storage tissues. Planta 175:520-526.

Oparka, K.J., R. Viola, K.M. Wright and D.A.M. Prior. 1992. Sugar transport and metabolism in the potato tuber. p. 91-114. In: C.J. Pollock, J.F. Farrar and A.J. Gordon (eds.). Carbon partitioning within and between organisms. Bios Scientific Publishers, Oxford.

Ou-Lee, T. and T.L. Setter. 1985. Effect of increased temperature in apical regions of maize ears on starch-synthesis enzymes and accumulation of sugars and starch. Plant Physiol. 79:852-855.

Palmer, C.E. and O.E. Smith. 1970. Effect of kinetin on tuber formation on isolated stolons of *Solanum tuberosum* L. cultured in vitro. Plant and Cell Physiologie 11:303-314.

Peterson, R.L. 1973. Control of cambial activity in roots of turnip (*Brassica rapa*). Can. J. Bot. 51:475-480.

Pierik, R.L.M. and M.A. Ruibing. 1973. Regeneration of bulblets on bulb scale segments of *Hyacinth*. Neth. J. Agr. Sci. 21:129-138.

Rapoport, H.F. and R.S. Loomis. 1985. Interaction of storage root and shoot in grafted sugarbeet and chard. Crop Sci. 1079-1084.

Rapoport, H.F. and R.S. Loomis. 1986. Structural aspects of root thickening in *Beta vulgaris* L. : comparative thickening in sugarbeet and chard. Bot. Gaz. 147:270-277.

Reeve, R.M., H. Timm and M.L. Weaver. 1973. Parenchyma cell growth in potato tubers. II Cell division vs cell enlargement. Amer. Potato J. 50:71-78.

Rice, R.D., P.G. Alderson and N.A. Wright. 1983. Induction of bulbing of tulip shoots in vitro. Scientia Hort. 20:377-390.

Rijven, A.H.G.C. 1986. Heat inactivation of starch synthase in wheat endosperm tissue. Plant Physiol. 81:448-453.

Rong, L., Wang, X. Qin, Wang, R. Zhen and Y. Jun. 1989. Tissue culture and rapid propagation of *Zantedeschia* hybrid. Guihaia 9:97-102.

Saftner, R.A. and R.E. Wyse. 1984. Effect of plant hormones on sucrose uptake by sugar beet root discs. Plant Physiol. 74:951-955.

Stimart, D.P. and P.D. Asher. 1981. Developmental responses of *Lilium longiflorum* bulblets to constant or alternating temperatures in vitro. J. Amer. Soc. Hort. Sci. 106:450-454.

Taeb, A.G. and P.G. Alderson. 1990. Effect of low temperature and sucrose on bulb development and on the carbohydrate status of bulbing shoots of tulip in vitro. J. Hort. Sci. 65:193-197.

Thorpe, T.T. and Murashige, T. 1970. Some histological changes underlying shoot initiation in tobacco callus cultures. Can. J. Bot. 48:277-285.

Ting, F.S.T. and M.J. Wren. 1980. Storage organ development in radish (*Raphanus sativus* L.). 2. Effects of growth promoters on cambial activity in cultured roots, decapitated seedlings and intact plants. Ann. Bot. 46:277-284.

Tovar, P., J.H. Dodds, R. Chandra and R. de Arcaute. 1987. Effect of medium composition on in vitro tuberization. Amer. Potato J. 64:462.

Trewavas, A.J. 1982. Growth substance sensitivity: the limiting factor in plant development. Physiol. Plant. 55:60-72.

Trip, P., G. Krotkov and C.D. Nelson. 1964. Metabolism of mannitol in higher plants. Amer. J. Bot. 51:828-835.

van Aartrijk, J. and G.J. Blom-Barnhoorn. 1980. Effects of sucrose, mineral salts, and some organic substances on the adventitious regeneration in vitro of plantlets from bulb-scale tissue of *Lilium speciosum* 'Rubrum.' Acta Hort. 109:297-302.

Vreugdenhil, D. and H.J. Bouwmeester. 1989. Effects of ethylene on tuberization in radish (*Raphanus sativus*). Plant Growth Regulat. 8:21-30.

Walker, A.J., L.C. Ho and D.A. Baker. 1978. Carbon translocation in the tomato: pathways of carbon metabolism in the fruit. Ann. Bot. 42:901-909.

