GENOTYPIC VARIATION OF DORMANCY IN WHEAT

(*Triticum aestivum L.*)

A Thesis presented in partial fulfilment of the
requirements for the degree of

Master of Agricultural Science

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1996

Abstract

Embryo dormancy and α - amylase dormancy are desirable in wheat to minimise pre-harvest sprouting damage. The current work focuses on the embryo and graincoat colour.

A loose association between grain redness and dormancy in wheat is common knowledge. But the causal relationships between colour and dormancy are not clear and need to account for dormancy variability in the gene - pool. The study's working hypothesis was that colour formation triggers hypo - oxia synthesis of ABA (*vs.* gibberellins) which triggers dormancy if the timing with embryo development is optimal.

Development profiles for eight attributes (including dormancy) of grain were investigated from five white and five red wheat cultivars representing a wide genetic base. Tagged ears were sampled from pollination to harvest ripeness (days after pollination to 12.5 % moisture). All the white - grained cultivars did not have dormancy at harvest ripeness, and there was considerable variation of dormancy levels in the red - grained cultivars. The total-grain abscisic acid was not associated with redness nor dormancy, and no evidence of ABA sensitivity could be found in cv. Brevor. The failure to detect the putative dormancy of cvs. Brevor and Kenya 321 was probably due to fine detail employed in the present work, but may also have been due to the single ripening environment used. Base α - amylase and flavanol levels did not contribute to the variation in embryo dormancy. Gibberellic acid insensitivity in the *Rht/Gai* genotypes was not expressed in terms of embryo dormancy. Examination of the profiles suggested that redness was necessary to permit dormancy, but that dormancy *timing* was independent of colour. This led to varying levels of dormancy at harvest ripeness. No association with ABA was evident, nor with colour precursor. However timing and duration of polymerisation (flavanol) development (hypo-oxia) did show a weak association with dormancy delay and level.

The new hypothesis suggests that colour formation hypo-oxia permits dormancy, but that its timing is flexible with respect to harvest ripeness. Broader genetic control (other than the Redness gene) is indicated. Heritability estimates indicated that timings, rather than levels, are more useful selection criteria. This included

embryo dormancy attributes, colour, and harvest ripeness. For plant breeders it suggested that grain sampled at harvest ripeness could be selected for dormanc as measured in this study.

ACKNOWLEDGEMENTS

I wish to thank:

Dr. I. L. Gordon, Senior Lecturer, Department of Plant Science, Massey University, New Zealand, without whose valuable guidance this thesis will not have been finished.

Dr. D. Woolley for his valuable help with the hormone analysis work.

The Seed Technology Centre, Massey University, for use of the facilities that were necessary in this work.

My wife for her encouragement and motivation in this study.

Norman Zvomuya
1996

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Chapter 1

The nature and extent of preharvest sprouting damage in the grain industry

Sprouting damage is an important problem in the grain industry. When sprouting damage occurs before the grain is harvested it is known as *preharvest sprouting damage*. All the major cereal crops, i.e. wheat, maize, rice and barley, may be affected when grown under weather conditions conducive to germination. The areas mostly affected by preharvest sprouting include the wet or humid regions of the world, such as, northwest Europe, North and South America, parts of Australia, and New Zealand (McEwan, 1967; Belderok, 1967). In New Zealand, sprouting damage occurs most frequently in the Manawatu region, the southern part of the Canterbury Plains and Southland although climatic conditions ideal for sprouting can occur in any of the wheat - growing districts (McEwan, 1967).

The problem manifests itself in the form of a high level of enzymes involved in germination, e.g., proteinases, pentosanases, glucanases and α-amylase (Gale, 1976; Olered, 1976; Bewly and Black, 1994), and starch phosphorylases and hemicellulases in the intact grain (Ching, 1972; Kruger and Preston, 1976). These enzymes are stimulated or activated by gibberellins which are secreted by the embryo and the scutellum. Such enzyme mobilization and activation may be observed in conditions which delay and are favourable for the synthesis of germination - type enzymes without apparent associated embryo germination (Flintham and Gale, 1988).

A high level of α - amylase activity causes hydrolysis of the carbohydrate reserves which are translocated to and are used by the embryo during germination. As a result, sprouting - damaged grain typically has reduced test weight, a mealy texture and a low milling yield. Flour from damaged grain tends to have a high paste viscosity (i.e. low Hagberg falling number). The bread-making quality of such flour is poor (Gordon, 1975; Olered, 1976; Gale, 1976) as reflected in bread having an excessively dark and sticky crumb which may cause the breakage of cutting blades during high speed slicing of bread in the bakery. The incidence of collapsed loaves may also be high. Once present α-amylase cannot normally be

inactivated, hence a maximum level of enzyme is usually specified for many uses (*Plant Breeding Institute*, 1990).

Table 1 below shows the extent of the problem in terms of its geographic distribution and economic cost. The cash loss to farmers in the United Kingdom in the years 1985 and 1987 was estimated at 50 - 60 million pounds sterling. Extra wheat was imported in order to compensate for the loss in home - grown material leading to substantial deficits in the national balance of payments (Bewley and Black, 1994). In the northern region of Brazil, preharvest sprouting is considered to be the most serious problem facing wheat growers and it causes great losses to the producer.

The tendency to sprout is influenced by the interaction between genotype of the grain and the environmental factors present both before and during the harvest season (Stoy, 1983). However, preharvest sprouting is intimately dependent on the absence or presence of dormancy. It is known that dormancy is strongly expressed at temperatures above about 13 °C (Reddy *et al.*, 1985; Bewley and Black, 1994), a condition referred to as *relative* dormancy, although sprouting may occur even in grains with relative dormancy when the temperature is low. This is one reason why preharvest sprouting is common in wheat when wet weather prevails at certain times during maturation, whereas in regions that are warm and dry during grain development and maturation wheat is not likely to suffer from preharvest sprouting.

Genetic variation within and amongst wheat cultivars makes it difficult to find a universal solution to the problem of preharvest sprouting damage although an integrated approach of the application of knowledge already accumulated from field and laboratory studies may lead to the formulation of effective regional strategies for protecting the crop. Kruger (1975) cautioned that sprouting in the ear under field conditions may be very different from seed germination under laboratory conditions.

The damage caused by preharvest sprouting in various parts of the world.

Year	Place	Amount damaged	Reference
1969	New South Wales (Australia)	1.8 million tonnes white-grained wheat	Bewley and Black (1994)
1976	Switzerland (Europe)	42 % of total wheat crop	Weilenmann (1980)
1977	Nebraska (USA)	12 % of total red-grained wheat 19 % of total durum wheat	Bewley and Black (1994)
1983/84	Australia	20 % of total wheat crop	McMaster (1987)
1985	United Kingdom (Europe)	20 % of total wheat crop	Bewley and Black (1994)
1987	United Kingdom (Europe)	20 % of total wheat crop	Bewley and Black (1994)
1984 - 1988	Poland (Europe)	US $ 276 million (total value of crop damaged)	Bewley and Black (1994)

However, results from laboratory studies have proved useful to designers of sprout - warning systems in various countries, e.g., Belderok (1968) in the Netherlands, Schrodter and Grahl (1978a, 1978b) in West Germany, Lalluka (1976) in Finland and Olssn and Mattsson (1976) in Sweden. In these systems, sprouting forecasts are transmitted by radio to wheat farmers who then use the information to plan appropriate strategies. In the USA, Briggle (1979) reported adequate control of the problem in Michigan. Apparently this was a result of the adoption of new production management methods whereby sprouted grain was mixed with unsprouted grain in a proportion that did not affect the quality of flour adversely. Wahl and O'Rourke (1993) used a simulation model (SWOPSIM) to examine the effects of minor sprout damage (*less than* 5%) in the major wheat exporting countries on world wheat markets. In the USA and the European Union, which allowed blending for minor levels of sprout damage, wheat producer revenues were not affected as severely as in Canada and Australia which did not allowed blending. World markets are affected as regional effects were transmitted to international markets. The main finding in the study was that, in general, regions not affected by sprout damage will gain, while, in some cases, the region affected by sprout damage may have aggregate gains even though individual growers or localities suffer heavy losses.

Resistance to preharvest sprouting by suppressing or preventing germination under cool and wet ripening conditions may be necessary as a means of protecting the cereal grain from damage in the field. Three factors were identified as determining the resistance of cereal cultivars to preharvest sprouting (Strand, 1979), viz., seed dormancy at harvest, germination inhibitors in chaff structures and moisture conditions of the ear due to cultivar genotype. Of these, seed dormancy was shown to be responsible for up to 64% of the variation in sprouting resistance while the other two factors accounted for the rest. Because of the close relationship between dormancy and resistance to preharvest sprouting, the approaches used in the study of preharvest sprouting damage involve examination of the factors that influence the onset and maintenance of dormancy in developing and mature grain.

None of the putative genes for seed dormancy have been described in the majority of wheat varieties. It is probable, however, that dormancy in the red - grained wheat varieties may, in part, be a consequence of the biological synthesis of the

polymers comprising the colour pigment (*phlobaphenes*) of the graincoat. Dormancy may also be influenced by the very physical nature and chemical properties of these polymers. Gordon (1979) discussed these issues at length and suggested that a period of increased oxidative activity in the graincoat of red - grained wheats during development could deprive the embryo of sufficient oxygen needed for normal respiration and the provision of energy for growth. This may occur during the polymerisation of flavanols to form the red pigment, phlobaphene, a process that requires molecular oxygen and is catalysed by the polyphenol oxidase group of enzymes. Gordon (1979) also noted that while abscisic acid (ABA) is degraded in the presence of oxygen (and light) the synthesis of gibberellic acid (GA) requires the presence of molecular oxygen. Hence, the available oxygen within the grain tissues may ultimately determine the turnover rate and amount of both inhibitor and promoter in the grain and, therefore, influence dormancy and germinability. Therefore, any mechanism that restricts the rate of diffusion of oxygen into the grain or the absolute amount of oxygen reaching the inner tissues of the grain may contribute to a greater or lesser extent to the onset of grain dormancy and suppression of grain germination, especially if such restriction occurs at the time when the embryo is gaining its basic germinative ability. In mature seeds, the situation may be quite different since Walker-Simmons (1987) observed that dormancy then no longer depended on absolute ABA levels but was related to tissue sensitivity to ABA. It should be noted, however, that equivalent amounts of flavanols (Gordon, 1975), and polyphenol oxidases (Verry, 1978) were reported in both the red - and white - grained wheats and that there were no striking patterns in the amounts or peaks of activity of free and conjugated abscisic acid associated with red - or white - grained wheats (Verry, 1978).

The tanning properties of the phlobaphenes enable these oligomers to form complexes with graincoat proteins leading to the observed denser structure of the graincoats of red wheats. During imbibition such graincoats absorb water and swell causing an increase in resistance to oxygen diffusion into the grain largely because of the poor solubility properties of oxygen in water. The increased resistance to oxygen entry into the grain leads to a state of hypo - oxia in the internal tissues. The consequence of this mechanism in developing wheat grains is a slowing down in embryo development and accumulation of dormancy - promoting inhibitors at the expense of germination stimulants. This situation may cause a general disability by the embryo to accumulate its basic germinative

apparatus. If the activity of inhibitors (e.g. ABA) rises to a level sufficiently high to allow for the onset of dormancy then the embryo will not germinate upon reaching maturity even if conditions suitable for germination are available. In any case, if embryo maturity is not attained by harvest ripeness dormancy will be observed.

According to Gordon (1979) variations in grain development involving the establishment of factors required for germination prior to imbibition contribute to the variability in germinability in the red and white wheats alike. The basic difference between these wheats is the presence of the red pigment, phlobaphene, in the graincoat of red wheats and its absence in white wheats. This difference contributes to a large extent to the differences in dormancy levels between the red - and white - grained wheats. Another important difference in these wheats is that of sensitivity to hormones, for example, gibberellic acid insensitivity in some dwarf varieties and ABA sensitivity in some white varieties.

The present study was carried out in order to investigate the timing of events in developing wheat grains from the time of pollination until after harvest ripeness. The main focus was on the relationships between the pattern of graincoat colour development, the concentration of flavanols in developing grains, the timing of ABA peaks of activity and duration of these peaks, embryo maturity and the level of dormancy in developing wheat grains of ten wheat cultivars representing a diverse genotypic base. This study, therefore, extends the scope of previous studies at by examining simultaneously several physiological characters expressed in wheat from a wide genetical base within a common environment. The information obtained will provide an important insight in the physiology and genetical correlations of preharvest sprouting damage.

Chapter 2

REVIEW OF LITERATURE

2. INTRODUCTION

The important agronomic characters of yield and quality in wheat are measured in terms of the grain which is the unit of economic importance. Preharvest sprouting affects both these characters often with disastrous consequences (*see* Introduction). The study of preharvest sprouting damage, therefore, demands a good understanding of grain ontogeny, grain morphology, and cereal technology. Grain ontogeny is influenced by genetical, physiological and environmental factors that interact to form a unique biological system for each individual grain. This review concerns mainly the first two of these factors.

2.1 The wheat grain

The wheat grain is a fruit which comprises a single seed completely surrounded by an adherent pericarp, except at the point of attachment to the rachilla. It is dry at maturity, is formed from a single carpel and has no special method of opening to liberate the seed (Evers and Betchtel, 1988). The embryo is located on the dorsal side of the grain.

2.1.1 Water relations of the wheat grain

There is little resistance to the entry of water in the mature wheat grain. Water enters the seed in the region where the epidermis is particularly thin (Bradbury *et al.*, 1956a; Hinton, 1955) or at the interface between embryo and seed coat on the dorsal side. Lee and Atkey (1984) described stomata close to the apex on the crease side of the grain. About 25% of the stomata were reported to remain open at maturity by these authors. Cracks in the cuticle at maturity have been implicated in the lack of resistance to water at this stage (Woodbury and Wiebe, 1983; Lee and Atkey, 1984).

2.1.2 Anatomical description

The main anatomical features of the wheat grain are the seed coat, the endosperm and the embryo

2.1.2.1 The seed coat

The seed coat of wheat is the outermost layer of the true seed and is derived from the integuments that surround the nucellus at the time of fertilization. The mature seed coat comes from only the inner integument, the outer integument having degenerated early in development (Morrison, 1975). The seed coat and the pericarp are physically united (to form a "graincoat") with the innermost tissues of the pericarp adhering firmly to the cuticle on the outer surface of the seed coat. In red - grained wheats red - brown pigmentation is present in both the seed coat and the pigment strand but white - grained wheats lack this pigmentation. In the seed coat, the pigment is found in the colour layer which consists of two layers of cells beneath the cuticle. The structure of the seed coat is brittle where it overlies the embryo and is particularly susceptible to breakage partly because of the lack of support resulting from the space between it and the embryo surface.

2.1.2.2 The endosperm

The endosperm forms the largest and most important economic part of the grain. The start of endosperm development in wheat is by free nuclear division before cellularisation. Cell division is completed by day 14 (Brocklehurst, 1977) at which stage the endosperm is only a fraction of its final size. There is no vascular connection between the developing endosperm and the parent plant and solutes have to pass through a series of transfer cells to reach the endosperm. At maturity, the inner cells of the endosperm are generally dead tissue, packed solidly with insoluble food reserves such that any surviving cytoplasm is very distorted and does not survive desiccation. The endosperm consists of the aleurone layer and the starchy endosperm.

2.1.2.2.1 Aleurone layer

This is the outermost layer of the endosperm tissue and surrounds the grain over the starchy endosperm and part of the embryo. In wheat, it is one cell-layer thick whereas in barley it comprises several layers of cells at maturity. The aleurone layer is the only living tissue of the endosperm at maturity and is the site where most of the enzymes responsible for food mobilization on subsequent germination

are synthesized. Gibberellic acid and / or calcium ions will trigger the release, in an active form, of enzymes contained in the aleurone tissue which are capable of initiating the process of food reserve breakdown. Alpha - amylase is synthesized *de novo* as germination begins and appears to be the most efficient enzyme for starch degradation. Protein bodies found in aleurone tissue contain both inactive enzymes (e.g. β - amylase and carboxypeptidases) and protein reserves which will be eventually broken down to amino acids and then reassembled as the more active hydrolases synthesized on germination. Aleurone tissue contains very little starch.

2.1.2.2.2 Starchy endosperm

Endosperm cells are filled with starch granules embedded in a protein matrix (Bradbury *et al.*, 1956a, 1956b; Simmonds, 1972; Fincher and Stone, 1974). The starchy endosperm of wheat at maturity contains primarily two types of starch granules although considerable variation in the characteristics of starch grains both between cultivars and different positions in the seed is known to exist. Large type A and small type B granules as well as a small proportion of underdeveloped type A granules in the sub-aleurone region are found in all except the peripheral cells. The large granules are lens - shaped while the small granules are near - spherical. The type A granules are the initially formed ones observed in the cytoplasm after cellularisation of the endosperm. The final number of type A granules is determined by the number of plastids present when cell division ceases. The final size of A granules varies from about 30 to 50 μm and may be influenced by cultivar (Dengate and Meredith, 1984) and environment (Baruch *et al.*, 1979). The type B granules are not initiated until after cell division stops (Parker, 1985). They are often found in association with the equatorial (peripheral) groove and tubuli of type A granules or in extensions of the type A plastids.

The proportional contributions of starch and protein vary according to cell position, with the peripheral cells having the lowest starch content and therefore the highest protein percentage since all the cells have approximately the same mass of protein. Starch is the principal carbohydrate store in the wheat caryopsis. Accumulation of starch begins immediately after cell division ceases and the grain filling rate is closely related to the number of endosperm cells produced during the

initial stage of development (Brocklehurst, 1977; Nicholas *et al.*, 1980; Herzog, 1986).

2.1.2.3 The embryo

The embryo is composed of the embryonic axis and the scutellum. It lies on the dorsal side of the caryopsis and is, in reality, a miniature living plant. The mature embryonic axis can be divided into three regions, viz. the *shoot* or *epicotyl*, the *mesocotyl*, and the *radicle*. The embryo is essentially complete structurally by the start of the grain-filling stage, but most of the storage reserves have not been deposited. Large amounts of rough endoplasmic reticulum (RER), Golgi bodies, mitochondria and polysomes are found in the cytoplasm of the embryonic cells. Storage reserves in the embryo begin to be deposited toward the end of the endosperm cell division phase. The primary reserves are lipid droplets and the protein bodies. Starch is only occasionally found in the mature embryo. Lipid droplets are found in the embryo tissues early in development but at low frequency. The droplets become more numerous during grain filling and continue to increase in number during embryo maturation. Protein bodies form in small vacuoles after the major structures of the embryo have formed.

2.2 Dormancy and germination

Grain germinability and dormancy are both desirable features but only if they occur at appropriate times in the history of the grain. While uniform and rapid germination is a desirable attribute in the seed bed, a short dormancy period is often necessary to protect the mature grain from sprouting before harvest. Seed dormancy and seed germination are mutually exclusive events. They cannot both occur at the same time since dormancy, if it is present, must be broken before germination can take place. According to Karssen and Groot (1987), there cannot exist a state of partial dormancy. However, dormancy is a relative rather than an absolute state and is highly variable from cultivar to cultivar, and even within the same seed lot (Pollock and Toole, 1961).

2.2.1 Dormancy

Much of the variation in sprouting resistance found among wheat cultivars was reported to be due to dormancy (Strand, 1979). A working definition of dormancy is that it is a state of arrested development of seed embryos, buds or spores under

conditions **otherwise suited for growth** (Amen, 1968; Villiers, 1972; Taylorson *et al.*, 1977; Strand, 1979). Dormancy may be important in wild cultivars where it is part of an intrinsic survival strategy operating differentially through the dispersal units (Mackey, 1975). Domestication of crop cultivars brought with it variable changes in the response of these plants to their physical and chemical environments. Moreover, increased farm mechanization and the need to meet demand for higher quality grain by processors have seen a deliberate selection for uniform germination in cereal crops by plant breeders. In wheat and other grain crops a short dormancy period is often needed to prevent germination before harvest (Taylorson *et al.*, 1977), yet selection for high protein content and vigorous seed as well as shifts in crop husbandry practices may have selected for sprouting in cereals (King, 1983).

2.2.2 Control of dormancy

Dormancy is a complex control system which is known to be influenced by the female parent (Mao *et al.*, 1983) and may be controlled by at least two recessive genes. It is difficult to manipulate in breeding programs. Roberts (1962) proposed that loss of dormancy is dependent on some oxidation reaction. However, all the common inhibitors of cytochrome oxidase (CN^-, Sodium azide, CO, H_2S and hydroxylamine) failed to prolong dormancy but were very effective dormancy - removing agents on rice, *Oryza sativa* L. (Roberts, 1964a, b). The NO_3^- and NO_2^- ions are stimulatory but reduced forms of nitrogen (e.g. NH_4^-, urea and amino acids) have no effect. Oxidized forms of nitrogen were effective by virtue of their ability to act as H acceptors rather than as a nitrogen source. A paradox exists, however, and Roberts (1964a) found that dormancy could be removed in rice by increased oxygen pressures or by the application of alternative hydrogen acceptors. Respiratory inhibitors could also remove dormancy. The common property of the successful germination stimulators was that they restrict oxygen uptake in the intact seed (Major and Roberts, 1968a).

Major (1966) proposed that it might be the oxidative pentose phosphate pathway (PPP) which is involved in loss of seed dormancy. However, the significance of this pathway has been discussed by several authors (Gordon, 1979; Kruger, 1989; Flintham, 1992) and it appears that the pentose phosphate pathway (PPP) may play only a minor role in the control of dormancy in wheat.

2.2.3 Variation in seed dormancy

There is considerable variation in dormancy levels within cultivars and between cultivars. In wheat, dormancy levels are generally higher in the red - grained wheats than in the white - grained varieties. However, the environment has considerable influence on seed dormancy. Strand (1983), in a study lasting 15 years, attributed 65.3% of the variation in seed dormancy to the environment. Hence, the level of dormancy, once established may show considerable instability due to seasonal conditions so that even slight differences in maturation timing could completely remove what was considered stable dormancy.

2.2.4 Dormancy mechanisms

The major dormancy mechanisms recognized in cereals are: (i) seed coat imposed dormancy, (iii) embryo - related processes (e.g. the presence of chemical inhibitor(s), lack of promoter or respiratory control due to inadequate oxygen supply to the embryo), and (iv) various types of combined dormancy (Nikolaeva, 1969). All these mechanisms fall under the general definition of *primary dormancy* described by Villiers (1972) as the inherent dormancy property of the organ or organism caused by endogenous factors. An additional and distinct mechanism, s*econdary dormancy*, occurs when seeds are imbibed under conditions unfavourable to the germination of that cultivar, e.g. when seeds are imbibed at temperatures unfavourable to germination they become unable to germinate at temperatures normally favourable for germination of that cultivar until some special releaser stimulus is applied (Villiers, 1972). Q*uiescence* describes a state of arrested development through which seeds pass on reaching maturity.

2.2.4.1 Coat imposed dormancy

Impermeability of the seed coats to water is one of the simplest yet most highly effective means of delaying germination. Such "hard-seededness" is exhibited by members of several families including *Leguminosae*, *Malvaceae*, *Chenopodiaceae*, *Liliaceae*, and *Solanaceae* but is not important in cereals. Dormancy in hard seeds is relieved by scarification or chemical treatment. The testa (the true seedcoat) also constitutes a physical barrier to oxygen diffusion needed for the provision of energy through oxidation processes (Villiers, 1972; Gordon, 1975; Verry, 1978). This is the case in dry mature cereal grains where the seed coats are freely permeable to water but swell on imbibition. Oxygen is

sparingly soluble in water: therefore, diffusion is the main way O_2 can pass through the seed coat. Removal or damage to parts of the seed coats (Wellington, 1956b; Belderok, 1968; Gordon, 1970; Lancaster and Wright, 1970) or increasing the oxygen tension of the surrounding air, e.g. by adding hydrogen peroxide to the imbibing solution (Belderok, 1976) leads to increases in the rate of embryo respiration in many cultivars of seeds, and frequently results in germination (Miyamoto *et al.*, 1961; Sanders, 1965; Villiers, 1972; Mao *et al.*, 1983; Black *et al.*, 1987). In developing grain the barrier to oxygen entry may occur through the enzymatic consumption of oxygen by the grain coat tissues thus depriving the embryo itself. Polyphenol oxidases have been found in cereal caryopses and are implicated in this process (Kruger, 1976; Gordon, 1979). Dormancy is also affected by temperatures above or below a critical value (*secondary dormancy*), e.g., dormancy is strongly expressed at temperatures above 13 ^{0}C (Reddy *et al.*, 1985; Bewley and Black, 1994). The reason for this may be the influence of temperature on membrane transitions (Black *et al.*, 1987) or the elevated oxygen uptake by the coats.

2.2.4.2 Embryo dormancy

The coat alone is not responsible for dormancy since immature embryos are incapable of germinating even in conditions which would release dormancy - imposing mechanisms. If the period of immaturity extends beyond harvest ripeness this would result in apparent dormancy at harvest ripeness. However, embryos of both dormant and non - dormant wheat varieties have been found to reach maturity in similar lengths of time (Gordon, 1975; Verry, 1978).

Besides immaturity it has been found in dormant grains that the embryos themselves suffer an internal constraint or inadequacy which, when exacerbated by the presence of the coat, prevents them from germination (Black *et al.*, 1987). Embryos from dormant grains germinate markedly slower than those from non - dormant grains. Dormant grains may lack the ability to synthesize key germination enzymes and hence be unable to express certain genes. Synthesis of certain novel proteins may be suppressed in dormant embryos so that germination cannot be completed. Respiratory inhibitors (e.g. cyanide), high oxygen levels and several electron acceptors (e.g. nitrate and nitrite) all break dormancy. Dormancy is also characterized by a low activity of the pentose phosphate pathway (PPP) but there is insufficient evidence to support the theory that when dormancy is broken

by inhibitors of the Tri-carboxylic Acid (TCA) cycle or terminal oxidation and by electron acceptors there is an accompanying elevated operation of the pathway.

2.2.4.3 The role of hormones in dormancy

There is no evidence of a hormonal basis to the maintenance of dormancy although hormones may be involved in dormancy induction (Kaarsen and Groot, 1987). Also, dormancy does not result from the presence of an inhibitor or the lack of a promoter. Verry (1978) reported significant quantities of abscisic acid in developing wheat grains but these were not related to either dormancy or to maturation and dehydration of the grain. In addition, no significant differences were found in the embryo and endosperm ABA content between dormant and non - dormant mature wheat grains (Black *et al.*, 1987; Walker - Simmons, 1987) and no convincing evidence that dormant grains contain less GA than non - dormant grains was available in the literature. However, Hilhorst and Karssen, (1992), have since stated that ABA plays a key role during the setting of primary dormancy while GA is needed in the induction of germination. The transition from the dormant to the germinative states involves change in the amounts of ABA and GA. Interactions between ABA, gibberellins and cytokinins in seed dormancy and germination were used to explain the promoter - inhibitor hypothesis of dormancy control (Amen, 1968; Khan, 1975) where the balance between the level of substances reflects relative dormancy. However, there has not been consistent experimental evidence to support this hypothesis and it was challenged by Karssen and Groot (1987) who categorically stated that only ABA was involved in the onset of dormancy during seed development and GA played no role, while GA was absolutely required for germination. This suggested that regulation of dormancy was not by antagonistic action between these two hormones. Some aspects of dormancy may be explained as a GA inadequacy or an over - production while the converse is suggested for ABA (King, 1989).

2.2.4.4 Gibberellins (GAs)

Gibberellins are the active stimulants of seed germination by mobilizing food reserves essential for embryo growth. More than 52 GAs have been identified but not all are effective (Reeve and Crosier, 1974). GA_4 and GA_7 are highly active. Immature seeds contain many free gibberellins usually in large quantities. These free GAs often decrease as seeds mature while the "bound" forms increase. At

anthesis there is a peak of biologically active GAs in whole ears of wheat but it is not known if these are characteristic of the seed or the shoot. The GAs present in developing grains, however, are different from those found in vegetative tissues. Shoot GAs have different hydroxylation patterns from GAs of developing seed, the latter of which are often cultivar - specific (Ingram *et al.*, 1986). These GAs may be important in embryogenesis and endosperm formation. However, the major peak of biologically active GAs occurs around the time of maximum grain volume (Wheeler, 1972; Radley, 1976; Mounla, 1978; Slominski *et al.*, 1979). In barley, peaks of GA - like activity appear 9 and 21 days after pollination (Mounla and Michael,1973).

The level of GA in seeds undergoing stratification increases although the increase is not always coincident with the termination of dormancy. Further, the germination of seeds (different from the termination of dormancy) usually coincides with the increase in GA but it is not clear when this increase begins. In barley seeds, the increase precedes radicle protrusion while α - amylase increases appear to be post - germinative since they are preceded by RNA and protein syntheses which are enhanced by GA (Chen and Park, 1973; Gordon, 1975; Higgins *et al.*, 1976). GA production is under the control of the embryo itself. In barley, the embryo / scutellum complex is essential for 24 to 36 h if hydrolytic enzymes are to accumulate in the non - embryo half of the grain.

2.2.4.5 Abscisic acid (ABA)

ABA is an endogenous component of many seeds, whether they are dormant or not (Wareing and Saunders, 1971) and is an effective inhibitor of germination when supplied exogenously. ABA inhibits germination by acting upon the alpha - amy genes which code for α - amylase. Alpha - Amy 1 and Alpha - Amy 2 are multi - gene families located on the group 6 and group 7 chromosomes, respectively (Lazarus *et al.*, 1985). Their differences in isoenzyme expression are due to differences in mRNA accumulation patterns. In aleurone tissue, accumulation of alpha - Amy 1 transcripts occurs in parallel with other genes that are regulated by GA, while accumulation of alpha - Amy 2 transcripts is sustained for 36 h longer than with alpha - Amy 1 (Lazarus *et al.*, 1985; *Annual Report of the Plant Breeding Institute*, 1983:1984). GA_3 specifically controls transient expression from the alpha - Amy 2/54 promoter only in aleurone protoplasts (Huttly and Baulcombe, 1989). This expression is inhibited by the presence of ABA in the

incubation media. The alpha - amylase genes in wheat and barley show more differences between alpha - Amy 1 and alpha - Amy 2 than within each gene in both cereals, indicating that the divergence of wheat and barley was later than the divergence of alpha - Amy 1 and alpha - Amy 2 (*Annual Report of the Plant Breeding Institute*, 1983 :1984).

2.2.4.6 Auxins (IAA)

Large amounts of free auxin (indole acetic acid) accumulate at the time of maximal grain fresh weight then decline rapidly in both wheat (Radley, 1976a; Wheeler, 1972) and barley (Mounla *et al.*, 1980). Most of the auxin is located in the endosperm but it is also present at high concentration in young embryos. There is little information on the function of auxin in developing grain but it is unlikely to be rate-limiting for cell expansion or auxin-directed assimilate transport. The precursor for auxin synthesis is tryptophan.

2.2.4.7 Cytokinins (Cks)

There is a transient peak of biologically active Cks in developing wheat grains during the first 6 days after anthesis. However, there is a paucity of reliable quantitative data on the cytokinin complex of young wheat grains. It has been suggested that the source of Cks may be roots but polar Cks are produced *in situ* in detached, cultured ears. Cks are important in initiating reproductive 'sink activity' but the mechanism is unknown. A possible mechanism is by indirectly stimulating endosperm nuclear and cell division or by inducing rapid expansion of the ovule and surrounding ovary tissues. It is also likely that Cks may simply direct assimilates to the young developing grains.

2.2.5 Tissue sensitivity to hormones

The reaction of tissues to hormones is not only influenced by the absolute amount of the hormone but also by the sensitivity of the tissue to the hormone. The sensitivity of dormant seeds to ABA or GA is different from that of the non - dormant seed and it has been suggested that it is the relative change in this sensitivity that determines whether the seed is in a dormant or a germinative state (Walker-Simmons, 1987).

2.2.5.1 Gibberellic acid insensitivity

An important aspect of grain physiology affected by dormancy is the ability of the aleurone layer to produce α - amylase in response to GA. In *Avena fatua*, aleurone tissue from high dormant lines are poorly responsive to GA. Low temperature alters responsivity of aleurone tissue from certain dwarf wheats (Singh and Paleg, 1984) although it is doubtful that this effect is related to dormancy. GA_3 caused changes in lipid and phospholipid composition of isolated aleurone tissue previously exposed to 5 ^{0}C for 20 h (Surinder *et al.*, 1984). The time course for these changes was very similar to the low temperature - induced increase in GA sensitivity and changes in α - amylase production suggesting that GA receptor sites could be membrane - based lipids (Surinder *et al.*, 1984).

The *Rht* genes responsible for dwarfism (in particular, *Rht1* and *Rht3*) are known to be associated with gibberellic acid insensitivity (Gale, 1987; Borner *et al.*, 1987; Borner and Mettin, 1988). For example, two samples of a dwarf white wheat, Tordo, which is known to carry the genes *Gai3/Rht3* were found to react differently during germination. The sample which appeared to be sprouting susceptible produced higher levels of α - amylase during germination than the sprouting resistant sample (Mares *et al.*, 1983). GA insensitivity is characterized by high levels of endogenous GA_1 and GA_{20}, the immediate precursor of GA_1, in the early 13 - hydroxylation pathway of GA biosynthesis that operates in vegetative tissue. This seems to indicate that the *Rht* alleles cause non-utilization of available GA_1 possibly by production of an inhibitor or by directly reducing the efficiency of a putative GA receptor (Gale, 1989).

Five *Rht* alleles all of which can be identified by an insensitive seedling growth response to applied GA relative to that of tall *rht* genotypes are known in wheat. The most potent of these is *Rht*3. *Rht*1 and *Rht*2 were derived from the Japanese variety Norin 10, *Rht*3 from Tom Thumb, *Rht*10 from the Chinese variety Ai-bian-1 and *Rht*1S from the Japanese variety Saitama 27. The alleles are reported to have additive effects on both GA response and plant height and are all carried on the group 4 chromosomes (Borner and Mettin, 1988; Gale, 1989). The GA insensitivity derived from Saitama 27 was shown to be controlled by a single gene located on chromosome 4A and allelic to *Rht*1 and *Rht*3 (Worland and Petrovic, 1988; Worland and Law, 1986). Genotypes carrying the *Rht* alleles in the homozygous condition are higher yielding than their corresponding tall genotypes

(Gale *et al.*, 1986; Worland and Law, 1987; Worland and Petrovic, 1988; Sastry, 1988; Uddin and Marshall, 1989) although the yield advantage may be negated by a reduction in grain weight (Gale *et al.*, 1986). The mode of action of *Rht* alleles is not known but it is known that the cell elongation responses in extending stem internodes are more sensitive to *Rht* action than the α - amylase response of aleurone cells to endogenous or applied GA. Only *Rht3* has a marked effect and can reduce enzyme levels produced by distal half-grains to about 1% of those obtained with *rht* genotypes (Gale and Marshall, 1973, 1975; Fick and Qualset, 1975). *Rht1* and *Rht2* have a minor effect on aleurone response. Aleurone responses of *Rht1S* and *Rht10* have not been reported.

Aleurone GA - insensitivity is entirely endosperm mediated and is allele - dose dependent. *Rht3Rht3rht* heterozygotes are similar in response to *Rht3* homozygotes, and *Rht3rhtrht* heterozygotes are responsive (Gale and Marshall, 1975). Response due to *Rht1* and *Rht2* may not be additive (Upadhyay *et al.*, 1987) and it appears that some genotypes may have other genes for high α - amylase activity and low GA sensitivity. Mature and sprouted grain from lines homozygous for *Gai3/Rht3* displayed reduced α - amylase activity (Flintham and Gale, 1979) while tiller numbers, spikelet numbers, dormancy, nitrogen levels and sedimentation values were unaffected to any major extent. Loss in GA - dependency may result in grains with enhanced GA content or premature germination, while grains with low ABA content may germinate precociously (Qureshi *et al.*, 1989).

2.2.5.2 ABA insensitivity

Abscisic acid is required for the setting of dormancy but not for its maintenance. Walker - Simmons (1987) explained the different sprouting reactions between two white wheat cultivars, Brevor and Greer, as being due to different tissue sensitivity to ABA. In these cultivars, similar amounts of ABA were found in both the endosperm and embryo tissues, yet Brevor had considerably more resistance to sprouting than Greer.

2.2.5.3 Interactions between phytohormones

ABA inhibited translocation of 14-C sucrose from the nutrient solution into the developing ears unless cytokinins (Cks) were also applied (Borkovec and Prochazka, 1990). However, with or without Cks, ABA reduced grain dry weight.

In the early stages of wheat grain filling ABA content is positively correlated with filling rate while at later stages it is negatively correlated (Bai *et al.*, 1989). Gurbaksh - Singh et al. (1989) reported that IAA and ABA reached maximum levels in developing wheat grain at 30 days after anthesis (DAA). However, Verry (1978) found genotypic variation in ABA content peak occurrence and a general tendency for these maxima to occur during the second or third week post anthesis around the time of the colour change or at about harvest-ripeness. ABA inhibits GA_3 - induced acid phosphate activity. *De novo* synthesis of acid phosphatase has been confirmed in GA_3 - treated half seeds (Saluja *et al.*, 1989).

2.2.5.4 Role of grain coat colour in dormancy

Both white - and red - grained wheat varieties are affected by pre - harvest sprouting damage although the frequency of occurrence is generally greater in the white varieties than it is in the red varieties. In the red - grained wheats there exists a variable level of resistance to pre - harvest sprouting damage (McEwan, 1979; Gordon, 1983). The work of Nilsson - Ehle (1914) was amongst the first to provide evidence for a possible association between red colour and prolonged dormancy in wheat.

Many of the varieties with exceptionally good dormancy e.g. cultivars RL 4137, Park, Thatcher and Hilgendorf 61, carry red - gene *(R)* alleles on all three group 3 chromosomes (McEwan, 1979; Gale, 1989). Heterozygotes (*R/r*) display only partial dominance for dormancy (Ibrahim, 1967). Inter - allelic additivity was demonstrated in comparisons of one - and two - allele and two - and three - allele genotypes (Freed, 1972; Freed *et al.*, 1976; Reitan, 1979). Gordon (1975) did an extensive survey on 97 genotypes and found that the red graincoat colour was poorly correlated with seed dormancy and there appeared to be no indication of a close association between the intensity of red graincoat colour and dosage of red grain gene (McEwan, 1979). Intensity of red seed coat colour is not therefore a useful selection criterion for dormancy.

The observed variation in sprouting resistance in red - grained wheats is not simply due to differences in gene dosage levels (Gordon, 1979; McEwan, 1979) but is likely to be due to either an interaction of the factors for red grain colour with some other component of the genetical background or independent "dormancy" genes closely linked to the *R* loci. It is now accepted, however, that the cause of this

association is pleiotropy rather than close genetic linkage of the genes for red colour with dormancy genes (Flintham and Gale, 1988). Certain factors of dormancy are not linked or pleiotropic with red pericarp and can hence be transferred to white - grained wheat cultivars, e.g. the white - grained wheat line, Losprout, released in 1984 in Canada, was derived from a cross between RL 4137 (red, spring) and 7722 (white, spring) (DePauw *et al.*, 1985). Losprout has high resistance to sprouting damage, greater than that of the moderately resistant white - grained Kenya 321.BT.1 and comparable to that of Pitic 62, Glenlea and Neepawa (all red - grained) and significantly less than that of RL 4137. RL 4137 has a genetic mechanism for sprouting resistance associated with red genes and one or more mechanisms not associated with grain colour.

2.2.5.5 Inheritance of red colour in wheat

Red grain colour in wheat is simply inherited. It is dominant over white colour and is determined by three independent, homoeologous loci on the long arms of the group 3 chromosomes, i.e. 3AL, 3BL and 3DL (Metzger and Silbaugh, 1970). Segregation for red colour occurs on the F2 plant rather than on the F1 plant.

2.2.5.5.1 Grain coat pigments

Although the grain coat pigments in wheat are yet to be fully identified they are known to be flavanoid. Most red grained wheats contain the reddish - brown flavanoid polymer, *phlobaphene* (Miyamoto and Everson, 1958; Pomeranz, 1971; Gordon, 1975). Phlobaphenes, condensed tannins, flavolans and procyanidins are all condensation products of flavan-3-ols, flavan-3, 4-diols and possibly anthocyanins (Ribereau-Gayon, 1972). In whole grain, differences in flavanol levels throughout the ontogenesis of red - and white - grained genotypes were not associated with graincoat colour (Gordon, 1975; Cross, 1977). Gale and Flavell (1971) found, in the variety Hope, that control of anthocyanin synthesis was exercised by genes on the homoeologous chromosomes 7A and 7B and that these genes displayed only partial dominance. Other dormancy associations that have been discovered include responses to catechin tannins, the presence of and response to various inhibitors, anthesis date, vernalization response and winter hardiness.

2.2.5.6 The flavanoids

1. The flavanoids are classified into four groups:
2. Benzoic acids, cinnamic acids and coumarins
3. Flavones, flavanols and related compounds
4. Chalcones, dihydrochalcones and aurones
5. Anthocyanins

They are characterized by having the $C_6 - C_3 - C_6$ structure in common in which the two benzene rings are linked by a C_3 group which is different in the different flavanoids (Ribereau - Gayon, 1972). The two benzene rings have different biosynthetic origins and differ in the amount of hydroxylation. The A ring (left - hand ring) is either *meta* - dihydroxylated (resorcinol type) or *meta* - trihydroxylated (phloroglucinol type), except for the chalcones where one of the hydroxyls of the A ring is combined in an oxygen heterocycle of five or six atoms. The B ring is either monohydroxylated, *ortho* - dihydroxylated or *vic* - trihydroxylated. Ring A is formed by the condensation of three molecules of acetic acid, whilst ring B is derived from sugars by the shikimic route (Ribereau - Gayon, 1972). Flavan-3, 4-diols may be derived directly from dihydroflavonone through a reduction process. Dehydroxylation of flavan-3, 4-diol produces a key intermediate, flav-3-en-3-ol, which is unsaturated between C_3 and C_4 carbons. Reduction of flav-3-en-3-ol gives flavan-3-ol and oxidation yields an anthocyanidin (Haslam, 1977). Much of the biochemistry of the flavanoids is still unknown. However, the following discussion establishes what is known about the structure and chemistry of the flavan-3-ols and the condensed tannins.

2.2.5.6.1 Structure of the flavan-3-ols (catechins)

The flavan-3-ols are related to the anthocyanins and flavones but they do not generally exist free in nature as the glycosides. The most common members differ only in the number of hydroxyl groups (1, 2 or 3) in the phenyl B ring and these groups are never methylated. The term 'catechin'refers specifically to the flavan-3-ol which has two hydroxyl groups in the side ring. All the compounds have two asymmetric carbon atoms (C - 2 and C - 3) thus giving four optical isomers; in the case of the catechin series these configurations are (+)-catechin, (-)-catechin, (+)-epicatechin and (-)-epicatechin. (+)-catechin and (-)-epicatechin are the common occurring forms. The central heterocyclic ring, being saturated, is not planar; the

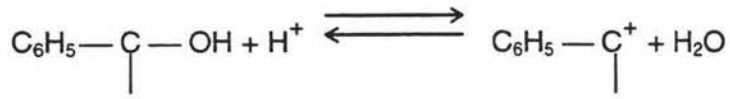
oxygen atom and C - 4 are in the plane of the A - ring but C - 2 and C - 3 lie either above or below the ring. The infra - red spectra show that there is strong hydrogen bonding between the hydroxyl group at C - 3 and the heterocyclic oxygen in both (+)-catechin and (-)-epicatechin.

The main difference between the flavan-3-ols and the flavan-3, 4-diols is the fact that on heating in acid solution the catechins give yellow-brown insoluble products of high molecular weight, whereas flavan-3, 4-diols give, besides these phlobaphenes, some anthocyanidin.

2.2.5.6.2 Structure of the condensed tannins

Condensed tannins from plants are a mixture of several polymers which are condensation products of the flavans. These polymers have been designated flavolans in which the flavan-3, 4-diols play a more important role than the flavan-3-ols. The tannins have a molecular weight between 500 and 3000; that of the flavans is of the order 250 to 300 which means that the flavolans contain 2 to 10 monomers and could be called oligomers. Polymerization plays an important role in determining the properties of the tannins, which vary depending on whether dimers, trimers or tetramers are present.

In the polymerization of flavan-3-ols, there is an increase in the number of hydroxyl groups on polymerization presumably due to the addition of water to the O—C(2) bond of the heterocyclic ring thus giving one phenolic and one benzylic alcoholic hydroxyl group extra. The latter readily forms a carbonium ion (with a reduced electron density) in acid milieu by the reaction:



This ion can then react with a strongly nucleophilic centre (one having an excess of electrons) such as is found at C-6 or C-8 of the flavan A ring of a second molecule of flavan-3-ol giving a dimer. Such a mechanism could involve a large number of molecules to yield a high molecular weight polymer.

Flavan-3, 4-diols polymerize more readily than the corresponding flavan-3-ols. This could be due to the fact that the hydroxyl group at C-4 is also benzylic in nature and can react with the nucleophilic centres of another hydroxyflavan. Condensation could proceed with a further flavan molecule through the free OH^-

group at C-4 in one of the ring structures. By treatment with acid, a dimer formed by condensation of two flavan molecules gives carbonium ions and rapidly yields insoluble high molecular weight compounds, the phlobaphenes. Hathway (1962) suggested that condensation of flavans was due to their oxidation catalyzed by tyrosinase.

In the case of flavan-3-ols, an *ortho*-quinone is obtained from ring-B which gives the ring an electrophilic character. Thus, this can react with a nucleophilic A ring of a second flavanol giving a dimer in a 'head to tail' manner and condensation progresses in this way. Gallocatechin will give mainly a 'tail to tail' linkage and give ellagic acid (two moles of gallic acid).

However, it is highly likely that in any one tannin, several different types of linkage exist simultaneously, perhaps involving several different monomers. Moreover, catechins may form dimers with other types of compound which could be incorporated into the tannin, e.g. flavylium salts especially those lacking an hydroxyl group at C-3.

The complex problem of structure of condensed tannins is important because it detremines their properties which are linked closely to the size of the flavolan. If the degree of polymerization could be determined by analytical means, it seems probable that one might interpret the transformations which these tannins undergo in plants and apply such knowledge e.g. in calculating the size and efficiency of the oxygen trap represented by these tannins and in plant and food chemistry.

2.2.6 Germination

Non - dormant seeds will germinate under suitable conditions of adequate moisture and temperature. Germination may be defined as comprising the processes occurring in the seed in the period between rehydration of the dry seed and embryo growth. These processes include physiological and biochemical changes under the influence of phytohormones, the environment and the genetic make - up of the grain.

Thus, the process of germination is preceded by hydration and reorganisation of the dry seed. A lag phase often occurs before the change to germinative metabolism and embryo growth. RNA and protein synthesis start soon after imbibition (Marcus *et al.*, 1966) and will continue at an increasing pace throughout

germination. Changes in RNA occur following reassembly of the pre - existing mRNA, and ribosomes aggregate to form polysomes. Synthesis of new mRNA and ribosomes may occur after 12 h or more. Membrane composition may change with the appearance of new endoplasmic reticulum but its synthesis may not occur until 24 h (Jones, 1980). Respiration increased rapidly soon after imbibition in wild oat (Chen and Varner, 1969), but new mitochondria appeared much later (Bewley and Black, 1978).

GA is the key hormone involved in the mobilization of reserves essential for embryo growth and is under the control of the embryo itself. In barley the embryo / scutellum complex is essential for 24 to 36 h if hydrolytic enzymes are to accumulate in the non - embryo half of the grain.

2.2.6.1 Enzyme activity

Important changes that occur in the grain as a result of preharvest sprouting include the reduction in grain bulk weight and the loss of quality of the milled flour. These changes are brought about by the action of enzymes that catalyse or facilitate starch hydrolysis in the endosperm. Following imbibition gibberellic acid is produced by the embryo and the scutellum. This plant growth hormone is responsible for activating endogenous enzyme systems in the early stages leading to germination and also stimulates the synthesis of new enzyme molecules by the aleurone layer of the endosperm.

Different enzymes or groups of enzyme systems increase in temporal sequence during the sprouting reaction. Active proteolytic enzymes, starch phosphorylases and hemicellulases appear before the amylases. The lag phase between the appearance of the first three groups of enzymes and α - amylase was found to be about 20 hours (Gordon, 1977).

Proteases catalyse the breakdown of the protein matrix surrounding starch granules and expose these granules to attack by α - amylase. The latter hydrolyses starch molecules to oligosaccharides and therefore provides the substrates for β - amylase. Beta - Amylase is an exo - enzyme which requires the action of an endo - enzyme, in this case, α - amylase, to cleave the starch molecule into smaller segments before it can have significant effect on starch degradation. Proteases, phosphorylases, hemicellulases occur in large amounts in the ungerminated grain but they do not normally cause damage to the grain

because they occur in bound or inactive forms until activated by hydrolysis. Release of the active forms takes place during the initial phases of imbibition and is accelerated by the action of gibberellic acid which also stimulates *de novo* synthesis of many enzymes. Abscisic acid (ABA) inhibits the action of GA and is known to promote dormancy. The cytokinins play a stimulatory role by blocking the action of ABA and facilitating the action of GA.

2.2.6.2 - Amylase

The total natural cereal α - amylase activity is made up of a basal level inherent in the dormant grain and the α - amylase activity that develops in the sprouting grain in wet conditions (Buchanan and Nicholas, 1979). There are varietal differences in the proportion and occurrence of these two types of α - amylase.

Basal α - amylase can be measured very rapidly by means of the Hagberg Falling Number method at the milling stage. However, bakers tend to express α - amylase activity in terms of Farrand Units using the Farrand method which measures total α - amylase activity, that is, natural cereal α - amylase activity plus any amylase supplements of a fungal origin (e.g. from the metabolism of bakers' yeast (*Saccharomyces cerevisiae*). The consequence of a high level of α - amylase in sprouted grain is an increase in operating costs in the bakery. This is because the physico-chemical properties of bread crumb are altered progressively as the activity of α - amylase increases: compressibility and stickiness increase, resilience and density decrease, and high molecular weight dextrins increase. In cold water, starch granules form a colloidal suspension which has little viscosity but the products of starch dextrinisation give dough its viscoelasticity. These changes affect bread sliceability since sticky crumbs provide high resistance to the movement of the high speed slicing blades which therefore require regular and frequent lubrication if breakages are to be minimised (Buchanan and Nicholas, 1979; Gordon, 1979; Orth *et al.*, 1987). Amylase resistant starch is subject to less damage at moderate sprouting and also gives desirable milling properties (Mackey, 1975). However, an amylase - deficient flour is undesirable because it prevents production of sugars on which the yeast acts during the fermentation and proof processes. Carbon dioxide gas, a by - product of the fermentation of sugar by yeast, is needed to raise bread.

2.2.6.3 Environmental influence

The environment and the genetic makeup of the wheat grain and their interaction have a large bearing on sprouting damage. In a study carried, over 16 years, on the effects of temperature, rainfall and global radiation on seed dormancy at different stages of seed development in barley, wheat and oats, environmental effects accounted for 65.3 % of the variation in seed dormancy (Strand, 1983). Significant differences were found between cultivar, year, harvest time and germination temperature. Seed dormancy reached a maximum at different stages of maturity during different years being 10 days after yellow ripeness in an average year.

2.2.6.4 Temperature

Germinability of harvest - mature wheat grain shows a marked temperature dependence. For example, the optimum temperature for complete germination varied from 20 ^{o}C for the non - dormant cultivar, Timgalen, to 10 ^{o}C for the strongly dormant cv. RL 4137 (Mares, 1984). Further, Mares (1984) found that germinability was increased by pre - treating imbibing grains at 5, 10 or 15 ^{o}C and the temperature for maximum germinability decreased with increasing grain dormancy level.

Developing seed harvested from the second to the seventh week post - anthesis showed a significantly higher rate of loss of sprouting tolerance when germinated at 17 ^{o}C compared with 25 ^{o}C (Plett and Larter, 1986) while genotypic differences were more marked in the 25 ^{o}C regime. Daily temperature fluctuations before physiological maturity and precipitation after physiological maturity significantly affected alpha - amylase activity in grain (Nielsen *et al.*, 1984). Sprouting was increased by large daily temperature fluctuations, low daily temperatures and high precipitation.

In controlled environment studies on the effects of temperature on wheat germination with water uptake restricted by resistances outside the seed Woobury and Wiebe (1983) found that at low temperatures the lag in germination or emergence increased but growth rate was unaffected. The amount of water required for germination was decreased by low temperature when water supply was interrupted but was increased if water supply was continuous.

2.2.6.5 Rain

During seed maturation, a crack is formed in the brush region of the pericarp, allowing rapid movement of water beneath the pericarp in the dorsal surface. This may transfer inhibitors to the embryo (Woodbury and Wiebe, 1983) and cause a block to germination. Differences in resistance to pre - harvest sprouting between cultivars increased when harvesting was delayed after maturity by 15 - 20 days (Nettevich *et al.*, 1986) and alpha - amylase activity in grain that had received rainfall during the grain filling and maturation period depended on genotype and growing conditions. Nielsen *et al.* (1984) found seven white wheat lines (*Triticum aestivum*) to be uniformly susceptible to pre - harvest sprouting after subjecting mature spikes to simulated rain while a red - grained cultivar was more resistant. However, response to pre - harvest rain, in a controlled environment rain simulation system, of dwarf and semi - dwarf wheat lines isogenic for the dwarfing genes *rht*, *Rht1*, *Rht2* and *Rht1* + *Rht2*, varied widely with both genotype and environment and no relationship between the response and the particular dwarfing genes involved was found (Mares *et al.*, 1983).

2.2.6.6 Nitrogen - based fertilizer

Protein content is positively correlated with sprouting (Morris and Paulsen, 1985). High levels of N fertilizer application were found to increase rain - induced pre - harvest sprouting in genotypes with moderate or low levels of resistance but did not affect preharvest sprouting of genotypes with strong resistance and all genotypes in areas where conditions were not conducive to preharvest sprouting (Morris and Paulsen, 1985). Differences in nitrogen nutrition have been shown to alter not only grain protein but also to increase amylase synthesis in wheat and diastatic activity in barley (King, 1983).

2.2.7 Viability

Sprouting affects both quality and germinative capacity of the grain. It was demonstrated that grain pre - sprouted until germination was just still invisible produced higher shoot and root dry matter (DM) and lower electrolyte leakage than grain pre - sprouted until germination was just visible with the coleoptiles about to penetrate the seed coat (Falkenstein and Steiner, 1985). Rain - affected seed lots exhibit electrolyte leaching due to increased quantities of water soluble compounds rather than to membrane deterioration and yield of crop raised from rain - affected seed was significantly lower than that from unaffected seed in spite

of compensation made for low germination percentage at sowing (Agrawal and Dadlani, 1984).

2.3 Breeding for preharvest sprouting tolerance

Many uses of wheat require white seed - coat as a desirable quality attribute. However, the majority of white - grained wheat varieties possess little or no tolerance to preharvest sprouting and quality is often compromised in these varieties especially under cool and wet ripening conditions. An important objective in wheat breeding therefore is the identification and selection of lines having sufficient dormancy to pass through the physiological window between the early ripening stage and harvest - ripeness without sprouting; at least 10 days of dormancy from the time harvest - ripeness is reached is considered adequate protection. Harvest - ripeness (HR) is defined differently in different climatic region, e.g. in temperate regions with cool ripening seasons HR is defined as the stage when grain moisture content has reached 18 % of grain dry weight, whereas in warmer ripening climates (such as the northern parts of Australia) HR may be defined as the stage when grain contains 12 % or less moisture based on grain dry weight.

Resistance to sprouting damage is a complex trait with multigenic inheritance and relatively low heritability (Flintham and Gale, 1989). Germplasm for resistance to preharvest sprouting is widely available in the red - grained varieties but only few white - grained wheats have sufficient dormancy to warrant the cost of a breeding program. Increasing knowledge on dormancy and its inheritance appears to make dormancy genes easier to handle than other factors (Derera, 1989) and it is not surprising that much breeding effort into pre - harvest sprouting resistance is based on dormancy factors.

The strategy employed in most breeding programmes into pre - harvest sprouting is to transfer dormancy factors through crossing a resistant red - grained parent and a white - grained cultivar having the desired agronomic ideotype. A series of backcrosses are then carried using the white variety as the recurrent parent. However, this is not an easy task because of the pleiotroic effects that exist between the dormancy genes and red seedcoat colour.

Recently, McCaig and DePauw (1992) have shown that white - grained cultivars having high levels of preharvest sprouting tolerance can be successfully bred. They have also pointed out that the observed differences in sprouting tolerance between wheat classes probably reflect current cultivars and applied selection pressure during cultivar development rather than inherent differences between classes. This observation is confirmed by the successful transfer of dormancy and preharvest sprouting resistance from RL 4137 (a red, spring wheat) to a white wheat using line 7722 as the white - seeded spring parent. Release of 'Losprout' (a white - grained, spring wheat with reasonable tolerance to preharvest sprouting) in 1984 (DePauw *et al.*, 1984) revealed that certain dormancy factors are not linked or pleiotropic with red pericarp and can therefore be transferred to white - seeded wheat cultivars (Derera, 1989).

Other factors that may be exploited in breeding programmes include the rate of water uptake in the caryopsis, starch resistance to amylases causing degradation, reduced rate of amylase production, and the physical characteristics of the spike (Derera, 1982).

Chapter 3

MATERIALS AND METHODS

3. INTRODUCTION

The review discussed in the previous chapter identified areas where there is a paucity of information regarding wheat physiology and genetics with respect to germinability and graincoat colour, e.g. the significance of the triploid nature of endosperm tissue in relation to dormancy and the specific relationships between different metabolic activities during development of the wheat grain are unknown. Clearly, if the problem of preharvest sprouting damage is to be controlled adequately greater effort must be directed not only towards the accumulation but also the understanding and correct utilization of knowledge in many such areas. Many of the relationships between developmental events are complex: therefore, experimental designs must be carefully chosen in order to gain optimum information from the research.

In this study, attributes related to harvest ripeness, grain growth, germinability, graincoat colour and flavanol concentration, the concentration of abscisic acid in developing grains, base α-amylase concentration and grain dormancy were measured, at intervals, from a few days after pollination until after grain maturity. The measured variables were fitted to appropriate functions using numbers of days from pollination as the independent variable.

The purpose of this study was to examine the relationship between graincoat colour development, the concentration of flavanols in developing grain, abscisic acid content and dormancy in wheat cultivars representing a diverse genetic background. Ten wheat cultivars that differed in colour (5 red-grained and 5 white-grained), in height (dwarf, semi-dwarf and tall), in hormone sensitivity (gibberellic acid insensitivity and abscisic acid sensitivity), and dormancy were used in the study.

The specific objectives were:

1. to investigate the relationship between graincoat colour development, the timing of peak ABA activity and the level of dormancy in these wheat genotypes.
2. to establish the relationship between the grain development characters studied in these diverse genotypes, and to intepret these relationships in terms of sprouting damage.
3. to estimate genetic properties of these sprouting attributes, e.g. heritability and genetic correlation.
4. to investigate the levels of α - amylase in relation to dormancy due to gibberellic acid insensitivity.

3.1 Plant material

Ten wheat cultivars (*Triticum aestivum* L.) differing in colour (*R* gene), height (*Rht3*/*Gai3* gene), and putative resistance to preharvest sprouting were investigated in order to study the relationship between potential to sprouting and relevant physiological characteristics. The experiment was conducted at the Massey University Crop and Pasture Blocks from October 1994 to April 1995, this being the usual growing period for spring wheat in this region (Manawatu District, New Zealand). The ten cultivars constituted the treatments which were arranged into a randomised complete block experiment with four blocks. An experimental unit consisted of two 3 m rows 0.5 m apart with plants in each row 20 cm apart. All cultivars had a spring habit with the exception of Brevor, which has 6-8 weeks requirement for vernalization (Walker-Simmonds, 1987). This was supplied artificially (*see* Section 3.1).

3.2 Growing conditions

Plants were grown under natural conditions without irrigation or fertilization although selective fertilization (with ten grams of KNO_3) was done in Block 1 for two cultivars, Tordo and Isis, to encourage tillering and flowering because emergence had been poor in these cultivars. On two occassions (December 1994 and January 1995) all plants were irrigated when the soil moisture content appeared low and the ground was cracking. On both occassions care was taken to water the plants *below* the canopy level in order to avoid water

falling on the developing ears. Weeds were controlled by spraying with Glean® at 20 g /ha and Bromoyinl® at 500 ml /ha. This controlled the broad-leaved weeds. Rust disease and aphids were both controlled by spraying with Tilt® at 500 ml /ha. The active ingredient in Tilt® is propiconazole (at 250 g / l).

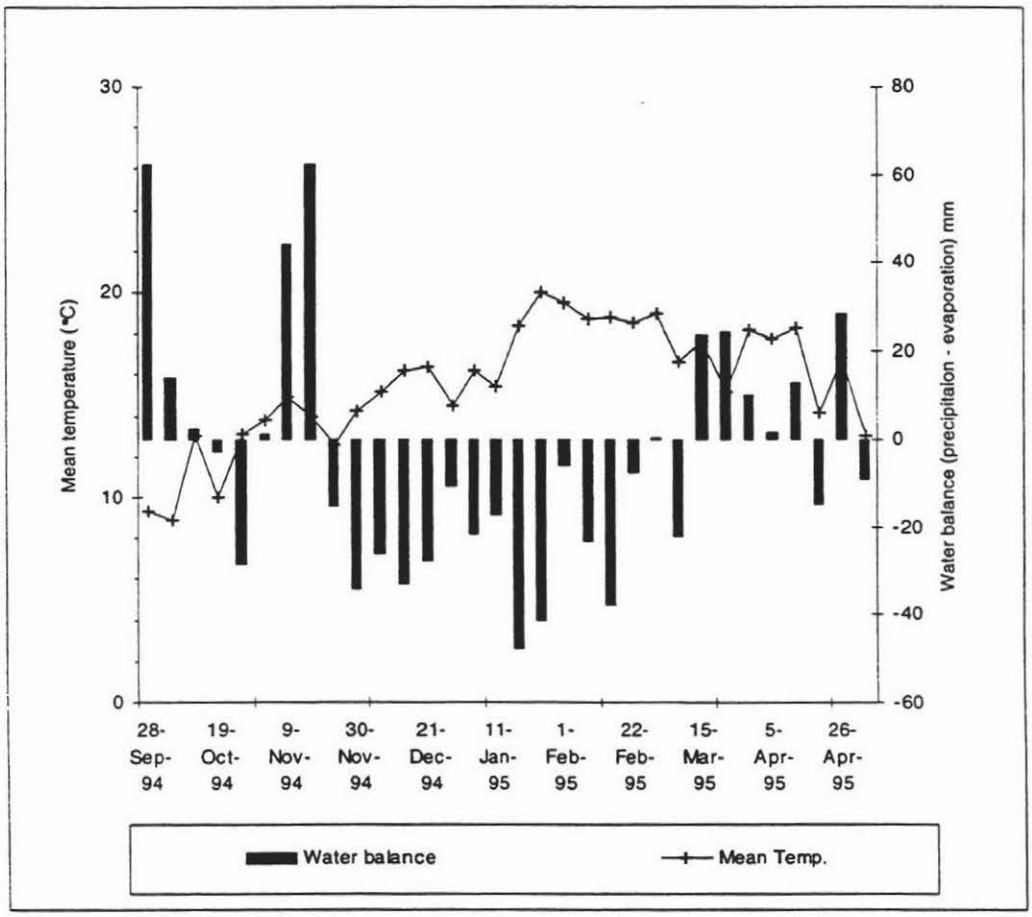
3.2.1 The weather

The mean weekly temperature and water balance during the growing season are shown in Figure 1 below. The data for Figure 3.1 were obtained from the daily climatological observations at 0900 h collected by the Grasslands Division of Agricultural Research Station No. E 05363 on Latitude 40 23^0 South and Longitude 175 37^0 East. This station is situated aproximately 2 km to the north of the experimental site. The mean temperature ranged from 10 ^{0}C in October, 1994 (i.e. at sowing time) to 20 ^{0}C in February, 1995 (i.e. ripening time) before declining gradually to about 11 ^{0}C in April. There were no serious fluctuations between minimum and maximum temperatures over the entire period of the experiment. Temperature conditions were thus suitable for the normal growth of wheat. However, the water balance as estimated from the difference between precipitation and evaporation (i.e. ignoring water losses due to drainage and runoff and water gains due to irrigation) was negative for most of the time during vegetative growth. This may have contributed to the formation of deep cracks in the top soil that put the root system at risk from becoming torn off the plants. Since the soil water potential was not measured in order to assess the extent of the associated risk from dehydration, the decision was made for the controlled irrigation in December, 1994, since frequent inspections of the crop showed that the crop was not at any great risk from dehydration.

3.3 Wheat cultivars in the study

As mentioned in section 2, the ten genotypes in the study differed primarily in graincoat colour (which is loosely associated with dormancy) and height (associated with sensitivity to gibberellic acid) and putative resistance to preharvest sprouting.

Figure 3.1. The mean temperature and water balance during the growing season (October, 1994 - May, 1995).



White-grained cultivars

1. Gamenya

A medium height wheat of Australian origin with no dormancy. *Rht* genes have not been reported in Gamenya. Registered in 1960 (Ferns *et al.*, 1975).

[1] *Pedigree*: Gabo*6 | Mentana (W1124) ‖ Gabo*2 | Kenya117A (W1347)

2. Brevor

medium height wheat with moderate dormancy which originates in the U.S.A. It possesses the *Rht1* gene for height reduction and low GA sensitivity. Brevor is insensitive to abscisic acid (Walker-Simmons, 1987) and has genes for high α-amylase production. It has a moderate vernalization requirement.

3. Isis

A tall, non - dormant wheat of Australian origin with no *Rht* genes.
Pedigree: Celebration | Macquarie | 3 | Macquarie ‖ Warigo | Ghurka.
Registered in 1969 (Ferns et al., 1975).
Isis was suspected to consist of two isotypes because of observed genetic variation during vegetative growth. One isotype remained prostrate during the entire vegetative period showing evidence of some vernalization requirement. The other isotype was upright from the beginning. There were three "bursts" of flowering in this variety at intervals of about three days, the earliest flowering being the upright isotype. **Sampling was only from the upright variety.**

4. **Tordo**

Tordo possesses the *Gai/Rht3* gene for gibberellin insensitivity and dwarfness (Mares *et al.*, 1983; Gale, 1982). It also has genes for high α-amylase. It is non-dormant and is of Mexican origin.

5. **Kenya 321**

A tall cultivar. It is one of the few white-grained wheats with low to moderate dormancy. Dormancy of this variety may involve inhibitor systems (Gale, 1989; Depauw and McCaig, 1989).

Red-grained cultivars

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[1] The notation used follows that used by L. H. Purdy *et al.* (1968) Crop Sc. 8, 405. An initial cross is indicated by a slash (/) and later crosses appear as //, /3/, /4/, etc. A backcross is shown by an asterisk (*) preceded by a number indicating how many times the recurring parent was used.

1. **Sonora 64 A**

   A semi-dwarf wheat with moderate dormancy. Sonora 64 A possesses one gene for red colour (*R1*) and the *RhtX* gene for reduced height. It is of Mexican origin.

2. **Karamu**

   A semi-dwarf wheat with considerable dormancy. Karamu has one gene for red colour (*R1*) and the *Rht1* gene for reduced height. Karamu is the New Zealand selection of the Australian breeding line, WW15.

   Pedigree: Lerma Rojo | Norin 10-Brevor ‖ Yaktana 54 | Norin 10-Brevor |3* Andes.

3. **La Prevision**

   A tall, highly dormant wheat which possesses two genes for red colour (*R1* and *R2*). It is of Mexican origin. There is no evidence of gibberellin insensitivity.

4. **Thatcher**

   A tall, dormant having three genes for red colour (*R1*, *R2*, and *R3*); it is of Canadian origin. There is no evidence of gibberellin insensitivity in this wheat.

5. **Hilgendorf 61.**

   This is a mid-tall, dormant wheat with three red genes for red colour (*R1*, *R2* and *R3*). It is highly sprouting resistant. Hilgendorf 61 is of New Zealand origin.

   Pedigree: S 1894 |7* Hilgendorf.

   The recurrent parent, Hilgendorf, was selected from Cross 7 | Tainui released in 1947. Hilgendorf 61 shows transgressive segregation for red graincoat colour and for resistance (McEwan, 1979). One parent, Tainui, has only a moderate level of sprouting resistance while the other parent, Cross 7, is white-grained and sprouting susceptible. Hilgendorf 61 is highly sprouting resistant.

With the exception of Brevor and Karamu (Karamu has Brevor in its pedigree), these cultivars are genetically independent.

3.4 Vernalization of Brevor seedlings

One hundred and fifty Brevor grains were placed on Whatman 1 filter paper, moistened with 2 ml 0.2% w/v KNO_3 solution, in a glass petri dish and allowed to imbibe to 50% of usual uptake (8-10 h). Germination was for 2-4 days at 5-10° C. Germinated grains were transferred to 5 cm^2 peat pots filled with a seedling soil mixture (*see* Appendix 3.2). The pots were placed in trays and moved to a bench in the 5° C cold room under two photosynthetic lights ("Eye" Self-Ballasted Mercury Lamp, white, 240 V SB/E28, 275 W, Japan). The light and dark periods were 8 h and 16 h, respectively. The plants were kept moist under these conditions for 8 weeks before being transferred to the field when they were approximately at the same morphological age as the plants sown directly in the field.

3.5 Tagging at flowering

The age of developing grain was to be determined from tagged wheat ears of known age. Ears from the primary and the second tillers were tagged, and from the third tiller where a shortage of material was anticipated. Ears were tagged at anthesis this being defined as the time when one-half of the plants in each plot had one or two anthers just extruding. This was taken to indicate the time at which pollination occurred. Tags were labeled with date, block number and treatment number. Eighty to 100 ears from the first and second tillers were tagged in each plot within a 2-4 day period. This meant that the age of every experimental ear was known.

3.6 Sampling procedures

Five to six tagged spikes were harvested at random from each plot at each sampling time from about 7 days after anthesis over the whole period of grain development. Successive sampling was at intervals of between 5 and 10 days depending on the rate of development of the grains; when it was noticed that events were occurring at a faster rate (e.g. from the moisture and germination tests) then the period between successive samples was reduced, and *vice versa*. Samples were immediately placed in sealed plastic containers after removal from the mother plant in the field. This reduced the risk of excessive moisture loss during transfer to the laboratory.

In the laboratory, grain was hand-plucked from the middle third of the ear and only the the two basal grains were used. This was necessary because of the differential development of grains within the ear. Size and maturity traits vary with position of the grain in the spikelet as well as along the spike (Rawson and Evans, 1970). Hand-plucking was necessary in order to prevent damage to the pericarp. Size and maturity differences between the grains were kept to a minimum by using only the basal and second grains of each spikelet (Rawson and Evans, 1970; Evans *et al.*, 1972); although, in the mature and drier ears, handling was difficult because grains tended to fall from spikelets. This was particularly the case with Sonora 64 A and Karamu. Free grains were temporarily stored in lidded and labeled petri dishes which were placed in a humidifier to prevent moisture loss. Each sample of grains was sub-divided either for immediate analysis or for storage in glass vials until required for analysis later. Ears that reached anthesis on the same date were used in the same sample. The age of a grain sample was expressed as days after pollination (DAP). The moisture, colour and germination tests were done immediately. After drying in the oven, the samples used in the measurement for grain moisture content were stored in labeled waterproof bags at 5 ^{0}C for future use in the α-amylase assays. Samples for flavanols and abscisic acid were quickly frozen using liquid nitrogen and stored at - 70 ^{0}C.

3.7 Characters measured

The following grain development characters were measured at each sampling time from about 7 days after anthesis until the grains reached maturity and beyond.

3.7.1 Grain moisture content, grain dry weight and harvest ripeness

The mass of twenty fresh grains was measured soon after plucking from the spike and dried in an oven at 60 ^{0}C for 48 hours, cooled in a dessicator over silica gel and reweighed to give the dry weight. This temperature regime was used because the dried sample would be used for measurement of α-amylase known which is known to be stable at this temperature (Barnes and Blakeney, 1974). Above this temperature, α-amylase becomes unstable and may become denatured.

The percentage grain moisture content of the grains was calculated on the basis of the fresh weight, as is customary in most seed studies (ISTA 1993), as:

$$\text{Moisture (\%)} = \frac{100 \times (FW - DW)}{FW}$$

where, FW = fresh weight (g)
DW = Dry weight (g)

The grain dry weight was expressed in mg per grain by dividing the total grain dry weight in the sample by the number of grains in the sample.

Harvest ripeness was defined as the time when the grain moisture content decreased to 12.5 % (Gordon, 1975). This was estimated by interpolation of the fitted function for grain moisture content (details later).

3.7.2 Normal germination, special germination and embryo maturity

Two samples of twenty grains each were subjected to one of two germination tests: normal or special. In the normal germination test, the grains were placed, crease down, on a substrate of Whatman 1 filter paper moistened with 2 ml distilled water in a 9 cm glass Petri dish. Germination was carried out at 20 ^{0}C in the dark (Anon., 1966) for 5 days. The special germination test (Gordon, 1975) was designed to break dormancy and therefore measured embryo maturity. Grains were placed crease down, as for normal germination, on a substrate of Whatman 1 filter paper in a 9 cm glass Petri dish. The test was read after 5 days. Four recognized dormancy - breaking mechanisms were applied in this test (ISTA, 1993). The test therefore could be used to determine embryo maturity as the mean ontogenic ability to germinate in a whole grain system. The dormancy - breaking mechanisms applied were,

1. 0.2 % w/v KNO_3 in the imbibing fluid
2. alternating light (16 hours) and dark (8 hours) periods, and
3. alternating low (15 ^{0}C) and high (20 ^{0}C) temperatures.
4. 10^{-4} M GA_3 in the imbibing fluid

The first three mechanisms had been used by Gordon (1975) and Verry (1978) in previous work. Inclusion of the fourth mechanism, involving the plant growth hormone, GA_3, in conjunction with the other three mechanisms is a modification of the original recipe of Gordon (1975) for this test. Gibberellin A_3 (Lot No. 49321, Cat. No. 16069, United States Biochemical Corporation, Cleveland, Ohio 44128) was used in this test. The four mechanisms together

should form a powerful means of breaking dormancy. The special germination test was carried out in an automatic germinator (Contherm Scientific Germinator M190 PHS with a precision logic control system). The change over from light to dark periods in this germinator is sudden but the temperature change is more gradual and is complete in approximately 1 hour.

Lack of response in the special germination test was regarded as due either to embryonic immaturity or dead caryopses. Gordon (1975) carried out extensive tests of grains which failed to germinate in the dormancy-breaking tests using tetrazolium and showed that embryo inviability was a very rare occurrence. In this study, therefore, the tetrazolium test was not repeated as inviability was considered trivial. The results were assumed to measure only embryo maturity which was expressed as

$$\% \text{ Embryo maturity} = \frac{G}{T} x100$$

where, G = number of germinated grains
T = total number of grains in the test.

The number of germinated grains in the normal and special germination tests was counted after 5 days of incubation. Grains were considered as germinated when there was evidence of embryo growth since it is difficult to detect the start of germinative processes, e.g. membrane reconstitution, in imbibed grains by the naked eye.

3.7.3 Dormancy

From the results of the normal and special germination tests, the level of dormancy in the grains was estimated as the difference between the special and normal germination for grain of the same age expressed as a fraction of the special germination (embryo maturity) as follows

$$\text{Dormancy } (\%) = \frac{SG - NG}{SG} x100$$

where, SG = special germination

NG = normal germination

Grain Colour Score

Three grains were used for a colour score test. The sodium hydroxide method of Quartley and Wellington (1962) was used. Grains were soaked in 5 ml of 5 % NaOH solution for one hour and compared with five standard cultivars which were similarly treated. The grain standards were ranked from 1 to 5, the lowest score being white and the highest dark red (Table 3.1). When the colour of the grain fell between two colour scores, this was recorded as the mean of the two scores.

Table 3.1. Standard cultivars for the graincoat colour score.

Standard Cultivar	Score
New Zealand Velvet	1
Spoetnik	2
Park	3
Pitic	4
Hope	5

The division line between "white" and "red" is at about 2.5-3.0 of the scores. Very young grains that had not yet developed colour and those in which the chlorophyll colour (green to yellowish green) of the pericarp was still present were scored as zero. This scheme gave 11 points on an the ordinal scale. With the intervals defined as they were, the scale may even approach being meristic.

Measurement of the Flavanols.

Flavan-3-ols (and flavan-3, 4-diols) were measured by a modification of the Swain and Hillis (1959) procedure (Gordon, 1975). This method is specific for flavan-3-ols and similar compounds with an undeactivated phloroglucinol or resorcinol A ring structure. Ten fresh grains were weighed, ground in a mortar and pestle and extracted in 10 ml of 10 % v/v aqueous Methanol in glass-stoppered vials for 24 hours. The vials were agitated continuously during the 24 hours on a gyrotory shaker (Model G2, New Brunswick Scientific Co. Inc.) at 100 RPM. At the end of the extraction period the samples were centrifuged at 5000 RPM for 10 minutes, and 2 x 1 ml aliquots were withdrawn from the supernatant for use in the vanillin / sulphuric acid method for the determination of flavanols (Swain and Hillis, 1959).

One aliquot (A) was reacted with 4 ml of 1 % vanillin in 70 % sulphuric acid (vanillin reagent) and the second aliquot (B) with 4 ml of 70 % sulphuric acid. Both aliquots were cooled rapidly in iced water and the colour of the solution allowed to develop for 15 minutes. The optical densities of the solutions were measured at 500 nm using as reference an aliquot prepared by adding 4 ml of 70 % sulphuric acid to 1 ml deionised water. A vanillin reagent blank (C) was prepared by adding 4 ml vanillin reagent to 1 ml deionised water and the optical density measured after each A and B set. The vanillin reagent blank was used to allow for reagent ageing in the final measured absorbance
(i.e. A-B-C).

A standard curve was determined using pure flavan-3-ol [(+)-catechin, $C_{15}H_{14}O_6$, Formula weight = 290.3; Lot 5637; Sigma Chemicals) at known concentrations. Four replicates were used. Each of the replicates was from one of 4 freshly prepared standard solutions at nine levels of concentration ranging from 5 $\mu g\ ml^{-1}$ to 65 $\mu g\ ml^{-1}$ in 10 % v/v aqueous MeOH. The predictive equation for the curve was:

$$Y = 5.75757 + 96.67282.\log(X1) + 11.26241.(X2) + 3.37004.\log(X3)$$

where, Y = concentration (μg / g)

X1 = (A-B-C)

X2 = Age of sample/10

X3 = C

A, B and C have the meanings assigned to them above.

The parameter estimates for the curve, their standard errors and t-statistics are given in Table 3.2 below. The analysis of variance is shown in Appendix 1, Table A 3.3. The regression coefficient of determination was 0.9745. The final concentration of flavan-3-ols in the grain samples was calculated as μg g^{-1} of grain extracted by multiplying by

$$\frac{10 \times V \times D}{m}$$

where, V = volume of extractant (10 ml here)

D = dilution factor (if any) used

m = mass of grain used

Table 3.2. Parameter estimates for the standard curve for estimating flavanols in developing wheat grains

Variable	DF	Parameter Estimate	Standard Error	T for H0: Pameter = 0	Prob > | T |
Intercept	1	5.75757	9.44040	0.610	0.5462
log(X1)	1	96.67282	2.82329	34.241	0.0001
X2	1	11.26241	4.42139	2.547	0.0159
log(X3)	1	3.37004	2.00785	1.678	0.1030

Table 3.3. Parameter estimates for the standard curve for estimating flavanols in developing wheat grains

Variable	DF	Parameter Estimate	Standard Error	T for H0: Pameter = 0	Prob > | T |
Intercept	1	2.76291	0.01655	166.9087	0.00077
X1	1	-1.18419	0.01431	-82.7507	0.0013

R^2 0.9745

Measurement of abscisic acid (ABA)concentration

Abscisic acid was measured using an enzyme linked immunosorbent assay procedure modified from Walker-Simmons (1987). The samples for the abscisic acid determination were kept frozen at -70 ^{0}C (*see* Section 5) until ready for use when the grain was allowed to thaw to room temperature still in the sealed vial. Between 200 and 500 mg (fresh weight) of the grains was measured and extracted in 5 ml cold 80 % MeOH containing 100 mg / l butylated hydroxytoluene (BHT) and 0.5 g / l citric acid monohydrate. BHT is an antioxidant and was included in order to prevent oxidation of ABA. Formation of Methyl (+) - abscisate (ABA - Me) was prevented by the addition of citric acid to the medium. ABA - Me can be formed as an artifact from conjugated ABA present in the plant material if alkaline methanol is used as the extraction medium.

The extraction was done overnight in the dark at 5 ^{0}C and 200 RPM on a gyrotory shaker. ABA is sensitive to light, the presence of which triggers isomerisation of the molecule, but is relatively stable to heat. The wheat grains were crushed in a motar and pestle and then transferred to 40 ml Eppendorf plastic tubes. At the end of the extraction, the samples were centrifuged at 4500 RPM for 5 minutes and the supernatant liquor transferred to 12 ml vacutainer plastic tubes. The supernatants were dried in a Savant Speed Vac Concentrator. The residues were dissolved in 5 ml of 0.02 M ammonium acetate and loaded onto sep-pak® columns for purification. The solution was allowed to run through the sep-pak® column. The sep-pak® was then rinsed with 5 ml 0.1 M acetic acid and 5 ml distilled water. ABA was eluted with 5 ml 80% v/v aqueous MeOH, collecting the eluent into a plastic vacutainer tube. The aliquot was dried under vacuum in the Savant vacuum concentrator, after which the residue was redissolved in 4 ml Tris-buffered saline (TBS) adjusted to pH 7.8. The ELISA assay materials used and sources of supply are given in Appendix A.

3.4.ELISA assay procedure (Walker-Simmons, 1987)

Nunc-Immuno Plate MaxiSorp™ microtitration plates having 96 wells were used in the procedure. The procedure described below was used on both standard and sample extract material. A synthetic ABA standard [(±)-2-cis, 4-trans-abscisic acid, M.W. 264.32 , 98 % pure] was used. Serial dilutions, ranging in concentration from 10 pg / 100 µl to 5000 pg / 100 µl, were

prepared from a standard stock solution containing 5 x 10^{-6} pg / 100 µl ABA. The method was sensitive to concentrations between 10 pg / 100 µl and 5000 pg / 100 µl.

Coating of Wells with ABA-4'-BSA Conjugate

A 200 µl aliquot of the conjugate was added to each well of the microtitration plate using an Eppendorf multipipette. The plates were incubated, in the dark, at 4 °C overnight. The conjugate binds strongly to the surface of the well and contains sites for binding ABA.

Incubation of ABA samples with Monoclonal antibody (Mab)

350 µl aliquots of the ABA sample and ABA standard solutions were delivered to labeled microcentrifuge tubes. Then 350 µl of Mab solution was added and mixed. Two additional microcentrifuge tubes were prepared, one having 350 µl TBS and 350 µl of Mab (labeled as 'B_0' = 100 % binding) and the other having 350 µl TBS + 350 µl of a solution containing 100 pg / 100 µl standard. The latter tube was labeled 'NSB' for non specific binding (= 0 %) The 700 µl in each microcentrifuge was enough for three replicate well assays. The solutions were wrapped in tinfoil and incubated overnight at 4 °C. The purpose of this step is to bind ABA molecules to Mab molecules because it is the Mab molecules that can bind with the enzyme molecules (antimouse-alkaline phosphatase or 2nd antibody).

Addition of ABA samples incubated with Mab

Plate wells coated with conjugate were washed three times with the washing buffer, TBST - BSA (*see* Appendix 3.4). The second and final washing solution was left in the plate for 5 minutes and then discarded. Then 200 µl aliquots of the samples incubated with Mab were added to two replicate wells. Plates were incubated, in the dark, for 2.5 hours at room temperature. The purpose of this step is to attach the ABA - Mab complex to the conjugate molecules fixed to the well surface. The binding is on a one - to - one basis between conjugate and the ABA - end of the ABA - Mab complex.

Addition of rabbit antimouse alkaline phosphatase (2nd antibody)

Wells were washed three times with washing buffer. Rabbit antimouse alkaline phosphatase (200 µl) was added to each well and the plates were incubated, in the dark, for 2 hours at room temperature. Washing removes any excess Mab molecules that are not complexed with ABA molecules. The 2nd antibody binds to the Mab end of the conjugate - ABA - Mab complex.

The concentration of ABA in the solution therefore determines the amount of Mab that reacts and also the amount of 2nd antibody taken up. The 2nd antibody catalyses the breakdown of *p*-nitrophenyl phosphatase to a yellow compound, *p*-nitrophenol, and a phosphate group. The optical density of *p*-nitrophenol is measured to determine the concentration of ABA in the original sample incubated with Mab.

1. *Measurement of Alkaline Phosphatase (PNPP)*

Wells were washed three times with washing buffer. *p*-Nitrophenyl phosphate substrate (200 µl) was added to each well. Plates were incubated, in the dark, for around 1 hour until the Absorbance at 410 nm of control samples containing no ABA (B_0), was approximately 1.0. The sample absorbance was measured at 410 nm using a Dynatech MR5000 Enzyme Immunoassay Reader. The absorbance of the samples is inversely proportional to the amount of ABA in the original sample incubated with Mab.

