THE PELLETING OF CLOVERS AND FERTILISER

The effects of the localized placement of fertiliser at seeding (pelleting) on germination, morphology and herbage yield of *Trifolium* species.

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by

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INTRODUCTION

"Clovers are essential to justify topdressing - subterranean clover, *Lotus major* and certified white clover are the great triumvirate of the hills." - - - - - E.B. Levy (1) *

The pelleting of clover seed and fertiliser may be valuabl as a method of introducing the essential clovers into the unploughable hill country.

The hill land of New Zealand is agriculturally important because of the large area involved, the animals and animal products produced and its value as a source of direct and indi employment. The hill country indirectly influences the valuab lowlands. There are two aspects; firstly, the supply of store stock for fattening and breeding (which results in a continuou transfer of minerals as animal skeletons); and secondly the importance of the hill catchment areas in the prevention of flood discharges, and the silting of rivers, flats and resevoi lower down.

The problem of much of the hill country is essentially on of low or declining stock carrying capacity, either actual or incipient. Two visible indications of this decline in fertilit are deterioration and reversion. Deterioration is the thinnin out of the sown pasture to a low fertility species, dominant sward of browntop (*Agrostis tenuis*) and danthonia (*Danthonia* s

* Figures in parenthesis refer to literature cited in the li of references.

Reversion is the gradual encroachment of fern (*Pteridium esculent*
and scrub (*Leptospermum scoparium*).

This need to improve the hill country is the purpose behind these trials studying the pelleting of clover seed and fertiliser

HILL COUNTRY ECOLOGY

Madden (2) and Grange (3) agree in their estimates of approximately $10\frac{1}{2}$ - 11 million acres of pastoral North Island hill country. Most of this area has a carrying capacity of 1-3 sheep per acre plus cattle. The sward consists of low producing specie stunted browntop, crested dogstail (*Cyanosarus cristatus*), sweet vernal (*Anthoxanthum odoratum*), much moss on the shady slopes and numerous flat weeds with a dearth of clovers. The clovers that a present are mainly low producing, annual clovers such as suckling clover (*Trifolium dubium*), clustered clover (*Trifolium glomeratum* striated clover (*Trifolium striatum*) and haresfoot trefoil (*Trifolium arvense*). The turf is open and is insufficiently dens and vigorous to prevent seedlings of weed and scrub growth establishing. There is often much bare ground exposed to wind an runoff water. (See Figure 1 for distribution of the North Island hill country).

Original sub-tropical rain forest (in the North Island) has given place to a stock-induced, mesophytic grassland.

The felling of the bush commenced with the earliest settleme The felled trees and undergrowth were burned and English seed mixtures sown, often on the still-warm ashes. Frequently the see mixtures thus sown on the earlier burns were not suitable. High

FIGURE 1 - Distribution of North Island hill country. (After N.Z. Royal Commission on the Sheep Industry, 1949)

is one of the most remarkable peculiarities of the N.Z. flora" (5) - and these are unsuitable for pastoral uses, mainly being shrubs.

The first phase in improving the hill country then, is to establish clovers in the pasture, mainly through surface sowing. Topdressing by air after the clovers are established has been shown (8) to be feasible and practicable, but phosphate application to many of the hill country pastures where clovers are missing would be futile waste.

The results from oversowing trials with clovers have been variable. Success of establishment depends on numerous factors seed sown, season and time of sowing, length and density of swa etc. and it is important that the grazing management consolidate this pasture improvement.

White clover establishment in the oversowing trials varied at different localities from 1-50% of seeds sown (6,7). The figures for subterranean clover were higher. "In all cases, establishment compared with number of seeds sown is extremely poor." (7) One objective in the pelleting of seed and fertilis was to find out whether this method could result in increased establishment of the sown clovers.

The problem of maintaining a permanent farming economy on hill country is linked with many other agricultural and economic problems. The progressive deterioration of several million acre of hill country has resulted in reversion to fern and scrub and has, in many catchments, led to the onset of soil erosion and flood problems. The initial step in improving the hill country is to establish clovers in the sward by oversowing. Topdressi with phosphates, together with efficient utilisation of the fe produced, and the fertility response from increased stocking, the key to restorative treatment designed to raise the fertili of the hill country, maintain a balanced pastoral economy and help fulfil the pledge held out through F.A.O. of freedom from want for all.

PELLETING OF SEED

The need to introduce clover in hill country pastures has been demonstrated. This work is a study of the pelleting of se and fertiliser to discover its possible future value in over-sowing hill country. The use of pelleted seed in aerial sowing has been kept in mind.

Pelleting of seed with fertiliser or inert materials is still new. In the U.S.A. this method has been practised with vegetable seeds which have been combined with materials such as fungicides and insecticides. Little work has been done with the pelleting of seed and fertilisers.

The advantages of pelleting seed and fertiliser were thougl to be economy in seeding rates, earlier and better establishment of seedlings, more efficient use of fertiliser (placement) and greater ease in aerial sowing (ballistics).

Disadvantages would be the technical costs of pelleting, germination injury due to toxicity of fertiliser in close proximity to seed and possible "burn" of the seedling roots.

No critical work evaluating this technique of pelleting has been published in New Zealand and it has not been possible to find overseas work dealing with the basic questions involved. This was the main objective of the present research: to determi the efficiency of the pelleting of fertiliser as compared with the standard method of broadcasting under experimental conditio Secondly, the aim was to try to find the reasons for the superi ity of pelleting, i.e. a study of the plans physiological respo This would seem to be the logical approach.

The major portion of the investigation involved trials measuring the yield of different clover species under the several treatments. Preliminary work of a trial and error nature was needed to find the maximum quantity of fertiliser which could be pelleted with a clover seed without causing germination injury.

ORGANISATION OF PRESENTATION

The work carried out falls naturally into three sections

1. The preliminary investigation into the method of pelleting the quantities of fertiliser permissible per pellet and germination tests.
2. Pot trials comparing pelleting, broadcasting and a combination treatment with half the fertiliser pelleted and half broadcast. These treatments were applied to several speci using different fertilisers at two levels.
3. Root studies, involving the removal of plants for detailed laboratory study of the root system, root/shoot ratio and degree of nodulation.

For ease and clarity of presentation this thesis has been sub-divided into sections corresponding with the above organisation.

TIME AND PLACE OF EXPERIMENT

This work was carried out at the Grasslands Division Station, Department of Scientific and Industrial Research, Fitzherbert West, Palmerston North. The preliminary work bega in December 1949 and the last yield cuts were taken on 15th December, 1950.

SECTION A

THE PRELIMINARY INVESTIGATION INTO THE METHOD OF PELLETING AND GERMINATION TESTS.

CHAPTER II

REVIEW OF LITERATURE

The literature covering pelleting, fertiliser injury and aerial sowing has been reviewed. A major consideration in the localized concentration of seed and fertiliser is the danger o germination injury. In the U.S.A. aerial sowing is one advantage of pelleted seed and would be of practical importanc in New Zealand.

PELLETING OF SEED AND FERTILISER

A coating was first put on vegetable seeds in the U.S.A. for ease of handling and precision planting; later nutrient an protective materials were added. Pelleted seeds have been use in aerial sowing for re-afforestation, and in erosion control projects. Both successes and failures have been experienced in the U.S.A. The coating of seed with fertiliser and protective substances is not easy. Various methods of pelleting are in use.

Saxby (9) lists the Tumbler, Extrusion, Compression and Continuous-belt methods. One process (10) involves seed tumbling in a rotating drum with the coating material until an approximately spherical pellet of the desired size is produced One commercial firm uses a water slurry of montmorillonite clay; another applies alternate layers of methyl-cellulose and a mixture of feldspar and ash. The moist pellets are graded to a size tolerance of 1/32 inch and dried. Several different additives can be incorporated into the pellet - fertiliser, plant hormones and fungicides.

A portable pelleting machine which is operated in the location where the seed is to be sown has been developed (11). Soil mixed with fertiliser, rodenticides etc. is used for the pelleting material. The prepared seed is sown by air. The Do Chemical Company has experimented with the pelleting of grass and clover seed in humid coastal Oregon (12). Their aims are -

(a) To cut the seeding rates of expensive seed, and

(b) To supply a better environment for germination.

Their pellets included a mercuric organic, plus 2% triple phosphate and pH correctors. Since all coated seed is made to order it is possible to adjust the coating for specific soil conditions. The application of fungicides in seed coatings has been especially effective. Although no protection is given th resultant plant by this treatment, the seed and subsequent root are effectively guarded for periods up to two months (10).

Fertiliser is commonly added. The inclusion of superphosphate in the coating equal to 10% of the weight of the seed gives a fertiliser concentration equal to 500 lbs. of superphosphate per acre in the cubic inch around the seed. This supply is rapidly exhausted but exerts a beneficial effect on the early growth and maturity rates. Care needs to be taken to prevent a concentration of soluble fertiliser salts around t seed sufficient to cause germination injury.

The advantages claimed for this method are;- improved germination, more efficient use of fertiliser, better establishment of plants, reduction in rate of seeding.

Saxby (9) summarizes the factors determining the physical nature of the pellets and concludes that the pellets should be shock and abrasion resistant, the fertiliser used must be commercially available and germination must not be impaired. New Zealand trials have not proved that pelleting is better or worse than broadcast seed. Subterranean clover pelleted gave much better results than white clover (9).

FERTILISER INJURY

The concentration of superphosphate immediately surroundi the clover seed led to some germination injury. There are two ways in which localized salt concentration may affect germinat

(a) There may be enough soluble salt in the seed bed to buil up the osmotic pressure of the soil solution to a point which will retard or prevent intake of necessary water.

(b) Certain constituent salts or ions may be toxic to the embryo or seedling.

Germination is retarded or inhibited by the presence of soluble salts in the soil and this effect is related primarily to the osmotic pressure of the soil solution (13, 14). As osmotic pressure increases, rate and per cent germination decrease (15).

There is evidence that certain salts or ions may be toxic to the embryo or seedling if occurring in sufficiently high concentrations. This toxicity may cause reduced germination and is frequently accompanied by abnormalities in the growth and development of the seedlings.

Various workers indicate that species and varieties of plants exhibit varying degrees of tolerance with respect to germination and seedling growth; cereals as a group being more tolerant of salt than the legumes (16, 17, 18). Stewart's (16) order of tolerance for legumes was peas, red clover, lucerne and white clover in descending order.