Wardlaw, I.F. 1968. The control and pattern of movement of carbohydrates in plants. Bot. Rev. 34:79-105.

Wardlaw, I.F. 1990. The control of partitioning in plants. New Phytologist 116:341-261.

Wareing, P.F. and J. Patrick. 1975. Source-sink relations and the partition of assimilates in the plant, p. 481-499. In: J.P. Cooper (ed.). Photosynthesis and productivity in different environments. Cambridge Univ. Press, Cambridge.

Woolley, D.J. and P.F. Wareing. 1972. Environmental effects on endogenous cytokinins and gibberellin levels in *Solanum tuberosum*. New Phytologist 71:1015-1025.

Wyse, R.E., E. Zamski and A.D. Tomos. 1986. Turgor regulation of sucrose transport in sugar beet taproot tissue. Plant Physiol. 81:478-481.

8 ECOLOGICAL AND HORTICULTURAL RELEVANCE, AND MECHANISMS OF CONTROL, OF DRY MATTER ACCUMULATION AND PARTITIONING IN *Zantedeschia* 'Best Gold' - AN INTEGRATIVE DISCUSSION

In addition to discussing the possible control mechanisms of dry matter partitioning in *Zantedeschia*, each of the preceding chapters has included discussion of the ecological and horticultural relevance of the results. However, each of these discussions has been presented in isolation, each focusing on a narrow range of subject areas. This final chapter integrates the findings contained in those preceding, presenting a broad overview of the relevance and consequences of this research. In addition, proposals for further experiments are presented to indicate possible avenues of research that should improve our understanding of the control mechanisms of dry matter partitioning in *Zantedeschia*.

8.1 Ecological relevance

Relative rates of growth and development of *Zantedeschia* 'Best Gold' increased with increasing temperature, with maximum total and tuber dry matter accumulation occurring between 21 and 26 C both in vivo and in vitro. This optimum temperature range for growth was close to the average daily air temperature during the growing season, i.e. 20 C, for the sites of natural habitat of the suggested parent specie, *Zantedeschia pentlandii*. With daily maximum air temperatures during the growing season averaging 25.4 C, it is apparent that *Zantedeschia* 'Best Gold' as a representative selection of this specie, is well adapted to optimise growth under the temperature regimes of its natural habitat.

The ability of *Zantedeschia* 'Best Gold' to alter the photosynthetic rate and the partitioning of the daily increment of dry matter into leaf area (LWP) in response to the photosynthetic photon flux (PPF) under which it was cultivated, indicates that this selection is shade tolerant. This tolerance of shade, rather than being an obligative response, has its parallel in the recorded sites of natural habitat of the suggested parent specie, *Zantedeschia pentlandii*, i.e., *"open grassland"* and *"forest margins"* (Letty, 1973). Such habitats, i.e., full sun and partial shade, would result in daily integrals of PPF similar to those used in the current study. While the photosynthetic acclimation to PPF was found to be less than complete, when considered in conjunction with changes in LWP, *Zantedeschia* 'Best Gold' can be regarded as being well adapted to optimise growth under the diversity of PPF regimes of its natural habitat.

The evolution of the geophytic growth habit has been suggested as arising from the selective pressures of the growing environment, as plants migrated or climates changed (Rees, 1981 & 1984). In conjunction with the evolution of eco- and/or endodormancy and the resultant differing forms of cyclic periodicity of growth and development, the geophytic growth habit provides an excellent survival strategy for a number of plant species. As a survival strategy it would therefore be advantageous for geophytic plants to partition a greater proportion of the daily increment of dry matter toward the storage organ (i.e., tuber weight partitioning; TWP) than to leaf development, when placed in a sub-optimal environment. The magnitude of TWP in *Zantedeschia* increased at progressively lower temperatures, supporting the theory of the tuber evolving as a survival strategy. Increased partitioning of dry matter to the storage organ at cooler temperatures has been noted in a number of geophytic species (Aoba, 1976). In addition, enhanced dry matter partitioning to the storage organ in response to other environmental pressures such as declining soil moisture status has been observed (Boeken and Gutterman, 1991). However, while total plant dry matter accumulation in *Zantedeschia* was limited under low PPF, TWP was sometimes lower under the low PPF regime than under the high PPF regime. This apparent contradiction suggests that either PPF has not been a primary factor in its evolution or other forms of adaptation to PPF have taken precedence. The ability of *Zantedeschia* 'Best Gold' to acclimate to a diversity of PPF regimes through photosynthetic rate, LWP, and leaf area duration suggests that the latter is more likely to be true.