2. *Calculations*

The absorbances of duplicate standards or samples were averaged and the percentage binding of each standard or sample point was calculated by the following:

$$\%\ \text{Binding} = \frac{\text{Standard / Sample Absorbance} - \text{NSB Absorbance}}{B_0\ \text{Absorbance} - \text{NSB Absorbance}} \times 100 \text{ where,}$$

B_0 and NSB were described in 2. above.

The % Binding (B/B_0 %) was plotted against the concentration (pg) of (±)-2-cis, 4-trans-abscisic acid standards. A linear curve was drawn using a log-logit function

$$\text{Logit } B/B_0 = \text{Ln}\left(\frac{B/B_0\%}{100 - B/B_0\%}\right)$$

The sample concentration was determined by interpolation of the sample % Binding from the best fit standard curve. All sample results were the average of the replicates within the linear range of the ABA standard curve. The predictive equation for the curve was:

$Y = 2.762906 - 1.18419 \times \text{logit}(B/B_0)$

where, Y = Log(ABA concentration).

The parameter estimates for the curve, their standard errors and t-statistics are given in Table 3 below. The analysis of variance is shown in appendix 3.3. The regression coefficient of determination was 0.9996.

Recovery Test

The efficiency of the extraction procedure was measured by means of a recovery test using an internal standard.

Sample preparation

A 100 µl aliquot of ^{3}H-ABA in methanol (97 Ci / mMol) was added to the original grain sample before extraction. After extraction and drying in the Savant Speed Vac concentrator and purifying through the sep - pak® column, the extract was dissolved in 4 ml TBS. 200 µl was withdrawn into a microcentrifuge tube and 1 ml of scintillation fluid added. The solution was mixed for 30 seconds.

Standard preparation

The standard sample consisted of 100 µl of ^{3}H-ABA and 1 ml scintillation fluid. The mixture was vortexed for 30 seconds as above. Radio activity in the sample and standard solutions was counted in a Wallac 1409 Liquid Scintillation Counter. The recovery was 99.5 %. ELISA assay validation test. A validation test (Walker-Simmons, 1987) for the ELIZA procedure for ABA was conducted using the ABA standard [(±)-2-cis, 4-trans-abscisic acid] at four levels, 20, 100, 200 and 2000 pg/100 µl.

The procedure was as follows

3. *No sample*: 200 µl standard solution + 200 µl TBS
4. *25 % sample*: 200 µl standard + 100 µl sample + 100 µl TBS
5. *50 % sample*: 200 µl sample + 200 µl sample

The final concentration of the standard ABA solutions were 10, 50, 100 and 1000 pg/100 µl. An assay was conducted exactly as in section 6.6.1. above with the exception that the solutions 1, 2 and 3 above replaced the undiluted

sample material. Validation of the procedure was done in order to confirm non-interference by the plant extract material with linearity of the immunoassay. Data for the validation test are given in appendix A 3.6. The results confirmed linearity for the procedure.

3.7.4 Measurement of base α-amylase concentration

The base α-amylase, is the endogenous level of α-amylase present in the developing grain in the absence of germinative processes. This was measured using the method of Barnes and Blakeney, 1974, using the dried grain sample from the moisture content measurement. Drying had been at 60 ^{0}C for 48 hours. Alpha-amylase is stable at this temperature (Barnes and Blakeney, 1974). The procedure uses Phadebas® tablets and is adapted for cereal α-amylase. The Phadebas® tablet contains a β-limit dextrin, derived from potato starch, which is labeled with a dye, Cibachron blue. The β-limit dextrin is α-amylase specific and completely resistant to attack by β-amylase (Barnes and Blakeney, 1974). Hydrolysis of the β-limit dextrin causes release of an equivalent amount of the blue dye which, in turn causes an increase in the optical density of the suspension

3.7.4.1 Alpha - amylase assay procedure

Approximaley 200 - 500 mg of the dried sample was extracted with 5 ml Tris-maleate buffer containing Ca^{++} ions. The extraction was done using a motar and pestle and the extract was quantitatively transferred to a 12 ml centrifuge tube after which the mixture was centrifuged at 2000 G (aproximately 4500 RPM) for five minutes.

Using a micropipette, 100 µl of the supernatant was added to 4 ml of Tris-maleate buffer in a glass test tube and mixed gently. Several test tubes containing sample extracts (A) mixed with tris-maleate buffer and a blank (B) containing tris-maleate buffer in place of the sample extract were initially equilibrated in a water bath maintained at 50 ^{0}C for five minutes. At the end of the equilibration period, one Phadebas® tablet was added to each tube at 15 second intervals. The tubes were shaken gently for 5 seconds and then incubated at 50 ^{0}C in the water bath for 15 minutes. The test - tubes were agitated at five - minute intervals during the 15 minutes. The reaction was stopped by adding 1 ml of 0.5 M NaOH solution after 15 minutes from the

beginning. The suspension was filtered through Whatman No. 1 filter paper and the absorbance of the clear liquid was measured at 620 nm in a spectrophotometer (Philips Pye Unicam PU 8600 UV/VIS). Disposable cuvettes (10 mm square, supplied by Elkay Products Inc., 800 Boston, USA) were used in the measurement. Phadebas® Amylase Test Tablets were supplied by Kabi Pharmacia Diagnostics together with a standard curve. However, the supplied standard curve was not used in this assay because it is recommended only for human serum and urine α-amylase analysis.

3.7.4.2 Standard curve for the α - amylase assay

A standard curve was therefore constructed using four replicates of known concentrations of an α-amylase standard (1,4-α-D-Glucan glucanohydrolase; EC 3.2.1.1; Lot 124F - 0244; Type VIII - A prepared from Barley Malt and supplied by Sigma Chemical Company). This α-amylase standard contained 2.3 EU/g. One EU (enzyme unit) of α-amylase is defined as the amount of enzyme catalyzing the hydrolysis of 1 μmol glucosidic linkage per minute at 37° C.

The predictive equation for the regression of concentration of α-amylase on absorbance at 620 nm was

$$Y = 10.50152 + 1228.75844 \times (A-B)$$

where,

Y = α-amylase concentration (EU / l)
A = sample absorbance
B = Blank absorbance.

The statistics from the regression analysis are shown in Table 3.4. Both statistics were highly significant (Pr > F = 0.0001) and the regression coefficient of determination was 0.9998. The analysis of variance for the regression is given in appendix A 3.5. Using the predictive equation obtained from the standard curve, sample absorbance values were converted to EU/g dry tissue using the following equation

$$EU / g = \frac{Y(EU / l)}{1000} \times \frac{1000}{DWT\ (mg)} \times 5$$

where, Y has the same meaning as above, and, DWT = dry weight

Validation test for the Phadebas® tablet method for assay of α - amylase content in wheat grains

The principle behind the validation test was the same as in the test for the efficiency of the ELIZA procedure for ABA content in wheat grains (*see* section 6.6.3 above). The test was done on 14 - day old grains. Three sets of solutions (1) standard α-amylase solution at four levels of concentration, (2) a 50 % dilution of the sample extract, and (3) a 25 % dilution of the sample extract were prepared as follows:

Table 3.4. Parameter estimates for the standard curve for estimating α-amylase in developing wheat grains.

Variable	DF	Parameter Estimate	Standard Error	T for H0: Pameter = 0	Prob > | T |
Intercept, β_0	1	10.50152	2.28318	4.600	0.0001
Slope, β_1	1	1228.75844	2.91204	421.957	0.0001

(1) 2 ml standard solution + 2 ml tris - maleate buffer
(2) 2 ml standard solution + 2 ml sample extract
(3) 2 ml standard solution + 1 ml sample extract + 1 ml tris - maleate buffer

The original concentration of the standard α - amylase solutions in each set were 20, 100, 500 and 1000 EU/l, respectively, so that the final concentration levels were 10, 50, 250 and 500 EU/l, respectively. The analysis procedure was the same as in the actual assay described above. The results of the validation test are shown in Appendix 1, Figure A 3.1 (b). The 1:2 diluted sample shows good agreement with the standard, thus, proving linearity of the assay.

3.8 Data Analysis

Data sets from each variable from the ten genotypes were used as the dependent variable (Y) in separate regression analyses. The age of the grains measured in days after pollination (DAP) was used as the independent variable (X) in all the regression analyses. Initial simple plots of the data were used to identify outliers in the few cases where these existed. Any outliers were removed and treated as missing data. Different regression functions were fitted on data from each experimental unit and for each character measured. Estimated statistics from these functions formed a set of variables which could beanalysed in the Randomised Complete Block design. Secondary variables (timings to key physiological thresholds) were also estimated plot by plot using the fitted functions.

Germination tests and the colour score test data sets were truncated at the tail ends. The truncated regions were those where zero values were observed for more than two time nodes or where maximum levels were observed for more than two time nodes. The justification for truncating data at the tail ends was that long and / or uneven tails influence the position of the asymptotes of the fitted sigmoidal curves. The choice of two time nodes as the cut off point is arbitrary, is based on experience. It enables the valid comparison of estimates of statistics obtained in the regressions for each character across the treatments.

All data that suggested sigmoidal growth (i.e. grain dry weight, germination, and the colour score data) or sigmoidal decline (i.e. grain moisture content and the

dormancy data) in the initial plots were fitted using the Richards function. This is a generalised sigmoidal function having four parameters, *viz.* A, describing the upper asymptote; B, describing the X-axis placement; K, the acceleration of rate statistic; and V, the inflexion placement statistic.

3.8.1 Non-linear regression

Least squares estimates were obtained in the analysis in the calculation of the Jacobian and gradient at each point. The Marquardt derivative method was used in the NLIN procedure in the SAS program (SAS Institute Inc., 1990). The Marquardt method is a compromise between the Gauss-Newton and the Steepest descent methods and is equivalent to performing a series of ridge regressions useful when the parameter estimates are highly correlated (as in the Richards function), or the objective function is not well approximated by a quadratic (SAS Institute Inc., 1990). In this method, the selected starting values for the four parameters in the procedure are updated after the first iteration, and NLIN, through a grid search that may involve many iterations, may or may not converge on the least squares for the regression. If convergence is met, NLIN prints

1. an analysis-of-variance table including as sources of variation Regression, Residual, Uncorrelated Total, and Corrected Total,
2. parameter estimates,
3. an asymptotically valid standard error of the estimate.
4. an Asymptotic 95% Correlation Interval for estimates of the parameters,
5. an Asymptotic Correlation Matrix of the parameters.

3.8.2 The Quadratic exponential function

The α-amylase data were analysed using a quadratic exponential function. Previous studies involving changes of α-amylase concentrations in developing and germinating wheat grains (Gordon, 1975; Gordon *et al.*, 1979; Sereeprasert, 1990) showed that the quadratic exponential function is suitable for analysis of α-amylase data. Simple plots of the raw data for the flavanols showed an exponential decline similar to that for the base α-amylase data and, therefore, the quadratic exponential was also preferred for these data.

The regression function for the quadratic exponential is of the form

$$Y = e^{[\alpha+\beta_1(X)+\beta_2(X)^2]} \qquad 3.1$$

where Y = base α-amylase concentration (EU/g), or flavanol concentration (μg/g),

X = age of grains in days

α, β_1, and β_2 are unknown parameters.

The logarithmic form of equation 3.1 gives

$$\ln(Y) = \alpha + \beta_1(X) + \beta_2(X)^2, \qquad 3.2$$

a second order polynomial which is biologically meaningful. The β_2 represents extra deceleration accentuting the main rate of change, β_1. Estimates for the values of base α-amylase at harvest ripeness, germination maturity (95% germination in the normal germination test) and embryo maturity (95% germination in the special germination test), and similarly for the flavanols, were derived by substituting X (days after pollination) with appropriate values.

3.8.3 Function analysis

The statistics from all regression functions fitted on the data from each experimental unit formed a new set of variables for analysis. Univariate analysis (ANOVA) was inappropriate, however, because of correlation amongst the statistics, and because they should be considered as a set (defining each curve function). These parameter estimates were analysed by multivariate analysis of variance (MANOVA). In addition, therefore, standardised discriminant functions were estimated that could be used to discriminate the cultivars into significance groups with regard to each character regression (profile) investigated. Parsimony was employed in order to choose the best discriminant(s) to use in the model. Only the discriminants that explained 70 % or more of the cumulative total variance, and were significant, were retained. Where only one discriminant passed the parsimony test, the univariate t-statistic was used to test for significance amongst the discriminant scores. It was intended to use Hotelling's T^2 when more than one Discriminant was needed, but this proved unnecessary. Discrimiation structure (between cultivars) and the standardized coefficients matrix were examined in order to establish meaning to the Discriminants and to estimate the relative discrimianatory power of the variables (regression statistics).

The method used to summarise the discriminatory importance of the function statistics was that of Gordon (*pers. comm.*, 1995). In this method, the between cultivars (treatments) structure and the standardized discriminany coefficients were

examined jointly, in order to compare both the associative and functional aspects of the discrimination. The structure and coefficients were given "importance points" of 3,2,1,1/2 and 0, respectively, for each descending quantile of their ranges. Functional importance (thus defined) was then modified by associative importance according to the scheme in Table 3.5. In this way, the discriminatory importance of each attribute was obtained, and this greatly facilitated understanding of the behaviour of the function.

The discriminant scores provided a means of discrimination amongst the cultivars on the joint basis of their four sigmoid statistics (i.e. of discriminating their development *profiles* for any one character). Like in a univariate discriminator (e.g. lsd) it was still necessary to distinguish the significance gaps (or overlaps) in the ranked discriminants. As these discriminants are normal (log linear functions of the original normal variates - Morrison, 1972) the t-test can be used for this, just as for lsd. Where only one discriminant is required, Student's usual t is appropriate: where more than one was needed, Hotelling's T^2 would be used.

3.8.4 Analysis of variance

The secondary variables, from the sigmoid profiles, were used to focus on specific issues of development interest, e.g. what is the level of each dependent variable at harvest ripeness? The analyses of variance on the secondary variables were done using the GLM procedure of SAS statistical package (SAS Institute Inc., 1990). The variables in the analyses were those variables derived from the regression equations. The model for the analysis of variance was the usual RCB model, as follows:

$$Y_{ij} = \mu + \alpha_i + \beta_j + \varepsilon_{ij}$$

where,

Y_{ij} = the observed value of the variable of the i^{th} treatment in the j^{th} block

μ = the mean for the population from which the sample of observations was taken

α_i = the effect of the i^{th} treatment

β_j = the effect of the j^{th} block

ε_{ij} = the random error associated with the individual observations

The significance of treatment or block effects was tested using the F - test.

Table 3.5. Interpretation of Discriminant functions (Gordon, *pers. comm.*, 1995).

Standardised coefficientsStructure Matrix

Proportion of largest coefficient	Points
0.8 - 1.0	3
0.6 - 0.8	2
0.4 - 0.6	1
0.2 - 0.4	1/2
0 - 0.2	0

Correlation	Points
0.99 -0.80	3
0.79 - 0.60	2
0.59 - 0.40	1
0.39 - 0.20	1/2
0.19 - 0.0	0

The combined analysis

Score from Structure Matrix	3	2	1	1/2	0
Score from the Std Coeff					
3	Strong	Strong	Medium	Suppressed	Suppressed
2	Strong	Medium	Medium	Weak	Suppressed
1	Enhanced	Enhanced	Weak	weak	Suppressed
1/2	Pseudo	Pseudo	Weak	Null	Null
0	Pseudo	Pseudo	Null	Null	Null

Chapter 4

4. RESULTS

The raw data collected in the experiment is shown in the Appendix 5, Table A 7.2.

Variability amongst the ten wheat cultivars was observed in the number of days from sowing to flowering (Table 4.1). The flowering date was recorded as the day when anthers had extruded from the middle third of the ear when these ears were tagged.

4.1 Grain dehydration

Grain dehydration is a key attribute as it defines "harvest ripeness", thereby providing the background to the other development attributes.

4.2 Development profiles for grain moisture content

The grain moisture content data were fitted with Richards functions as discussed in Chapter 3. Estimates of the four parameters in the Richards function and the standard errors associated with each statistic are given in the Appendix (Tables 4.1(a) to 4.1(j)). The statistics estimated in the functions are (i) A, the upper asymptote, (ii) B, the X - axis displacement, (iii) K, the rate of acceleration or decelleration, and, (iv) V, the inflexion placement. Also shown are the F values and the coefficients of regression, R^2, for each individual function estimated. All the functions were not different from logistic curves (V = 1 in the logistic curve, *see* chapter 3). Previous studies have used the logistic function to fit grain moisture content data (e.g. Verry, 1978). The grain moisture content values at the start of the experiment (7 days after anthesis) ranged from 76 % to 80 %. In the majority of cases the functions fitted the data very well as the coefficients of regression show. Graphs of the estimated Richards functions and data points for each experimental unit are shown in Appendix 2, Figures A 4.1(a) to A 4.1(j).

Table 4.1. The mean number of days from sowing to pollination of ten wheat cultivars and tests for significant differences.

Cultivar	Mean (days)[1]	t-group
Isis	104.875	A
Thatcher	94.900	B
La Prevision	94.350	BC
Hilgendorf 61	90.975	BCD
Kenya 321	90.825	CD
Gamenya	90.500	CD
Karamu	89.850	D
Brevor	87.550	DE
Sonora 64 A	84.525	DEF
Tordo	82.550	F

[1] Means with the same letter are not significantly different

4.2.1 Analysis of the Richards function for grain dehydration

The Richards functions for grain moisture content were analysed in a MANOVA. The variables used in the analysis were the four statistics estimated. These four statistics captured the effect of time and, therefore, it was not necessary to consider time, as a variable, in the analysis. Parsimony for the model was set at 70 % of total variance and the first and second discriminants satisfied this requirement. The manova test for the hypothesis of no overall treatment (cultivar) effect was highly significant ($P < 0.0011$) but the block effect was not. Of the two discriminants, the first was highly significant ($P < 0.0011$) but the second was not. Only the first discriminant was used in the analyses.

Table 4.2 shows some of the proportion and significance of all 4 discriminants. The first canonical correlation was very strong, the second and third moderate, while the fourth canonical correlation was weak. The canonical structure between the discriminants and their variables and the standardised canonical coefficients for the variables are reported in Table 4.3

Table 4.4 shows the mean values of the statistics obtained from the Richards functions for each treatment. Discriminant scores were calculated for each cultivar to yield two sets of discriminants on which tests of significance were done. Figures 4.1 and 4.2 show, superimposed, diagrams for the mean Richards functions fitted on data for grain moisture content for the ten wheat cultivars. The table within each figure shows significance groups amongst the cultivars using the ordinary t-test. These tables are sub-sets of one table (Table 4.5). There were no clear divisions between the white - and the red - grained cultivars. This confirms the results from previous studies (Gordon, 1975; Verry, 1978).

Using the method developed by Gordon (*pers. comm.*, 1995) described in chapter 3, Table 3.5, the following results were obtained:

1. The upper asymptote (A) had no influence on the first discriminant.
2. The influence of the 'X - axis placement' factor (B) was suppressed, probably by a combination of the weak influences from both the 'acceleration' factor (K) and the 'inflexion placement' factor (V) on the first discriminant.

From this analysis, it was concluded that the first discriminant determined the 'dehydration value' of the grain during development since "large" values (i.e. less

negative, tending to 0) of K would cause a slowing down of water loss from the grain and, therefore, delay harvest ripeness and *vice versa*. The second discriminant contributed to the control of grain dehydration by having a strong influence from B, which effects the horizontal displacement of the function to the right and, therefore, causes a delay in harvest ripeness.

4.3 Harvest ripeness (HR)

The analysis of variance for harvest ripeness was not significant (F = 1.07). Both the genotypic and block effects were non significant. Table 4.6 contains the mean number of days from pollination to harvest ripeness and the test for significant differences amongst the cultivars for this character. There were two overlapping groups.

The variance components estimates and the heritabilty estimates for harvest ripeness are shown in Table 4.7. The genotypic variance component was significant ($P < 0.05$). Harvest ripeness proved to be largely influenced by the environment ($h^2 = 0.252$, narrow sence). The definition of harvest ripeness is based on grain moisture content which itself is largely influenced by environmental variables, e.g. temperature and humidity. Therefore, it is not surprising that the environment had a large influence.

Table 4.2. The four discriminants for grain dehydration and their contributions to total variation.

Discriminant	Proportion	P > F
1	0.6837	0.0011
2	0.1631	0.1936
3	0.1196	0.3689
4	0.0337	0.7158

Table 4.3. The correlation between the variables and the first discriminant (between canonical structure), and the standardized coefficients for each variables.

Variable	Correlation	Coefficient
A	-0.1396	0.8094
B	0.0885	4.5039
K	-0.2397	-3.1380
V	0.4869	2.3881

Table 4.4. Estimated statistics for the mean Richards functions for grain moisture content of developing grain of ten wheat cultivars.

Statistic	A		B		K		V	
Cultivar	Mean	**s.e. A**	Mean	**s.e. B**	Mean	**s.e. K**	Mean	**s.e. V**
Gamenya	76.2481	**0.4927**	-4.3121	**0.9191**	-0.1250	**0.0288**	0.9763	**0.0516**
Tordo	77.7073	**1.2501**	-3.7356	**0.4271**	-0.1000	**0.0001**	1.0009	**0.0721**
Kenya 321	77.7392	**1.5004**	-3.9320	**0.2291**	-0.0998	**0.0002**	1.0173	**0.0287**
Brevor	78.2187	**1.2641**	-4.8738	**1.2419**	-0.1248	**0.0288**	1.0272	**0.0622**
Isis	78.7304	**0.9904**	-3.3385	**0.3106**	-0.0999	**0.0004**	0.9884	**0.0548**
Sonora 64 A	77.2117	**1.8646**	-4.2492	**1.5542**	-0.1251	**0.0499**	1.0364	**0.0474**
Thatcher	77.9684	**0.8229**	-4.0865	**1.0949**	-0.1123	**0.0251**	1.0449	**0.0634**
La Prevision	77.0885	**1.3227**	-3.2268	**0.3578**	-0.0969	**0.0090**	0.9430	**0.0131**
Hilgendorf 61	77.7271	**1.4999**	-3.6978	**0.0427**	-0.0999	**0.0001**	0.9404	**0.0232**
Karamu	78.0000	**2.3094**	-3.5831	**0.2187**	-0.0988	**0.0023**	0.9866	**0.0536**

Table 4.5. Discriminant scores for the grain moisture content functions of developing grain of ten wheat cultivars and t-tests for significant differences.

Cultivar	Discriminant score[1]	
La Prevision	5.9348	A
Isis	2.7545	AB
Gamenya	2.1726	AB
Hilgendorf 61	1.0227	BC
Karamu	0.6794	BCD
Sonora 64 A	0.5471	BCD
Tordo	-0.5999	BCD
Thatcher	-2.2227	CD
Kenya 321	-3.1173	DE
Brevor	-7.1839	E

[1]Discriminant scores with the same letter are not significantly different

Figure 4.1. Diagrams for the mean Richards functions for grain dehydration of cvs. La Prevision, Hilgendorf 61, Sonora 64 A, Thatcher and Brevor.

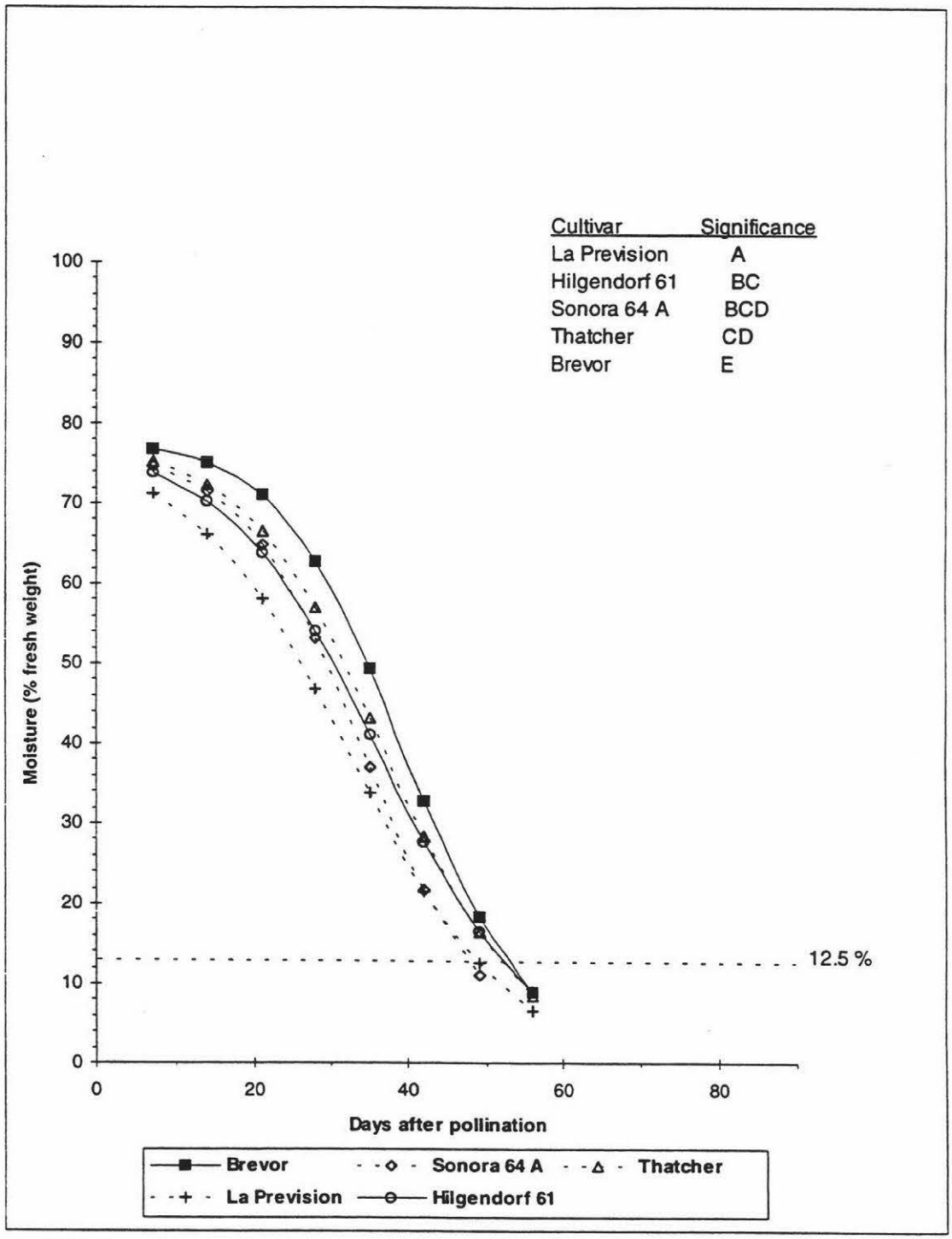


Figure 4.2. Diagrams for the mean Richards functions for grain dehyadration of cultivars Isis, Gamenya, Karamu, Tordo and Kenya 321.

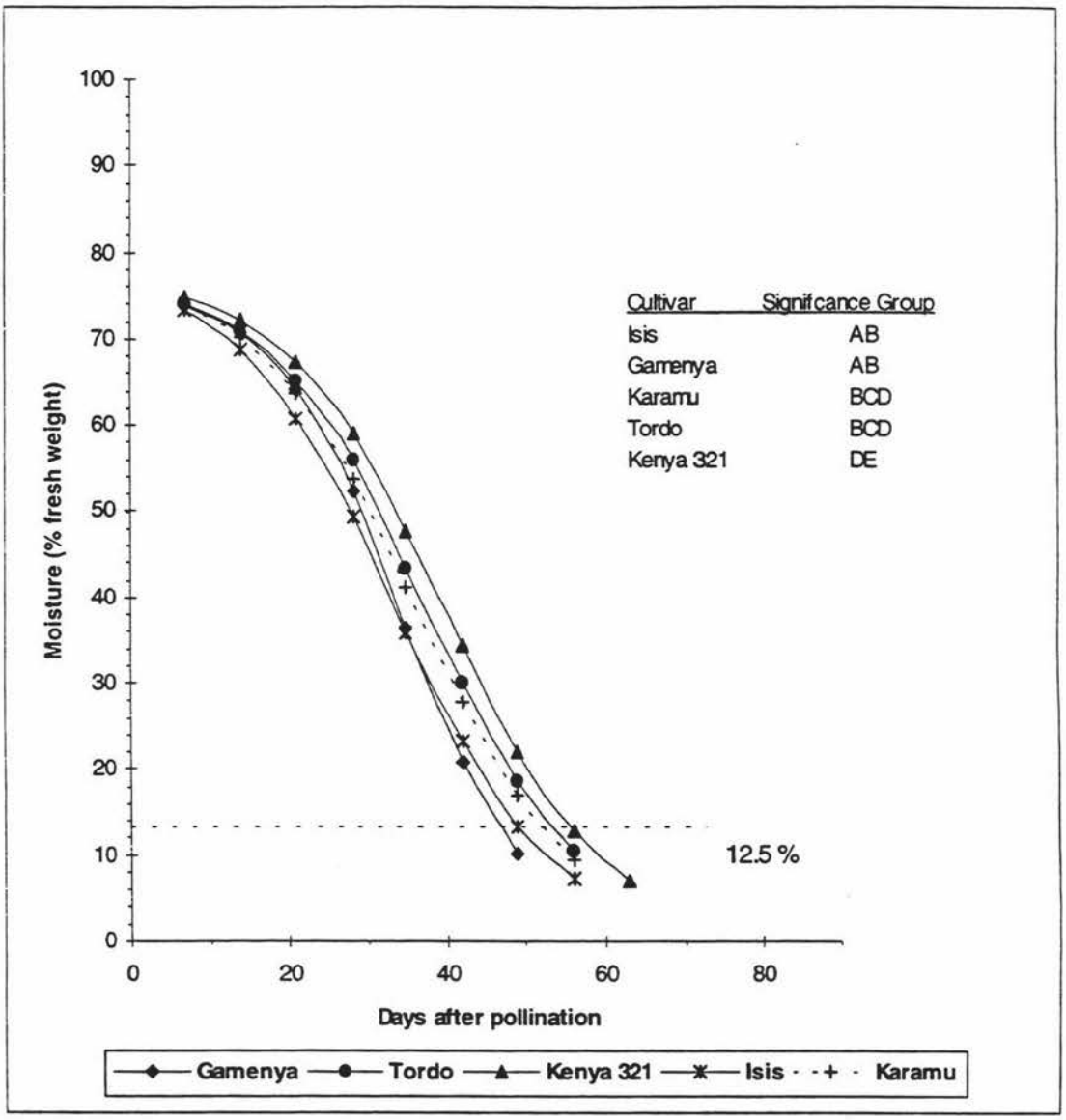


Table 4.6. The mean number of days to reach harvest ripeness and t - tests for significant differences among ten wheat cultivars.

Cultivar	Mean[1]	
Kenya 321	58.01	A
Tordo	55.68	AB
Karamu	54.55	AB
Brevor	54.43	AB
Thatcher	53.72	AB
Hilgendorf 61	51.69	AB
Isis	51.64	AB
La Prevision	51.09	B
Sonora 64 A	50.64	B
Gamenya	49.38	B

[1] Means with the same letter are not significantly different.

Table 4.7. Variance components and heritability estimates for harvest ripeness of ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F (ANOVA)	P > F
Genotype	9	7.0023	2.98579	0.252	0.11194	1.07	0.4193
Block	3	0.5261	0.33272				
Error	27	20.8236	5.46854				

5. GRAIN MASS

Grain mass is a character commonly used to monitor grain development, and is an important agronomic character.

5.1 Development profiles for grain mass

Grain growth was measured as dry mass (mg) of the grain using ordinary gravimetric methods. Simple plots of the data for grain growth showed a sigmoidal

5.1 Development profiles for grain mass

Grain growth was measured as dry mass (mg) of the grain using ordinary gravimetric methods. Simple plots of the data for grain growth showed a sigmoidal pattern and, therefore, Richards functions were fitted on these data. Tables A 4.2 (a) to A 4.2 (j), in Appendix 2, show the statistics obtained from the Richards functions fitted on the data from all the experimental units. The regression fits were excellent and the coefficients of regression were all high. Graphs of the functions for each experimental unit and the observed data points are shown in Appendix 2 (Figures A 4.2 (a) to A 4.2 (j)). Table 4.8 shows the MANOVA mean values of the statistics estimated from the Richards functions and Figure 4.3 shows diagrams of the mean Richards functions for grain mass. The Richards functions were used to estimate the following secondary variables:

Mnemonic	Description of variable
GMHR	Grain mass at harvest ripeness
T95GM	Number of days from pollination to maximum grain mass

5.1.1 Analysis of the Richards functions for grain mass

Tables 4.9 and 4.10 show properties of the canonicals, the structure matrix between the first discriminant and its variables and the standardized coefficients for the variables, respectively. The Manova hypothesis for no overall genotypic effects was highly significant ($P < 0.0001$) but that for blocks was not. Only the first and the second discriminants showed strong correlations. The first discriminant was highlly significant ($P < 0.0001$) and the second was only significant at the 10 % significance level. However, the first discriminant satisfied the 70 % required by parsimony and all the other discriminants were dropped from any further consideration.

The first discriminant showed strong correlations with all 4 variables. Examination of the structure matrix and the standardized coefficients (Tables 4.10) gave the following results:

Large A values tended to give steep slopes to the curves while small A values gave rise to slowly rising, lazy curves (Figure 4.3). This enhanced the effect of B and also gave rise to the pseudo effects of K and V. Thus it would be expected that the first discriminant affected both the grain filling rate and the grain filling time.

The discriminant scores extracted were tested for significant differences amongst the cultivars using the t-test (Table 4.11). Three significance groups and three cultivars that were significantly different from the others were separated.

Table 4.8. Estimated statistics for the mean Richards functions for grain mass of developing grain of ten wheat cultivars.

Statistic	A		B		K		V	
Cultivar	mean	**s.e. A**	mean	**s.e. B**	mean	**s.e. K**	mean	**s.e. V**
Gamenya	38.465	**4.1547**	3.4725	**1.0162**	0.1753	**0.0728**	0.9294	**0.1295**
Tordo	48.742	**2.0486**	3.2301	**0.3882**	0.1749	**0.0301**	1.1634	**0.1654**
Kenya 321	50.83	**1.5810**	3.6536	**0.7142**	0.1634	**0.0460**	1.0562	**0.0694**
Brevor	54.703	**4.6618**	3.362	**0.6696**	0.148	**0.0494**	1.0382	**0.1062**
Isis	48.48	**4.3504**	3.1564	**0.5890**	0.1467	**0.0478**	1.1153	**0.1116**
Sonora 64 A	43.745	**1.6891**	3.299	**0.2120**	0.1508	**0.0002**	1.0407	**0.0485**
Thatcher	34.941	**2.2688**	4.7839	**0.8815**	0.2404	**0.0141**	0.7883	**0.1937**
La Prevision	45.24	**2.2113**	3.5632	**0.5799**	0.1633	**0.0247**	1.0007	**0.0401**
Hilgendorf 61	46.251	**7.1858**	3.3499	**0.0417**	0.1635	**0.0250**	1.0632	**0.0940**
Karamu	47.1	**1.2611**	3.9028	**0.3611**	0.1627	**0.0250**	1.0099	**0.0879**

Figure 4.3. Diagrams of the mean Richards functions for grain mass of developing grains of ten wheat cultivars.

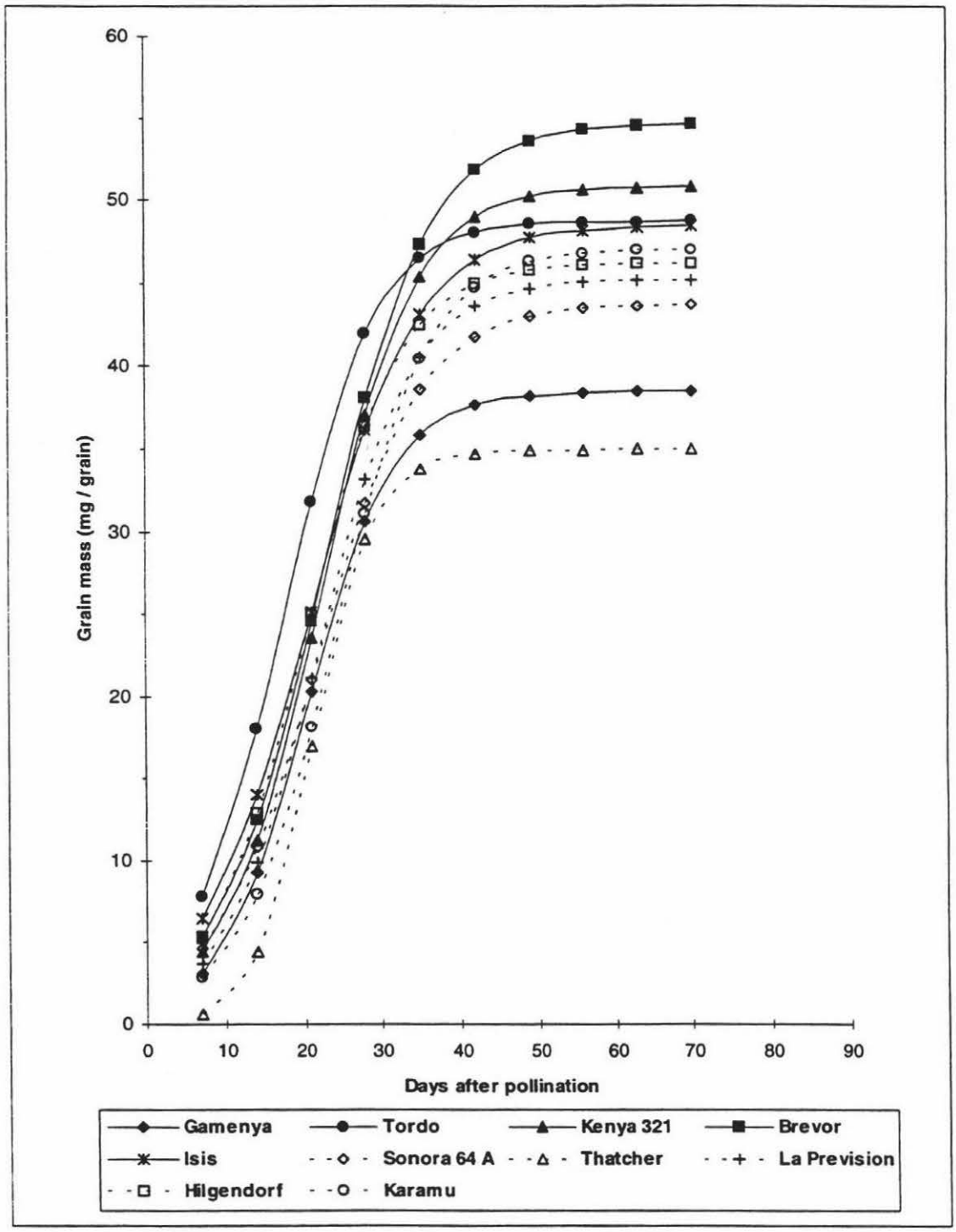


Table 4.9. The four discriminants for grain mass and their contributions to total variation.

Discriminant	Proportion	Pr > F
1	0.7469	0.0001
2	0.1963	0.0513
3	0.0448	0.7120
4	0.0121	0.8729

Table 4.10. The correlation between the variables and the first discriminant (between canonical structure) for grain mass.

Variable	DISCR1	DISCR1
A	0.9655	1.6661
B	-0.7951	-0.8807
K	-0.8549	-0.0009
V	0.8869	0.1589

Table 4.11. Discriminant scores for the grain mass functions of developing grain of ten wheat cultivars and tests for significant differences.

Cultivar	Discriminant score[1]	
Brevor	17.5752	A
Kenya 321	8.8502	B
Tordo	8.1641	B
Isis	7.8578	B
Hilgendorf 61	2.3325	C
Karamu	0.2812	CD
La Prevision	-1.1399	D
Sonora 64 A	-2.0706	D
Gamenya	-13.387	E
Thatcher	-28.463	F

[1] Discriminant scores with the same letter are not significantly different

5.2 Grain mass at harvest ripeness (GMHR)

Grain mass at harvest ripeness is an important attribute for obvious reasons. It is one of the components of yield. Grain mass was measured as the dry weight of the grain after drying in an oven for 48 hours at 60 ^{0}C.

Table 4.10 shows the mean grain mass (mg) at harvest ripeness and the results of the test for significant differences amongst the ten cultivars. The analysis of variance was highly significant ($P < 0.01$).

5.2.1 Heritability estimates for grain mass at harvest ripeness of ten wheat cultivars

Table 4.13 shows the variance components and the heritability estimates and their standard errors for grain mass at harvest ripeness, respectively. The genotypic component was significant at the 0.05 significance level. The results showed that grain mass is mainly controlled by the environment ($h^2 = 0.271$, narrow sence).

5.3 Number of days from pollination to maximum grain weight (T95GM)

This variable represents total time of growth of the grain, i.e. building of tissue material and accumulation of storage materials. The grain was considered fully grown when it attained 95 % of the maximum observed grain mass. All the cultivars completed grain growth by day 30 after pollination (Table 4.14). The analsis of variance showed no significant differences at the 0.1 level. This period did not depend on grain size since small grains such as Gamenya or Sonora 64 A (Table 4.11) were no different from large grains such as Brevor or Kenya 321.

5.3.1 Heritability estimates of number of days from pollination to maximum grain weight

Variance components and heritability estimates for T95GM are shown in Tables 4.15. The heritability estimates were slightly higher than those for grain mass at harvest ripeness showing a larger genetic influence. This variable includes two distinct stages of grain growth, namely, cell division and storage material accumulation. The former of these is largely under the control of the genotype whereas the latter is mainly dependant on environmental conditions.

Table 4.12. The mean grain mass (mg) at harvest ripeness and tests for significant differences amongst ten wheat cultivars

Cultivar	Mean (mg) [1]	
Brevor	54.703	A
Kenya 321	50.830	AB
Tordo	48.742	AB
Isis	48.480	AB
Karamu	47.100	ABC
Hilgendorf 61	46.251	ABC
La Prevision	45.240	ABC
Sonora 64 A	43.745	BCD
Gamenya	38.465	CD
Thatcher	34.941	D

[1] Means with the same letter are not significantly different.

Table 4.13. Variance components and heritability estimates for grain mass at harvest ripeness and the ANOVA F value and significance.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F (ANOVA)	P > F
Genotype	9	17.4640	7.44667	0.271	0.11577	3.53	0.0031
Block	3	6.8751	4.34823				
Error	27	47.0034	12.34369				

Table 4.14. The mean number of days from pollination to maximum grain mass and tests for significant differences amongst ten wheat cultivars.

Cultivar	Mean (mg) [1]	
Kenya 321	29.355	A
Karamu	29.270	A
Sonora 64 A	28.498	AB
Hilgendorf 61	27.003	ABC
La Prevision	25.415	ABC
Gamenya	24.895	ABC
Brevor	24.368	ABC
Isis	24.023	BC
Thatcher	23.675	BC
Tordo	22.618	C

[1] Means with the same letter are not significantly different.

Table 4.15. Variance components and heritability estimates for number of days from pollination to maximum grain mass of ten wheat cultivars and the ANOVA F value.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F (ANOVA)	P > F
Genotype	9	6.01103	2.56311	0.330	0.12517	1.79	0.1012
Block	3	1.55662	0.98450				
Error	27	12.22691	3.21095				

5. EMBRYONIC MATURITY

The special germination test was used four dormancy - breaking mechanisms. This test measured directly the grain's potential ability to germinate and, therefore, reflected embryonic maturity (*see* Materials and Methods, Chapter 3). Embryonic maturity was estimated as the number of grains that germinated under the dormancy - breaking conditions.

5.1 Development profiles for embryonic maturity

The observed data for the special germination test showed that embryonic maturity was complete in all the cultivars, except La Prevision, at the end of the experimental period. Therefore, the upper asymptote, A, of the estimated functions was restricted to 100% in all experimental units, except the four EU's belonging to the cv. La Prevision. The statistics estimated by the Richards functions are shown in Tables A 5.1 (a) to A 5.1 (j), in Appendix 3 and the mean values and standard errors of the these estimates are reported in Table 5.1. The regression fits were all very good. Graphs of the functions and the observed data points for each experimental unit are shown in Appendix 3 (Figures A 5.1 (a) to A 5.1 (j)). The functions were used to estimate secondary variables that could be tested for significant differences amongst the cultivars. The following is a list of these variables:

Mnemonic	Variable description
1. **EMHR**	Embryonic maturity at harvest ripeness
2. **T50EM**	Number of days to 50 % embryonic maturity (median embryonic maturity)
3. **T95EM**	Number of days to 95 % embryonic maturity (full maturity)
4. **HR-T50EM**	Number of days between harvest ripeness and median embryonic maturity
5. **HR-T95EM**	Number of days between harvest ripeness and days to 95 % embryonic maturity

5.1.1 Analysis of the Development Profiles for embryo maturity

Only the first discriminant passed the parsimony test of 70 % (Table 5.2). Its correlation was strong and significant at P = 0.0001. A summary of the correlations between the variables and the first discriminant and the standardized

discriminant coefficients for each of these variables are given in Table 5.3. Analysis of this information using Table 3.5 in chapter 3 gave the following results:

1. The upper asymptote, A, had no influence on the first discriminant.
2. Both B and K were suppressed.
3. V was identified as a pseudo variable.

It appears, therefore, that the important influences on the first discriminant came from B and K although these influences were dampened because of the poor correlations. Large B values were expected to cause a rightward displacement of the function but this would be reduced due to suppression. Similarly, large K values were associated with steep rises of the functions.

Discriminant scores for the cultivars separated them into two distinct groups and two individuals at either end of the scale. The test for significant differences amongst the cultivars was done at the 0.05 significance level using the ordinary t - test (Table 5.4). Brevor had the largest positive score whereas La Prevision had the largest negative score. The majority of the white cultivars were early maturing (the exception being Tordo) and the majority of the red - grained cultivars were late maturing, with the exception of Sonora 64 A. The pseudo effect of V, the inflexion placement factor, is noticeable in the middle third of the curves where the point of inflexion would normally be expected in these functions. In view of these observations, it was concluded that the first discriminant affected grain maturity and may be described as a 'maturity factor'.

5.2 Embryonic maturity at harvest ripeness (EMHR).

The mean values for the percentage germination under dormancy - breaking conditions at harvest ripeness and the t-tests for differences amongst the cultivar are given in Table 5.5. Two overlapping significance groups were found but La Prevision was not in any of them.

5.2.1 Heritability estimates for embryonic maturity at harvest ripeness

Heritability estimates for EMHR (Table 5.6) showed that there was a relatively large genetic influence ($h^2 = 0.589$) on this character. The environment also had a considerable influence on development of the embryo since development can

occur, normally, in the absence of adequate nutrients, water or a suitable temperature. Environmental stresses can retard development.

5.3 Days from pollination to median embryonic maturity (T50EM).

The mean values for days from pollination to median embryonic maturity are shown in Table 5.7. The range for the ten cultivars was from 26.04 days (Gamenya) to 46.76 days (La Prevision). Four overlapping groups were separated at P = 0.05. La Prevision was different from every other cultivar. This character showed strong heritability estimates (h^2 = 0.730) as shown in Table 5.8. These estimates were almost 13 % higher than the corresponding estimates for embryonic maturity at harvest ripeness (Table 5.6). T50EM is a measure of time whereas EMHR is a measure of the rate of development. The connection between the two measures lies in the fact that both are correlated with the rates of growth and development of the embryo. Therefore, a comparison of heritabilities for the two measures is justified. The differences in heritabilities are probably due to the fact that T50EM occurred at a point when events, within the developing embryo, were occurring at or near their maximum rates[1] whereas the embryo would have been entering a quiescent phase by the time it gained full maturity. Therefore, variation amongst the cultivars was expected to be greater for T50EM than for EMHR

5.4 Days from pollination to full embryonic maturity (T95EM).

Four overlapping groups were identified amongst the ten cultivars in the significance test for differences on days from pollination to full embryonic maturity (Table 5.9). La Prevision was different from all the other cultivars. Variance components and heritability estimates for this character are shown in Tables 5.10. There was strong genetic influence on this character (h^2 = 0.782), stronger than for T50EM and almost 20 % higher than that for embryonic maturity at harvest ripeness. The same reasons mentioned above (section 5.4) for differences between T50EM and EMHR may explain the observed differences in heritabilities

[1] All the Richards functions fitted could be described as logistic functions ($V \cong 1$). In a logistic function, the maximum rate of change occurs at the point of inflexion. This point is located midway between the two asymptotes of the function. During growth and development, changes in a character such as rate of increase in embryonic maturity increase to a maximum at the point of inflexion before decreasing to a minimum at maturity.

between T95EM and EMHR. The difference between heritability estimates for T95EM and similar estimates for T50EM is due to the extra variation amongst the cultivars during the space of time between T50EM and T95EM. The embryo is in the declining phase of growth and/or development during this period but the rate of decline may not be the same in all the cultivars. Variation in microenviroments amongst the experimental units was trivial.

5.5 Difference between harvest ripeness and days from pollination to median embryonic maturity (HR-T50EM).

This represented a measure of the later phase of embryonic growth and/or development when the rates were declining. It is also the amount time that would be allowed, after T50EM, for the grain to complete its growth and/or development before being harvested. Median embryonic maturity was observed before harvest ripeness in all the ten cultivars studied. Kenya 321 had the largest separation between median embryonic maturity and harvest ripeness ripeness (Table 5.11).

5.5.1 Heritability estimates for HR-T50EM

Heritability estimates for this measurement were moderate (h^2 = 0.580). The variance components and the heritability estimates are given in Tables 5.12. Care must be exercised, however, in interpreting these estimates since estimates for harvest ripeness and days from pollination to median embryonic maturity (T50EM) are based on functions fitted on different characters, namely grain moisture content and embryonic maturity. Harvest ripeness was found to be largely under the control of the environment (*see* Section 4.4 in Chapter 4). As discussed above (*see* Section 5.4), T50EM was found to have a large genetic component. Therefore, the individual variations in each of these characters amongst the cultivars must be taken into consideration in any explanation for the differences in HR-T50EM.

5.6 Difference between harvest ripeness and days from pollination to embryonic maturity (HR-T95EM)

Three overlapping groups were found and one cultivar, La Prevision, was different from all the others at P = 0.05. Embryonic maturity occurred after harvest ripeness in La Prevision and midway towards harvest ripeness in Kenya 321. Kenya 321 was different from Tordo, Karamu, Hilgendorf 61, Isis, Thatcher and La Prevision

(Table 5.13). The genotypic variance components and heritability estimates are shown in Table 5.14. The analysis of variance was highly significant ($P < 0.001$). A strong genetic influence was found for HR-T95EM ($h^2 = 0.646$).

Table 5.1. The mean values of the statistics estimated from the Richards functions for special germination of ten wheat cultivars.

Attribute:	A		B		K		V	
Cv.	mean	**s.e. A**	mean	**s.e. B**	mean	**s.e. K**	mean	**s.e. V**
Gamenya	98.7548	**0.9591**	8.1051	**1.8203**	0.2842	**0.0529**	0.9803	**0.0231**
Tordo	99.8750	**0.2500**	10.1824	**2.2382**	0.2634	**0.0477**	1.0164	**0.1013**
Kenya 321	99.7485	**0.4914**	7.3144	**2.4493**	0.2385	**0.0855**	1.0155	**0.0297**
Brevor	99.4955	**0.5722**	4.7823	**1.0982**	0.2385	**0.0855**	1.0155	**0.0297**
Isis	99.7497	**0.4943**	6.2384	**2.1621**	0.1759	**0.0646**	0.9133	**0.0747**
Sonora 64 A	99.3753	**0.4788**	9.6028	**3.1224**	0.3259	**0.1189**	0.9855	**0.0240**
Thatcher	99.4991	**0.5763**	5.1359	**1.4666**	0.1256	**0.0296**	0.9846	**0.0231**
La Prevision	99.3878	**0.4838**	8.6334	**0.3218**	0.1646	**0.0212**	1.5471	**0.2846**
Hilgendorf 61	99.2500	**0.5000**	7.9421	**3.3919**	0.2242	**0.0977**	1.0275	**0.0346**
Karamu	99.3752	**0.4788**	8.7740	**2.3244**	0.2386	**0.0751**	0.9980	**0.0406**

Table 5.2. The proportional contribution and significances of the four discriminants for special germination

	Proportion	Pr > F
1	0.8690	0.0001
2	0.0696	0.0043
3	0.0431	0.0498
4	0.0183	0.2490

Table 5.3. The correlation between the variables and the first discriminant (Between discriminant structure) and the standardized coefficients for special germination.

Variable	Correlation	Standardized Coeffients
A	-0.0743	0.6122
B	-0.2574	-4.4239
K	0.3754	4.7111
V	-0.9178	-1.4997

Table 5.4. Discriminant scores for the Richards functions for embryonic maturity in ten wheat cultivars and the t-test among the cultivars.

Cultivar	Discriminant score[1]	t-test
Brevor	14.8404	A
Sonora 64 A	7.8272	B
Gamenya	7.0266	B
Kenya 321	5.2393	B
Isis	3.9309	B
Hilgendorf 61	-0.5985	C
Thatcher	-0.8927	C
Karamu	-0.9278	C
Tordo	-2.6637	C
La Prevision	-25.2365	D

[1] Discriminant scores with the same letter are not significantly different

Figure 5.1. Diagrams of the mean Richards functions for embryonic maturity of ten wheat cultivars.

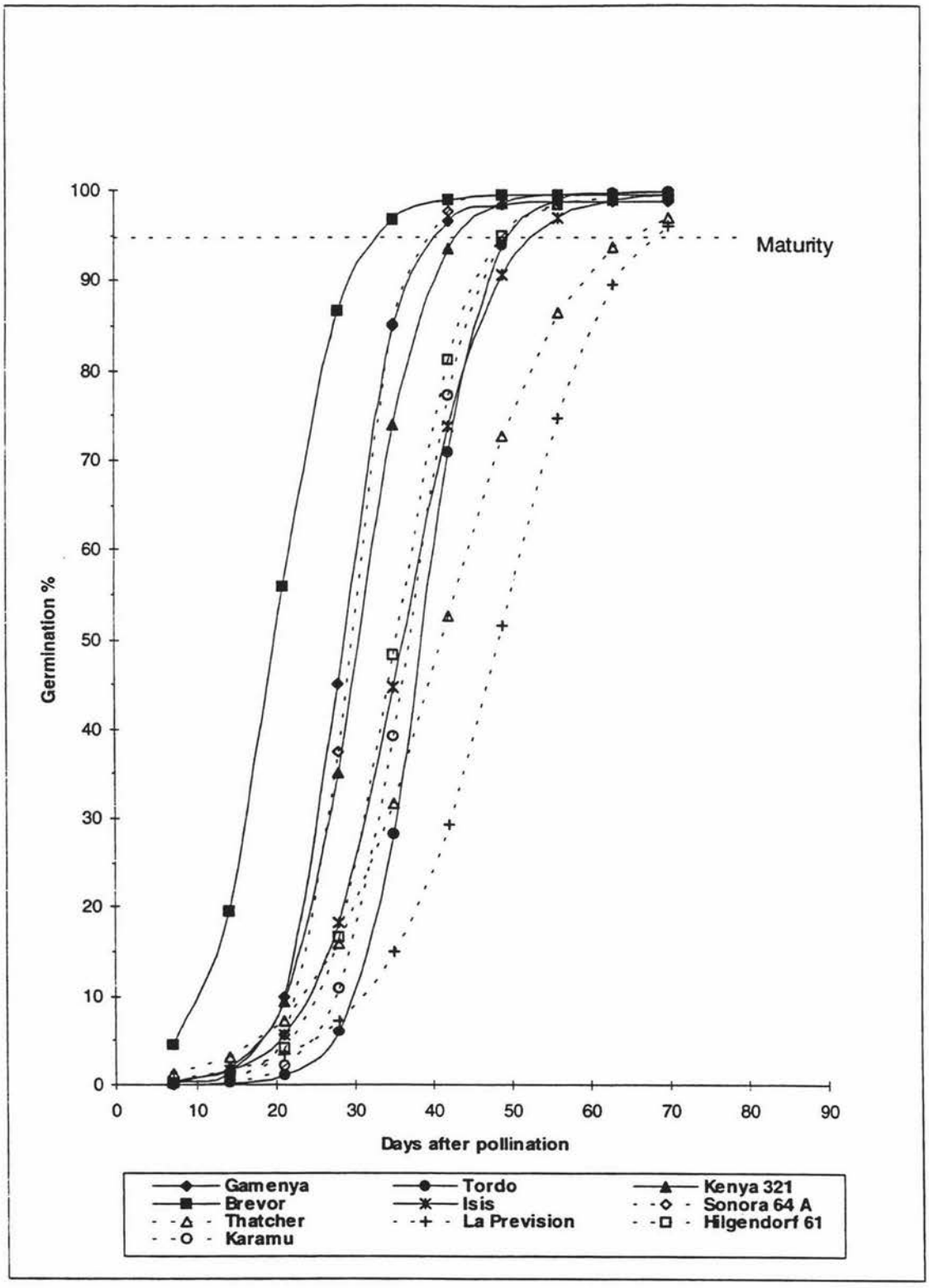


Table 5.5. The mean % embryonic maturity at harvest ripeness and significance symbols for ten wheat cultivars.

Cultivar	Mean[1]	t-test
Kenya 321	99.02	A
Gamenya	98.23	A
Sonora 64 A	97.56	AB
Tordo	97.09	AB
Karamu	94.44	AB
Brevor	93.79	AB
Isis	90.99	AB
Hilgendorf 61	89.06	AB
Thatcher	82.06	B
La Prevision	54.81	C

[1] Means with the same letter are not significantly different.

Table 5.6. Variance components estimates and heritability estimates for embryonic maturity at harvest ripeness for ten wheat cultivars and the ANOVA test for significance.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	176.9372	75.44630	0.589	0.12692	4.49	0.0006
Block	3	9.4350	5.96725				
Error	27	123.4984	32.43227				

Table 5.7. The mean days to median embryonic maturity and t - tests for differences amongst ten wheat cultivars

Cultivar	Mean[1]	t-test
La Prevision	46.76	A
Tordo	35.76	B
Thatcher	34.94	B
Karamu	34.26	B
Isis	31.95	BC
Hilgendorf 61	31.90	BC
Brevor	27.71	BCD
Sonora 64 A	27.67	BCD
Kenya 321	27.63	BCD
Gamenya	26.04	D

[1] Means with the same letter are not significantly different.

Table 5.8. Variance components and heritability estimates for days after pollination to median embryonic maturity of ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	37.2752	15.89419	0.730	0.10119	8.46	0.0001
Block	3	1.9265	1.21845				
Error	27	13.7891	3.62120				

Table 5.9. The mean number of days to full embryonic maturity and the t- test for differences amongst ten wheat cultivars

Cultivar	Mean[1]	t-test
La Prevision	52.77	A
Thatcher	40.17	B
Tordo	38.27	B
Karamu	37.16	B
Isis	35.60	BC
Hilgendorf 61	35.24	BCD
Brevor	31.67	CDE
Kenya 321	30.63	CDE
Sonora 64 A	29.94	DE
Gamenya	28.31	E

[1] Means with the same letter are not significantly different.

Table 5.10. Variance components estimates for number of days after pollination to median embryonic maturity for ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	49.93831	21.29376	0.782	0.08702	11.70	0.0001
Block	3	1.30538	0.82559				
Error	27	13.92988	3.65817				

Table 5.11. The mean differences between number of days to median embryonic maturity and harvest ripeness and t - tests for differences amongst ten wheat cultivars

Cultivar	Mean[1]	t - test
La Prevision	4.32	A
Thatcher	18.78	B
Isis	19.69	B
Hilgendorf 61	19.79	B
Tordo	19.92	B
Karamu	20.29	B
Sonora 64 A	22.97	BC
Gamenya	23.35	BC
Brevor	26.72	BC
Kenya 321	30.37	C

[1]Means with the same letter are not significantly different.

Table 5.12. Variance components and heritability estimates for the mean differences between number of days to median embryonic maturity and harvest ripeness for ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	46.2723	19.73058	0.580	0.12792	4.36	0.0007
Block	3	2.9556	1.86926				
Error	27	33.5124	8.80079				

Table 5.13. The mean differences between number of days to full embryonic maturity and harvest ripeness and t - tests for differences amongst ten wheat cultivars

Cultivar	Mean[1]	t - test
La Prevision	(1.69)[2]	A
Thatcher	13.55	B
Isis	16.04	BC
Hilgendorf 61	16.45	BC
Karamu	17.40	BC
Tordo	17.41	BC
Sonora 61 A	20.71	BCD
Gamenya	21.07	BCD
Brevor	22.77	CD
Kenya 321	27.37	D

[1] Means with the same letter are not significantly different.

[2] figures within parentheses: embryonic maturity occurred after harvest ripeness

Table 5.14. Variance components estimates for the mean differences between number of days to full embryonic maturity and harvest ripeness for ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	59.3008	25.28596	0.646	0.11875	5.64	0.0001
Block	3	2.1497	1.35957				
Error	27	32.4887	8.53194				

6. NORMAL GERMINATION

Normal germination estimated grain germinability under standard conditions, using distilled water as the imbibant and Whatman filter paper as substrate. The observed differences in normal germination amongst the cultivars suggested differences in levels of dormancy during development. The data observed were used to estimate the level of dormancy in the grains. This was defined as the difference between special and normal germination, expressed as a percentage of special germination. All the data used for estimating a dormancy data point were from the same sample.

6.1 Development profiles

Richards functions were fitted on data for normal germination and good fits were obtained. The coefficients of regression ranged from 0.9439 to 0.9966 (Tables A 5.2 (a) to A 5.2 (i) in Appendix 3). However, one cultivar, La Prevision, did not respond in the test and there are no function statistics to report. Dormancy for this cultivar was 100 %. Table 5.15 shows the mean values and standard errors of the estimates of statistics from the Richards functions and Figures A 5.2 (a) to A 5.2 (j) are graphs of the fitted functions and data points for each experimental unit.

6.2 Function analysis

Both the first and second discriminants passed the parsimony test and were retained in the analysis of the fitted functions. They were both significant at P = 0.0001. The third discriminant was significant at P = 0.01. Information about the discriminants, the structure matrix between the variables and the two discriminants and standardised coefficients for each of the variables are shown in Tables 5.16 and 5.17, respectively. The strongest correlations were between K and the first discriminant, and B and the second discriminant. A summary of the joint analysis of the structure matrix between the variables and the discriminants, and the standardized coefficients of the variables is given below:

1. The influence of the upper asymptote, A, on the first discriminant was suppressed but A had a medium influence on the second discriminant.
2. B had a weak influence on discriminant 1 but a strong influence on discriminant 2.
3. K had a strong influence on discriminant 1 and only a weak one on discriminant 2.
4. V had no influence on both discriminants.

Since the first discriminant contributed to variation in the function by more than twice the contribution of the second discriminant, the influence of K was more important than that of B. Small K values were associated with early and rapid responses in the normal germination test. The curves would have steep slopes and, therefore, would approach the upper asymptote rapidly. Large K values effected lazy, slow approaches to the upper asymptote. The expected horizontal shift of the function due to large B values was negated or, at least reduced, because of the minor contribution of the second discriminant to variation in the function.

Pairwise comparisons between the cultivars using the two discriminant scores are presented in Table 5.18, and show the strong discrimination between cultivars. Of note is cv. La Prevision which differed significantly from every other cultivar. Figure 5.2 shows diagrams of the mean Richards functions estimated for the ten cultivars. The curve for La Prevision is shown as a straight line on the horizontal axis for completeness.

Table 5.15. The mean values of the statistics estimated from the Richards functions for normal germination of ten wheat cultivars.

Attribute:	A		B		K		V	
Cv.	mean	**s.e. A**	mean	**s.e. B**	mean	**s.e. K**	mean	**s.e. V**
Gamenya	98.9197	**0.6478**	9.9312	**1.8998**	0.3126	**0.0467**	1.0667	**0.1065**
Tordo	98.8750	**0.6292**	11.0500	**0.2944**	0.2635	**0.0250**	1.0249	**0.1041**
Kenya 321	99.2290	**0.9362**	7.0502	**1.2354**	0.1635	**0.0251**	1.0136	**0.0612**
Brevor	98.8944	**1.0093**	11.0973	**1.1975**	0.2636	**0.0478**	0.9149	**0.0238**
Isis	99.9890	**0.0140**	10.1983	**1.2400**	0.2519	**0.0410**	1.0421	**0.1053**
Sonora 64 A	99.0088	**0.8083**	6.7658	**1.9044**	0.1627	**0.0637**	1.0298	**0.0960**
Thatcher	97.0001	**3.0274**	10.0751	**1.8822**	0.1510	**0.0408**	0.9498	**0.1077**
La Prevision	0.0000	**0.0000**	0.0000	**0.0000**	0.0000	**0.0000**	0.0000	**0.0000**
Hilgendorf 61	97.7306	**1.9861**	8.2248	**2.6666**	0.1635	**0.0629**	1.2359	**0.1312**
Karamu	99.1442	**0.7637**	8.9625	**2.5811**	0.1760	**0.0645**	1.0252	**0.2727**

Table 5.16. The proportional contribution of the four discriminants for the normal germination and their significances

Discriminant	Proportion	Pr > F
1	0.5666	0.0001
2	0.2458	0.0001
3	0.1438	0.0038
4	0.0438	0.1388

Table 5.17. The correlation between the variables and the first discriminant (Between discriminant structure) for nomal germination.

	Dicriminant 1		**Discriminant 2**	
Variable	Correlation	Standardized coefficient	Correlation	Standardized coefficient
A	0.6227	0.3032	0.4757	1.3935
B	0.2260	-1.7459	0.6856	2.1744
K	0.8608	2.8911	0.2991	-1.3957
V	0.0604	-0.1823	-0.3223	-0.1783

Table 5.18. Pair - wise comparisons of ten wheat cultivars using the first two discriminants for normal germination

Cultivar	Gam	Tordo	K321	Brevor	Isis	Sonora	Thatch	La P	Hil61	Karam
Gamenya	----------	(*)	*	(*)	*	**	*	***	***	ns
Tordo	6.52619	----------	***	ns	ns	***	***	***	***	*
Kenya 321	23.995	57.038	----------	***	***	ns	ns	***	ns	*
Brevor	7.55737	-0.034	61.188	---------	ns	***	***	***	***	*
Isis	15.5442	1.9195	80.121	1.2483	---------	***	***	***	***	*
Sonora 64 A	38.4962	78.1	1.6311	82.86	104.72	---------	ns	***	ns	*
Thatcher	22.0706	54.152	0.0354	58.225	76.712	2.15	----------	***	ns	*
La Prevision	95519.6	97120	92452	97276	97988	91677	92568	---------	ns	ns
Hilgendorf 61	48.6751	91.407	3.3727	96.085	119.86	0.006	4.1196	1.73	--------	**
Karamu	0.45862	13.19	15.305	15.288	25.373	26.96	13.842	0.004	34.43	---------

Significance symbols in upper triangle are explained below:

ns : no significance difference (P > 0.1)

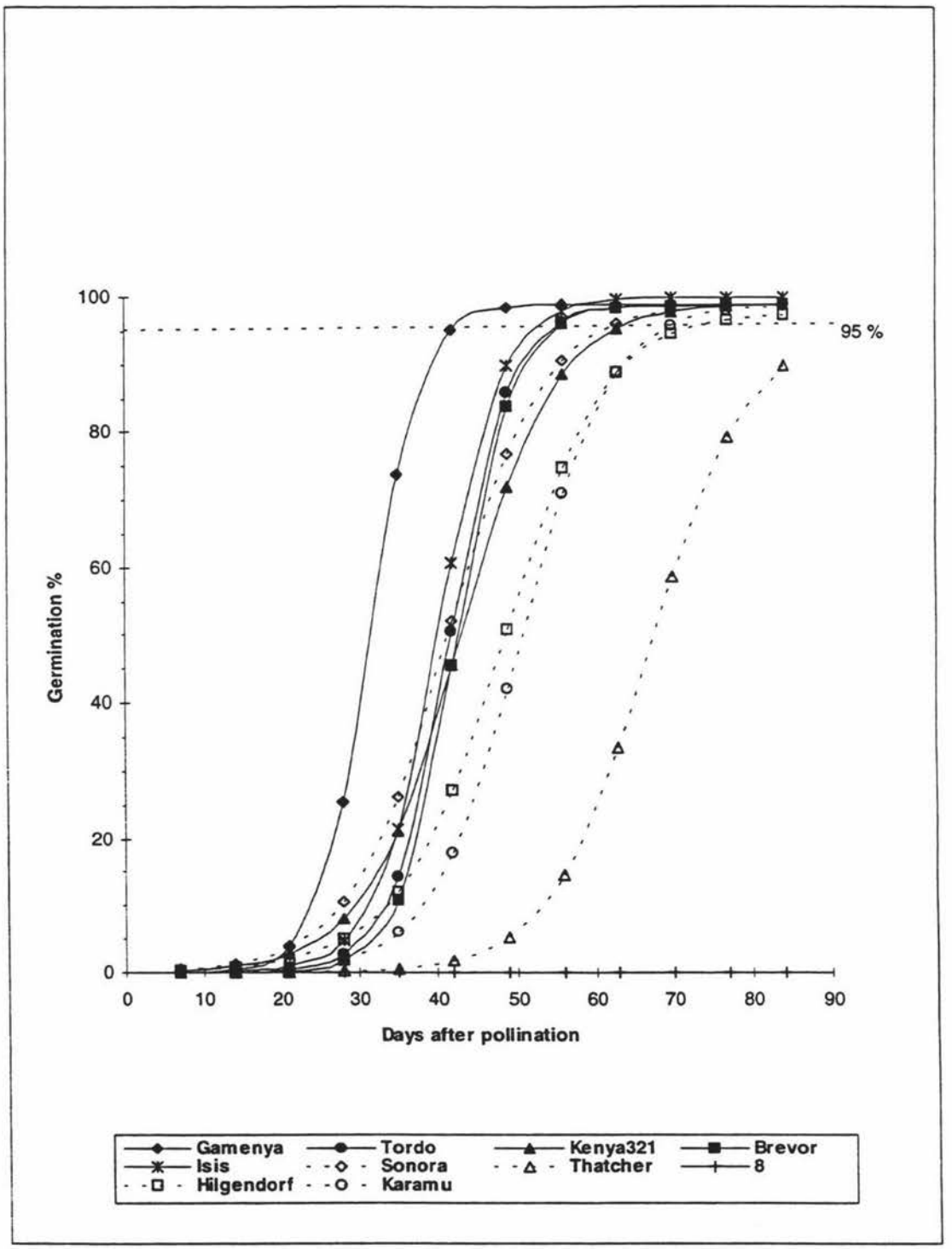
(*) : 0.1 > P > 0.05

* : 0.05 > P > 0.01

** : 0.01 > P > 0.005

*** : 0.005 > P

Figure 5.2. Diagrams of the mean Richards functions for normal germination of ten wheat cultivars.



7. DORMANCY

Dormancy was estimated, in this study, from the difference between the special and normal germination measurements. Both these measures were done on sub - samples taken from the same harvest.

7.1 Development profile for grain dormancy

In fitting the Richards functions, the upper asymptote, A, was set at 100 % because young grains from the ten cultivars did not germinate under the conditions of the special germination test indicating immaturity. Estimates of the statistics obtained from the Richards functions for dormancy levels are shown in Tables A 5.3 (a) to A 5.3 (j) in Appendix 3. The estimates of statistics and standard errors for the mean Richards functions are given in Table 5.19. Graphs of the fitted functions and data points for each experimental unit are shown in Figures A 5.3 (a) to A 5.3 (j). The functions were used to estimate the level of dormancy at harvest ripeness and dormancy levels at times of agronomic interest. The ordinary lsd test was used to test for significant differences among the cultivars. Secondary variables estimated were:

Mnemonic	Variable description
1. **DHR**	Dormancy at harvest ripeness
2. **T50D**	Number of days from pollination for dormancy level to decline to 50 %
3. **T5D**	Number of days from pollination for dormancy level to decline to 5 % dormancy
4. **HR-T50D**	Number of days between harvest ripeness and days to 50 % dormancy
5. **HR-T5D**	Number of days between harvest ripeness and days to 5 % dormancy

7.2 Analysis of the dormancy function

Dormancy in the developing wheat embryos was described by a declining function, having negative values for the statistics B and K, in all the cultivars except La Prevision in which dormancy remained at 100 % throughout the period of the experiment. The discriminant correlation was very strong for the first three

discriminants but moderate for the fourth discriminant. The first three discriminants were all highly significant ($P < 0.0001$) but the fourth was not (Table 5.20). The first discriminant accounted for almost all of the variation in the function and, therefore, the other three were not considered in subsequent analyses. The correlation between the variables and the discriminant standardized coefficients are given in Tables 5.21.

The following results were obtained from the discriminant correlational analysis:

1. The upper asymptote, A, had no influence on the discriminant
2. The inflexion placement factor, V, had a strong influence on the discriminant
3. Enhancement occurred for the effect of B, and
4. K was a pseudo variable.

Large positive values of V were associated with slowly declining functions, therefore, enhancing the effect of large (less negative) B values. Cultivars having such functions lost dormancy slowly in comparison to those having functions with small V's and small B's. The first discriminant described the shape of the function by influencing dormancy release and may be succinctly described as an 'attenuation' factor.

The discriminatory power of the first discriminant was used to separate the cultivars into significantly different groups using the t-test at the 5 % significance level. The discriminant separated the cultivars into two overlapping groups (Table 5.22). La Prevision was left out of the test because only one statistic, A, was available for this cultivar. As observed above, the upper asymptote was unimportant in discriminating amongst the cultivars. The biggest influence came from V (with a high positive standardized coefficient) although the pseudo effect of K was apparent on examining the curves for Thatcher and Gamenya (Figure 5.3).

Table 5.19. The mean values of the statistics estimated from the Richards functions for grain dormancy of ten wheat cultivars.

Attribute:	A		B		K		V	
Cv.	mean	**s.e. A**	mean	**s.e. B**	mean	**s.e. K**	mean	**s.e.V**
Gamenya	100.4593	**0.536**	-6.950	**3.018**	-0.250	**0.091**	0.9330	**0.057**
Tordo	99.7214	**0.557**	-8.918	**0.848**	-0.224	**0.049**	0.9465	**0.007**
Kenya 321	100.4819	**0.716**	-4.813	**1.904**	-0.123	**0.034**	0.9457	**0.067**
Brevor	99.3862	**0.797**	-9.450	**0.711**	-0.249	**0.057**	0.9732	**0.021**
Isis	100.5815	**1.528**	-5.120	**0.631**	-0.150	**0.041**	0.9636	**0.010**
Sonora 64 A	100.0000	**0.000**	-4.825	**1.645**	-0.113	**0.025**	0.9625	**0.048**
Thatcher	100.2482	**0.493**	-9.094	**1.260**	-0.137	**0.025**	0.9985	**0.004**
La Prevision	100.0000	**0.000**	0.000	**0.000**	0.000	**0.000**	0.000	**0.000**
Hilgendorf 61	100.8717	**0.842**	-4.141	**2.472**	-0.077	**0.033**	0.9200	**0.086**
Karamu	98.9648	**2.132**	-6.850	**1.769**	-0.138	**0.048**	0.9728	**0.024**

Figure 5.3. Diagrams of the mean Richards functions for grain dormancy of ten wheat cultivars.

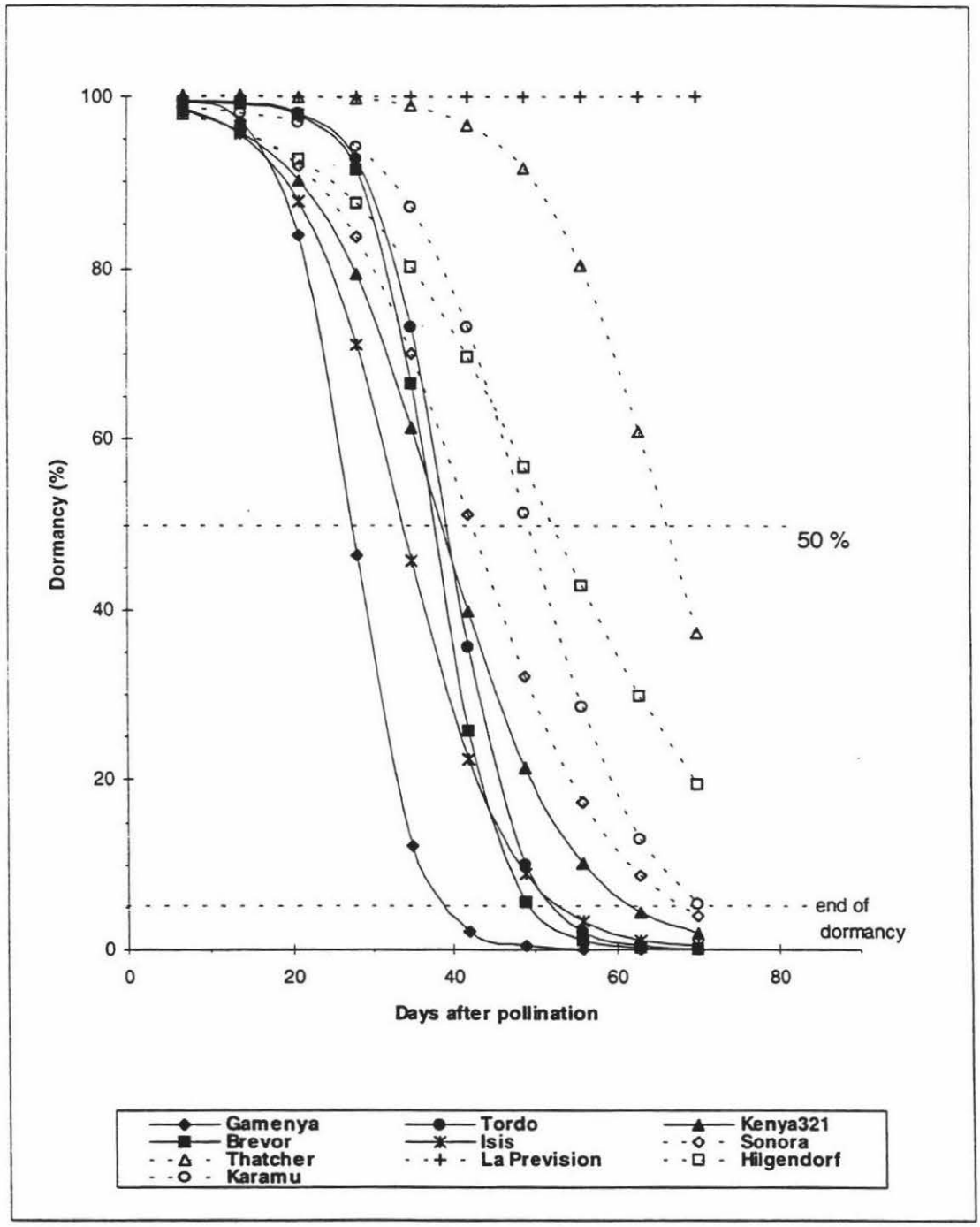


Table 5.20. Properties of the four discriminants for the Richards function statistics for dormancy

Discriminant	Proportion	P > F
1	0.9760	0.0001
2	0.0158	0.0001
3	0.0071	0.0001
4	0.0011	0.1851

Table 5.21. The correlation between the variables and the first discriminant (structure) and the standardized coefficients for the variables for the dormancy function.

Variable	Correlation	Standardized coefficient
A	0.1091	0.1453
B	-0.6384	8.0852
K	-0.6423	-5.0955
V	0.9743	17.0716

Table5.22. Discriminant scores for the Richards functions of dormancy levels of ten wheat cultivars and the t - groups amongst the cultivars.

Cultivar	Discriminant score	t - test
Isis	87.6377	A
Sonora 64 A	81.0222	B
Karamu	74.1015	C
Brevor	74.0064	C
Thatcher	71.6304	CD
Kenya 321	70.8015	CD
Gamenya	68.9654	D
Tordo	53.4686	E
Hilgendorf 61	47.6605	F

7.3 Dormancy at harvest ripeness (DHR)

The level of dormancy at harvest ripeness is useful information for both the plant breeder and the farmer. The plant breeder may use such information in selection programs whereas, for the farmer, the information enables him/her to predict the potential risk (to preharvest sprouting) he/she takes for delaying the harvest.

Significant differences for the level of dormancy at harvest ripeness were observed amongst the cultivars at the 5 % level of significance (Table 5.23). Four groups were identified and one of the cultivars, Karamu, belonged to two of the groups There were no significant differences between Thatcher and La Prevision, the two most dormant red - grained wheats, at harvest ripeness. All the white - grained wheat cultivars fell in one significance group. Isis, Kenya 321 and Brevor had very weak dormancy at harvest ripeness but the dormancy levels in both Gamenya and Tordo were trivial (less than 5 %). Hilgendorf 61 had lost 50 % of its dormancy at the time of harvest ripeness. Sonora 64 A had a significantly lower level of dormancy at harvest ripeness than 3 of the red - grained wheats but was not different from Karamu.

The dormancy level estimates at harvest ripeness were obtained by interpolation, using the observed harvest ripeness data as the independent variable, X (time), in the Richards function for each individual experimental unit. This is the usual method for estimation of the level of growth and development variables at harvest ripeness.

7.3.1 Heritability estimates for dormancy at harvest ripeness (DHR)

The variance components and heritability estimates for DHR are shown in Table 5.24. The ANOVA test for significance and the genotypic variance component were both highly significant ($P < 0.0001$). A high heritability estimate was observed in DHR. This variable is, therefore, useful and can be used in selection programs.

7.4 Number of days from pollination for the dormancy level to decline to 50 % (T50D)

The analysis of variance was highly significant ($P < 0.0001$), as was the genotypic variance component. Three overlapping significance groups and two individual

cultivars, at either end of the groups, were separated at the 5 % level of significance (Table 5.25). Only 9 cultivars were considered in this variable because La Prevision remained with a 100 % dormancy level throughout the period of the study. For this reason, La Prevision was different from all the other cultivars. The mean values for T50D in Table 7.7 show that the rate of dormancy release was faster in Gamenya than in all the other cultivars. Thatcher had the least rate of dormancy release amongst the 9 cultivars considered and La Prevision did not lose any of its dormancy at all.

7.4.1 Heritability estimates for number of days from pollination for the dormancy level to decline to 50 % (T50D)

A high estimate of heritability for T50D was found (Table 5.26). It was lower than the heritability estimate for DHR by only 5 %. The genotypic variance component for this variable was highly significant ($P < 0.0001$). Fifty percent, in the fitted Richards functions for dormancy, represents the point of inflexion for the function when the rate of dormancy release is at its maximum. After this point, the rate slows down. Therefore. T50D is an interesting and, perhaps, useful variable to measure.

7.5 Number of days from pollination to 5 % dormancy (T5D)

This variable may be considered to represent the time it takes for the grain to lose all of its original dormancy and can provide useful information about residual dormancy at harvest ripeness. It is an important variable, therefore, because it shows the total time period, during growth and development, and after harvest ripeness, during which the grain is protected against preharvest sprouting. Table 5.27 shows the mean number values of T5D and comparisons using the t-test at the 5 % significance level for nine of the cultivars. La Prevision does not appear in Table 7.9 for the same reasons mentioned above. Four significance groups were observed. Hilgendorf 61 and Thatcher were similar and took the longest time to lose dormancy compared to all the other cultivars. Combining Table 4.6 in Section 4 (mean harvest ripeness) and Table 7.9, Hilgendorf 61 had 39 days and Thatcher 35 days of useful dormancy after harvest ripeness. Gamenya had lost all its dormancy 40 days after pollination, well before harvest ripeness (mean HR = 52.54 days after pollination).

7.5.1 Heritability estimates for number of days from pollination to 5 % dormancy

Table 5.28 shows the variance components and the estimated heritabilities for number of days from pollination to 5 % dormancy. The genotypic variance component was highly significant (P = 0.0001). Estimated heritabilities for T5D were lower than for T50D or DHR but were still very high showing that it is a useful variable for use by plant breeders and farmers alike.

Table 5.23. The mean levels of dormancy at harvest ripeness and t-tests.

Cultivar	Mean[1]	t-group
La Prevision	100.00	A
Thatcher	83.51	A
Hilgendorf 61	49.02	B
Karamu	35.27	BC
Sonora 64 A	29.13	C
Isis	11.20	D
Kenya 321	7.75	D
Brevor	5.16	D
Tordo	3.37	D
Gamenya	0.59	D

[1]Means with the same letter are not significantly different.

Table 5.24. Variance components and heritability estimates for % dormancy at harvest ripeness.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	1236.71619	527.33759	0.890	0.04937	24.42	0.0001
Block	3	6.0071	3.79924				
Error	27	152.57221	40.06741				

Table 5.25. The mean number of days from pollination to 50 % dormancy for nine wheat cultivars.

Cultivar	Mean[1]	t group
Thatcher	71.95	A
Hilgendorf 61	59.78	B
Karamu	56.33	BC
Sonora 61 A	48.43	CD
Kenya 321	43.84	D
Tordo	43.44	D
Brevor	41.80	D
Isis	40.03	D
Gamenya	29.95	E

[1]Means with the same letter are not significantly different.