The fertiliser effect varies with the soil moisture conte[nt]. High rates of application of soluble salts produced no germination injury of soy beans when the moisture content of the soi[l] was high, but the same rate of application on drier soils caused germination injury.

Collings (19) quotes unpublished work of his and stated that although good germination of soy beans may be secured wit[h] high rates of application of fertiliser salts, the roots of th[e] seedlings soon become stunted and injured, probably resulting from plasmolysis induced by the high concentration of the soil solution. Root burn may result from lower fertiliser concentration than is capable of producing serious germination injury.

Collings cites old literature showing superphosphate les[s] injurious when in contact with seed than the common carriers of Nitrogen and Potash. Injury from phosphates, sulphates, chlorides and nitrates increases in order (20).

The literature deals mainly with plant growth in respect to saline and alkalie soils. No reference has been found dealing with concentration of superphosphate in the substrate and the effect upon the germination and development of clover seedlings.

AERIAL SOWING

The use of the aeroplane as a modern implement of the hill has been eagerly accepted by labour-hungry farmers. Trials (8) however, have shown that satisfactory distribution of the individual components of a mixture of grass and clover seeds is not obtained in aerial sowing, owing to the differing ballistic properties of the seeds. The different densities, sizes and shapes of clover seed are reflected in a band distribution (21)

One advantage of pelleting seed and fertiliser is the satisfactory aerial spread of pellets of uniform size and shape

American workers (22, 23) concluded that the distribution of grass seed sown by aeroplane was unsatisfactory and suggest that pelleted seed may offer a solution. Only one report (24) on the subsequent establishment and growth of pelleted seed has been seen. The pellets contained various species of grass and legume seed combined with a mixture of clay, rodent-insect repellant and growth promoting chemicals. The number of seeds per pellet varied from 2 to 59. Pelleting affected the viabili of the seed to some extent; and in the case of *Agropyron cristatum*, germination dropped from 80% to 15% after pelleting.

The results of this aeroplane pellet seeding on Indian reservations was not satisfactory. There seemed several reasons - the areas sown varied greatly from desert scrub to mountain forest; there was not the proper management of grazing that appears essential; and a better distribution of seed within the pellets would have been desirable.

CHAPTER III

THE EXPERIMENT, OBJECTIVES, MATERIALS AND METHOD OF PELLETING

The preliminary investigation was of a trial and error nature to determine the limits within which fertiliser and seed could safely be incorporated.

OBJECTIVES

The first need was to find the maximum amounts of fertilis which could be pelleted with clover seeds without seriously impairing germination. American figures quoted (10) of amount of fertiliser used were too small to supply any appreciable par of the plant's fertiliser requirements. One possible advantag of pelleting was kept in mind, i.e. the supplying of the clover seedlings phosphate requirement for the initial year in pellete form. This would result in economy of fertiliser, labour etc.

A second objective was the determination of the extent of root injury and deformation of emerged seedlings as contrasted with complete germination failure.

The third aim was to approximately evaluate the physical characteristics of the pellets and the effect of the different fertilisers on the physical nature of the pellets, and to try t amplify field observations.

MATERIALS

<u>Seed</u>: Government Stock white clover seed of 99% purity and (95 ± 2)% germination was used throughout the germination and

pelleting tests. This same line of seed was used in the pot trials and root studies.

Fertiliser: The fertilisers and materials used were -

(a) "Elephant" Sulphate of Ammonia, guaranteed minimum analysi of 21% Nitrogen.

(b) "K.P." Superphosphate, 44/46 from the Wanganui works.

(c) Finely ground Bentonite, ex Plant Chemistry Laboratory stores (records lost).

The superphosphate and sulphate of ammonia were finely gro dried in electric ovens and kept for use in dessicators. A reserve of the ground fertilisers was placed in store in case o future needs.

These same fertilisers were used throughout the experiment period.

Pellets: The pellet sizes varied with the different levels of fertiliser used. The two sizes most used were 0.01 gram of fertiliser per seed which resulted in a pellet approximately 0. centimeter diameter and 0.1 centimeter thick, and 0.02 gram o fertiliser per seed, which compacted to form a pellet approxima ly 0.4 centimeter diameter and 0.1 centimeter thick.

These pellets resembled small flattened tablets (see Figur 2).

PELLETING

Each pellet was made by hand to ensure accurate control of the quantity of fertiliser used. The required amounts of fertiliser were weighed out and an allowance made for the small losses which occur in the pelleting process. The initial

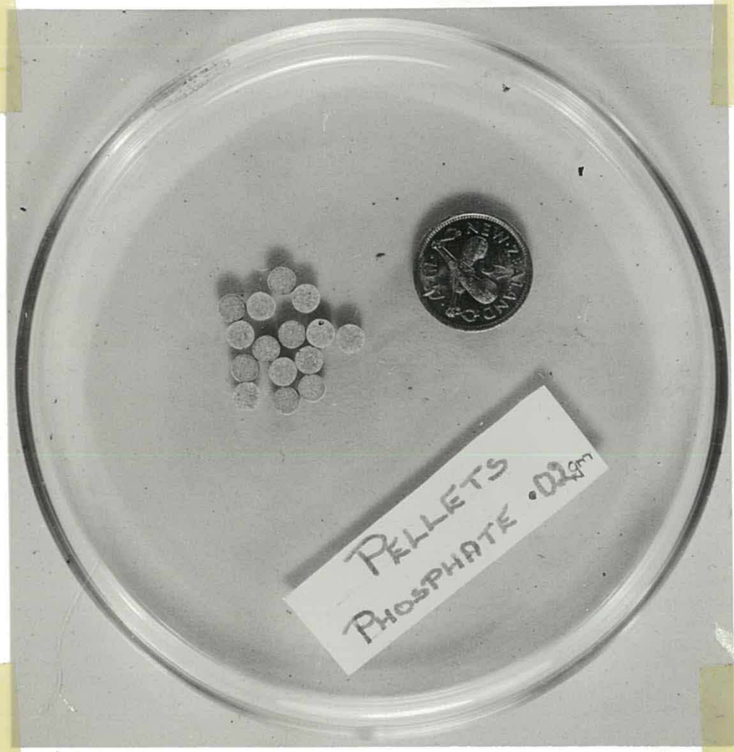


FIGURE 2. Illustrates size of pellets compared with N.Z. threepenny piece.

quantity of fertiliser required to form a pellet of a certain weight was found by trial and error.

Half inch bolts were drilled in a lathe to form steel cylinders of $\frac{1}{8}$, 3/15 and 1/10 inch diameters respectively. Lengths of spring steel of these diameters were cut to form closely fitting plungers or rams. The heads of the bolts were ground to form a plane surface and the thread end cut off to giv a height of 1 - $1\frac{1}{2}$ inches. This new end was counter sunk to receive the end of a glass funnel. The rest of the pelleting equipment consisted of a flat steel base-plate, light hammer and a glass funnel.

The pellets were formed by pressure. The size of bolt requ ed was selected, eg. $\frac{1}{8}$ inch diameter bolt for making the 0.01 gr pellets and the larger bolts for the bigger pellets. The drille bolt was set up on the base plate and the weighed fertiliser poured into the bolt. The glass funnel was removed and the clos fitting plunger inserted and given a sharp tap with the hammer. This was sufficient to compress the fertiliser - clay mixture into a hard pellet. These pellets were placed in labelled containers. (See figure 3 for apparatus used in pelleting)

This was the case when making pellets of fertiliser alone. When making pellets containing an enclosed clover seed, half the fertiliser mix was poured into the bolt, then the seed was dropped down followed by the remainder of the fertiliser. The pellet was then compacted as before. It is readily realised tha the above method, although permitting rigid control of the finished pellets, is slow and laborious. (ten pellets take fourteen minutes to make)

FIGURE 3. Equipment used in making pellets. From left to right - glass funnel set in drilled bolt standing on base plate, weighed fertiliser, drilled bolt and plunger of a different size, tweezers, metal spoon and hammer.

Chapter IV

GERMINATION TESTS AND STUDIES OF THE PHYSICAL CHARACTERISTICS OF THE PELLETS

Germination tests: To determine the limits within which seed and fertiliser could safely be incorporated, P, N and PN* pellets were tested for viability at differing levels of the fertilisers.

The germination tests were carried out in a hotwater bath germinator with alternating temperature. The pellets were cou ed out in lots of ten and twenty and placed on damp filter pap in 4 inch glass petri dishes. Moist cotton wool was placed und the filter-paper to act as a resevoir and prevent drying.

The emerged seedliegs were counted daily and in certain cases they were examined under a binocular microscope to see i radicle development was normal or whether the seedling had suffered in any degree from fertiliser injury.

After about twelve days any germinated pellets were opene and examined under the binocular microscope to try to determin the cause of germination failure.

* Symbol abbreviations used in this work are -

P = superphosphate

N = sulphate of ammonia

PN = a 50 : 50 mix of superphosphate and sulphate of ammonia.

These remain constant throughout the thesis. Pellets of these fertilisers contain an equal weight of bentonite clay.

Preliminary tests showed little difference between the germination of the different clovers pelleted, i.e. red, white and subterranean clovers. White clover was then used as the standard clover in germination trials. Germination results (o white clover) are set out in the following table.

TABLE I. RESULTS FROM GERMINATION TESTS

(The figures are the average of duplicate lots of 20 and concer only seedlings with normal cotyledons and radicle)

% Germination

Fert.	Levels gms/ seed	Days from start of test													
		1	2	3	4	5	6	7	8	9	10	11	12	13	14
Control	0	-	-	-	-	69	91	95							
	.01	-	-	40	70		80								
P	.02	-	-	-	-	-	20	50	75					90	90
	.1													20	20
	.02					40	50	(i) 60							
PN	.05						10						(ii) 50		
	.25													40	40
N	.02						30								

Key: (i) Ungerminated pellets opened and seeds dissected - two seeds later germinated, one with deformed radicle. (Batch of 20 pellets)

(ii) Ungerminated pelleted seed when dissected failed to germinate.

There is a general decrease in viability and a rise in toxicity from pelleted P, PN to N. In the same way there is a lowering of viability accompanied by a retarding of germinatio as the quantities per pellet of any one fertiliser increase. (Fig. 4 illustrates these trends)

Examination of some of the ungerminated pellets in the tes showed the seeds split and soft with decay. The splitting of t seed coat during pelleting was thought to be a possible cause of germination failure. (The seed and fertiliser are compacted a hammer tap on a movable plunger) Newly made pellets were broken and the seeds inspected under the low power microscope showed cracks in the seed coat. These damaged seeds failed to germinate and it is concluded that the pelleting accounts for some of the germination failure. Probably toxic fertiliser solutes are able to penetrate the cracks in the seed coat and injure the sensitive embryo.