8.2 Mechanisms of control

In reviewing 60 publications from the last three decades which utilized plant growth analysis, Poorter (1990) indicated that the partitioning of dry matter into leaf area was found to be *"a crucial attribute determining the potential rate of dry matter accumulation (RGR_W) of a species."* In contrast, unless grown under conditions which result in large changes in the efficiency of leaves to produce dry matter (i.e., NAR, Konings, 1990), NAR *"seems only of secondary importance."* Data presented in this thesis also indicate the importance of partitioning of dry matter into leaf area as a primary determinant of RGR_W in *Zantedeschia* 'Best Gold'. Across a 15 C range in daily mean temperature and a single-fold difference in PPF, the rate of dry matter accumulation (RGR_W) of *Zantedeschia* 'Best Gold' was highly correlated with the partitioning of dry matter into leaf development (LWP). In contrast, only a weak correlation was determined between RGR_W and the efficiency of these leaves to produce additional dry matter, i.e., net assimilation rate (NAR). While many examples have been reported where this correlation is weak, there is evidence to suggest that this weak correlation results from a strong negative correlation between NAR and leaf area ratio (LAR), or as presented here LWP (Poorter, 1990). Hence, while NAR is recognised as an important component of RGR_W, its direct

contribution is frequently masked by concomitant changes in LAR. With photosynthetic rate being a primary component of NAR, the strong correlation between photosynthetic rate and RGR_W in *Zantedeschia* 'Best Gold' supports the interpretation that NAR is also an important determinant of plant growth. While photosynthetic rate was correlated with RGR_W, it did not correlate with the dry matter accumulation rate of the tuber (RGR_T). Clearly the photosynthetic process must be involved in contributing photoassimilates for tuber growth, but it was suggested that the plant's response to dry matter partitioning into the leaf, i.e., LWP, and the tuber, i.e., TWP, had a greater influence on the relative growth rate of the tuber than could be accounted for by photosynthetic rate alone.

In terms of RGR_T it was evident that the mechanism of acclimation to temperature under the low PPF regime was different from that under the high PPF regime. However, the mechanisms of acclimation under both PPF regimes suggested that tuber growth was source limited. Source limitation was expressed either in terms of:

1) enhanced intersink competition for assimilates, as occurred under the low PPF regime, where enhanced leaf area development (LWP) was in direct competition with enhanced tuber growth (RGR_T). This was also confirmed in vitro where dry matter partitioning to the tuber and tuber dry weight was reduced under limited source strength.

2) efficiency of dry matter accumulation of leaf area present, as occurred under the high PPF regime, where large increases in RGR_T were highly correlated with increased NAR. This was also confirmed in vitro where increased source strength increased partitioning to the tuber and tuber dry weight.

The possibility of sink limitation of growth, as well as source limitation, was clearly portrayed in the in vitro studies. However, at temperatures commonly encountered during the growing season in warm-temperate climates, source strength would appear most limiting to tuber dry matter accumulation. While sink strength was found to be most limiting at temperatures above and below this range, i.e., 31 and 16 C, this was under conditions of controlled source strength in vitro. Since under in vivo conditions such temperatures would potentially inhibit both source and sink strength, it is possible that an increase in either source or sink strength would have the potential to increase dry matter accumulation. Hence, while source and sink strengths must be in balance, in most cases dry matter accumulation can be considered to be simultaneously source and sink limited.

Under in vitro conditions of minimal source limitation, it was suggested that the sink limitation at the temperature extremes of 16 and 31 C resulted from temperature-induced

limitations on growth and respiration. While this sink limitation of tuber growth occurred at both 16 and 31 C, an additional form of sink limitation was evident at 31 C. At 31 C partitioning of dry matter towards the tuber was restricted, reflecting the possibility of high temperature inactivation of starch metabolising or sucrose unloading enzymes (Krauss and Marschner, 1984; Hanft and Jones, 1986; Rijven, 1986; MacLeod and Duffus, 1988). While it is possible that the reduced maximum total and tuber dry weights in vivo at 28 C, compared with 25 C, may have resulted from high temperature inactivation of starch metabolizing or sucrose unloading enzymes, 28 C was also associated with reduced values for LAP, LWP, and leaf area duration. Hence in vivo, at the temperature extreme of 28 C, a primary determinant of the reduced maximum total and tuber dry weights was also high temperature source limitation, through inhibition of leaf area expansion and duration.