Table 5.26. Variance components and heritability estimates for number of days from pollination to 50 % dormancy.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	8	207.7710	92.91805	0.841	0.07117	11.70	0.0001
Block	3	3.0956	1.95783				
Error	27	39.4012	10.79203				

Table 5.27. The mean number of days from pollination to 5 % dormancy for nine wheat cultivars and test for significant differences.

Cultivar	Mean[1]	t-group
Hilgendorf 61	90.69	A
Thatcher	89.22	A
Karamu	74.01	B
Sonora 64 A	68.67	B
Kenya 321	62.44	BC
Isis	55.75	C
Tordo	53.46	C
Brevor	51.20	CD
Gamenya	39.39	D

[1] Means with the same letter are not significantly different.

Table 5.28. Variance components estimates for the number of days from pollination to 5 % dormancy.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	8	301.5808	134.87103	0.813	0.07997	12.88	0.0001
Block	3	6.4796	4.09809				
Error	27	69.32883	18.20666				

7.6 The difference between harvest ripeness and number of days from pollination to 50 % dormancy (HR-T50D)

This time segment represents the stage when the embryo is nearing maturity and dormancy levels are declining but at a slower rate than before. Again, La Prevision was left out from the analysis for reasons mentioned above. The mean values for HR-T50D and the t-test significane groups are shown in Table 5.29. All the white - grained cultivars formed one significance group and T50D was smaller than harvest ripeness (positive values). Only one of the red - grained cultivars had a positive value for HR-T50D, the others having negative values.

7.6.1 Heritability estimates for the difference between harvest ripeness and days from pollination to 50 % dormancy

Tables 5.30 shows the variance components estimates and heritability estimates, respectively, for HR-T50D. The ANOVA significance test was highly significant at (P = 0.0001). The heritability estimates for this variable were quite high and explained 78.9 % of the phenotypic variation. This variable, however, may be of academic interest only since it is estimated from variables that are already useful and easier to measure.

7.7 The difference between harvest ripeness and number of days from pollination to 5 % dormancy (HR-T5D)

This variable measures the length of time, after harvest ripeness, that the grain has some protection against sprouting, i.e. residual dormancy at harvest ripeness. Farmers usually are able to locate the harvest ripeness stage of the grain but may not be in a position to harvest the grain immediately. Information about the length of time available, after harvest ripeness, before the grain loses all its dormancy is very useful information agronomically. Plant breeders can use information about HR-T5D in selection programs.

The mean values and t-tests for HR-T5D are shown in Table 5.31. Negative values indicate the presence of a residual dormancy at harvest ripeness. The tests for significant differences amongst the cultivars revealed four groups, two of them overlapping. The white cultivars did not have residual dormancy at harvest ripeness.

7.7.1 Heritability estimates for the difference between harvest ripeness and number of days from pollination to 5 % dormancy

The variance components and the heritability estimates for HR-T5D are shown in Table 5.32. The heritability estimates were only slightly higher than those for HR-T50D. Because HR-T5D is easier to measure than HR-T50D and it shows similar heritability estimates it should be the preferred method for measuring the 'window' of protection against sprouting damage *after* harvest ripeness.

Table 5.29. The difference between harvest ripeness and number of days from pollination to 50 % dormancy of ten wheat cultivars.

Cultivar	Mean[1]	t - group
Thatcher	18.23	A
Hilgendorf 61	8.09	B
Karamu	1.78	BC
Sonora 64 A	-2.22	C
Isis	-11.61	D
Tordo	-12.24	D
Brevor	-12.63	D
Kenya 321	-14.17	D
Gamenya	-19.44	D

[1]Means with the same letter are not significantly different. Negative values show cultivars where 50 % dormancy occurred before harvest ripeness.

Table 5.30. Variance components and heritabilty estimates for the difference between harvest ripeness and number of days from pollination to 50 % dormancy of ten wheat cultivars.

Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
8	151.4277	67.72053	0.789	0.08779	4.36	0.0007
3	0.9255	0.58532				
27	40.4808	10.63079				

Table 5.31. The difference between harvest ripeness and number of days from pollination to 5 % dormancy of ten wheat cultivars.

Cultivar	Mean[1]	t-group
Hilgendorf 61	39.00	A
Thatcher	35.49	A
Karamu	19.46	B
Sonora 64 A	18.03	B
Kenya 321	4.43	C
Isis	4.11	C
Tordo	-2.22	CD
Brevor	-3.23	CD
Gamenya	-10.00	D

[1]Means with the same letter are not significantly different. Negative values show that dormancy was lost before harvest ripeness.

Table 5.32. Variance components and heritability estimates for the difference between harvest ripeness and number of days from pollination to 5 % dormancy of ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	8	301.1776	134.69070	0.799	0.08470	11.69	0.0001
Block	3	4.0668	2.57204				
Error	27	75.8825	19.92773				

6. GRAINCOAT COLOUR

Two types of graincoat are recognized in wheat, red and white. The interest in the colour of wheat arises because colour has been associated with resistance to sprouting.

6.1 Development profiles for graincoat colour

Colour development showed a sigmoidal developmental pattern. Richards functions were found appropriate. For each experimental unit, the fits were excellent (Appendix 4, Tables A 6.1 (a) to A 6.1 (j)), and graphs of the fitted functions and observed data points agreed well (Figures A 6.1 (a) to A 6.1 (j), in Appendix 4). Estimates of statistics from the mean Richards functions for each cultivar are shown in Table 6.1, and these estimates were used to plot the diagrams for the mean Richards functions in Figure 6.1. Height differences between the red - and white - grained cultivars are indicated in these diagrams.

Secondary variables at points and levels of interest were estimated using the Richards functions. The following is a list of these variables:

Mnemonic	Description of variable
COLHR	Graincoat colour score at harvest ripeness
T95COL	Number of days from pollination to maximum colour score (colour maturity)
HR-T95COL	Difference between harvest ripeness and number of days to 95 % colour intensity
$COLGR_0$	Initial growth rate of colour development
MEANGRCOL	Mean growth rate of colour development

6.2 Function analysis

The manova for the graincoat colour function was highly significant ($P < 0.0001$) and, from the univariate anaylses of variance, the upper asymptote, A, showed highly significant differences amongst the cultivars ($P < 0.0001$).

moderate correlations and were insignificant at 10 % (Table 6.2). The first discriminant contributed 85.78 % of the total variation in the model and was sufficient to explain the parsimonious model. The discriminant structure between the first discriminant and its variables and the standardized coefficients for the variables are shown in Table 6.3. Using this information the following results were obtained:

1. The upper asymptote (A) had a strong influence on the first discriminant
2. Both B and V were unimportant in the first discriminant
3. K had a weak influence on the first discriminant.

Based on these results, the emphasis of discrimination amongst the cultivars was found to be on intensity of colour at maturity and on growth rate. Cultivars having large A values were more red than those with small A values. All the red - grained cultivars had large A values. Variation was apparent amongst the white - grained varieties, in colour intensity (A) and, also, in the profiles of the functions (Figure 6.1).

Discrimination amongst the cultivars was measured using the t-test at the 0.05 level of significance (Table 6.4). Three groups, two of them overlapping, and a single cultivar were separated from each other. The overlap occurred within the white group of cultivars. There was a clear division between the red and white cultivars. However, variation was observed amongst the red varieties since Hilgendorf 61 was separated from the other four cultivars

Table 6.1. Estimated statistics for the mean Richards functions for graincoat colour of developing grain of ten wheat cultivars.

Statistic	A		B		K		V	
Cultivar	mean	**s.e. A**	mean	**s.e. B**	mean	**s.e. K**	mean	**s.e. V**
Gamenya	1.9678	**0.1873**	7.3020	**1.2019**	0.2369	**0.0631**	0.9487	**0.0706**
Tordo	2.2530	**0.5886**	5.2736	**1.8402**	0.2201	**0.0628**	0.9564	**0.0962**
Kenya 321	2.4496	**0.3420**	7.7405	**2.7578**	0.1999	**0.0816**	0.8183	**0.1034**
Brevor	2.2075	**0.3752**	4.8102	**0.8587**	0.1225	**0.0269**	0.7316	**0.1450**
Isis	2.1503	**0.5327**	7.2632	**2.5884**	0.2125	**0.0853**	0.7846	**0.1233**
Sonora 64 A	4.0033	**0.0817**	7.0374	**3.1713**	0.2403	**0.1049**	0.7262	**0.3822**
Thatcher	4.2248	**0.9142**	8.4635	**1.6846**	0.2550	**0.0451**	0.8129	**0.1548**
La Prevision	4.0242	**0.4843**	8.6985	**1.7588**	0.2673	**0.0533**	0.8513	**0.1295**
Hilgendorf 61	5.0750	**0.2062**	8.0625	**0.4270**	0.2250	**0.0289**	0.9395	**0.0895**
Karamu	4.3104	**0.5750**	4.5245	**0.6977**	0.1501	**0.0401**	0.8148	**0.1144**

Figure 6.1 Diagrams of the mean Richards functions for graincoat colour of ten wheat cultivars showing the harvest ripeness for each cultivar.

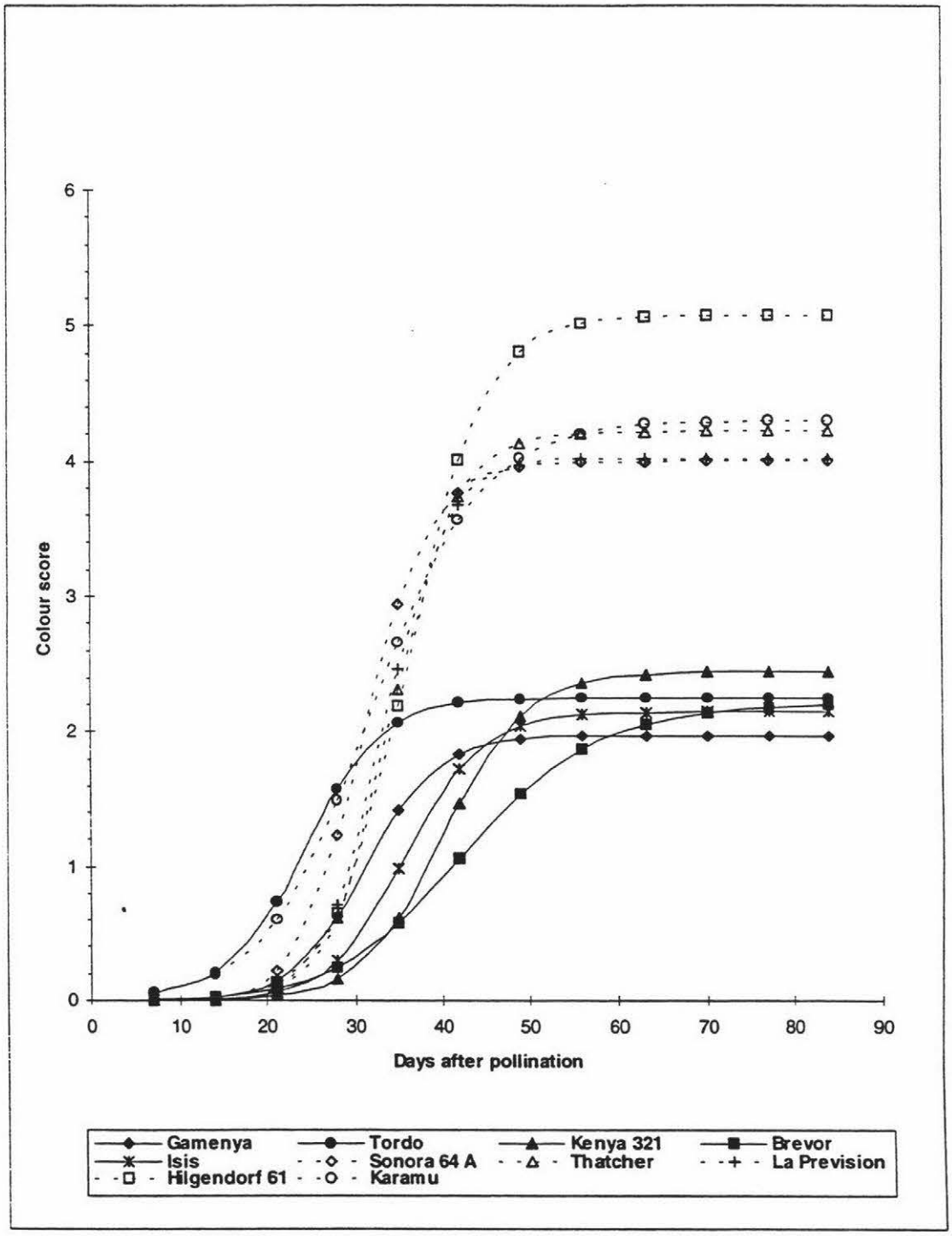


Table 6.2. Properties of the four discriminants for the Richards function statistics for graincoat colour.

Discriminant	Proportion	Pr > F
1	0.8578	0.0001
2	0.0867	0.0047
3	0.0362	0.1043
4	0.0193	0.2563

Table 6.3. The correlation between the variables and the first discriminant (structure) for graincoat colour.

Variable	Correlation	Standardized coefficient
A	0.9857	3.1848
B	0.3146	-0.9440
K	0.3396	1.4912
V	-0.0747	-0.8643

Table 6.4. Discriminant scores for the Richards functions estimated for graincoat colour development in ten wheat cultivars and tests for significant differences

Cultivar	Discriminant score[1]	t - group
Hilgendorf 61	21.2373	A
Karamu	12.9012	B
Thatcher	12.8220	B
Sonora 64 A	11.6588	B
La Prevision	10.2065	B
Kenya 321	-11.5183	C
Tordo	-12.1616	C
Isis	-13.9030	CD
Brevor	-14.3563	CD
Gamenya	-16.8867	D

[1] discriminant scores with the same letter are not significantly different at P = 0.05

6.3 Graincoat colour at harvest ripeness (COLHR)

Graincoat colour is one of the developmental characters used by farmers to spot the harvest ripeness stage of growth and development. The importance of colour identification in wheat arises, primarily, from the need to separate white - grained wheats from red - grained wheats because their uses in manufacturing differ.

Differences in the intensity of graincoat colour were observed between the red wheat and white wheat cultivars, as well as within the red wheat cultivars, investigated (Table 6.5). The white wheats formed one significance group, indicating that any observed differences in colour intensity, within this group, were due to chance alone. Two overlapping groups were identified within the red - grained cultivars.

6.3.1 Variance estimates and heritability estimates for graincoat colour

Table 8.6 shows the variance estimates and the heritability estimates for graincoat colour intensity at harvest ripeness. The ANOVA F test was highly significant ($P < 0.0001$). The heritability estimates for graincoat colour at harvest ripeness were very high ($h^2 = 0.852$).

6.4 Number of days from pollination to colour maturity (T95COL)

This variable represented colour maturity. To the farmer, knowledge of when the grain is expected to reach grain maturity (T95COL), in specific environments, may be useful for the scheduling of activities at harvest time.

Variation, amongst the ten cultivars, was observed in T95COL. Table 6.7 shows three overlapping significance groups for this variable. Brevor took the longest to reach colour maturity and Tordo, the least. A moderate heritability estimate for T95COL was found ($h^2 = 0.378$, Table 6.8).

6.5 Initial growth rate of graincoat colour development ($COLGR_0$)

This variable was estimated because of its possible connection with later developmental events. Graincoat colour formation, in wheat, is an oxygen - demanding process which may affect the growth of internal tissues by depriving

them of sufficient oxygen. The initial rate of colour formation in the graincoat may, therefore, provide important information, e.g., the oxygen consumption rate of the graincoat.

Table 6.9 shows the initial growth rates of colour development of the ten wheat cultivars. Two overlapping significance groups were found. There was no clear distinction between the red - grained cultivars and the white - grained cultivars such as was found for level of colour at harvest ripeness (Section 6.4). It seems, therefore, that initial growth rate of colour development did not influence with the final intensity of redness in these wheats. The reddest cultivar, Hilgendorf 61, had a medium starting growth rate.

Low heritability estimates (h^2 = 0.262) were obtained for $COLGR_0$ in the single environment used. These estimates are shown in Table 6.10.

6.6 Mean growth rate of graincoat colour development (MEANGRCOL)

There was greater separation amongst the cultivars for the mean growth rate (Table 6.11) than for the initial growth rate of colour development. Three overlapping groups were found. In general, the pattern found for $COLGR_0$ was maintained for MEANGRCOL. The mean growth rates of colour development in Karamu and Brevor were lower than those of La Prevision and Thatcher.

Moderate heritability estimates (h^2 = 0.299) were found for MEANGRCOL (Table 6.12) indicating that this variable could find use in breeding programs, where it may be combined with the duration (h^2 = 0.378) to colour maturity, to select for early maturing cultivars having high levels of dormancy at harvest ripeness.

6.7 The difference between duration of colour formation and harvest ripeness (HR - T95COL)

It was suggested in Section 6.5, above, that this variable could be more meaningful to the farmer than T95COL. The reason for this is that knowledge of HR-T95COL, within specific environments, can assist the farmer in locating the harvest ripeness stage with great accuracy. Indeed, it may be more useful to know on which side of harvest ripeness colour maturity occurs than to know the absolute duration of colour formation.

The mean number of days between harvest ripeness and the timing of colour maturity are shown in Table 6.13. Colour maturity occurred after harvest ripeness in Brevor and Kenya 321 but before harvest ripeness in the rest of the cultivars. In Kenya 321, Isis and Gamenya, colour maturity occurred within a day of harvest ripeness. In these cultivars, therefore, colour maturity is a good indicator of harvest ripeness under the environment used in this study. Colour maturity was achieved 14 days before harvest ripeness, in Tordo, and ten days before harvest ripeness in Thatcher. Therefore, in these cultivars, colour on its own is a poor indicator of harvest ripeness.

Although three overlapping significance groups were found (Table 6.13), sub - groups are reported here in order to show the within group variations. Thus, Brevor occupied sub - group **A**; Kenya 321, Isis, and Gamenya were in sub - group **AB**; Hilgendorf 61, in sub - group **ABC**; Karamu, La Prevision, Sonora 64 A and Thatcher, in sub - group **BC**; and, Tordo, in sub - group **C**. With the exception of Tordo, which reached colour maturity well before harvest ripeness, the red cultivars achieved colour maturity earlier than the white cultivars. Other than this general pattern, there appears to be no further information to be gained from Table 6.13.

Variance components estimates and heritability estimates for HR-T95COL are shown in Tables 6.14. A moderate heritability estimate ($h^2 = 0.322$) was obtained. The environment has a large influence on this variable but the genotypic effect may be sufficient to enable reasonable genetic gains in breeding programs.

Table 6.5. The mean colour score at harvest ripeness and tests for significant differences amongst ten wheat cultivars (five red - and five white - grained).

Cultivar	Mean[1]	t-group
Hilegndorf 61	4.8025	A
Sonora 64 A	4.6125	AB
Thatcher	4.1800	AB
Karamu	4.0250	B
La Prevision	3.9100	B
Tordo	2.2450	C
Kenya 321	2.1975	C
Isis	1.9500	C
Gamenya	1.8675	C
Brevor	1.6900	C

[1] Means with the same letter are not significantly different.

Table 6.7. Variance components and heritability estimates for graincoat colour score of five red - and five white - grained wheat cultivars at harvest ripeness.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	1.5796	0.67355	0.852	0.06406	17.47	0.0001
Block	3	0.0286	0.01806				
Error	27	0.2754	0.07232				

Table 6.7. The mean number of days from pollination to colour maturity of five red - and five white - grained wheat cultivars.

Cultivar	Mean[1]	t-group
Brevor	62.74	A
Kenya 321	58.35	AB
Isis	51.46	ABC
Hilgendorf 61	48.658	BC
Gamenya	48.54	BC
Karamu	48.31	BC
Thatcher	43.30	C
La Prevision	43.26	C
Sonora 64 A	42.46	C
Tordo	40.55	C

[1] Means with the same letter are not significantly different.

Table 6.8. Variance components and heritability estimates for number of days from pollination to colour maturity of five red - and five white - grained wheat cultivars at harvest ripeness.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	51.3428	21.89263	0.378	0.13040	2.13	0.0507
Block	3	10.1328	6.40857				
Error	27	84.3996	22.16442				

Table 6.9. The initial growth rate of graincoat colour development in five red - and five white - grained wheat cultivars.

Cultivar	Mean[1]	t-group
Thatcher	0.32400	A
La Prevision	0.31955	A
Isis	0.27555	AB
Sonora 64 A	0.25785	AB
Gamenya	0.25420	AB
Kenya 321	0.25145	AB
Hilgendorf 61	0.24010	AB
Tordo	0.23980	AB
Karamu	0.18280	B
Brevor	0.17033	B

[1] Means with the same letter are not significantly different.

Table 6.10. Variance components and heritability estimates for the initial growth rate of graincoat colour in five red - and five white - grained wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	0.0025	0.00105	0.262	0.11413	1.14	0.3689
Block	3	0.0002	0.00014				
Error	27	0.0069	0.00182				

Table 6.11. The mean growth rate of colour development in the graincoats of five red - and five white - grained wheat cultivars.

Cultivar	Mean[1]	t-group
La Prevision	0.14480	A
Thatcher	0.14158	A
Sonora 64 A	0.12433	AB
Gamenya	0.12248	AB
Isis	0.11930	ABC
Hilgendorf 61	0.11595	ABC
Tordo	0.11475	ABC
Kenya 321	0.11115	ABC
Karamu	0.08215	BC
Brevor	0.07078	C

[1] Means with the same letter are not significantly different.

Table 6.12. Variance components and heritability estimates for the mean growth rate of graincoat colour development in five red - and five white - grained wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	0.0005	0.00023	0.299	0.12196	1.40	0.2265
Block	3	0.0001	0.00004				
Error	27	0.0012	0.00033				

Table 6.13. The mean difference between harvest ripeness and the duration of colour formation in ten wheat cultivars.

Cultivar	Mean[1]	t- group
Brevor	-8.31[2]	A
Kenya 321	-0.34	A
Isis	0.18	AB
Gamenya	0.85	AB
Hilgendorf 61	3.03	AB
Karamu	6.24	ABC
La Prevision	7.83	BC
Sonora 64 A	8.19	BC
Thatcher	10.42	BC
Tordo	15.13	C

[1] Means with the same letter are not significantly different.
[2] Negative values indicate that colour maturity was attained after harvest ripeness

Table 6.14. Variance components estimates for the mean difference between harvest ripeness and the duration of colour formation in ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	44.1214	18.81343	0.322	0.12375	1.77	0.1058
Block	3	12.3504	7.81105				
Error	27	92.1385	24.19677				

7. FLAVANOLS

Colour formation in wheat is due to the polymerisation of flavan-3-ols and flavan-3,4-diols. The process is catalysed by polyphenol oxidases in the pericarp, and requires molecular oxygen. A barrier to oxygen movement, by diffusion, may be occur causing deficits in the internal tissues. Oxygen diffusion, into the internal tissues, is especially important because it is essential in the Citric Acid Cycle to generate ATP. Therefore, the high energy requirements of young developing grains may not be adequately met, especially, in those grains that also have a high oxygen requirement for colour formation. Thus, the rate of decrease of flavanols in developing grains may give an indication to the rate of deposition of colour pigments in the pericarp and the rate of oxygen consumption by the coats.

7.1 The Quadratic exponential functions for concentration of flavanols in developing wheat grains

The concentration of flavanols, in developing wheat grains, followed an exponential decline. Quadratic exponentials were found appropriate. Good fits were obtained for each experimental unit (Tables A 6.2 (a) to A 6.2 (j)). Graphs of the fitted functions and the observed data points for each experimental unit are given in Appendix 6 (Figures A 6.2 (a) to A 6.2 (j)). The regressions were all significant at $P = 0.05$, many of them, at $P < 0.001$. Table 7.1 shows the estimated mean values of the three statistics obtained for each cultivar. The mean diagrams of the functions estimated for each cultivar are shown in Figures 7.1 and 7.2.

7.2 Analysis of the Quadratic exponential functions for flavanol concentration in developing wheat grains

The estimated statistics for the quadratic exponential functions fitted to each individual experimental unit were analysed using multivariate analysis of variance. The first discriminant had a strong correlation and was significant at $P < 0.01$. Both the second and third discriminants were not significant. Only the first discriminant was, therefore, used because of parsimony. Table 7.2 shows the proportional variance contributions of the discriminants. The correlations between discriminant 1 and the variables were all very strong.

1. The intercept had no influence on the first discriminant
2. The slope had a strong influence on the first discriminant.
3. The acceleration (decelleration) had a strong influence on the first discriminant.

The strong influence of the slope on the first discriminant was associated with the rapid loss of flavanols in the developing grains. In the red - grained cultivars, this may represent a rapid polymerisation of flavanols. It may not be true, however, because flavanol levels equally decline in the white - grained cultivars as in the red - grained cultivars, yet, colour is formed in only the latter.

There were no important differences amongst the cultivars in the initial amount of flavanols (intercept) in the grain. There were three overlapping significance groups amongst the cultivars. No consistent pattern was seen that could indicate differences between the red - grained cultivars and the whites (Table 7.4).

7.3 Flavanol concentration at harvest ripeness

The level of flavanols at harvest ripeness were high in all the cultivars. The only significant difference observed amongst the cultivars, for this variable, was that between Hilgendorf 61 and Brevor (Table 7.5). The ANOVA was insignificant ($P > 0.3$). The heritability estimates were low ($h^2 = 0.240$). These and the variance components are given in Table 7.6.

Table 7.1. Estimated statistics for the mean Quadratic exponential functions for Flavanol levels of developing grain of ten wheat cultivars.

Statistic	B_0		B_1		B_2	
Cv.	mean	s.e. B_0	mean	s.e. B_1	mean	s.e. B_2
Gamenya	6.1298	**0.3911**	-0.1382	**0.0176**	0.00139	**0.00025**
Tordo	6.0095	**0.3655**	-0.1231	**0.0332**	0.00121	**0.00054**
Kenya 321	6.0654	**0.3589**	-0.1050	**0.0187**	0.00085	**0.00023**
Brevor	6.2589	**0.4109**	-0.1424	**0.0222**	0.00139	**0.00030**
Isis	5.9249	**0.3037**	-0.1069	**0.0431**	0.00079	**0.00090**
Sonora 64	5.8594	**0.1798**	-0.1058	**0.0066**	0.00089	**0.00003**
A Thatcher	6.4084	**0.5071**	-0.1481	**0.0319**	0.00148	**0.00042**
La Prevision	6.0515	**0.6041**	-0.1159	**0.0252**	0.00100	**0.00039**
Hilgendorf 61	6.2082	**0.3690**	-0.1174	**0.0133**	0.00102	**0.00015**
Karamu	6.8917	**0.2055**	-0.1677	**0.0246**	0.00164	**0.00040**

Table 7.2. Properties of the four discriminants for the statistics estimated from the quadratic exponential functions for flavanol concentration.

	Proportion	Pr > F
1	0.6720	0.0095
2	0.2314	0.2616
3	0.0966	0.5218

Table 7.3. The correlation between the variables and the first discriminant (between discriminant structure) for flavanol concentration.

Variable	Correlation	Standardized coefficients
β_0	-0.7348	1.9720
β_1	0.9018	7.2773
β_2	-0.8461	4.7125

Table 7.4. Discriminant scores for the for the statistics estimated from the quadratic exponential functions for flavanol concentration.

Cultivar	Discriminant score[1]	t-group
Kenya 321	4.2959	A
Hilgendorf 61	2.5706	A
Sonora 64 A	2.5211	A
Tordo	1.4258	AB
La Prevision	1.3862	AB
Isis	0.5398	AB
Gamenya	-1.9596	B
Thatcher	-2.6794	B
Brevor	-2.9812	B
Karamu	-5.1193	B

[1] discriminant scores with the same letter are not significantly different at P = 0.05

Figure 7.1. The mean diagrams of the quadratic exponential functions fitted on flavanol concentration data for ten wheat cultivars

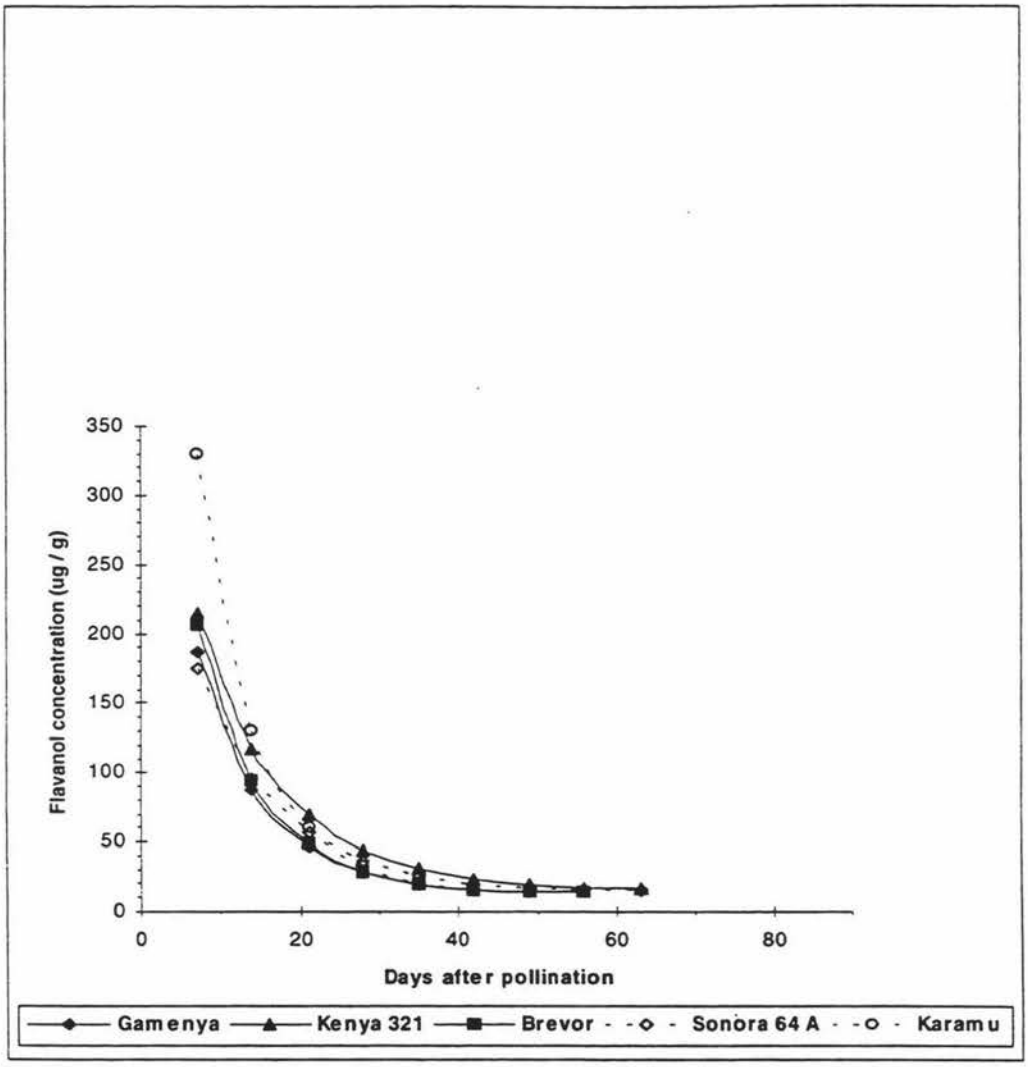


Figure 7.2. The mean diagrams of the quadratic exponential functions fitted on flavanol concentration data for ten wheat cultivars

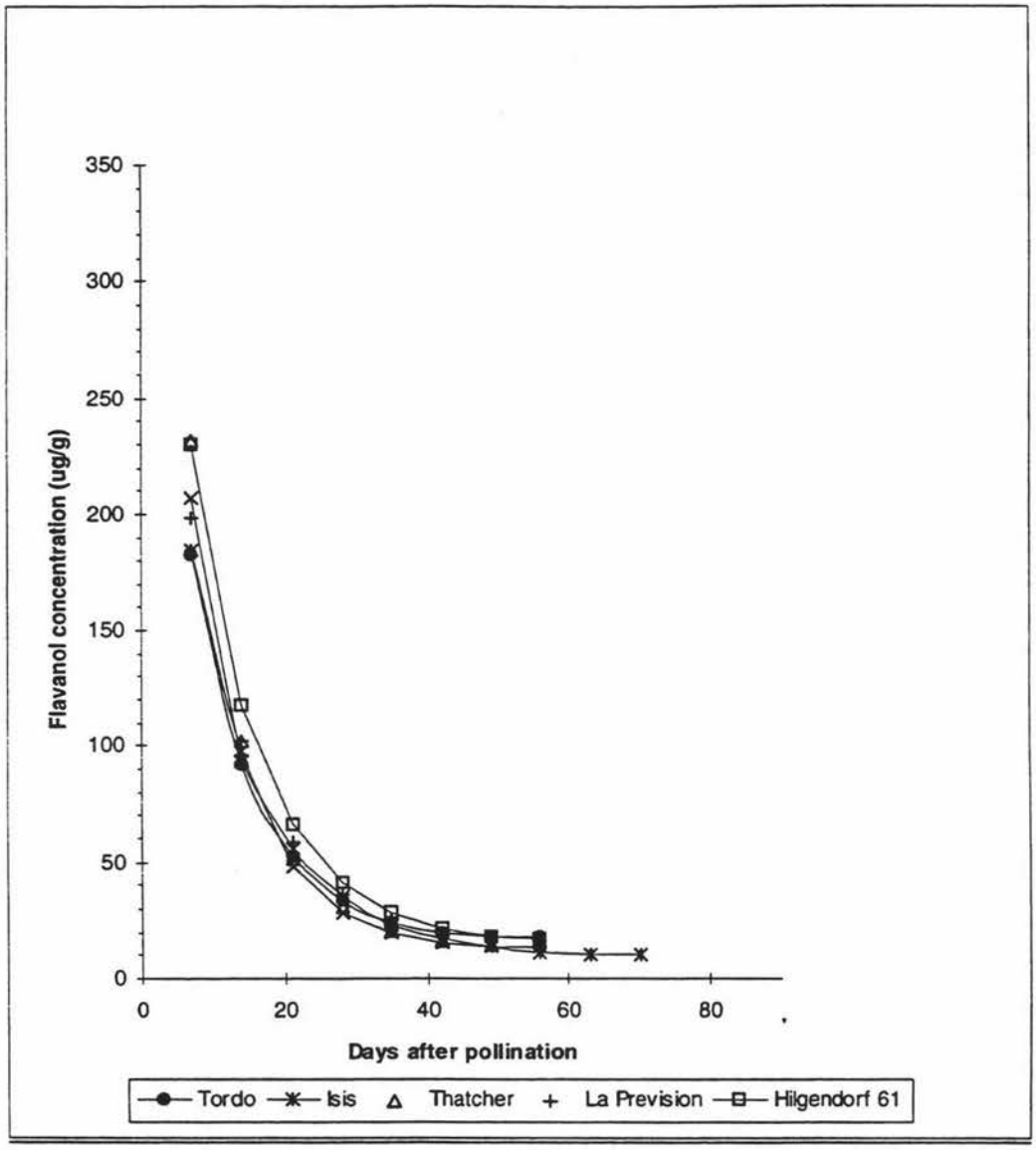


Table 7.5. The mean values for flavanol concentration (µg/g) at harvest ripeness and tests for significant differences amongst ten wheat cultivars.

Cultivar	Mean[1]	t-group
Hilgendorf 61	18.041	A
Tordo	17.175	AB
Kenya 321	17.105	AB
Sonora 64 A	16.416	AB
La Prevision	15.816	AB
Gamenya	15.174	AB
Thatcher	15.159	AB
Isis	14.530	AB
Karamu	14.339	AB
Brevor	13.460	A

[1] means with the same letter are not significantly different at P = 0.05

Table 7.6. Variance components and heritability estimates for the flavanol concentration at harvest ripeness in ten wheat cultivars.

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	2.1066	0.89827	0.240	0.10952	1.23	0.3116
Block	3	0.7576	0.47913				
Error	27	6.6571	1.74825				

8. ALPHA - AMYLASE

The level of alpha - amylase, in developing wheat grains, may be an important variable to measure because it indicates the responsivity of the aleurone layer to endogenous changes in hormone levels. There is usually a spill over of alpha - amylase that originates from the maternal parent. This alpha - amylase may be important in initiating starch deposition in the endosperm. In this way, the significance of basal alpha - amylase may differ from that of germinative alpha - amylase. Germinative alpha - amylase is involved in the breakdown of starch reserves in the endosperm and embryo. Only the basal alpha - amylase level was measured in this study.

8.1 Quadratic exponential functions for alpha - amylase activity

The decline of alpha - amylase activity in the developing grains followed an exponential pattern. Quadratic exponentials were appropriate on biological grounds. These functions were fitted to the data from each individual experimental unit. Good fits were obtained. The coefficients of regression for the functions were all high. The estimated statistics obtained from the regressions and standard errors are shown in Tables A 7.1 (a) to A 7.1 (j), and Figures A 7.1 (a) to A 7.1 (j) show graphs of the fitted function and observed data points for each experimental unit.

8.1.1 Analysis of the quadratic exponential functions fitted on alpha - amylase data

The statistics (β_o, β_1, and, β_2) estimated by the quadratic exponential functions were used in a discriminant correlational analysis to study the behaviour and meaning of the functions. The correlation for the first discriminant was very strong (Table 8.2), and, the first discriminant explained the 70 % of total variance required by parsimony. The first discriminant was significant ($P < 0.05$) but the second and third discriminants were not (Table 8.2). Table 8.3 shows the structure between the first discriminant and its variables, and the standardized coefficients for the variables. Analysis of the information contained in these tables revealed that the intercept (β_0) had a strong influence on the first discriminant and that the influence of the slope (β_1) was suppressed. Acceleration of the growth of the function (β_2)

had no influence on the first discriminant. Therefore, the initial level of alpha - amylase in the developing grains was important in discriminating amongst the cultivars.

Table 8.1 shows the mean estimates of β_o, β_1, and, β_2 in the fitted quadratic exponential functions, and Figures 8.1 and 8.2 show diagrams of the mean functions estimated for each cultivar. This figure was sub - divided into two portions for clarity reasons. The tables inside the figures were extracted from Table 8.4 showing the t-grouping of the discriminant scores for each cultivar. The cultivars were discriminated into five overlapping significance groups.

Table 8.1. Estimated statistics for the mean Quadratic exponential functions for alpha - amylase in developing grain of ten wheat cultivars.

Statistic	B_0		B_1		B_2	
Cultivar	mean	**s.e. B_0**	mean	**s.e. B_1**	mean	**s.e. B_2**
Gamenya	4.6786	**0.9669**	-0.1827	**0.1201**	0.00260	**0.00052**
Tordo	5.0325	**0.3531**	-0.2211	**0.0371**	0.00250	**0.00071**
Kenya 321	5.9096	**1.0232**	-0.2767	**0.1501**	0.00361	**0.00318**
Brevor	5.3507	**0.5120**	-0.1973	**0.0739**	0.00186	**0.00125**
Isis	4.9742	**0.6146**	-0.2567	**0.0567**	0.00212	**0.00321**
Sonora 64 A	5.2334	**0.6566**	-0.2004	**0.0759**	0.00213	**0.00099**
Thatcher	5.2922	**0.3860**	-0.2365	**0.0433**	0.00235	**0.00058**
La Prevision	4.1398	**0.8917**	-0.1265	**0.1517**	-0.00005	**0.00423**
Hilgendorf 61	4.6208	**0.3453**	-0.1538	**0.0710**	0.00123	**0.00126**
Karamu	5.4517	**1.2871**	-0.2219	**0.1149**	0.00196	**0.00154**

Table 8.2. Properties of the three discriminants for the Quadratic exponential function fitted for alpha-amylase

	Proportion	Pr > F
1	0.8030	0.0143
2	0.1231	0.6676
3	0.0738	0.6513

Table 8.3. The correlation between the variables and the first discriminant and the standardized coefficients for alpha-amylase.

Variable	Correlation	Standardized coefficients
β_o	0.8203	3.4151
β_1	-0.3658	2.7795
β_2	0.4793	0.1351

Figure 8.1. Diagrams of the mean quadratic exponential functions for alpha - amylase activity in developing wheat grains from five cultivars.

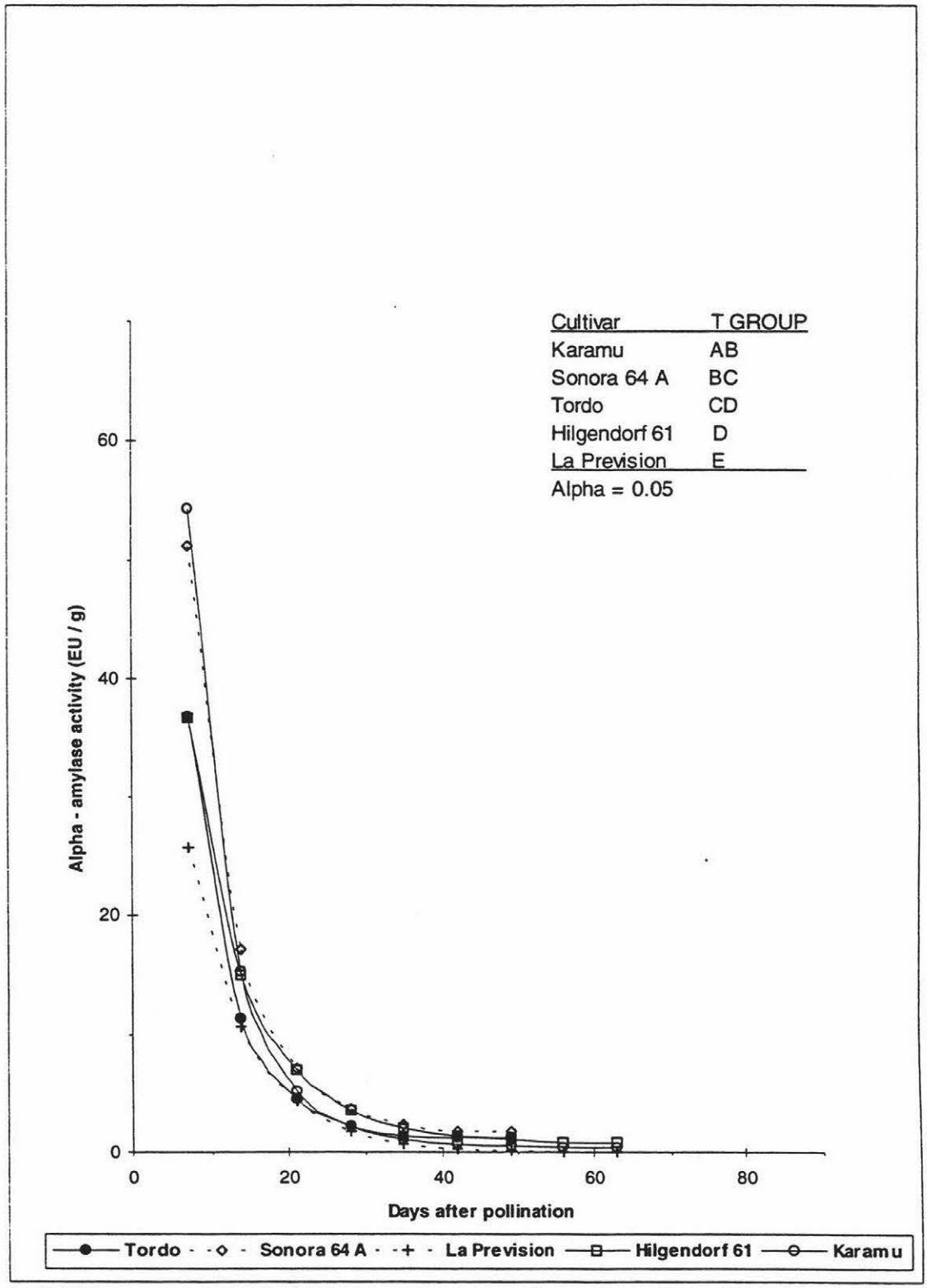


Figure 8.2. Diagrams of the mean quadratic exponential functions for alpha - amylase activity in developing wheat grains from five cultivars.

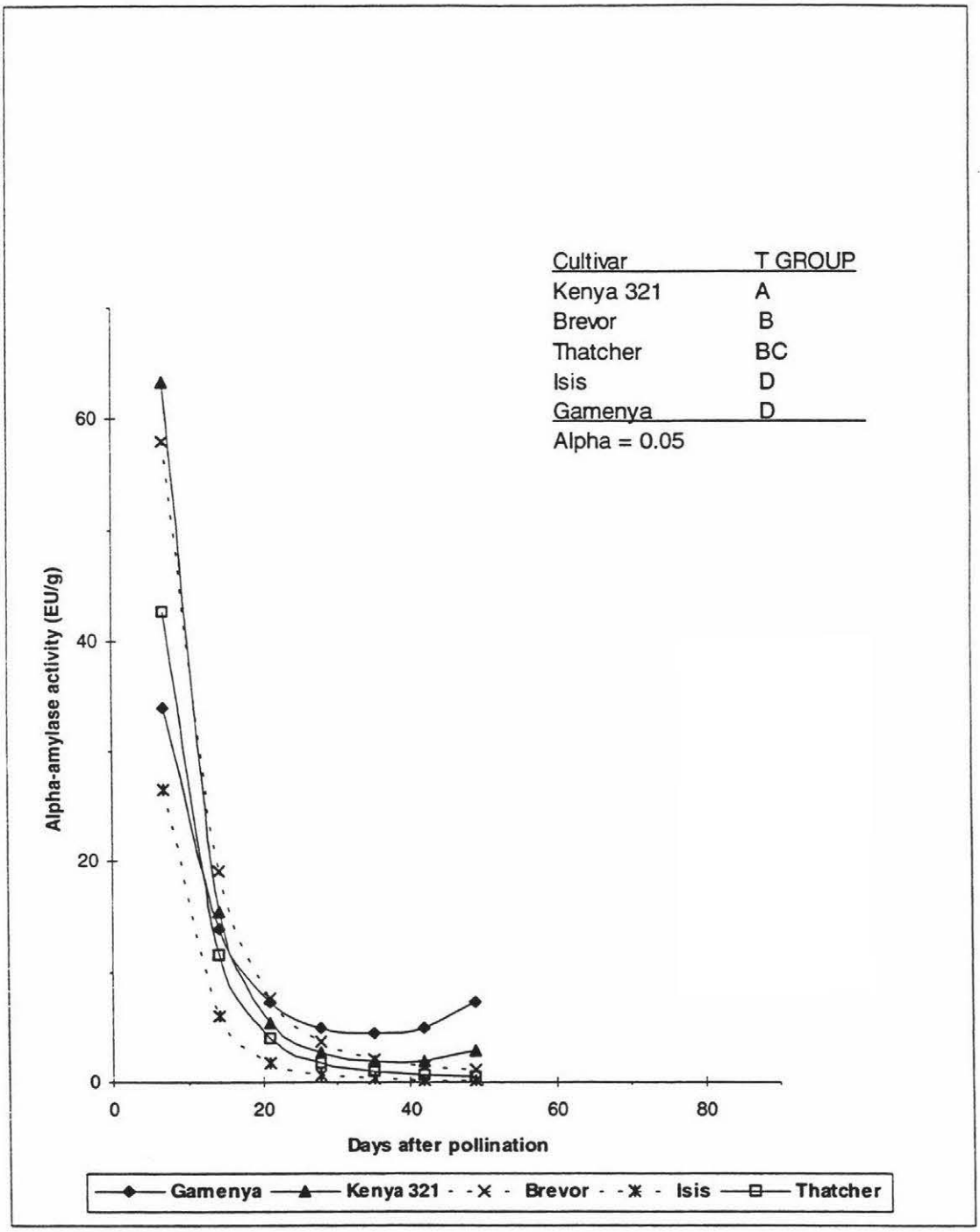


Table 8.4. The discriminant scores for alpha - amylase activity in developing grains of ten wheat cultivars and the t-test for differences.

Cultivar	Mean[1]	t-group
Kenya 321	4.0639	A
Brevor	3.1038	A
Karamu	2.6887	AB
Sonora 64 A	1.9003	ABC
Thatcher	0.4835	ABCD
Tordo	-1.0210	BCDE
Hilgendorf 61	-1.2471	BCDE
Gamenya	-2.1346	CDE
Isis	-3.5491	DE
La Prevision	-4.2884	E

[1] Means with the same letter are not significantly different.

The following secondary variables were derived from the alpha - amylase data:

Mnemonic	Description of variable
AMYHR	Level of alpha - amylase at harvest ripeness
AMYABA1	Level of alpha - amylase at the first peak of ABA activity
AMYABA2	Level of alpha - amylase at the second peak of ABA activity
AMYD50	Level of alpha - amylase at the time of median dormancy

8.2 Level of alpha - amylase at harvest ripeness (AMAHR)

The level of alpha - amylase at harvest ripeness is one of the characters that is considered routinely, in wheat, because it has a direct bearing to the quality of wheat flour. The level of basal α - amylase at harvest ripeness may indicate the level of risk involved should the grain be exposed to conditions suitable for sprouting.

Sonora 64 A had the highest level of α - amylase activity at harvest ripeness. La Prevision and Karamu had the lowest levels (Table 8.5). The three overlapping significance groups were found for α - amylase activity at harvest ripeness. A moderate heritability estimate (h^2 = 0.282) was found for this character indicating the importance of the environment. Most of the basal α - amylase in developing grains originates from the maternal parent. The variance components and heritability estimates for α - amylase at harvest ripeness are reported in Table 8.6.

8.3 Level of alpha - amylase at the first peak of ABA activity (AMYABA1)

Abscisic acid in the grain acts directly to inhibit alpha - amylase synthesis by the endosperm. However, ABA is not involved in deactivating alpha - amylase already present in the grain. In the absence of germinative conditions, therefore, the alpha - amylase present in the grain may have arisen through transfer from the mother plant or may have been produced early in development before inhibiting levels of ABA activity occurred. The first ABA peak of activity had a direct bearing only on future alpha - amylase production in the endosperm tissue.

No significant differences were found, amongst all ten cultivars, in the level of alpha - amylase activity at the time of the first peak of ABA activity (Table 8.7). The large variation in the data (MSE = 4131.452), however, makes this test insensitive, e.g. Isis, having a mean of 93.67 EU/g, was grouped with Gamenya (mean activity = 2.66 EU/g).

8.4 Level of Alpha - amylase at the second peak of ABA activity (AMYABA2)

The levels of alpha - amylase activity, at the time of the second ABA peak, amongst the ten cultivars fell in three overlapping significance groups (Table 8.8). A comparison of Table 8.5 and Table 10.8 indicates that, for Isis and Gamenya, alpha -amylase activity increased from AMYABA2 to AMYHR. This could be an indication of germinative processes having occured in these cultivars. In all the other cultivars, alpha - amylase activity continued to decline.

Table 8.5. The mean alpha - amylase activity at harvest ripeness in developing grains of ten wheat cultivars and t-tests for differences.

Cultivar	Mean[1]	t-grouping
Sonora 64 A	2.5901	A
Kenya 321	2.3359	AB
Isis	2.0538	ABC
Tordo	1.8868	ABC
Brevor	1.1866	ABC
Hilgendorf 61	0.9426	ABC
Gamenya	0.8883	ABC
Hilgendorf 61	0.7858	BC
La Pravision	0.5442	C
Karumu	0.5149	C

[1] Means with the same letter are not significantly different.

Table 8.6. Variance components and heritability estimates for α - amylase activity at harvest ripeness

Type	Degrees of freedom	Variance component	s.e.	h^2	s.e.	F	P > F
Genotype	9	0.5955	0.25390	0.282	0.11774	1.39	0.232
Block	3	0.1273	0.08054				
Error	27	1.5194	0.39901				

Table 8.7. The mean alpha - amylase activity, at the first peak of ABA activity, in developing grains of ten wheat cultivars.

Cultivar	Mean[1]	t-group
Isis	93.67	A
Brevor	68.08	A
Kenya 321	62.40	A
Thatcher	44.41	A
Karamu	37.32	A
Sonora 64 A	32.24	A
La Prevision	25.70	A
Hilgendorf 61	24.05	A
Tordo	23.87	A
Gamenya	2.66	A

[1] Means with the same letter are not significantly different.

Table 8.8. The mean alpha - amylase activity, at second peak of ABA activity, in developing grains of ten wheat cultivars and t-tests for differences.

Cultivar	Mean[1]	t-group
Brevor	6.038	A
Sonora 64 A	5.548	A
Tordo	4.820	AB
Karamu	4.509	ABC
Kenya 321	3.012	ABC
La Prevision	2.289	ABC
Hilgendorf 61	2.262	ABC
Thatcher	1.108	BC
Isis	0.967	BC
Gamenya	0.363	C

[1] Means with the same letter are not significantly different.

9. ABSCISIC ACID

The abscisic acid found in young grains may include residual amounts from the vegetative tissues of the maternal plant. This abscisic acid may prevent the young grain from precociously germinating in response to suitable environments.

Abscisic acid activity was measured using the Enzyme-Linked Immunosorbent Assay (ELIZA) method of Walker-Simmons *et al.* (1987). No consistent pattern was found amongst the ten cultivars studied or between the two groups of red - and white - grained cultivars in amount, timing of activity or duration of ABA acivity. However, very early peaks of activity (before the tenth day after pollination) were observed for some cultivars (Table 9.1). The other cultivars had their first peak of ABA activity occurring between day 12 and day 23 after pollination, and all cultivars had at least two peaks of ABA activity that occurred before harvest ripeness (Table A 7.2, in Appendix 7). Gamenya had the second peak of ABA activity in only one replicate, and no third peak. The third peak (pre-harvest ripeness) of ABA activity was observed in, at least, one replicate of each of the remaining cultivars. A post harvest ripeness peak was observed in all the cultivars, and the second replicate of Tordo also had a fifth ABA peak, 78 days after pollination, (Table A 7.2). The level of abscisic acid activity varied widely, in the different cultivars, throughout the entire period of the experiment, and there was no obvious pattern to suggest which characters were influenced by these different levels of ABA activity.

Therefore, no function was fitted to the ABA data, but the timing of appearance (days after pollinantion), width of peak (days), and, in some cases, the height of peak (ng/grain), of ABA activity could be analysed multivariately.

9.1 Analysis of timing, width (duration) and height (amount) of ABA peaks of activity

The first three (pre - harvest) peaks of ABA activity were analysed, separately, in a MANOVA to investigate the relative importance of the timing, duration and level of activity. The post harvest peaks (4 and 5) were ignored in these analyses because they were considered unimportant for dormancy at harvest ripeness.

9.2 ABA peak 1

The univariate analysis of variance for timing of peak 1 was highly significant ($P < 0.001$). The genotypic effect was significant at 0.001. Neither the width nor the height of peak 1 showed any significant effects at the 0.1 level. All three variables were used in the MANOVA. A strong correlation was found for the first discriminant. The correlations for the second and third discriminants were only moderate. The first discriminant was highly significant ($P < 0.0001$) and the second was significant at $P = 0.05$. Parsimony for the model (70 %) was satisfied by the first discriminant alone (Table 9.2).

Correlations between the first discriminant and the variables and the standardized coefficients of the variables are shown in Table 9.4. Timing had a strong influence on the first discriminant. This indicated greater importance for the timing of ABA activity in relation to the other variables. The effect of the width (duration) of ABA activity (days between the two troughs on either side of the peak) was enhanced, although, this had a negative coefficient. Height (amount of ABA) had no effect on the first discriminant. The importance of width and timing of peak, and lack of influence from height of ABA activity, seems to suggest a significant role for tissue sensitivity. One can imagine a role, for the first discriminant, of an "ABA sensor". Such a "sensor" would reside in tissues that normally respond to ABA, such as, the aleurone layer of the endosperm (e.g. α - amylase response) and embryonic tissue (e.g. growth and development, and dormancy setting

The ten cultivars were discriminated into five overlapping significance groups (Table 9.5). There was no obvious separation between the red - grained cultivars and the white - grained cultivars.

9.3 ABA Peak 2

The mean values for timing, width and height of peak 2 are shown in Table 9.2. Both timing and height of peak 2 were not significant. The genotypic effect was significant at ($P = 0.1$) for width of peak. Moderate correlations were recorded for the first and the second discriminants. These two discriminants were required by parsimony to describe the variance of the model, but, only the first discriminant was significant at $P = 0.05$ (Table 9.6). The correlation between timing and the first discriminant was weak, but that between width and the first discriminant was

strong (Tables 9.7). Height had a moderate correlation with the first discriminant. A very strong correlation was observed between timing and the second discriminant, while that between width and the second discriminant was of moderate strength.

Width of peak 2 had the strongest influence on the first discriminant, the effect of timing was suppressed while height of peak had only a weak influence. Timing of peak had a strong influence on the second discriminant. Thus, the first discriminant had a 'duration of peak' orientation, while the second discriminant had an emphasis on timing of peak 2.

The multivariate test for significant differences between pairs of cultivars, using Hotelling's T^2 statistic and the F approximation, showed differences at the 0.1 level between Isis and five other cultivars, *viz*, Tordo, Kenya 321, Brevor, Sonora 64 A and La Prevision (Table 9.8). There were no other significant differences between pairs of cultivars. Figure 9.1 shows scatter plots obtained as a result of the separation of the cultivars by the two discriminants.

Table 9.1. The mean timing (days after pollination), width (days) and height (ng/grain) of the first peak of Abscisic acid activity in developing grains of ten wheat cultivars.

Cultivar	TIMING		WIDTH		HEIGHT	
	Mean (DAP)[1]	**s.e (days)**	Mean (DAP)	**s.e (days)**	Mean (ng/grain)	**s.e. (ng/grain)**
Gamenya	22.00	**3.3665**	29.25	**8.5000**	3.6823	**3.27797**
Tordo	9.75	**2.9861**	8.00	**4.3205**	8.2965	**3.62088**
Kenya 321	15.00	**9.5568**	19.75	**14.6600**	1.2133	**0.88095**
Brevor	6.25	**0.5000**	10.50	**4.4347**	7.6018	**2.66893**
Isis	22.25	**5.2520**	28.00	**9.6953**	8.1138	**8.94865**
Sonora 64 A	12.50	**7.5055**	18.75	**19.8221**	2.7733	**4.32537**
Thatcher	8.25	**4.5000**	20.50	**12.8970**	9.9725	**5.92498**
La Prevision	8.25	**2.8723**	12.00	**8.8694**	0.9325	**0.77393**
Hilgendorf 61	13.00	**8.0829**	21.25	**15.0859**	5.3525	**5.25742**
Karamu	9.50	**7.0000**	11.25	**5.85230**	12.3565	**10.71600**

[1] DAP : days after pollination

Table 9.2. The mean timing (days after pollination), width (days) and height (ng/grain) of the second peak of Abscisic acid activity in developing grains of ten wheat cultivars.

Cultivar	TIMING		WIDTH		HEIGHT	
	Mean (DAP)[1]	**s.e (days)**	Mean (DAP)	**s.e (days)**	Mean (ng/grain)	**s.e. (ng/grain)**
Gamenya	36.00	**18.0000**	3.50	**7.0000**	0.21550	**0.43100**
Tordo	27.50	**13.0256**	19.25	**5.5603**	9.39975	**9.69318**
Kenya 321	38.50	**10.3763**	28.00	**13.8323**	8.18825	**8.68890**
Brevor	28.00	**14.8324**	26.00	**7.7889**	4.31825	**3.65970**
Isis	41.25	**10.2103**	11.00	**4.0825**	5.38275	**2.20282**
Sonora 64 A	17.75	**12.1758**	15.50	**11.1505**	14.41975	**5.00260**
Thatcher	24.75	**19.7210**	17.25	**14.3614**	0.46575	**0.45767**
La Prevision	26.75	**8.0571**	19.75	**11.7863**	9.74550	**12.61065**
Hilgendorf 61	16.75	**19.4487**	11.00	**14.2829**	3.15000	**5.21673**
Karamu	25.50	**6.3509**	19.25	**2.6300**	1.39975	**0.80777**

[1] DAP : days after pollination

Table 9.3. Properties of the three discriminants for the first ABA peak in developing wheat grains of ten wheat cultivars.

Discriminant	Proportion	Pr > F
1	0.7140	0.0001
2	0.2236	0.0463
3	0.0624	0.4334

Table 9.4. The correlation between the variables and the first discriminant and the standardized coefficients of the variables for the first ABA peak in developing wheat grains of ten wheat cultivars.

Variable	Correlation	Standardized coefficient
Timing	0.9490	2.9485
Width	0.6629	-1.5738
Height	-0.2579	-0.1346

Table 9.5. Discriminant scores for the first ABA peak activity in ten wheat cultivars and the T - test for differences amongst the cultivars.

Cultivar	Discriminant Score[1]	t-group
Isis	8.2796	A
Gamenya	7.8370	A
Kenya 321	2.4325	AB
Sonora 64 A	-0.2885	BC
Hilgendorf 61	-0.5297	C
Tordo	-0.7233	CD
Karamu	-2.1250	CD
La Prevision	-3.2758	DE
Brevor	-5.5048	E
Thatcher	-6.1021	E

[1] Discriminant scores with the same letter are not significantly different.

Table 9.6. Properties of the three discriminants for the second ABA peak in developing wheat grains of ten wheat cultivars.

Discriminant	Proportion	Pr > F
1	0.5630	0.0417
2	0.3241	0.2310
3	0.1129	0.5734

Table 9.7. The correlation between the variables and the first and second discriminants and the standardized coefficients of the variables for the second ABA peak in developing wheat grains of ten wheat cultivars.

	Dicriminant 1		**Discriminant 2**	
Variable	Correlation	Standardized coefficient	Correlation	Standardized coefficient
Timing	-0.0927	-1.1130	0.9932	1.0170
Width	0.7287	1.5139	0.6317	0.0576
Height	0.4792	0.5343	0.3153	0.1420

Table 9.8. The multivariate t-test for pairwise comparisons between ten wheat cultivars using the two discriminants for the second peak of ABA activity (the upper triangle contains significance symbols and the lower F^1 statistics).

Cv.	Gam	Tordo	Ken321	Brevor	Isis	Sonora	Thatch	La Pre	Hil61	Kar
Gamenya	----------	ns	ns	ns	ns	ns	ns	ns	ns	ns
Tordo	-0.4355	----------	ns	ns	*	ns	ns	ns	ns	ns
Kenya 321	-2.3248	-0.4968	----------	ns	*	ns	ns	ns	ns	ns
Brevor	1.825	0.4562	-0.5313	----------	*	ns	ns	ns	ns	ns
Isis	-4.3037	5.2275	7.8726	8.9844	----------	*	ns	*	ns	ns
Sonora 64 A	2.4798	-0.1863	-2.015	-0.3496	6.196629	----------	ns	ns	ns	ns
Thatcher	-0.7276	0.1855	-0.431	1.3678	2.231535	0.89736	----------	ns	ns	ns
La Prevision	0.111	0.0196	-0.6555	0.2772	5.887753	-0.24479	0.3921	---------	ns	ns
Hilgendorf 61	0.0335	-0.283	-1.6619	1.109	0.052982	1.26628	-0.28	0.019	---------	ns
Karamu	-0.3836	0.0056	-0.6953	0.8105	3.54616	0.36708	0.0725	0.123	-0.204	-------

[1] $F = (n_1 + n_2 - p - 1)T^2$

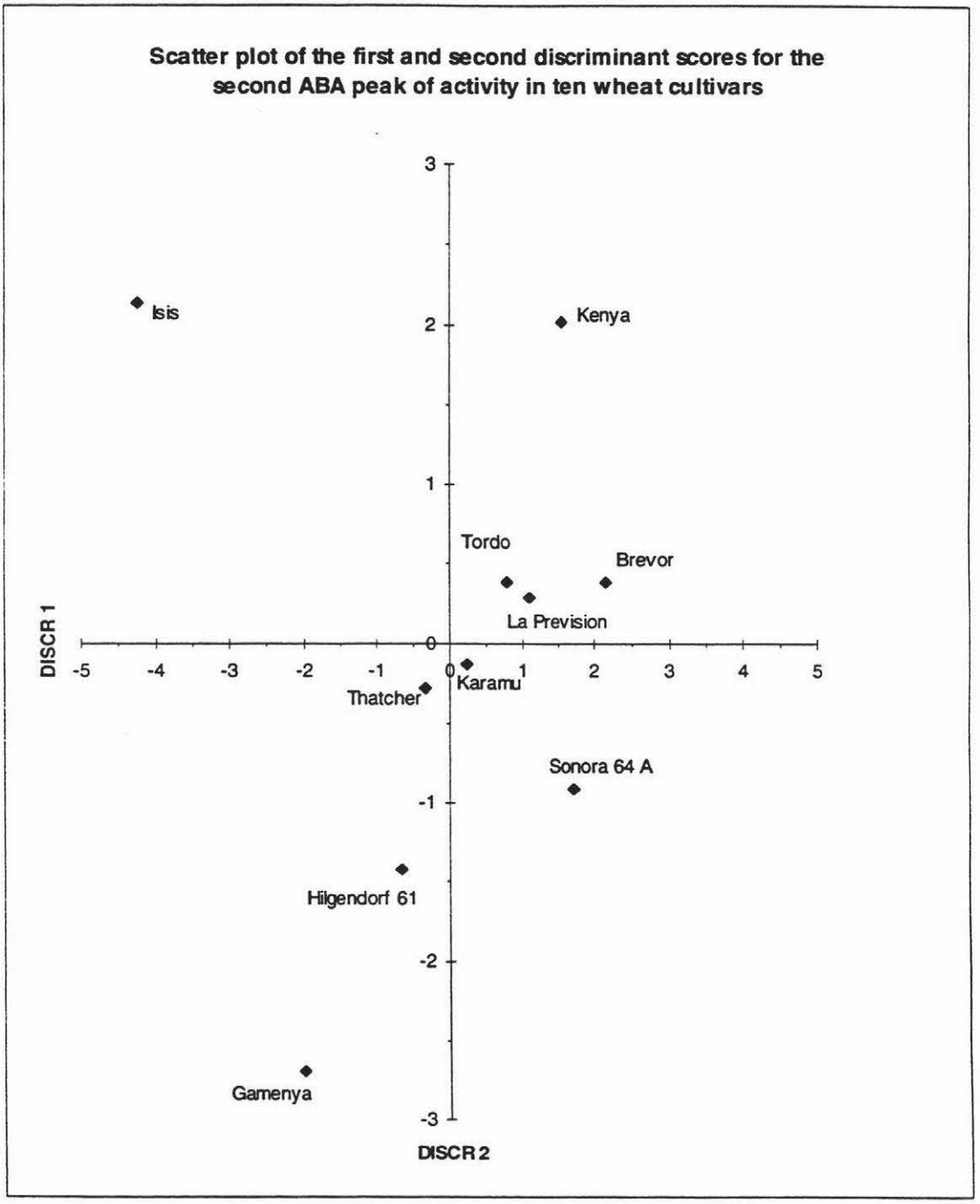
$T^2 = (n_1 n_2 (x_1 - x_2)' C^{-1} (x_1 - x_2))/(n_1 + n_2)$

Significance symbols in upper triangle are explained below:

ns : not siginificantly different ($P > 0.1$)

* : $0.1 > P > 0.05$

Figure 9.1. Scatter plot of the first and second discriminant scores for the second ABA peak in ten wheat cultivars.



Chapter 5

Discussion

5.1 Redness and Dormancy

Red graincoat colour was shown to be associated with embryo dormancy, but not its cause. All the red wheat cultivars showed some level of dormancy at harvest ripeness but the white cultivars did not. The variation in the level of dormancy was not influenced by the number of red (*R*) genes present. The cultivars having three red genes did not have the highest levels of dormancy. Reitan (1980) deduced from a study of a diallel cross involving 8 parents that recessive genes for long dormancy duration had a greater effect than differences between number of *R* alleles. Sereeprasert (1990) reported interallelic interaction between the gene(s) for grain redness and some unknown dormancy genes; and gene effects were found also to be dependent on environmental conditions. Dormancy in the white - grained wheats was lost rapidly and the curves showed steep descents. An exception was Kenya 321 the dormancy function of which was parallel to that of Sonora 64 A, although at a lower level. However, even in this case, dormancy had declined to a trivial level by harvest ripeness.

5.2 ABA and Embryo Dormancy

Dormancy does not appear to be related to the timing, amount or duration of whole grain abscisic acid activity. The timing, width and duration of peaks of ABA did not correspond with the observed dormancy levels amongst the cultivars. The lack of association between ABA activity and dormancy has a bearing on one aspect of the working hypothesis. The results did show that redness was needed for dormancy, but not that ABA was the intermediate link. The part of the hypothesis concerning hypo - oxia is, therefore, still viable, as the formation of colour has been confirmed as a pre-requisite of embryo dormancy.

5.3 The Working Hypothesis

According to the working hypothesis, the formation of colour during development of the grain would cause an increase in oxygen demand in the graincoat, which, in turn, would lead to oxygen depletion in the internal tissues of the grain. Because of gibberellin / ABA biochemistry, it was further postulated that there would be an increase in ABA production, triggering the onset of dormancy, especially, if the embryo was immature. More detail is needed here, because there is no information available, in the literature, on the *magnitude* of hypo-oxia that is sufficient to stimulate dormancy in the embryo. It could be that localised embryonic ABA *is* responsible, but more refined techniques would be required. No evidence could be found about the rate of oxygen consumption by the graincoats in wheat. Lenoir *et al.* (1986) investigated oxygen consumption by the glumellae in barley where it was observed that a delay in oxygen uptake existed, for up to 10 h, in imbibed non - dormant barley caryopses, but no such delay could be found in the glumellae of dormant caryopses. This delay was enough to allow germination. Similar studies need to be conducted in wheat if the connection between ABA, hypo-oxia, redness and dormancy is to be fully understood. However, as discussed in 5.2. the present results do not support ABA involvement, but they do support, still, a role for colouration, possibly through hypo-oxia.

5.4 Hypo - oxia and Colour formation

The evidence accumulated, in this study, suggests a role for the *timing* of colour formation in embryo dormancy. The colour profiles showed that colour formation was more rapid in the red wheats than in the white wheats and, with the exception of cv. Tordo, colour formation was completed earlier in the red than the white wheats. Since colour formation in wheat is an oxidative process, involving the polyphenol oxidases (Kruger, 1976; Gordon, 1979), it is probable that oxygen consumption in the graincoats of red wheats was greater than in the white wheats at any time during colour development. This would intensify the reduction in oxygen available at the internal tissues, such as the embryo.

The implications for low oxygen tensions in the grain are many and varied but the ones that are relevant to the present study are (a) a reduced rate of oxidative respiration, (b) stimulation of ABA synthesis, and, (c) suppression of GA synthesis. If hypo-oxic conditions did stimulate ABA synthesis, then it can only be assumed

that the amount and/or rate of synthesis of ABA must have been trivial, or localised, because no relationship was found in this work between whole grain ABA activity and dormancy. (GA activity in the developing grains was not measured in this study, time and available resources being limiting). This suggests that ABA may not be the causal agent in any hypo - oxic condition, suggesting that restriction of oxidative respiration may be an alternative reason for the variation in dormancy levels observed.

Oxidative respiration occurs via the Tri-Carboxylic Acid Cycle (TCA) in living tissue. It is the main energy source for growth and development. Under normal conditions, the TCA cycle generates enough ATP (adenosine tri - phosphate) molecules to support normal growth and development, but, when insufficient oxygen is available, operation of the TCA cycle is depressed or may cease altogether, when it is replaced by anaerobic respiration. This causes the production of ethanol and other metabolic waste products accompanied by a reduction in the number of ATP molecules produced. Dormant wheat grains, mid - way through development may not be capable of overcoming this restraint, e.g., by resorting to alternative energy - generating pathways, such as, the pentose phosphate pathway (Roberts, 1973). It is suggested, therefore, that such a situation may exist in the developing red wheat grains, during the rapid phase of colour development, and that this destabilised normal embryonic development.

5.5 Embryo maturity

Embryonic maturity was noticeably delayed in the most-dormant red - grained wheats (La Prevision and Thatcher) but not in the least dormant ones (Hilgendorf 61, Karamu and Thatcher). A similar pattern was noticed in the white - grained wheats, with the exception of Tordo. This cultivar is a gibberellic acid - insensitive wheat, and it showed an initial delay in germinative response, under these dormancy - breaking conditionsfollowed by a steep rise later. This contradicts previous reports that embryos reach maturity in similar lengths of time under dormancy - breaking conditions, e.g. germination of excised embryos (Miyamoto *et al.*, 1961), indicating differences in embryo maturity may arise from the choice of material used. However, embryo maturity was considered an important component for germinability (Sereeprasert, 1990).

5.6 Timing and Dormancy

The prerequisite for dormancy to occur in the wheats studied appeared to be the presence of redness but other factors were also involved. The timing of colour development and the timing of embryonic development were among some of those factors. The variation in the timing of colour maturity amongst the cultivars represented in the gene pool was moderate (Table 6.7), but those of median embryonic maturity and full embryonic maturity (95 %) were high (Tables 5.7 and 5.9, resp.). It would appear that the moderate genotypic variation amongst the cultivars found in timing of colour development was *amplified* into the larger variation that was observed in the timing of both embryonic development variables. The latter could have been responsible for the higher variation observed in the dormancy timing variables (Tables 5.23, 5.25 and 5.27).

Table 5.1 shows the location of inflexion times on the time axis, of the colour, embryo maturity and dormancy curves. A comparison of the colour inflexion point and embryonic inflexion point shows that the majority of the non - dormant wheats reached the colour inflexion point *after* the embryonic inflexion point. The exception was the dwarf cv. Tordo which also had a delayed start to embryonic development. On the other hand, within the group of red wheats, inflexion time for the colour development profile was generally *before* the embryonic inflexion point. For the cultivars Isis (white), Sonora 64 A (red) and Hilgendorf 61 (red), the inflexion points for colour and embryo maturity were almost coincident. The inflexion point on the sigmoidal functions that described each of these developmental events represented the point of maximum rate of change in the appropriate developmental attribute. For the colour development function, this point represented the point of maximum flavanol polymerisation and putative oxygen consumption by the graincoats which probably caused a deficit of oxygen amounts within the grain. It is probable that the location of the colour inflexion point relative to the embryo maturity inflexion point may influence subsequent germinability of the embryo. The discussions in Sections 5.4 and 5.5 above are relevant here.

Table 9.9. The mean inflexion times for the colour, embryo and dormancy development curves of ten wheat cultivars.

Cultivar	Presence of Dormancy [1]	Embryo maturity inflexion time (DAP)[1]	Colour inflexion time (DAP)	Dormancy inflexion time (DAP)
Gamenya	N	28.59	31.05	27.52
Tordo	(N)	38.60	24.16	39.57
Kenya 321	(N)	30.60	39.72	38.68
Brevor	(N)	19.99	41.82	37.84
Isis	N	35.98	35.32	33.89
Sonora 64 A	(Y)	29.51	30.62	42.36
Thatcher	Y	41.01	34.00	66.37
La Prevision	Y	49.80	33.14	not available
Hilgendorf 61	Y	35.30	36.11	52.70
Karamu	Y	36.78	31.51	49.44

[1] N = no dormancy at HR; (N) = putative dormancy; Y = dormancy at HR; (Y) = little or no dormancy at HR.

5.7 Flavanols and Dormancy

The flavanol content of red - and white - grained wheats have been shown to be similar (Gordon, 1975; Cross, 1977; Verry, 1978). The *timing* of oxygen deprivation in the internal tissues as a result of the enzyme-catalysed polymerisation of flavanols remains, however, a possible explanation for the difference in dormancy profiles amongst the wheat cultivars, and for the variation observed between the red cultivars. This implies that flavanols, in red wheat grains, are solely substrates for the polyphenol oxidases but are not *in vivo* inhibitors of embryo germination, as observed earlier by Gordon (1979).

The lack of striking differences in the flavanol levels between the white - grained and the red - grained cultivars is a surprise because there should be a steeper decrease in the monomeric flavanol levels in red - grained cultivars, as colour deposition progresses, compared to white - grained cultivars. The decrease of flavanol levels in the white - grained cultivars, which follows a similar pattern to that of the decrease of flavanol levels in the red - grained cultivars, also raises the question of whether these decreases are not merely due to some metabolic degradation process, not related to colour formation. If this is the case, then what is the role of the flavanols in the wheat grain? There appears a need, here, to establish this role and the mechanism by which metabolic degradation of the flavanols occurs in the wheat grains. However, the evidence also suggests that the *R* gene *is* the gene for the polymerising enzyme.

The change of colour from green to white, in white - grained wheat, may simply be a consequence of the progressive dehydration of the pericarp during growth, and the concurrent disappearance of chloroplasts from the pericarp. This was, probably, the reason why colour development in white wheats did not have any observable influence on embryonic maturity or dormancy. One would expect the colour timings of white wheat to be concomitant with their dehydration timings.

5.8 "White grained" dormancy

The putative dormancy of some of the white - grained cultivars (e.g., cvs. Brevor and Kenya 321) has not been observed in this study. One possible reason for this contradiction could be genotype X environment interactions (Gordon, 1975; Cross, 1977; Moss, 1979; Baker, 1981; Strand, 1983; Sereeprasert, 1990; Mares, 1993).

Another possibility is variation between researchers' methods in measuring dormancy including differences in composition of the specific dormancy - breaking conditions used. The effect of the environment on reported dormancy levels may also be indirectly due to differences in the developmental time allowed, after pollination, before measurements are taken, e.g., harvesting at "physiological" maturity (mass based) as opposed to harvesting at morphological maturity (colour and hardness based). Brevor was classified as being highly dormant (DeMacon *et al.*, 1992) after harvesting at "physiological" maturity and allowing 6 weeks of after - ripening. Abscisic acid sensitivity was reported as among the causes for dormancy in this cultivar (Walker-Simmons, 1987). However, most of the studies on ABA responsiveness in wheat have been done on isolated embryos (Walker-Simmons, 1987, 1988) and the *in vivo* situation may be different. Therefore, unless the exact test conditions are known, it is difficult to make comparisons between dormancy measurements with certainty.

In the present work, Tordo did not show any non-trivial dormancy at harvest ripeness. However, if the curve juxtapositions were different (in another environment), dormancy could be observable. In section 5.6, three timing statistics (colour/embryo maturity, embryo dormancy/embryo maturity, and colour/embryo dormancy) *did* show incipient behaviour in Tordo which was similar to the dormant red wheats. This is very reassuring, not only with respect to these Tordo results, but also with respect to the whole approach and methodology used in this study.

5.9 Grain dehydration

The lack of significant differences between the white - and the red - grained cultivars, with regard to moisture content, implies that differences in graincoat structure (Bradbury *et al.*, 1965a; Belderok, 1976) may not have played an important role in this study in the control of grain dehydration. The control of dehydration in developing grains is probably bi - phasic. In young, developing grains, the maternal plant plays an important role by transferring nutrients, water and minerals, via the transfer cells, to the young grain. The efficiency of the maternal plant in these processes is reflected by the rate of growth and development of the young grain. After the connection with the maternal plant is cut off, the environment exerts a greater influence on the rate and amount of water loss from the grain. King *et al.* (1992) reported that rapid dehydration at maturity

was associated with dormancy, and that drying could cause cessation of development and dormancy (King, 1993). Evidence in this study did not support this view, but the ripening environments were probably very different, with less dehydration stress in the present case.

5.10 Base α - amylase and Dormancy

The level of endogenous α - amylase has shown no important differences amongst the cultivars at harvest ripeness and no association with embryo dormancy, colour development or embryonic maturity. Further, the ABA activity did not appear to influence these endogenous α - amylase levels. Germinative α - amylase levels were not measured due to resource limitations. Base α - amylase is located in the graincoat (Kruger, 1976; Olered, 1976; Daussant and Renard, 1976; Marchylo *et al.*, 1979), and this would account for these findings.

5.11 Gibberellic acid insensitivity

The three cultivars having *Gai / Rht* genes (cvs. Tordo, Sonora 64 A and Karamu) did not show significant differences from the *rht* cultivars in the overall pattern of embryonic development. However, Tordo and Karamu were slower to show signs of embryonic growth (Figure 5.1) compared with all other cultivars. The *Rht* alleles probably cause the non-utilization of available $\mathbf{GA_1}$, through the production of an inhibitor or by directly reducing the efficiency of a GA receptor (Gale, 1989). GA_1 occurs early in the metabolism of the gibberellins. The presence of GA_3 in the imbibing fluid used in the dormancy - breaking test may have overcome this and caused the rapid test response over-riding the inherent condition in the grain. Moreover, the presence of other dormancy - breaking mechanisms in the special germination test may have evoked a "non - sensitivity" response in these cultivars.

5.12 Heritability measurements

5.12.1 Grain mass

The heritability estimates for harvest ripeness and grain mass were low (h^2 = 0.252 and 0.271, respectively). This may have been due to the genotype X environment interactions. The experiment was conducted in a single environment and it is not

possible to partition out these interactions. Another possibility was the lack of genetic variability for the cultivars in this genepool for these characters.

5.12.2 Graincoat Colour

Heritability estimates were high for colour intensity at maturity ($h^2 = 0.852$) showing relative independence from environmental influences. Grain coat colour is known to be controlled by three homoeologous genes, and genotypes may possess one, two, three or none of these genes. Further, red colour is dominant over white colour. The cultivar set widely sampled the gene-pool for this character, and so heritability estimates were not biased by lack of genetic variability. The estimates for colour timing showed values less than one half of those for colour intensity. Number of days from pollination to colour maturity had only a moderate heritability estimate ($h^2 = 0.378$) showing that there was a strong environmental influence on this variable. This seems reasonable because this variable is largely determined by the rate of development of the colour profile which would be expected to be influenced by the environment. In section 5.6, the effects of colour timing on embryo maturity and embryo dormancy were discussed. That showed thatselection for this character would be for earliness of colour maturity, so that embryo maturity might be delayed and the dormancy level might be increased. The other three characters examined in connection with colour (i.e. number of days from pollination to colour maturity, mean growth rate of colour and the difference between harvest ripeness and number of days to colour maturity) showed lower heritabilities, and would not be useful selection criteria. The breeder may, therefore, choose to use intensity of colour at maturity and days from pollination to colour maturity as useful characters in selection programs. However, these should be considered only as broad screening characters because of the poor association between colour intensity and dormancy found in this experiment (and in previous studies). Subsequently, direct selection for dormancy level would be efficacious (*see* 5.12.4)

5.12.3 Embryo maturity

Embryonic maturity at harvest ripeness had a fairly high heritability estimate ($h^2 = 0.589$). The importance of embryonic maturity in germinability has been discussed earlier. The higher heritability estimates obtained for the embryo maturity timing measures, days from pollination to median embryonic maturity ($h^2 = 0.730$) and

days from pollination to full (95 %) embryonic maturity (h^2 = 0.782), are worthy of serious consideration by breeders. The latter measure is a very practicable one for the breeder. These high heritabilities emphasise that this is an important character, and is an important assessment of true physiological maturity.

5.12.4 Dormancy

Dormancy level at harvest ripeness showed very high heritability estimates (h^2 = 0.890). The estimate is unbiased, as a wide sampling of the gene-pool was represented. Clearly, a different mix of cultivars will yield different heritability estimates. The inclusion of additional ripening environments, and of genotype X envronment interactions, would probably lower these estimates. Strand (1983) attributed 65.3 % of the variation in seed dormancy to the environment. The other timing variables measured, days from pollination to 50 % dormancy, or to the end of dormancy, also showed high heritability estimates. Dormancy at harvest ripeness should, therefore, be the most useful measure to take, and very appropriate to the problem of sprouting damage resistance.

However, a plant breeder is unlikely to take serial samples over time, and so will not be able to fit profiles, such as done here. The breeder will take a single sample as near to harvest ripeness as he/she can judge. Therefore the following simple relationship between dormancy levels and grain moisture, around harvest ripeness, was investigated, to see if adjustment of this single measure would improve its reliability. The regression relationships found were:

1. Where only the variability in red - grained cultivars is considered

$$D = 171.6343 - 8.4864\ M$$

where, D = dormancy level, and,
M = moisture level.

s.e. β_0 = 130.73604 s.e. β_1 = 10.16681 R^2 = 0.0379
F = 0.697 (not significant)

2. Where variability in dormancy levels for both white and red cultivars is considered.

$$D = 157.99594 - 9.53368\ M$$

where, D = dormancy level, and,
M = moisture level.

s.e. β_0 = 118.99951 s.e. β_1 = 9.21220 R^2 = 0.0274
F = 1.071 (not significant)

It is noted that neither of these regressions showed any utility (low R^2 and non - significance). So there is no value in adjusting, as far as these results show. These results suggest that the "errors" in dormancy assessment arising from small variation in time (about harvest ripeness) would be trivial.

5.12.5 Future research

These results highlight possible areas of future research, as follows.

1. Study of the biology and metabolism of the graincoats of developing wheat grains would be useful to test the hypo-oxia / dormancy proposal. Such studies should include investigating the enzyme kinetic properties of the polyphenol oxidases and the level of hypo-oxia.
2. A reappraisal of flavanol chemistry needs to be conducted in wheat grains with the view of explaining the causes for the decline in flavanol levels in both red and white wheat grains, even though the latter do not lose lose it because of colouring.

5.13 Conclusions / Summary

1. The study has shown that the presence of red colour is necessary for dormancy to occur in wheat.

2. This association between colour and dormancy may be due to hypo - oxia imposed by colour development, but not (probably) because of ABA increase. The *timing* of these constraints at particular stages of embryonic growth and development may determine onset of embryonic dormancy.