A further test was made with pellets formed with the minim compaction. Ten pellets each of P, N and PN 0.02 gm./seed were broken and the seeds dissected out. All the seeds were sound a appeared free from seed coat cracks under the microscope. These seeds were tested for germination. After 4 days the counts were

P - 50% germinated
PN - 100% "
N - 90% "

Notes taken after nine days record that the P pellets comprise (from a batch of 10) :

2 Mouldy pellets
1 Seedling with damaged radicle
2 Seedlings lacking cotyledons and having thin radicles
5 Seedlings germinated normally.

The N pellets comprised :

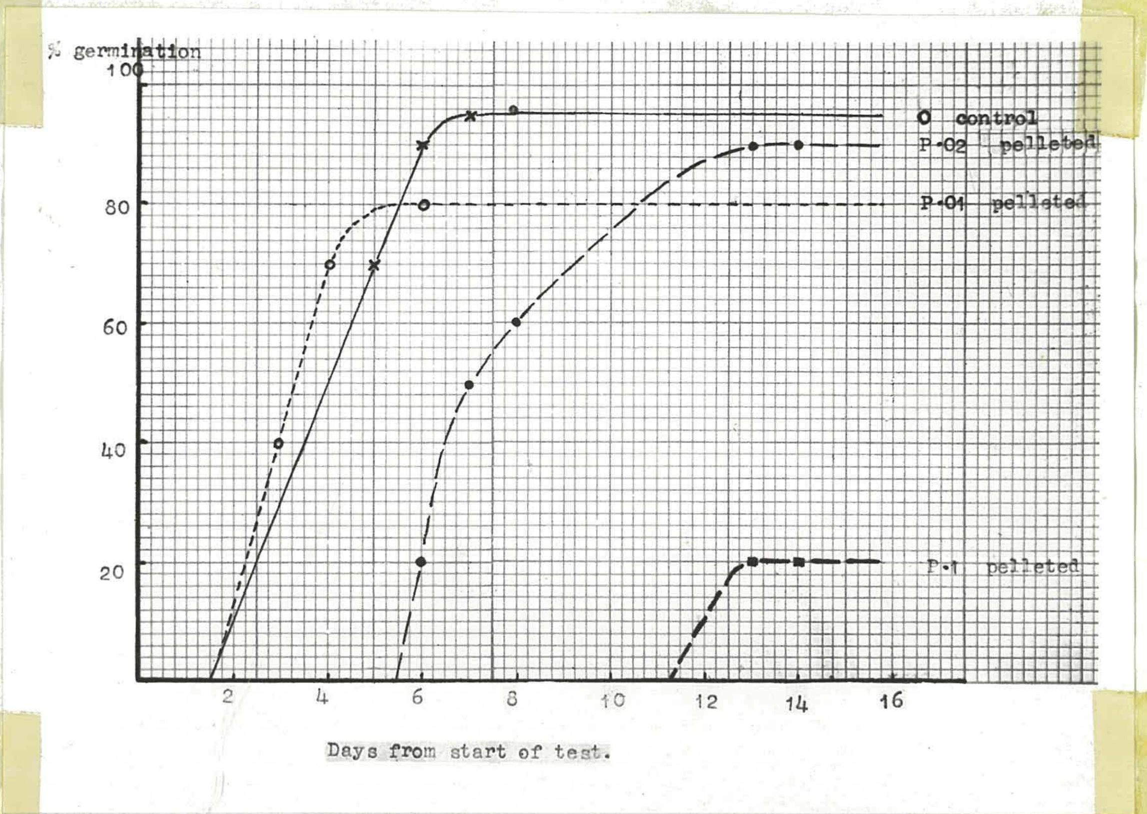


FIGURE 4. Germination tests - Pelleted seed, P series.

P.01 = 0.01 gms. of superphosphate pelleted per seed

P.02 = 0.02 " " " " " "

P.1 = 0.1 " " " " " "

Note: (i) decrease in germination as amount of fertiliser per seed increases.

(ii) delay in germination with increase in amount of fertiliser pelleted.

1 seedling with stunted radicle
9 seedlings germinated normally

Apparently the method of pelleting, in the pelleted seeds, caused some unobservable damage.

The proportion of germination losses (when germinated on warm, moist filter paper) was approximately:

Control 0 fertiliser - 6% non germinated, 2% radicle injury

PN pelleted 0.02 gm/seed - 20-30% non germinated, 10% radicle injury

A series of germination tests, 15 months later than those in Table I, gave lower germination counts:

P	pelleted 0.02 gm/seed	- 35% germination
PN	" " "	- 15% "
N	" " "	- 10% "

Some unknown factor affecting germination is operating. The pellets became covered with mould after 17 days. Black, white, red and green fungal mycelium covered the bottom of the petri dishes. Delay in germination may have allowed the growth of saprophytic fungi. Fungal growth associated with delayed germination had been noticed before. Ungerminated pellets were broken open for inspection to try to determine the cause of non-germination. It was decided that fertiliser injury and the retarding of germination allowing extensive fungal growth, were the probable explanations of the germination failure. (See Figs. 5 and 6 for illustrations of dissected seedlings)

STUDIES OF THE PHYSICAL CHARACTERISTICS OF THE PELLETS

The object was to approximately evaluate the solubility and

PELLETED PN. FERTILISER GERMINATION INJURY.

(x 25 approx.)

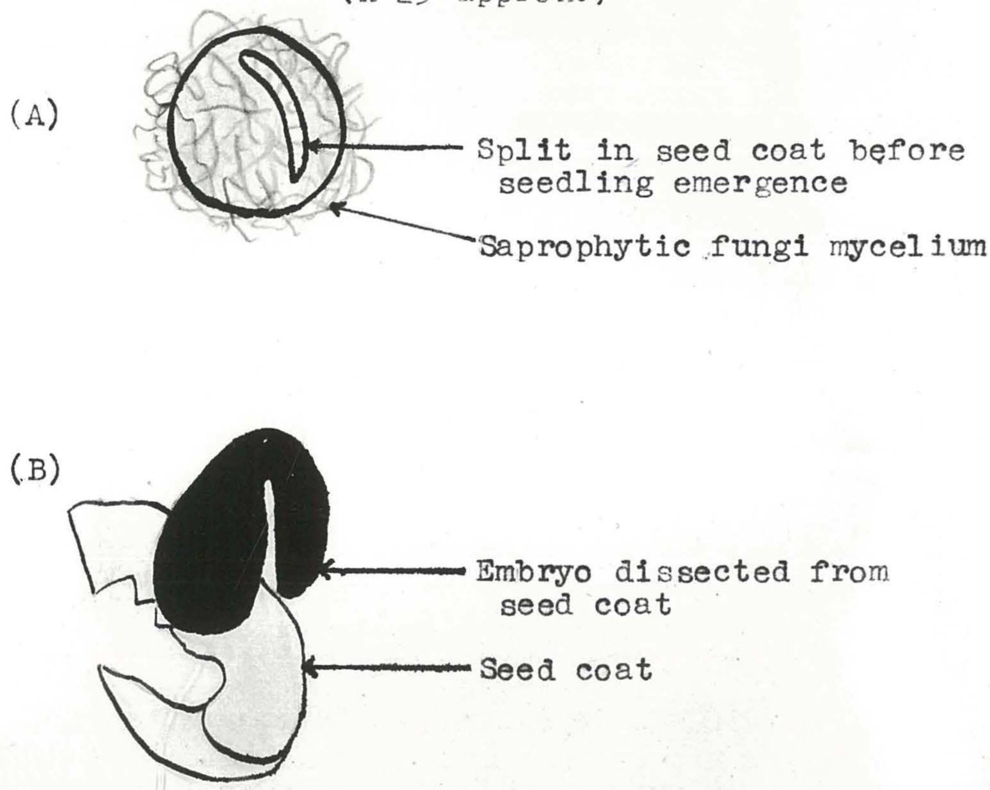


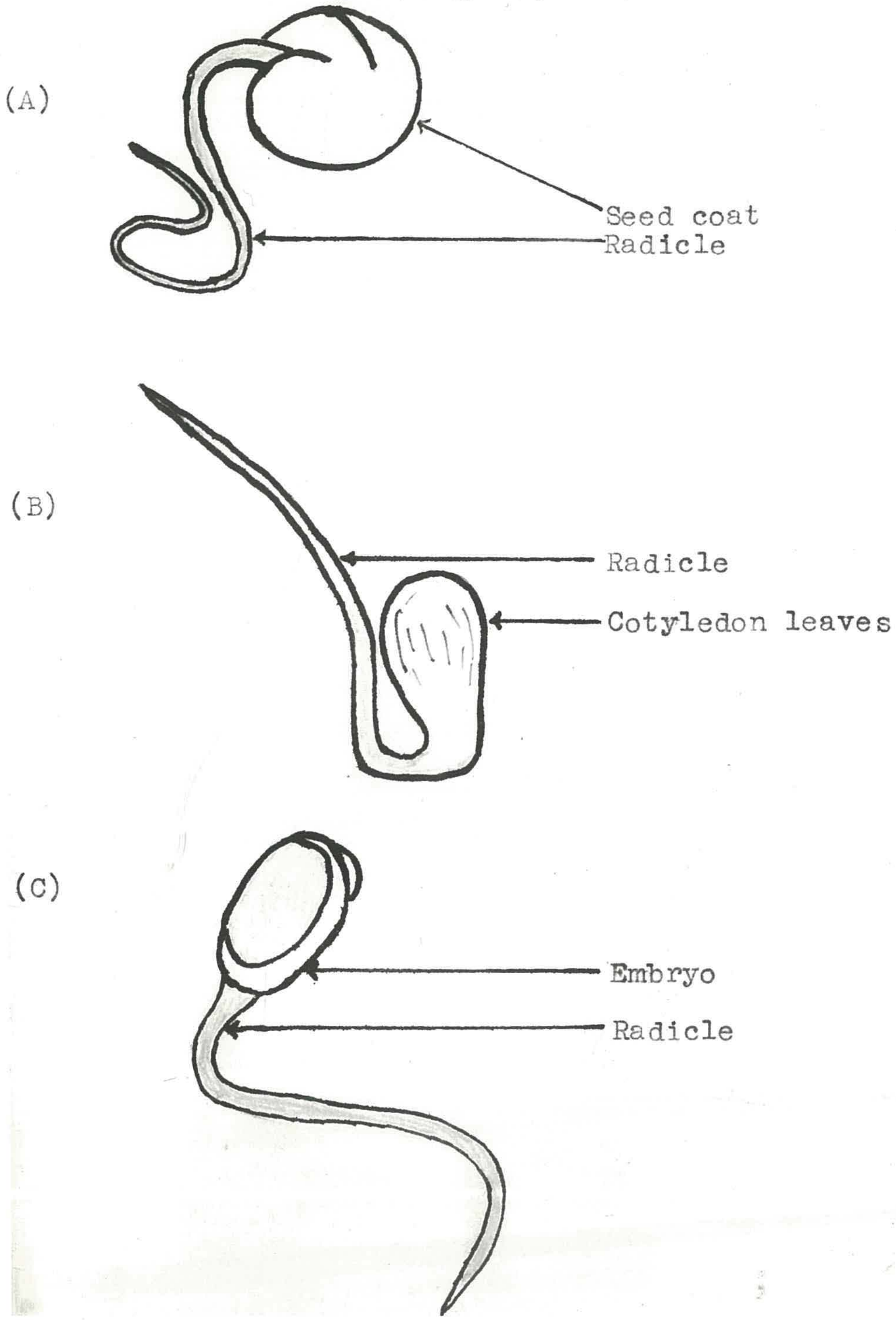
FIGURE 5. Dissection of seed from ungerminated PN pellets, 0.02 gm.fertiliser/seed, after 3 weeks.