The depression in activity of the starch metabolising enzymes, ADPG-pyrophosphorylase and starch phosphorylase, in potatoes (*Solanum tuberosum* L.) at these high temperatures, could be used as the basis of a testable hypothesis to determine the rate limiting step of high temperature reduction of tuber growth in *Zantedeschia*. Similarly, as found in maize (*Zea mays* L.) kernels, examination of the activity of sucrose unloading enzymes such as acid invertase, would determine if this process was limiting. If conducted in vitro with controlled source strength, examination of the activity of these enzymes in *Zantedeschia* at temperatures inhibiting tuber growth, compared with those promoting tuber growth, would determine if in fact these enzymes and their activity might be associated with the rate limiting step for tuber growth. Conducting such an experiment at controlled source strength would enable the hypothesized control of enzyme concentration via assimilate supply (Farrar, 1990; Farrar and Williams, 1991) to be eliminated. Since the flux of soluble carbohydrates into the tubers of potato continued at temperatures which inhibited starch synthesis (Krauss and Marschner, 1984) it is suggested that with *Zantedeschia* any influence of assimilate supply on enzyme concentration would be eliminated. However, quantification of carbohydrate fluxes, through use of radioactive labelling, would be required to validate this. Both C^{14} and C^{11} have been successfully used to investigate carbohydrate fluxes, and their relationship to source and sink strength, in a range of plant species (Ho 1979; Minchin and Thorpe, 1992). In addition, conducting this experiment in vitro with controlled source strength would also eliminate the high temperature influence on inhibition of leaf area expansion and duration, and presumably therefore reduced source strength if conducted in vivo. The examination of cell numbers and their rates of division and enlargement, concomitant with the above proposed experiment, would indicate whether these processes might also be a primary determinant of growth of the storage organ. Across a diversity of storage organs, e.g. fruit, stolon-tuber or hypocotyl-root, the production of cells and their enlargement have been shown to be integral to the final size and the amount of assimilates stored (Reeve et al., 1973; Rapoport and Loomis, 1986;

Cheng and Breen, 1992). These processes of cell division and enlargement have been suggested as contributing to the high temperature inhibition of tuber growth in potato (Krauss and Marschner, 1984).

The thickening of the hypocotyl-root of sugar beet (*Beta vulgaris* L.) can be regarded as a continuation from germination of successive initiation and enlargement of cambia and associated parenchymatous tissues (Rapoport and Loomis, 1986). In contrast, the stolon-tuber of the potato arises from the summation of two separate processes, i.e., stolon development and tuberization at the stolon tip. Therefore in contrast to sugar beet, the definite cellular differentiation that occurs in the formation of the potato tuber is more likely to involve a physiological trigger mechanism such as that involving phytohormones (Booth, 1963). There is an obvious paucity of information on the anatomical structure of the compacted stem (tuber) of *Zantedeschia* species in group 2. However, since this storage organ is derived from the stem and its initiation required no obligate environmental trigger, its development is most likely similar to the continuation from germination of cellular division and enlargement, as reported for sugar beet. Therefore it remains to be seen just how important cell division and enlargement are in contributing to the rate limiting steps of tuber formation in *Zantedeschia.*

The initiation of tuber growth, as denoted by increases in both structural and starch dry weights, did not require an obligate environmental trigger. Hence, rather than looking to the tuber for the point of control of the onset of tuber growth, it is more likely that this was dictated by the establishment of a photosynthetic leaf area large enough to provide a carbohydrate flux beyond the demands for continued leaf and root development, and respiration. A future experiment which is able to differentially manipulate the competitive sink strengths of leaf development and tuber development would test the validity of this hypothesis. Differential manipulation of competitive sink strength could be achieved by removing a variable number of dominant buds from tubers prior to planting. Through the influence of apical dominance, the application of such a treatment would create a range of potential leaf area development rates and carbohydrate fluxes. It is suggested that those treatments with a greater number of dominant buds will take longer to reach a stage where the sink strength of continued leaf and root development, and respiration, is less than the sink strength of the tuber. As a result of this delay, the timing of the onset of tuber growth would also be delayed, hence supporting the hypothesis. In addition, radioactive labelling could be used in conjunction with the above experiment to test the hypothesis that differing sink strengths were associated with differing carbohydrate fluxes.