3. Variability of dormancy levels among the red wheats was found showing that the number of red genes present is immaterial in determining the level of dormancy.

4. Variation in intensity of redness between cultivars having the same number of red genes and amongst cultivars having different numbers of red genes was observed suggesting that colour intensity is not simply due to additive gene dosage.

5. The importance of timing of events, in addition to redness, was demonstrated, indicating that dormancy may be a polygenic phenomenon. This suggests that selection for dormancy should be direct, e.g. using harvest ripeness as the base-line for selection.

6. The high heritability estimates found for colour level at harvest ripeness indicate that selection for colour can be useful in selection programs aimed at increasing the genetic advance of characters that are correlated with graincoat colour. As far as dormancy is concerned, however, further direct selection (using methods such as these) should follow.

7. The flavanol levels were found to be similar in all cultivars and did not show a relationship with dormancy, embryo maturity or colour development.

8. Dormancy was not found in these white cultivars previously reported to have it. This may have come about because of the characters juxtapositions arising from the single environment involved. However, the precision and definition of methods may have been important also. Nevertheless, some of the timing data did reveal "hidden" dormancy in one of the wheats (Tordo).

9. The timing, duration or amount of whole grain abscisic acid was not associated with dormancy at harvest ripeness.

10. The gibberellin insensitivity of the *Rht / Gai* cultivars was not apparent in this study. This may have been because the dormancy breaking methods compensated for natural early development blocks to GA metabolism.

Let the thick curtain fall; I better know than all
How little I have gained, how vast the unattained.

Others shall sing the song, others shall right the wrong,
Finish what I begin, and all I fail of win.

What matter I or they? Mine or another's day,
So the right word be said, and life the sweeter made?

Whittier

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Appendices

A 3.1

Seedling Potting mixture used in vernalization of Brevor seedlings.

Dolamite	3 kg/m^3
Agricultural Lime	3 kg / m^3
PG mix	1.5 kg / m^3
Substrate blend	100 % bark

A 3.2

Materials used in ELISA assay

1. Chemicals

a) **Tris (hydroxymethyl) aminomethane** (TRIS), enzyme grade
Formular: $C_4H_{11}NO_3$,
MWt : 121.14
Purity: 99.8 %
M.P. range: 173 -175 °C
Lot number: 66836
Supplier: United States Biomedical Corp., Cleveland, Ohio 44128

b) **ABA Monoclonal antibody** (Phytodetek™). Mab to free *cis, trans*(+)ABA (Mertens *et al.*, 1983)
Lot number: 66836; Part number: 8015
Expiry date: 9/06/96
Supplier: Idetek, Inc., 1057 Sneath Lane, San Bruno, CA 94066.

Two mg MAb was mixed into 135 mL TBS, containing 0.2 % (w/v) BSA. Aliquots of 1 mL of Mab solution were stored at - 20 °C. Before ELISA assay a 1 mL aliquot was thawed and diluted in TBS to a final volume of 11 mL which is sufficient for the assay wells of one microtitration plate.

c) **Rabbit Anti-Mouse IgG (whole molecule) alkaline phosphatase conjugate** (affinity isolated antibody developed in rabbit) 052H-8845, referred to as 'second antibody' in assay procedure.

Lot number: 034H8807; Product number: A-4312

Supplier: Sigma Immuno Chemicals, P. O. Box 14508, St Louis, MO 63178 USA

d) **Bovine Serum Albumin** (BSA), ELISA grade

Rabbit anti-mouse alkaline phosphatase conjugate was diluted 1:1000 in TBS.

Purity: 96-99 % albumin

Nitrogen content: 15.9 %

H_2O content: 0.9 %

Initial fraction by cold alcohol precipitation, Fraction V, remainder mostly globulins; Dessicate.

Lot number: 64H0643, A-3350

e) ***p*-Nitrophenyl Phosphate**, disodium (Sigma 104® phosphatase substrate), 6 H_2O per mole. Referred to as PNPP in assay procedure.

Supplier: Sigma Chemical Company, P. O. Box 14508, St Louis, MO 63178, USA.

Tel. 314 - 771 - 5750

Anhydrous MWt: 263.1

Lot number: 123H50061

Supplier: Sigma Diagnostics, St Louis, MO 63178, USA

The substrate, p-nitrophenyl phosphate, was prepared at a concentration of 1 mg/mL in 0.05 M $NaHCO_3$ (pH 9.6).

1. **Tween®20** (Polyethylene (20 sorbitan mono-laurate)

   Lot number: 005886

   Supplier: AJAX Chemicals, 9 Short Street, Auburn, N.S.W. 2144, Australia

a) **(±)-2-cis, 4-trans-Abscisic acid**; synthetic

Purity: 98 %

FWt: 264.32

M.P.: range: 188-190 °C

Lot number: 1 Sept. 1989

Packaging Cadpak PA-131

Supplier: Aldrich Chemical Company, P.O. Box 355, Milw. WI53201

Tel. 414-273-3850

Dissolve 5 mg (±)-2-cis, 4-trans-ABscisic acid in 2mL methanol and make up to 100 mL with TBS. The resulting solution contains 5 x 10^{-6} picograms ABA/100 µl. Do a dilution series with TBS to give a range of stndards between 0.1 - 5000 pg/100µl.

b) TBS (Tris buffered saline):

6.05 g TRIS; 0.2 mg $MgCl_2$; 8.8g NaCl per litre

Adjust to pH 7.8

c) Washing buffer (TBST - BSA):

TBS containing 0.05 % (v/v) Tween®20 and 0.1 % (w/v) BSA prepared on day of use

d) $NaHCO_3$ 0.05 M:

4.2 g $NaHCO_3$, Analar grade, dissolved in 1 litre H_2O

Adjust pH to 9.6

e) ABA-4'-BSA conjugate:

The conjugate was prepared according to Weiler (1979) and lyophilized. Conjugate was suspended in 0.05 M $NaHCO_3$ (pH 9.6), at a concentration of 7 mg/mL and stock aliquots 30 µl were kept frozen at -20 °C. On day of use, each 30 µl aliquot was diluted with 20 mL 0.05 M $NaHCO_3$ (pH 9.6), enough to coat the assay wells of one microtitration plate.

A 3.5. Analysis of Variance for (±)-2-cis, 4-trans-Abscisic acid standard sample.

Dependent variable: Log(concentration)

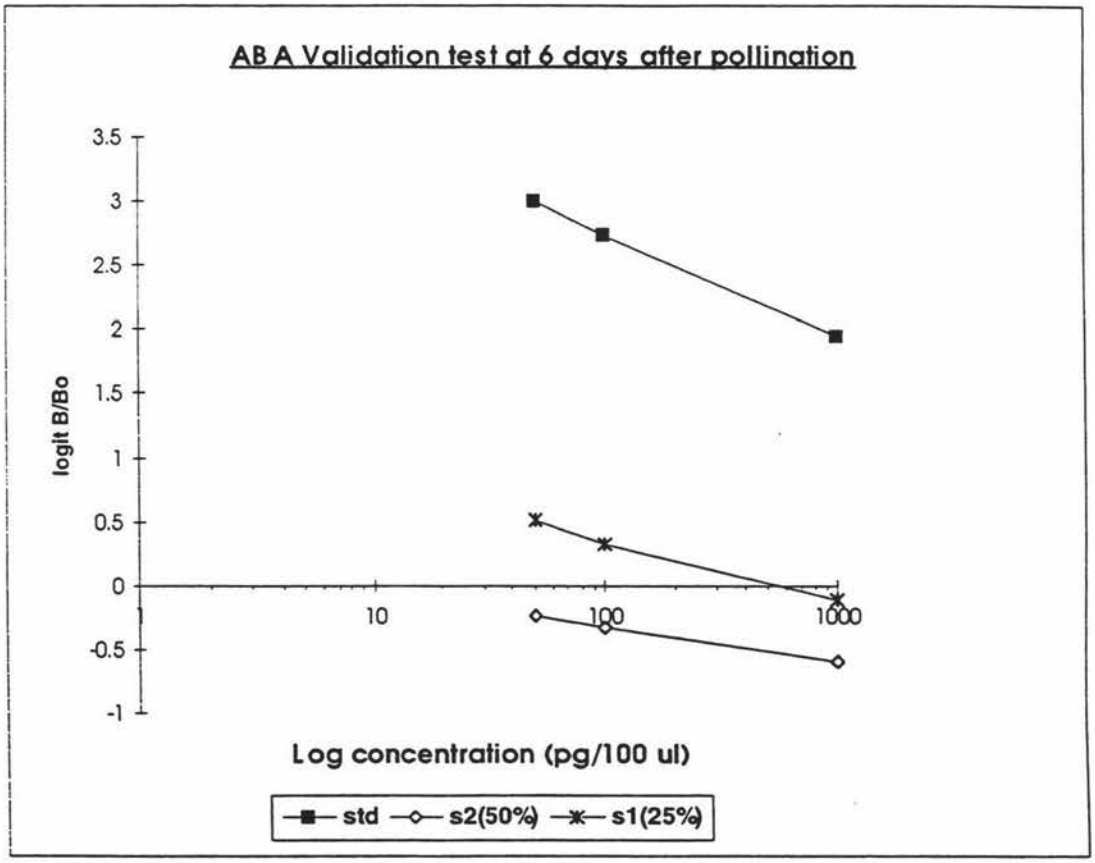
Source	DF	Sum of Squares	Mean Square	F Value	Prob > F
Model	1	8.48672	8.486719	6847.69	0.0000036
error	3	0.00372	0.001239		
Corrected Total	4	8.49044			

Root MSE	0.035204are	R-square	0.999562
C.V.	10.43949	Adj R-sq	0.999416

A 3.6. Data for validation test for the ELIZA procedure for ABA analysis in wheat grains: Absorbances at 410 nm

ABA added	Std Abs	Sample 1 (1:4) Abs	Sample 2 (1:2) Abs
50	0.938	0.640	0.475
100	0.926	0.601	0.453
1000	0.867	0.500	0.395
NSB			
B_0			

Figure A 3.1. Validation for the ELIZA procedure for ABA analysis in wheat grains



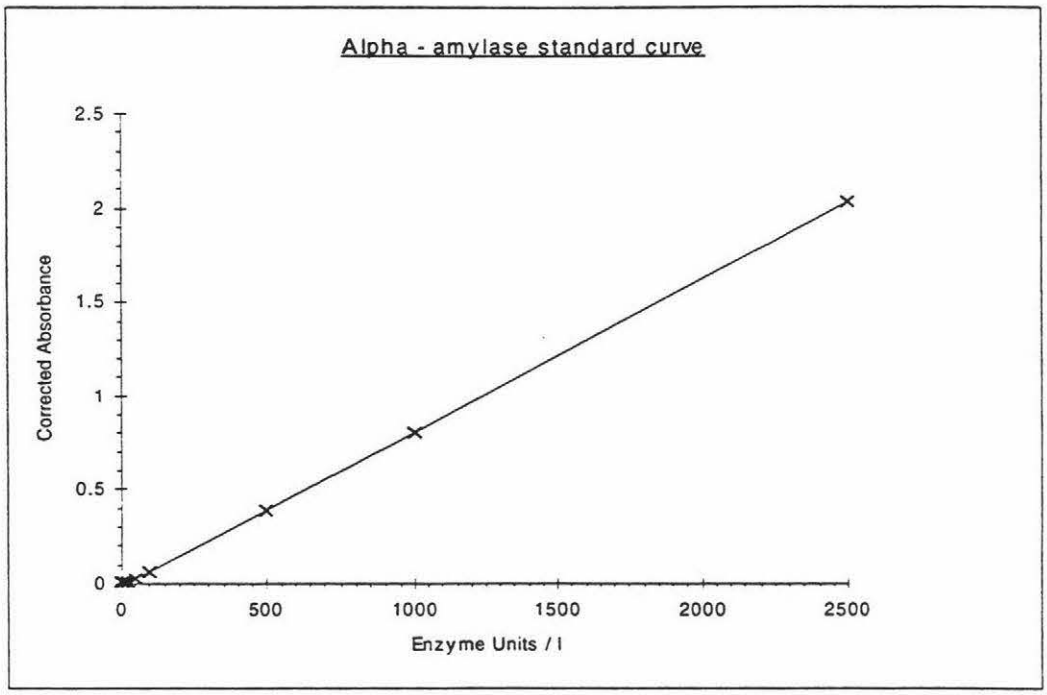
A 3.7. Tris - maleate buffer (α - amylase assay)

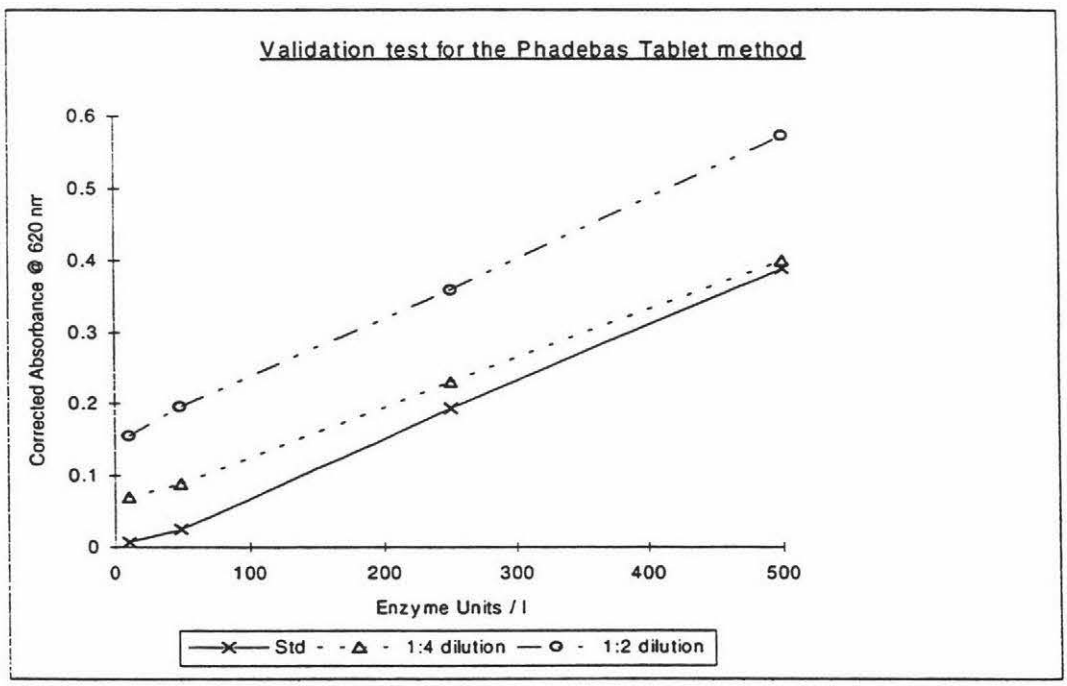
Measure 6.05 g Tris, 5.8 g Maleic cid, 153.9 ml 0.5 M NaOH, 1.11 g $Ca Cl_2$ and add 600 ml distilled water. Adjusted pH to 6.2 and make up the solution to 1 litre.

A 3.8. Data for validation test for the Phadebas® Tablet method for the estimation of α - amylase activity in developing wheat grains: values are the mean absorbance at 620 nm for two replicates

α - amylase added	Std Abs	Sample 1 (1:4) Abs	Sample 2 (1:2) Abs
10	0.054	0.117	0.202
50	0.073	0.135	0.244
100	0.0	0.278	0.405
1000	0.848	0.4471	0.620

A 3.2. (a) Standard curve for Alpha - amylase (b) Validation test for the Phadebas tablet method.





Appendix 2.

Table A 4.1 (a). The Richards function for grain moisture content (statistics by replicate).

cv. Gamenya

Replicate:	1	2	3	4
Statistic				
A	76.00000	76.98707	76.00372	76.00160
s.e. A	16.37485	8.83413	10.26534	9.36251
B	-3.15227	-4.79884	-4.04629	-5.25104
s.e. B	1.52752	1.79329	1.42365	1.62053
K	-0.10004	-0.14999	-0.09997	-0.14991
s.e. K	0.10117	0.11562	0.11362	0.10925
V	0.89896	1.00173	1.00126	1.00329
s.e. V	1.27410	1.95038	1.28329	1.05698
F_{regr}	174.8324***	128.3165***	648.7795***	276.3575***
R^2	0.9836	0.9676	0.9947	0.9883

significance level
*** $P < 0.001$

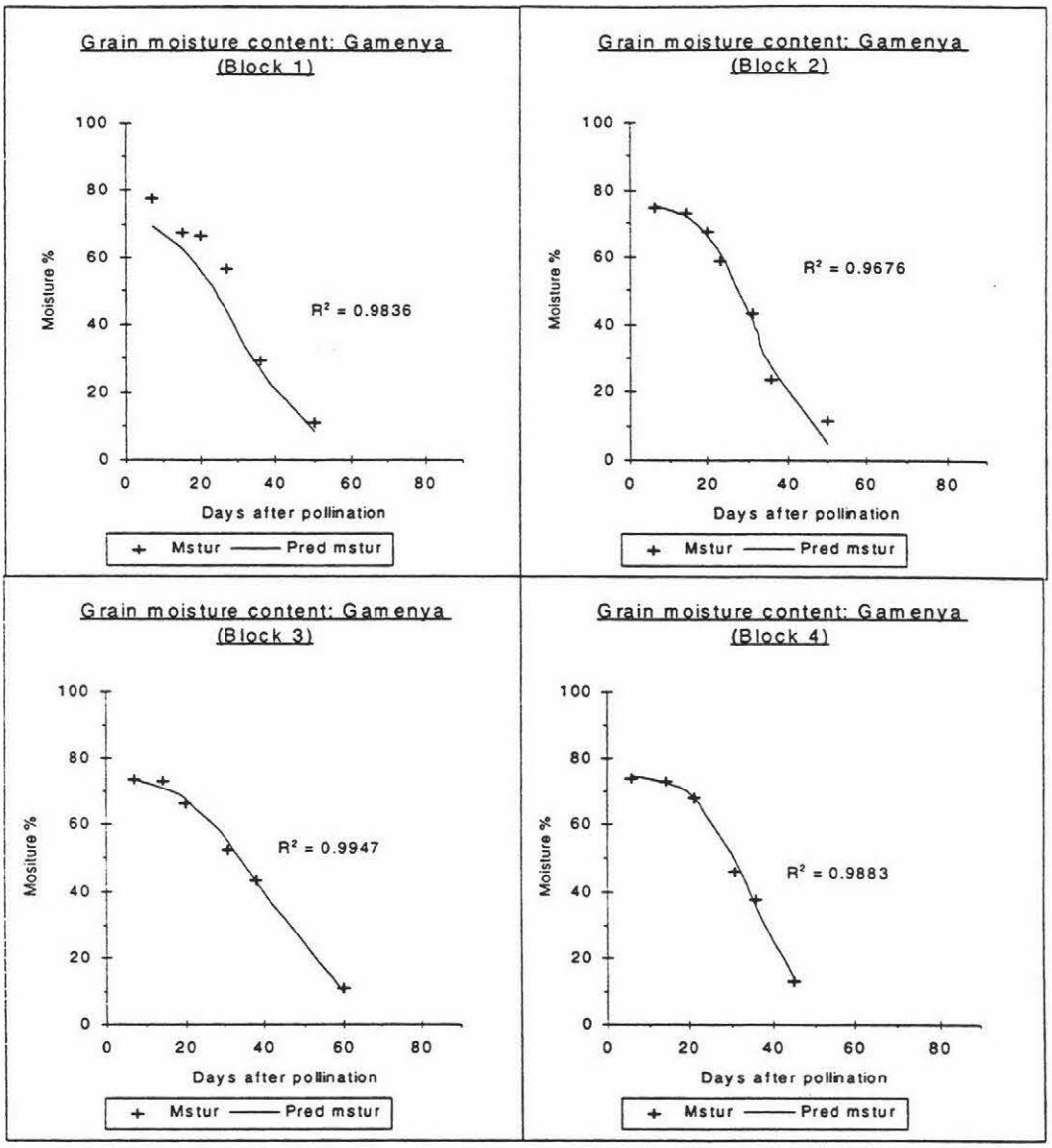

Grain moisture content: Gamenya (Block 1)
Moisture %
Days after pollination
R² = 0.9836
Mstur
Pred mstur
Grain moisture content: Gamenya (Block 2)
Moisture %
Days after pollination
R² = 0.9676
Mstur
Pred mstur
Grain moisture content: Gamenya (Block 3)
Mositure %
Days after pollination
R² = 0.9947
Mstur
Pred mstur
Grain moisture content: Gamenya (Block 4)
Moisture %
Days after pollination
R² = 0.9883
Mstur
Pred mstur

Table A 4.1 (b). The Richards function for grain moisture content (statistics by replicate).

cv. Tordo

Replicate:	1	2	3	4
Statistic				
A	79.00000	78.00735	77.82172	76.00031
s.e. A	16.80056	7.43526	14.30413	34.34203
B	-4.00000	-4.10078	-3.69083	-3.15074
s.e. B	1.64028	0.87876	1.49154	2.50878
K	-0.10000	-0.09988	-0.09990	-0.10005
s.e. K	0.14102	0.07394	0.10806	0.23973
V	1.05000	1.05351	1.00128	0.89883
s.e. V	4.54513	2.36768	2.56141	3.26903
F_{regr}	86.3665***	295.7898***	104.1294***	120.8597***
R^2	0.9428	0.9758	0.9631	0.9604

significance level
*** $P < 0.001$

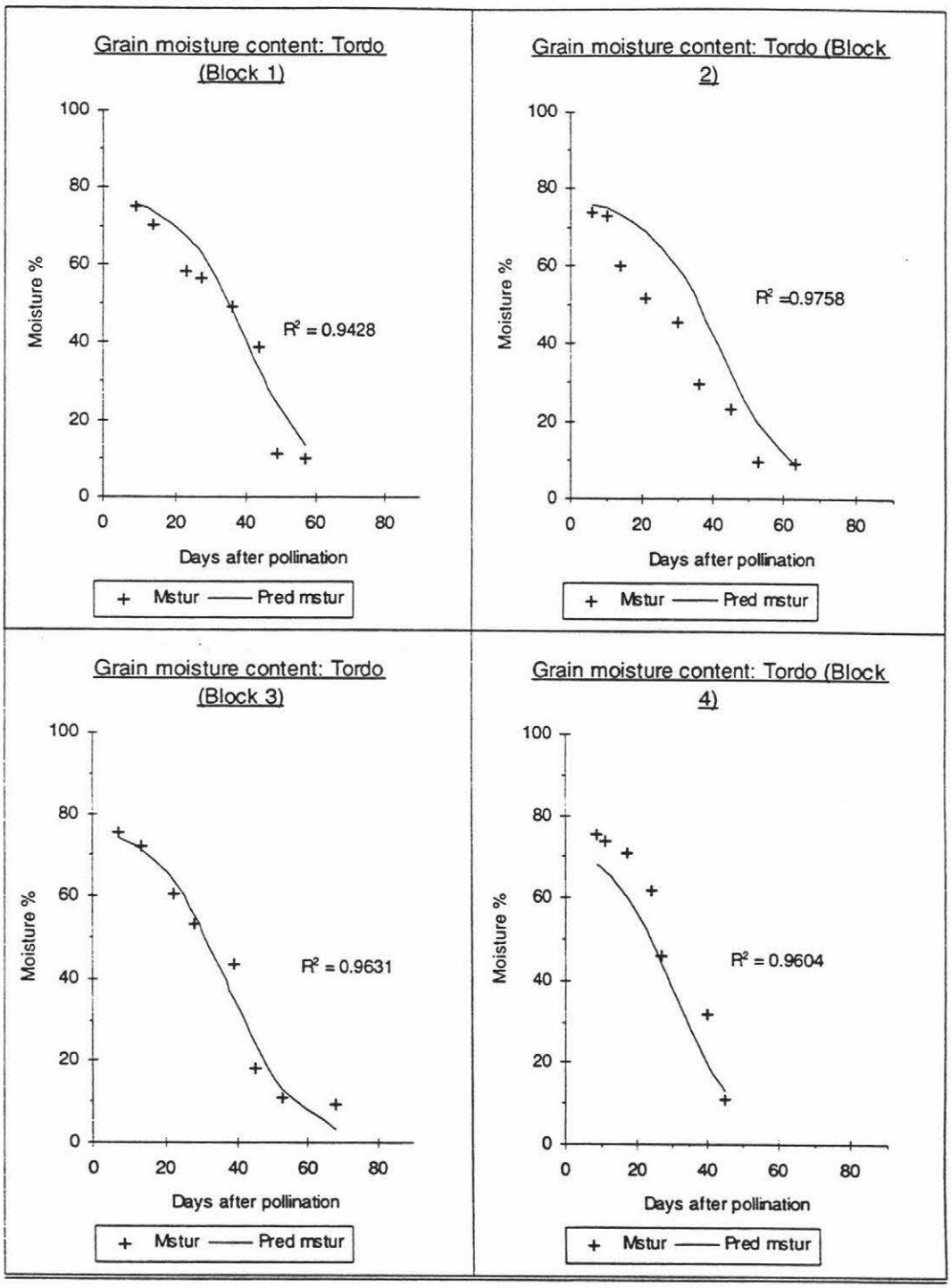

Grain moisture content: Tordo (Block 1)
Moisture %
Days after pollination
R² = 0.9428
+ Mstur —— Pred mstur
Grain moisture content: Tordo (Block 2)
Moisture %
Days after pollination
R² =0.9758
+ Mstur —— Pred mstur
Grain moisture content: Tordo (Block 3)
Moisture %
Days after pollination
R² = 0.9631
+ Mstur —— Pred mstur
Grain moisture content: Tordo (Block 4)
Moisture %
Days after pollination
R² = 0.9604
+ Mstur —— Pred mstur

Table A 4.1 (c). The Richards function for grain moisture content (statistics by replicate).

cv. Kenya 321

Replicate:	1	2	3	4
Statistic				
A	78.98443	76.00000	79.00000	76.97221
s.e. A	10.91419	8.09775	11.88682	8.65524
B	-4.15103	-4.08160	-3.65000	-3.84519
s.e. B	1.19392	1.13861	1.01565	0.98254
K	-0.09968	-0.09972	-0.10000	-0.09998
s.e. K	0.10475	0.07618	0.09648	0.06807
V	1.06024	1.00000	1.00000	1.00243
s.e. V	3.35232	1.07618	2.41001	1.56734
F_{regr}	157.8572***	265.4525***	257.1138***	233.6193***

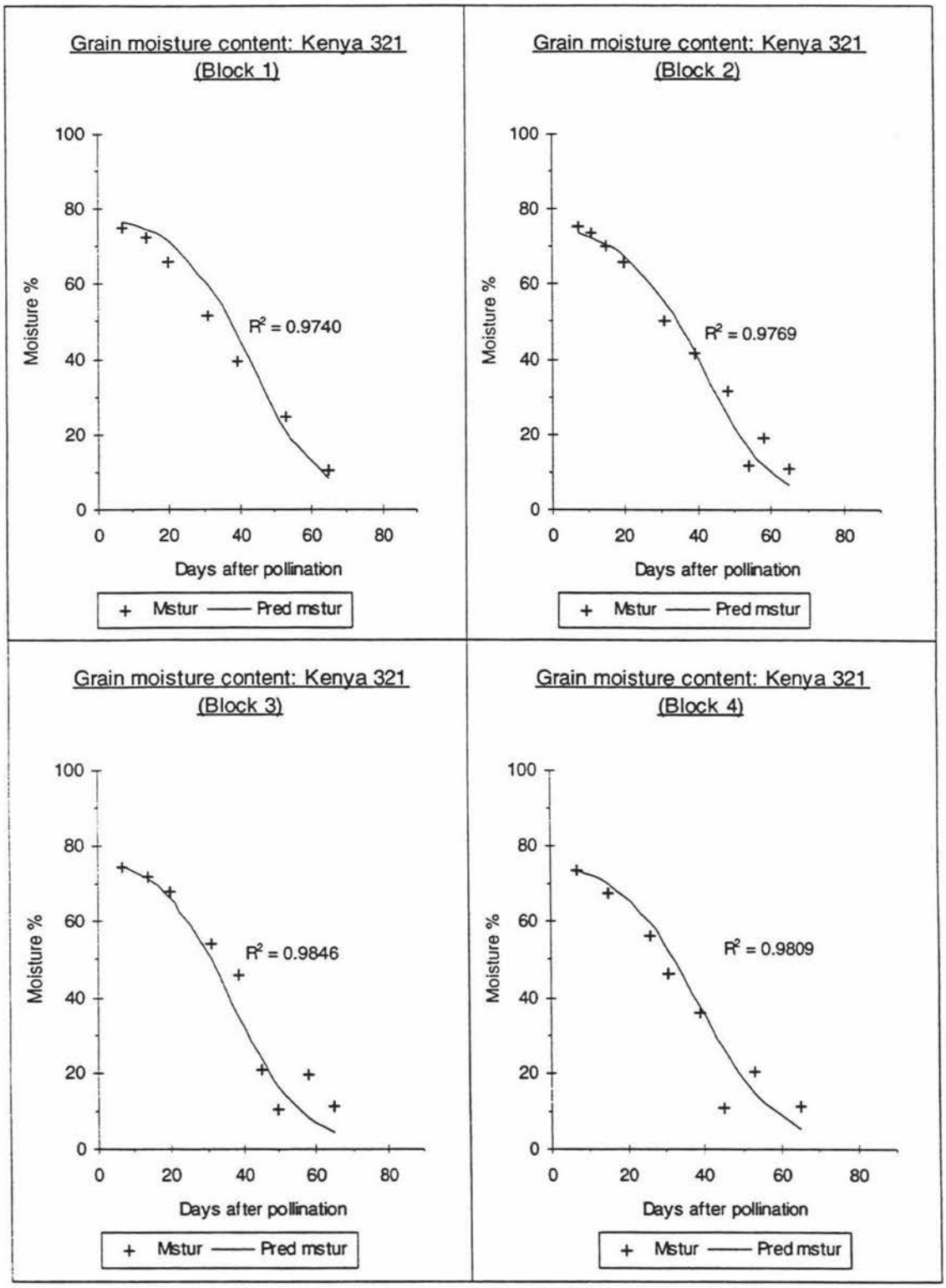

Grain moisture content: Kenya 321 (Block 1)
Moisture %
Days after pollination
R² = 0.9740
+ Mstur —— Pred mstur
Grain moisture content: Kenya 321 (Block 2)
Moisture %
Days after pollination
R² = 0.9769
+ Mstur —— Pred mstur
Grain moisture content: Kenya 321 (Block 3)
Moisture %
Days after pollination
R² = 0.9846
+ Mstur —— Pred mstur
Grain moisture content: Kenya 321 (Block 4)
Moisture %
Days after pollination
R² = 0.9809
+ Mstur —— Pred mstur

Table A 4.1 (d). The Richards function for grain moisture content (statistics by replicate).

cv. Brevor

Replicate:	1	2	3	4
Statistic				
A	77.01793	77.85491	80.00000	78.00199
s.e. A	9.67748	8.63123	8.71577	9.83679
B	-3.65656	-5.93537	-5.95325	-3.95012
s.e. B	1.19726	2.03652	2.79256	1.35032
K	-0.09980	-0.14963	-0.14995	-0.09999
s.e. K	0.07098	0.19962	0.20960	0.07840
V	0.95522	1.05688	1.09647	1.00024
s.e. V	1.26658	1.63259	1.39170	1.72660
F_{regr}	229.8569***	156.8867***	121.7258***	169.5529***

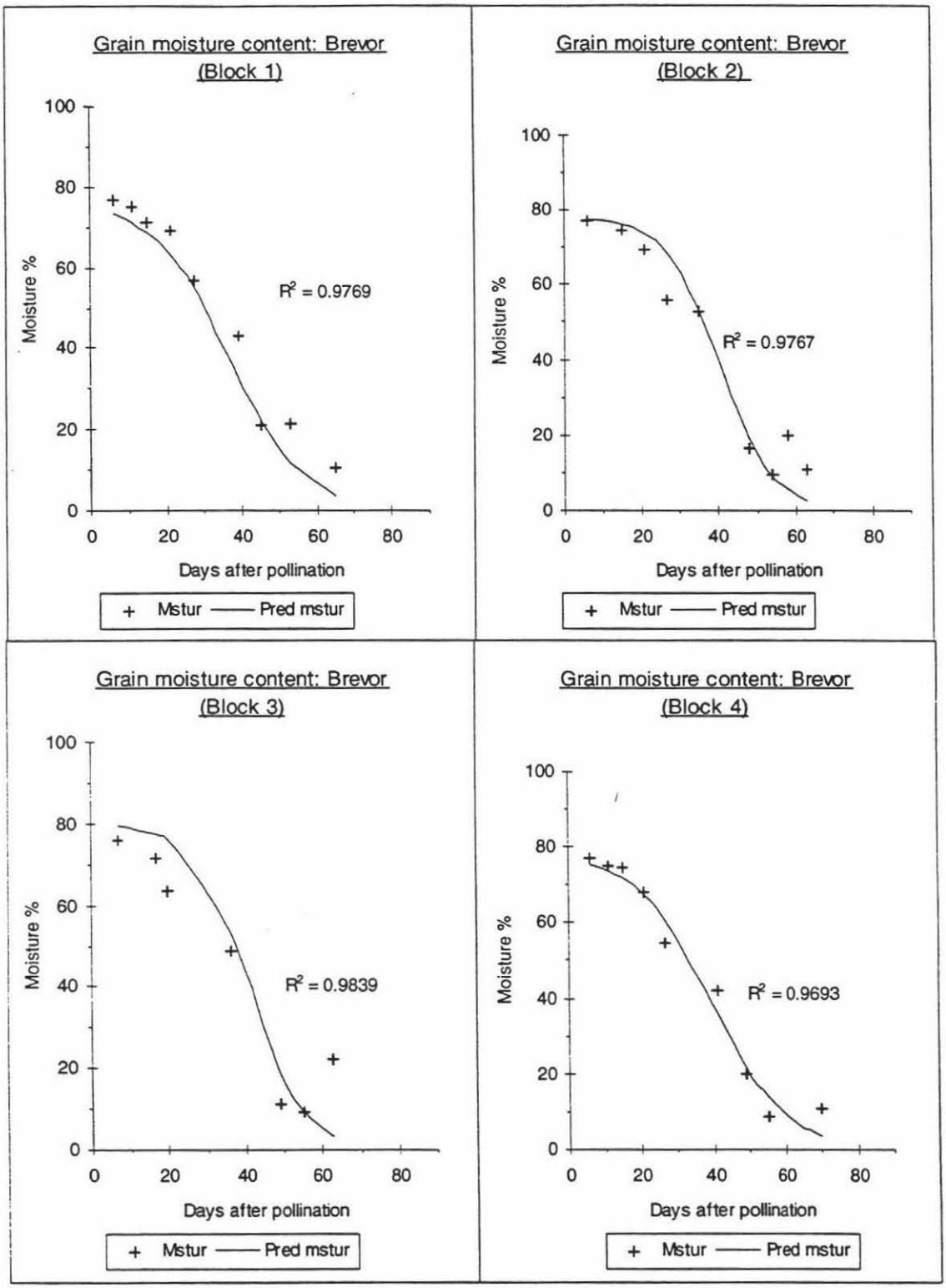

Grain moisture content: Brevor (Block 1)
Moisture %
Days after pollination
R² = 0.9769
+ Mstur —— Pred mstur
Grain moisture content: Brevor (Block 2)
Moisture %
Days after pollination
R² = 0.9767
+ Mstur —— Pred mstur
Grain moisture content: Brevor (Block 3)
Moisture %
Days after pollination
R² = 0.9839
+ Mstur —— Pred mstur
Grain moisture content: Brevor (Block 4)
Moisture %
Days after pollination
R² = 0.9693
+ Mstur —— Pred mstur

Table A 4.1 (e). The Richards function for grain moisture content (statistics by replicate).

Isis

Replicate:	1	2	3	4
Statistic				
A	78.00000	79.99552	77.88599	79.0399
s.e. A	12.42478	20.42189	13.54478	24.16238
B	-3.20000	-3.15468	-3.19586	-3.80332
s.e. B	1.048178	1.37510	1.14729	2.22168
K	-0.10000	-0.09958	-0.10035	-0.09964
s.e. K	0.07806	0.11418	0.08467	0.19854
V	0.95000	1.00009	0.94250	1.06112
s.e. V	1.25044	2.27663	1.29588	6.18408
F_{regr}	253.0443***	133.5798***	220.7324***	37.4548**

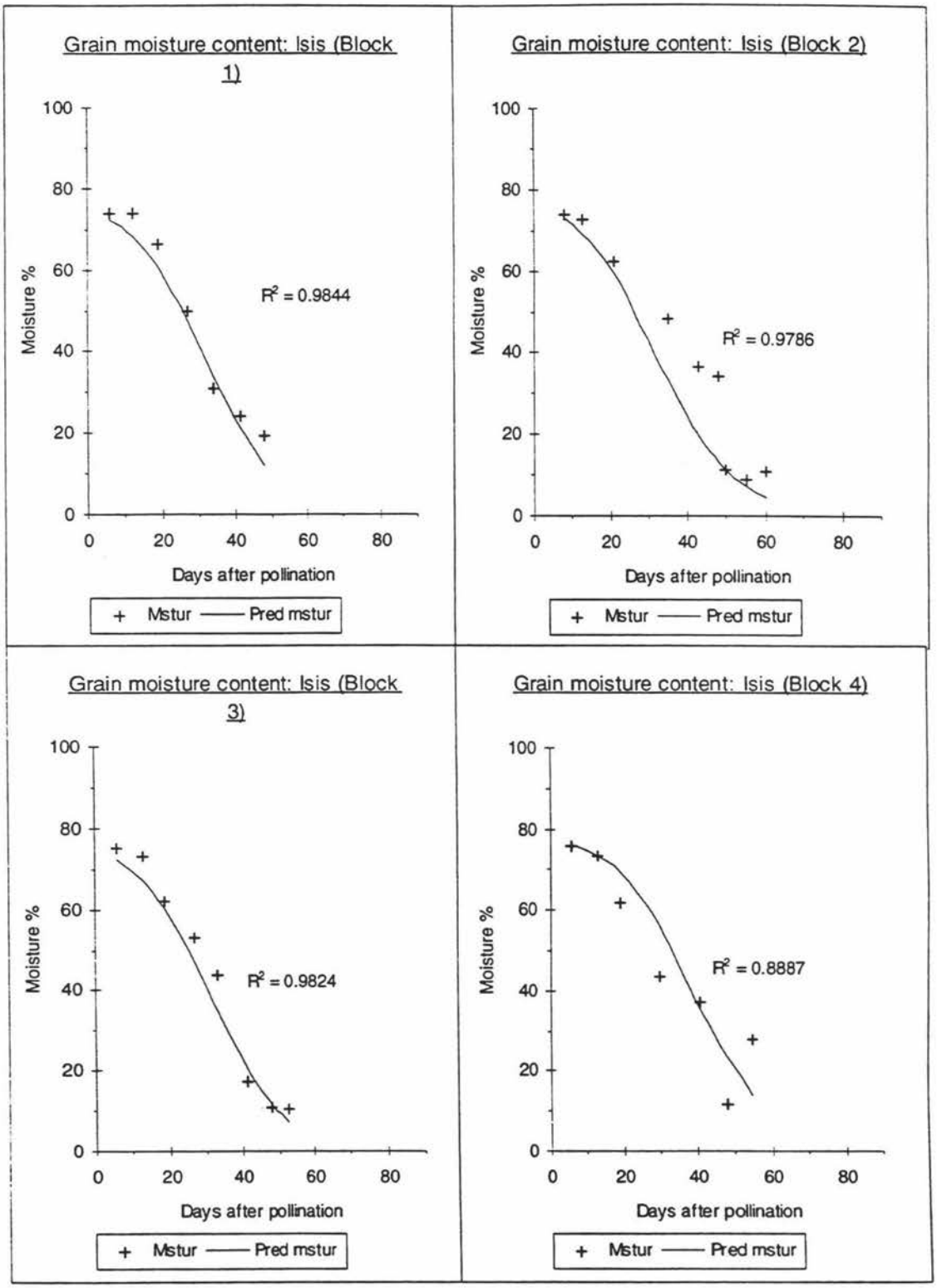

Grain moisture content: Isis (Block 1)
Moisture %
Days after pollination
R² = 0.9844
+ Mstur —— Pred mstur
Grain moisture content: Isis (Block 2)
Moisture %
Days after pollination
R² = 0.9786
+ Mstur —— Pred mstur
Grain moisture content: Isis (Block 3)
Moisture %
Days after pollination
R² = 0.9824
+ Mstur —— Pred mstur
Grain moisture content: Isis (Block 4)
Moisture %
Days after pollination
R² = 0.8887
+ Mstur —— Pred mstur

Table A 4.1 (f). The Richards function for grain moisture content (statistics by replicate).

cv. Sonora 64A

Replicate:	1	2	3	4
Statistic				
A	76.90721	76.00156	79.93445	76.00350
s.e. A	5.40657	10.32695	20.23952	16.90477
B	-6.54701	-3.84994	-3.25105	-3.34882
s.e. B	1.86671	1.50246	1.24805	1.45110
K	-0.19995	-0.09998	-0.10035	-0.09995
s.e. K	0.15535	0.13962	0.12221	0.10712
V	1.10146	1.00037	1.04182	1.00186
s.e. V	3.28029	2.25601	3.13760	2.21370
F_{regr}	197.7360***	575.4724***	113.4710***	104.6183***

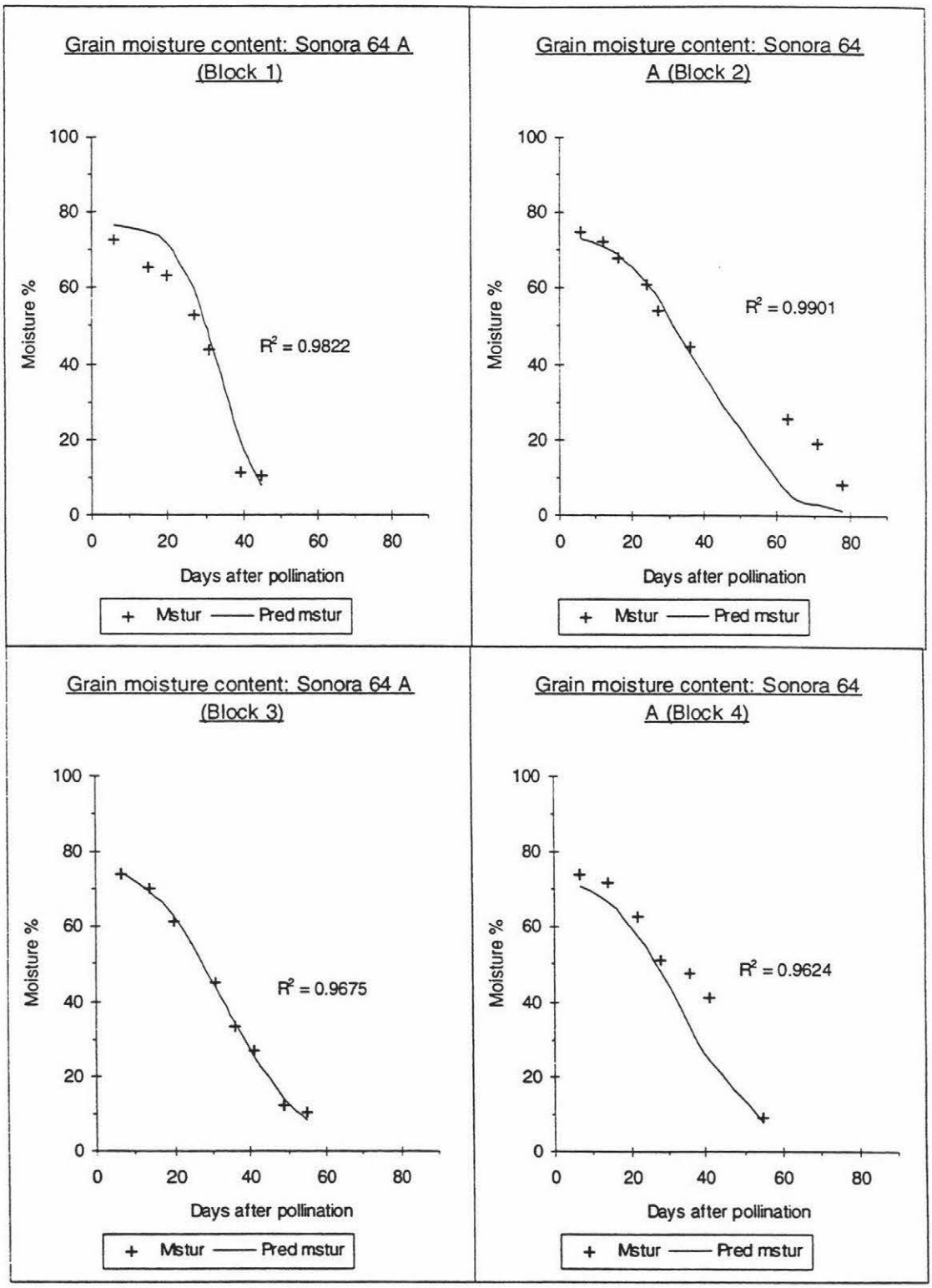

Grain moisture content: Sonora 64 A (Block 1)
Moisture %
Days after pollination
R² = 0.9822
+ Mstur —— Pred mstur
Grain moisture content: Sonora 64 A (Block 2)
Moisture %
Days after pollination
R² = 0.9901
+ Mstur —— Pred mstur
Grain moisture content: Sonora 64 A (Block 3)
Moisture %
Days after pollination
R² = 0.9675
+ Mstur —— Pred mstur
Grain moisture content: Sonora 64 A (Block 4)
Moisture %
Days after pollination
R² = 0.9624
+ Mstur —— Pred mstur

Table A 4.1 (g). The Richards function for grain moisture content (statistics by replicate).

cv. Thatcher

Replicate:	1	2	3	4
Statistic				
A	77.96004	76.98124	77.93637	78.99594
s.e. A	14.79917	14.23977	20.19785	6.47058
B	-3.79710	-3.80050	-3.09662	-5.65197
s.e. B	1.39743	1.45008	1.64266	1.54356
K	-0.09961	-0.09956	-0.09994	-0.14993
s.e. K	0.11672	0.10684	0.12560	0.10902
V	1.06071	1.06150	0.95458	1.10288
s.e. V	3.74793	3.27707	1.89874	3.27859
F_{regr}	88.3389***	91.6983***	92.5065***	162.7736***

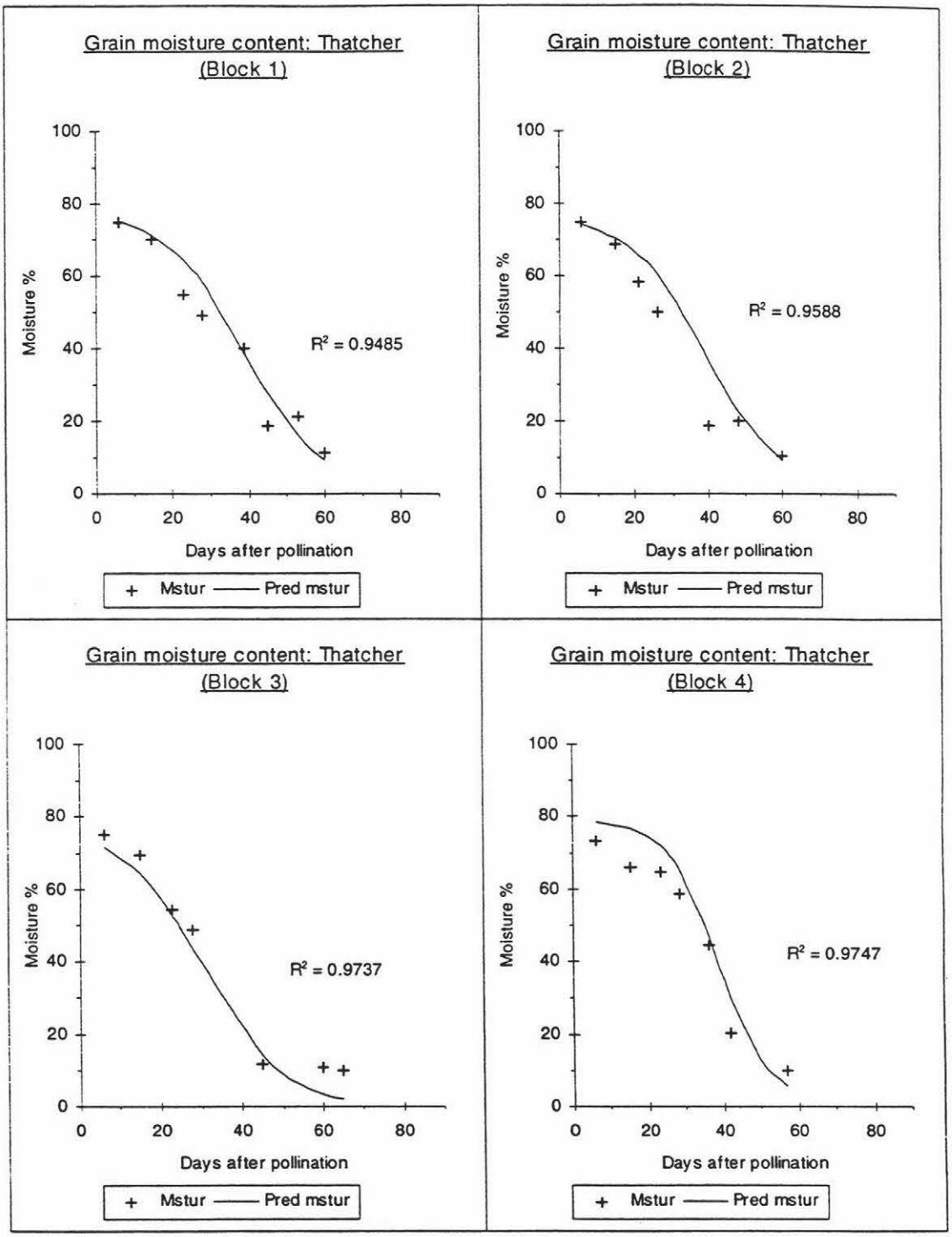

Grain moisture content: Thatcher (Block 1)
Moisture %
100
80
60
40
20
0
0
20
40
60
80
R² = 0.9485
Days after pollination
Mstur
Pred mstur
Grain moisture content: Thatcher (Block 2)
Moisture %
R² = 0.9588
Days after pollination
Mstur
Pred mstur
Grain moisture content: Thatcher (Block 3)
Moisture %
R² = 0.9737
Days after pollination
Mstur
Pred mstur
Grain moisture content: Thatcher (Block 4)
Moisture %
R² = 0.9747
Days after pollination
Mstur
Pred mstur

Table A 4.1 (h). The Richards function for grain moisture content (statistics by replicate).

cv. La Prevision

Replicate:	1	2	3	4
Statistic				
A	76.053497	76.00000	77.54273	78.75772
s.e. A	9.72501	9.6430	9.82160	10.43216
B	-3.19933	-3.64571	-2.08454	-3.28736
s.e. B	1.30213	1.26307	1.41015	1.63040
K	-0.9737	-0.10583	-0.08454	-0.09991
s.e. K	0.062361	0.07540	0.07398	0.06329
V	0.92538	0.94261	0.94743	0.95652
s.e. V	1.13645	1.10195	1.11011	0.99288
F_{regr}	120.5586***	232.6131***	122.7959***	199.7354***

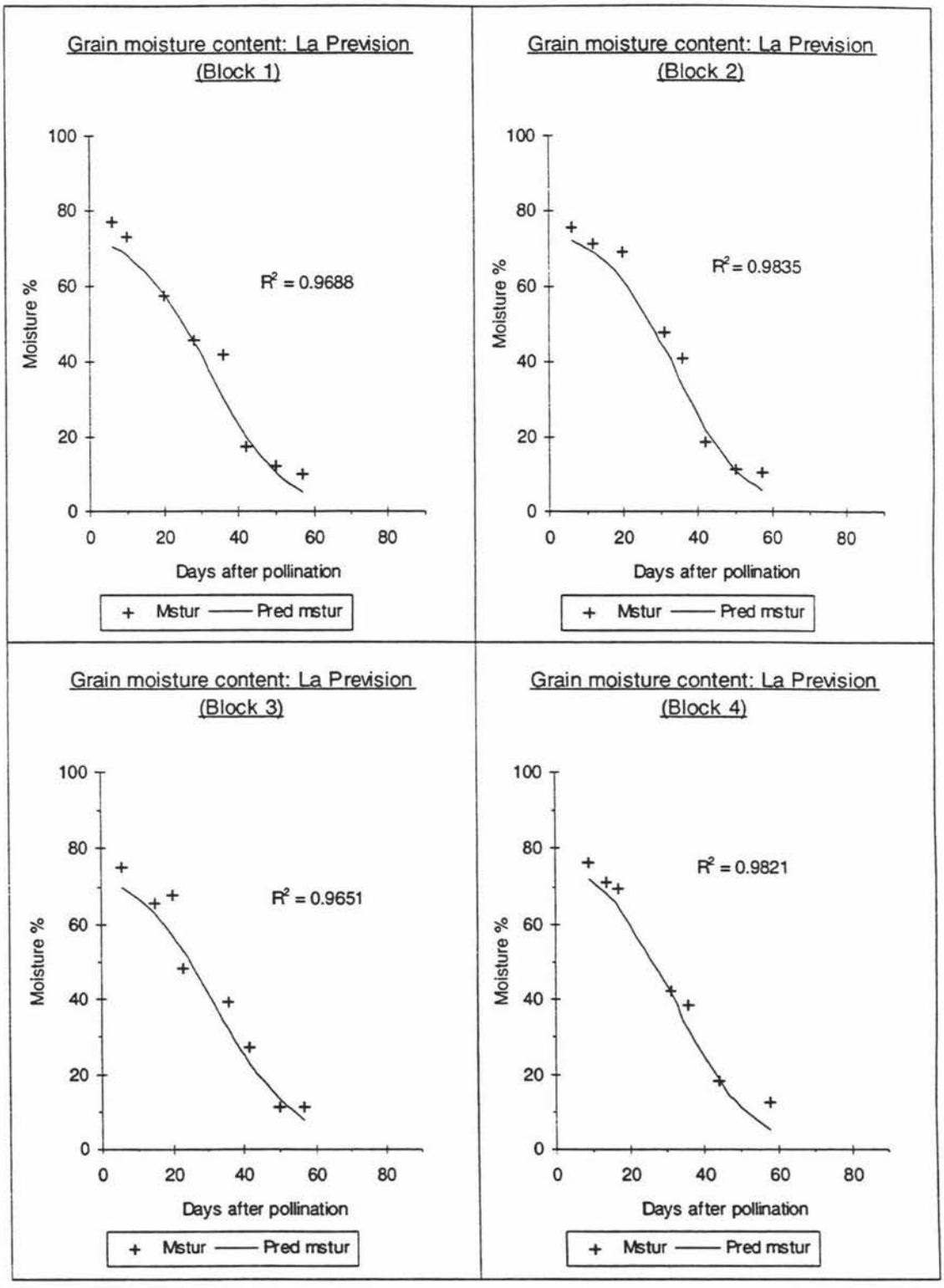

Grain moisture content: La Prevision (Block 1)
Moisture %
Days after pollination
R² = 0.9688
+ Mstur —— Pred mstur
Grain moisture content: La Prevision (Block 2)
Moisture %
Days after pollination
R² = 0.9835
+ Mstur —— Pred mstur
Grain moisture content: La Prevision (Block 3)
Moisture %
Days after pollination
R² = 0.9651
+ Mstur —— Pred mstur
Grain moisture content: La Prevision (Block 4)
Moisture %
Days after pollination
R² = 0.9821
+ Mstur —— Pred mstur

Table A 4.1 (i). The Richards function for grain moisture content (statistics by replicate).

cv. Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
A	79.97682	76.96438	77.00000	76.96739
s.e. A	31.27929	6.85400	10.33848	22.46988
B	-2.69711	-3.69866	-3.75000	-3.64552
s.e. B	2.32375	0.84729	1.41214	3.05283
K	-0.09987	-0.10006	-0.10000	-0.09985
s.e. K	0.14148	0.05068	0.07154	0.16150
V	0.90611	0.94861	0.95000	0.95706
s.e. V	1.50927	0.86763	1.14313	2.45852
F_{regr}	60.4617***	459.4409***	180.6236***	44.0529**

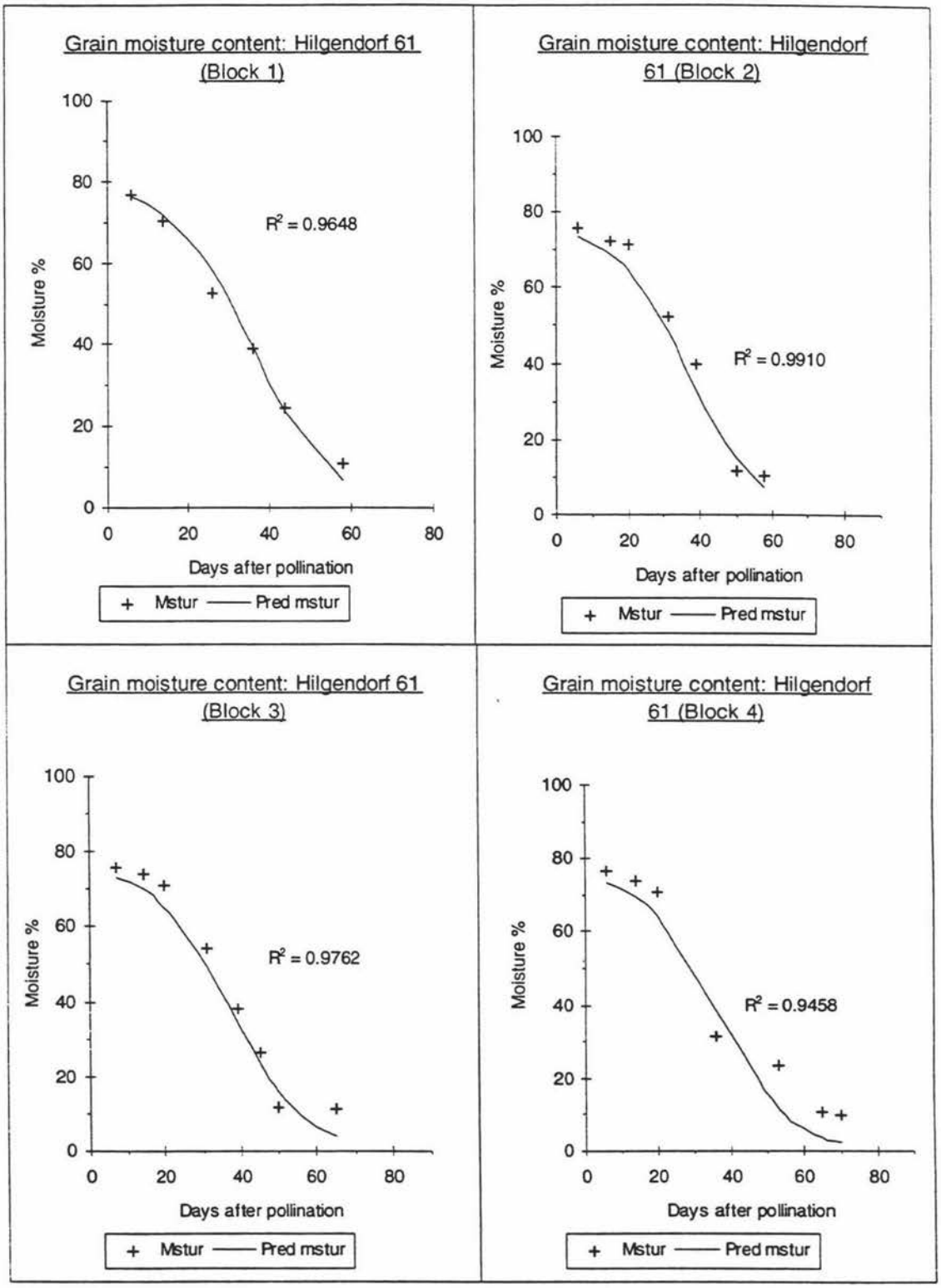

Grain moisture content: Hilgendorf 61 (Block 1)
Moisture %
Days after pollination
R² = 0.9648
+ Mstur —— Pred mstur
Grain moisture content: Hilgendorf 61 (Block 2)
Moisture %
Days after pollination
R² = 0.9910
+ Mstur —— Pred mstur
Grain moisture content: Hilgendorf 61 (Block 3)
Moisture %
Days after pollination
R² = 0.9762
+ Mstur —— Pred mstur
Grain moisture content: Hilgendorf 61 (Block 4)
Moisture %
Days after pollination
R² = 0.9458
+ Mstur —— Pred mstur

Table A 4.1 (j). The Richards function for grain moisture content (statistics by replicate).

cv. Karamu

Replicate:	1	2	3	4
Statistic				
A	80.00000	76.00000	76.00000	80.00000
s.e. A	11.03936	15.31874	13.40618	12.64782
B	-3.60000	-3.28120	-3.65000	-3.80118
s.e. B	1.011242	1.31664	1.59433	1.10872
K	-0.10000	-0.09530	-0.10000	-0.09976
s.e. K	0.07904	0.10567	0.10023	0.09675
V	1.00000	0.93964	0.95000	1.05662
s.e. V	1.79164	1.91194	1.69966	2.98743
F_{regr}	214.7756***	169.7865***	129.1745***	142.9464***
R^2	0.9803	0.9778	0.9673	0.9646

significance level

*** $P < 0.001$

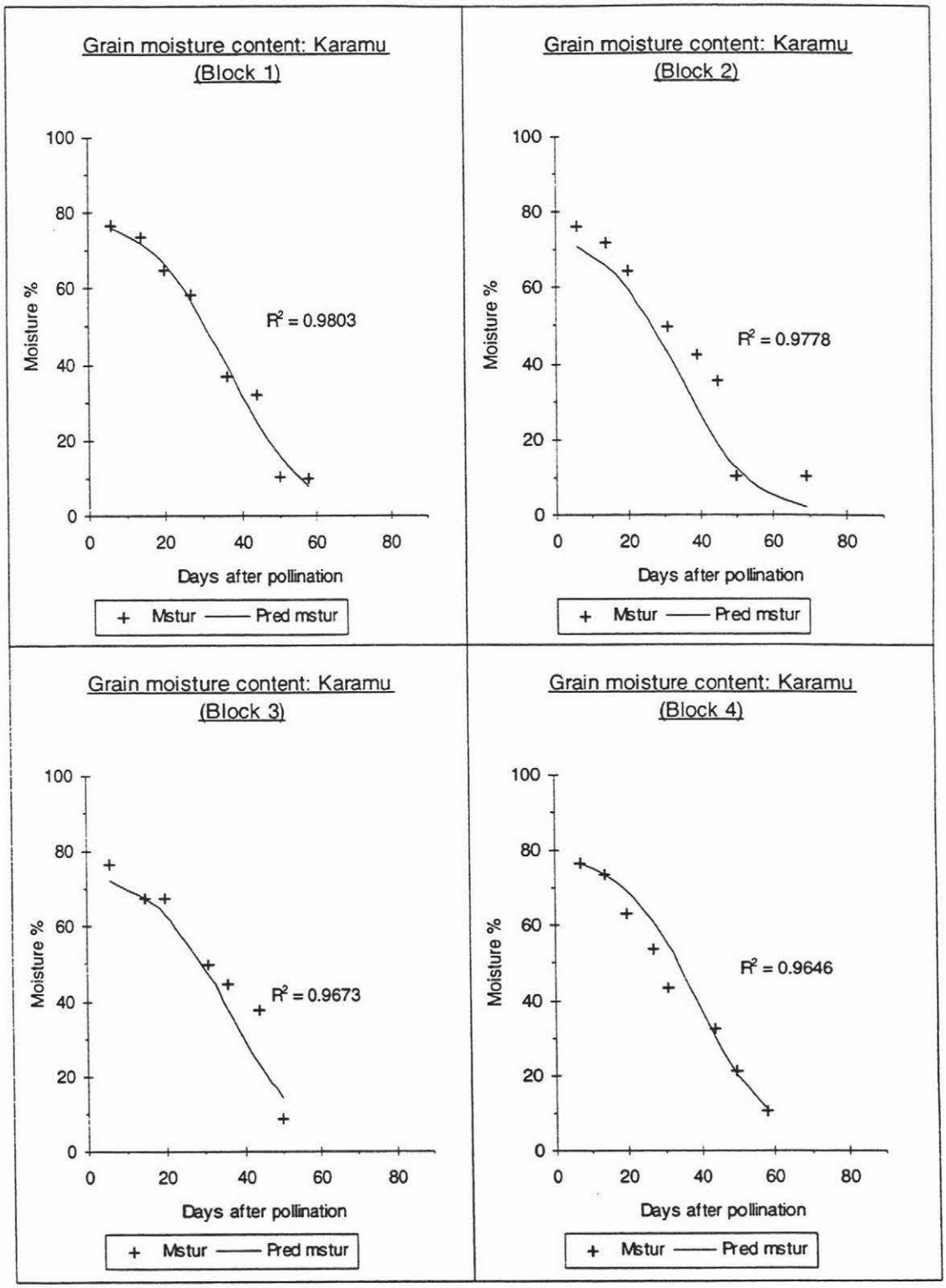

Grain moisture content: Karamu
(Block 1)
Moisture %
Days after pollination
R² = 0.9803
+ Mstur —— Pred mstur
Grain moisture content: Karamu
(Block 2)
Moisture %
Days after pollination
R² = 0.9778
+ Mstur —— Pred mstur
Grain moisture content: Karamu
(Block 3)
Moisture %
Days after pollination
R² = 0.9673
+ Mstur —— Pred mstur
Grain moisture content: Karamu
(Block 4)
Moisture %
Days after pollination
R² = 0.9646
+ Mstur —— Pred mstur

Table 4.2 (a). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Gamenya

Replicate:	1	2	3	4
Statistic				
A	35.02545	44.50746	37.32603	37.00000
s.e. A	4.71671	2.79972	3.83413	2.02614
B	2.96652	4.62994	2.34356	3.95000
s.e. B	1.72869	2.04906	3.60215	1.09854
K	0.15122	0.26005	0.08884	0.20100
s.e. K	0.21638	0.26210	0.07418	0.19085
V	0.92128	1.04544	0.75102	1.00000
s.e. V	2.59154	3.47841	2.0941	0.99564
F_{regr}	32.877 **	112.0398 ***	225.4536 ***	249.2388 ***
R^2	0.8803	0.9531	0.9867	0.9917

significance level

*** $P < 0.001$

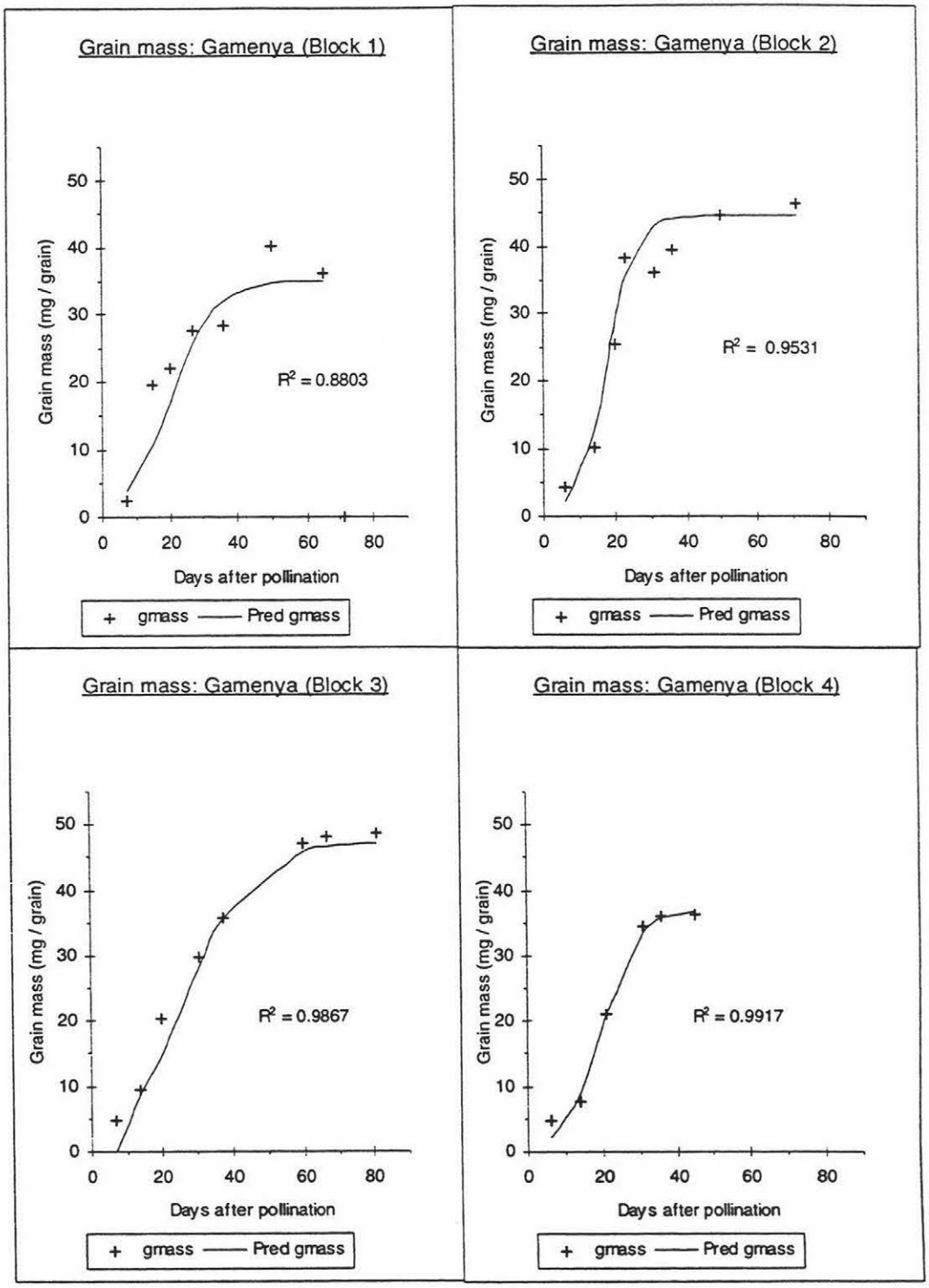

Grain mass: Gamenya (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.8803
+ gmass —— Pred gmass
Grain mass: Gamenya (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9531
+ gmass —— Pred gmass
Grain mass: Gamenya (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9867
+ gmass —— Pred gmass
Grain mass: Gamenya (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9917
+ gmass —— Pred gmass

Table 4.2 (b). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Tordo

Replicate:	1	2	3	4
Statistic				
A	46.02363	48.94165	50.99862	49.00434
s.e. A	4.67062	4.458373	2.27186	2.28133
B	3.05486	2.76489	3.54923	3.55135
s.e. B	2.86745	2.11375	1.56982	1.07473
K	0.15080	0.14705	0.20088	0.20096
s.e. K	0.20893	0.18149	0.22443	0.16869
V	1.14242	1.01189	1.10163	1.39763
s.e. V	1.75730	1.15834	1.18365	1.89531
F_{regr}	89.07914 ***	67.765 ***	126.2204 ***	366.581 ***
R^2	0.9396	0.9092	0.9693	0.9907

significance level
*** $P < 0.001$

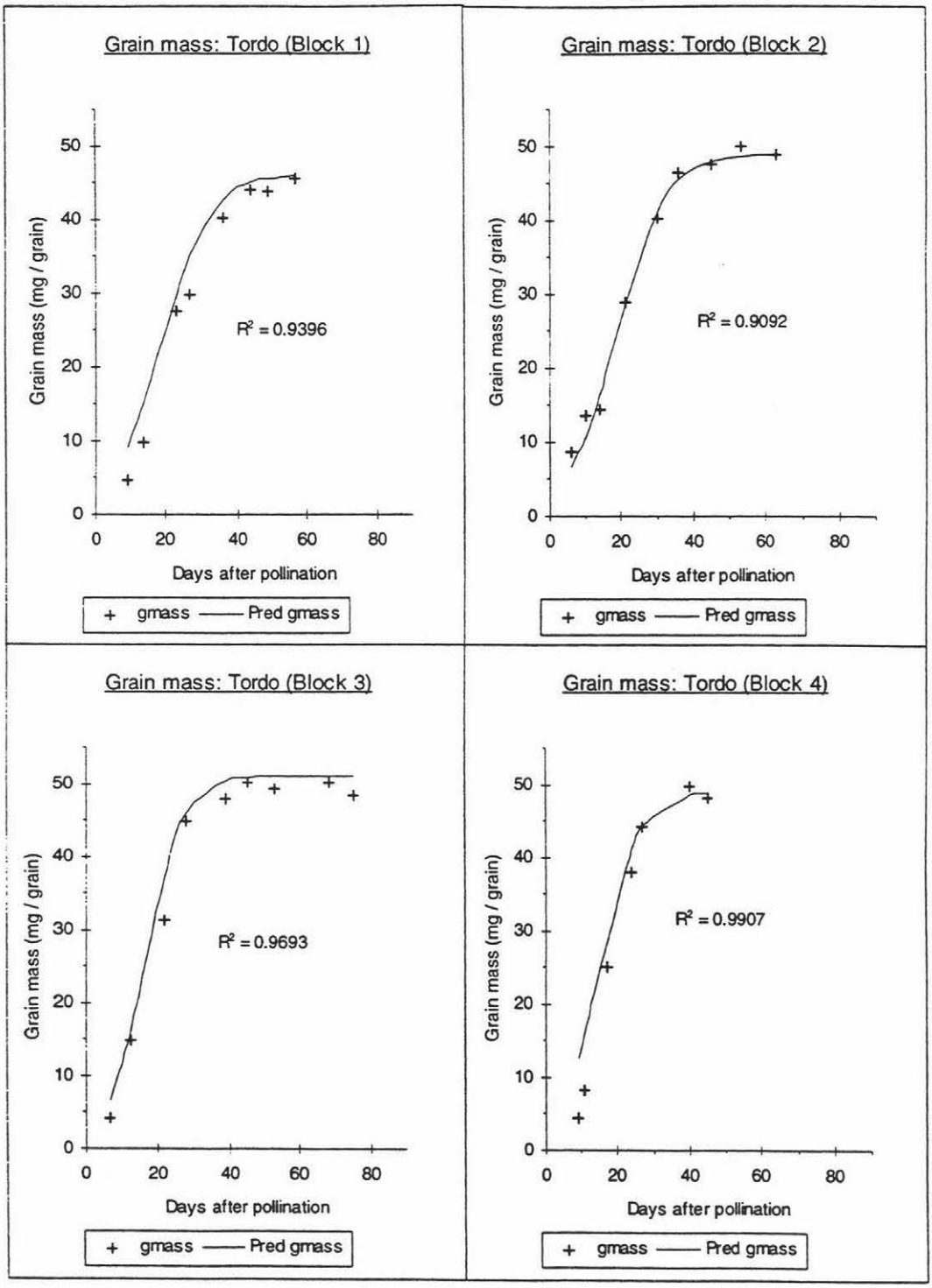

Grain mass: Tordo (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.9396
+ gmass —— Pred gmass
Grain mass: Tordo (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9092
+ gmass —— Pred gmass
Grain mass: Tordo (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9693
+ gmass —— Pred gmass
Grain mass: Tordo (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9907
+ gmass —— Pred gmass

Table 4.2 (c). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Kenya 321

Replicate:	1	2	3	4
Statistic				
A	52.06058	52.11186	50.34165	48.80439
s.e. A	2.07763	0.73478	2.14625	2.13066
B	4.10392	4.40666	2.91662	3.18739
s.e. B	2.33966	1.72050	1.44875	2.43569
K	0.19987	0.20105	0.10559	0.14718
s.e. K	0.15072	0.04089	0.0199	0.21365
V	1.15087	1.05103	0.98446	1.03854
s.e. V	4.42259	0.67333	0.37778	0.75102
F_{regr}	215.6257 ***	1964.3378 ***	788.4844 ***	76.3405 ***

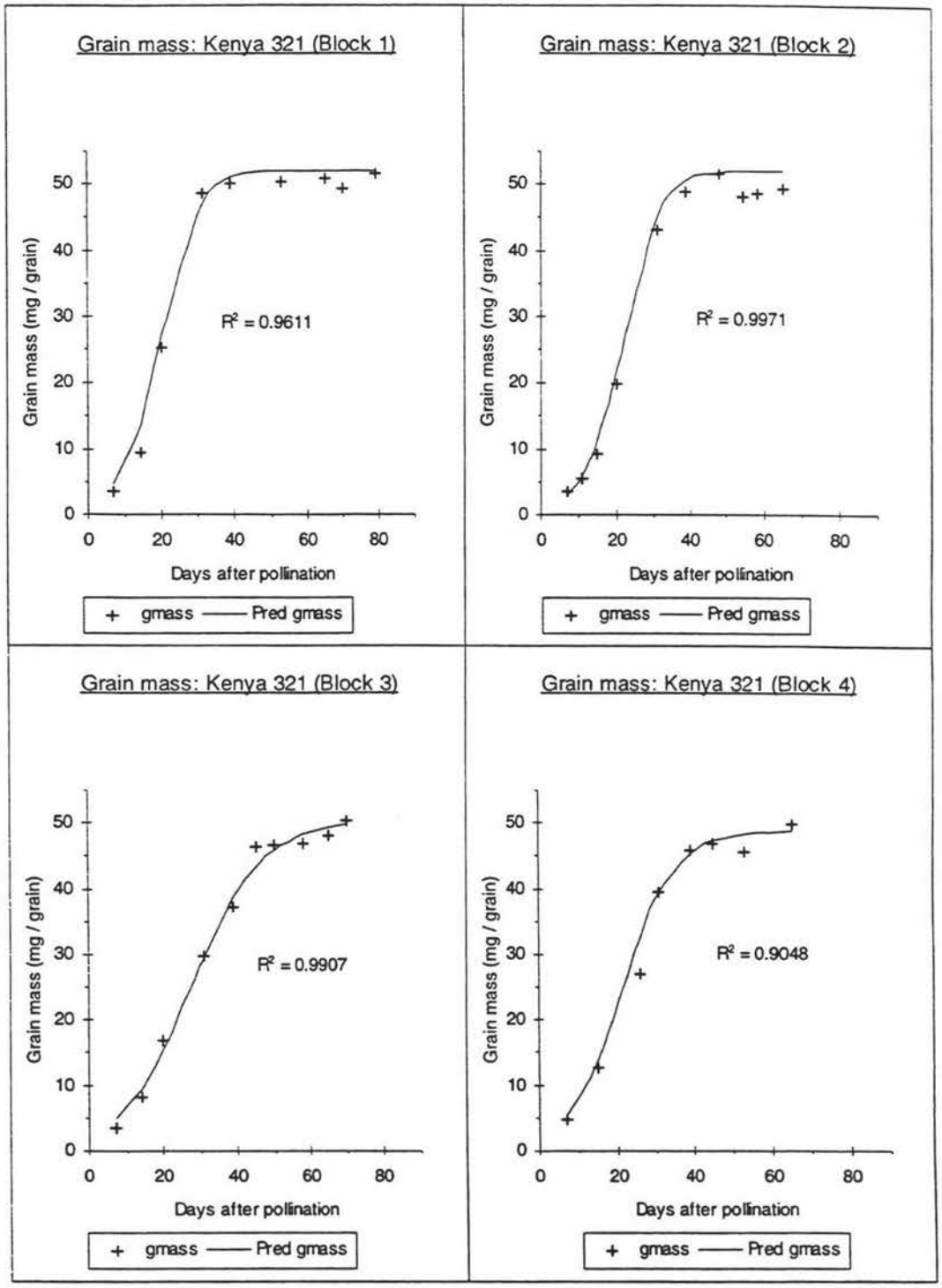

Grain mass: Kenya 321 (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.9611
+ gmass —— Pred gmass
Grain mass: Kenya 321 (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9971
+ gmass —— Pred gmass
Grain mass: Kenya 321 (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9907
+ gmass —— Pred gmass
Grain mass: Kenya 321 (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9048
+ gmass —— Pred gmass

Table 4.2 (d). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Brevor

Replicate:	1	2	3	4
Statistic				
A	49.00652	52.82290	58.99871	57.98299
s.e. A	3.79196	22.71821	3.95229	1.51050
B	3.44875	2.40622	3.94900	3.64403
s.e. B	4.45512	8.83945	14.32021	3.45300
K	0.15061	0.08182	0.20102	0.15854
s.e. K	0.10814	0.16091	0.33077	0.06807
V	0.95238	0.94309	1.15171	1.10556
s.e. V	1.28651	3.01835	9.39906	1.77982
F_{regr}	130.171 ***	66.3001 ***	82.4793 ***	739.3489 ***

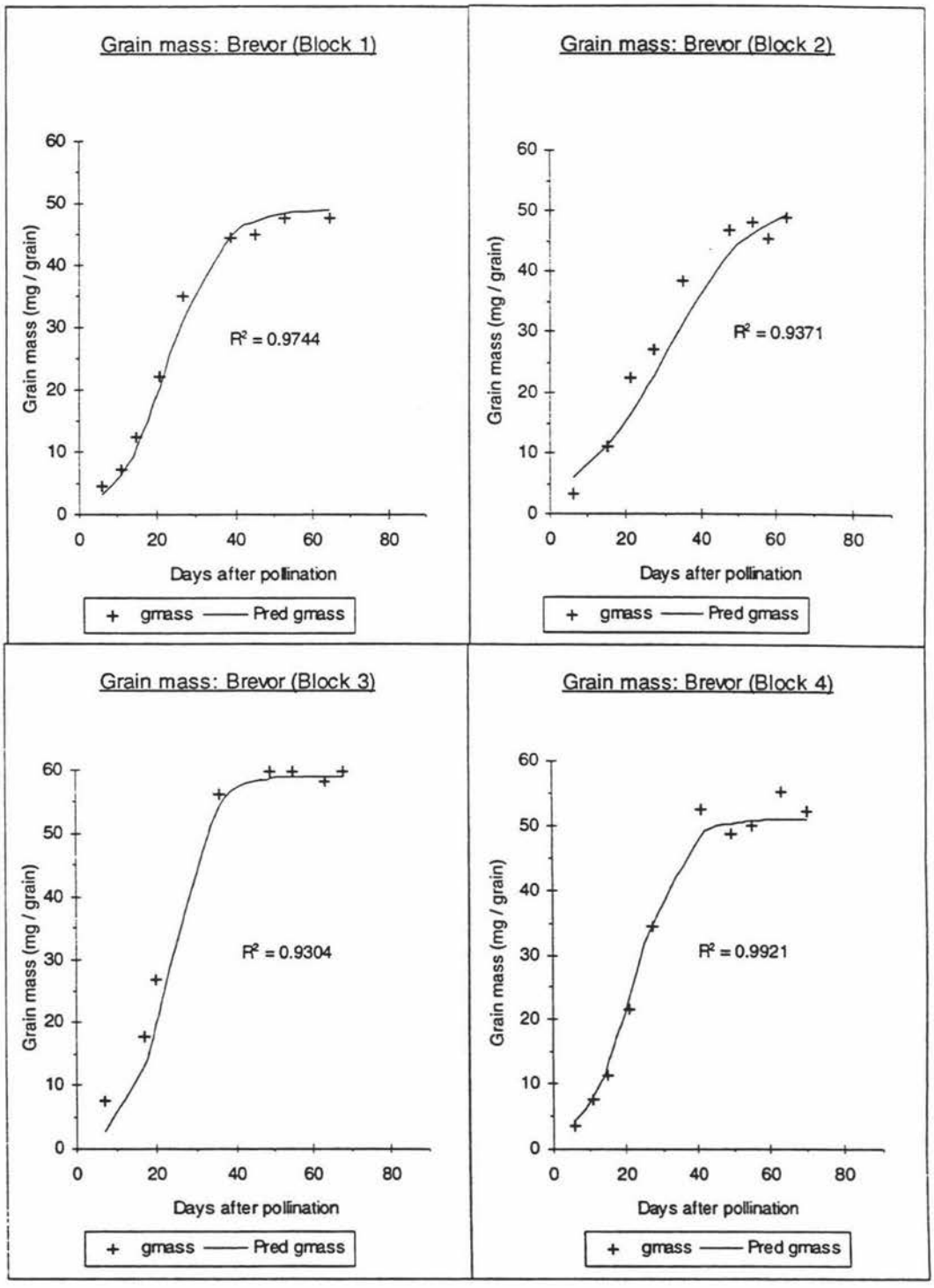

Grain mass: Brevor (Block 1)
Grain mass (mg / grain)
R² = 0.9744
Days after pollination
gmass
Pred gmass
Grain mass: Brevor (Block 2)
Grain mass (mg / grain)
R² = 0.9371
Days after pollination
gmass
Pred gmass
Grain mass: Brevor (Block 3)
Grain mass (mg / grain)
R² = 0.9304
Days after pollination
gmass
Pred gmass
Grain mass: Brevor (Block 4)
Grain mass (mg / grain)
R² = 0.9921
Days after pollination
gmass
Pred gmass