A. Ungerminated seed covered with saprophytic fungal mycelium.

B. Dissected embryo

PELLETED P. FERTILISER GERMINATION INJURY.

(x 25 approx.)



<u>FIGURE 6</u>. Dissection of seed from ungerminated P pellets, 0.02 gm. fertiliser/seed, after 3 weeks.

A. Seed germinating after 3 weeks. Note thin radicle

B. Seed coat removed from swollen seed.

C. Most of the dissected seeds contained an embryo with an extended stability of the pellets formed from the different fertilisers, and obtain figures which would confirm field observations. I h been noted that the P pellets persisted longer and appeared les soluble under weathering than the PN and N pellets.

Solubility studies -

The aim was to get a measure of the water soluble constitu in the P, N and PN pellets.

Two variations of water extraction were used:

(i) Stationary - in which ten 0.075 gm. pellets of P, N and PN were placed in fibre extraction thimbles and immersed in 750 ml. beakers of water.

(ii) Running extraction-in which the same quantity of pellets we placed in extraction thimbles in Soxhlet extractors. Tap water constantly dripped through the apparatus and automatically siphoned away at intervals.

The pellets remained in the beaker of water from the 25th January 1951 to 29th January 1951. The running extraction apparatus was in operation from 26th January 1951 to 29th Janua 1951. The thimbles containing the pellets were then removed, allowed to drain, dried in an electric oven and re-weighed.

Table II gives the results from solubility studies with th two methods of water extraction.

TABLE II. RESULTS FROM SOLUBILITY TESTS.

Method	Fert.	Wt.thimble (gms.)	Wt.pellets (gms.)	Wt.pellets after extraction (gms)
Stationary extraction (4 days)	P	1.0593	1.4877	1.3675
	N	1.0015	1.4972	0.7821
	PN	0.9899	1.5143	1.0929
Running extraction (3 days)	P	1.0567	1.5000	0.9043
	N	1.0578	1.5012	0.6978
	PN	0.9603	1.5018	0.7472

Allowing for the 50% insoluble bentonite content of the pellets, the solubility losses of the fertiliser constituents a as follows:-

		% solubility loss (of fert.)
Stationary extraction	P	16
	N	96
	PN	55
Running extraction	P	72
	N	100
	PN	93

The percentage solubility losses of the fertilisers follow the order expected. The running extraction method, as compared with the stationary, caused a large increase in the P fertilise loss. This loss would presumably be the water soluble phosphat together with most of the gypsum.

Stability tests -

Pellets in the pot trials and root studies had been noted as lasting and visible fourteen days after sowing. It was thought that the N pellets, of soluble ammonium sulphate, would readily absorb soil moisture and break up, or would disintegrat during rain. (This had been noted in the pot trials.)

A simple apparatus was set up to gauge the relative stabilities of the P, N and PN pellets. The pellets were place on thin strips of blotting paper, one end of which dipped into a petri dish of water. Metal rods, 1½ inches long and 0.12 inches diameter, were set on top of each pellet. (See figure 7

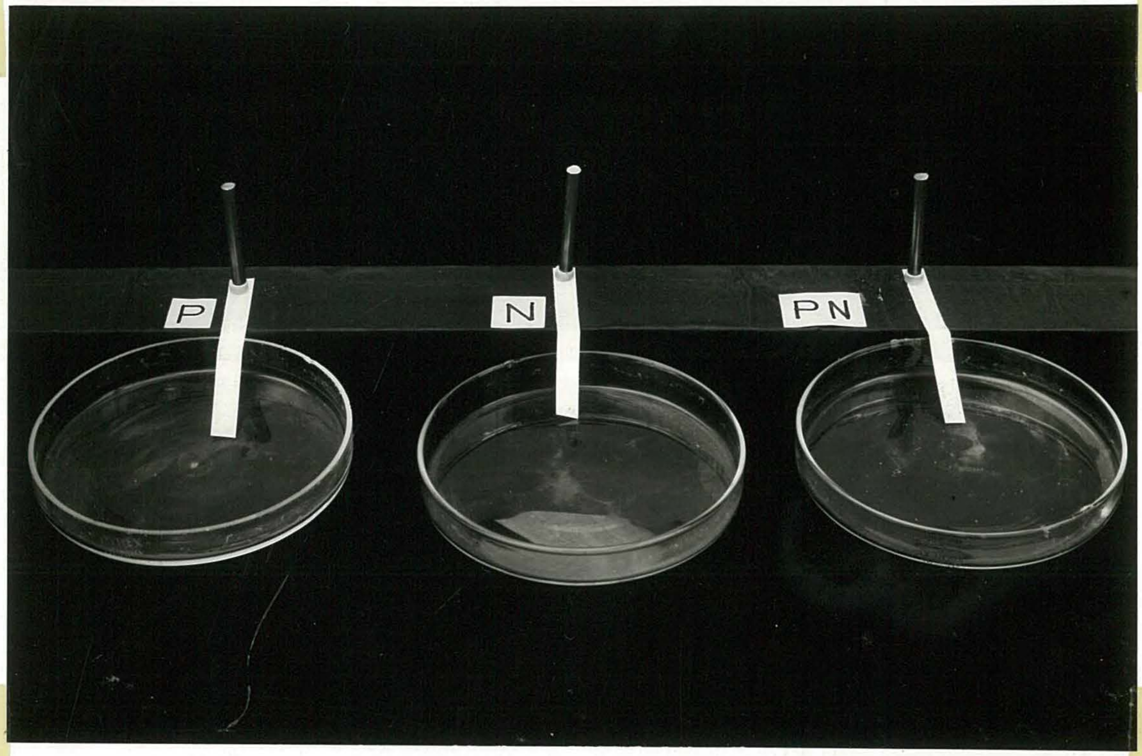


FIGURE 7. Layout for stability tests: metal indicator rod upright on top of pellet; water absorbed through blotting paper wick.

for layout of apparatus)

The pellets absorbed water from the moist blotting paper wicks. When sufficient water had been absorbed the pellets swelled and crumbled and the metal rods fell. The times taken by the pellets to dislodge their indicator rods were recorded. Ten runs were timed. The results are recorded in Table III.

TABLE III. STABILITY TEST

Times of fall of indicator rods. (Times taken from the first rod to fall in each run)

N	PN	P
0	1 min.35 sec.	3 min. 35 sec.
1 min.15 sec.	45 sec.	0
2 min. 7 sec.	0	50 sec.
0	2 min. 40 sec.	6 min. 30 sec.
2 min. 50 sec.	0	1 min. 55 sec.
0	1 min.	5 min. 15 sec.
o	3 min. 40 sec.	9 min. 30 sec.
0	1 min. 30 sec.	3 min. 30 sec.
0	20 sec.	3 min. 40 sec.
1 min. 5 sec.	0	1 min. 40 sec.

phosphates in the surface layers of the soil. (27, 28)

Two methods for improving the efficiency of phosphate utilisation are:

(i) Placement of fertiliser in special positions, and

(ii) Granulating the superphosphate.

These two methods are combined in the practice of pelletin

PLACEMENT OF FERTILISER

Introduction - The term "placement of fertiliser" has come into use in recent years to describe the differing places or positions in which fertiliser can be arranged in the soil, (as opposed to broadcasting) in relation to the roots of the plants they are nourishing. Different placement positions have widely differing results and correct placement helps ensure the plant obtains the maximum benefit from the fertilisers used.

It is not to be expected that a single pattern of fertilis placement would be found superior with all crops under all conditions, but with many crops the placing of fertiliser in bands at the sides of the seeds or plants has been the most efficient placement.

Band Placement - American workers (29) describe the use of a starter fertiliser with meadow seeding - placed so that it is available to seedlings immediately after germination. This is done by banding both seed and fertiliser. Germination injury resulted when large amounts of mixed fertiliser were used in contact with the legume seed.

Preliminary tests indicated that seeding rates with the band method may be quarter to one-third less than those needed by broadcasting. Accurate placement over the fertiliser bands was important - lucerne seedlings perform as unfertilised plants when separated by only half an inch from the fertiliser bands.

Coating seed with fertiliser - Gusav (30) obtained a utilisation of 70-80% of the phosphate by treating cereal and other seeds with phosphate fertiliser before sowing. The seed was either moistened with a concentrated solution of phosphatic fertiliser, or coated with a mixture of starch paste and fertiliser. Roberts (31) has obtained striking increases in the yields of wheats and oats by soaking the seed in 5% K_2HPO_4 and K_3PO_4 solutions. The yield figures suggest that the phosphate was about sixty times as efficient when absorbed by the seed as whe applied to the soil. Nearly 75 per cent of the applied phospha was recovered from the harvested grain. The limitation of this novel method would seem to be the small amount of phosphate which can be supplied in this way.