During the last decade the frequently accepted theory that plant development is regulated by the concentration of phytohormones has been challenged (Trewavas, 1982; Firn, 1986).

Such challenges have not denounced the involvement of phytohormones, but have alerted the scientific community to consider an alternative interpretation of their involvement. Stated simply, these authors have suggested that it is the plant's sensitivity to phytohormones that changes rather than the concentration of phytohormones *per se.* Regardless of the potential for changes in sensitivity with development, phytohormone activity will involve interaction with some form of receptor within the plant, presumably a protein. While it is acknowledged that changes in sensitivity may also involve changes in affinity of receptors and the capacity of the cells to produce a response, techniques are available to determine receptor concentrations and their activities, as well as phytohormone concentrations. While it is unlikely that a phytohormone switch is involved in the tuber growth in *Zantedeschia,* the application of immunoassay techniques such as enzyme linked immunosorbent assay (ELISA), to determine their concentrations and activities, may prove beneficial in determining the potential role of phytohormones as a control mechanism for tuber development in *Zantedeschia.* Similarly, the synthesis of DNA and RNA associated with the synthesis of proteins can now be readily detected using DNA finger-printing techniques. Proteins such as patatin which have been associated with tuberization in potato, have been identified using immunoassays (Paiva et al., 1983; Park et al., 1983). The accumulation of patatin in potato plants was found to be inversely related to the concentration of exogenously applied gibberellic acid (GA_3), and therefore supports the hypothesis of phytohormone regulation of development (Hannapel et al., 1985). However, the greater sensitivity of patatin accumulation to GA_3 than starch accumulation may be indicative of the lack of a direct causal link between these events, rather than being a *"surprising"* result reported by Hannapel et al. (1985). Re-examination of the role of proteins in tuberization, which encompasses the hypothesis of altered plant sensitivity as well as phytohormone regulation, may provide a more effective avenue of research to determine the control mechanisms of storage organ development in crops such as *Zantedeschia.*

Since publication of the paper most frequently cited as first interrelating the derived quantities of plant growth analysis (equation (3.1), West et al., 1920), it might appear that our understanding of plant growth has not progressed to any degree. However, when considered in terms of the necessity for scientific hypotheses to be presented with complete unambiguity, the fact that the scientific community still utilizes this notational expression to describe plant growth can be viewed as an indication of its scientific rigour. Attempts to present some of the components of growth analysis in alternative forms (e.g. LAP and LWP), as used in the current experiments, and proposed by Jackson (1963) and Potter and Jones (1977), can still be related back to the concept that *"the growth rate of the plant depends simultaneously upon the efficiency of its leaves as producers of new material and upon the leafiness of the plant itself"* (Hunt, 1982). However, in contrast to the

conventionally used harvest index, application of this alternative expression of plant growth to partitioning of dry matter to the tuber (TWP), has provided a more sensitive measure of short term changes in partitioning as well as information on how it is influenced by growing environment (Boerboom, 1978; Keating et al., 1982), i.e., equation (4.1). In the current study the superior sensitivity of TWP over that of harvest index was evident when the harvest index continued to decline well after the commencement of tuber growth, compared with the TWP which increased immediately.

In this thesis plant growth analysis has been used to provide an empirical description of growth and dry matter partitioning, with at least the beginnings of a mechanistic model being developed, i.e., equations (4.18) to (4.23). In addition to further investigating the mechanisms and rate limiting steps of tuber growth, as outlined above, future research could also focus on developing this empirical data into an assimilate partitioning model for *Zantedeschia*. Such models have been developed for a number of species (Fick et al., 1973; Rees, and Thornley, 1973; Thornley and Johnson, 1990) and, since their robustness will be highly dependent on an understanding of the mechanisms of control, both forms of research will need to be developed concurrently.

8.3 Horticultural relevance and consequences

The temperature treatments employed in the current study included a range of mean temperatures (13 to 28 C) frequently encountered by N.Z. producers of *Zantedeschia* during the growing season, under both protected and non-protected cultivation. Similarly, the two PPF regimes employed equated to a daily integral PPF received by plants in Palmerston North, N.Z. under peak, unshaded, mid-summer (high PPF), and winter (low PPF) conditions. While it is appreciated that both temperature and PPF regimes vary with diurnal and seasonal progression, plant growth and dry matter accumulation are readily related to temperature and light integrals (Tollenaar et al., 1979; Johnson and Thornley, 1985; Warrington and Norton, 1991), and therefore the findings of this study should have direct relevance to commercial horticulturalists.