Table 4.2 (e). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Isis

Replicate: / Statistic	1	2	3	4
A	54.00206	49.91681	45.00000	45.00000
s.e. A	7.45961	8.38133	8.96065	8.93652
B	3.14989	2.37770	3.29787	3.80000
s.e. B	2.31658	4.74027	2.28643	2.560321
K	0.15074	0.08441	0.15068	0.20100
s.e. K	0.09641	0.07889	0.03954	0.08456
V	1.20314	0.95169	1.15624	1.15000
s.e. V	1.35624	1.70558	1.51436	1.20312
F_{regr}	478.2947 ***	301.9890 ***	326.6654 ***	426.4786 ***

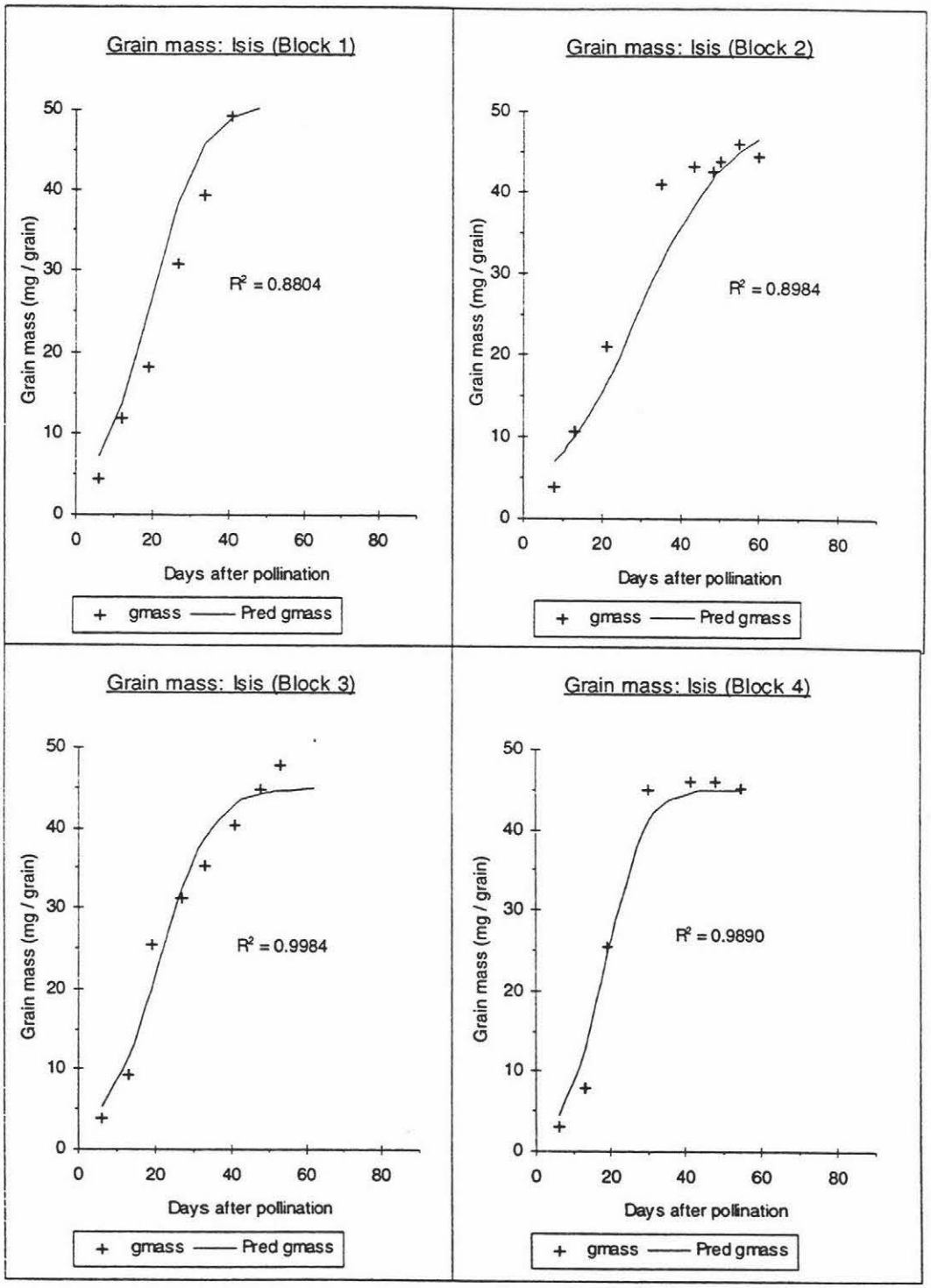

Grain mass: Isis (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.8804
+ gmass —— Pred gmass
Grain mass: Isis (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.8984
+ gmass —— Pred gmass
Grain mass: Isis (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9984
+ gmass —— Pred gmass
Grain mass: Isis (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9890
+ gmass —— Pred gmass

Table 4.2 (f). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Sonora 64A

Replicate:	1	2	3	4
Statistic				
A	43.00505	44.00142	45.96493	42.00779
s.e. A	5.60145	2.09154	2.91662	3.14718
B	2.99991	3.44976	3.29695	3.44954
s.e. B	2.63410	2.14758	2.97870	3.01564
kK	0.15072	0.15095	0.15075	0.15058
s.e. K	0.13564	0.10291	0.14967	0.14401
V	1.00307	1.05112	1.10514	1.00345
s.e. V	1.33326	2.25658	1.40236	1.31156
F_{regr}	584.37856 ***	172.37289 ***	1079.79587 ***	239.36486 ***

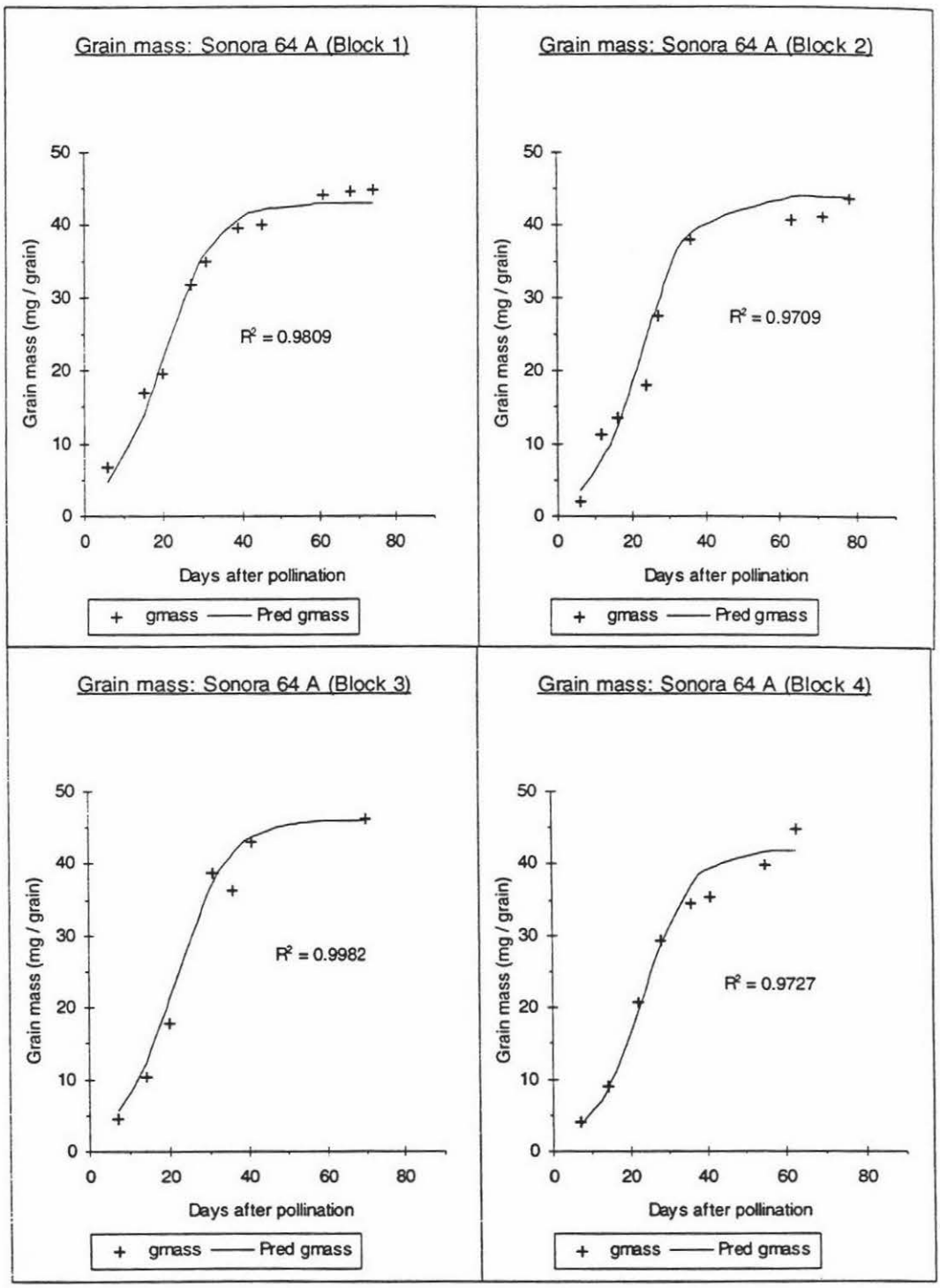

Grain mass: Sonora 64 A (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.9809
+ gmass —— Pred gmass
Grain mass: Sonora 64 A (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9709
+ gmass —— Pred gmass
Grain mass: Sonora 64 A (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9982
+ gmass —— Pred gmass
Grain mass: Sonora 64 A (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9727
+ gmass —— Pred gmass

Table 4.2 (g). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Thatcher

Replicate:	1	2	3	4
Statistic				
A	45.00000	35.23453	41.00374	37.53003
s.e. A	5.45145	3.61205	4.00771	4.95687
B	3.55000	3.88676	3.55060	5.99826
s.e. B	2.13654	2.23065	1.75614	1.95874
K	0.20100	0.22068	0.15080	0.24001
s.e. K	0.10245	0.09265	0.11425	0.09875
V	1.35000	0.85064	1.00211	0.90221
s.e. V	0.98267	0.734568	0.85996	0.69514
F_{regr}	81.49002 ***	124.5623 ***	204.5631 ***	159.2634 ***

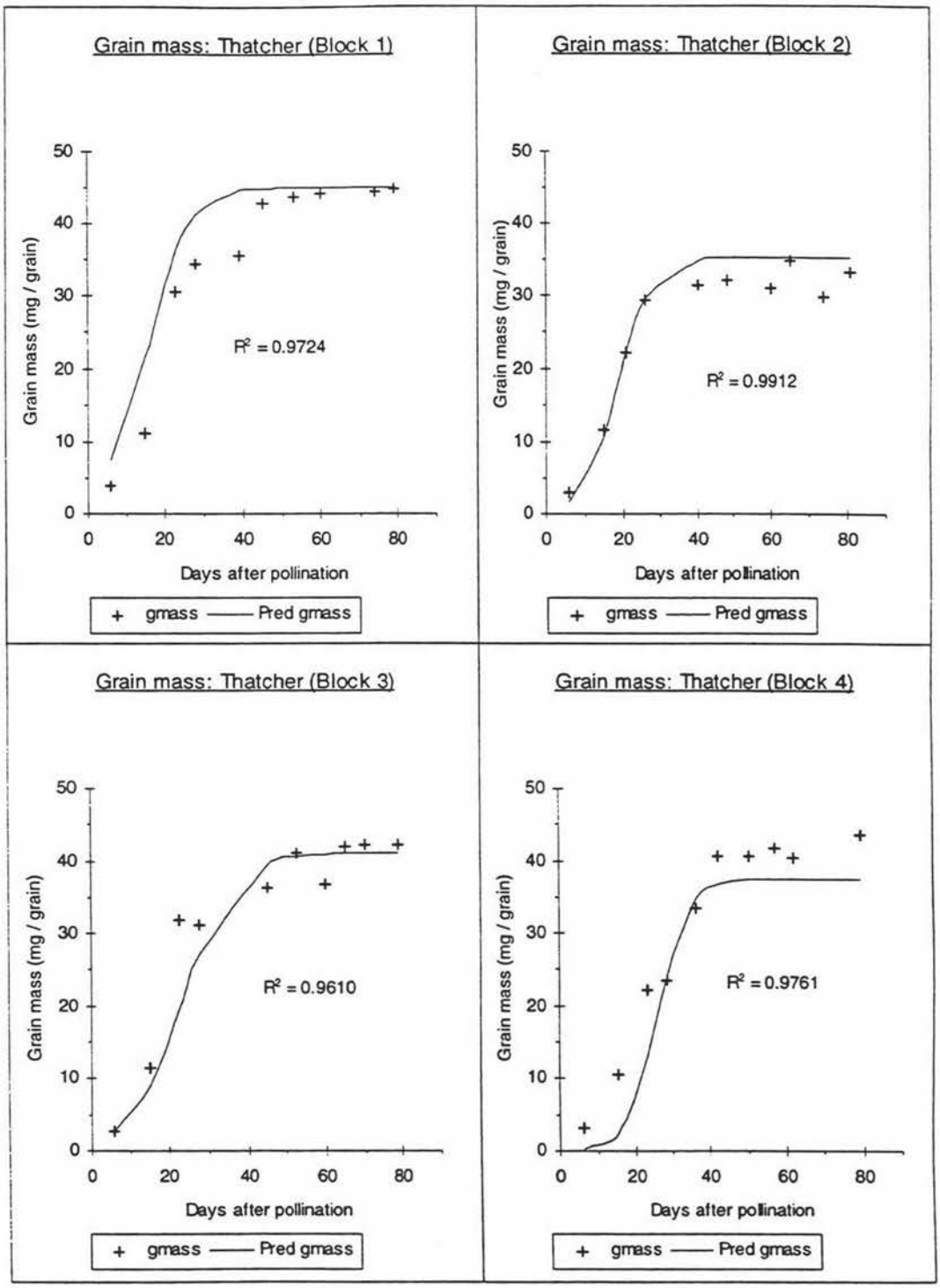

Grain mass: Thatcher (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.9724
+ gmass —— Pred gmass
Grain mass: Thatcher (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9912
+ gmass —— Pred gmass
Grain mass: Thatcher (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9610
+ gmass —— Pred gmass
Grain mass: Thatcher (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9761
+ gmass —— Pred gmass

Table 4.2 (h). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. La Prevision

Replicate:	1	2	3	4
Statistic				
A	46.00000	42.00105	46.99265	45.96740
s.e. A	3.99825	3.40666	2.07852	3.10540
B	3.20000	3.50000	3.15098	4.40174
s.e. B	1.25698	2.51461	3.28789	2.99541
K	0.15100	0.15095	0.15092	0.20039
s.e. K	0.098506	0.09839	0.07241	0.08221
V	1.00000	0.95086	1.04888	1.00319
s.e. V	1.00319	0.009830	1.50920	1.01368
F_{regr}	1396.5961 ***	173.0879 ***	433.0992 ***	459.3722 ***

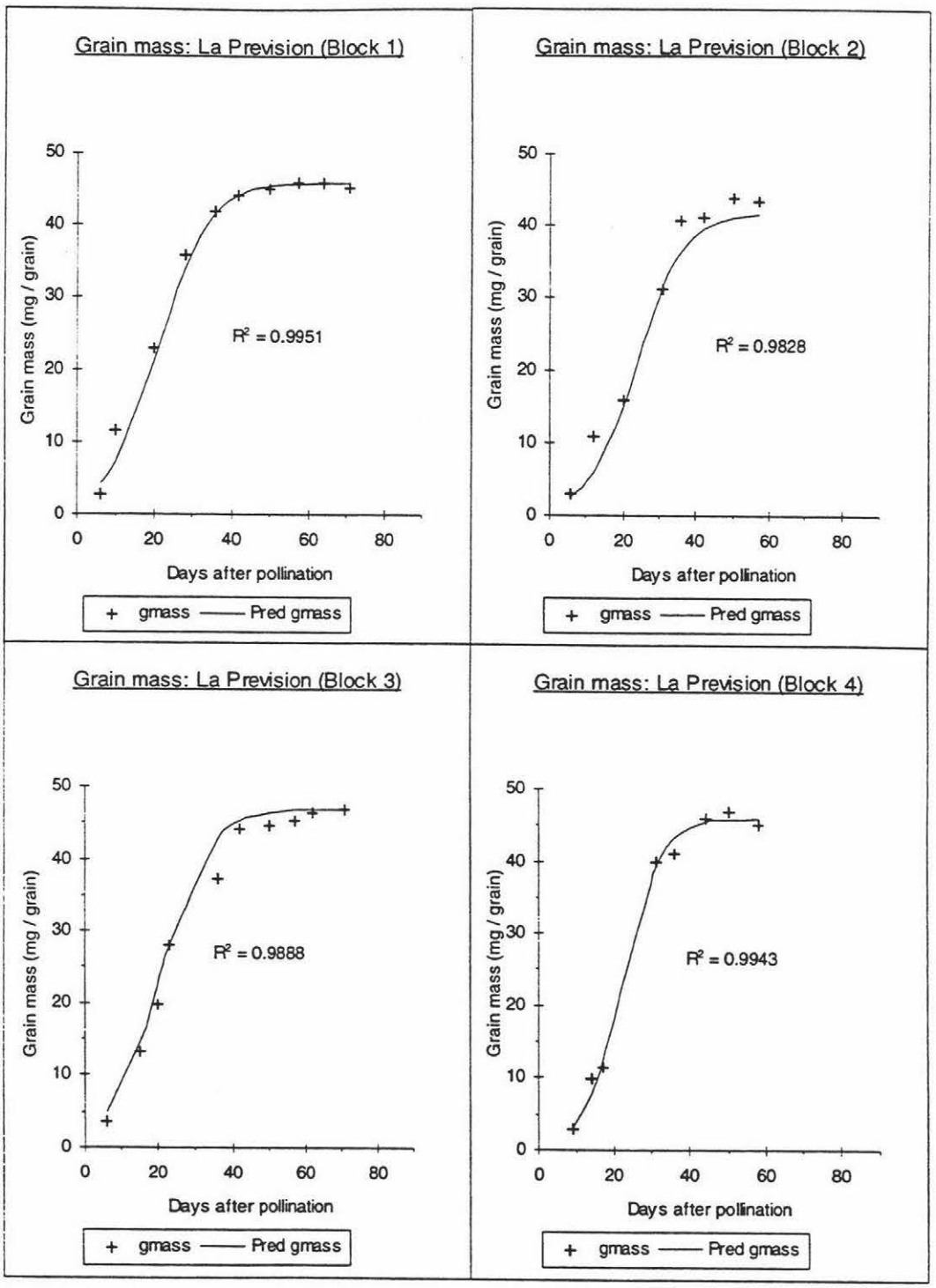

Grain mass: La Prevision (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.9951
+ gmass —— Pred gmass
Grain mass: La Prevision (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9828
+ gmass —— Pred gmass
Grain mass: La Prevision (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9888
+ gmass —— Pred gmass
Grain mass: La Prevision (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9943
+ gmass —— Pred gmass

Table 4.2 (i). Estimated statistics of the Richards functions fitted on data for grain mass.

Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
A	42.00000	49.009826	55.0000	38.99280
s.e. A	5.33214	3.83921	4.86008	3.81527
B	3.40000	3.35159	3.35000	3.29792
s.e. B	2.13526	2.69671	2.50561	1.52940
K	0.20100	0.15085	0.15100	0.15103
s.e. K	0.063124	0.05730	0.03719	0.03432
V	1.05000	1.00070	1.20000	1.00219
s.e. V	1.59981	1.03257	1.93418	0.57817
F_{regr}	93.6800 ***	518.735 ***	2215.337 ***	1140.9276 ***

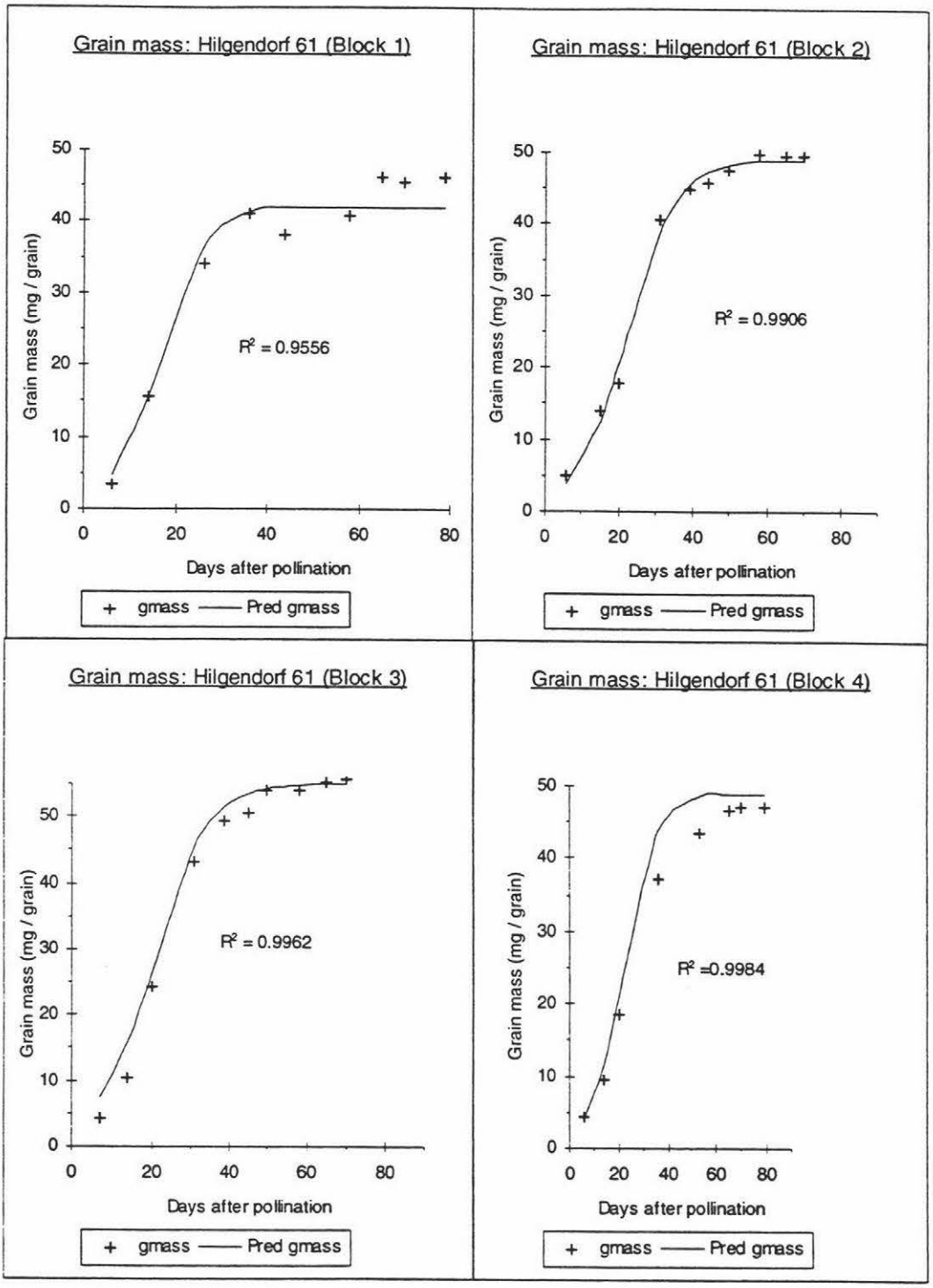
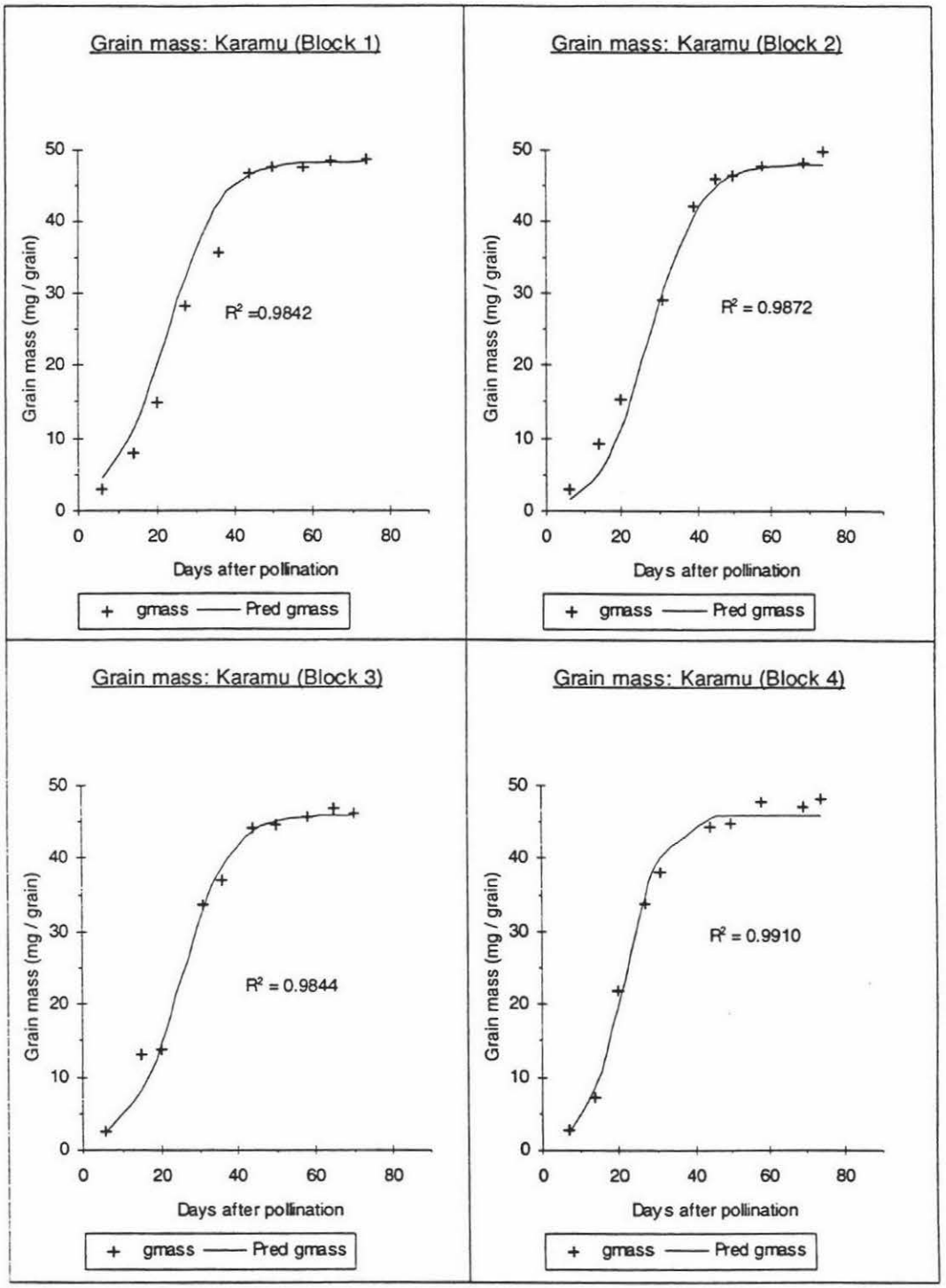

Grain mass: Hilgendorf 61 (Block 1)
Grain mass (mg / grain)
Days after pollination
R² = 0.9556
+ gmass —— Pred gmass
Grain mass: Hilgendorf 61 (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9906
+ gmass —— Pred gmass
Grain mass: Hilgendorf 61 (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9962
+ gmass —— Pred gmass
Grain mass: Hilgendorf 61 (Block 4)
Grain mass (mg / grain)
Days after pollination
R² =0.9984
+ gmass —— Pred gmass

Table 4.2 (j). Estimated statistics of the Richards functions fitted on data for grain mass.

cv. Karamu

Replicate:	1	2	3	4
Statistic				
A	48.36774	48.00000	46.00000	46.03060
s.e. A	2.24572	2.14256	4.09337	3.33145
B	3.51132	4.10000	3.70000	4.30008
s.e. B	5.41498	3.21653	4.54768	2.15698
K	0.14860	0.15100	0.15100	0.20008
s.e. K	0.09251	0.09826	0.10219	0.13102
V	1.13631	0.95000	0.95000	1.00324
s.e. V	1.25581	0.78015	1.29548	0.99215
F_{regr}	258.9513 ***	549.2584 ***	180.9962 ***	665.834 ***
R^2	0.9842	0.9872	0.9844	0.9910

significance level
*** $P < 0.001$


Grain mass: Karamu (Block 1)
Grain mass (mg / grain)
Days after pollination
R² =0.9842
+ gmass —— Pred gmass
Grain mass: Karamu (Block 2)
Grain mass (mg / grain)
Days after pollination
R² = 0.9872
+ gmass —— Pred gmass
Grain mass: Karamu (Block 3)
Grain mass (mg / grain)
Days after pollination
R² = 0.9844
+ gmass —— Pred gmass
Grain mass: Karamu (Block 4)
Grain mass (mg / grain)
Days after pollination
R² = 0.9910
+ gmass —— Pred gmass

Appendix 3.

Table A 5.1 (a). Estimated statistics of the Richards functions fitted on data for special germination .

cv. Gamenya

Replicate:	1	2	3	4
Statistic				
A	98.00000	98.00000	99.01913	100.00000
s.e. A	2.42271	3.30302	2.56235	12.83829
B	6.81995	7.85000	6.99945	10.75110
s.e. B	2.57845	3.93568	4.50123	14.32616
K	0.23365	0.30100	0.251073	0.35097
s.e. K	0.06031	0.10018	0.09630	0.37598
V	0.97328	1.00000	0.99725	0.95078
s.e. V	0.55300	0.91897	0.80156	2.04853
F_{regr}	658.7893 ***	493.1770 ***	541.2078 ***	42.3879 *
R^2	0.9975	0.9967	0.9981	0.9760

significance level
* P < 0.05
*** P < 0.001

Figure A 5.1 (a). The Richards function fits for special germination of the four replicates of cv. Gamenya.

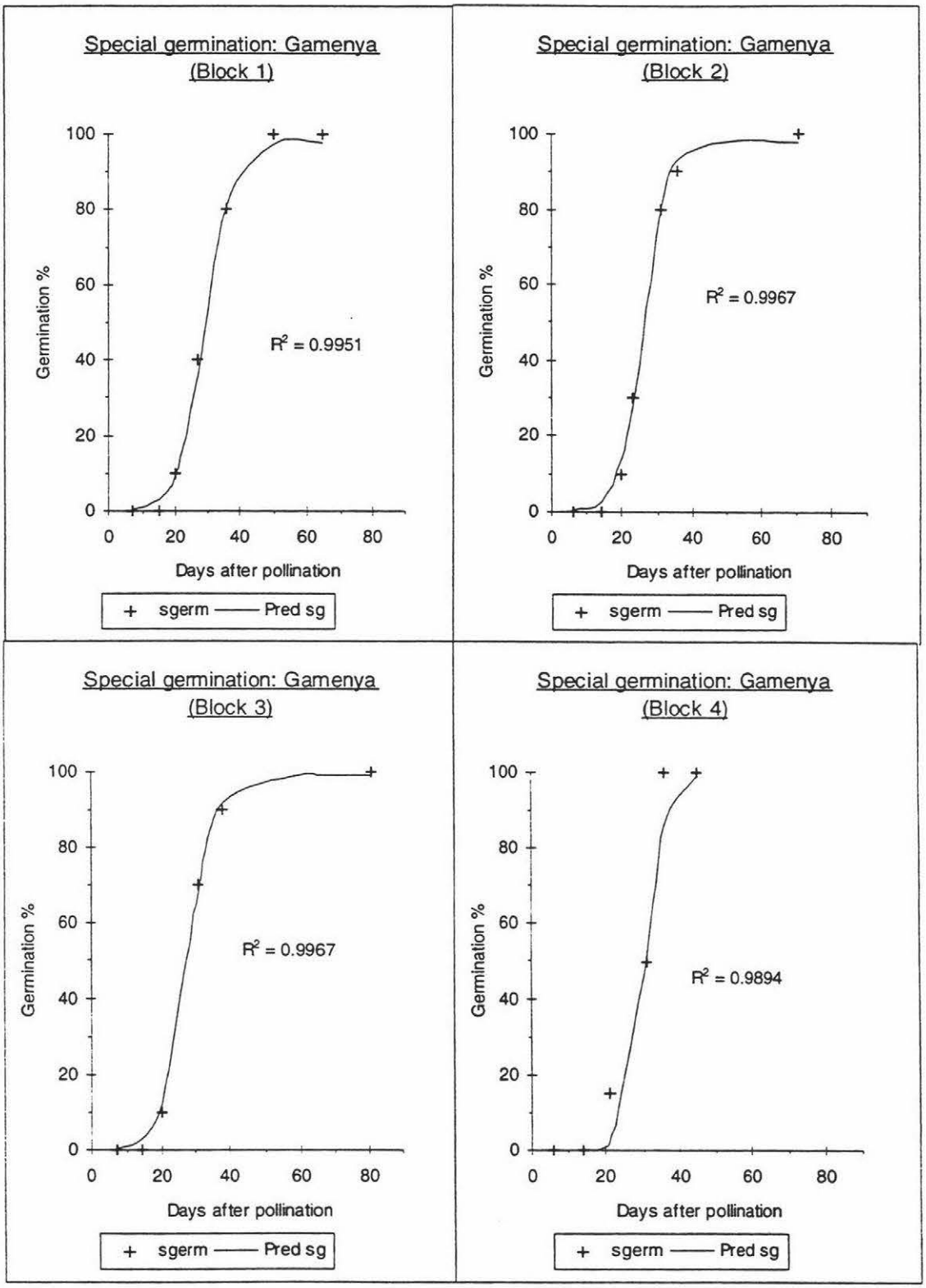


Table A 5.1 (b). Estimated statistics of the Richards functions fitted on data for special germination.

cv.Tordo

Replicate:	1	2	3	4
Statistic				
A	100.00000	100.00000	99.99996	99.50000
s.e. A	7.93079	3.23658	7.19100	7.34317
B	11.20008	12.02880	10.55083	6.95000
s.e. B	13.58905	8.93233	13.27083	2.93237
K	0.30107	0.30040	0.25101	0.20100
s.e. K	0.25151	0.16304	0.23348	0.66971
V	1.14626	1.02204	0.99730	0.90000
s.e. V	4.59849	1.71392	2.54876	3.73768
F_{regr}	109.3575 **	367.1908 ***	82.77364 ***	16.2032 NS
R^2	0.9883	0.9946	0.9762	0.9690

significance level
NS not significant
*** $P < 0.001$

Figure A 5.1 (b). The Richards function fits for special germination of the four replicates of cv. Tordo.

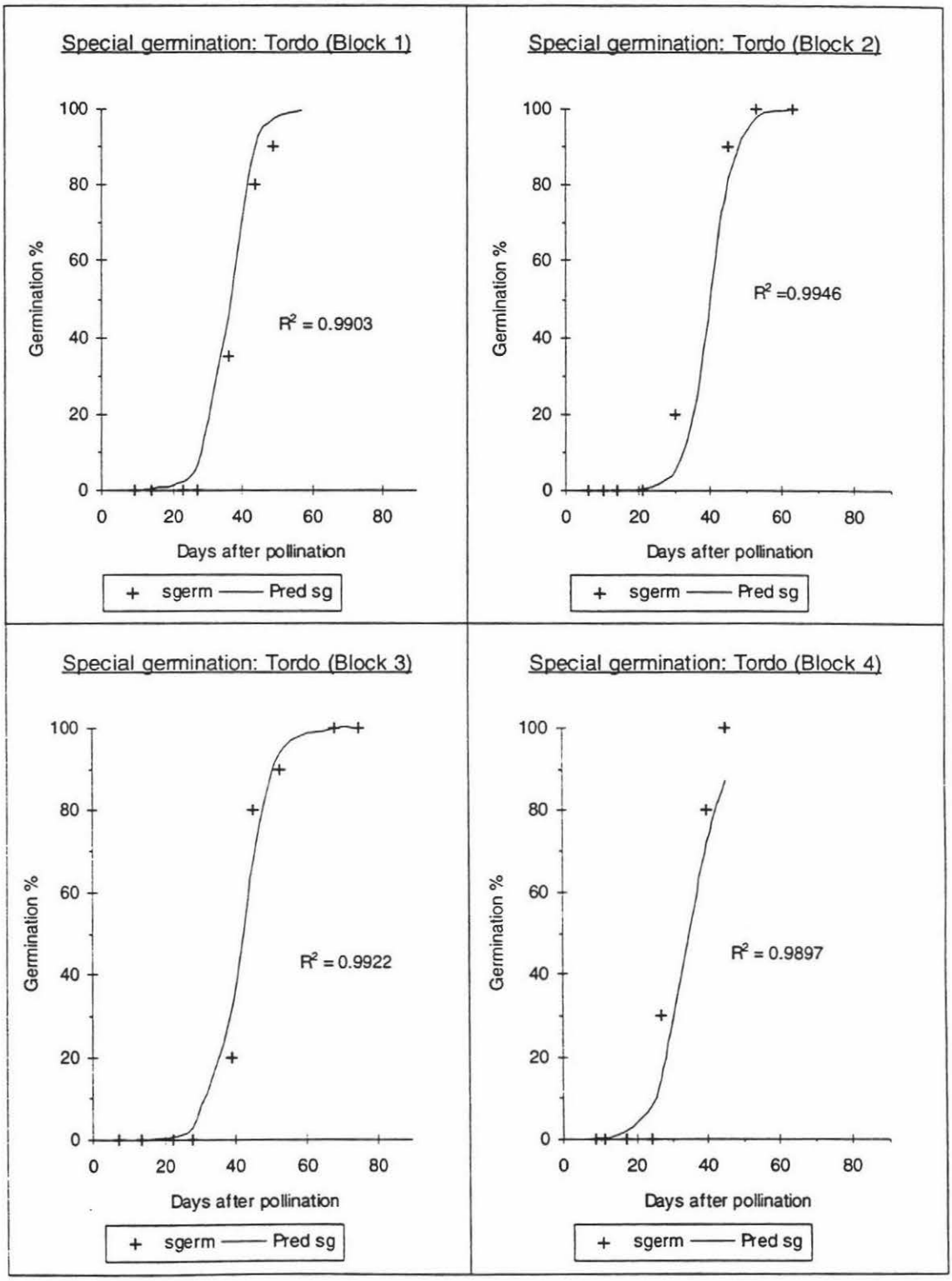


Table A 5.1 (c). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Kenya 321

Replicate:	1	2	3	4
Statistic				
A	99.01144	100.00000	100.00000	99.98238
s.e. A	1.28590	8.02660	3.59307	5.15692
B	10.49979	6.15000	7.79586	4.81175
s.e. B	4.66924	11.03098	4.76155	3.16838
K	0.35100	0.20100	0.25114	0.15084
s.e. K	0.12319	0.22435	0.10649	0.06000
V	0.99871	1.05000	1.02914	0.98402
s.e. V	0.75187	3.40555	1.19909	0.90805
F_{regr}	1876.1038 ***	77.5362 ***	355.2079 ***	341.2921 ***
R^2	0.9972	0.9538	0.9931	0.9913

significance level
*** $P < 0.001$

Figure A 5.1 (c). The Richards function fits for special germination of the four replicates of cv. Kenya 321.

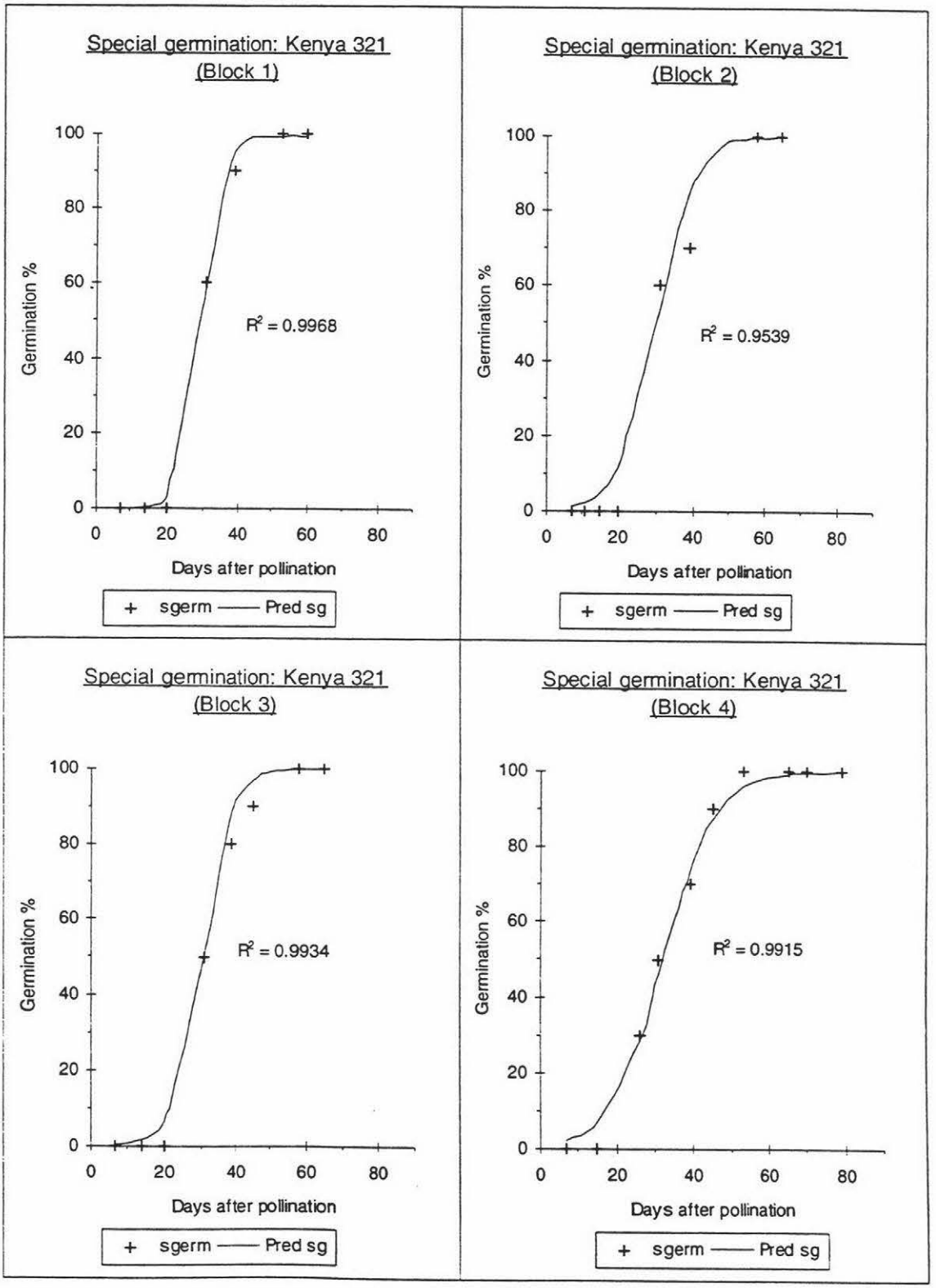


Table A 5.1 (d). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Brevor

Replicate:	1	2	3	4
Statistic				
A	100.00000	99.00000	99.00000	99.98186
s.e. A	6.31423	5.60000	9.51902	6.62775
B	5.50000	5.60000	4.22224	4.80680
s.e. B	2.29105	6.58398	8.80984	4.5513
K	0.15100	0.20100	0.10187	0.15077
s.e. K	0.47493	0.15634	0.21735	0.08524
V	0.90000	0.95000	0.75001	0.99416
s.e. V	3.46716	1.42456	0.70053	1.30523
F_{regr}	13.0655 NS	91.8347 ***	43.4873 *	210.0685 ***
R^2	0.9119	0.9795	0.9748	0.9873

significance level
NS not significant
* $P < 0.05$
*** $P < 0.001$

Figure A 5.1 (d). The Richards function fits for special germination of the four replicates of cv. Brevor.

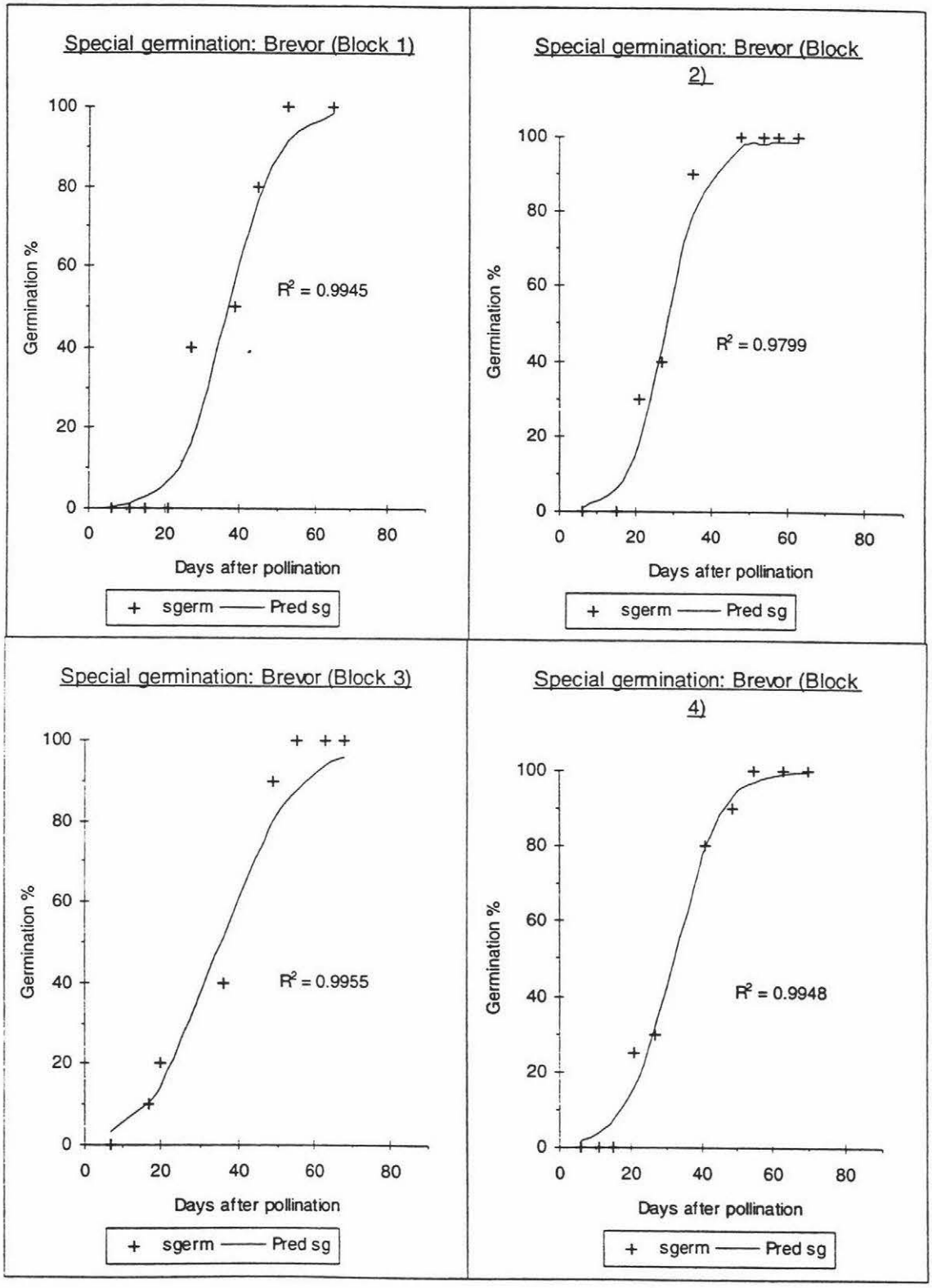


Table A 5.1 (e). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Isis

Replicate:	1	2	3	4
Statistic				
A	100.00000	100.00000	99.00831	99.99042
s.e. A	12.73105	9.98285	9.70521	10.00236
B	8.40000	7.75000	4.80249	4.00119
s.e. B	9.72854	8.96242	5.12689	3.21650
K	0.25100	0.20100	0.15079	0.10094
s.e. K	0.22589	0.16305	0.101123	0.094851
V	0.95000	0.95000	0.95209	0.80124
s.e. V	1.48224	1.76084	1.36021	1.23110
F_{regr}	96.2501 ***	130.0247 ***	158.0624 ***	22.0187 *
R^2	0.9849	0.9728	0.9837	0.9206

significance level
* P < 0.05
*** P < 0.001

Figure A 5.1 (e). The Richards function fits for special germination of the four replicates of cv. Isis.

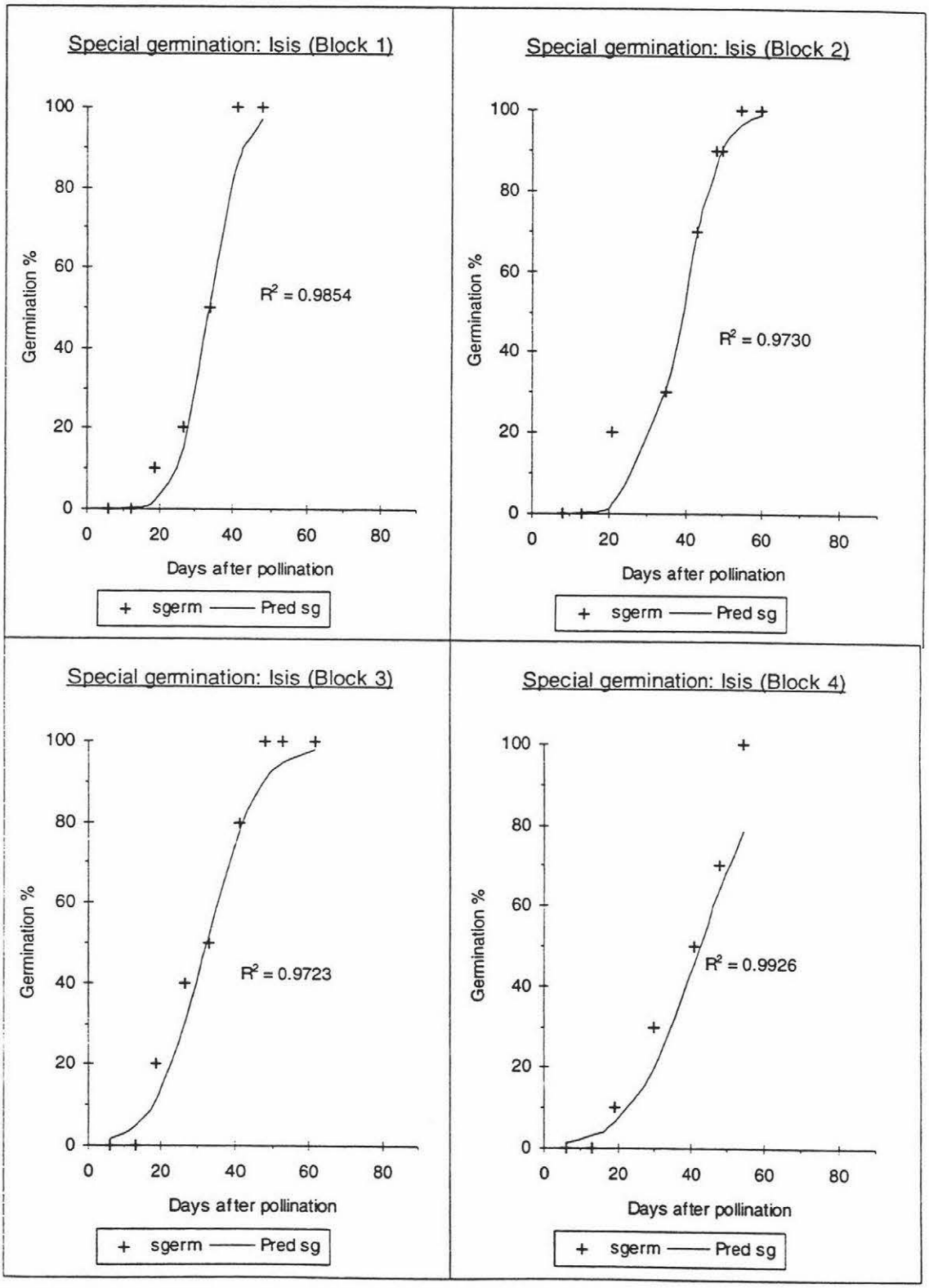


Table A 5.1 (f). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Sonora 64A

Replicate:	1	2	3	4
Statistic				
A	99.00000	99.00000	100.00000	99.50111
s.e. A	3.42389	3.12726	14.31064	2.70546
B	9.95000	11.35000	5.10000	12.01100
s.e. B	6.65581	7.20782	6.28974	4.65733
K	0.35100	0.40100	0.15100	0.40064
s.e. K	0.17585	0.20161	0.13605	0.12062
V	1.00000	1.00000	0.95000	0.99202
s.e. V	1.24374	1.20208	1.24696	0.80672
F_{regr}	329.6030 ***	378.4946 ***	155.1566 ***	653.0228 ***
R^2	0.9933	0.9956	0.9888	0.9981

significance level
*** P < 0.001

Figure A 5.1 (f). The Richards function fits for special germination of the four replicates of cv. Sonora 64 A.

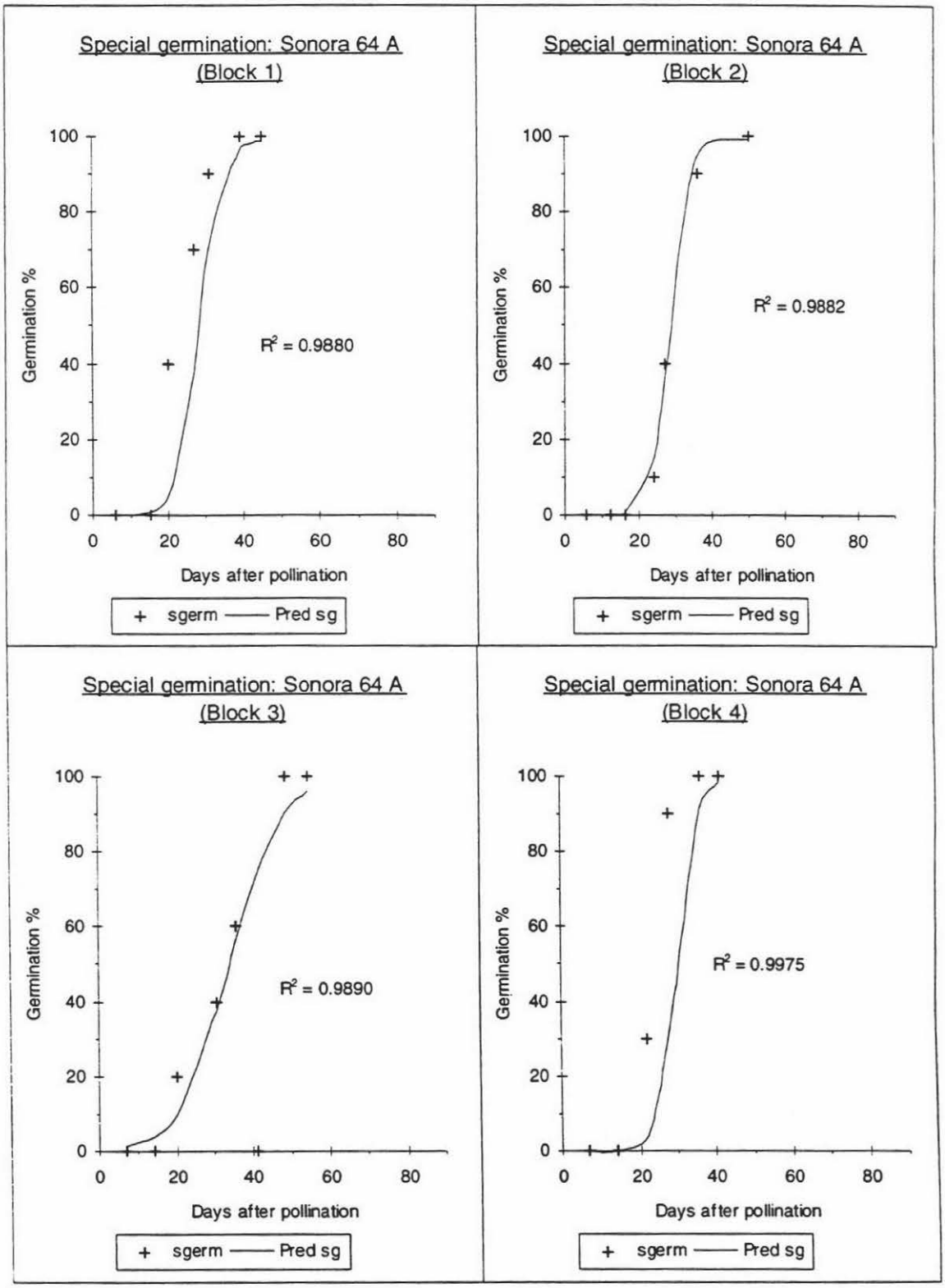


Table A 5.1 (g). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Thatcher

Replicate:	1	2	3	4
Statistic				
A	99.00000	99.99638	100.00000	99.00000
s.e. A	5.95201	4.39710	6.29112	12.03018
B	4.14452	6.65613	6.10000	3.64276
s.e. B	2.89981	4.72245	6.62987	5.24536
K	0.10109	0.15140	0.15100	0.09898
s.e. K	0.04364	0.07156	0.10747	0.07770
V	0.95020	0.99499	1.0000	0.99331
s.e. V	0.73041	1.19052	1.55291	1.77599
F_{regr}	381.9713 ***	304.3329 ***	265.8752 ***	100.0415 ***
R^2	0.9888	0.9920	0.9915	0.9640

significance level
*** $P < 0.001$

Figure A 5.1 (g). The Richards function fits for special germination of the four replicates of cv. Thatcher.

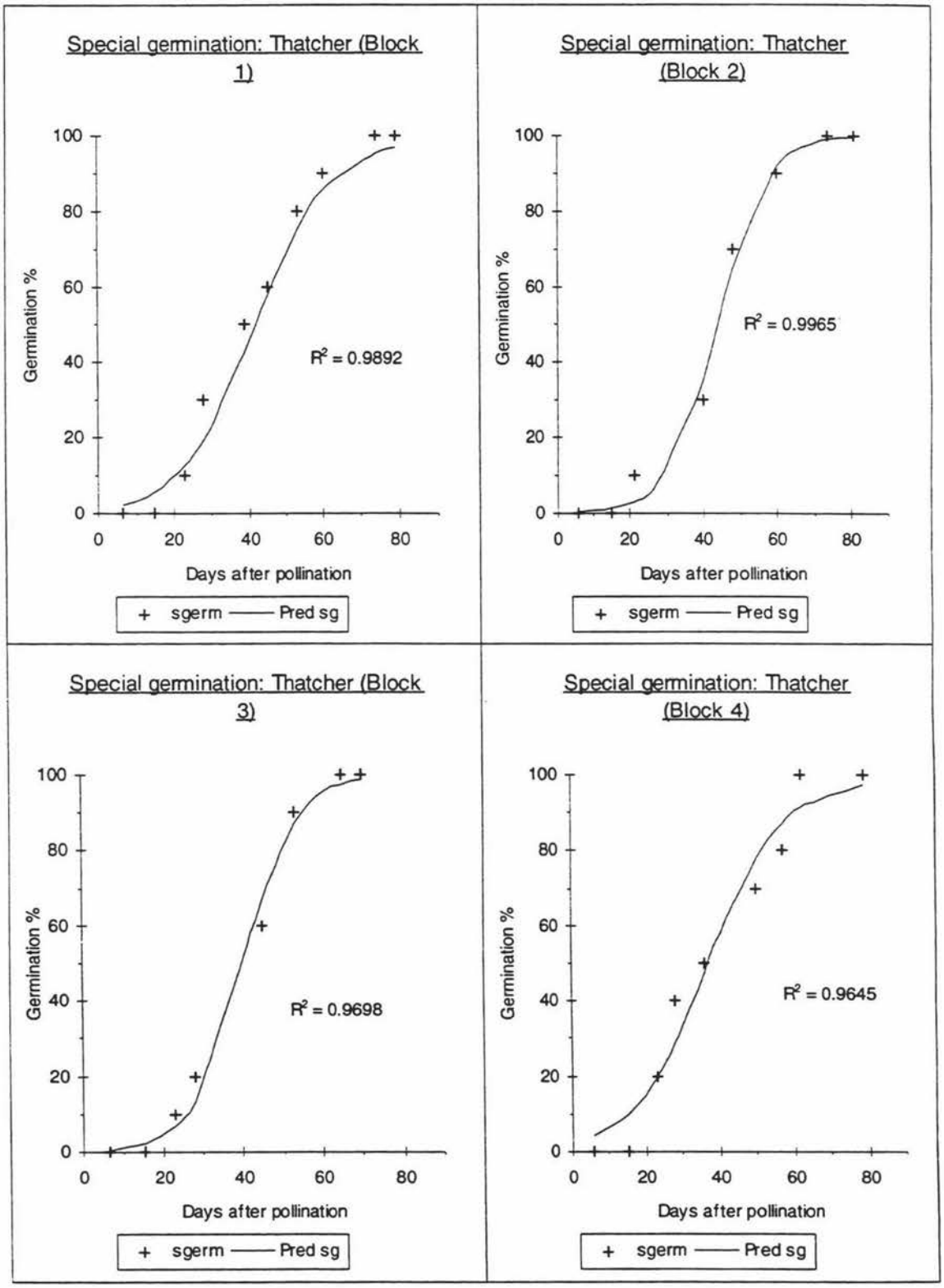


Table A 5.1 (h). Estimated statistics of the Richards functions fitted on data for special germination.

cv. La Prevision

Replicate:	1	2	3	4
Statistic				
A	99.55128	100.00000	99.00000	99.00000
s.e. A	19.36535	33.79426	10.25647	25.25567
B	8.92085	8.90000	8.39735	8.31536
s.e. B	12.14003	26.61380	6.18456	3.86084
K	0.15153	0.15100	0.16005	0.19571
s.e. K	0.06074	0.26865	0.02845	0.14303
V	1.46514	1.25000	1.54154	1.93167
s.e. V	17.24796	14.64461	11.81900	32.12627
F_{regr}	46.1541 *	114.2276 ***	64.2561 *	19.8939 *
R^2	0.9747	0.9862	0.9837	0.9439

significance level
* P < 0.05
*** P < 0.001

Figure A 5.1 (h). The Richards function fits for special germination of the four replicates of cv. La Prevision.

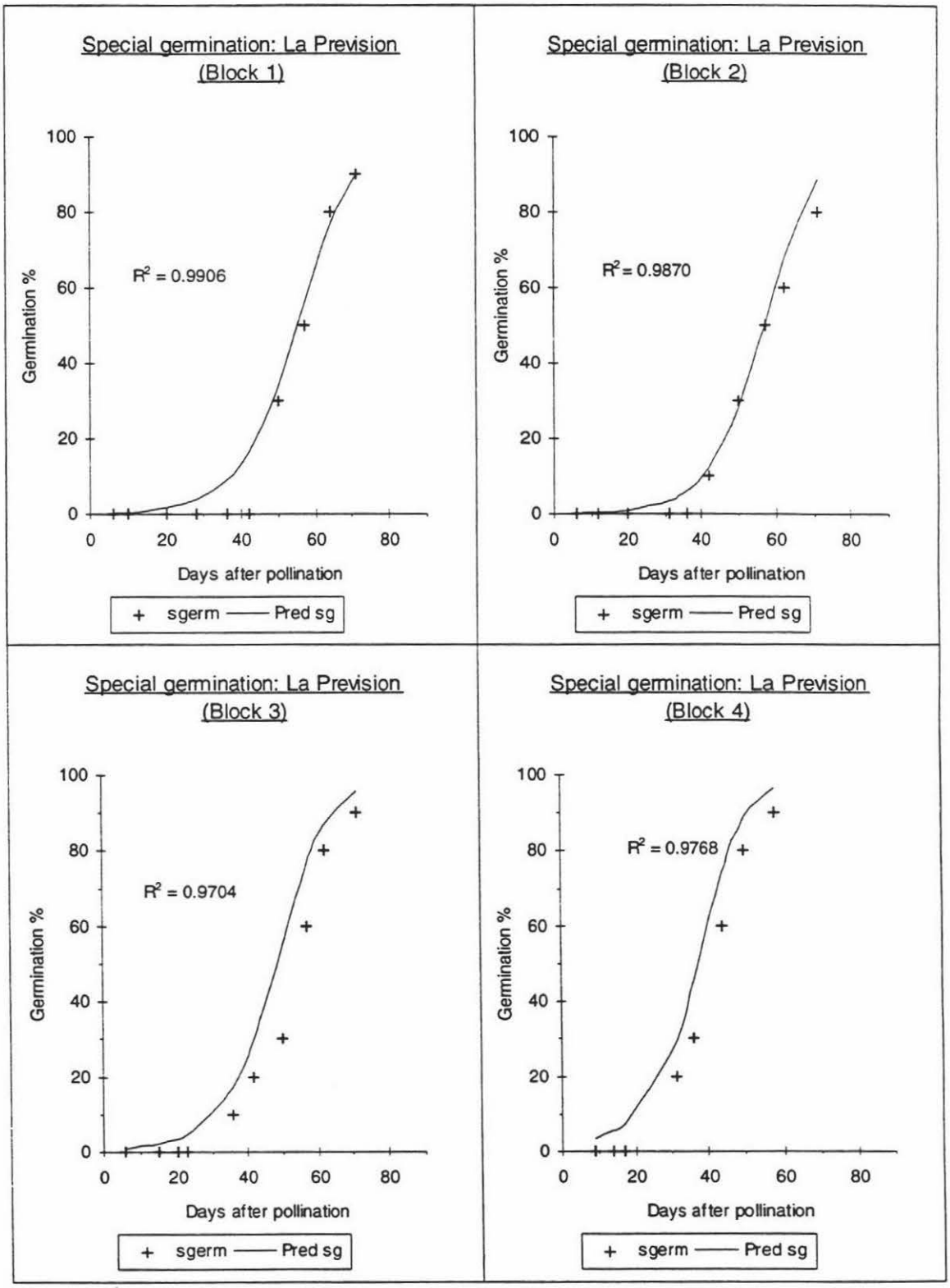


Table A 5.1 (i). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
A	99.00000	99.00000	99.00000	100.00000
s.e. A	3.84604	4.40473	6.80693	6.03170
B	10.24681	5.02119	5.05000	11.45029
s.e. B	12.96855	3.55897	5.88352	4.25219
K	0.25101	0.14370	0.15100	0.35107
s.e. K	0.23113	1.09249	0.10460	9.59094
V	1.01622	1.01579	1.00000	1.07818
s.e. V	2.56663	1.09249	1.72264	83.38299
F_{regr}	266.2741 ***	415.9758 ***	149.7373 ***	106.3936 ***
R^2	0.9913	0.9888	0.9676	0.9765

significance level
*** $P < 0.001$

Figure A 5.1 (i). The Richards function fits for special germination of the four replicates of cv. Hilgendorf 61.

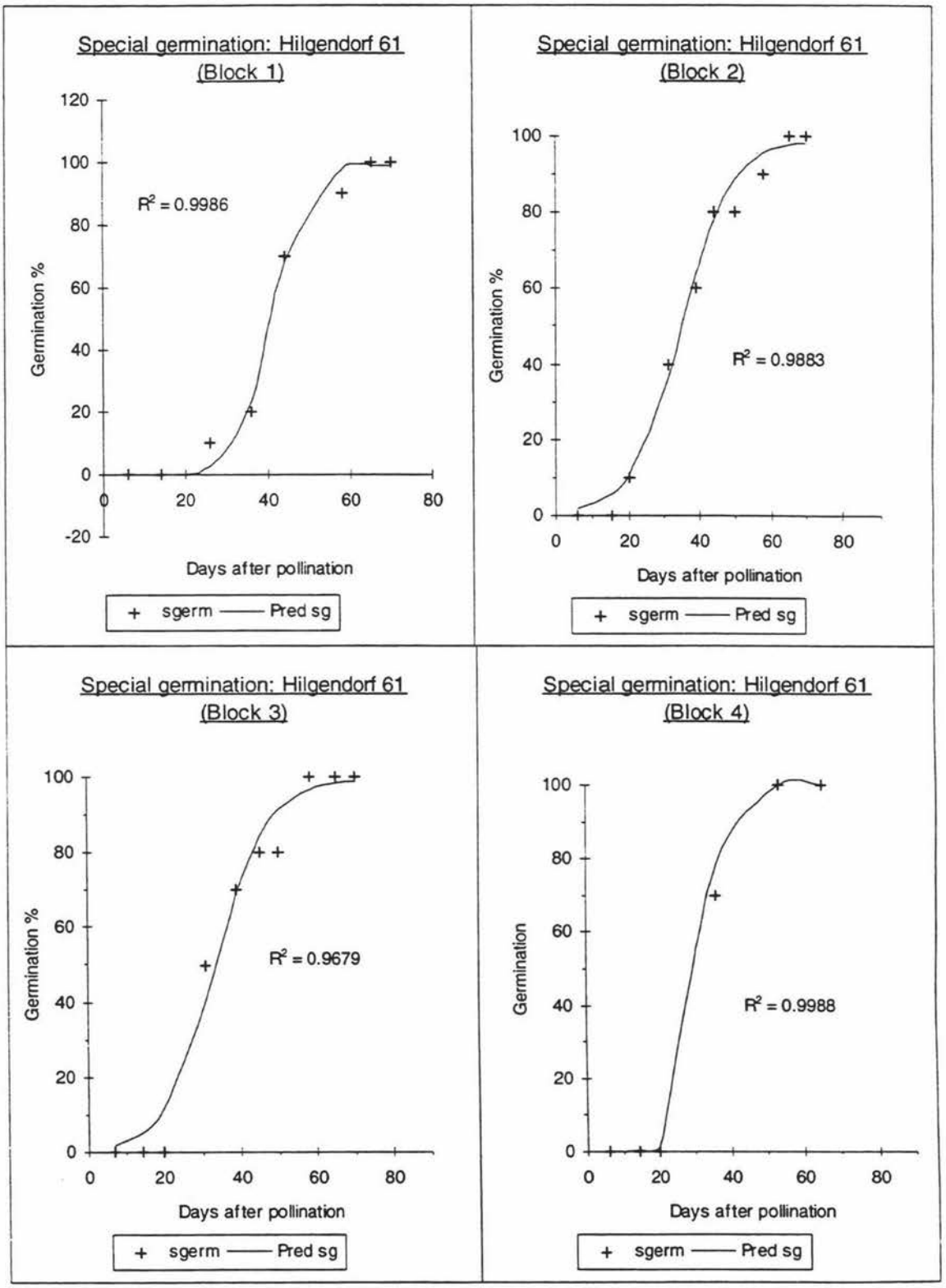


Table A 5.1 (j). Estimated statistics of the Richards functions fitted on data for special germination.

cv. Karamu

Replicate:	1	2	3	4
Statistic				
A	99.50090	99.00000	100.00000	99.00000
s.e. A	6.04718	4.89434	3.70448	2.74744
B	10.50019	6.30000	10.99566	7.30000
s.e. B	14.61622	4.02817	5.86336	3.76926
K	0.30100	0.15100	0.30124	0.20100
s.e. K	0.31408	0.06649	0.12391	0.06626
V	1.04909	0.95000	0.99297	1.00000
s.e. V	3.01256	0.81514	1.00934	0.91389
F value	118.4939 ***	320.5324 ***	437.2649 ***	585.5569 ***
R^2	0.9795	0.9887	0.9960	0.9958

significance level
*** P < 0.001

Figure A 5.1 (j). The Richards function fits for special germination of the four replicates of cv. Karamu.

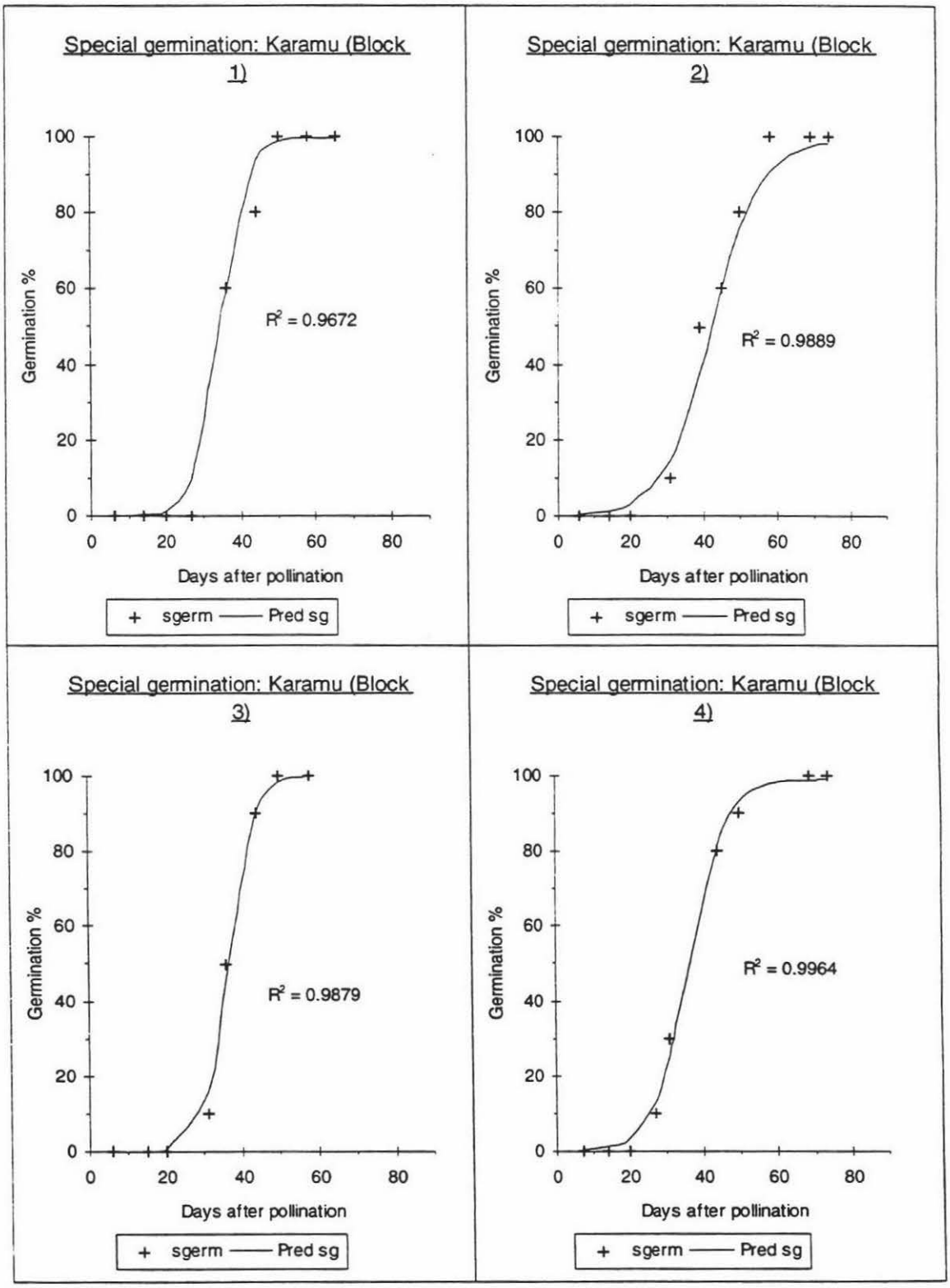


Table A 5.2 (a). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Gamenya

Replicate:	1	2	3	4
Statistic				
A	99.11538	99.51916	99.04431	98.00000
s.e. A	2.49734	5.64516	3.62019	5.32997
B	11.54514	8.44538	8.13411	11.60012
s.e. B	2.54154	15.90772	12.30621	8.97331
K	0.34710	0.30091	0.25133	0.35100
s.e. K	0.57684	0.31591	0.41206	0.20765
V	1.02174	1.19869	1.09678	0.94968
s.e. V	4.21780	7.88709	2.30597	1.34131
F_{regr}	516.1019 ***	137.8153 ***	135.1358 ***	252.6103 ***
R^2	0.9960	0.9841	0.9900	0.9966

significance level
*** $P < 0.001$

Figure A 5.2 (a). The Richards function fits for normal germination of the four replicates of cv. Gamenya.

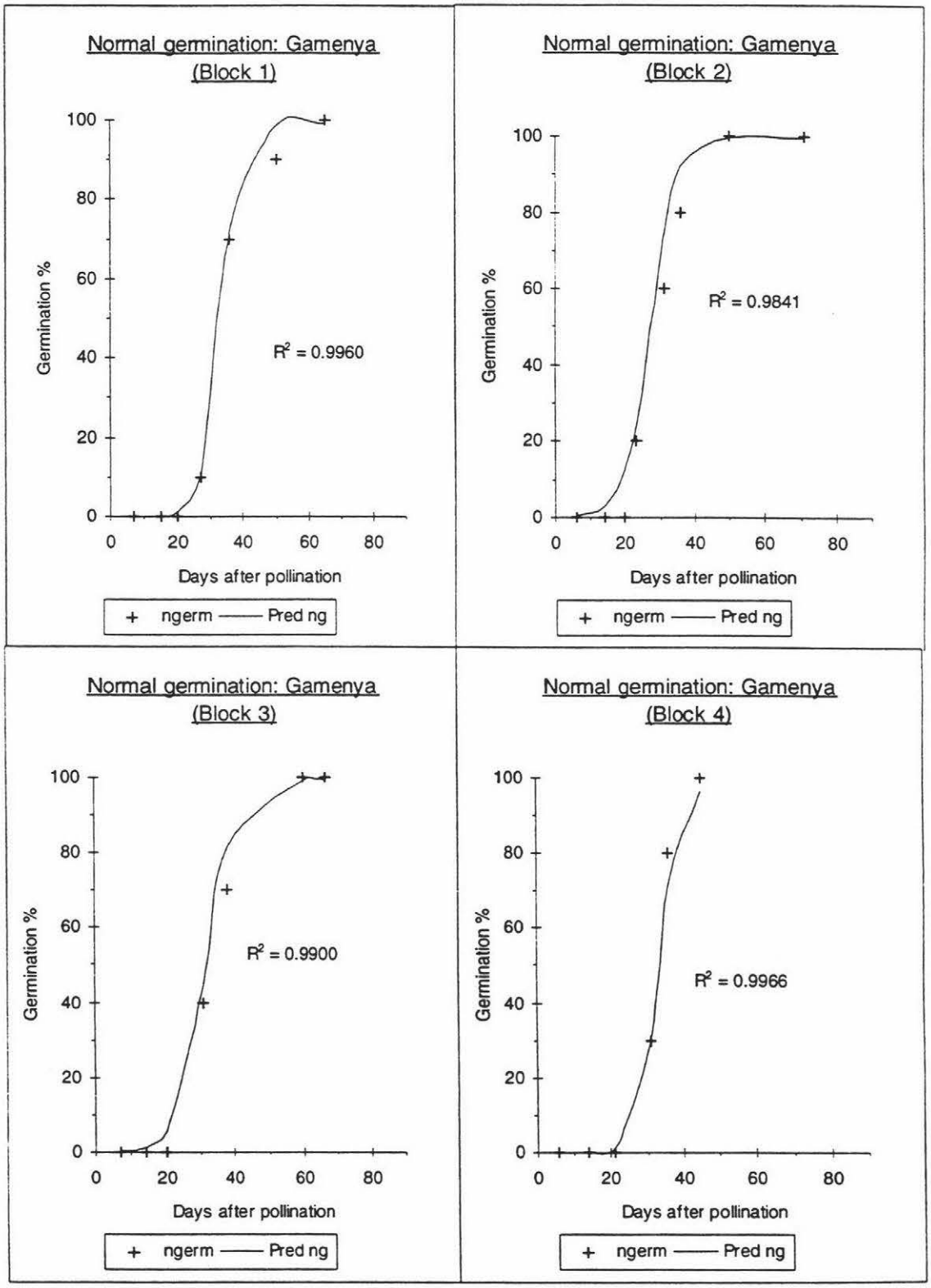


Table A 5.2 (b). Estimated statistics of the Richards functions fitted on data for normal germination.

cv.Tordo

Replicate:	1	2	3	4
Statistic				
A	98.00000	99.00000	99.00000	99.50000
s.e. A	3.979494	9.36089	4.01623	7.42589
B	10.65000	11.05003	11.35	11.15000
s.e. B	5.58439	1.74776	5.68921	6.53234
K	0.25100	0.251001	0.251	0.30100
s.e. K	0.89676	0.29926	0.26015	0.29104
V	1.50000	1.04968	1.00000	0.90000
s.e. V	1.87468	3.56094	1.21032	1.20564
F_{regr}	12.2621 NS	54.7561 *	30.4160 *	10.3245 NS
R^2	0.9510	0.9744	0.9548	0.9958

significance level

*** P < 0.001

Figure A 5.2 (b). The Richards function fits for normal germination of the four replicates of cv. Tordo.

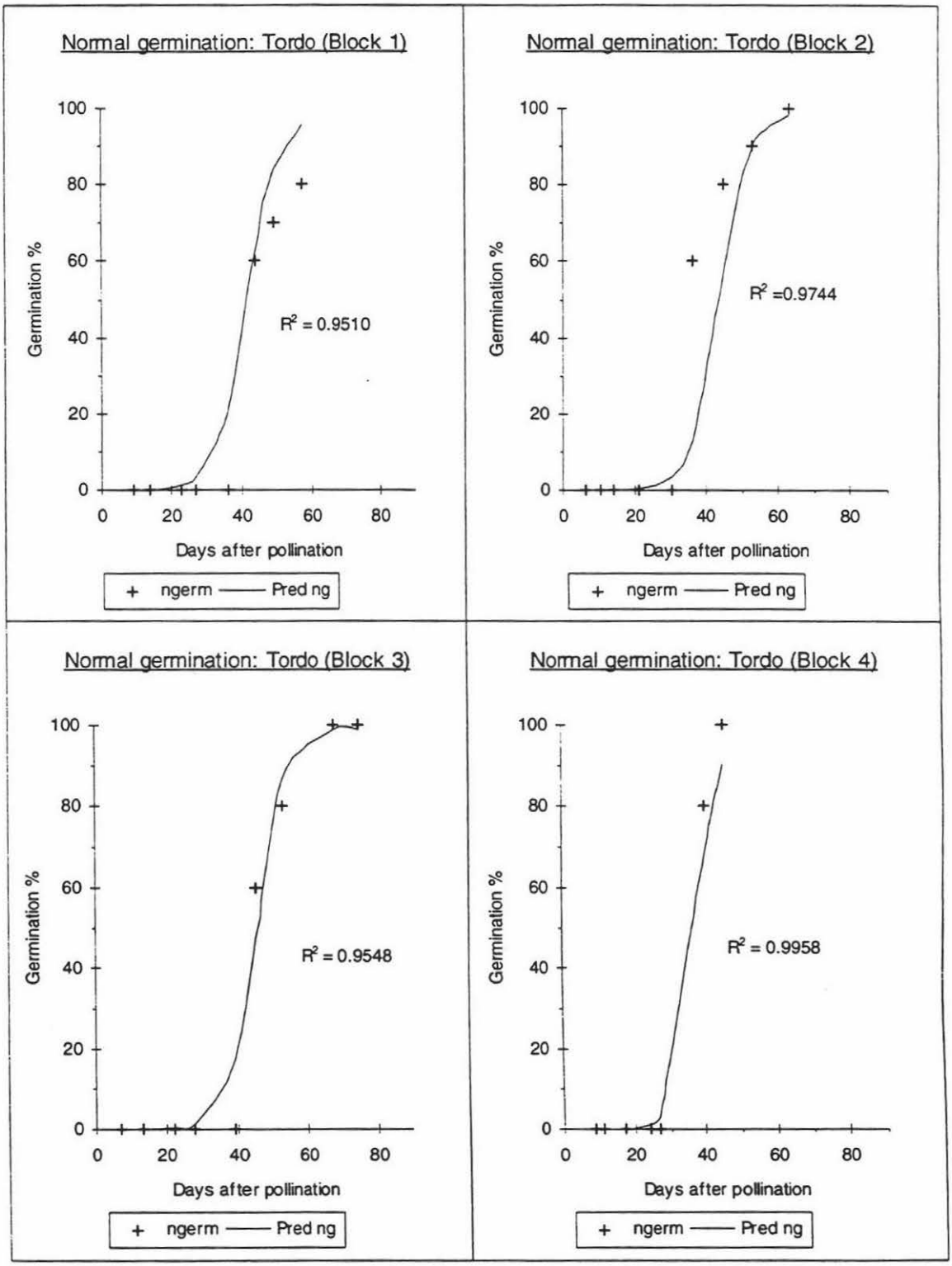


Table A 5.2 (c). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Kenya 321

Replicate:	1	2	3	4
Statistic				
A	99.00000	98.00000	99.91592	100.00000
s.e. A	2.81158	8.69301	8.88206	3.68409
B	6.45000	8.90093	6.49969	6.35000
s.e. B	3.46218	10.49812	4.80224	2.92302
K	0.15100	0.20113	0.15091	0.15100
s.e. K	0.05222	0.15836	0.08152	0.04724
V	1.00000	1.09956	0.95466	1.00000
s.e. V	0.89143	3.27024	0.91116	0.70343
F_{regr}	623.8957 ***	192.3180 ***	279.0464 ***	755.5193 ***
R^2	0.9943	0.9877	0.9900	0.9960

significance level
*** $P < 0.001$

Figure A 5.2 (c) The Richards function fits for normal germination of the four replicates of cv. Kenya 321.