Granulation - Band placement is one technique in minimising the fixation of water soluble phosphate. Preparing the fertiliser in a granular form is another method. Often the two methods are combined in practice. Granulation will result in a reduction of fertiliser surface in contact with the soil, with a corresponding reduction in rate of fixation. Each granule becomes the centre of steady phosphate supply, and the plant root actually concentrate themselves round these zones. (25, 32)

Many papers (32, 33, 34) show granulated superphosphate to be more effective than broadcast applications, especially on acid, podsol soils. It is difficult though to evaluate the response due to granulation and that resulting from placement. Terman (35) found that results from pot culture experiments an field tests did not agree, thus sounding a note of warning in accepting results based on pot trials only.

A modification of granulation was the technique of brique ing fertiliser devised by Hance. (36,37) The method however, does not seem to have attracted much further attention. Vario workers (33, 38, 39) recommend "nest" applications of super-phosphate for strongly phosphate fixing soils. Considerably better development of root systems of wheat resulted when larg granules of superphosphate were placed in the centre of small plots as nests. (33)

Advantages of placement - The greater efficiency of fertili correctly placed with respect to the plant is considered due t

(i) increased uptake of nutrients by the plant, i.e. plant/ fertiliser relationship, and

(ii) minimised losses by leaching and fixation in unavailable forms, i.e. fertiliser/soil relationship.

Detailed advantages that have been recorded (40) followin the placement of fertiliser for various crops are increase in yield, reduction in weed growth, better establishment and earl growth, earlier maturity and increased resistance to wire-worm attack. It does not follow that all advantages are enjoyed by all crops under all conditions. The difference in favour of placement fertilising is generally greatest on soils of low nutrient availability and when comparatively small amounts of fertiliser are added.

Localized concentration of fertiliser may have injurious effects on seed germination and seedling establishment.

Plant responses to placement - The early rapid growth of plants resulting from localized placement is due to the increa concentration of plant nutrients available to the young plant. Cook and Millar (41) state that phosphate starvation is usuall most pronounced during the early growth of plants; this is especially true if the growing conditions during this time are not ideal. At that time, owing to slow growth, the roots are not extending to the phosphate and because of its immobile conditions the phosphate is not moving to the roots.

Thus localized high concentration of phosphate near to th roots stimulates rapid early growth, a view expressed by Cooke (42) Haynes and Thatcher (29) report lucerne seedlings show remarkably fast growth when they are given supplementary phosphate. The active root development at an early stage may resu in such plants being able to utilise the soil reserves of minerals more efficiently and earlier than unfertilised plants and render them less vulnerable to adverse environmental conditions.

Recent American data (43) indicate that it is possible to effect early growth stimulation by one treatment and to increa further the uptake of fertiliser phosphate at later stages of growth by another placement. In view of the characteristic phosphate utilisation by various crops, it appeared that multiple placement of phosphate could be "tailored" to the cro

The extensive recent literature dealing with aspects of fertiliser placement and the efficiency of utilisation by plan may be summarised as follows:-

There are many factors that influence the effectiveness of fertiliser placement and perhaps the one with the greatest effect is the soil complex. Much additional knowledge yet remains to be learnt. Advantages in yield, quality and efficiency of use of applied fertiliser are gained by correct placement, and American work has shown that side placement in band is often superior to contact drilling. The present tendency is to depart from the old method of "fertilising the soil" to the practice of "fertilising the crop".

CHAPTER VI.

THE EXPERIMENT, OBJECTIVES, LAYOUT, MATERIALS AND METHOD

The Experiment - No critical work evaluating the pelleting of seed and fertiliser has been found in the literature. That is the main objective of this experiment: to determine the efficiency of the pelleting of seed and fertiliser as compared with the standard method of broadcasting. Pot trials comparing pelleting, broadcasting and a combination treatment with half the fertiliser pelleted and half broadcast were laid down in the autumn of 1950. These treatments were applied to three clover and one grass species using phosphatic and nitrogenous fertilisers at two levels.

Small-scale pot trials were decided upon in preference to plots, because of the slow, tedious manufacture of the pellets. By the use of "identical pots" it was hoped to remove much of the variability associated with soil heterogeneity, and thus allow efficient measurements with few samples on a small scale. Data from pot experiments and field trials were found by Coffey and Tuttle(44) to agree closely in fertiliser experiments. Care needs to be taken however, in applying the results from pot experiments to field conditions.

The responses to the various treatments were measured as dry weight yields of herbage. Routine observational notes on developments, incidence of extraneous factors such as disease, etc. were recorded. Dry weight determinations were used as a uniformly comparable basis of herbage yield estimates over the experimental period. The disadvantages of green weight determinations are manifold and where drying facilities are available, serious criticism can be directed at any work employing green weight fig

An experimental standard of four plants per square foot was decided upon. This figure was the maximum obtained in any of th white clover oversowing trials (6, 7) and it would be deemed satisfactory if four plants per square foot would result consistently from oversowing. The size of the pots used, 8 inch diamet very nearly corresponds to an area of ¼ square foot per plant. Each treatment then consisted of one plant per pot.

The localized placement of fertiliser modifies the plant's root system. Root studies are difficult and laborious and the interpretation of results is not easy. Roots and shoots are closely interdependent and since herbage yields reflect the net response of the plant to the applied treatments and are easy to measure, these are taken as recording treatment results. Root studies of pelleted and broadcast fertiliser plants were done in supplementary pilot trial. The aim of these supplementary studi was to try to understand in what manner the plant responds to pelleting, and to attempt to explain results obtained in the pot trials.

OBJECTIVES

The major objective of the experiment was to compare the pelleting of fertiliser with broadcasting and a combination treatment of half the fertiliser pelleted and half broadcast. It was thought that pelleting might show superior results during the early stages of seedling growth, due to greater nutrient uptake from the localized concentration of fertiliser. The broadcast fertiliser is spread over a wider area. The plant's response to the fertiliser is delayed until the root system has developed sufficiently to tap this supply. Thus the combination treatment of half the fertiliser pelleted and half broadcast was to see whether it was possible to combine the initial "boost" of the pelleted fertiliser with the later and continued response from broadcast fertiliser - whether the combination of both treatments was better than each individually.

These treatments were compared, using phosphate and nitrogen fertilisers, at two levels, with three clover and one grass species. Superphosphate and sulphate of ammonia were chosen for a number of reasons - they are the two fertilisers likely to be of commercial importance in pelleting, and also they offer contrasting properties and modes of action. The one is soluble readily available and gives a rapid response; superphosphate is less readily available, gives a response over a longer period, and efficiency of utilisation by plants is low. They differ in degree of germination injury caused by pelleting. New Zealand soils are largely deficient in phosphates, hence superphosphate is the main fertiliser applied to pastures. Nitrogen fertilisers are not generally applied to pasture, the grass growth being dependent on the nitrogen "fixed" by clovers made vigorous with phosphatic topdressing. Some workers (62, 71) believe that clovers as well as grasses benefit from the addition of a small quantity of nitrogen fertiliser at seeding.

Two levels of fertiliser were used, 0.02 gm. per seed and 0.01 gm. per seed. The higher level was the maximum amount found during the preliminary germination trials which did not seriously reduce germination. It was thought that the differe in favour of pelleting would be greater if a smaller quantity fertiliser was used, i.e. the lower the amount of fertiliser p seed.

The species used in the trials were red clover (*Trifolium pratense*), white clover, subterranean clover (*Trifolium subterraneum*) and perennial ryegrass. The three clovers were chosen as these are the species likely to be of importance in the over-sowing of hill country, also they have differing growt forms - white clover a perennial, red clover a bi- or triennial and subterranean clover an annual. It was not known whether these differences in the species' characteristics would influe treatment responses. Perennial ryegrass was included to compa the pelleting of a monocotyledon possessing a different root system and seedling morphology. The pelleting of grass seed a fertiliser was not thought to be as commercially suitable as wa clover, but the pelleting of grass seed had been advocated for the prevention of uneven distribution with aerial sowing. (8)

A fifth series was a combination treatment, with a central white clover plant surrounded by four ryegrass plants. The pelleting treatments were applied only to the central white clover, and results were measured as white clover herbage yield (although grass yields were recorded). This series was identi al with the white clover series except for the competition of the ryegrasses. The quicker growing grasses with their extensi fibrous root system would use a large part of the broadcast fertiliser, and the white clover would suffer a double check from:

(i) direct competition for fertiliser uptake, and

(ii) the grasses would obtain an advantage and check the clove still more by shading and competition for moisture and nutrients.

The advantages of pelleting should be emphasised. This series most closely corresponds with what would occur in pract with oversowing, i.e. with clover seed surface sown on pasture the established grasses strongly compete for available nutrien and moisture.

The objects of the pot trial in detail, are as follows -

To compare (i) the pelleting of seed and fertiliser with broadcasting

(ii) the treatment responses with nitrogen and phosphate fertilisers and combinations of the fertilisers

(iii) these fertiliser treatments at 2 levels, 0.01 gm/seed and 0.02 gm/seed

(iv) the fertiliser treatment responses with diffe ing species - red clover
White clover
subterranean clover
perennial ryegrass

(v) the effect of plant competition on treatment response

(vi) all possible interactions of these factors.

LAYOUT

An adequate experimental design involves not only a satis factory plan for conducting the trial, but also an appropriate statistical method for evaluating the results. The two can il be divorced. (Snedecor (45))

The size of the experiment was limited by the number of

8 inch pots available. These were not quite 200 pots, and the experiment was restricted to these. The design of the trial wa a 3 X 3 X 3 X 5 factorial with one replication.

There were three treatments of 3 levels of nitrogen and three levels of phosphate fertilisers on five varieties. The symbolism used was as follows –

Treatments p = pelleted fertiliser

b = broadcast fertiliser

pb = half fertiliser pelleted and half broadcast

Fertilisers O = no fertiliser, control

N_1 = 0.01 gm. of sulphate of ammonia

N_2 = 0.02 gm. " " " "

O

P_1 = 0.01 gm. of superphosphate

P_2 = 0.02 gm. " "

Varieties Aa = red clover

Ac = white clover

Ak = subterranean clover

Ba = perennial ryegrass

M = combination of Ac and Ba

There were twenty-seven readings for each species. A diagrammatical outline for a single species is presented below.