Since dry matter accumulation of *Zantedeschia* 'Best Gold' was found to be highly adaptive to PPF regimes, an important horticultural management consequence of this study is that the establishment and maintenance of an effective leaf area will be critical if growth is to be maximised. The optimum PPF under which to grow *Zantedeschia* 'Best Gold' was also dependent on temperature, with a maximum total plant dry weight occurring under the high PPF regime at 25 C.

While the estimated maximum tuber dry matter accumulation occurred under the low PPF

regime, the commercial relevance of such a finding must be interpreted with care. With financial pressure growers frequently seek crops and/or production systems that allow a short cropping period. Hence, while tubers of a greater weight may be attained eventually if plants are grown at 25 C under low PPF, tuber weight under the high PPF regime would be greater than that under the low PPF regime until 140 days (i.e., 4.7 months) of growth. Therefore producers wishing to maximise tuber weight, but requiring a shorter production period, would be more likely to meet these goals by utilizing a high PPF regime. These findings are directly applicable to growers involved in the first season's growth from tissue cultured material, where protected greenhouse cultivation is typically used. In contrast, beyond the first season's growth much of New Zealand's *Zantedeschia* production is from unprotected cultivation, and is therefore exposed to the cooler temperatures typical of a temperate to warm-temperate climate. Utilizing such a comparatively low cost production system, there is frequently less pressure to ensure a short annual cropping period. Using climate data from Palmerston North, New Zealand (40°23'S), at least 4 months would exist during the growing season where daily *"normal"* temperatures ([maximum + minimum]/2) average 16 C or greater (Shearer, 1973). Interpolation of data presented in Figure 4.2 clearly indicates that after 120 days at this temperature tuber dry weight would be greater under the low PPF regime than under the high PPF regime. Considering the additional early and late periods of the growing season where growth continues to occur, it is suggested that as long as plant spacing was appropriate and leaf area duration was maintained, shading would result in a greater final tuber dry weight compared with non-shaded crops. Following back-transformation of the data presented in Figure 4.2 after 140 days growth, tuber dry weight would be more than 60% greater under the low PPF regime than under the high PPF regime. A similar interpretation of results is attained for Kerikeri, New Zealand (35°14'S), where more than 5 months would exist during the growing season where daily *"normal"* temperatures average 16 C or greater (Shearer, 1973). Hence shading of *Zantedeschia* crops in this region also has the potential to result in a greater final tuber dry weight compared with non-shaded crops. In contrast, in Invercargill, New Zealand (46°25'S), *"normal"* temperatures during the growing season do not reach 16 C or greater, and only 2 months of the year receive a *"normal"* temperature of 13 C or greater. In the current study at 13 C, tubers of a greater weight were not attained under the low PPF regime compared with the high PPF regime until 160 days (i.e., 5.3 months). Hence use of shading in Invercargill can not be recommended. Even considering the lower daily integrated PPF in Invercargill than either Palmerston North or Kerikeri (De Lisle, 1966), the natural growing season would appear to be too short to take advantage of the use of shading. Tuber producers in Invercargill would therefore need to examine the cost effectiveness of providing heating before considering the application of additional shading.

The paucity of any quantified data for *Zantedeschia* on just what is an appropriate plant spacing to optimise plant productivity makes it near impossible to give grower recommendations at this time. The current inability of commercial producers to offer any guarantee of adequate leaf area duration, places serious doubt on the commercial viability of making a general recommendation for the use of shade for all tuber producers in regions with climates similar to Palmerston North and Kerikeri. Leaf area duration will not only be influenced by temperature and PPF regime, but also irrigation, nutrition and weed control practices (refer Section 1.5.3). In addition, the cost effectiveness of erecting shade structures for open-ground production areas will need to be evaluated before final commercial recommendations can be made.