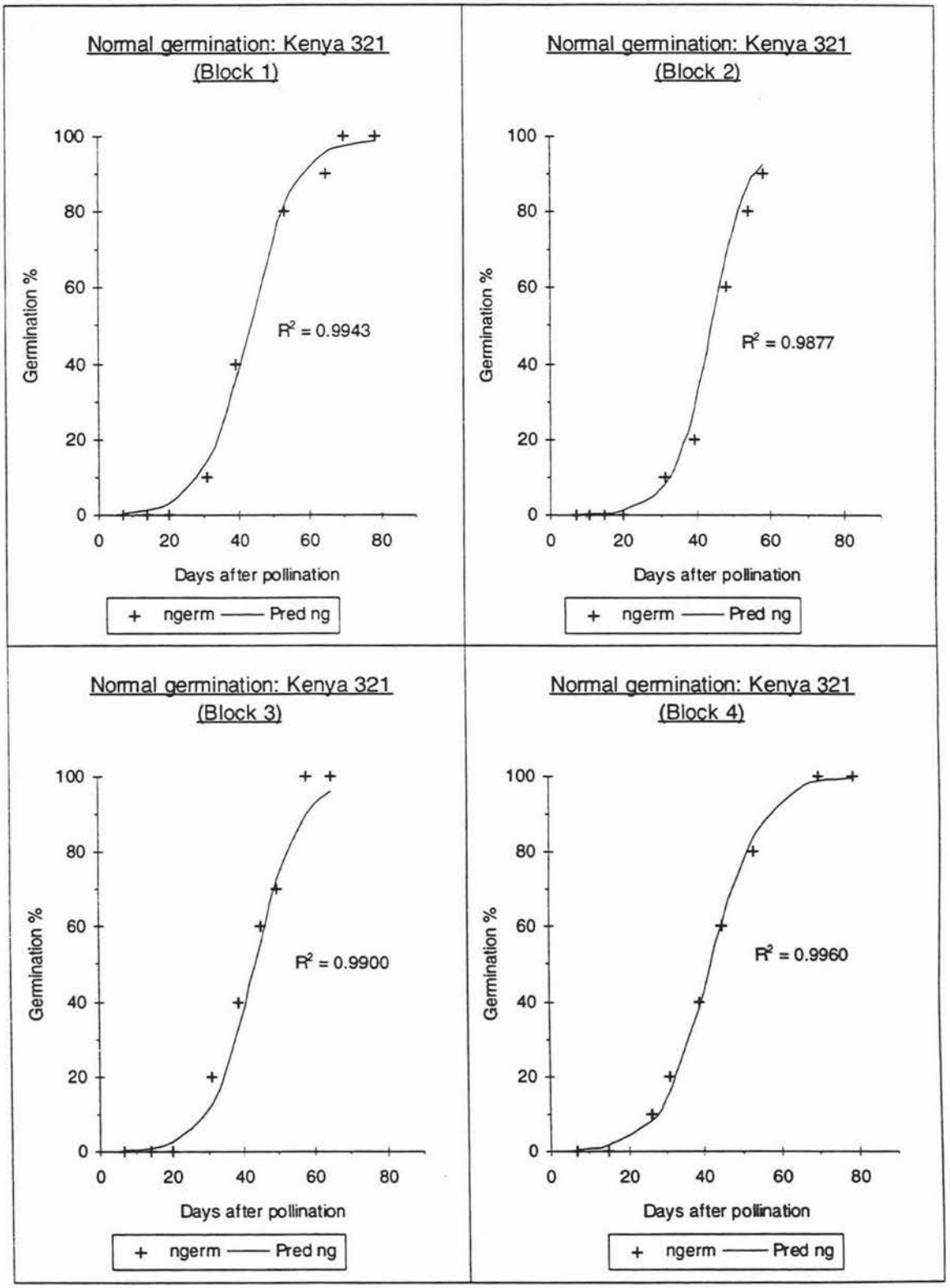


Table A 5.2 (d). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Brevor

Replicate:	1	2	3	4
Statistic				
A	98.00000	100.00000	100.00000	98.07757
s.e. A	18.76743	9.25457	9.25457	15.88393
B	12.00000	11.85000	11.85000	9.38906
s.e. B	20.53837	11.13308	11.13308	13.05879
K	0.30100	0.25100	0.25100	0.20125
s.e. K	0.41888	0.19650	0.19650	0.22110
V	0.90000	0.95000	0.95000	0.90973
s.e. V	2.44481	1.20585	1.20585	1.64272
F_{regr}	25.2899 NS	166.8690 ***	166.8690 ***	76.5378 *
R^2	0.9790	0.9790	0.9920	0.9848

significance level
NS not significant
* $P < 0.05$
*** $P < 0.001$

Figure A 5.2 (d) The Richards function fits for normal germination of the four replicates of cv. Brevor.

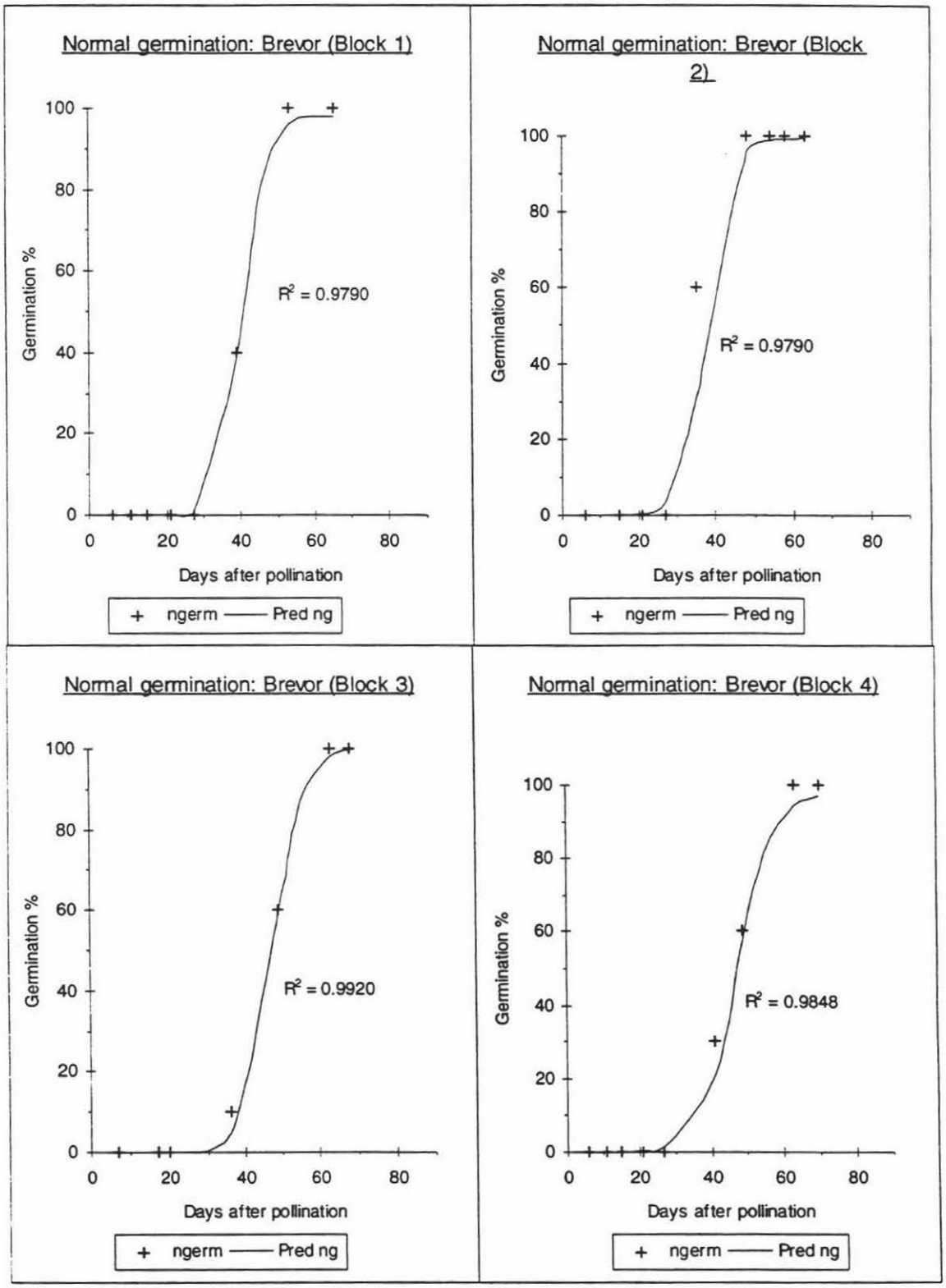


Table A 5.2 (e). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Isis

Replicate:	1	2	3	4
Statistic				
A	99.98539	99.97067	100.00000	100.00000
s.e. A	9.56102	5.72849	6.33739	6.69880
B	10.94923	11.33647	9.95000	8.55760
s.e. B	8.51712	8.73453	8.86848	5.19241
K	0.30142	0.25413	0.25100	0.20116
s.e. K	0.18895	0.12988	0.16397	0.09267
V	0.94260	1.18574	1.05000	0.99021
s.e. V	1.11191	3.38909	1.96852	0.96184
F_{regr}	127.2968 **	543.6129 ***	2.05.5702 ***	409.7426 ***
R^2	0.9927	0.9953	0.9895	0.9959

significance level
** P < 0.01
*** P < 0.001

Figure A 5.2 (e) The Richards function fits for normal germination of the four replicates of cv. Isis.

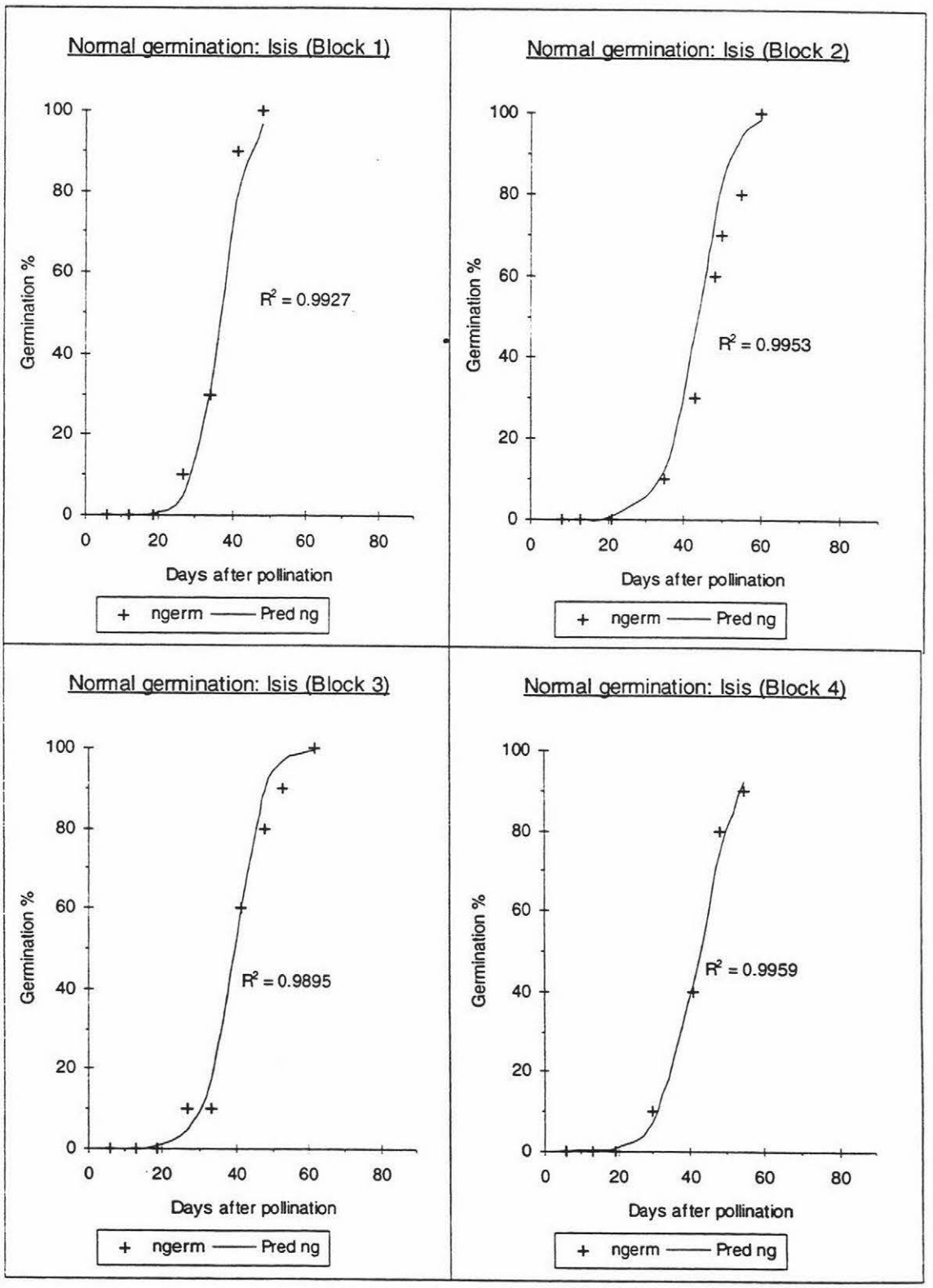


Table A 5.2 (f). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Sonora 64A

Replicate:	1	2	3	4
Statistic				
A	99.00337	98.02019	100.00000	99.01156
s.e. A	9.87774	10.68823	11.23333	9.69578
B	5.40108	9.30241	5.20598	7.15138
s.e. B	12.28617	7.31115	9.53628	3.56987
K	0.15100	0.25082	0.09830	0.15086
s.e. K	0.17591	0.28835	0.11245	0.16478
V	1.14775	1.05136	0.91784	1.00233
s.e. V	6.21843	3.66514	1.79542	4.35914
F_{regr}	72.6629 ***	73.2650 ***	195.6048 ***	364.6718 ***
R^2	0.9650	0.9773	0.9886	0.9929

significance level
*** P < 0.001

Figure A 5.2 (f) The Richards function fits for normal germination of the four replicates of cv. Sonora 64 A.

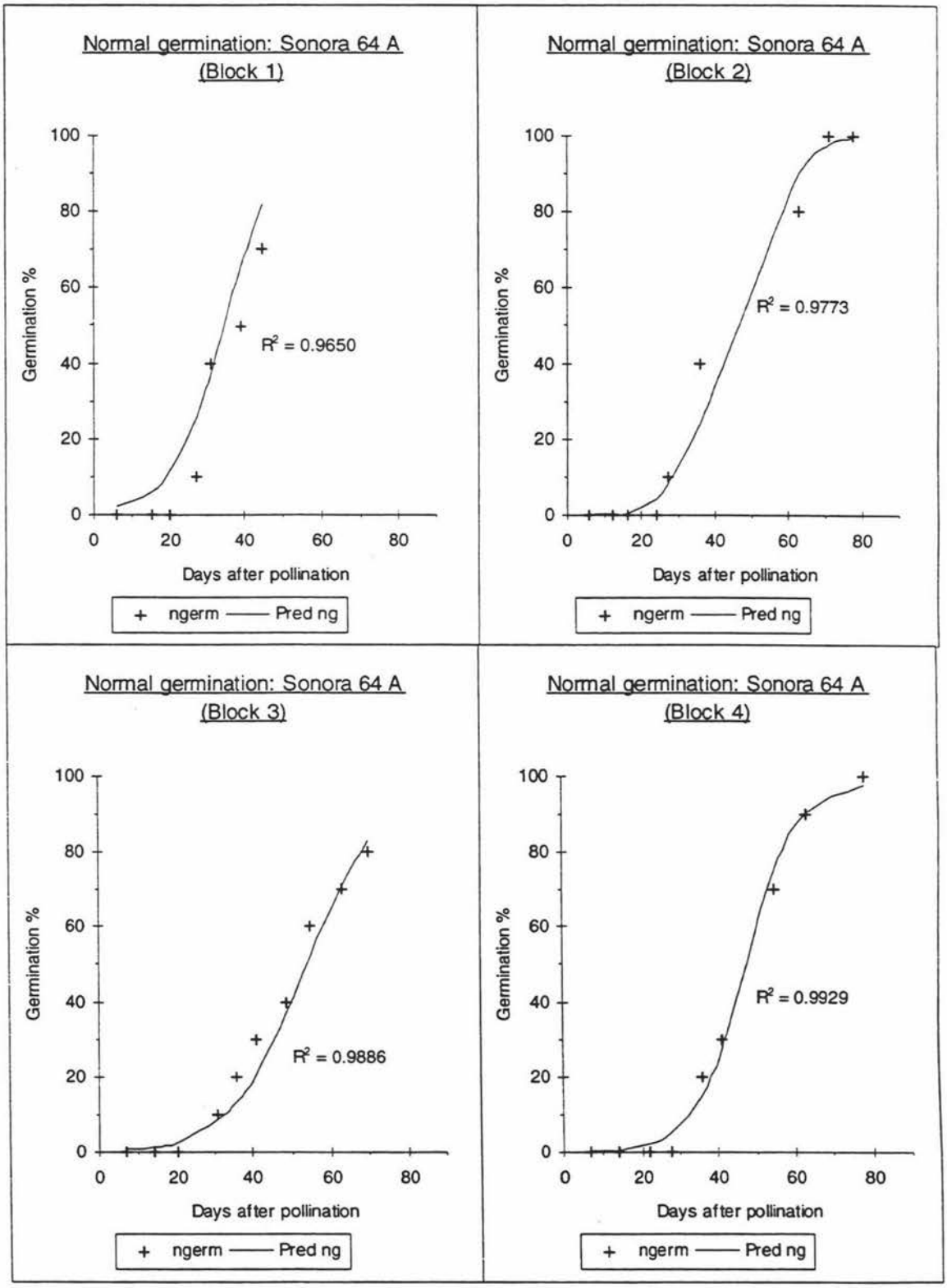


Table A 5.2 (g). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Thatcher

Replicate:	1	2	3	4
Statistic				
A	92.50049	99.00000	98.50000	98.00000
s.e. A	15.90833	12.56347	7.58323	36.39324
B	12.00026	7.50000	10.20000	10.60000
s.e. B	2.68610	12.65483	2.79475	7.01294
K	0.20100	0.10100	0.15100	0.15100
s.e. K	0.34765	0.12863	0.37396	1.05474
V	1.09928	0.95000	0.90000	0.85000
s.e. V	1.99653	1.56247	2.39137	3.21913
F_{regr}	85.8427 ***	278.6268 ***	26.7667 *	36.1818 NS
R^2	0.9751	0.9943	0.9439	0.9891

significance level
NS not significant
* $P < 0.05$
*** $P < 0.001$

Figure A 5.2 (g) The Richards function fits for normal germination of the four replicates of cv. Thatcher.

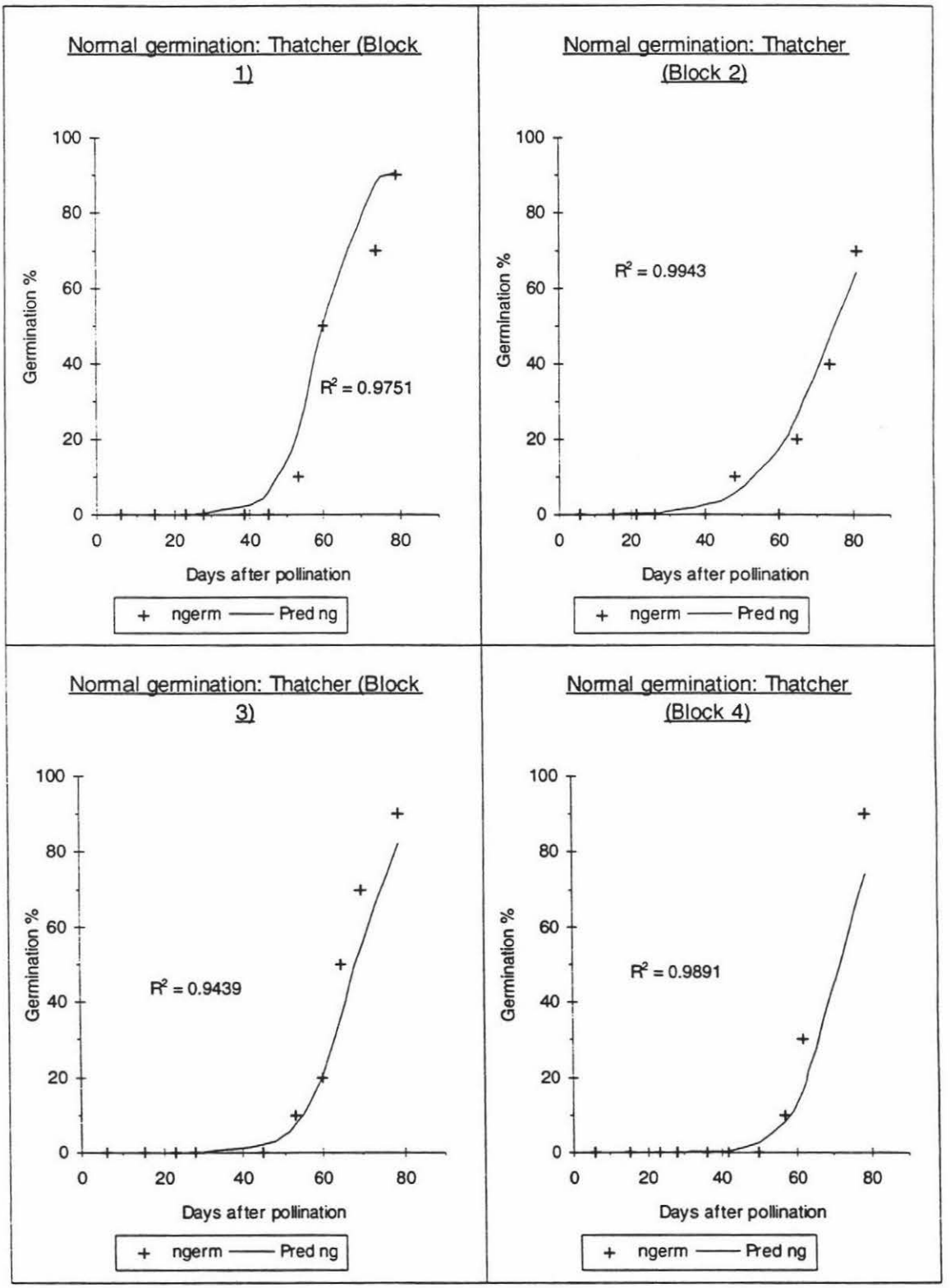


Table A 5.2 (h). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
A	94.901313	98.52124	99.49999	98.00000
s.e. A	30.94065	30.51125	27.84177	30.62907
B	10.79895	8.49928	4.50117	9.09997
s.e. B	12.00012	15.64154	2.54996	13.26810
K	0.25106	0.15104	0.10100	0.15100
s.e. K	0.23406	0.20322	0.18103	0.16238
V	1.34847	1.09766	1.34758	1.15007
s.e. V	3.06254	4.38907	2.78568	2.67815
F_{regr}	62.7517 ***	154.0595 ***	65.0885 ***	90.9984 *
R^2	0.9718	0.9891	0.9597	0.9899

significance level
* P < 0.05
*** P < 0.001

Figure A 5.2 (h) The Richards function fits for normal germination of the four replicates of cv. Hilgendorf 61.

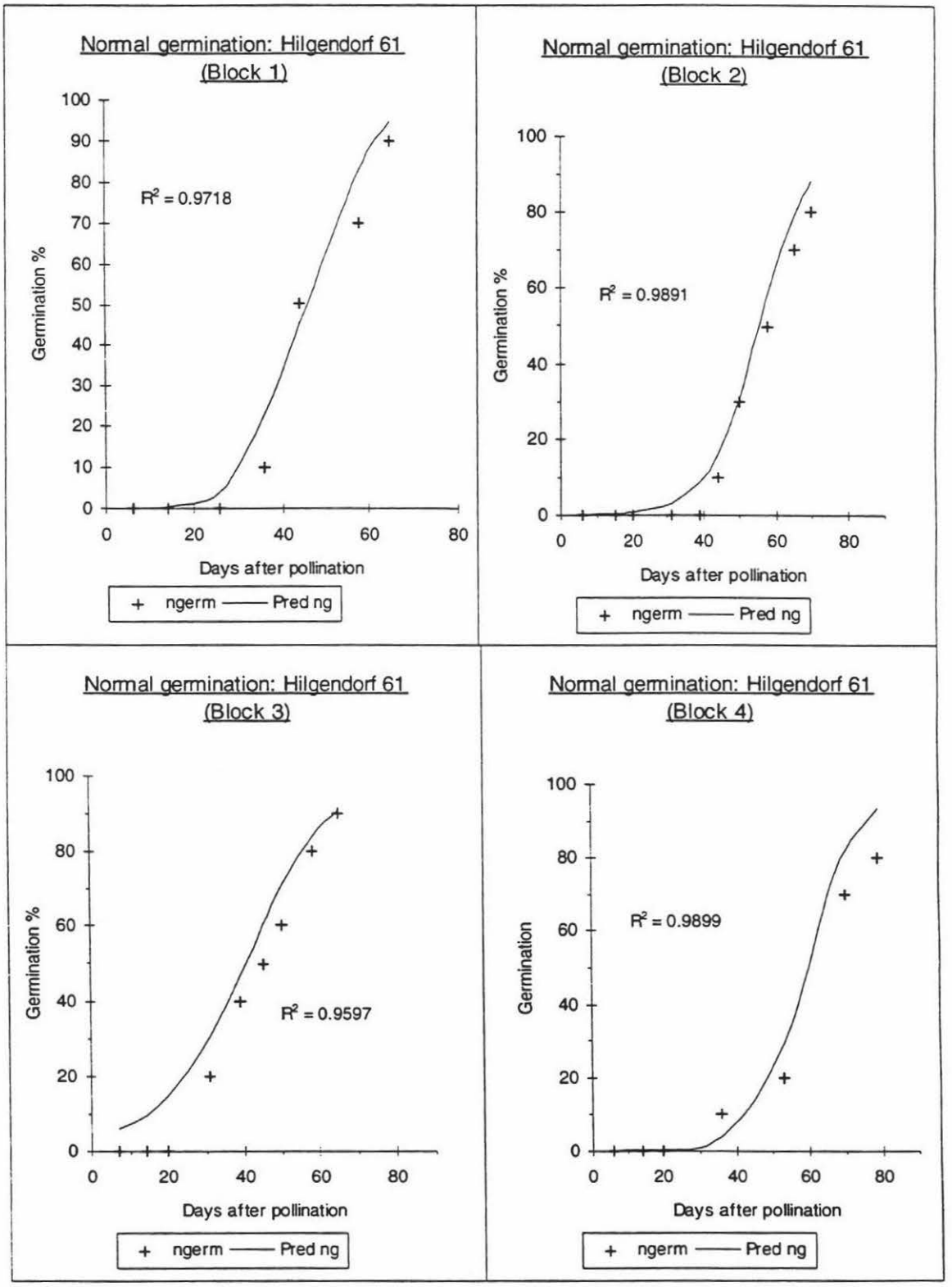


Table A 5.2 (i). Estimated statistics of the Richards functions fitted on data for normal germination.

cv. Karamu

Replicate:	1	2	3	4
Statistic				
A	91.99182	92.04024	99.50016	93.00018
s.e. A	16.89258	6.46048	15.94555	6.01555
B	6.50129	10.79994	7.00013	11.55003
s.e. B	4.06149	15.41856	8.95639	10.67492
K	0.10098	0.20091	0.15100	0.25100
s.e. K	0.11406	0.15774	0.13557	0.17020
V	0.60007	1.20153	1.04962	1.04969
s.e. V	0.21070	0.99950	2.40550	244825
F_{regr}	224.8425 ***	238.5067 ***	166.8035 ***	154.7747 ***
R^2	0.9933	0.9898	0.9889	0.9919

significance level
** P < 0.01
*** P < 0.001

Figure A 5.2 (i) The Richards function fits for normal germination of the four replicates of cv. Karamu.

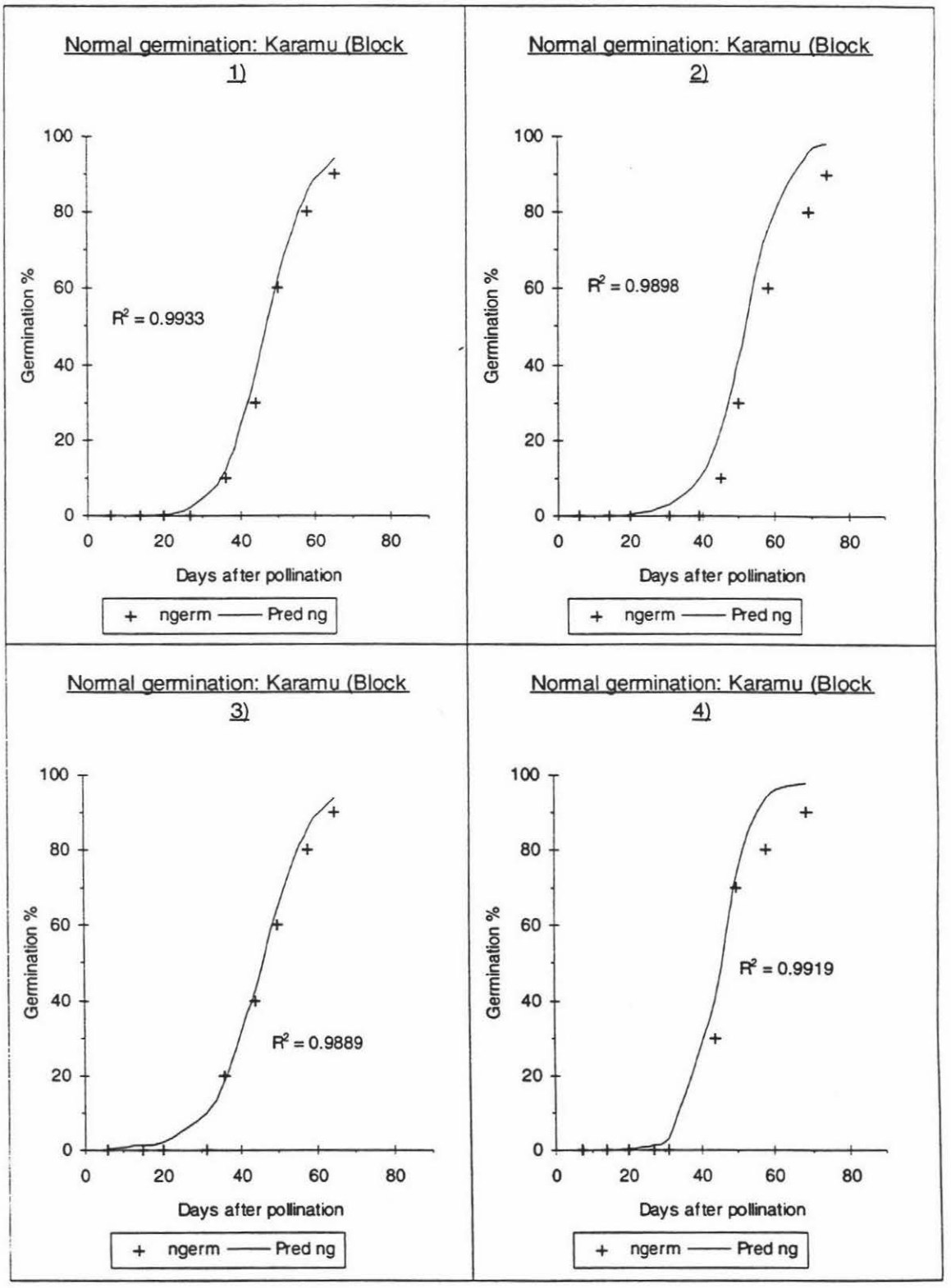


Table A 5.3 (a). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Gamenya

Replicate:	1	2	3	4
Statistic				
A	100.86785	100.00000	99.99260	100.00000
s.e. A	8.65298	14.52961	8.79370	9.75084
B	-9.14962	-3.10000	-6.00164	-9.55000
s.e. B	2.33561	1.56980	2.77468	2.56321
K	-0.30074	-0.15000	-0.19997	-0.35000
s.e. K	0.12908	0.11011	0.15899	1.96541
V	0.97006	0.85000	0.95197	0.95000
s.e. V	1.23508	1.00410	1.59393	1.33551
F_{regr}	291.9199 ***	20.0632	139.5540	191.4140
R^2	0.9924	0.9144	0.9844	0.9941

significance level
*** $P < 0.001$

Figure A 5.3 (a) The Richards function fits and observed data for dormancy of the four replicates of cv. Gamenya.

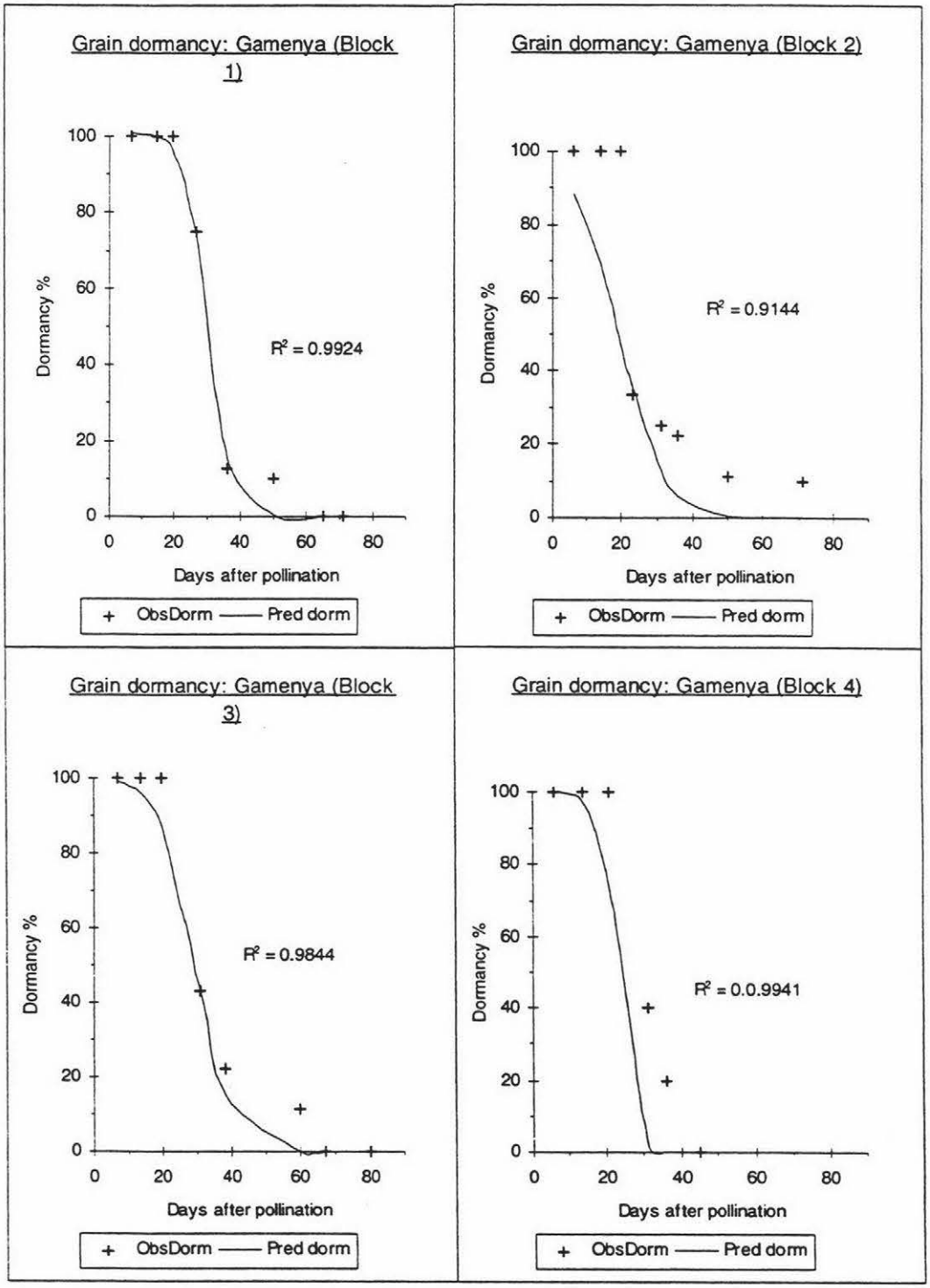


Table A 5.3 (b). Estimated statistics of the Richards functions fitted on data for dormancy.

cv.Tordo

Replicate:	1	2	3	4
Statistic				
A	100.00000	100.00000	100.00000	98.88543
s.e. A	10.91496	13.93590	12.60016	14.55982
B	-8.35000	-8.45000	-8.70000	-10.17069
s.e. B	3.10015	5.08661	4.12396	2.46789
K	-0.20000	-0.20000	-0.20000	-0.29709
s.e. K	0.08352	0.12046	0.14946	0.133924
V	0.95000	0.95000	0.95000	0.93590
s.e. V	1.30264	0.89701	0.79648	0.76527
F_{regr}	67.8018 ***	131.9733 ***	56.4191 ***	254.2704 ***
R^2	0.9457	0.9642	0.9415	0.9883

significance level
*** $P < 0.001$

Figure A 5.3 (b) The Richards function fits and observed data for dormancy of the four replicates of cv. Tordo.

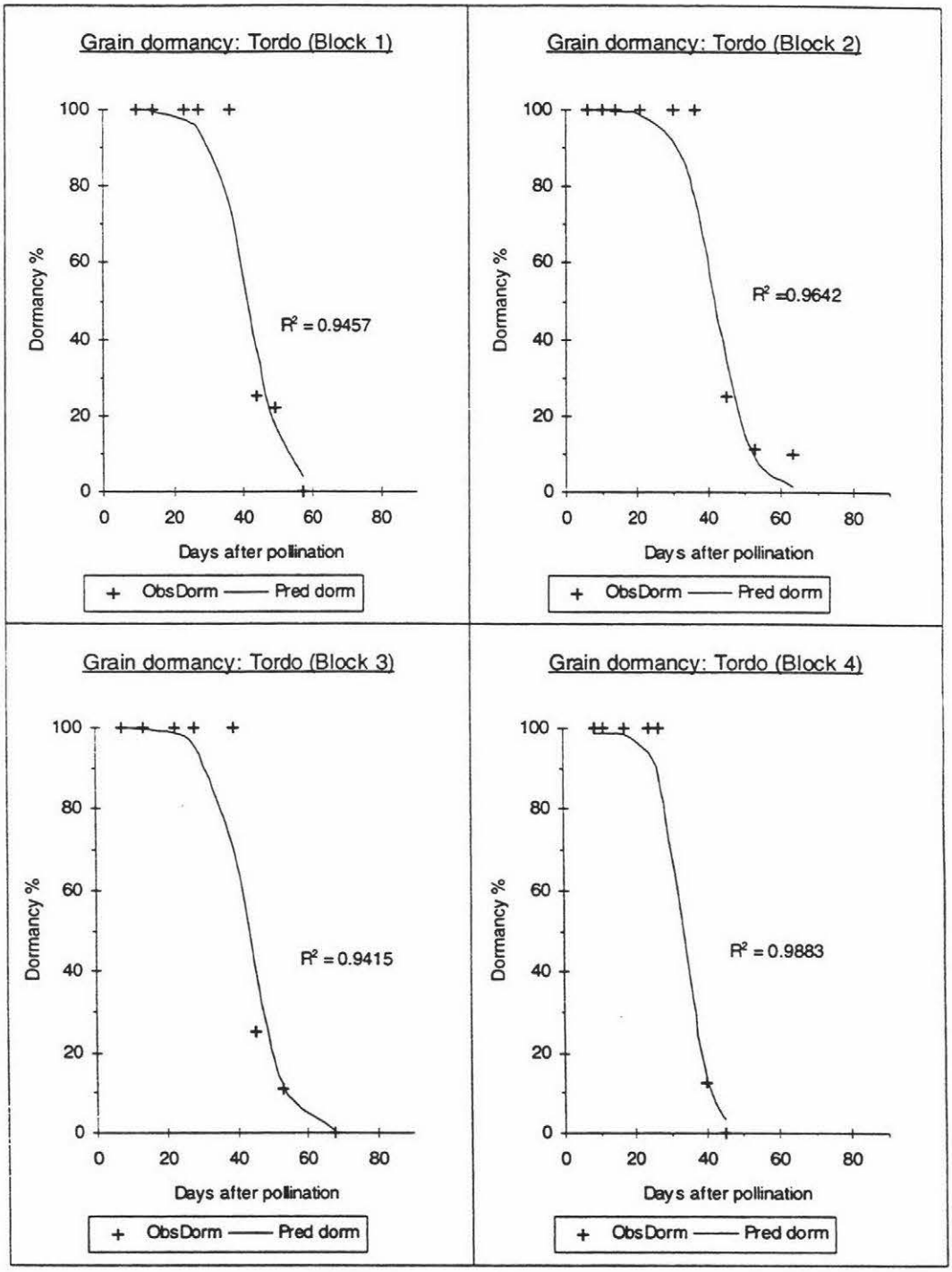


Table A 5.3 (c). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Kenya 321

Replicate:	1	2	3	4
Statistic				
A	100.00000	101.48171	99.93115	100.51445
s.e. A	13.02641	10.98564	16.71333	11.99931
B	-6.25000	-6.62371	-3.54798	-2.83084
s.e. B	3.52641	2.53104	1.71482	1.61015
K	-0.15000	-0.15396	-0.10016	-0.08660
s.e. K	0.088425	0.09830	0.08143	0.06327
V	1.00000	1.00651	0.89556	0.88061
s.e. V	0.91064	0.90102	0.92277	0.98585
F_{regr}	561.1335 ***	545.0029 ***	144.7886 ***	216.3536 ***
R^2	0.9942	0.9897	0.9763	0.9844

significance level
*** P < 0.001

Figure A 5.3 (c) The Richards function fits and observed data for dormancy of the four replicates of cv. Kenya 321.

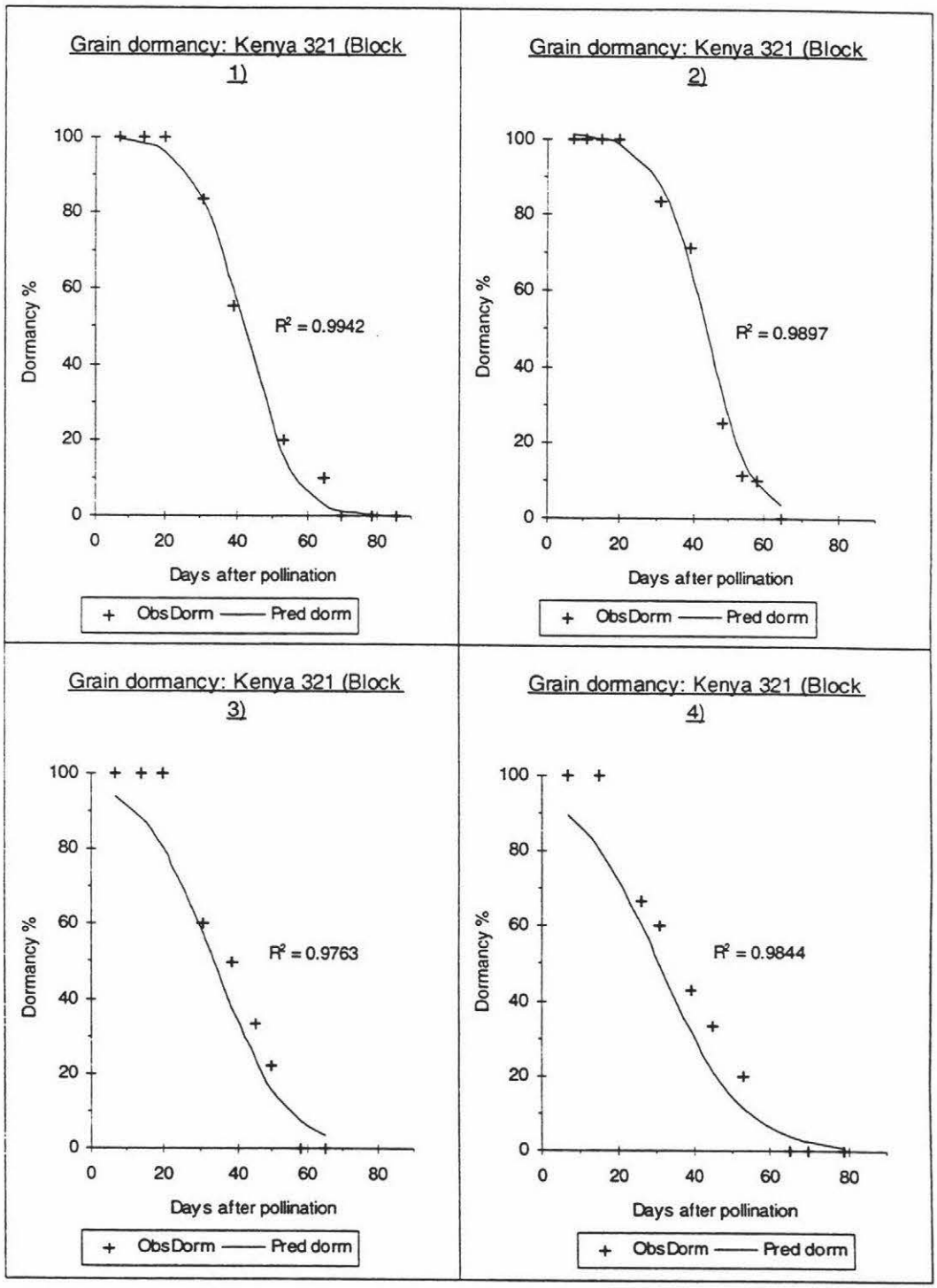


Table A 5.3 (d). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Brevor

Replicate:	1	2	3	4
Statistic				
A	98.52680	100.00000	98.89329	100.12484
s.e. A	11.62803	9.12803	9.68014	11.00901
B	-10.20882	-9.90000	-8.89114	-8.79847
s.e. B	3.02839	4.32831	2.12843	3.52829
K	-0.29602	-0.30000	-0.19964	-0.2001
s.e. K	0.20992	0.20992	0.20992	0.20992
V	0.96011	0.98010	0.79018	0.96237
s.e. V	1.98252	1.98252	1.98252	1.98252
F_{regr}	786.6536 ***	306.4819 ***	81.7848 ***	518.4746 ***
R^2	0.9959	0.9918	0.9732	0.9919

significance level
*** P < 0.001

Figure A 5.3 (d) The Richards function fits and observed data for dormancy of the four replicates of cv. Brevor.

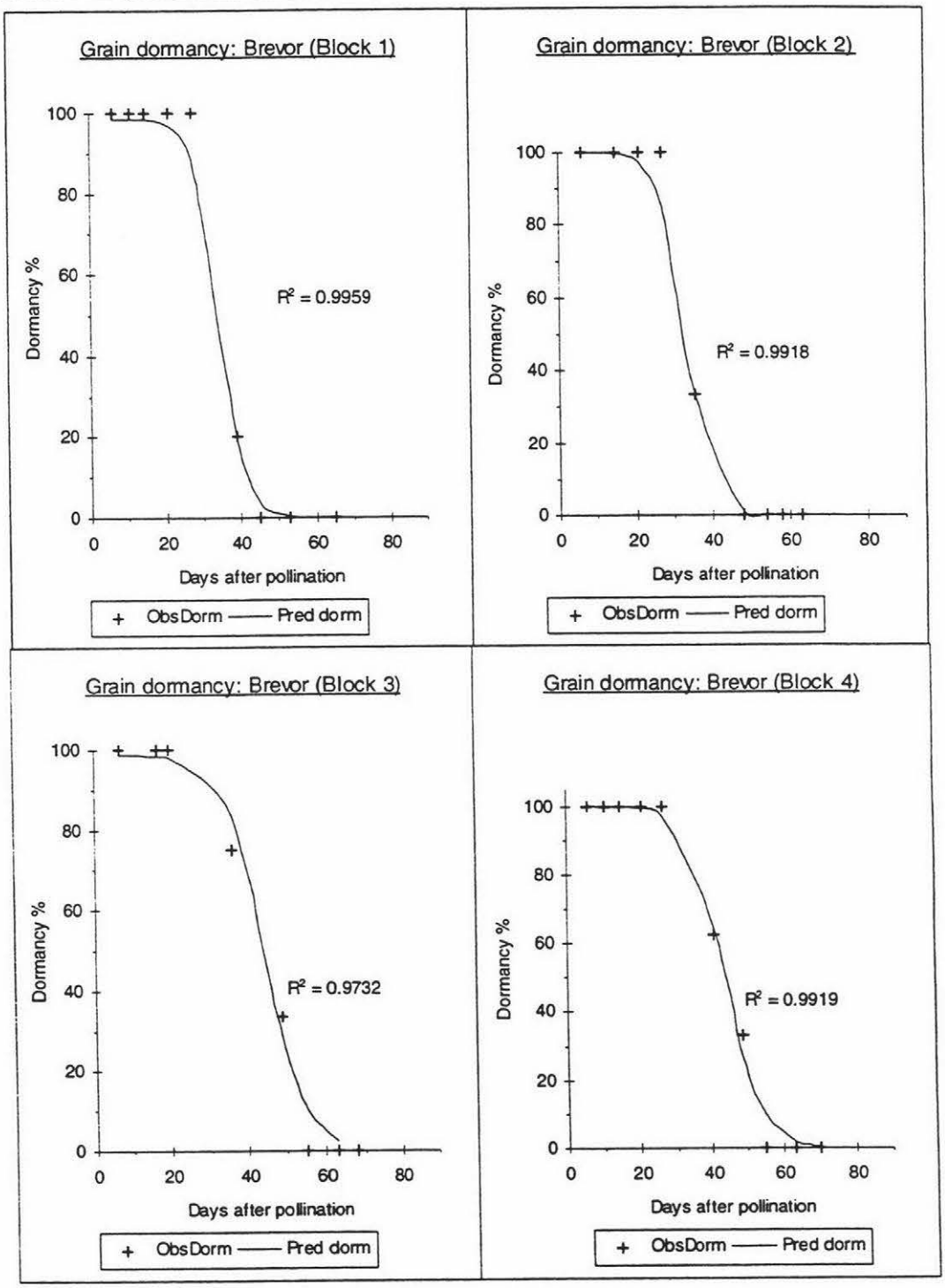


Table A 5.3 (e). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Isis

Replicate:	1	2	3	4
Statistic				
A	99.73389	102.87095	99.89738	99.82362
s.e. A	11.92803	8.84350	5.98340	6.78014
B	-5.79812	-4.27444	-5.15879	-5.24813
s.e. B	3.52833	1.11322	1.31053	1.76452
K	-0.19963	-0.10043	-0.14956	-0.15053
s.e. K	0.20992	0.06433	0.07062	0.05326
V	0.96319	0.97572	0.96309	0.95221
s.e. V	1.98252	1.34667	0.86386	0.92015
F_{regr}	66.6017 ***	362.7782 ***	349.6586 ***	159.4547 ***
$\sigma_{y.x}$	11.2670	5.30697	5.36160	7.54378
R^2	0.9664	0.9849	0.9895	0.9837

significance level
*** $P < 0.001$

Figure A 5.3 (e) The Richards function fits and observed data for dormancy of the four replicates of cv. Isis.

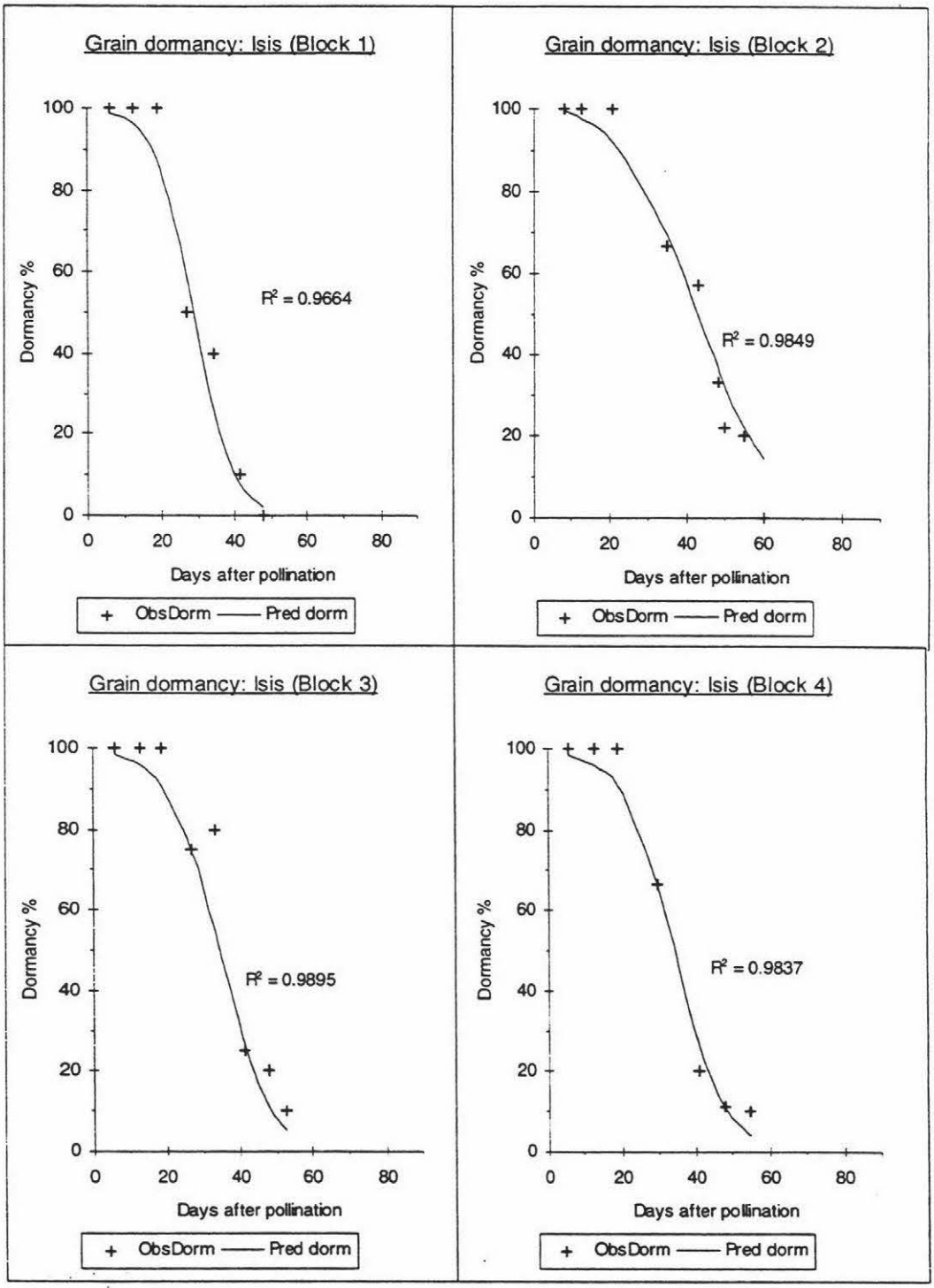


Table A 5.3 (f). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Sonora 64A

Replicate:	1	2	3	4
Statistic				
A	100.00000	100.00000	100.00000	100.00000
s.e. A	26.76715	15.23041	11.21023	13.20136
B	-3.35000	-3.95000	-4.90000	-7.10000
s.e. B	2.31457	1.68021	1.93863	2.43022
K	-0.10000	-0.10000	-0.10000	-0.15000
s.e. K	0.12336	0.09658	0.09256	0.097142
V	0.90000	0.95000	1.00000	1.00000
s.e. V	1.51615	1.23798	2.34483	1.03560
F_{regr}	61.2683 ***	145.4939 ***	105.1011 ***	642.6330 ***
R^2	0.9224	0.9729	0.9494	0.9930

significance level
*** P < 0.001

Figure A 5.3 (f) The Richards function fits and observed data for dormancy of the four replicates of cv. Sonora 64 A.

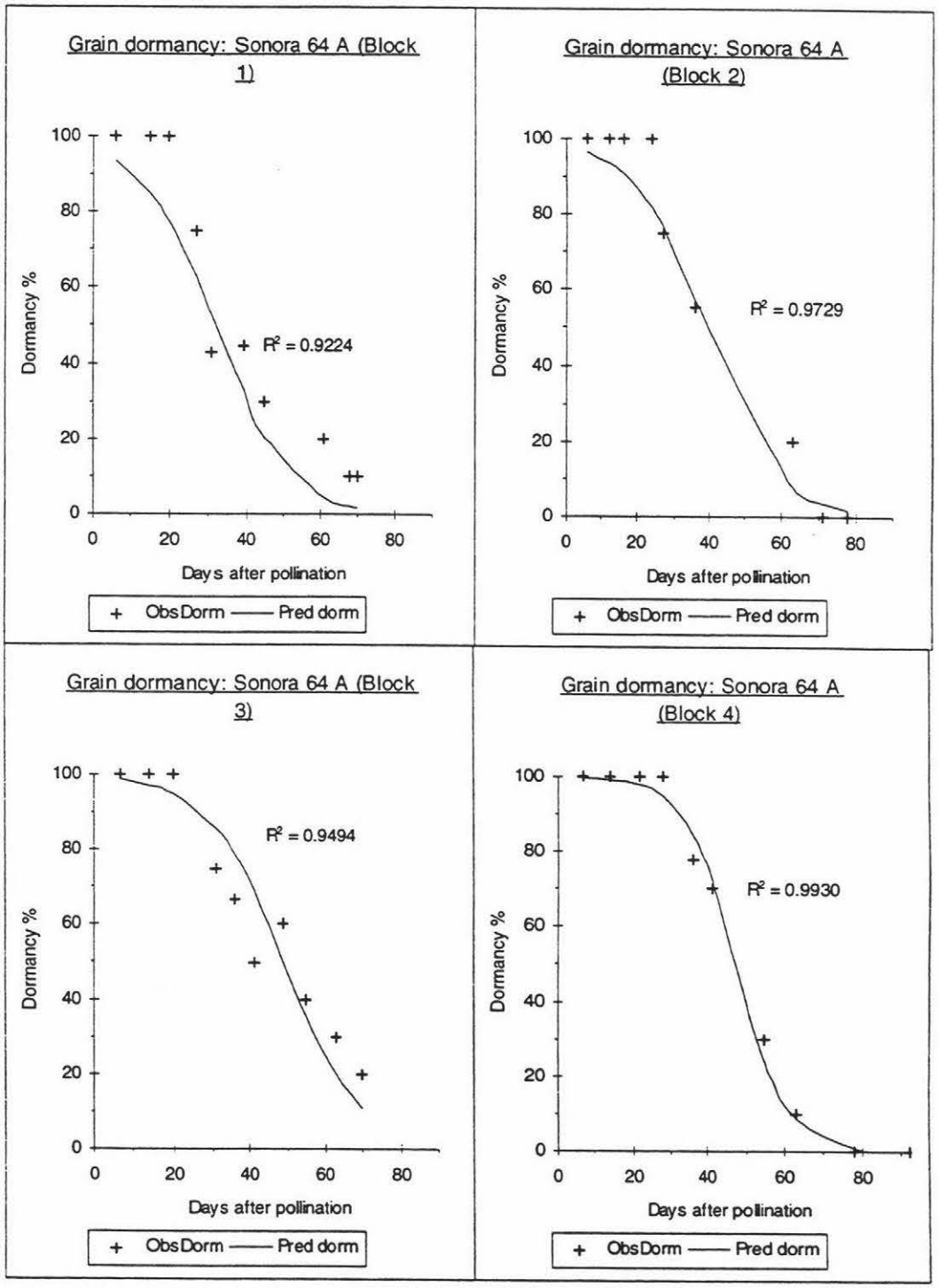


Table A 5.3 (g). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Thatcher

Replicate:	1	2	3	4
Statistic				
A	100.00000	100.00000	100.00000	100.98833
s.e. A	1.65342	1.93254	1.89530	1.77103
B	-9.40000	-7.25134	-9.65000	-10.07518
s.e. B	1.32654	1.83364	1.61195	2.36899
K	-0.15000	-0.09997	-0.15000	-0.14946
s.e. K	0.043352	0.03956	0.03945	0.05789
V	1.00000	1.00149	1.00000	0.99251
s.e. V	0.85641	0.72914	0.73961	1.03366
F_{regr}	1918.167 ***	1501.148 ***	1191.091 ***	1554.305 ***
R^2	0.9504	0.9873	0.9920	0.9892

significance level
*** $P < 0.001$

Figure A 5.3 (g) The Richards function fits and observed data for dormancy of the four replicates of cv. Thatcher.

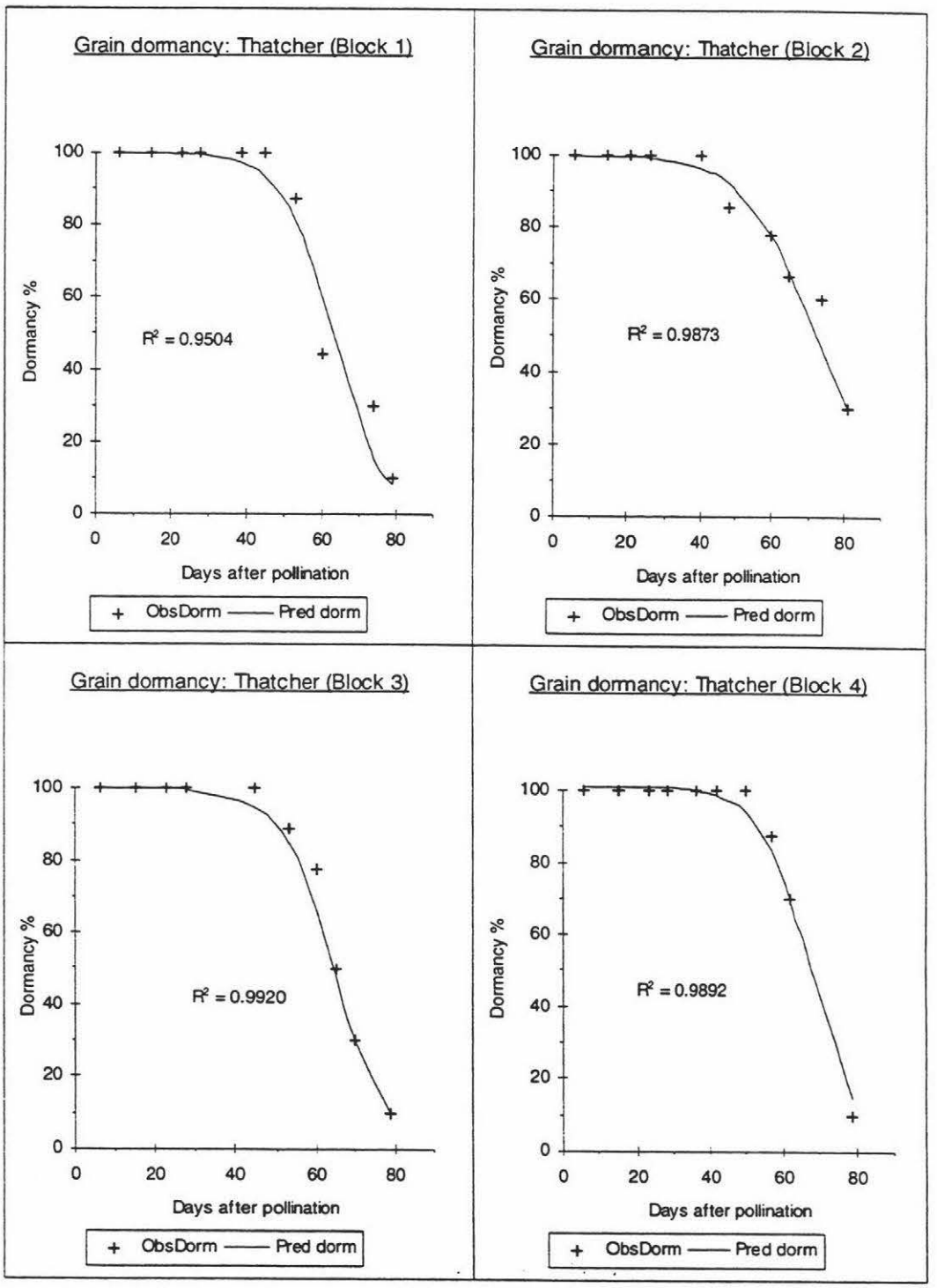


Figure A 5.3 (h) The Richards function fits and observed data for dormancy of the four replicates of cv. La Prevision.

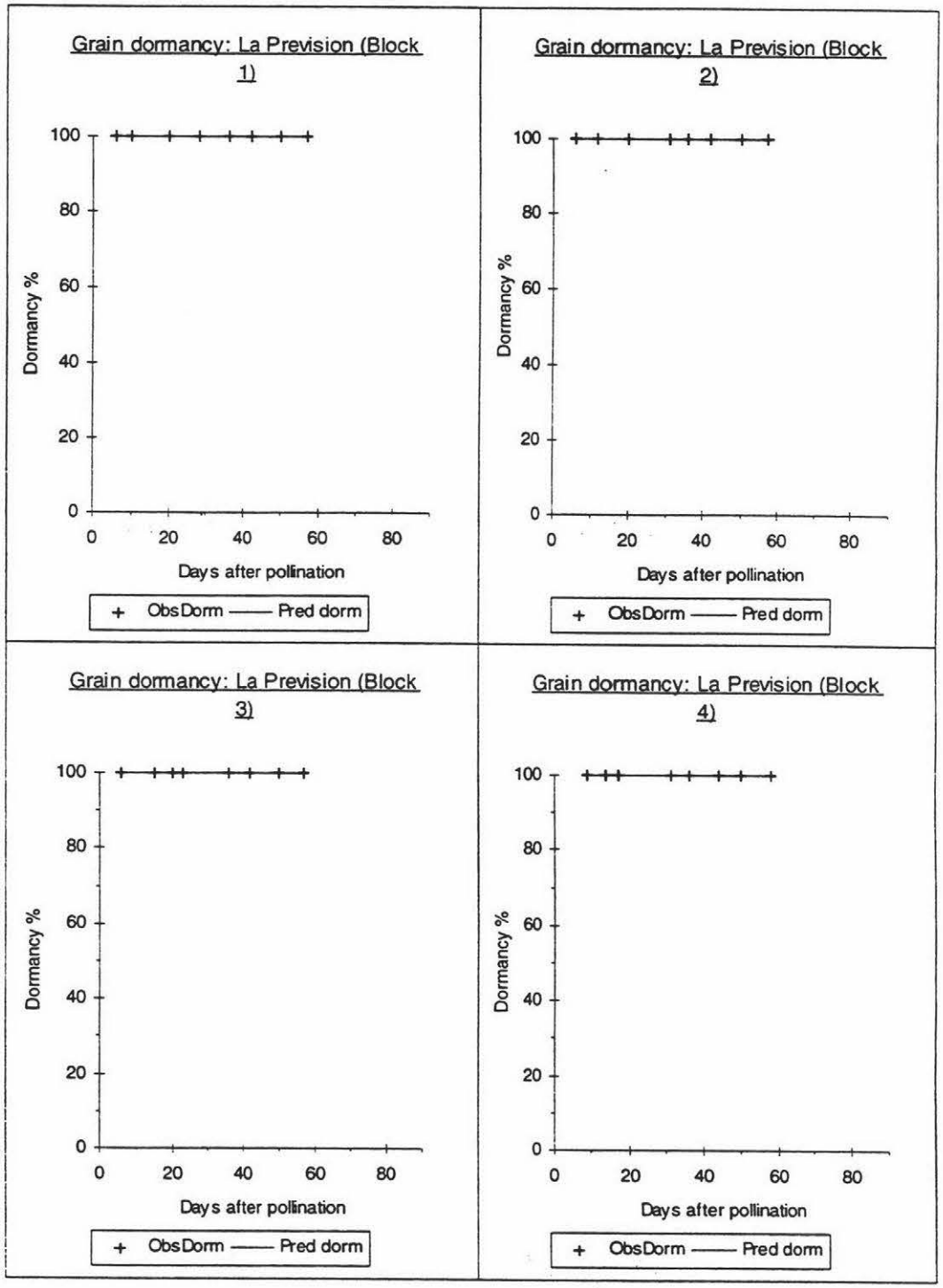


Table A 5.3 (i). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
A	101.36722	101.13245	101.37004	99.61982
s.e. A	4.52160	4.12688	4.54772	4.19750
B	-2.00091	-6.31327	-2.0004	-6.25120
s.e. B	1.72651	1.34123	1.53934	1.63490
K	-0.04861	-0.11203	-0.04853	-0.09979
s.e. K	0.01240	0.05339	0.06350	0.06481
V	0.84649	0.97789	0.84638	1.00905
s.e. V	0.71425	1.25925	1.23389	1.93584
F_{regr}	34.7268 ***	495.8106 ***	305.2613 ***	374.7106 ***
R^2	0.8712	0.9792	0.9626	0.9874

significance level
** P < 0.01
*** P < 0.001

Figure A 5.3 (i) The Richards function fits and observed data for dormancy of the four replicates of cv. Hilgendorf 61.

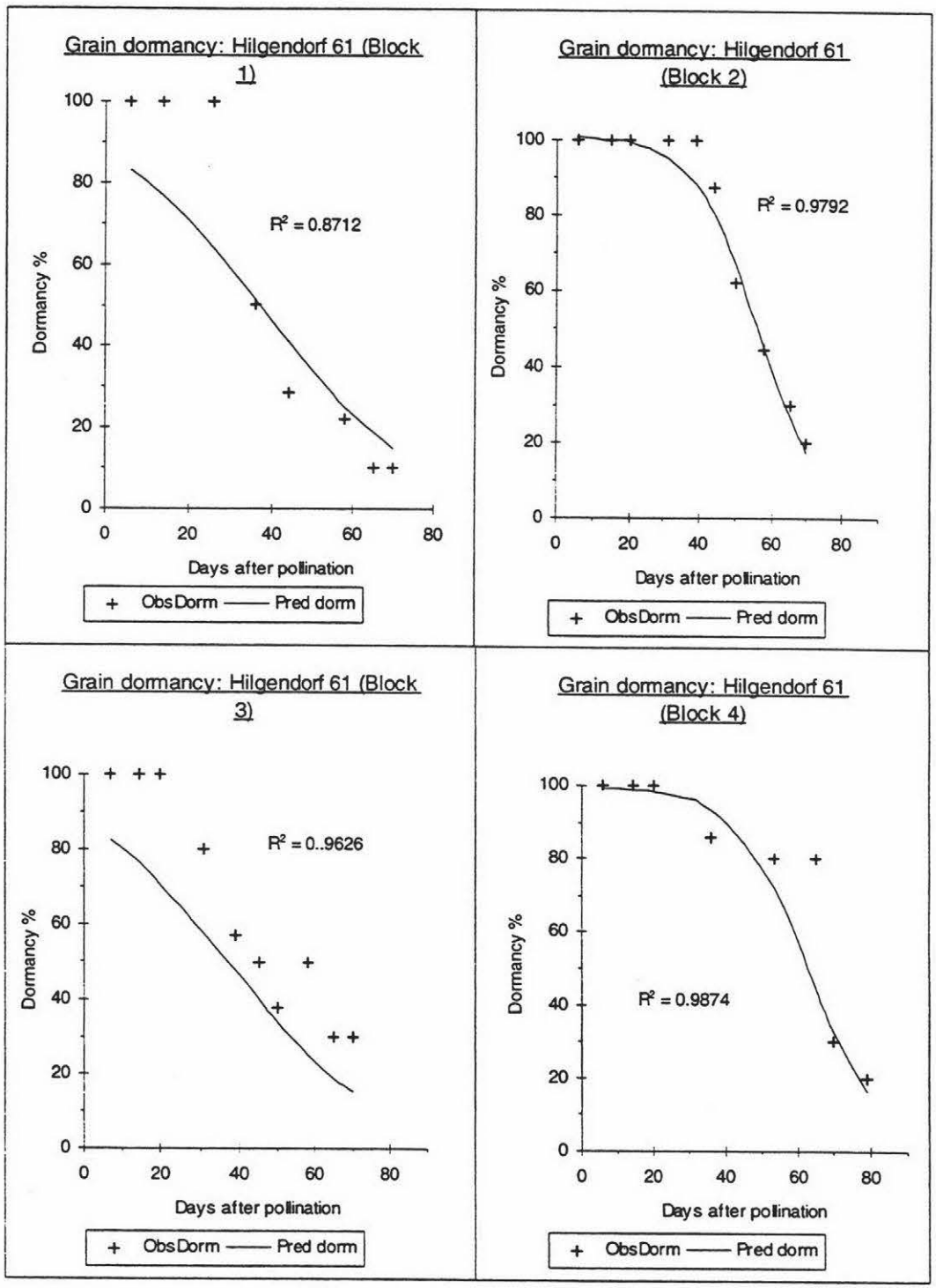


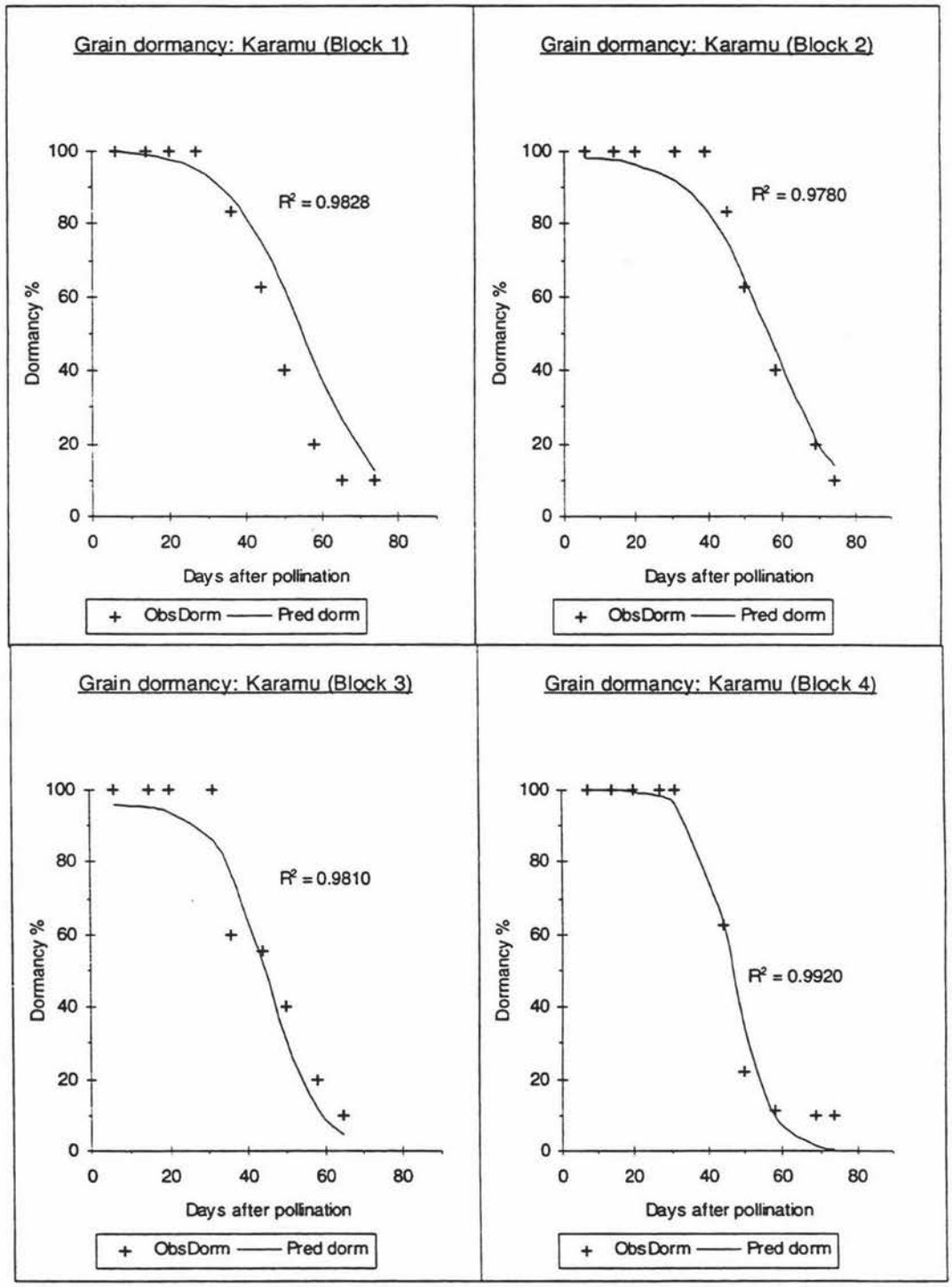
Table A 5.3 (j). Estimated statistics of the Richards functions fitted on data for dormancy.

cv. Karamu

Replicate:	1	2	3	4
Statistic				
A	100.89270	98.977	95.99015	100.0000
s.e. A	4.46125	4.45012	6.35140	4.85269
B	-5.50157	-5.7004	-6.8498	-9.35000
s.e. B	0.98057	1.23605	0.99031	3.26452
K	-0.10025	-0.10000	-0.14996	-0.20000
s.e. K	0.05417	0.09625	0.08631	0.09342
V	0.98702	0.95187	0.95242	1.00000
s.e. V	1.63391	1.02236	1.32064	1.68952
F_{regr}	653.7207 ***	642.7136 ***	544.2634 ***	667.8596 ***
R^2	0.9828	0.9780	0.9810	0.9920

significance level
*** P < 0.001

Figure A 5.3 (i) The Richards function fits and observed data for dormancy of the four replicates of cv. Karamu.



Appendix 4.

Table A 6.1 (a). The Richards function for Grain coat colour (statistics by replicate).

cv. Gamenya

Replicate: Statistic	1	2	3	4
A	1.97803	1.69968	2.09675	2.09675
s.e. A	0.17783	0.91606	0.51506	0.51506
B	5.52639	7.59924	8.04113	8.04113
s.e. B	7.65389	28.88721	22.63049	22.63049
K	0.14942	0.300016	0.24907	0.24907
s.e. K	0.13208	0.91513	0.50545	0.50545
V	1.01729	0.84975	0.96378	0.96378
s.e. V	2.16657	3.07072	4.37240	4.37240
F_{req}	79.3552 ***	6.3932 NS	7.2955 NS	7.2955 NS
R^2	0.9761	0.9147	0.9349	0.9349

significance level

*** $P < 0.001$

NS not significant (5% level)

Figure A 6.1(a). The Richards function fits for graincoat colour of the four replicates of cv. Gamenya.

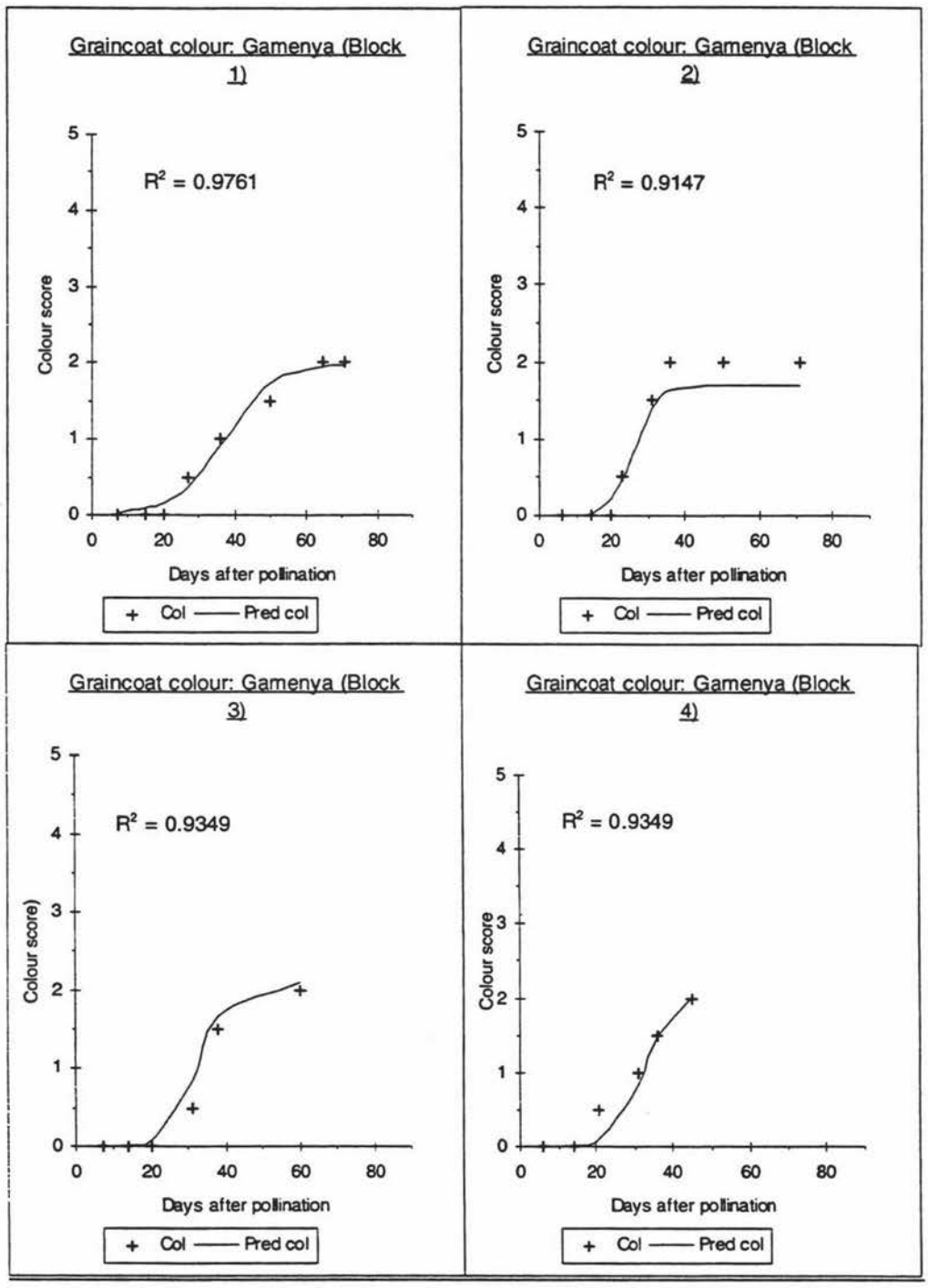


Table A 6.1 (b). The Richards function for Grain coat colour (statistics by replicate).

cv. Tordo

Replicate: Statistic	1	2	3	4
A	1.41595	2.79675	2.40000	2.39936
s.e. A	0.35963	0.63355	0.23130	0.53758
B	4.39237	4.08606	4.60017	8.01567
s.e. B	15.67052	10.58857	6.27925	36.65155
K	0.23150	0.14943	0.20000	0.29951
s.e. K	0.50401	0.24870	0.17970	1.09880
V	0.89951	0.89696	0.93016	0.93549
s.e. V	3.13265	2.10050	1.31741	4.88995
F_{reg}	19.4663 NS	28.8252 **	99.1192 *	12.5916 NS
R^2	0.9571	0.9260	0.9844	0.9193

significance level

* $P < 0.05$

** $P < 0.01$

NS not significant (5% level)

Figure A 6.1(b). The Richards function fits and data points for graincoat colour of the four replicates of cv. Tordo.

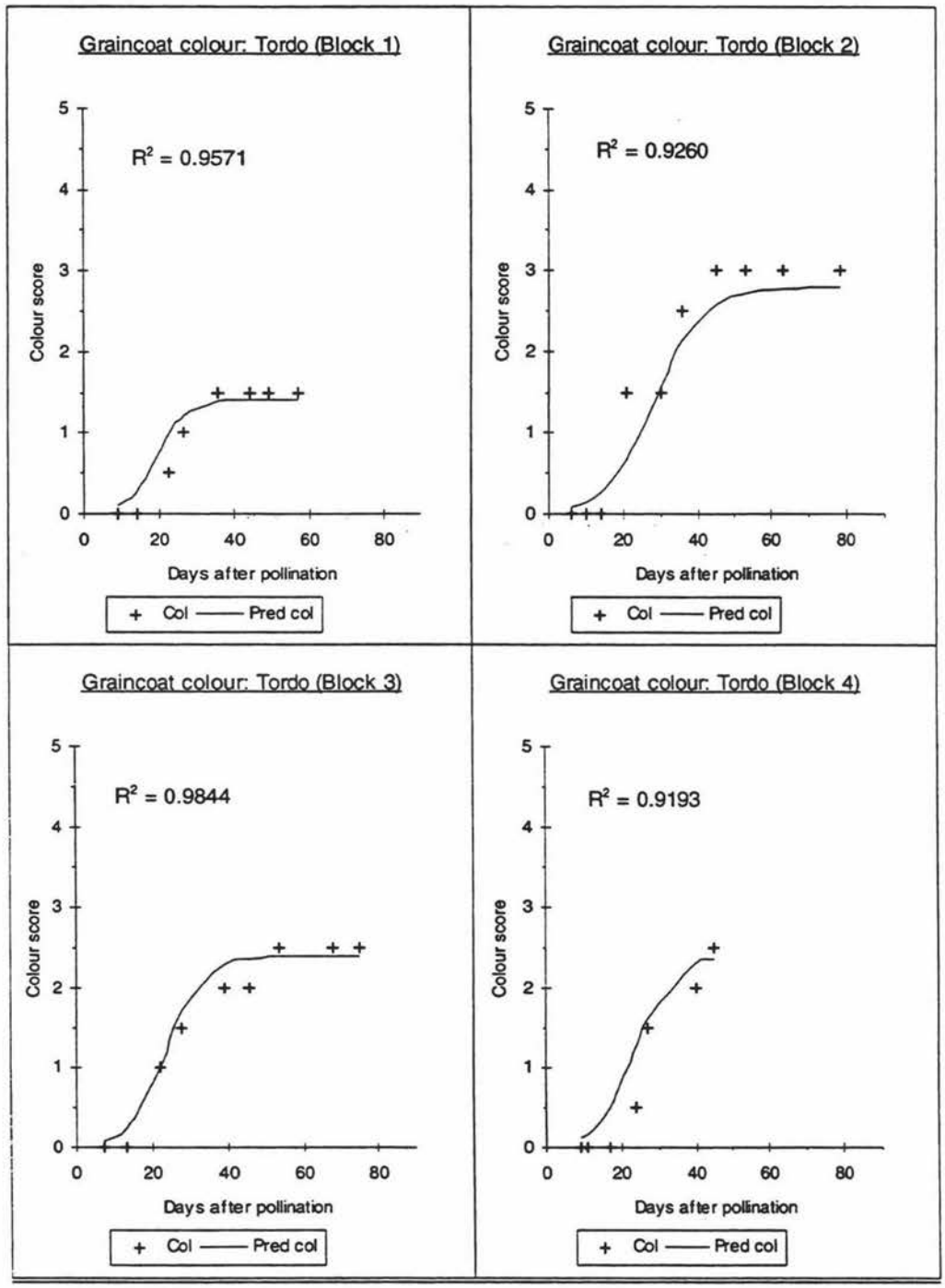


Table A 6.1 (c). The Richards function for Grain coat colour (statistics by replicate).