Ak

P	O	N_1	N_2	P_1	P_2	N_1P_1	N_2P_2	N_1P_2	N_2P_1
b	"	"	"	"	"	"	"	"	"
pb	"	"	"	"	"	"	"	"	"

eg. N_1P_2 p = (0.01 gm N + 0.02 gm P) pelleted/seed

N_2P_1 pb = (0.01 gm N + 0.005 gm P) pelleted

+ (0.01 gm N + 0.005 gm P) broadcast

The pots were in double rows sunk into the ground. Treatments wer allotted at random. (See Appendix II for arrangement of pots and treatments.) Figure 8 shows the layout of the pot trial.

MATERIALS

Seed - The same line of white clover was used in the preliminar pelleting tests. The other seed used was:-

Government Stock, Montgomery red clover, 99% purity and (74 + 16) % germination.

Certified Mt.Barker subterranean clover, 99.7% purity and (87 + 9) % germination.

Government Stock, perennial ryegrass, 99.6% purity and 93% germination.

Fertiliser - The same fertilisers and pellets were used in the preliminary pelleting tests.

Soil - The soil used in this experiment came from a fairly unifor bank thrown up by a bulldozer when widening and straightening the Tiritea stream, Fitzherbert West. This soil was chosen as it was accessible, of a sandy nature and relatively impoverished. It was felt a marked response to fertiliser treatments would be obtained.

Three cubic yards of this loamy sand, as it was tentatively classified, were sieved, free from stones, organic debris, etc. through a wire mattress onto a tractor trailer. The soil was transported to a glass house and laid out on tarpaulins to dry. After thorough mixing and a final fine sieving, 8 lbs. of air dry soil were weighed into each pot.

FIGURE 3. Layout of the pot trial. Taken November, 1951

The aim was to maintain all other factors as constant as possible in order to emphasize any differences due to treatments. It was hoped in this manner and by use of "identical pots" to largely eliminate the otherwise considerable variation of soil heterogenity.

A mechanical analysis of the soil was carried out (see Appendix III for method), resulting in the determination on an air dry basis of:

3.6% moisture
2.9% loss by ignition
0.7% loss by solution
6.9% clay
9.4% silt
76.5% sand

These proportions of soil fractions result in classification as sandy loam. (46) The soil properties and plant reactions that could be expected to result from this large proportion of sand must be borne in mind in interpretating the experimental results.

Pots — The flower pots used were 8 inch diameter, unglazed, porous clay pots. These were well washed before using.

This size was chosen because they would be large enough to prevent the pots becoming pot-bound during the twelve month experimental period.

METHODS

Potting — Eight pounds of sifted, air-dry soil were weighed into each pot. The pots were placed in double rows three feet apart and sunk into the ground. This was to simulate natural conditions as much as possible. Figure eight shows the layout of the pots. The pots were thoroughly watered the night before sowi

Sowing - The treatments were sown on 13th March 1950. The treatments were chosen at random and labelled pegs showing the respective treatments were stuck in each pot. Three seeds of the particular species were sown per pot, the fertiliser pellet being placed on top of the seeds. The aim was to allow for unforeseen losses due to birds, hard seeds, etc. and to ensure at least one seedling grew per pot. The broadcast fertiliser was uniformly spread using an improvised shaker - a round tin with holes punched in the lid.

The avoid much of the germination injury, the pelleted fertil er was placed on top of the seed instead of the seed being inside the pellet. It was felt that this experimental method was justifi and would not seriously invalidate any conclusions drawn from the experiment. Because of the small number of samples, it was essent to avoid as far as possible any misses.

As a further precaution, a duplication of the white clover an pelleted nitrogen treatments were laid down in some spare pots. This was to allow for any germination injury due to the concentrat of Sulphate of Ammonia. Seven spare pots were left bare in case some treatment should later require checking, or in case they were required for any purpose later on.

From the three sown seeds the healthiest seedling was selecte and the others pulled out, leaving one seedling per pot. In spite of these precautions, a few treatments failed to germinate and these were resown eleven days later.

0.26 inches of rain fell the day after sowing.

Cutting - The herbage was cut to a level of half an inch using modified sheep shears as adopted by Marryatt and Simpson (47). Wooden blocks, half an inch thick, were placed on either side of the plant as height guards. The times of cutting varied. Whenever there was sufficient growth a particular species was cut. The quicker establishing ryegrass series were cut on 20th April 1950 to try to evaluate the treatment effects on early growth, and the ryegrass plants in the M series on 20th June 1950 to prevent undue shading of the clover seedlings.

If some treatments had not enough herbage to clip, they were left until the next cutting period. The last cut was on 15th December 1950, nine months after sowing. Refer to Appendix IV for details of cutting dates and yields.

Weighing - Clipped herbage was placed in small, metal patty tins together with a numbered metal disc. The total cut series were placed in a forced draught, drying oven at the Grasslands Division, Department of Scientific and Industrial Research. They were dried from 3-4 hours at 70°C. The samples were then transferred in small batches from the oven to a dessicator and weighed as rapidly as possible.

The pots were cut in sequence and by reference to the plan of the experiment and the numbered metal disc showing the sequence of the sample, recording was rapid and efficient.

An air-damped, Satorius-Werke balance was used. This balance is reasonably fast, very easy to operate and accurate to 0.0002 gm. This accuracy of weighing was used in evaluating the treatment effects on early growth, when herbage weights were small. Weighings were made to the third decimal place of a gram, as small weights were anticipated and weighings had to be accurat to establish real differences. Weight sheets for the separate species were prepared and data at each weighing entered under the treatments.

Routine Care and Notes - Observations were made on general grow and development, incidence of disease, insect damage etc. and notes entered in the field book.

Routine care consisted of periodic weeding of the pots. The principal weeds were spurrey (*Spergula arvensis*), mouse-eared-chickweed (*Cerastium glomeratum*) and *Poa annua*. The spaces between the pots were weeded, and the paths between the rows were hoed occasionally. The pots were watered at intervals during the three weeks after sowing. The dry conditions were accentuated by the sandy pot soil, and the seedlings were watered to aid surviva

CHAPTER VII

RESULTS - POT TRIALS

General Development - Some subterranean clovers had germin and a few red clovers were beginning to germinate twelve days after sowing. The dry conditions were accentuated by the sand nature of the pots and were a severe natural hazard. The pots were watered by night with a fine spray from a sprinkler jet. The conditions in the pots were markedly different from the micro-environment provided by the shade and shelter of the gra in an oversown pasture.

Rain on the 4th March 1950 had largely dissolved the N pellets, while the P.N. pellets were mainly intact. Phosphate pellets still remained eleven days after sowing.

The treatments were classified by eye with respect to speed of establishment and growth, one month after sowing. Th stages of development were recognized in the clovers and two i the ryegrass. Results are recorded in Table IV.

TABLE IV. Establishment assessment. Date: 4.4.50

Key to symbols - Ba (ryegrass) - U = unfolded shoot
S = shoot of first leave

Clovers - c = Cotyledons showing
f = First leaf
t = first trifoliate lea

	N	N_2	P_1	P_2	N_1P_1	N_2P_2	N_2P_1	N_1P_2	O
				As (Red clover)					
p	f	c	c	c	f	c	f	c	f
b	f	O	f	f	f	O	c	f	f
pb	t	c	c	f	f	t	f	c	f
				Ac (White clover)					
p	f	c	f	f	f	f	c	f	c
b	f	f	f	c	c	f	c	c	f
pb	c	f	f	f	O	c	c	f	c
				Ak (sub. clover)					
p	f	f	t	t	t	t	t	t	t
b	c	t	t	s	t	t	t	t	t
pb	[illegible]	un-emerged	t	t	t	t	t	f	t
				Ba (per. ryegrass)					
p	s	s	s	s	very vigorous	u	u	u	s
b	s	s	s	s	third leaf	s	s	u	s
pb	s	s	s	s	vigorous	s	s	s	s
				M (white clover + ryegrass) Refers to Ac only					
p	f	f	f	f	f	c	f	f	c
b	c	c	c	f	f	f	f	f	c
pb	f	c	f	f	f	t	f	c	c

There is no definite trend resulting from the treatments. The subterranean clovers show quicker establishment.

There is considerable variation between the Ac series and th clovers in the M series, although these should be similar as ther was no competition at this stage from the ryegrass seedlings in t

M series. The establishment assessment does not agree with from the root study plot trials. There pelleting consistent. speeded the growth and development of the clover seedlings.

Figures 9, 10, 11 and 12 show graduation in response from Ba O, N_1P_1 b, N_1P_1 pb to N_1P_1 p. The pelleted plant had greate tillering, spread and height of herbage. This isolated series typifying exactly the expected results, must be accepted with reservation. The apparent differences may not be due to the treatments, and until confirmed by statistical examination must regarded with caution.

The cotyledon leaves of four plants had been chewed by insects and these were resown on 11th April 1950.

<u>Compilation of results</u> - The analysis of variance developed b R.A. Fisher consists essentially in the portion and apportionmen of the total experimental variation to its known causes with a residual portion ascribable to unknown or uncontrolled variation and therefore termed experimental error. When the variability i measured in suitable terms, i.e. sums of squares of deviations about the means, the variability ascribed to the various causes will be strictly additive.