The linear relationship between temperature and RGR_T indicated a PPF dependent base temperature for tuber growth between 4.8 and 6.1 C. This generally linear response of RGR_T with temperature, between the base temperature and 28 C, can be used as the basis of a crop model (e.g. Karlsson et al., 1989) to monitor and predict tuber growth. The development of a crop model into a grower useable system of graphical tracking (e.g. Heins et al., 1987) would require further development of nondestructive measurements of leaf area and/or weight, but would eventually provide a much needed improvement in predicting and potentially improving tuber yield.

A more immediate horticultural management consequence of the response of final tuber weight to temperature is that tuber producers must compare the cost effectiveness of growing under protected greenhouse cultivation with supplemental heating in the optimal range of 21 to 26 C, with the cost of utilizing unprotected cultivation in regions with daily *"normal"* temperatures near this optimal temperature range. Even warmer regions of New Zealand such as Kerikeri have *"normal"* temperatures during the growing season that only range between 17 and 19 C. Hence improvements in tuber yield should be obtainable in all regions of New Zealand through the use of supplemental heating. Clearly, the potential tuber yield from unprotected cultivation in warmer regions such as Kerikeri is greater than that of cooler regions such as Palmerston North or Invercargill. Considering the lack of naturally occurring environments in New Zealand with temperatures near the optimum range of 21 to 26 C, it is possible that *Zantedeschia* tuber production may be more cost effective in countries other than New Zealand. If it is more cost effective to produce tubers in countries other than New Zealand an appropriate industry development strategy must be followed to ensure New Zealand's current dominance in the international industry is not lost. An integral component of such an industry development strategy would be the continued development of breeding and selection programmes, as well as quality assurance programmes.

8.4 References

Aoba, T. 1976. Effect of temperature on the seasonal periodicity of bulb formation in plants. Environ. Control in Biol. 14:81-90.

Boeken, B. and Y. Gutterman. 1991. The effect of water on the phenology of the desert geophytes *Bellevalia desertorum* and *Bellevalia eigii*. Israel J. Bot. 40:17-31.

Boerboom, B.W.J. 1978. A model of dry matter distribution in cassava (*Manihot esculenta* Crantz). Neth. J. Agr. Sci. 26:267-277.

Booth, A. 1963. The role of growth substances in the development of stolons, p. 99-113. In: J.D. Ivins and F.L. Milthorpe (eds.). The growth of the potato. Butterworths, London.

Cheng, G.W. and P.J Breen. 1992. Cell count and size in relation to fruit size among strawberry cultivars. J. Amer. Soc. Hort Sci. 117:946-950.

De Lisle, J.F. 1966. Mean daily insolation in New Zealand. N.Z. J. Sci. 9:992-1005.

Farrar, J.F. 1990. The carbon balance of fast-growing and slow-growing species, p. 241-256. In: H. Lambers, M.L. Cambridge, H. Konings and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague.

Farrar, J.F. and M.L. Williams. 1991. The effects of increased atmospheric carbon dioxide and temperature on carbon partitioning, source-sink relations and respiration. Plant, Cell and Environ. 14:819-830.

Fick, G.W., R.S. Loomis and W.A. Williams. 1973. Computer simulation of dry matter distribution during sugar beet growth. Crop Sci. 13:413-417.

Firn, R.D. 1986. Growth substance sensitivity: the need for clearer ideas, precise terms and purposeful experiments. Physiol. Plant. 67:267-272.

Hanft, J.M. and R.J. Jones. 1986. Kernel abortion in maize. I. Carbohydrate concentration patterns and acid invertase activity of maize kernels induced to abort in vitro. Plant Physiol. 81:503-510.

Hannapel, D.J., J.C. Miller and W.D. Park. 1985. Regulation of potato tuber protein accumulation by gibberellic acid. Plant Physiol. 78:700-703.

Heins, R.D., J. Erwin, M. Karlsson, R. Berghage, W. Carlson and J.A. Biernbaum. 1987. Tracking easter lilies height with graphs. Grower Talks 51(8):64-65.

Ho, L.C. 1979. Regulation of assimilate translocation between leaves and fruits in the tomato. Ann. Bot. 43:437-448.

Hunt, R. 1982. Plant growth curves. The functional approach to plant growth analysis. Edward Arnold, London.

Jackson, J.E. 1963. Relationship of relative leaf growth rate to net assimilation rate and its relevance to the physiological analysis of plant yield. Nature 200:909.

Johnson, I.R. and J.H.M. Thornley. 1985. Temperature dependence of plant and crop processes. Ann. Bot. 55:1-24.