(c) Kenya 321

Replicate: Statistic	1	2	3	4
A	2.89997	2.30000	2.49978	2.09865
s.e. A	0.84063	0.13974	0.14794	0.06755
B	5.30023	6.95000	11.70300	7.00863
s.e. B	14.26804	7.39445	8.94232	2.88049
K	0.09995	0.20000	0.29994	0.19964
s.e. K	0.19131	0.16602	0.17031	0.06463
V	0.95306	0.80000	0.81835	0.70189
s.e. V	2.83579	0.64689	0.59611	0.15327
F_{req}	28.4995 *	128.7172 ***	56.8291 ***	670.6712 ***
R^2	0.9318	0.9836	0.9990	0.9962

significance level
* P < 0.05
*** P < 0.001

Figure A 6.1(c). The Richards function fits and data points for graincoat colour of the four replicates of cv. Kenya 321.

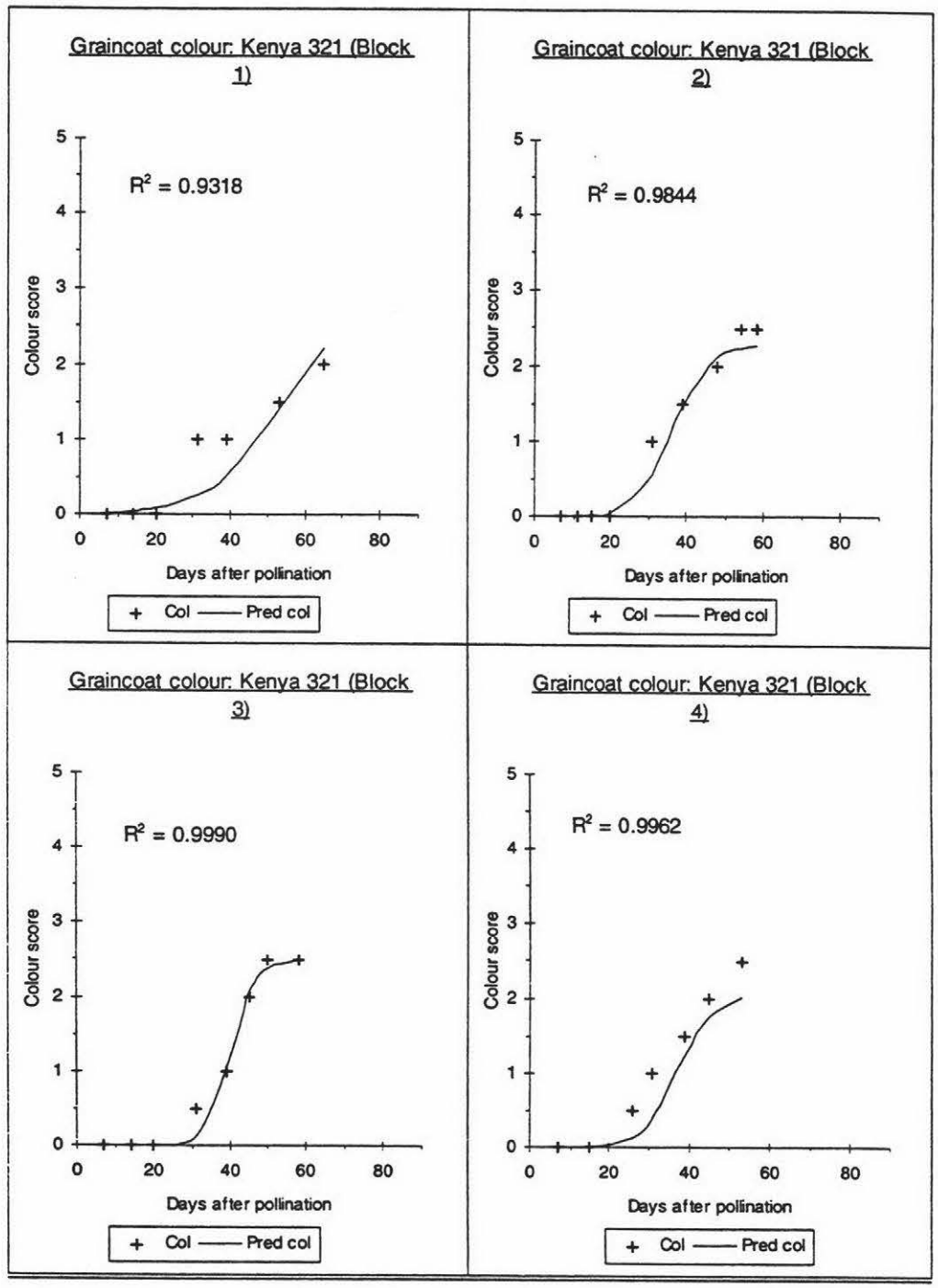


Table A 6.1 (d). The Richards function for Grain coat colour (statistics by replicate).

cv. Brevor

Replicate: Statistic	1	2	3	4
A	2.40236	2.00029	2.62676	1.80043
s.e. A	0.49511	0.71887	0.34233	0.15805
B	3.89807	4.49673	5.93830	4.90752
s.e. B	4.30939	5.53847	8.51128	5.83460
K	0.09862	0.10005	0.14183	0.14948
s.e. K	0.08832	0.11978	0.14420	0.13413
V	0.74792	0.59936	0.92849	0.65076
s.e. V	0.37895	0.17358	1.54786	0.25496
F_{reg}	119.4488 ***	110.0544 ***	70.7858 *	78.9279 ***
R^2	0.9789	0.9845	0.9827	0.9784

significance level
* P < 0.05
*** P < 0.001

Figure A 6.1(d). The Richards function fits and data points for graincoat colour of the four replicates of cv. Brevor.

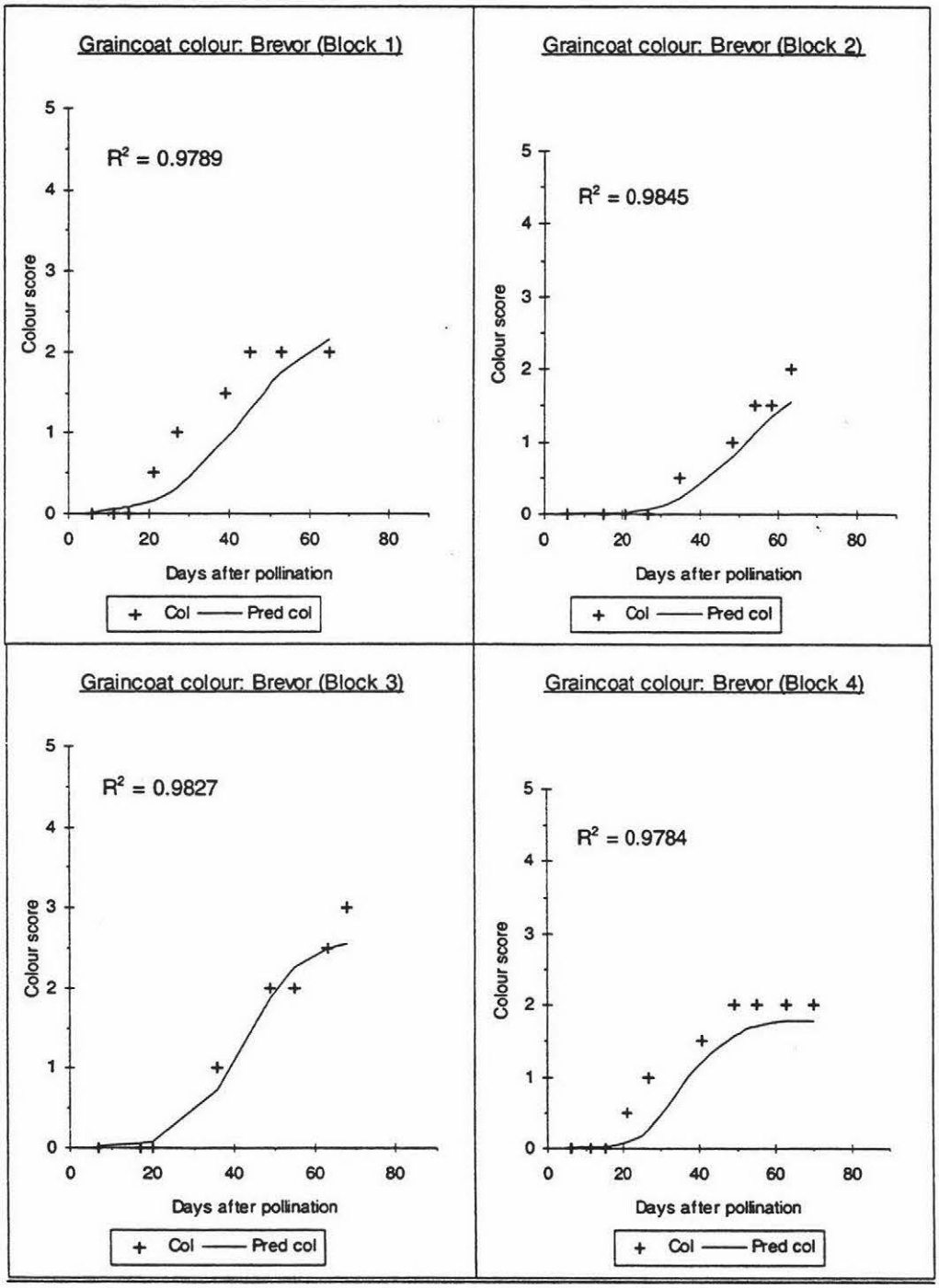


Table A 6.1 (e). The Richards function for Grain coat colour (statistics by replicate).

cv. Isis

Replicate: Statistic	1	2	3	4
A	1.80000	2.70069	2.50052	1.59995
s.e. A	0.29445	0.88324	0.23331	0.14779
B	7.99991	3.79603	10.00819	7.24876
s.e. B	12.18610	6.22239	11.67812	7.09426
K	0.25001	0.10013	0.29980	0.20004
s.e. K	0.32118	0.12890	0.29282	0.16516
V	0.84962	0.84732	0.84169	0.59979
s.e. V	1.10066	0.91671	0.96557	0.16566
F_{req}	62.5262 *	103.1216 ***	65.2234 *	222.8122 ***
R^2	0.9788	0.9823	0.9795	0.9949

significance level
* P < 0.05
*** P < 0.001

Figure A 6.1(e). The Richards function fits and data points for graincoat colour of the four replicates of cv. Isis.

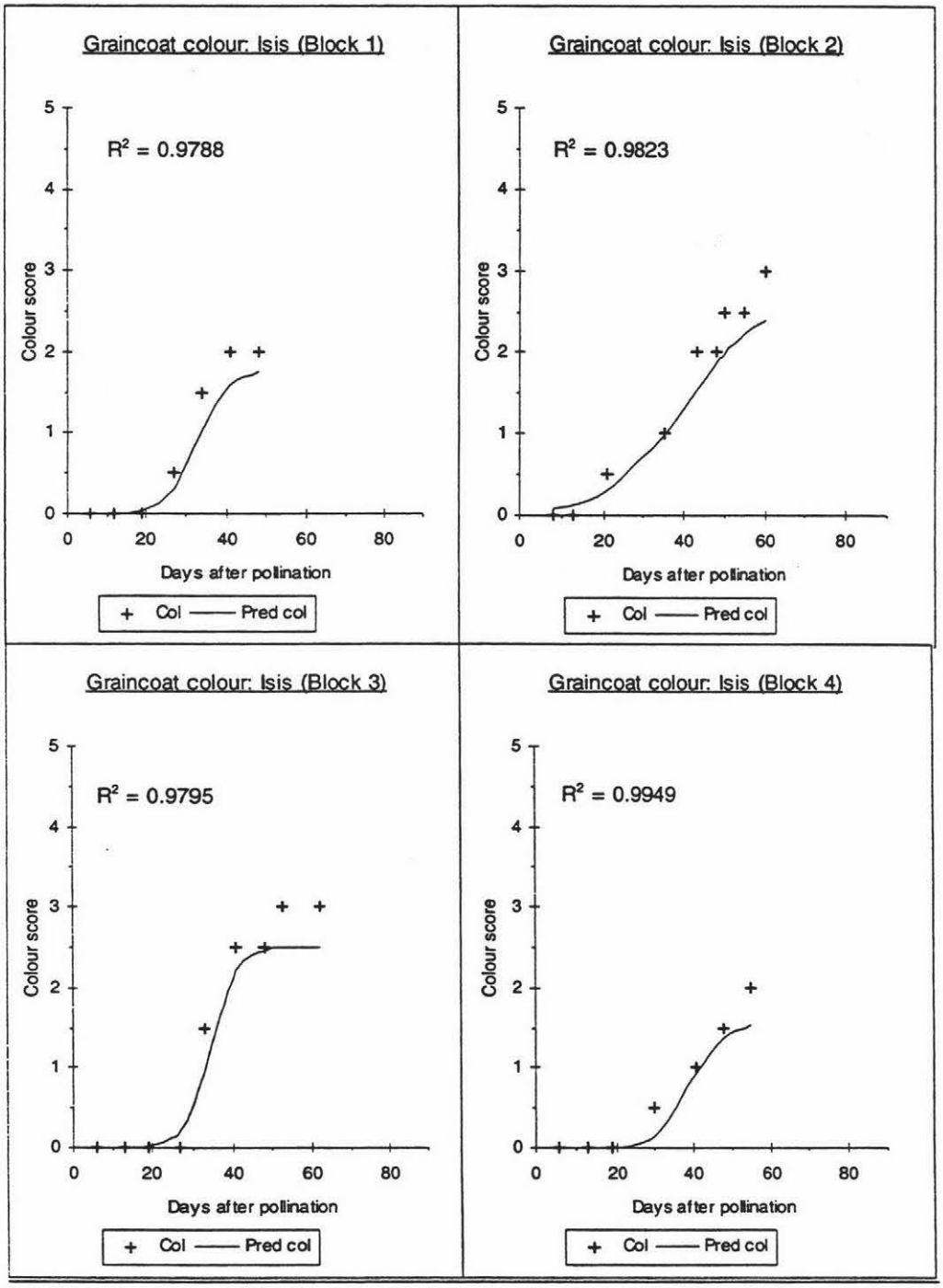


Table A 6.1 (f). The Richards function for Grain coat colour (statistics by replicate).

cv. Sonora 64 A

Replicate: Statistic	1	2	3	4
A	4.80000	4.70396	4.30009	5.00910
s.e. A	0.18011	0.307238	0.43946	0.82805
B	9.95000	9.59528	3.99930	4.60510
s.e. B	5.39642	14.59964	5.92714	7.53524
K	0.35000	0.31014	0.14996	0.15117
s.e. K	0.14835	0.33558	0.14149	0.15703
V	0.95000	0.94664	0.85097	0.94951
s.e. V	0.78035	2.29772	0.96347	1.81862
F_{req}	373.9671 ***	80.5971 *	95.4025 ***	47.2096 ***
R^2	0.9966	0.9851	0.9752	0.9588

significance level
* P < 0.05
*** P < 0.001

Figure A 6.1(f). The Richards function fits and data points for graincoat colour of the four replicates of cv. Sonora 64 A.

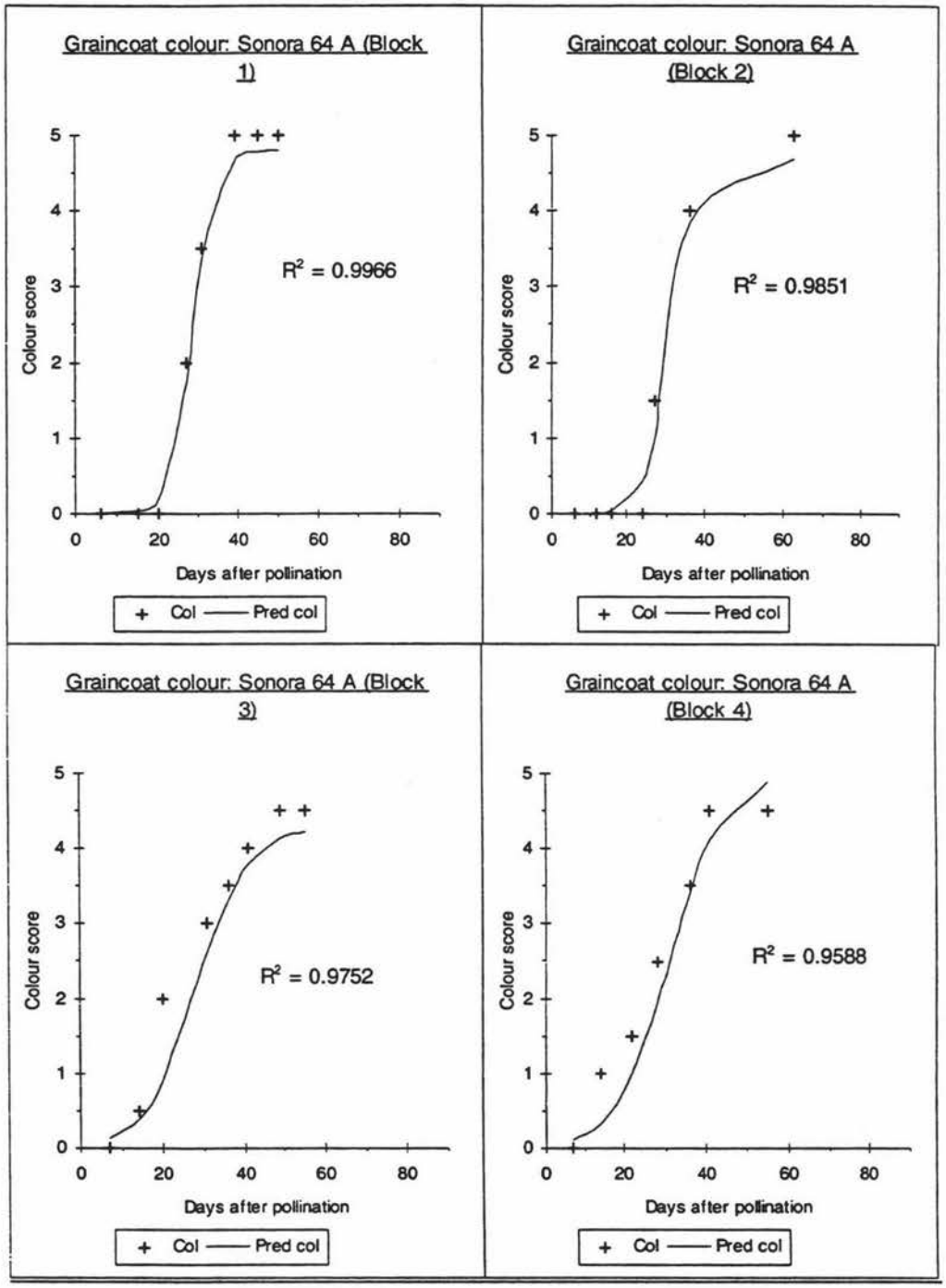


Table 6.1 (g). The Richards function for Grain coat colour (statistics by replicate).

cv. Thatcher

Replicate: Statistic	1	2	3	4
A	5.09997	3.00003	4.09924	4.69993
s.e. A	0.34899	0.06620	0.16344	0.41805
B	6.84997	7.65098	8.60271	10.75044
s.e. B	5.22457	9.56169	4.22819	10.43125
K	0.20000	0.24997	0.26005	0.30999
s.e. K	0.10753	0.23421	0.10871	0.25295
V	0.94993	0.60006	0.80084	0.90068
s.e. V	0.95001	0.11580	0.29651	1.08786
F_{reg}	305.6379 ***	1393.672 ***	509.9675 ***	115.9483 **
R^2	0.9950	0.9992	0.9983	0.9912

significance level
** P < 0.01
*** P < 0.001

Figure A 6.1(g). The Richards function fits and data points for graincoat colour of the four replicates of cv. Thatcher.

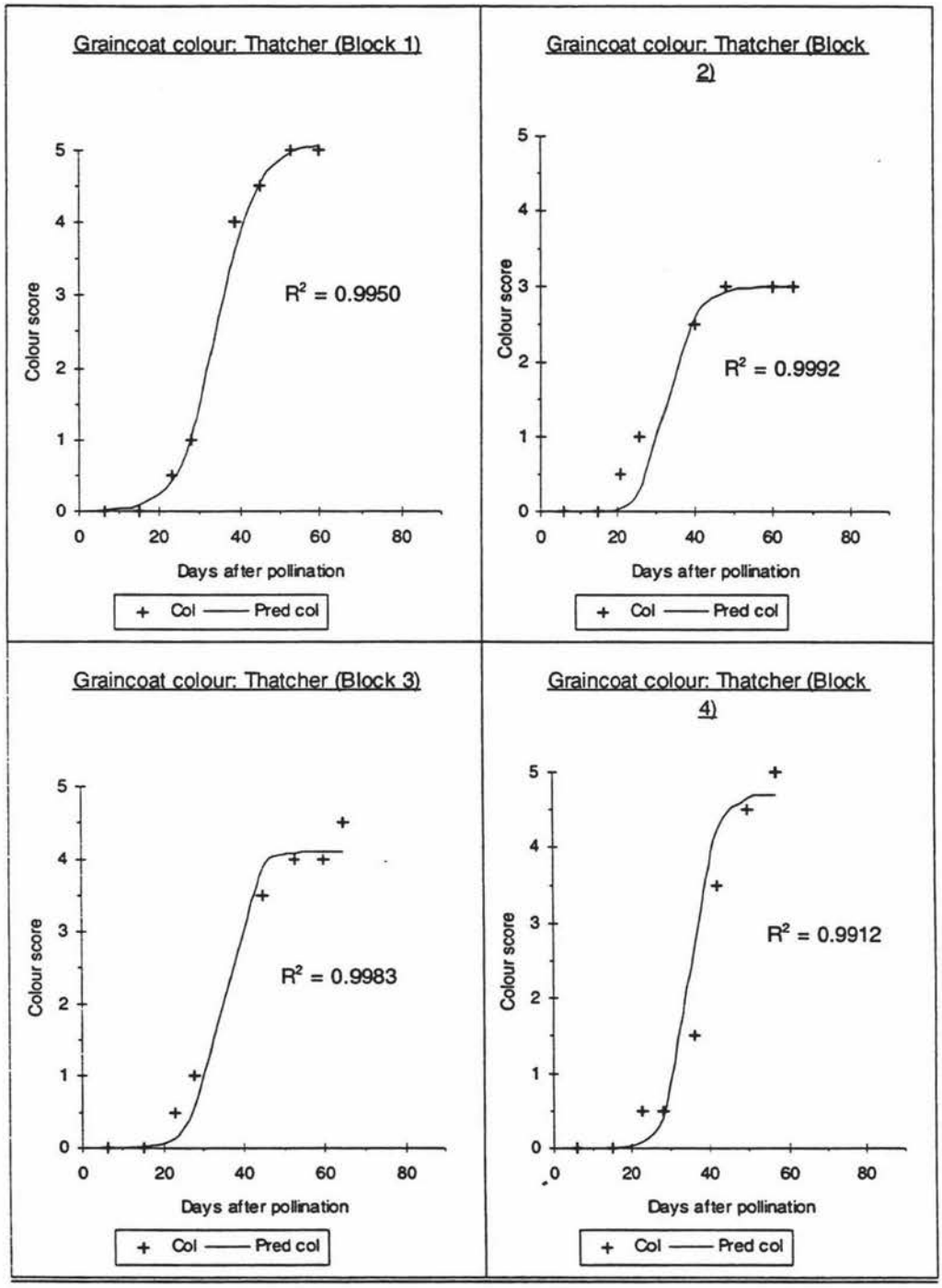


Table A 6.1 (h). The Richards function for Grain coat colour (statistics by replicate).

cv. La Prevision

Replicate: Statistic	1	2	3	4
A	3.39968	4.29996	3.90117	4.49580
s.e. A	0.11643	0.62378	0.42666	0.63978
B	11.29637	7.39922	8.04808	8.05049
s.e. B	6.03362	10.06167	16.73325	20.06519
K	0.31007	0.20005	0.31018	0.24877
s.e. K	0.14108	0.21684	0.48780	0.46213
V	0.69949	0.79910	0.99673	0.90985
s.e. V	0.23707	0.88455	3.32676	3.07315
F_{req}	350.0158 ***	57.1286 ***	43.5310 *	20.1031 *
R^2	0.9924	0.9699	0.9712	0.9445

significance level
** $P < 0.05$
*** $P < 0.001$

Figure A 6.1(h). The Richards function fits and data points for graincoat colour of the four replicates of cv. La Prevision.

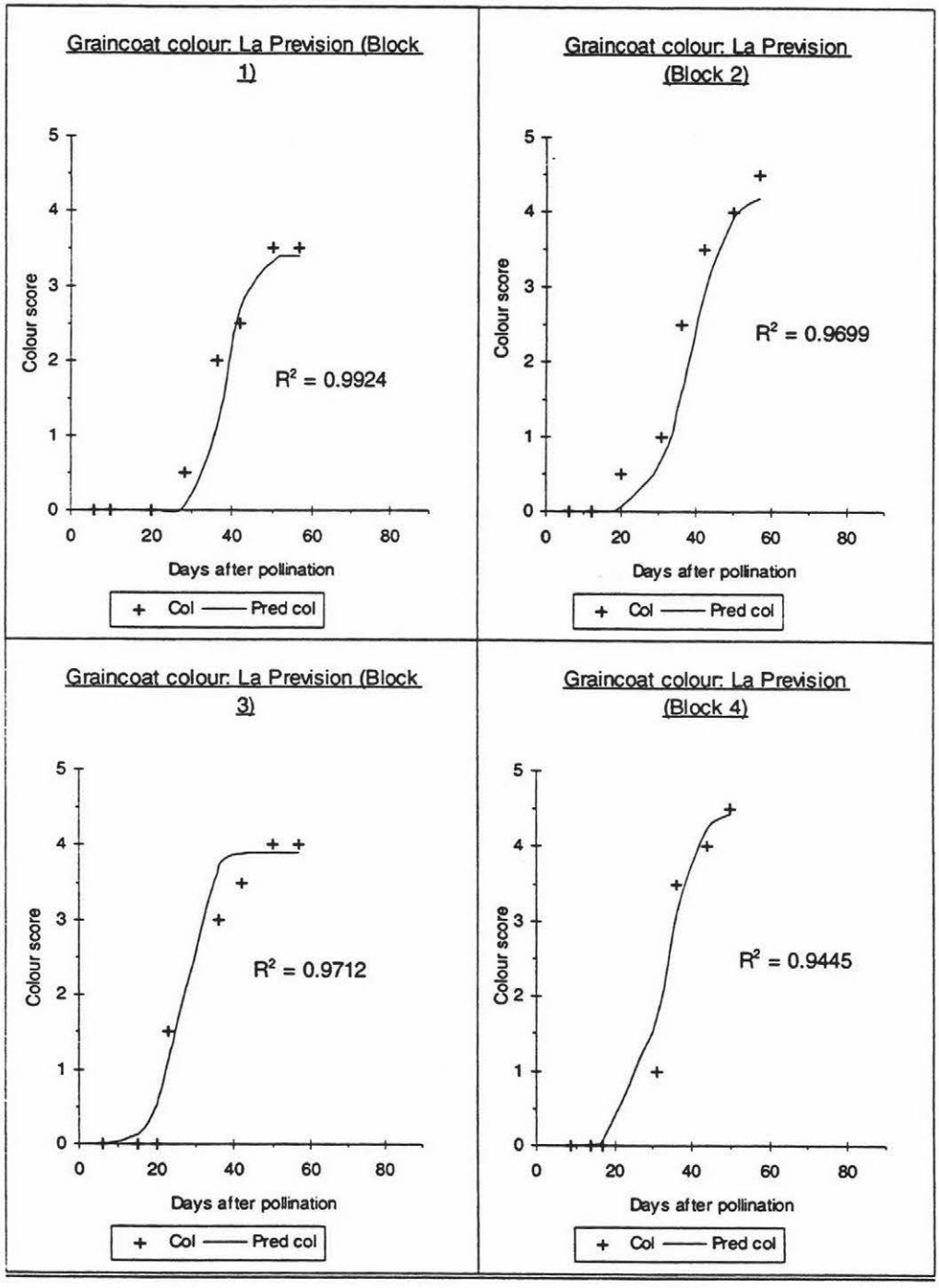


Table A 6.1 (i). The Richards function for Grain coat colour (statistics by replicate).

cv. Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
A	5.10000	5.30013	5.0999	4.79998
s.e. A	0.20349	0.51097	0.32010	0.26327
B	8.09998	7.70021	8.65012	7.79985
s.e. B	6.82361	7.84179	6.93210	33.77397
K	0.25000	0.20002	0.20000	0.25001
s.e. K	0.14387	0.14554	0.13444	0.78066
V	1.00007	0.99839	0.80994	0.94972
s.e. V	1.46971	1.67998	1.56723	4.56442
F_{reg}	421.5993 ***	157.9122 ***	165.3122 ***	155.7772 NS
R^2	0.9973	0.9887	0.9888	0.9961

significance level

*** $P < 0.001$

NS not significant (5% level)

Figure A 6.1(i). The Richards function fits and data points for graincoat colour of the four replicates of cv. Hilgendorf.

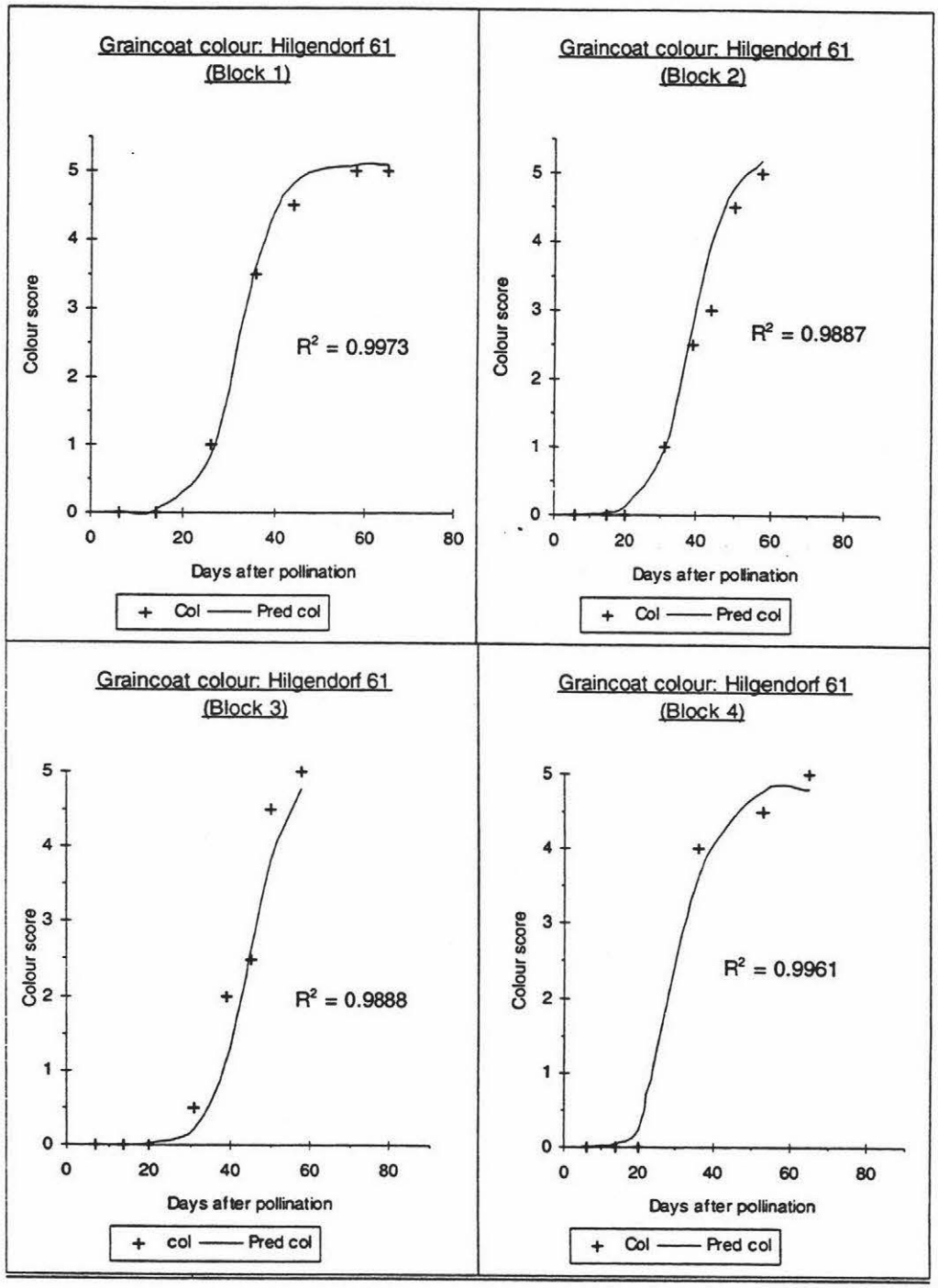


Table A 6.1 (j). The Richards function for Grain coat colour (statistics by replicate).

cv. Karamu

Replicate: Statistic	1	2	3	4
A	4.69855	3.50003	4.73999	4.30305
s.e. A	1.09622	0.44987	0.82907	0.37584
B	4.64774	3.54792	4.69875	5.20365
s.e. B	7.34125	3.27813	10.80477	7.76758
K	0.15016	0.10008	0.15192	0.19829
s.e. K	0.17789	0.07155	0.24042	0.20659
V	0.84912	0.64941	0.91291	0.84760
s.e. V	1.02047	0.19178	1.97265	0.99790
F_{req}	58.3476 ***	153.0698 ***	43.0830 ***	94.7366 *
R^2	0.9694	0.9744	0.9566	0.9874

significance level
** P < 0.05
*** P < 0.001

Figure A 6.1(j). The Richards function fits and data points for graincoat colour of the four replicates of cv. Karamu.

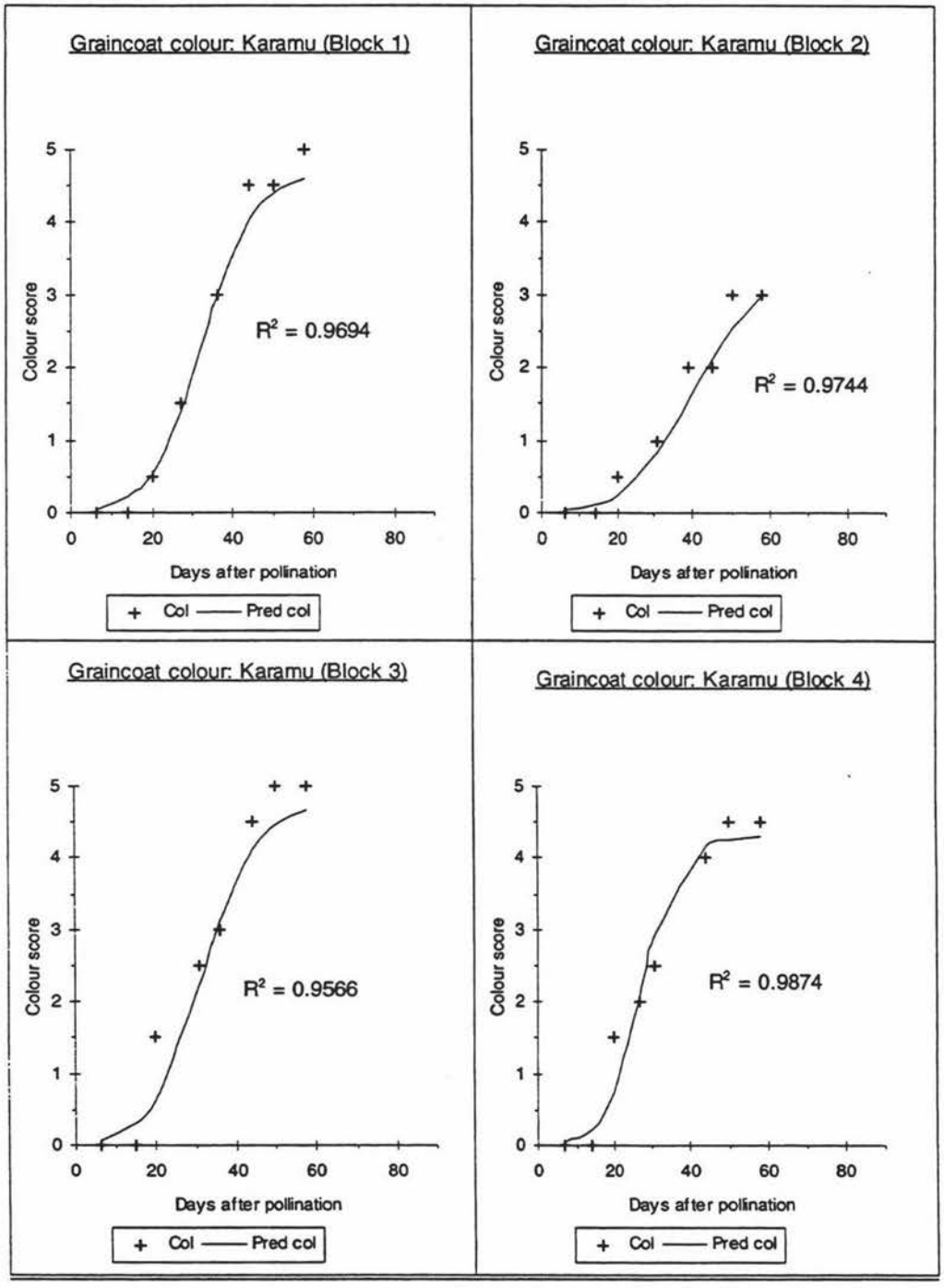


Table A 6.2 (a). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Gamenya

Replicate: Statistic	1	2	3	4
β_o	6.45853	5.66714	5.94413	6.44959
s.e. β_o	0.28938	0.26855	0.22854	0.12854
β_1	-0.14048	-0.11733	-0.13487	-0.16000
s.e. β_1	0.01929	0.01765	0.01521	0.01173
β_2	0.00132	0.00115	0.00133	0.00174
s.e. β_2	0.00026	0.00024	0.00020	0.00023
F_{Reg}	65.36 **	44.42 **	80.50 **	426.22 **
R^2	0.9703	0.9467	97.58	0.9965

** $P < 0.01$

Figure A 6.2 (a). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Gamenya.

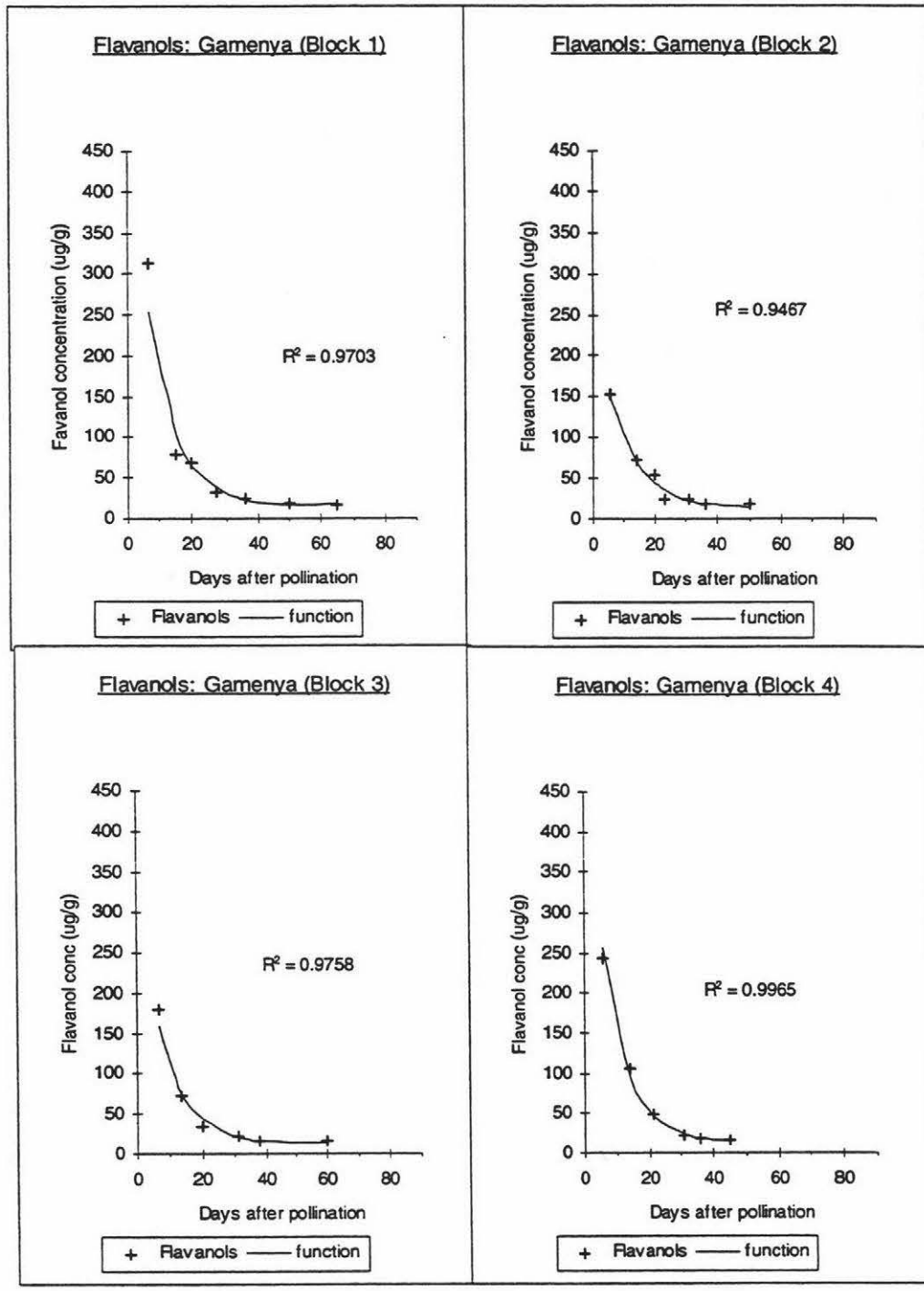


Table A 6.21 (b). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Tordo

Replicate: Statistic	1	2	3	4
β_o	6.29020	5.57840	5.83491	6.33457
s.e. β_o	0.41817	0.38586	0.41882	0.52810
β_1	-0.13777	-0.08131	-0.11461	-0.15870
s.e. β_1	0.02979	0.02116	0.02687	0.04653
β_2	0.00135	0.00057	0.00105	0.00187
s.e. β_2	0.00044	0.00024	0.00036	0.00085
F_{Reg}	37.25 **	25.88 **	22.05 **	25.64 **
R^2	0.9490	0.8961	0.8982	0.9277

** $P < 0.01$

Figure A 6.2 (b). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Tordo.

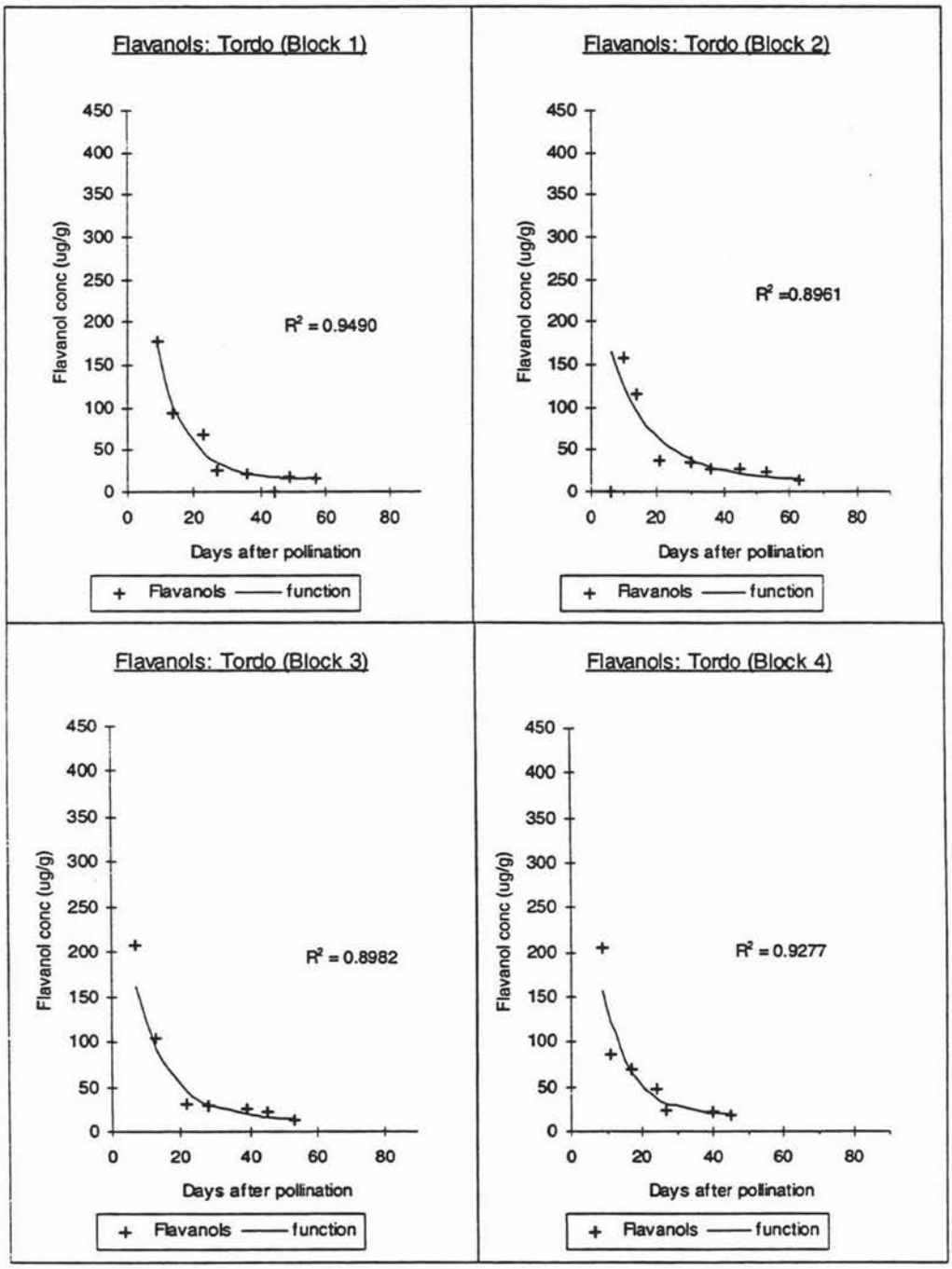


Table A 6.21 (c). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Kenya 321

Replicate: Statistic	1	2	3	4
β_o	6.28786	6.44396	5.84556	5.68412
s.e. β_o	0.26983	0.28833	0.16308	0.24695
β_1	-0.09870	-0.12544	-0.11375	-0.08219
s.e. β_1	0.01819	0.01894	0.00974	0.01481
β_2	0.00073	0.00108	0.00100	0.00059
s.e. β_2	0.00025	0.00025	0.00012	0.00020
F_{Reg}	69.84 ***	75.92 ***	175.62 ***	63.38 ***
R^2	0.9723	0.9499	0.9805	0.9621

*** $P < 0.001$

Figure A 6.2 (c). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Kenya 321.

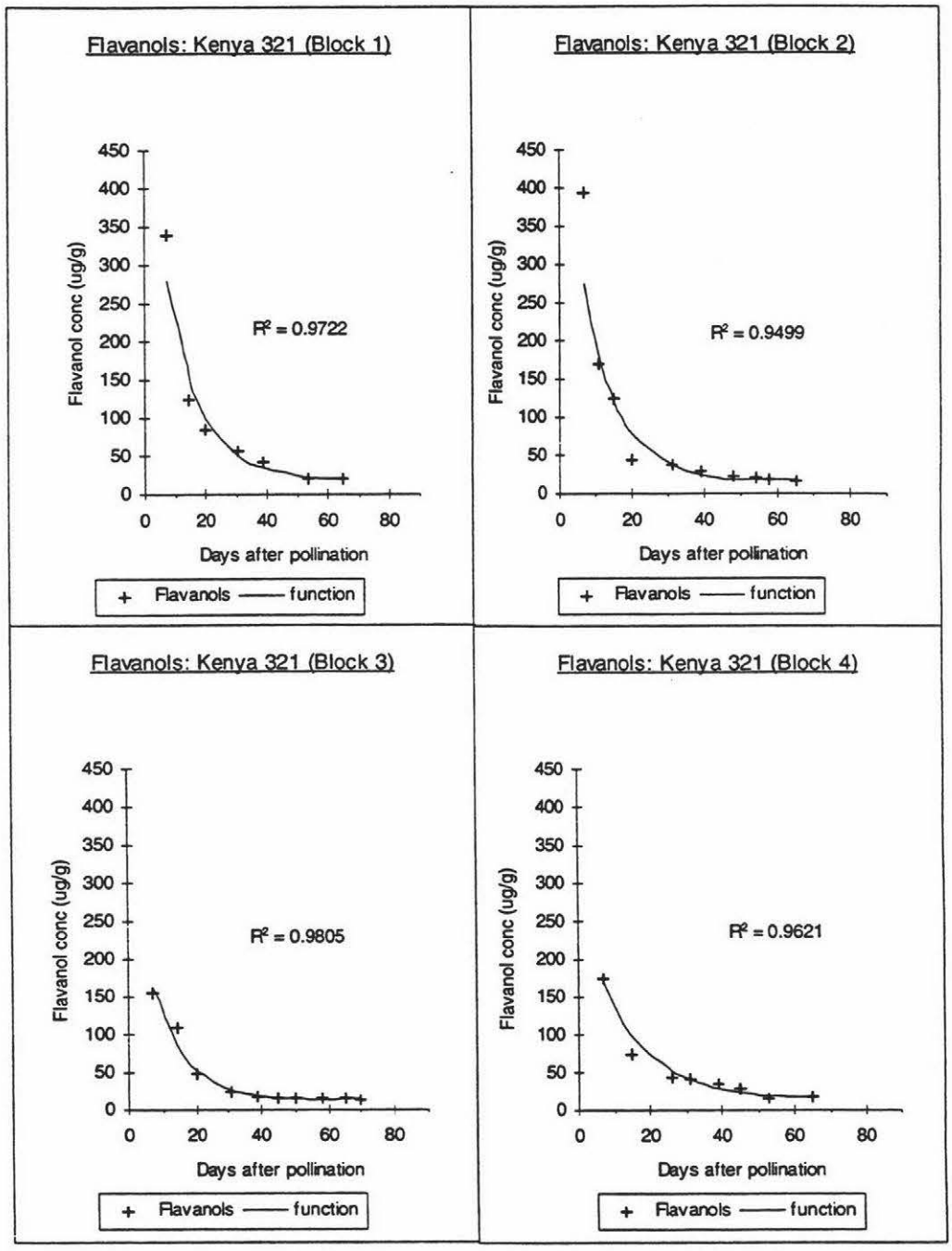


Table A 6.21 (d). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Brevor

Replicate: Statistic	1	2	3	4
β_o	6.76313	6.42182	5.96385	5.88664
s.e. β_o	0.33897	0.46293	0.40109	0.33384
β_1	-0.16309	-0.15203	-0.14302	-0.11137
s.e. β_1	0.02549	0.03055	0.02921	0.02232
β_2	0.00161	0.00149	0.00149	0.00095
s.e. β_2	0.00038	0.00041	0.00041	0.00029
F_{Reg}	67.81 ***	31.26 ***	31.09 **	41.72 ***
R^2	0.9576	0.9124	0.9396	0.9226

** $P < 0.01$
*** $P < 0.001$

Figure A 6.2 (d). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Brevor.

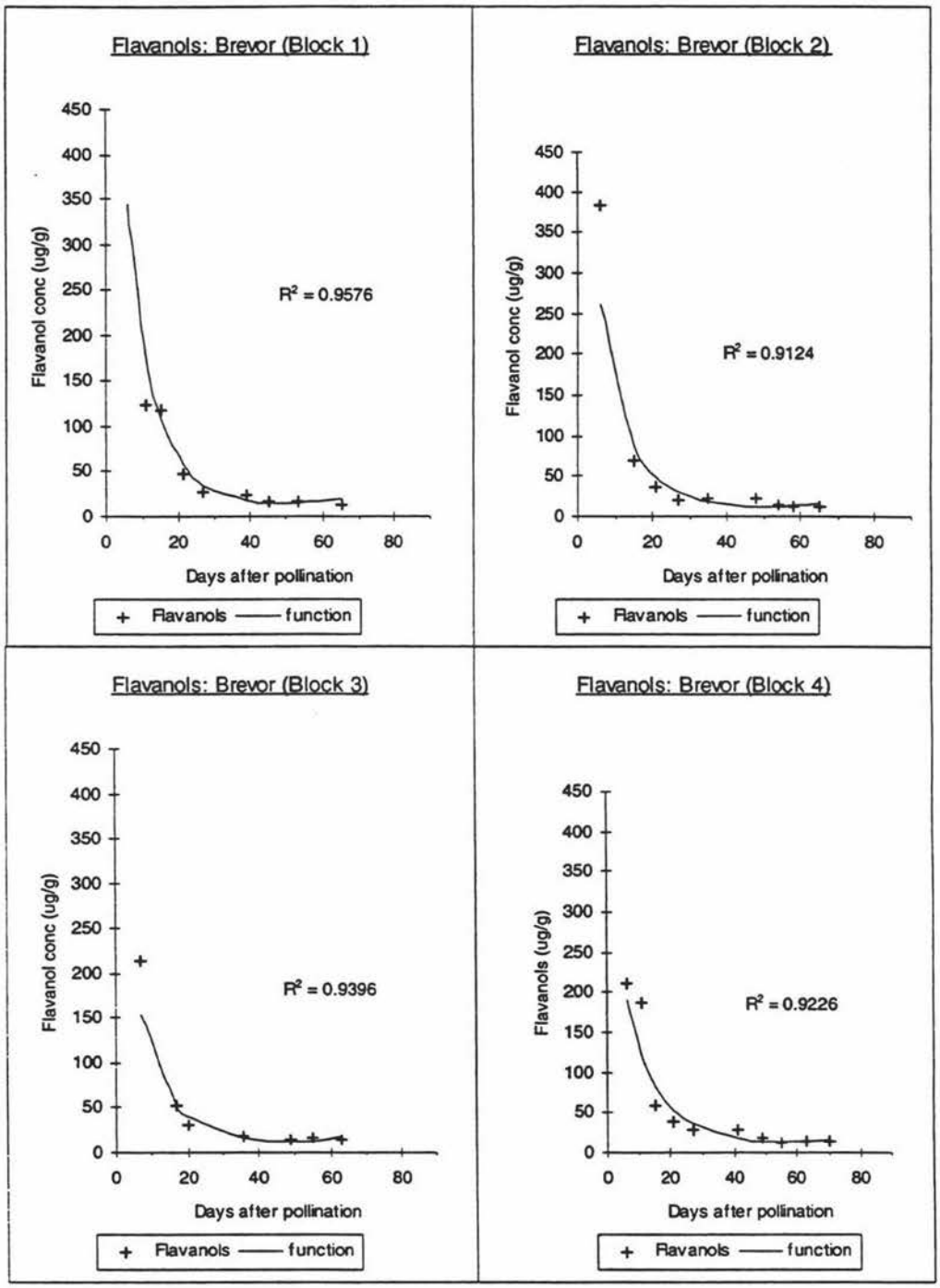


Table A 6.21 (e). Quadratic exponential function for flavanols data (statistics by replicate).

(e) Isis

Replicate: Statistic	1	2	3	4
β_o	5.62464	5.70449	6.15091	6.21964
s.e. β_o	0.43378	0.22176	0.20120	0.18997
β_1	-0.05690	-0.08499	-0.14260	-0.14327
s.e. β_1	0.04577	0.01591	0.01722	0.01683
β_2	-0.00037	0.00054	0.00150	0.00150
s.e. β_2	0.00096	0.00023	0.00031	0.00027
F_{Reg}	40.81 **	133.76 ***	144.63 ***	176.18 ****
R^2	0.9645	0.9781	0.9864	0.9916

** $P < 0.01$
*** $P < 0.001$

Figure A 6.2 (e). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Isis.

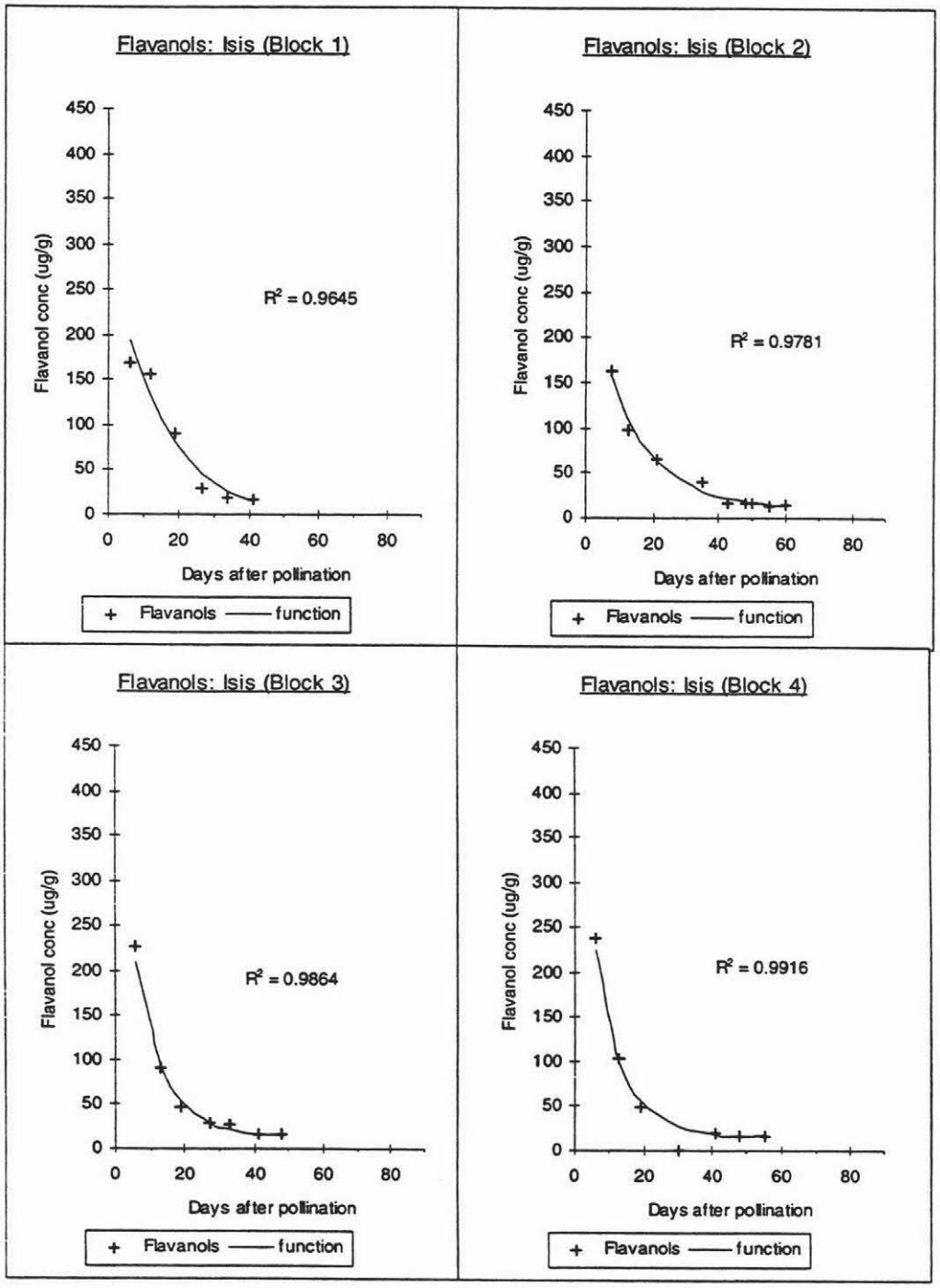


Table A 6.21 (f). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Sonora 64A

Replicate: Statistic	1	2	3	4
β_o	5.65133	5.91489	5.79617	6.07530
s.e. β_o	0.18552	0.33510	0.41720	0.30052
β_1	-0.10325	-0.10656	-0.09881	-0.11445
s.e. β_1	0.01031	0.02123	0.02836	0.01669
β_2	0.00088	0.00086	0.00087	0.00093
s.e. β_2	0.00012	0.00024	0.00040	0.00019
F_{Reg}	107.91 ***	38.47 ***	21.87 **	53.22 ***
R^2	0.9643	0.9277	0.8974	0.9466

** $P < 0.01$
*** $P < 0.001$

Figure A 6.2 (f). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Sonora 64 A.

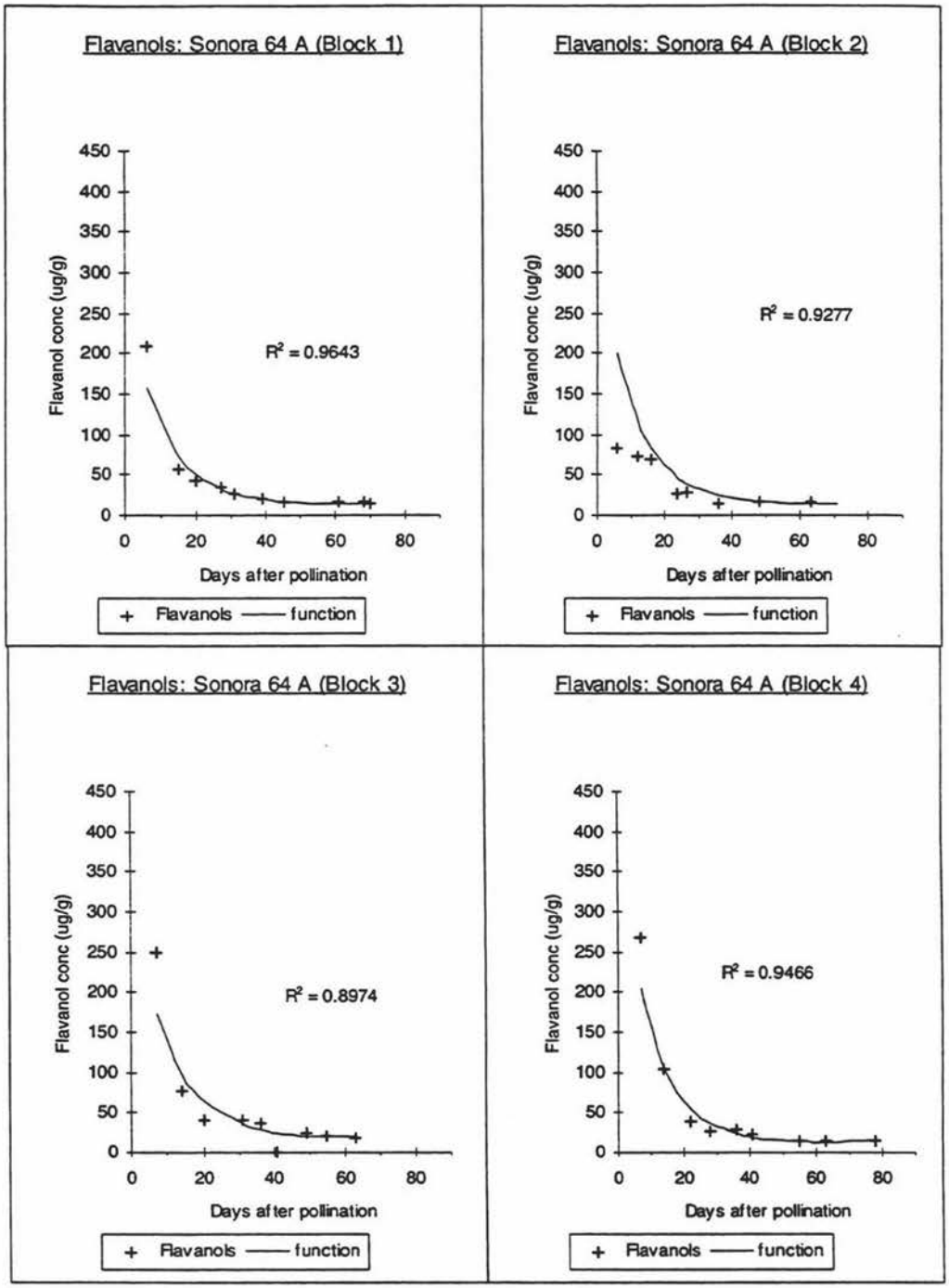


Table A 6.21 (g). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Thatcher

Replicate: Statistic	1	2	3	4
β_o	6.04245	6.63271	7.01643	5.94207
s.e. β_o	0.26035	0.31385	0.65624	0.20411
β_1	-0.13056	-0.15913	-0.18738	-0.11525
s.e. β_1	0.01492	0.02249	0.04431	0.01447
β_2	0.00124	0.00162	0.00199	0.00105
s.e. β_2	0.00018	0.00033	0.00060	0.00022
F_{Reg}	61.91 ***	63.54 ***	17.14 **	115.36 ***
R^2	0.9538	0.9695	0.8727	0.9788

** $P < 0.01$
*** $P < 0.001$

Figure A 6.2 (g). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Thatcher.

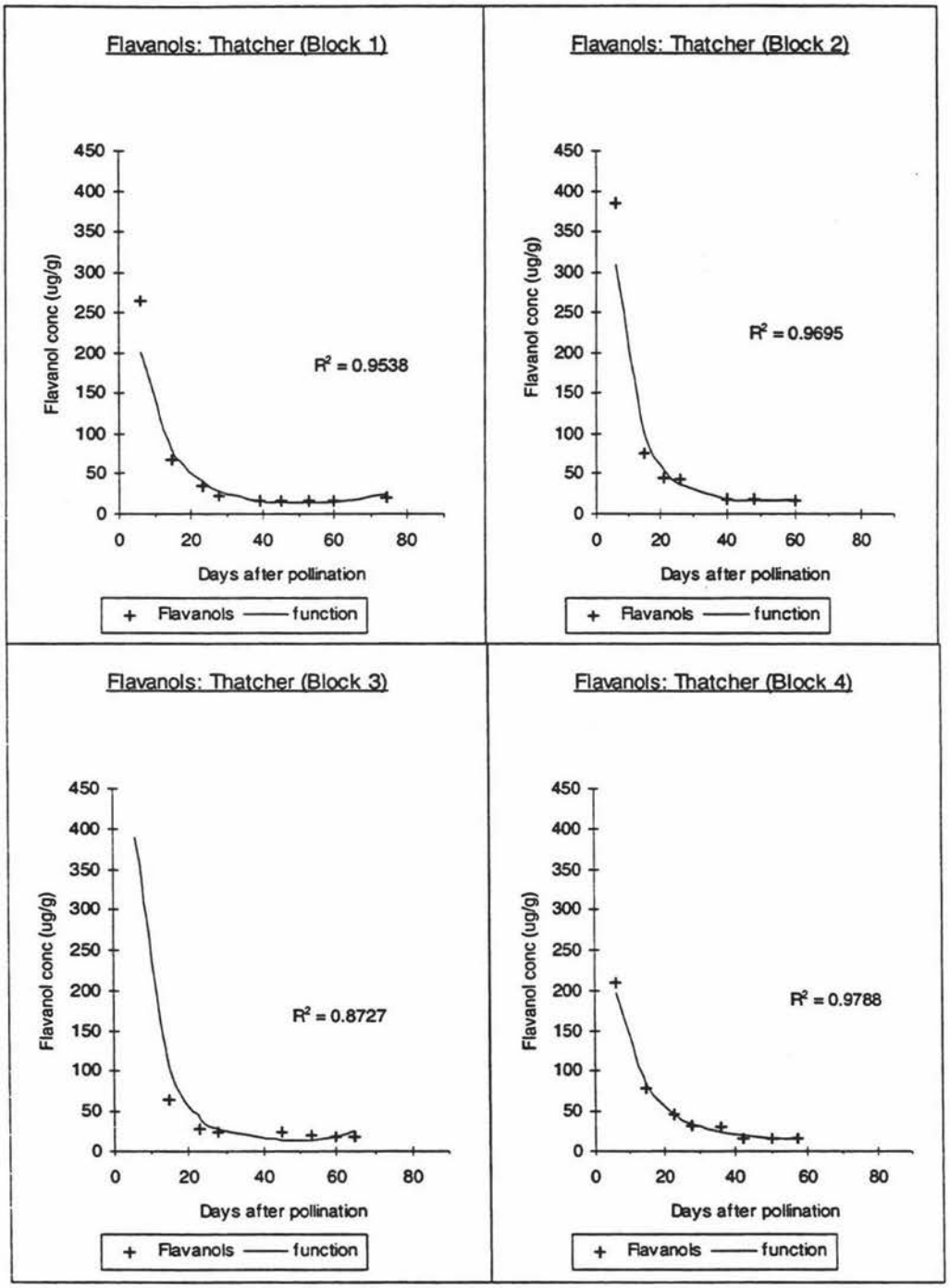


Table A 6.21 (h). Quadratic exponential function for flavanols data (statistics by replicate).

cv. La Prevision

Replicate: Statistic	1	2	3	4
β_o	6.55778	5.75918	5.34018	6.54869
s.e. β_o	0.40692	0.42987	0.24316	0.22928
β_1	-0.14891	-0.08934	-0.10598	-0.11935
s.e. β_1	0.02935	0.02178	0.01556	0.01657
β_2	0.00150	0.00057	0.00108	0.00086
s.e. β_2	0.00045	0.00023	0.00022	0.00025
F_{Reg}	38.50 ***	21.63 **	88.35 ***	192.13 ***
R^2	0.9390	0.8782	0.9779	0.9872

** $P < 0.01$
*** $P < 0.001$

Figure A 6.2 (h). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. La Prevision.

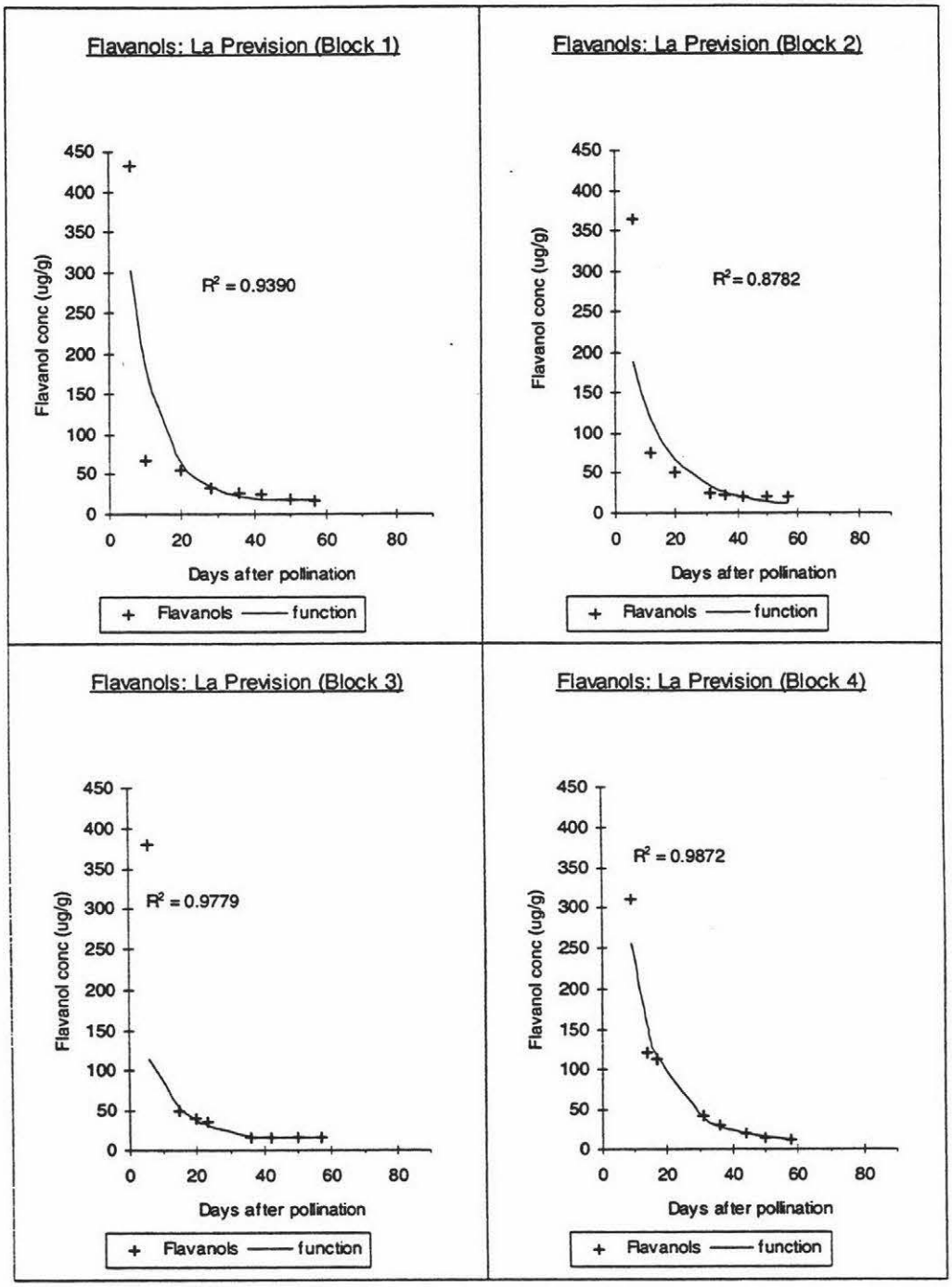


Table A 6.21 (i). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Hilgendorf 61

Replicate: Statistic	1	2	3	4
β_o	6.58518	5.85253	5.93330	6.46175
s.e. β_o	0.17266	0.27950	0.24923	0.62249
β_1	-0.13350	-0.10757	-0.10542	-0.12324
s.e. β_1	0.01100	0.01800	0.01614	0.04539
β_2	0.00121	0.00094	0.00086	0.00108
s.e. β_2	0.00015	0.00025	0.00022	0.00062
F_{Reg}	194.57 ***	56.49 ***	83.14 ***	13.37 *
R^2	0.9898	0.9496	0.9652	0.8991

* $P < 0.05$
*** $P < 0.001$

Figure A 6.2 (i). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Hilgendorf 61.

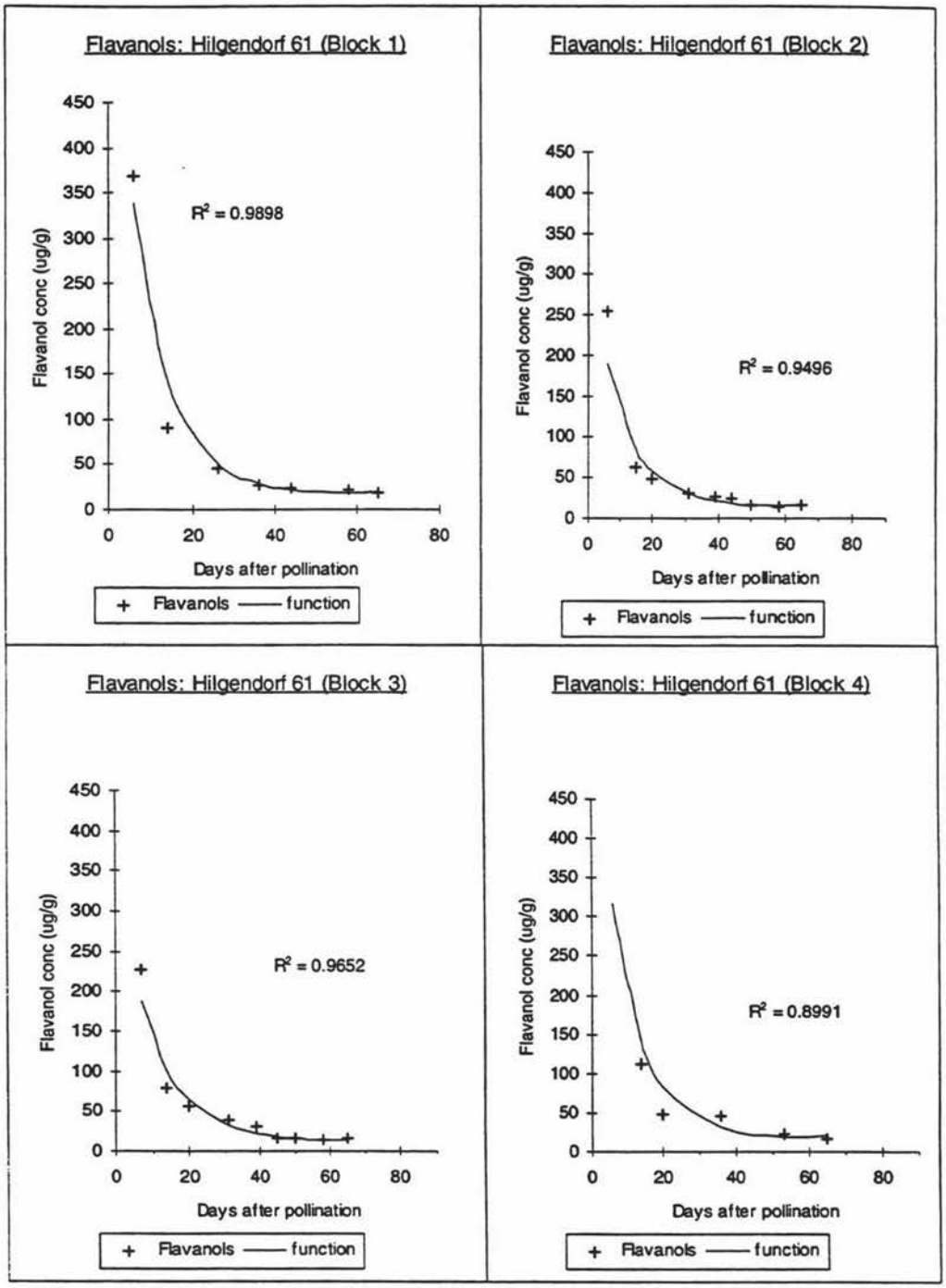


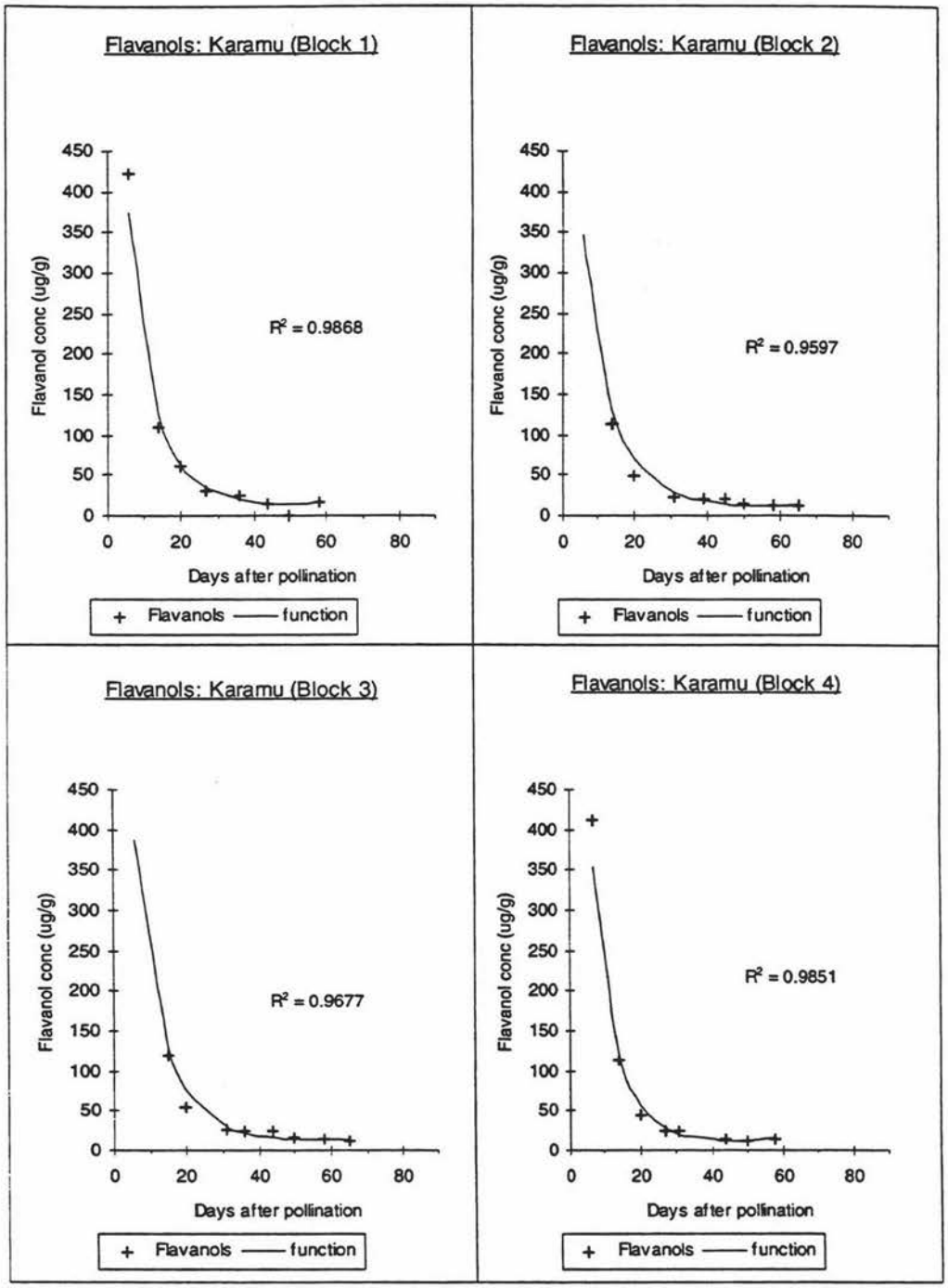
Table A 6.21 (j). Quadratic exponential function for flavanols data (statistics by replicate).

cv. Karamu

Replicate: Statistic	1	2	3	4
β_o	6.89695	6.68933	6.80805	7.17236
s.e. β_o	0.22002	0.31927	0.30074	0.24212
β_1	-0.17210	-0.14844	-0.14934	-0.20074
s.e. β_1	0.01599	0.01968	0.01929	0.01735
β_2	0.00175	0.00132	0.00132	0.00216
s.e. β_2	0.00024	0.00026	0.00026	0.00026
F_{Reg}	149.53 ***	71.43 ***	89.98 ***	165.72 ***
R^2	0.9868	0.9597	0.9677	0.9851

** $P < 0.01$
*** $P < 0.001$

Figure A 6.2 (j). The Quadratic exponential function fits and data points for Flavanol concentration of the four replicates of cv. Karamu.



Appendix 5.

Table A 7.1 (a). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Gamenya

Replicate: Statistic	1	2	3	4
β_o	5.19324	4.64436	3.33777	5.53921
s.e. β_o	0.80952	1.46340	0.38533	0.69906
β_1	-0.26216	-0.21246	-0.00540	-0.25078
s.e. β_1	0.06043	0.17435	0.04016	0.06452
β_2	0.00296	0.00187	-0.00260	0.00298
s.e. β_2	0.00078	0.00463	0.00087	0.00122
F_{reg}	12.43 ns	7.09 ns	142.67**	25.41*
R^2	0.9255	0.8764	0.9930	0.9621

significance level

* $P < 0.05$

** $P < 0.01$

n.s. not significantly different (5% level)

Figure A 7.1(a). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Gamenya.