Four treatments were missing at the completion of the exper ment and five plants appeared abnormal. Four of these abnormal plants were subterranean clovers which appeared to be affected b a virus disease. Similar symptoms had appeared in subterranean plants in a neighbouring experiment. The final figures were -

3% missing pots

4% abnormal results (due to natural causes)

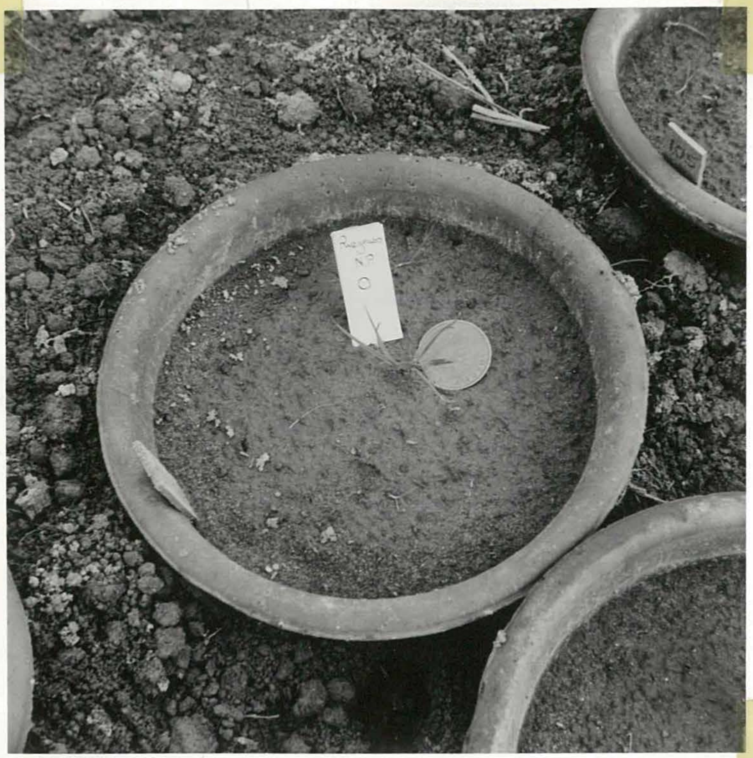


FIGURE 9. Ba series: Ryegrass control 0 fertiliser. 18/4/50

FIGURE 10. Ba series: Ryegrass N_1P_1 broadcast. Note slight response over control above. 18/4/50

FIGURE 11. Ba series: Ryegrass N_1P_1 pb (half the fertiliser pelleted and half broadcast). Note intermediate response compared with Figures 10 and 11. 18/4/50

FIGURE 12. Ba series: Ryegrass N_1P_1 pelleted. Note increase in growth and general development in series from control, N_1P_1 broadcast, N_1P_1 half pelleted and half broadcast, to N_1P_1 pelleted. 18/4/50

The statistically expected values were supplied by the Applied Mathematics Laboratory using the method outlined in Kendall's "Advanced Theory of Statistics" (48). In the analysis of the results, 9 degrees of freedom were subtracted from the possible total because these calculated values were considered "fixed", being dependent on the other values.

The results were arranged as in Table V for statistical analysis of the data.

TABLE V. Pot trials - Mean yields of main effects.

Grouping criteria	Groups	Mean average yield per pot (mgs.)
V (varieties)	Aa (red clover)	6384
	Ac (white clover)	11152
	Ak (subterranean clover)	3000
	Ba (per.ryegrass)	2446
	M (modified Ac series)	9055
Tr (treatments)	p (pelleted fertiliser)	6751
	b (broadcast ")	5644
	pb (½ fertiliser pelleted & ½ broadcast)	7126
N (sulphate of ammonia)	0 (control)	6314
	1 (0.01 gm. per seed)	6184
	2 (0.02 gm. per seed)	7024
P (superphosphate)	0 (control)	5708
	1 (0.01 gm. per seed)	6367
	2 (0.02 gm. per seed)	7669
	General mean =	6507

The totalled treatment results are tabulated in Appendix IV. From these figures the sums of squares of main effects and

1st and 2nd order interactions were calculated.

Analysis of results - The pot trial was analysed as a 3 X 3 X 3 X 5 factorial, with one replication, using a pooled error term made up of all the second and higher order interact: (except P X Tr X N the only second order interaction not involving V). The interaction (P X Tr X N) was separated and examined, as, disregarding species, this would be the only high order interaction easily interpreted.

The mean squares of the various high-order interactions which were pooled were calculated, for the validity of the pool Error Mean Square depended on these being of the same order of magnitude, so that the error term would not be biassed by an excessively large or small component here.

The analysis of variance is as follows:

TABLE VI. Analysis of Variance of Pot Trial

Source of Variance	Degrees of freedom	Sum of Squares	Mean Square	F variance ratio	5% Ratio required	Significance
V	4	15,858,513	3,964,628	24.3	2.50	ss
Tr	2	534,137	267,069	1.7	3.13	-
N	2	183,717	91,859	0.6	3.13	-
P	2	693,999	347,000	2.1	3.13	-
V X Tr	8	961,386	120,173	0.7	2.07	-
V X N	8	742,340	92,793	0.6	2.07	-
V X P	8	1,131,252	141,407	0.9	2.07	-
Tr X N	4	852,846	213,212	1.3	2.50	-
Tr X P	4	233,206	58,302	0.3	2.50	-
N X P	4	1,122,447	280,612	1.7	2.50	-
Tr X N X P	8	1,324,750	165,594	1.0	2.07	-
V X Tr X N	16	2,286,095	Pooled error term			
V X Tr X P	16	2,686,234				
V X N X P	16	2,994,993				
V X Tr X N X P	23	3,580,668				
Error	71	11,347,990	162,648			
Total	125	35,186,583				

ss = significant at the 1% level of probability. Standard deviation = 4033

TABLE VII. Table of varietal means (per pot basis)

Aa	-	6,384	mgs.
Ac	-	11,152	"
Ak	-	3,000	"
Ba	-	2,446	"
M	-	9,055	"

The difference between a pair of means (per plot basis) required for significance at the 5% level is given by

$$d_{.05} = 1865 \text{ mgs.}$$

Ac yielded higher than the other species. The difference betwee the Ac and M means, 2097 mgs. is greater than the difference required for significance (see above); thus the Ac and M white clovers are from different populations. This results from the competition of the four ryegrass plants per pot, in the M series

Analysis of 1st cuts: The treatment effects were more marked and showed less variability in the first yield cuts (see Appendix IV). Accordingly the yields from the 1st cut for the Ac and M series were analysed separately.

(a) The analysis of variance for the Ac 1st cut is shown in Table VIII.

TABLE VIII. Analysis of Variance of Ac 1st cut.

Source of variance	Degrees of freedom	Sum of Squares	Mean Square	F variance ratio	F 5% ratio required	Signi-fican
Tr	2	4238	2119	1.7	5.14	.
N	2	8989	4495	3.6	5.14	.
P	2	24149	12075	9.8	5.14	
Tr X N	4	9562	2391	2.0	4.53	.
Tr X P	4	8215	2054	1.7	4.53	.
N X P	4	7808	1952	1.6	4.53	.
Error = N X Tr X P	6	7448	1241			
Total	24	70409				

S = significant at the 5% level

Standard deviation 35.2

Coefficient of variation 75%

The variance due to P was significant at the 5% level. The diffe ence between means required by the "t" test for significance at t 5% level is 100 mg.s.

Thus there is a significant increase in yield from - --

Po - mean 112 mgs.

P_1 - " 383 "

to P_2 - " 768 "

There is also a trend for N_2 to have a depressing effect -

No - mean 429 mgs.

N_1 - " 618 mgs.

N_2 - " 216 mgs.

(b) In the same way the 1st M cut was analysed as a separate

3 X 3 X 3 experiment.

TABLE IX. Analysis of variance of M 1st cut

Source of variance	Degrees of freedom	Sum of Squares	Mean Square	F variance ratio	F 5% ratio req'd	Significance
Tr	2	2,514,735	1,257,368	12.57	4.74	S
N	2	366,819	183,410	1.85	4.74	-
P	2	351,724	175,862	1.77	4.74	-
Tr X N	4	749,108	187,277	1.89	4.12	-
Tr X P	4	1,839,351	459,838	4.63	4.12	S
N X P	4	848,673	212,168	2.14	4.12	-
Error Tr X N X P	7	695,051	99,293			
Total	25	7,365,461				

S = significant at the 5% level

Standard deviation = 315

Coefficient of variation = 31.5%

Variance due to treatments, and the interaction Tr X P are significant. This means that the variance due to p, pb and b is significant, and that the treatment effects vary within the different levels of P.

Table X. Table of means of 1st M cut. (mg.s)

	p	b	pb	Phosphate means
P_0	765	665	1381	937
P_1	1541	318	920	926
P_2	1031	813	1677	1174
Method means	1112.2	598.7	1325.9	1012.3

(i) 't' test for difference of means of main effect Tr,

$$d_{.05} = 352.0 \text{ mg.s}$$

From the table of means (Table X) it is seen that the yields fr p or pb are significantly greater than b. There is no signific difference between p or pb, but this may be due to the Po value which were allotted by chance: (Po p = 765 gms.

P_o pb = 1381 gms.)

(ii) Interaction (Tr X P) - or treatments within levels of phosphate.

The difference required by the "t" test to establish significance,

$$d_{.05} = 610 \text{ mg.s.}$$

Thus within P_1 level of phosphate, p gives greater yields than pb, or pb gives increase over b. For P_2, pb is better than p o b, with no significant difference between p or b. P_2 pb is significantly greater than P_1 pb. It appears that pelleted phosphate has a depressing effect at the higher level of fertil er. The treatment that gave the greatest yield was P_2 pb, i.e. the higher phosphate level with half the fertiliser pelleted an half broadcast.

<u>Summarising the results</u> - The analysis of variance of the pot trial as a 3 X 3 X 3 X 5 factorial with a pooled error term only gave significant differences due to varieties. White clov yielded higher than the other species. The competition from the ryegrass plants in the M series caused the M white clover result to differ significantly from the Ac series.

Analysis of the 1st Ac cut showed a significant yield increase due to levels of phosphate - P2 was greater than P1, and

P_1 greater than P_0.

Testing for treatment effect, and Tr X P interaction within the 1st M cut resulted in p and pb being significantly greater than b, and showed that the methods have differing effects within the levels of phosphate. At the higher P level pelleting appeared to have a depressing effect, and pb was superior.

SECTION C

ROOT STUDIES

CHAPTER VIII

REVIEW OF LITERATURE

ROOT DEVELOPMENT

A knowledge of the behaviour of the root system is of fundamental importance when considering the responses of the pla brought about by changes in the soil environment. (49) Roots and shoot are closely inter-dependent, (cf. review (50)), while underground development may be profoundly influenced by nutrient supply and placement. The advantages of localized placement (pelleting) arise from an extensive early root development (42) and ease and rapidity of phosphate absorption by the roots. Consequently the literature concerning root growth, its morpholo and development, and the factors governing these, was reviewed i an attempt to understand the underlying reasons for placement advantage and also any possible disadvantages that might accrue.

General: The general physical and physiological characteristi of root systems depend primarily on their heredity. Neverthele wide variations in the development of root systems within the limits of their hereditary potentialities are brought about by variations in certain environmental factors. Among these are soil fertility, soil moisture and competition with other root systems. "Three physical properties of the soil, i.e. texture structure and depth, principally determine the development and functioning of the root system." (49) These same soil propert affect and modify soil moisture and aeration.