Karlsson, M.G., R.D. Heins, J.E. Erwin and R.D. Berghage. 1989. Development rate during four phases of chrysanthemum growth as determined by preceding and prevailing temperatures. J. Amer. Soc. Hort. Sci. 114:234-240.

Keating, B.A., J.P. Evenson and S. Fukai. 1982. Environmental effects on growth and development of cassava (*Manihot esculenta* Crantz). III. Assimilate distribution and storage organ yield. Field Crops Res. 5:293-303.

Konings, H. 1990. Physiological and morphological differences between plants with a high NAR or a high LAR as related to environmental conditions, p. 101-124. In: H. Lambers, M.L. Cambridge, H. Konings and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague.

Krauss, A. and H. Marschner. 1984. Growth rate and carbohydrate metabolism of potato tubers exposed to high temperatures. Potato Res. 27:297-303.

Letty, C. 1973. The genus *Zantedeschia*. Bothalia 11(1&2):5-26.

MacLeod, L.C. and C.M. Duffus. 1988. Reduced starch content and sucrose synthase activity in developing endosperm of barley plants grown at elevated temperatures. Australian J. Plant Physiol. 15:367-375.

Minchin, P.E.H. and M.R. Thorpe. 1992. Carbon-11 in the study of phloem translocation. p. 225-247. In: C.J. Pollock, J.F. Farrar and A.J. Gordon (eds.). Carbon partitioning within and between organisms. Bios Scientific Publishers, Oxford.

Paiva, E., R.M. Lister and W.D. Park. 1983. Induction and accumulation of major tuber proteins of potato in stems and petioles. Plant Physiol. 71:161-168.

Park, W.D., C. Blackwood, G.A. Mignery, M.A. Hermodson and R.M. Lister. 1983. Analysis of the heterogeneity of the 40,000 molecular weight tuber glycoprotein of potatoes by immunological methods and NH_2-terminal sequence analysis. Plant Physiol. 71:156-160.

Poorter, H. 1990. Interspecific variation in relative growth rate: on ecological causes and physiological consequences, p. 45-68. In: H. Lambers, M.L. Cambridge, H. Konings and T.L. Pons (eds.). Causes and consequences of variation in growth rate and productivity of higher plants. SPB Academic Publishing, The Hague.

Potter, J.R. and J.W. Jones. 1977. Leaf area partitioning as an important factor in growth. Plant Physiol. 59:10-14.

Rapoport, H.F. and R.S. Loomis. 1986. Structural aspects of root thickening in *Beta vulgaris* L. : comparative thickening in sugarbeet and chard. Bot. Gaz. 147:270-277.

Rees, A.R. 1981. Concepts of dormancy as illustrated by the tulip and other bulbs. Ann. Applied Biol. 98:544-548.

Rees, A.R. 1984. Dormancy, flowering and periodicity in ornamental bulbs. The Plantsman 6(1):33-44.

Rees, A.R. and J.H.M. Thornley. 1973. A simulation of tulip growth in the field. Ann. Bot. 37:121-131.

Reeve, R.M., H. Timm and M.L. Weaver. 1973. Parenchyma cell growth in potato tubers. II. Cell division vs cell enlargement. Amer. Potato J. 50:71-78.

Rijven, A.H.G.C. 1986. Heat inactivation of starch synthase in wheat endosperm tissue. Plant Physiol. 81:448-453.

Shearer, A.R. 1973. Summaries of climatological observations to 1970. N.Z. Meterological Service Miscellaneous Publication 143. Government Printer, Wellington, N.Z.

Thornley, J.H.M. and I.R. Johnson. 1990. Plant and crop modelling - a mathematical approach to plant and crop physiology. Clarendon Press, Oxford.

Tollenaar, M., T.B. Daynard and R.B. Hunter. 1979. Effect of temperature on rate of leaf appearance and flowering date in maize. Crop Sci. 19:363-366.

Trewavas, A.J. 1982. Growth substance sensitivity: the limiting factor in plant development. Physiol. Plant. 55:60-72.

Warrington, I.J. and R.A. Norton. 1991. An evaluation of plant growth and development under various daily quantum integrals. J. Amer. Soc. Hort. Sci. 116:544-551.

West, C., G.E. Briggs and F. Kidd. 1920. Methods and significant relations in the quantitative analysis of plant growth. New Phytol. 19:200-207.