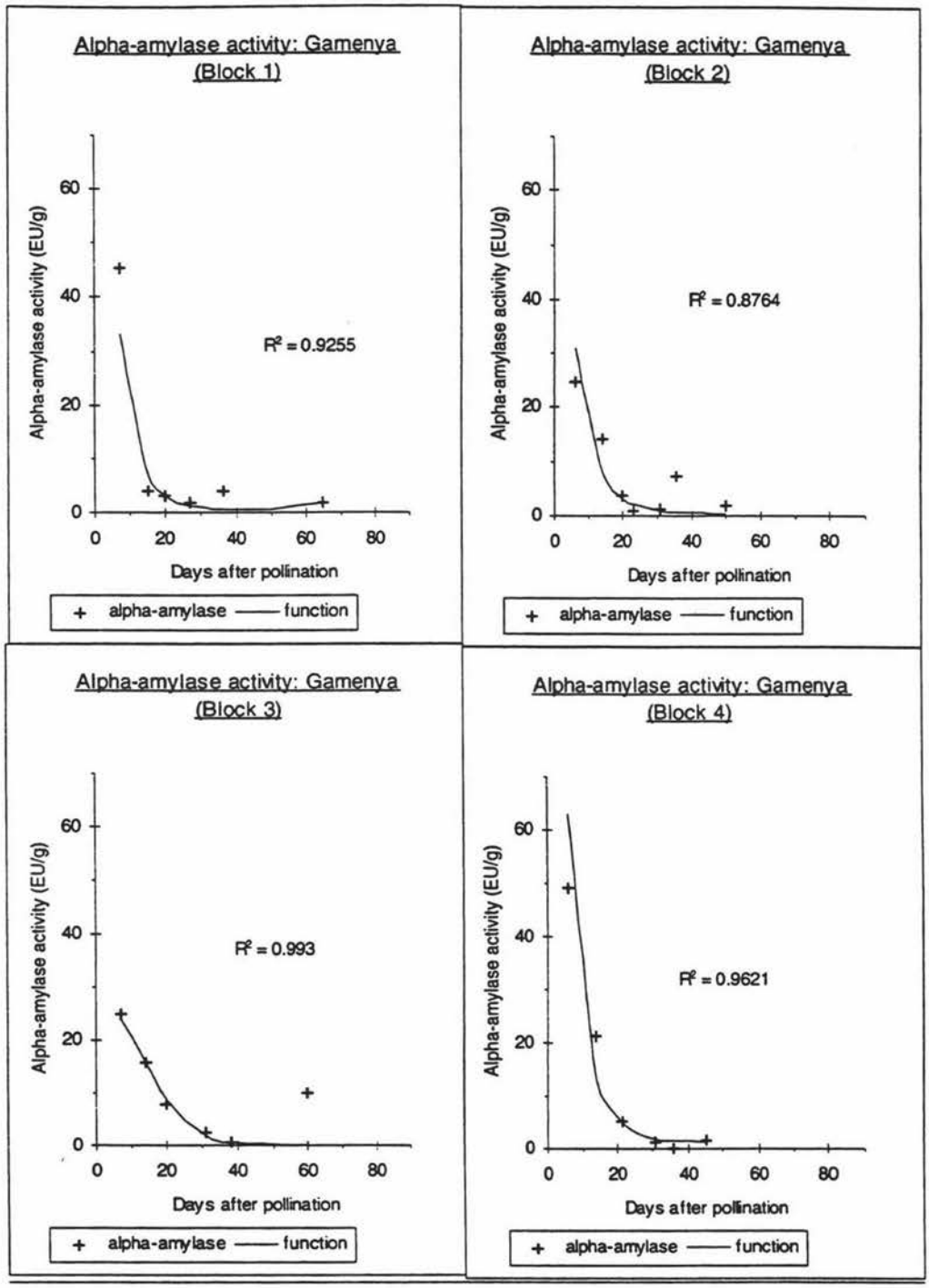


Table 7.1 (b). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Tordo

Replicate:	1	2	3	4
Statistic				
β_o	4.99203	4.63380	5.01045	5.49387
s.e. β_o	1.59509	0.44855	1.00344	0.80014
β_1	-0.23861	-0.16835	-0.22405	-0.25320
s.e. β_1	0.11509	0.03292	0.09889	0.06550
β_2	0.00300	0.00151	0.00247	0.00302
s.e. β_2	0.00180	0.00047	0.00189	0.00116
F_{reg}	5.12 ns	41.53*	16.08 ns	22.68*
R^2	0.9110	0.9765	0.9415	0.9578

significance level
* $P < 0.05$
n.s. not significantly different (5% level)

Figure A 7.1(b). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Tordo.

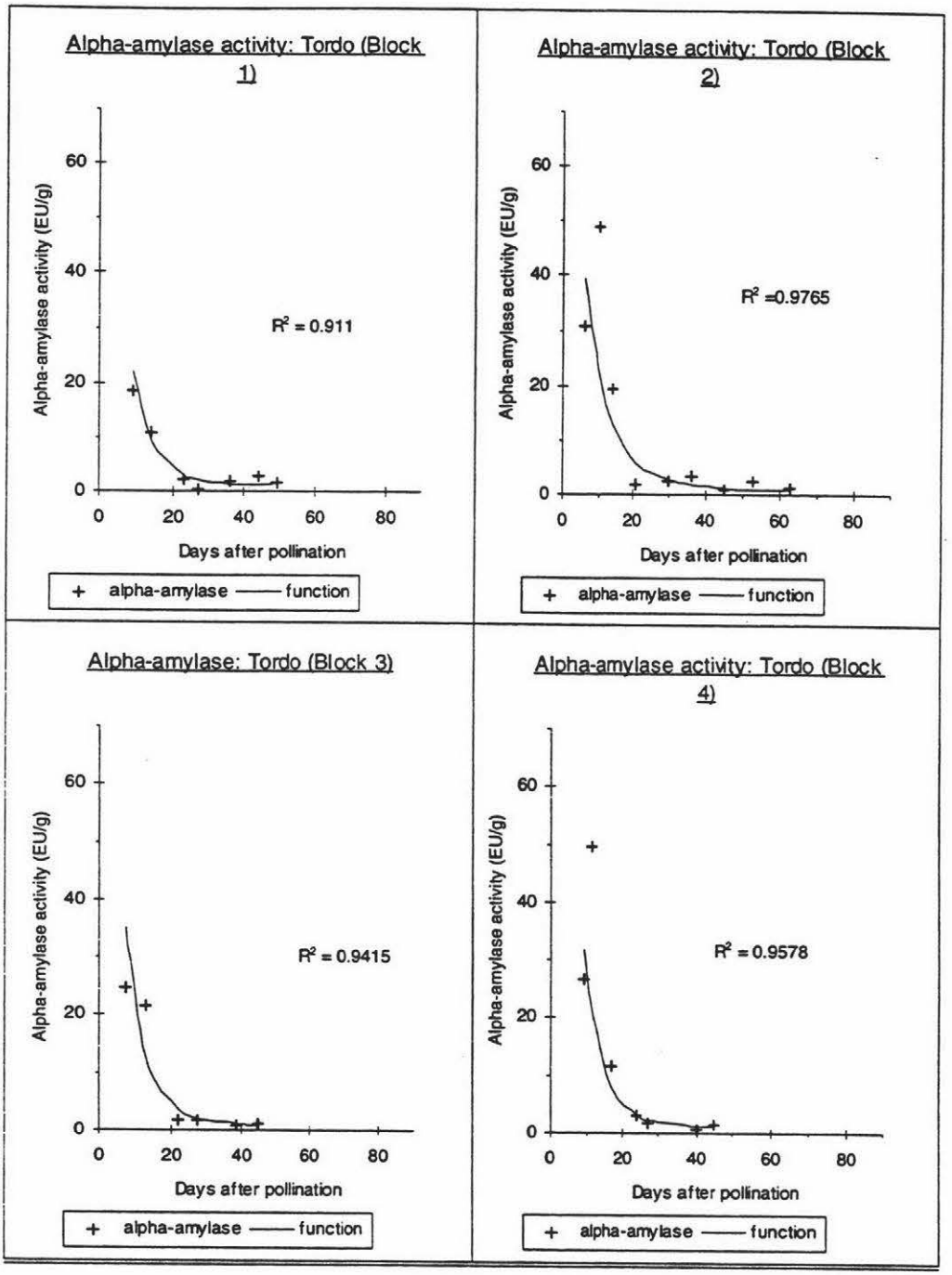


Table 7.1 (c). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Kenya 321

Replicate:	1	2	3	4
Statistic				
β_o	4.65272	7.06631	5.62933	6.28992
s.e. β_o	0.24876	1.37930	0.41437	0.48245
β_1	-0.14308	-0.47872	-0.18491	-0.29989
s.e. β_1	0.01803	0.16523	0.02825	0.04265
β_2	0.00115	0.00806	0.00151	0.00372
s.e. β_2	0.00024	0.00420	0.00039	0.00080
F_{reg}	152.46***	16.61 ns	97.95***	72.10*
R^2	0.9903	0.9432	0.9751	0.9863

significance level
* $P < 0.05$
*** $P < 0.001$
n.s. not significantly different (5% level)

Figure A 7.1(c). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Kenya 321.

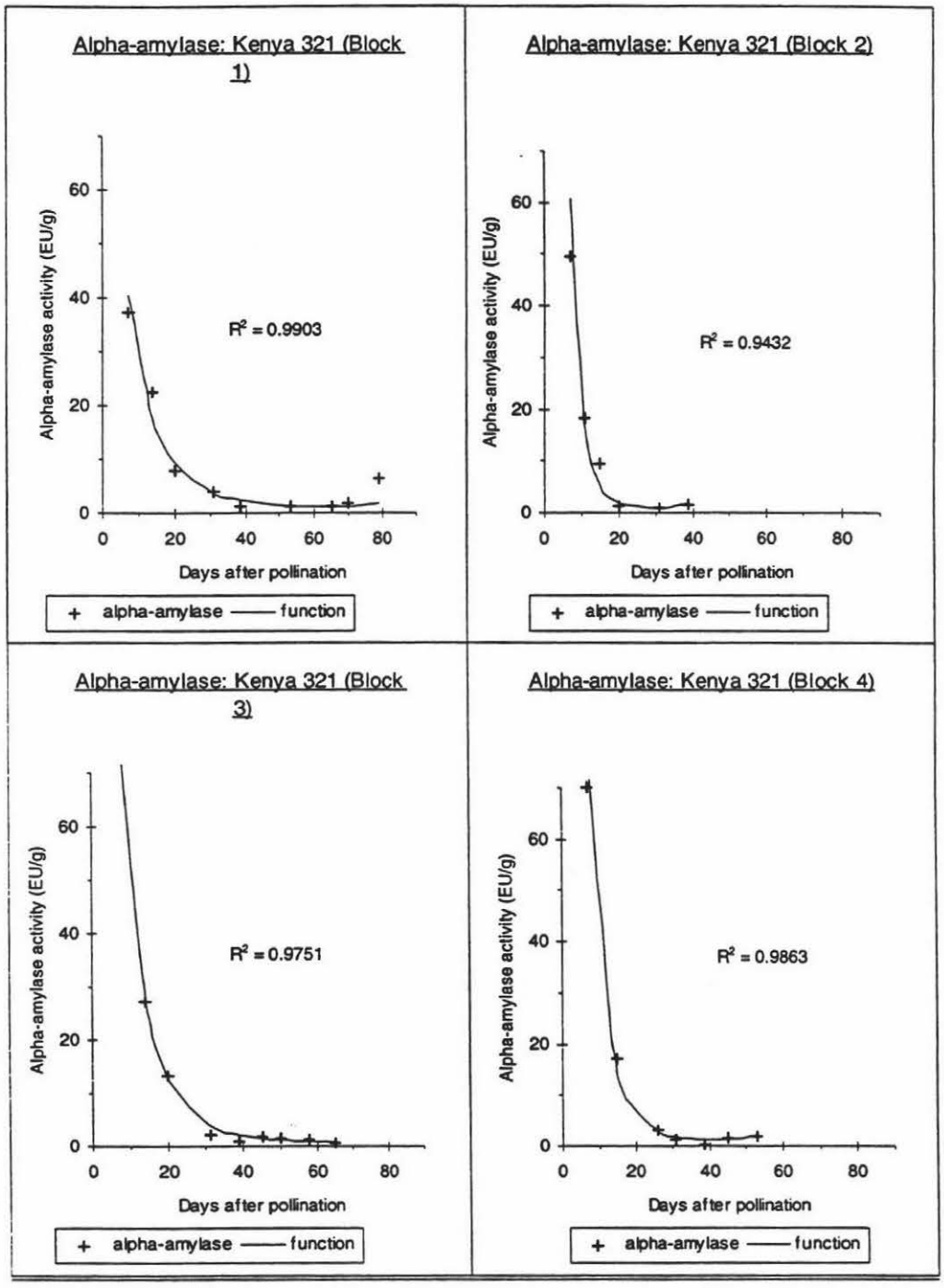


Table 7.1 (d). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

(b) Brevor

Replicate: Statistic	1	2	3	4
β_o	5.87834	5.36829	4.65436	5.50183
s.e. β_o	1.32944	0.95271	1.02668	0.73814
β_1	-0.28135	-0.20432	-0.10115	-0.20241
s.e. β_1	0.12479	0.07671	0.07395	0.06579
β_2	0.00337	0.00193	0.00032	0.00183
s.e. β_2	0.00234	0.00120	0.00101	0.00116
F_{reg}	8.11 ns	16.56*	15.45*	28.94**
R^2	0.8439	0.9169	0.9115	0.9354

significance level

* $P < 0.05$

** $P < 0.01$

*** $P < 0.001$

n.s. not significantly different (5% level)

Figure A 7.1(d). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Brevor.

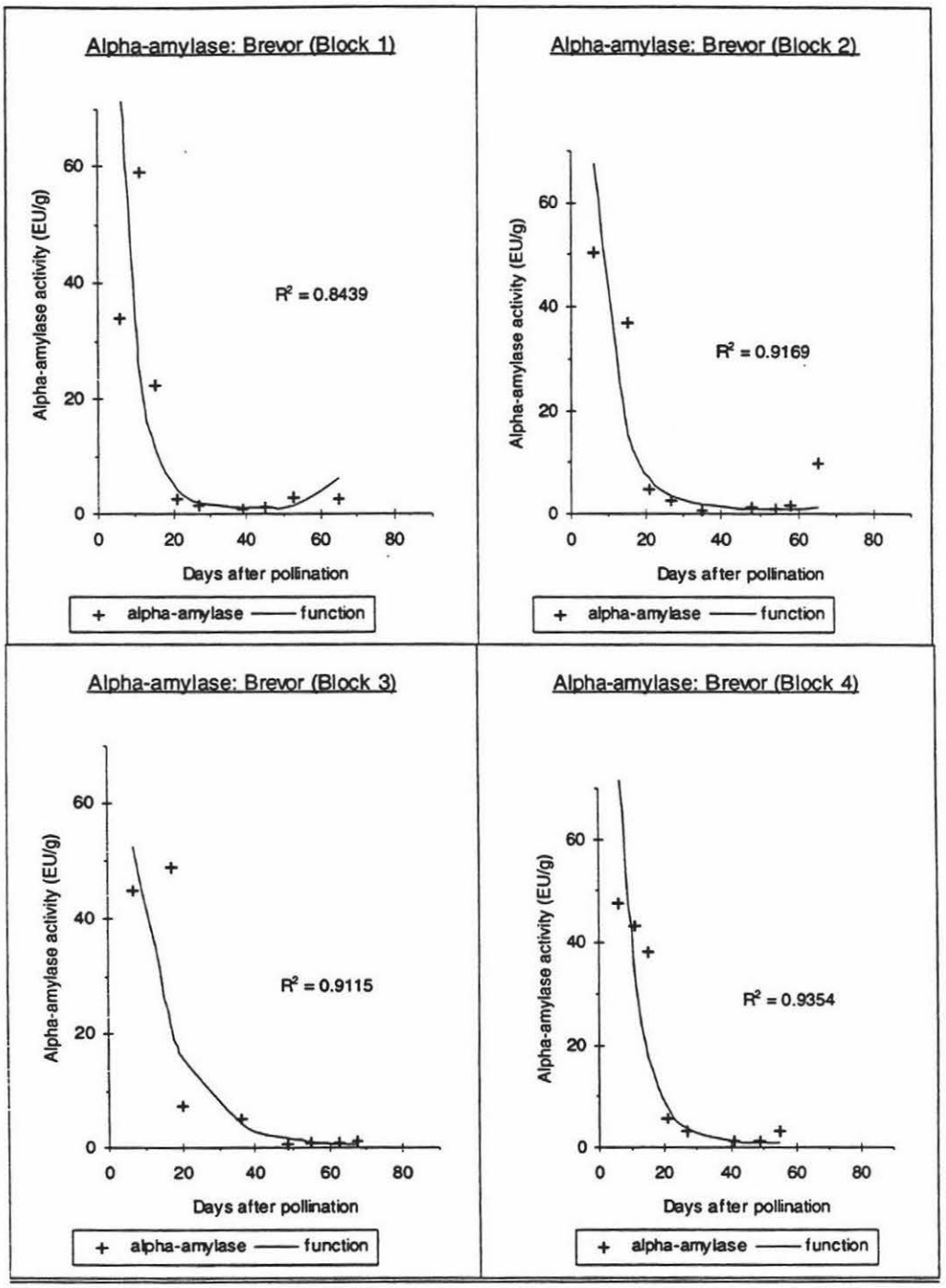


Table A 7.1 (e). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Isis

Replicate:	1	2	3	4
Statistic				
β_o	4.18037	4.80377	5.38816	5.52448
s.e. β_o	0.07053	0.68344	1.22257	2.13436
β_1	-0.19640	-0.22205	-0.31668	-0.29150
s.e. β_1	0.00849	0.06477	0.12122	0.21633
β_2	0.00256	0.00284	0.00467	0.00353
s.e. β_2	0.00020	0.00112	0.00248	0.00374
F_{reg}	1644.67***	24.66*	7.88 ns	3.52 ns
R^2	0.9997	0.9610	0.8874	0.8757

significance level

* $P < 0.05$

*** $P < 0.001$

n.s. not significantly different (5% level)

Figure A 7.1(e). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Isis.

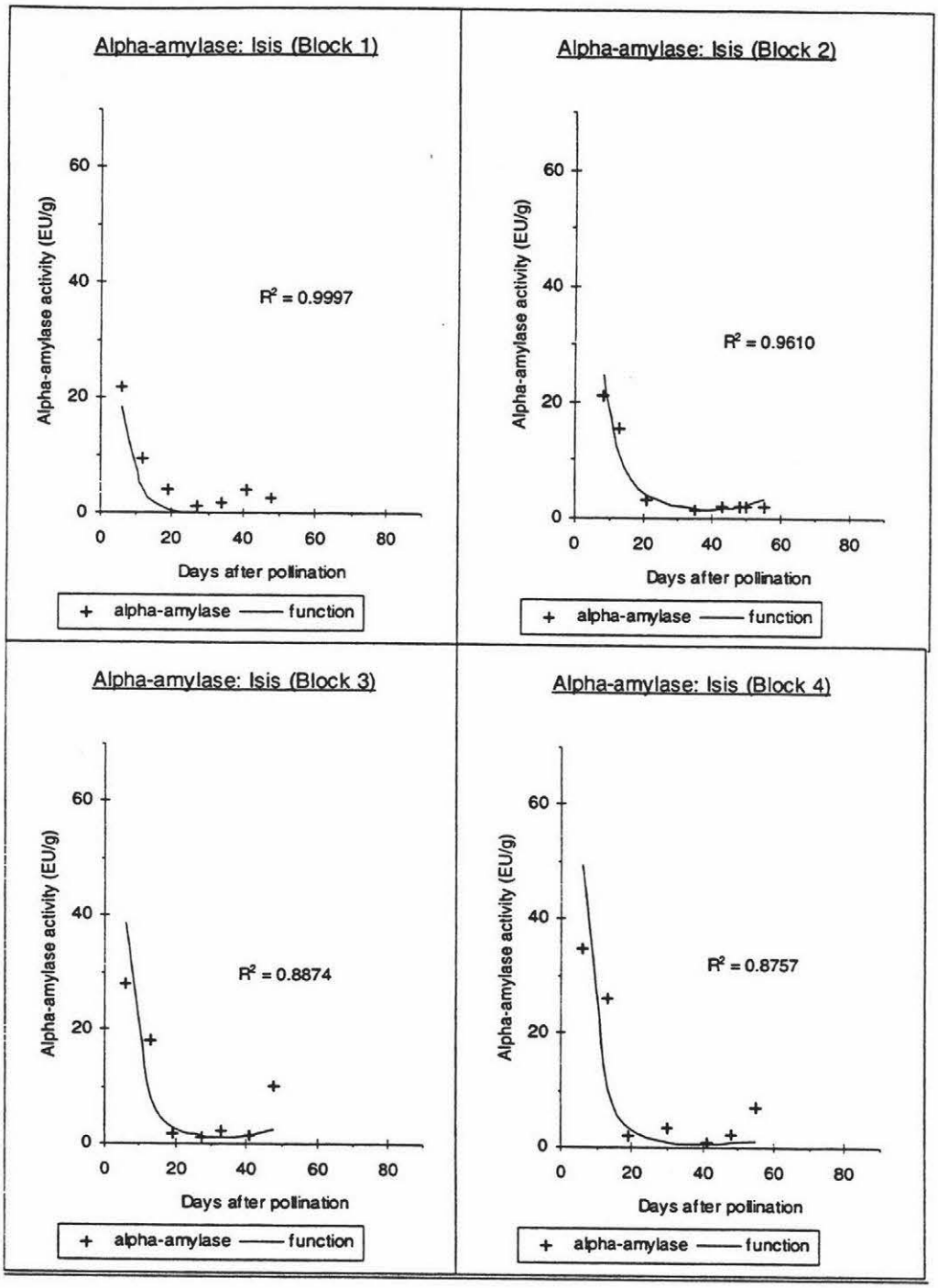


Table A 7.1 (f). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Sonora 64A

Replicate:	1	2	3	4
Statistic				
β_o	4.61187	4.75535	5.59111	5.97536
s.e. β_o	0.49185	0.53388	0.32384	1.52716
β_1	-0.11901	-0.15472	-0.24752	-0.28015
s.e. β_1	0.06785	0.03571	0.02410	0.11209
β_2	-0.00134	0.00125	0.00271	0.00323
s.e. β_2	0.00188	0.00044	0.00039	0.00178
F_{reg}	83.63**	24.71**	130.43***	6.34 ns
R^2	0.9824	0.9251	0.9849	0.7603

significance level
** $P < 0.01$
*** $P < 0.001$
n.s. not significantly different (5% level)

Figure A 7.1(f). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Sonora 64 A.

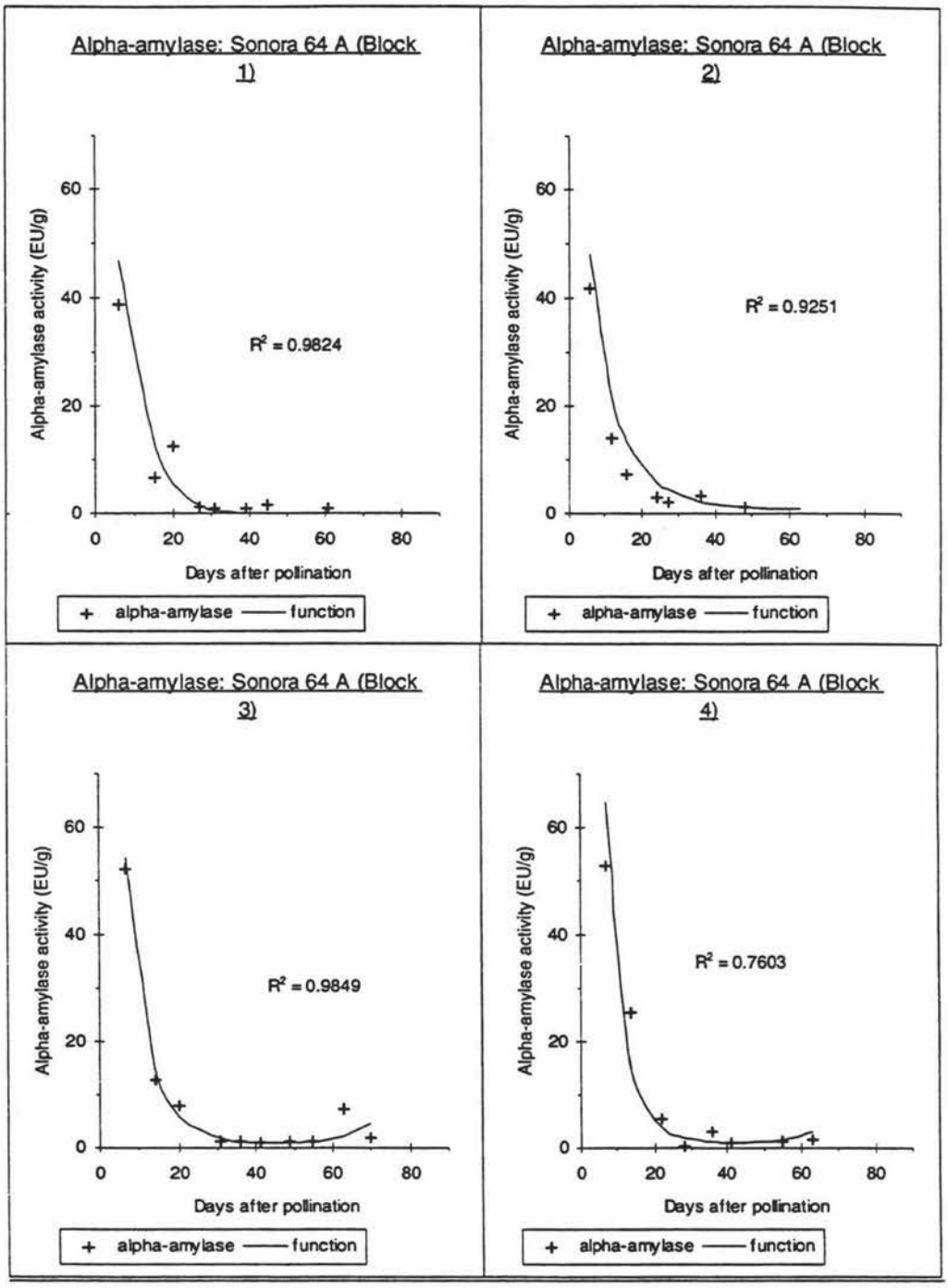


Table A 7.1 (g). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Thatcher

Replicate:	1	2	3	4
Statistic				
β_o	5.77956	5.36408	4.857813	5.16721
s.e. β_o	2.39951	0.64514	0.61778	0.58994
β_1	-0.27544	-0.25329	-0.17497	-0.24221
s.e. β_1	0.16307	0.04866	0.04384	0.04710
β_2	0.00323	0.00288	0.00154	0.00259
s.e. β_2	0.00240	0.00070	0.00059	0.00081
F_{reg}	4.03 ns	25.47*	28.85*	39.20**
R^2	0.8011	0.9444	0.9506	0.9631

significance level
* $P < 0.05$
** $P < 0.01$
n.s. not significantly different (5% level)

Figure A 7.1(g). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Thatcher.

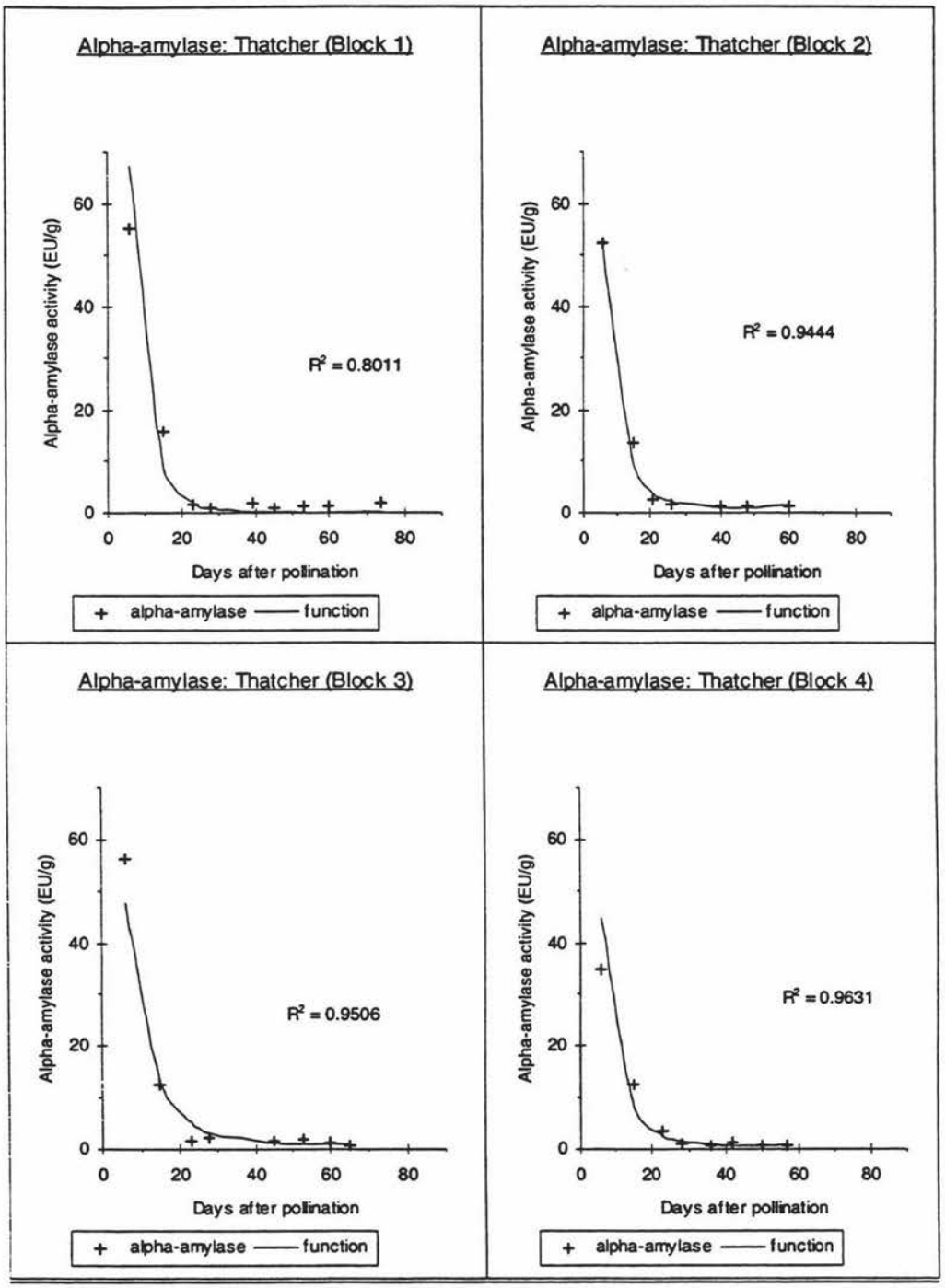


Table A 7.1 (h). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. La Prevision

Replicate:	1	2	3	4
Statistic				
β_o	5.00576	4.35642	4.30660	2.89058
s.e. β_o	0.68258	0.42679	0.47914	1.92135
β_1	-0.23542	-0.17339	-0.19498	0.09778
s.e. β_1	0.05593	0.031766	0.03937	0.20198
β_2	0.00274	0.00146	0.00195	-0.00634
s.e. β_2	0.00097	0.00050	0.00067	0.00437
F_{reg}	23.06*	59.85***	46.57**	27.86*
R^2	0.9389	0.9677	0.9688	0.9653

significance level
* $P < 0.05$
** $P < 0.01$
*** $P < 0.001$

Figure A 7.1(h). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. La Prevision.

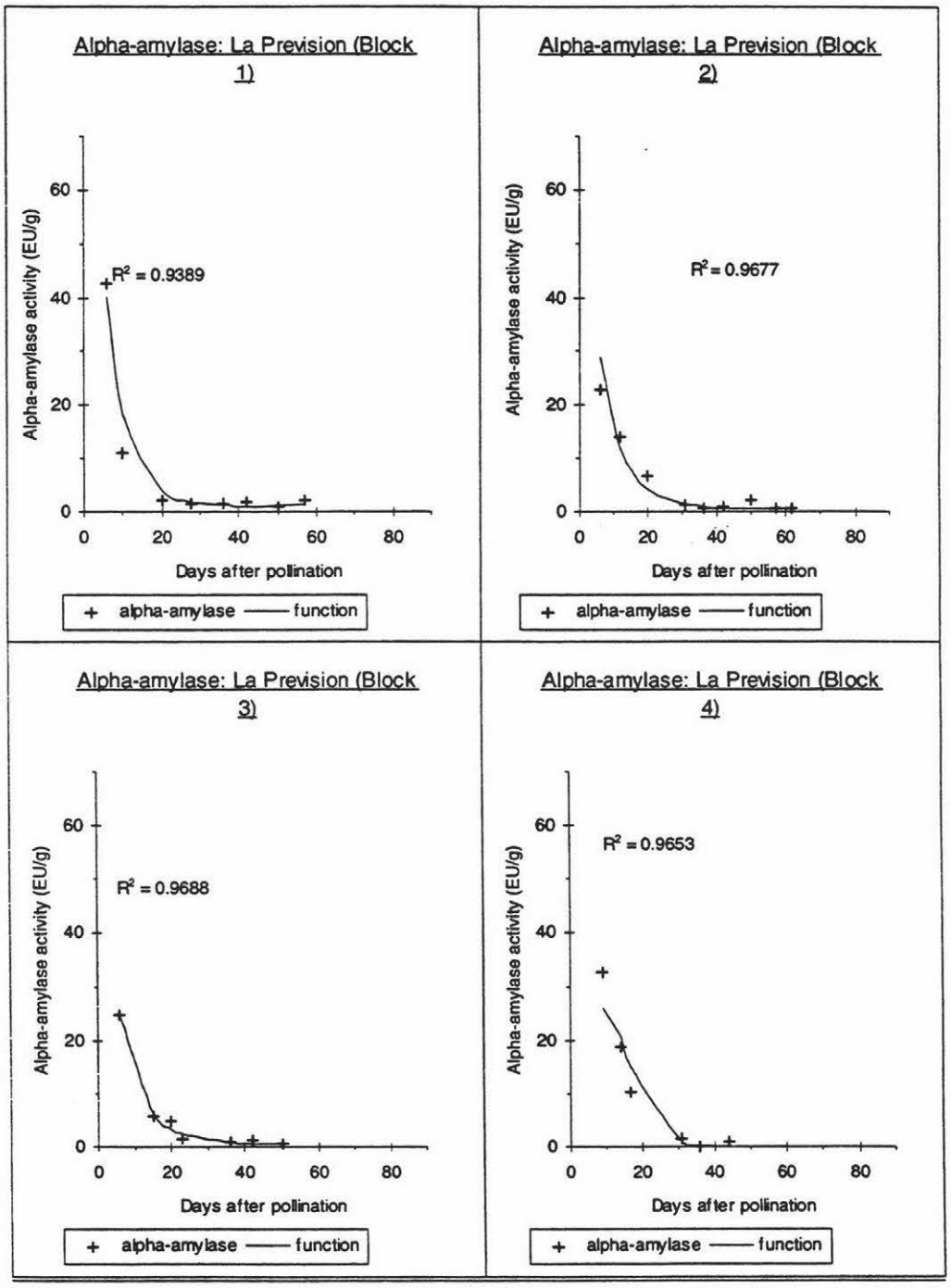


Table A 7.1 (i). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Hilgendorf 61

Replicate:	1	2	3	4
Statistic				
β_o	4.48100	4.25196	4.68424	5.06585
s.e. β_o	0.53144	0.55268	0.69618	0.49324
β_1	-0.17017	-0.06126	-0.23311	-0.15081
s.e. β_1	0.03456	0.04171	0.05417	0.04201
β_2	0.00160	-0.00035	0.00268	0.00098
s.e. β_2	0.00044	0.00063	0.00080	0.00069
F_{reg}	25.42*	47.08**	20.20*	54.25*
R^2	0.9443	0.9593	0.9309	0.9819

significance level
* $P < 0.05$
** $P < 0.01$
n.s. not significantly different (5% level)

Figure A 7.1(i). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Hilgendorf.

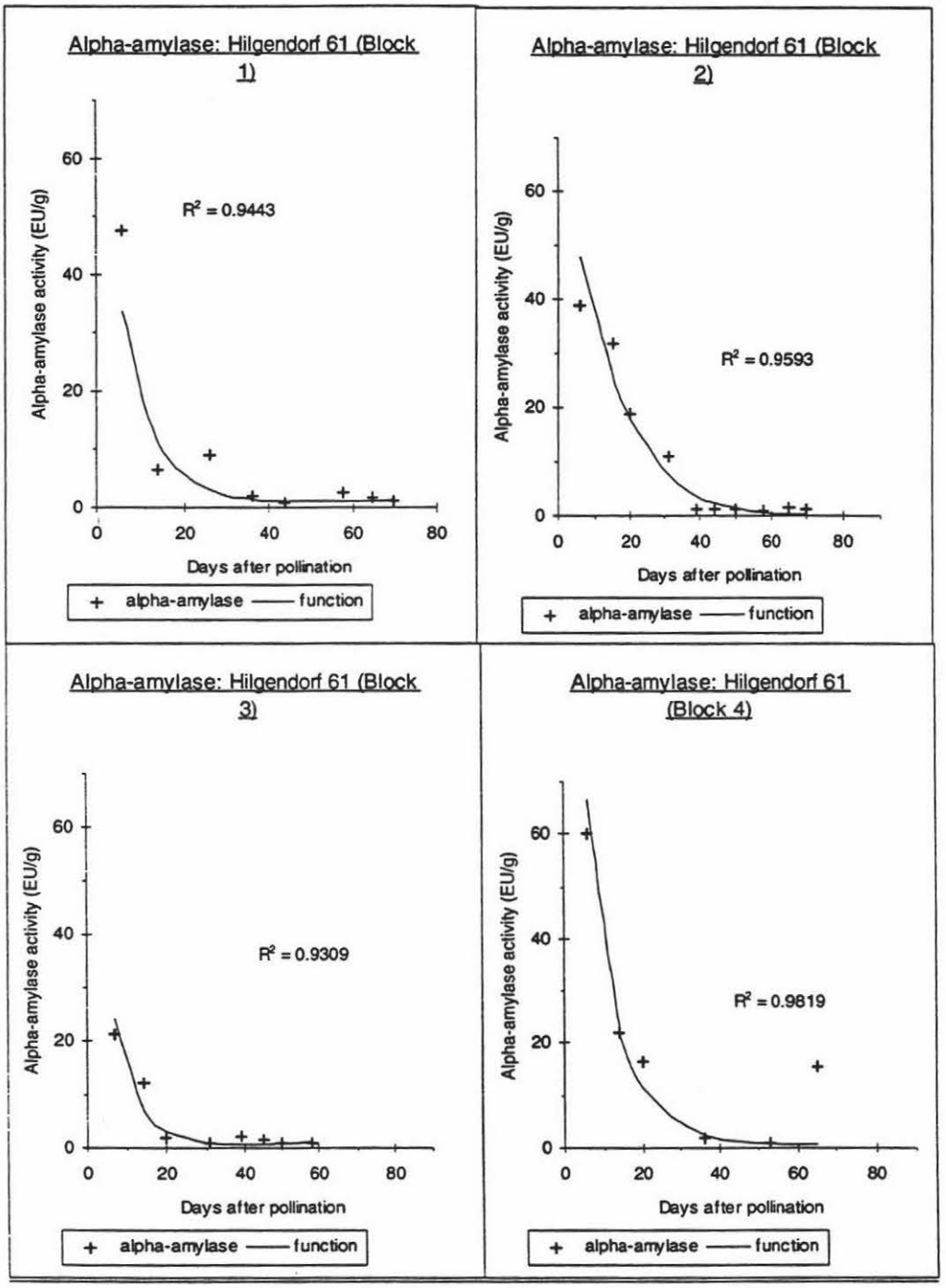


Table A 7.1 (j). Estimates of statistics for the Quadratic exponential functions fitted on α-amylase data from developing wheat grains (statistics by replicate).

cv. Karamu

Replicate:	1	2	3	4
Statistic				
β_o	4.75264	5.13850	4.56660	7.34898
s.e. β_o	0.58587	0.18350	0.52850	2.14392
β_1	-0.15848	-0.18632	-0.15012	-0.39267
s.e. β_1	0.05015	0.01449	0.03780	0.18124
β_2	0.00115	0.00160	0.00085	0.00422
s.e. β_2	0.00085	0.00022	0.00058	0.00300
F_{reg}	43.24**	411.44***	54.50**	7.91 ns
R^2	0.9665	0.9964	0.9646	0.8878

significance level

** $P < 0.01$

*** $P < 0.001$

n.s. not significantly different (5% level)

Figure A 7.1(j). The Quadratic exponential function fits and data points for alpha - amylase activity in grain from the four replicates of cv. Karamu.

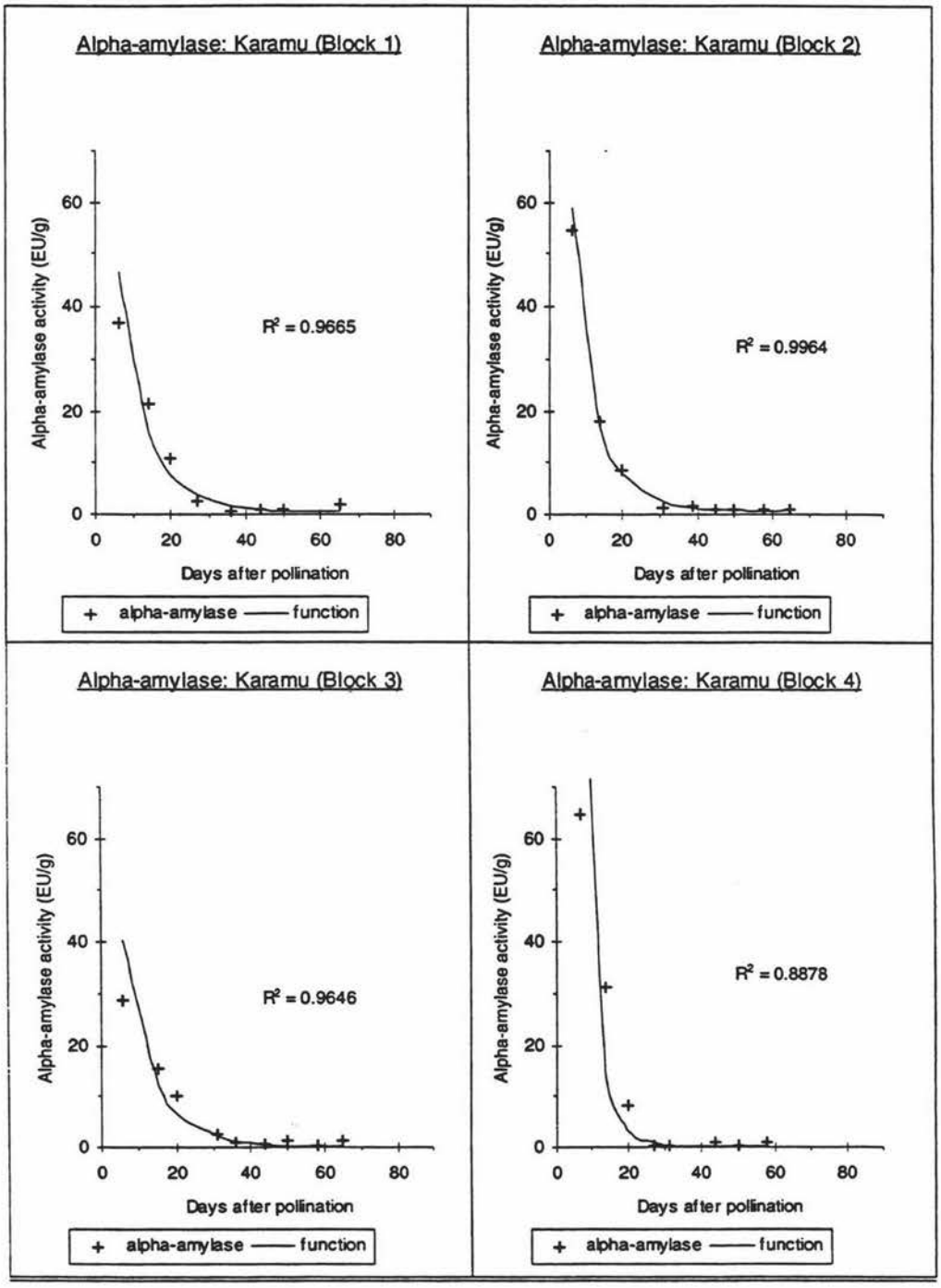


Table A 7.2. The timing (days after pollination), width (days) and height (ng/grain) of peaks of Abscisic acid activity in developing grains of ten wheat cultivars.

		1			2			3			4			5			HR
[1]Cv	Blk	day	wth	ht	day	wth	ht	day	wth	ht	day	wth	ht	day	wth	ht	
1	1	27	42	3.692	0	0	0	0	0	0	71	21	1.189	0	0	0	47.7
1	2	20	25	1.581	0	0	0	0	0	0	71	40	2.468	0	0	0	44.1
1	3	20	25	1.148	0	0	0	0	0	0	60	36	0.405	0	0	0	58.6
1	4	21	25	8.308	36	14	0.862	0	0	0	0	0	0	0	0	0	47.1
2	1	9	6	6.301	23	22	23.24	0	0	0	57	21	4.09	0	0	0	59.4
2	2	6	4	12.93	14	11	8.812	30	24	5.2	53	18	5.24	78	15	1.33	60.4
2	3	13	14	9.258	28	23	3.813	53	23	1.55	0	0	0	0	0	0	55.3
2	4	11	8	4.697	45	21	1.734	0	0	0	0	0	0	0	0	0	47.7
3	1	7	13	0.834	31	45	2.739	0	0	0	79	9	22.12	0	0	0	61.3
3	2	7	4	0.194	31	33	2.933	54	10	17.1	65	7	8.724	0	0	0	59.2
3	3	20	24	2.202	39	14	6.061	0	0	0	58	20	4.739	0	0	0	54.9
3	4	26	38	1.623	53	20	21.02	0	0	0	0	0	0	0	0	0	56.7
4	1	6	5	6.692	15	16	9.269	39	38	4.05	0	0	0	0	0	0	54
4	2	6	15	6.493	27	27	1.489	0	0	0	58	17	1.373	0	0	0	52.6
4	3	7	13	11.55	49	35	1.612	0	0	0	63	13	0.584	0	0	0	53.3
4	4	6	9	5.672	21	26	4.903	55	29	5.3	0	0	0	0	0	0	57.8
5	1	19	27	2.427	34	8	3.02	48	7	1.56	0	0	0	0	0	0	49.4
5	2	21	22	4.498	43	15	7.857	0	0	0	60	10	2.885	0	0	0	50.3
5	3	19	21	4.06	33	14	6.52	48	7	0.95	0	0	0	0	0	0	49
5	4	30	42	21.47	55	7	4.134	0	0	0	0	0	0	0	0	0	57.8
6	1	15	14	0.257	27	25	1.26	0	0	0	0	0	0	0	0	0	42.8
6	2	6	6	1.295	24	15	51.81	0	0	0	63	27	3.031	0	0	0	56.6
6	3	7	7	0.319	20	22	4.609	41	13	0.17	63	12	3.154	0	0	0	51.7
6	4	22	48	9.222	0	0	0	0	0	0	78	23	1.583	0	0	0	51.6
7	1	6	17	9.406	28	22	0.763	53	15	0.81	0	0	0	0	0	0	57.6
7	2	6	17	16.53	48	34	0.943	0	0	0	0	0	0	0	0	0	57.6
7	3	15	39	11.67	0	0	0	0	0	0	53	20	0.494	0	0	0	48.5
7	4	6	9	2.284	23	13	0.157	36	14	2.46	50	15	3.284	0	0	0	51.3
8	1	6	9	1.283	20	16	28.66	36	11	8.79	57	7	1.092	0	0	0	50
8	2	12	25	1.856	36	19	3.542	0	0	0	57	7	3.145	0	0	0	50.5
8	3	6	9	0.365	20	8	3.218	36	19	2.85	50	8	1.328	0	0	0	53.3
8	4	9	5	0.226	31	36	3.562	0	0	0	58	8	2.307	0	0	0	50.5
9	1	6	8	1.041	36	30	1.72	0	0	0	58	26	4.367	0	0	0	43.8
9	2	6	9	4.55	31	14	10.88	44	26	2.37	70	5	4.375	0	0	0	54.2
9	3	20	38	12.94	0	0	0	0	0	0	65	20	0.6	0	0	0	54.8
9	4	20	30	2.879	0	0	0	0	0	0	65	29	2.803	0	0	0	53.9
10	1	6	8	24.95	20	22	0.747	50	22	1.14	0	0	0	0	0	0	54.6
10	2	6	8	16.83	20	17	0.964	39	14	0.45	58	24	0.813	0	0	0	52.2
10	3	6	9	0.641	31	21	2.556	50	22	0.48	65	7	0.601	0	0	0	53.6
10	4	20	20	7.005	31	17	1.332	50	14	2.19	0	0	0	0	0	0	57.8

Figure A 7.3. Original data collected in the experiment inr the study of the Genotypic variation of dormancy in wheat.

Cultivar	Block	dap	mstur	gnwt	sg	ng	dorm	col	flavs	amy	aba	DTF
Gamenya	1	7	77.6	2.33	0	0	100.00	0	312.29	45.118	0.922	94
Gamenya	1	15	67.4	19.51	0	0	100.00	0	79	3.937	1.429	98
Gamenya	1	20	66.3	21.97	10	0	100.00	0	68.6	2.899	1.623	90
Gamenya	1	27	56.4	27.56	40	10	75.00	0.5	32.5	1.853	3.692	86
Gamenya	1	36	29.1	28.23	80	70	12.50	1	25.1	3.961	0.948	98
Gamenya	1	50	10.9	40.27	100	90	10.00	1.5	18.3	19.612	0.11	98
Gamenya	1	65	11.3	36.13	100	100	0.00	2	16.8	1.865	0.061	90
Gamenya	1	71	11.7	41.81	100	100	0.00	2	.	60.072	1.189	93
Gamenya	2	6	74.8	4.26	0	0	100.00	0	151.8	24.682	0.225	97
Gamenya	2	14	73	10.26	0	0	100.00	0	73.3	14.237	0.324	89
Gamenya	2	20	67.5	25.43	10	0	100.00	0	55.2	3.475	1.581	89
Gamenya	2	23	58.6	38.4	30	20	33.33	1	24.2	1.013	0.631	89
Gamenya	2	31	43.2	36.21	80	60	25.00	1.5	23.7	1.106	0.018	89
Gamenya	2	36	23.8	39.42	90	70	22.22	2	18.5	7.253	0.318	89
Gamenya	2	50	11.5	44.58	90	80	11.11	2	17.4	1.867	0.675	89
Gamenya	2	71	10.1	46.2	100	90	10.00	2.5	17.1	81.985	2.468	89
Gamenya	3	7	73.5	4.71	0	0	100.00	0	179.7	24.741	0.501	90
Gamenya	3	14	73.2	9.35	0	0	100.00	0	73.1	15.781	0.665	90
Gamenya	3	20	66.1	20.34	10	0	100.00	0	34.8	7.744	1.148	90

Gamenya	3	31	52.3	29.62	70	40	42.86	0.5	22.8	2.394	0.158	90
Gamenya	3	38	43.5	35.72	90	70	22.22	2	15.9	0.493	0.234	88
Gamenya	3	60	10.9	49.02	90	80	11.11	2	16.9	9.939	0.405	88
Gamenya	3	67	11.8	48.06	90	90	0.00	2	15.8	15.236	0.139	88
Gamenya	3	81	23.4	48.65	100	100	0.00	2 .	.	.		88
Gamenya	4	6	74.1	4.62	0	0	100.00	0	243	49.240	0.319	94
Gamenya	4	14	73.1	7.68	0	0	100.00	0.5	104.9	21.211	0.396	89
Gamenya	4	21	68	21.06	15	0	100.00	0.5	47.7	5.033	8.308	85
Gamenya	4	31	46.1	34.42	50	30	40.00	1	21.9	1.339	1.79	89
Gamenya	4	36	37.5	35.81	100	80	20.00	1.5	18.6	36.135	0.862	89
Gamenya	4	45	13.2	36.11	100	100	0.00	2	16.4	1.513	0.491	94
Tordo	1	9	74.9	4.66	0	0	100.00	0	176.4	18.454	6.301	95
Tordo	1	14	70.3	9.76	0	0	100.00	0.5	93.1	10.905	4.221	90
Tordo	1	23	58.2	27.5	0	0	100.00	1	67.6	2.123	23.242	77
Tordo	1	27	56.6	29.61	0	0	100.00	1.5	24.9	0.382	1.497	77
Tordo	1	36	49.1	31.22	35	0	100.00	1.5	22.1	1.772	0.258	77
Tordo	1	44	38.6	36.02	80	60	25.00	1.5 .		2.750	0.387	90
Tordo	1	49	11.3	41.04	90	70	22.22	1.5	16.9	1.527	0.393	77
Tordo	1	57	9.9	45.36	80	80	0.00	1.5	16.5 .		4.09	83
Tordo	2	6	.	.	0	0	100.00	0 .		30.751	12.928	86
Tordo	2	10	73.5	8.64	0	0	100.00	0	158.1	48.912	4.867	86
Tordo	2	14	72.7	13.45	0	0	100.00	0	114.4	19.354	14.17	86

Tordo	2	21	59.7	34.31	0	0	100.00	1.5	36.7	1.693	2.024	79
Tordo	2	30	51.8	38.87	0	0	100.00	1.5	35.3	2.425	3.37	76
Tordo	2	36	45.9	40.18	20	0	100.00	2.5	27.3	3.440	0.482	94
Tordo	2	45	29.9	40.3	80	60	25.00	3	26.7	0.977	0.241	94
Tordo	2	53	23.3 .		90	80	11.11	3	23.7	2.277	5.24	76
Tordo	2	63	9.4	52.9	100	90	10.00 .		12.9	1.119	0.134	76
Tordo	2	78 .		53.72	100	100	0.00 .		13.9	19.554	1.328	76
Tordo	3	7	75.5	4.16	0	0	100.00	0	208	24.609	0.707	90
Tordo	3	13	72.1	14.95	0	0	100.00	0	103.8	21.429	9.258	88
Tordo	3	22	60.4	31.29	0	0	100.00	0	30.3	1.715	3.329	88
Tordo	3	28	53.1	41.84	0	0	100.00	1	28.7	1.781	3.813	85
Tordo	3	39	43.6	40.08	20	0	100.00	2	25.2	0.720	1.589	95
Tordo	3	45	18.1	47.95	80	60	25.00	2	21.4	1.237	0.104	95
Tordo	3	53	10.9	50.04	90	80	11.11	2	13.5	12.528	1.548	95
Tordo	3	68	9.2 .		100	100	0.00	2 .		23.311	0.928	85
Tordo	3	75	.		100	100	0.00	2 .	.	.		85
Tordo	4	9	75.4	4.54	0	0	100.00	0	204.3	26.709	1.128	94
Tordo	4	11	73.8	8.15	0	0	100.00	0	86.4	49.554	5.8	85
Tordo	4	17	70.8	15	0	0	100.00	0	68.6	11.729	0.16	79
Tordo	4	24	61.5	28.02	0	0	100.00	0.5	47.3	3.229	0.585	76
Tordo	4	27	45.9	31.05	30	0	100.00	1.5	23.8	1.760	0.644	76
Tordo	4	40	31.8	32.77	80	70	12.50	2	21.5	0.693	1.106	85

Tordo	4	45	10.8	36.17	100	100	0.00	3	18.9	1.301	1.734	94
Kenya 321	1	7	75.1	3.56	0	0	100.00	0	338.8	37.201	0.834	90
Kenya 321	1	14	72.5	9.42	0	0	100.00	0	124.8	22.573	0.118	90
Kenya 321	1	20	65.7	21.22	0	0	100.00	0	85.6	8.002	0.52	90
Kenya 321	1	31	51.5	42.44	60	10	83.33	1	57.6	3.877	2.739	95
Kenya 321	1	39	39.6	37.01	90	40	55.56	1	41.2	1.262	1.748	95
Kenya 321	1	53	24.7	43.71	100	80	20.00	1.5	19.3	1.345	0.407	95
Kenya 321	1	65	10.6	45.91	100	90	10.00	2	19.5	1.269	0.012	90
Kenya 321	1	70	11.9	46.31	100	100	0.00	2 .		1.797	0.058	90
Kenya 321	1	79	18	51.5	100	100	0.00	3 .		6.334	22.115	90
Kenya 321	1	86	12	44.22	100	100	0.00	3 .	.	.		90
Kenya 321	2	7	75.4	3.48	0	0	100.00	0	394	49.447	0.194	89
Kenya 321	2	11	73.8	5.51	0	0	100.00	0	170.2	18.237	0.181	94
Kenya 321	2	15	70.2	9.07	0	0	100.00	0	123.4	9.303	0.424	89
Kenya 321	2	20	65.9	19.89	0	0	100.00	0	43.3	1.187	0.732	94
Kenya 321	2	31	50.1	43.02	60	10	83.33	1	36.2	1.076	2.933	94
Kenya 321	2	39	41.4	48.72	70	20	71.43	2	28.1	1.608	0.872	85
Kenya 321	2	48	31.8	51.63	80	60	25.00	2.5	21.1	1.380	0.344	85
Kenya 321	2	54	11.7	48.08	90	80	11.11	2.5	20.4	1.426	17.131	89
Kenya 321	2	58	19.1	48.67	100	90	10.00	3	17.3	1.085	0.404	89
Kenya 321	2	65	10.7	49.22	100	100	0.00	3	16.3		8.724	85
Kenya 321	3	7	74.4	3.59	0	0	100.00	0	153.4	92.373	0.11	90

Kenya 321	3	14	72	8.1	0	0	100.00	0	108.4	27.422	0.186	90
Kenya 321	3	20	68	16.97	0	0	100.00	0	47	13.368	2.202	90
Kenya 321	3	31	54	29.62	50	20	60.00	0.5	23.4	2.269	0.225	95
Kenya 321	3	39	45.7	37.17	80	40	50.00	1	18.4	0.815	6.061	95
Kenya 321	3	45	20.8	46.34	90	60	33.33	3	15.8	1.732	0.035	95
Kenya 321	3	50	10.4	46.51	90	70	22.22	3	14.3	1.619	4.64	90
Kenya 321	3	58	19.6	46.93	100	100	0.00	3	15.9	1.097	4.74	90
Kenya 321	3	65	11.1	48	100	100	0.00	3	15.6	0.743	0.179	90
Kenya 321	3	70	9.5	50.21	100	100	0.00	3	14.1	.		90
Kenya 321	4	7	73.4	4.81	0	0	100.00	0	174	70.061	0.079	89
Kenya 321	4	15	67.7	12.73	0	0	100.00	0	73.1	17.103	0.335	89
Kenya 321	4	26	56.2	27.11	30	10	66.67	0.5	43.8	3.105	1.623	94
Kenya 321	4	31	46.4	46.71	50	20	60.00	1	40.4	1.331	1.44	94
Kenya 321	4	39	35.9	36.91	70	40	42.86	2	34.5	0.307	0.631	94
Kenya 321	4	45	10.9	39.31	90	60	33.33	2.5	27.9	1.498	0.059	94
Kenya 321	4	53	20.3	45.65	100	80	20.00	3	14.8	1.795	21.015	94
Kenya 321	4	65	11.3	49.77	100	100	0.00	3	17.9	1.716	1.18	89
Kenya 321	4	70	10.7	51.77	100	100	0.00	3 .		.		89
Kenya 321	4	79	20.2	43.6	100	100	0.00	3 .		.		89
Brevor	1	6	76.8	4.38	0	0	100.00	0	489.2	33.763	6.692	95
Brevor	1	11	74.9	6.98	0	0	100.00	0	122.7	59.121	0.082	86
Brevor	1	15	70.9	12.49	0	0	100.00	0	118.7	22.197	1.31	86

Brevor	1	21	69.1	22.11	0	0	100.00	0.5	47.9	2.449	7.848	86
Brevor	1	27	56.7	34.99	40	0	100.00	1	26.7	1.468	3.369	86
Brevor	1	39	42.9	44.59	50	40	20.00	1.5	22.9	0.814	4.047	95
Brevor	1	45	20.8	45.06	100	100	0.00	2	16.8	1.192	0.657	95
Brevor	1	53	21.3	44.63	100	100	0.00	2.5	15.5	2.701	0.517	95
Brevor	1	65	10.5	57.61	100	100	0.00	3	12.7	2.451	0.009	95
Brevor	2	6	77.2	3.31	0	0	100.00	0	383.8	50.597	6.493	94
Brevor	2	15	74.3	11.14	0	0	100.00	0	68.1	36.823	2.26	85
Brevor	2	21	69.2	22.34	30	0	100.00	0	37	4.669	3.36	85
Brevor	2	27	55.8	26.95	40	0	100.00	0.5	20.9	2.528	1.489	85
Brevor	2	35	52.9	36.51	90	60	33.33	1	22.5	0.788	1.406	85
Brevor	2	48	16.3	36.83	100	100	0.00	2	22	1.379	0.225	85
Brevor	2	54	9.6	49.99	100	100	0.00	2	14.2	0.826	0.281	85
Brevor	2	58	20	55.47	100	100	0.00	2.5	12.4	1.465	1.373	89
Brevor	2	65	11	50.91	100	100	0.00	3	12.9	9.726	0.669	89
Brevor	3	7	76.1	7.51	0	0	100.00	0	213.6	44.746	11.548	90
Brevor	3	17	71.6	17.85	10	0	100.00	0	52.3	48.718	0.821	90
Brevor	3	20	63.4	26.74	20	0	100.00	0	31.6	7.302	4.53	90
Brevor	3	36	48.6	51.06	40	10	75.00	1	18.1	4.977	1.441	85
Brevor	3	49	11.2	59.84	90	60	33.33	2	13.9	0.525	1.612	85
Brevor	3	55	9.1	42.68	100	100	0.00 .		16.1	0.808	0.095	85
Brevor	3	63	22	51.09	100	100	0.00	2.5	14.4	0.751	0.584	85

Brevor	3	68	11	49.64	100	100	0.00	3 .		1.112	0.374	85
Brevor	4	6	77	3.62	0	0	100.00	0	210.7	47.570	5.672	94
Brevor	4	11	74.7	7.51	0	0	100.00	0	185.9	43.236	0.079	85
Brevor	4	15	74.5	11.26	0	0	100.00	0	58.1	38.038	0.067	85
Brevor	4	21	68.1	21.59	25	0	100.00	0.5	37.8	5.777	4.903	85
Brevor	4	27	54.5	34.27	30	0	100.00	1	28.1	3.149	1.408	85
Brevor	4	41	42	52.49	80	30	62.50	2	27.7	1.105	0.144	84
Brevor	4	49	19.9	48.71	90	60	33.33	2	18.4	1.173	0.146	84
Brevor	4	55	8.6	50.04	100	100	0.00	2.5	13	3.115	5.302	84
Brevor	4	63	17.8	55.31	100	100	0.00	2.5	13.9	13.965	3.442	84
Brevor	4	70	10.9	52.14	100	100	0.00	3	14.4	11.294	0.227	84
Isis	1	6	73.9	4.45	0	0	100.00	0	169.3	21.825	0.002	107
Isis	1	12	74	11.89	0	0	100.00	0	155.2	9.199	0.017	114
Isis	1	19	66.4	18.11	10	0	100.00	0	91.5	3.881	2.427	107
Isis	1	33	49.7	30.79	20	10	50.00	1	29.2	1.025	0.229	107
Isis	1	34	31.1	40.27	50	30	40.00	1.5	18.7	1.597	3.02	114
Isis	1	41	24.1	42.76	100	90	10.00	2	17.1	3.878	0.45	114
Isis	1	48	19.3	44.72	100	100	0.00	2 .		2.516	1.559	107
Isis	2	8	74	3.9	0	0	100.00	0	162.4	21.041	0.006	104
Isis	2	13	72.7	10.56	0	0	100.00	0	98.3	15.535	0.054	99
Isis	2	21	62.1	21.08	20	0	100.00	0.5	65.9	3.219	4.498	99
Isis	2	35	48.3	34.82	30	10	66.67	1.5	38.9	1.412	0	104

Isis	2	43	36.7	38.18	70	30	57.14	2	16.7	1.866	7.857	104
Isis	2	48	34.2	41.65	90	60	33.33	2	16.8	1.887	2.983	99
Isis	2	50	11.4	43.79	90	70	22.22	2.5	16.3	2.032	0.384	104
Isis	2	55	9	49.98	100	80	20.00	2.5	13.4	1.938	1.229	99
Isis	2	60	10.8	50.3	100	100	0.00	3	14.2		2.885	99
Isis	3	6	75.3	3.76	0	0	100.00	0	226.4	27.945	0.002	107
Isis	3	13	72.9	9.35	0	0	100.00	0	90.9	18.045	0.226	100
Isis	3	19	61.7	25.36	20	0	100.00	0	46.5	1.755	4.06	107
Isis	3	27	53.2	31.25	40	10	75.00	0	29.9	1.180	0.105	107
Isis	3	33	43.9	35.37	50	10	80.00	1.5	26.8	2.247	6.52	107
Isis	3	41	17.3	40.37	80	60	25.00	2.5	15.6	1.366	0.26	107
Isis	3	48	11	44.73	100	80	20.00	3	15.7	10.177	0.947	107
Isis	3	53	10.3	47.8	100	90	10.00	3 .			1.397	107
Isis	3	62	19.4	51.37	100	100	0.00	3 .			1.282	107
Isis	4	6	75.9	3.08	0	0	100.00	0	237.9	34.671	0.001	106
Isis	4	13	73.2	7.94	0	0	100.00	0	102.9	26.039	0.206	99
Isis	4	19	61.7	25.36	10	0	100.00	0	48.3	1.932	2.008	106
Isis	4	30	43.5	37.71	30	10	66.67	0.5 .		3.481	2.24	106
Isis	4	41	36.9	44.86	50	40	20.00	2	20	0.744	0.352	106
Isis	4	48	11.6	45.93	90	80	11.11	2.5	16.4	2.128	0.326	99
Isis	4	55	27.8	45.9	100	90	10.00	2.5	16.5	6.999	4.134	99
Isis	4	60	13	45.13	100	100	0.00	2.5 .			1.372	99

Sonora 64 A	1	6	72.8	6.76	0	0	100.00	0	208.1	38.853	0.046	95
Sonora 64 A	1	15	65.5	16.88	0	0	100.00	0	57	6.691	0.257	95
Sonora 64 A	1	20	63	19.64	0	0	100.00	0	43	12.423	0.021	77
Sonora 64 A	1	27	52.6	31.73	40	10	75.00	2	33.8	1.061	1.26	77
Sonora 64 A	1	31	43.7	34.99	70	40	42.86	3.5	27	0.822	0.602	95
Sonora 64 A	1	39	11.1	39.65	90	50	44.44	5	19.7	0.844	0.073	95
Sonora 64 A	1	45	10.3	40.05	100	70	30.00	5	17	1.387	0.024	95
Sonora 64 A	1	61	.	44.1	100	80	20.00	5	16.1	1.005	.	87
Sonora 64 A	1	68	.	44.51	100	90	10.00	5	16.2	8.020	.	87
Sonora 64 A	1	70	.	44.72	100	90	10.00	5	14.2	1.509	.	85
Sonora 64 A	2	6	75.1	1.94	0	0	100.00	0	274.5	29.392	1.295	97
Sonora 64 A	2	12	72.4	11.25	0	0	100.00	0	82.5	41.744	0.12	84
Sonora 64 A	2	16	67.8	13.54	0	0	100.00	0	73.3	14.075	2.43	84
Sonora 64 A	2	24	61	18.04	10	0	100.00	0	69.1	7.214	2.63	76
Sonora 64 A	2	27	53.9	27.47	40	10	75.00	1.5	26.8	3.147	0.067	76
Sonora 64 A	2	36	44.8	38.08	90	40	55.56	4	29.2	1.998	0.835	76
Sonora 64 A	2	63	25.7	40.82	100	80	20.00	5	13.6	3.403	3.031	76
Sonora 64 A	2	71	19.1	41.21	100	100	0.00	5	16.6	1.117	.	76
Sonora 64 A	2	78	8.4	43.75	100	100	0.00	5	15.8	35.936	.	76
Sonora 64 A	3	7	74.2	4.44	0	0	100.00	0	248.6	52.073	0.319	90
Sonora 64 A	3	14	70.1	10.46	0	0	100.00	0.5	77	12.702	0.159	90
Sonora 64 A	3	20	61.5	17.85	20	0	100.00	2	40.4	7.904	4.609	90

Sonora 64 A	3	31	45.1	38.76	40	10	75.00	3	39.6	1.272	0.916	90
Sonora 64 A	3	36	33.3	36.18	60	20	66.67	4	36	1.288	0.126	90
Sonora 64 A	3	41	26.7	42.92	60	30	50.00	4 .		1.008	0.167	85
Sonora 64 A	3	49	12.3	41.74	100	40	60.00	5	24.7	1.157	0.145	85
Sonora 64 A	3	55	10.3 .		100	60	40.00	5	20.8	1.213	0.341	85
Sonora 64 A	3	63	19.8	41.51	100	70	30.00	5	18	7.171	3.154	85
Sonora 64 A	3	70	10.5 .		100	80	20.00	5 .		1.958	0.247	85
Sonora 64 A	4	7	73.9	4.05	0	0	100.00	0	268.1	52.874	0.176	89
Sonora 64 A	4	14	71.8	8.98	0	0	100.00	0.5	102.4	25.577	0.349	89
Sonora 64 A	4	22	62.7	20.73	0	0	100.00	1	39.3	5.526	9.222	84
Sonora 64 A	4	28	51	29.31	30	0	100.00	2.5	25.4	0.343	1.48	84
Sonora 64 A	4	36	47.5	34.54	90	20	77.78	3	29	3.175	0.204	84
Sonora 64 A	4	41	41.1	35.4	100	30	70.00	5	22.3	1.053	0.094	84
Sonora 64 A	4	55	8.9	39.87	100	70	30.00	5	15.2	1.189	0.028	84
Sonora 64 A	4	63	20.2	44.82	100	90	10.00	5	13.5	1.546	1.542	84
Sonora 64 A	4	78	10.1	45.72	100	100	0.00	5	14.2	5.149	1.583	76
Sonora 64 A	4	92		45.61	100	100	0.00	5 .		2.952 .		76
Thatcher	1	6	74.9	3.76	0	0	100.00	0	265.5	55.244	9.406	95
Thatcher	1	15	70.1	11.07	0	0	100.00	0	66.3	15.685	0.587	95
Thatcher	1	23	54.6	30.58	10	0	100.00	0.5	34.5	1.522	0.162	98
Thatcher	1	28	49.2	34.33	30	0	100.00	1	22.5	0.913	0.763	98
Thatcher	1	39	39.9	35.55	50	0	100.00	4	16.7	1.750	0.319	95

Thatcher	1	45	18.9	42.75	60	0	100.00	5	17.1	0.814	0.009	95
Thatcher	1	53	21.1	.	80	10	87.50	5	16.6	1.121	0.812	95
Thatcher	1	60	11.1	.	90	50	44.44	5	17	1.337	0.257	95
Thatcher	1	74	.	.	100	70	30.00	5	19.5	1.779	.	95
Thatcher	1	79	.	.	100	90	10.00	5	.	.		90
Thatcher	2	6	74.8	2.84	0	0	100.00	0	385.2	52.372	16.53	94
Thatcher	2	15	68.6	11.65	0	0	100.00	0	75.8	13.479	0.358	94
Thatcher	2	21	58.4	22.06	10	0	100.00	0.5	45.1	2.442	0.092	99
Thatcher	2	26	50.1	29.38	10	0	100.00	.	42.9	1.671	0.11	99
Thatcher	2	40	18.9	31.52	30	0	100.00	4.5	19.1	1.293	0.289	99
Thatcher	2	48	19.8	32.2	70	10	85.71	5	17.5	1.346	0.943	99
Thatcher	2	60	10.6	31.05	90	20	77.78	5	16.9	1.348	0.516	94
Thatcher	2	65	10.9	34.85	90	30	66.67	5	.	.		94
Thatcher	2	74	.	.	100	40	60.00	5	.	.		94
Thatcher	2	81	.	.	100	70	30.00	5	.	.		94
Thatcher	3	6	74.9	2.6	0	0	100.00	0	665.9	56.331	6.147	95
Thatcher	3	15	69.5	11.52	0	0	100.00	0	65.5	12.485	11.667	95
Thatcher	3	23	54.3	31.79	10	0	100.00	0.5	27.3	1.680	0.554	98
Thatcher	3	28	48.8	31.08	20	0	100.00	1	24.3	2.091	0.106	98
Thatcher	3	45	11.6	36.32	60	0	100.00	5	25.1	1.567	0.007	95
Thatcher	3	53	.	.	90	10	88.89	5	19.3	2.015	0.494	95
Thatcher	3	60	10.8	.	90	20	77.78	5	18.6	1.387	0.46	95

Thatcher	3	65	10.1	.	100	50	50.00	5	17.2	0.653	0.305	90
Thatcher	3	70	.	.	100	70	30.00	5	.	.	.	90
Thatcher	3	79	.	.	100	90	10.00	5	.	.	.	90
Thatcher	4	6	73.4	3.08	0	0	100.00	0	210.3	34.701	2.284	97
Thatcher	4	15	65.9	10.43	0	0	100.00	0	77.9	12.538	0.066	97
Thatcher	4	23	64.6	22.12	20	0	100.00	0.5	45.7	3.477	0.157	97
Thatcher	4	28	58.6	23.59	40	0	100.00	0.5	33	1.095	1.36	97
Thatcher	4	36	44.3	33.42	50	0	100.00	3	30.7	0.647	2.455	97
Thatcher	4	42	20.2	40.65	50	0	100.00	5	16.1	1.135	0.858	97
Thatcher	4	50	.	.	70	0	100.00	5	17.1	0.716	3.284	97
Thatcher	4	57	9.9	41.68	80	10	87.50	5	16.2	0.755	0.475	97
Thatcher	4	62	.	.	100	30	70.00	5	.	.	.	97
Thatcher	4	79	.	.	100	90	10.00	5	.	.	.	89
La Prevision	1	6	77.1	2.75	0	0	100.00	.	431.9	42.549	1.283	95
La Prevision	1	15	73.1	11.58	0	0	100.00	0	66.2	11.094	0.125	95
La Prevision	1	20	57.3	22.85	0	0	100.00	0	54.1	2.054	2.75	90
La Prevision	1	31	45.8	35.83	0	0	100.00	0.5	31.8	1.650	0.094	90
La Prevision	1	36	41.7	41.84	0	0	100.00	2.5	27.1	1.413	8.785	98
La Prevision	1	42	17.2	44.21	0	0	100.00	4	23.9	1.976	0.028	98
La Prevision	1	50	12	45.12	30	0	100.00	4.5	17.5	0.975	0.53	98
La Prevision	1	57	9.9	45.85	50	0	100.00	5	16.1	2.181	1.092	98
La Prevision	1	62	.	46.04	80	0	100.00	5	.	.	.	98

La Prevision	1	71	.	.		90	0	100.00	5	.	.	98
La Prevision	2	6	75.6	2.83	0	0	100.00	.	364.6	22.754	0.782	97
La Prevision	2	12	71.3	10.87	0	0	100.00	0	75.6	13.941	1.856	97
La Prevision	2	20	69	15.83	0	0	100.00	0	51.6	6.837	1.174	89
La Prevision	2	31	47.7	31.32	0	0	100.00	0.5	25.2	1.119	0.179	89
La Prevision	2	36	40.8	40.83	0	0	100.00	2.5	22.5	0.754	3.542	97
La Prevision	2	42	18.6	41.15	10	0	100.00	4.5	19.8	0.845	1.968	97
La Prevision	2	50	11.5	43.93	30	0	100.00	5	19.9	2.056	1.03	97
La Prevision	2	57	10.4	.	50	0	100.00	5	20.2	0.486	3.145	97
La Prevision	2	62	.	.	60	0	100.00	.	.	0.660	.	97
La Prevision	2	71	.	.	80	0	100.00	.	.	.		97
La Prevision	3	6	74.9	3.55	0	0	100.00	0	380.1	24.816	0.365	98
La Prevision	3	15	65.5	13.13	0	0	100.00	0	50.7	5.811	0.227	98
La Prevision	3	20	67.6	19.84	0	0	100.00	0	39.4	4.730	3.218	90
La Prevision	3	23	48.3	28	0	0	100.00	1.5	36.3	1.586	1.691	90
La Prevision	3	36	39.1	37.17	10	0	100.00	3.5	16.6	0.924	2.853	98
La Prevision	3	42	27	44.26	20	0	100.00	4	16.5	1.073	0.145	98
La Prevision	3	50	11.2	44.6	30	0	100.00	4	16.2	0.552	1.328	98
La Prevision	3	57	11	45.31	60	0	100.00	4	16.4	.		98
La Prevision	3	62	.	.	80	0	100.00	4	.	.		98
La Prevision	3	71	.	.	90	0	100.00	4	.	.		98
La Prevision	4	9	76.3	2.83	0	0	100.00	0	311.2	32.611	0.226	94

La Prevision	4	14	71.3	9.84	0	0	100.00	0	121.1	18.763	0.087	89
La Prevision	4	17	69.5	11.33	0	0	100.00	0	113.2	10.305	1.721	89
La Prevision	4	31	42.1	39.93	20	0	100.00	1	42.4	1.571	3.562	89
La Prevision	4	36	38.5	40.98	30	0	100.00	4	30.2	0.113	1.342	89
La Prevision	4	44	17.9	45.93	60	0	100.00	4	20.5	0.939	0.968	89
La Prevision	4	50		46.78	80	0	100.00	4.5	14.1		0.185	89
La Prevision	4	58	12.7		90	0	100.00	5	12.5		2.307	89
Hilgendorf 61	1	6	76.8	3.28	0	0	100.00	0	367.5	47.488	1.041	95
Hilgendorf 61	1	14	70.2	15.47	0	0	100.00	0	90.3	6.593	0.245	95
Hilgendorf 61	1	26	52.5	33.95	10	0	100.00	1	45.8	8.946	0.649	95
Hilgendorf 61	1	36	38.8	34.93	20	10	50.00	3.5	27.8	2.055	4.34	90
Hilgendorf 61	1	44	24.6	38.07	70	50	28.57	5	23.5	0.969	0.043	90
Hilgendorf 61	1	58	10.9	40.74	90	70	22.22	5	20.9	2.486	4.367	90
Hilgendorf 61	1	65	10.4	46.23	100	90	10.00		17.7	1.562	1.828	90
Hilgendorf 61	1	70			100	90	10.00			1.234	1.326	90
Hilgendorf 61	2	6	75.8	4.97	0	0	100.00	0	253.3	38.675	4.55	94
Hilgendorf 61	2	15	72.2	13.89	0	0	100.00	0	61.8	31.886	0.4	94
Hilgendorf 61	2	20	71.3	17.9	10	0	100.00	0	48.5	18.654	0.92	89
Hilgendorf 61	2	31	52.4	40.58	40	0	100.00	0.5	29.9	10.919	10.877	94
Hilgendorf 61	2	39	40	44.75	60	0	100.00	3	25.9	1.206	0.311	94
Hilgendorf 61	2	44	40.8	45.8	80	10	87.50	4	23.9	1.353	2.366	89
Hilgendorf 61	2	50	11.6	47.41	80	30	62.50	5	16.5	1.232	1.926	89

Hilgendorf 61	2	58		20 .		90	50	44.44	5	14.3	0.845	0.421	89
Hilgendorf 61	2	65		11.7	51.1	100	70	30.00	5	16.2	1.603	0.419	89
Hilgendorf 61	2	70		10.9 .		100	80	20.00	5 .		1.333	4.375	89
Hilgendorf 61	3	7		75.6	4.33	0	0	100.00	0	227.5	21.251	1.18	90
Hilgendorf 61	3	14		74.1	9.42	0	0	100.00	0	79.6	12.085	1.236	90
Hilgendorf 61	3	20		71	18.36	0	0	100.00	0	56.3	1.783	12.935	90
Hilgendorf 61	3	31		54.1	37.09	50	10	80.00	0.5	39.4	1.051	1.489	95
Hilgendorf 61	3	39		38.3	43.36	70	30	57.14	3.5	29.4	2.180	0.714	95
Hilgendorf 61	3	45		26.5	45.58	80	40	50.00	5	16	1.447	0.113	95
Hilgendorf 61	3	50		11.5	44.02	80	50	37.50	5	15.9	0.972	0.244	90
Hilgendorf 61	3	58			44.07	100	50	50.00	5	14.3	1.051	0.296	90
Hilgendorf 61	3	65		11.1	45.17	100	70	30.00	5	16.1		0.6	90
Hilgendorf 61	3	70		10.7	46.81	100	70	30.00	5 .		.		90
Hilgendorf 61	4	6		76.7	3.15	0	0	100.00	0	470.2	60.177	0.005	94
Hilgendorf 61	4	14		73.9	10.26	0	0	100.00	0	111.5	22.006	0.356	89
Hilgendorf 61	4	20		70.6	16.06	0	0	100.00	0	48.6	16.318	6.36	89
Hilgendorf 61	4	36		31.6	35.21	70	10	85.71	4	45.6	1.850	0.816	89
Hilgendorf 61	4	53		23.5	38.51	100	20	80.00	5	23.4	0.905	1.091	94
Hilgendorf 61	4	65		10.6	38.68	100	20	80.00	5	17.3	15.605	2.803	89
Hilgendorf 61	4	70		9.6 .		100	70	30.00	5 .		.		89
Hilgendorf 61	4	79	.	.		100	80	20.00	5 .		.		89
Karamu	1	6		76.6	3.02	0	0	100.00	0	420.7	37.005	24.951	95

Karamu	1	14	73.7	7.95	0	0	100.00	0	109.6	21.407	0.095	90
Karamu	1	20	64.9	14.95	0	0	100.00	1.5	61.6	10.574	0.747	90
Karamu	1	27	58.5	28.11	0	0	100.00	1.5	30.2	2.411	0.375	86
Karamu	1	36	37	35.57	60	10	83.33	4	25.2	0.502	0.014	90
Karamu	1	44	32		80	30	62.50	5	14.4	0.951	1.07	90
Karamu	1	50	10.6	39.41	100	60	40.00	5		0.816	1.14	90
Karamu	1	58	9.8	40.5	100	80	20.00	5	16	1.755	0.114	90
Karamu	1	65		46.53	100	90	10.00	5				95
Karamu	1	74			100	90	10.00	5				95
Karamu	2	6	76	2.87	0	0	100.00	0	482.3	54.611	16.834	94
Karamu	2	14	72	9.3	0	0	100.00	0	113.1	18.067	0.087	89
Karamu	2	20	64.6	15.28	0	0	100.00	1.5	48.7	8.525	0.964	89
Karamu	2	31	49.8	29.14	10	0	100.00	2	22.4	1.363	0.375	94
Karamu	2	39	42.3	42.08	50	0	100.00	3	20.4	1.420	0.445	94
Karamu	2	45	35.7	49.91	60	10	83.33	4	19.3	0.834	0.089	94
Karamu	2	50	10.4	49.91	80	30	62.50	4.5	15.1	1.071	0.494	89
Karamu	2	58	20.6	48.69	100	60	40.00	5	13.1	0.818	0.813	89
Karamu	2	69	10.5	52.16	100	80	20.00	5	12.4	0.823	0.127	85
Karamu	2	74	10.9	49.72	100	90	10.00	5				85
Karamu	3	6	76.5	2.44	0	0	100.00	0	496.6	28.814	3.12	98
Karamu	3	15	67.7	12.98	0	0	100.00	0	118.7	15.449	0.134	98
Karamu	3	20	67.5	13.63	0	0	100.00	1.5	55.6	10.220	2.295	90

Karamu	3	31	49.6	33.58	10	0	100.00	2	26.8	2.423	2.556	90
Karamu	3	36	44.4	43.97	50	20	60.00	4	24	0.918	1.79	90
Karamu	3	44	37.8	46.05	90	40	55.56	4	23.8	0.478	0.315	90
Karamu	3	50	8.8	46.59	100	60	40.00	5	15.4	1.104	0.475	90
Karamu	3	58	.	.	100	80	20.00	5	13.2	0.351	0.238	90
Karamu	3	65	.	.	100	90	10.00	5	12.5	1.156	0.601	90
Karamu	4	7	76.6	2.6	0	0	100.00	0	411.8	64.738	4.2	89
Karamu	4	14	73.5	7.22	0	0	100.00	0	113.2	31.344	1.088	89
Karamu	4	20	63	21.89	0	0	100.00	1.5	44.9	8.052	7.005	89
Karamu	4	27	53.8	33.87	10	0	100.00	2	24.7	0.257	0.677	85
Karamu	4	31	43.4	38.06	30	0	100.00	3.5	24.1	0.387	1.332	89
Karamu	4	44	32.4	44.29	80	30	62.50	5	14.8	1.035	1.157	89
Karamu	4	50	21.4	44.84	90	70	22.22	5	12.7	0.207	2.192	89
Karamu	4	58	10.3	47.79	90	80	11.11	5	14.7	1.058	0.629	89
Karamu	4	69	9.9 .		100	90	10.00	5 .		.		89
Karamu	4	74	.	48.23	100	90	10.00	5 .		.		89

Key:

dap:	Days after pollination	flavs:	Flavanol concentration (ug/g)
mstur :	Grain moisture content (%)	amy:	Alpha-amylase activity (EU/g)
gnwt:	Grain mass (mg)	aba:	Abscisic acid activity (ng/grain)
sg:	Special germination (%)	DTF:	Days from sowing to pollination
ng:	Normal germination (%)	Col:	Colour score
dorm:	Dormancy (%)		