Since roots are dependent on their shoots for carbohydrate supplies, those factors which affect photosynthesis also affect root growth, eg. clipping and grazing, (50) and it has been demonstrated that shading reduces growth and the ratio of roots t shoots. (52)

Phosphate placement affecting root habit. It is well known tha when roots penetrate into areas of the soil containing an abunda of minerals they branch profusely but reports deal almost entire with the gramineae. Jacques (53) concludes that a relatively better penetration was found in the roots of unfertilised cocksf (*Dactylis glomerata*) and perennial ryegrass (*Lolium perenne*) plan Surface fertility tends to cause root accumulation in the fertil zone and less root is developed in proportion in the deeper-seat layers.

The same worker found white clover had almost 38 per cent o the root weight present in the top inch of soil and that with this species root penetration was little affected by fertiliser (superphosphate) placement.

Reports on placement and granulation of superphosphate (32, 33) state that bunches of fine roots develop round the granules, and plant roots concentrate around these zones of steady phospha supply. The distribution of lateral roots was therefore investigated to try to determine whether pelleting caused localize root concentration.

Phosphate uptake affecting the physiology of the plant - The mo efficient zone of absorption is usually near the root tip, hence the number of young root tips is an important factor in nutrient uptake and plant growth. Using isolated wheat roots, Burstrom

(54) found phosphate promoted cell multiplication over a wide range of concentrations, but had a negligible influence on cell elongation. Nitrates caused a rapid increase in rate of cell elongation but did not affect the rate of cell division.

The hypothesis that phosphate manuring increased the weigh and development of roots to a greater proportion than top growt was frequently advanced in the older text books. Muller (55) with barley and Eaton (56) with soy-beans found that phosphorus deficiency affected the top more than the roots so that the top root ratio of the minus phosphate plants was lower than that of the plus-phosphate ones. This relationship was investigated further.

NODULATION

Interest was first aroused in nodulation when it was obser ed at the lifting of the four week old clover seedlings for roo studies, that the pelleted treatments were nodulated while the broadcast or control plants were conspicuous by their lack of nodules. This study was pursued further and the relevant liter ature reviewed concerning both nodulation in geleral, and nodul ation in the process of symbiotic nitrogen fixation and the factors affecting it. The immense significance of legume bacteria in the economy of nature and man has primarily called the attention of research workers to the nodule bacteria, to th formation and activity of root nodules, and to other facts associated with them.

General - Development of Nodules. The general course of infection and nodule formation is well known. (57)

The bacteria normally enter the plant by penetrating the root hairs near the distal extremity, although there is evidenc that they may, more rarely, enter the other epidermal cells. The plant secretes from the roots a substance which assists this infection, (58) but the small percentage of root hairs infected even in the presence of large numbers of bacteria, indicates th the plant tissues can resist excessive infection. (59)

The infected root hairs usually show a characteristic curl ing of the tip, believed due to the production of indole-acetic acid, a common metabolic product of bacteria. (61) The point of infection is generally in this curled region. The bacteria within the root hair form one or more "infection threads" which grow down the hair and penetrate the cortical cells of the root Penetration of the cortical cells is accompanied by their rapid division producing a mass of young cells through which the infection threads ramify. It is known that the penetration of th infection thread through the cortex takes place contemporaneous with, and not subsequent to, the development of the pericycle o cortical cells into the initial nodular tissue. (62) The close examination fails to establish whether the penetration of the slime thread from an infected root hair into the cortex of the root is a consequence of meristematic activity in the root, or whether cell division follows immediately upon penetration. (63

The two opposing views on infection may be briefly formula as follows:-

1. The successful penetration of the infection thread stimulates the cells of the root to divide, the morphology of the resulting nodule being merely a consequence of the realized potentialities of any root meristem; or

2. the infection thread in the root hair is able to penetrate

the cortex only at points of incipient meristematic activity i.e. the foci of infection are predetermined in the plant, possibly at the site of lateral root initials.

The latter view has been propounded by Nutman (63, 64) who advances evidence in support of his hypothesis.

Nodulation has been interpreted by Thimann (5) in terms of the initiation and infection of lateral root primordia. He regar as an important factor the production of B-indolyacetic acid by the bacteria, which in very low concentrations is held to stimula the formation of the initials. This substance, a produce of bacterial metabolism inside the root tissues, causes enlargement the cells in which it is produced, and also, being readily diffus ible enters the pericycle and there also stimulates growth and division, giving rise to the first stages of a lateral root initi However since auxin production continues, this lateral root is prevented from elongating. Instead its cells increase in size, while certain of the uninfected cells are stimulated to division by auxin diffusing out of the infected area. He offers no explan ation of the formation and differentiation of the vascular system within the nodule; and is criticized by Nutman (63) on the failur of this view to adequately explain the greater number of nodules produced by innoculations with ineffective bacterial strains.

Some of the bacteria escape from the infection thread and come to lie scattered in the cytoplasm. After their escape the bacteria multiply rapidly and increase in size, usually becoming irregular and branched - the so-called bacteroid stage. Abundanc of this bacteriod stage seems to be correlated with active nitrogen fixation within the nodule. As soon as the bacteria become numerous in the cytoplasm, the host cell ceases to divide but increases in size with a corresponding hypertrophy of the nucleus. This effect of the bacteria upon the infected cells varies according to the physiology of the host plant, and age of the bacteroidal tissue. (60)

<u>Structure of the Nodule.</u> (65) The legume nodule consists of a mass of central cells containing <u>Rhizobia</u> bacteria, surrounded b a zone of parenchyma with a single vascular strand (Trifolium). The xylem consists of tracheids or parenchyma. The phloem is simply narrow, very elongated cells, and the strand is surrounde by a kind of bundle sheath. The nodule is protected by a varyin number of layers of cells developed from a meristemmatic layer, which may become dead and empty, or in some cases thickened.

<u>Pigmentation in Root Nodules.</u> Virtanen (66) discusses the significance of the red pigment in root nodules in his review. At Wisconsin station it was found (67) that the nodules formed b poor strains are usually small, round and white, and lie scatter over the entire root system. Nodules from a good strain are relatively few in number, fairly large, pink in colour and elong ed, and are located near the tap roots.

The red pigment of effective root nodules is haemoglobin, termed by Virtanen, leghaemoglobin. The observations so far mad show a distinct parellelism between nitrogen fixation and the occurrence of leghaemoglobin. If the pigment is not present in nodule or if it is decomposed, nitrogen fixation does not occur.

<u>Influence of Plant Nutrient Relationships.</u> Because nodulation occurs within a two phase symbiotic system, nutrient changes act

in two ways -

(a) The effect on the host plant.

(b) The effect on the Rhizobia organisms in the soil.

(1) Nutrient effect on host plant:

(a) C/N ratio - The carbohydrate level of the plant and particularly the relation of carbohydrate to nitrogen strongly effects symbiotic nitrogen fixation. The inhibiting effect of combined nitrogen on nodulation has been known for many years, an it is known also that an adequate supply of soluble carbohydrate must be maintained to permit nodulation and maximum fixation. (67

Orcutt and Fred (68) noted that innoculated soy-beans grown in sunlight of high intensity entered the "nitrogen-hunger" stage and remained there, although numerous and well-developed nodules were formed on the roots. When the plants at this stage were shaded, or fertilised with nitrates, nitrogen fixation set in. Thus too much as well as twoo little carbohydrate may inhibit nitrogen fixation.

The evidence is that restricted nodulation and nitrogen fixation resulting indirectly from some nutrient deficiency or unfavourable conditions in the host plant, is due in most cases to restricted supply of soluble carbohydrates to the nodules, or to adverse C/N relationship.

Jensen (69) found the provision of combined nitrogen in quantities similar to, or somewhat higher than, the amounts fixed in free nitrogen series has comparatively little influence on the actual number of nodules, but tends to lower the numbers in proportion to the weight of the roots. The average size of the nodu is greatly diminished and the proportional weight of the nodule fraction is reduced roughly one fourth.

The inhibitory effect of combined nitrogen on nitrogen fixation seems due to reduced development of nodule tissue (70) as well as lower efficiency of the tissue formed, probably because the available carbohydrates are largely used for protein synthesis with the combined nitrogen (cf. Wilson (67)). It is of interest to note that intermediate concentrations of nitrate may increase nodulation and also the amount of nitrogen fixed by increasing leaf area and photosynthesis. (71) Higher nitr concentrations decrease the soluble sugar level, and also the number and size of nodules.

(b) Phosphate status - Anderson at the Commonwealth Specialist Conference in Agriculture (72) stated that clovers l in phosphate are usually well nodulated, but German, Indian and American workers have reported that phosphate fertilisation increases nodulation. (73, 74, 75, 76) Jacques (53) working under New Zealand conditions found the number of nodules varied considerably regardless of fertiliser treatment, and some facto or factors other than phosphate content of the soil governed their distribution. Results in other trials varied with the ye but superphosphate mixed in the top six inches of the soil gave greatest nodulation.

It is difficult in seeking to evaluate the influence of phosphate on nodulation, to separate the indirect effects of phosphate fertilisers on the root system and on the C/N ratio o the plant. Roberts and Olway (73) suggest that a deficiency of phosphate may inhibit nitrogen fixation by interrupting protein synthesis.

(c) Calcium - Superphosphate contains approximately 60 lbs. of gypsum ($CaSO_4$) per hundredweight, therefore the influence of calcium on nodulation was reviewed. There is evidence that nodulation is influenced by the amount of calcium taken up by the plant. (77, 78)

(2) Nutrient effect on the development of Rhizobium species in the soil.

(a) Calcium - The separate influence of calcium and pH for the nodulation and development of legumes has been clearly demonstrated by Albrecht. (79) Added calcium may increase both growth and percentage nitrogen of legumes. (77) It is however not clear whether the improved nitrogen status of the plant is the result of the influence of calcium inside the plant, or on the Rhizobium species in the soil.

Various workers (80, 81, 82) have shown that the effect of lime on nodulation may be extremely localized.

(b) Soil pH - Jensen (69) found that the mass of root nodule tissue is little affected by the reaction because the smaller number of nodules in acid sand is compensated by increased size of individual nodules. When once formed nodules can continue to fix nitrogen at external reactions too low for the formation of new nodules (83).

(c) Phosphate supply - It is possible nodulation may become limiting at low phosphate supply level, following initial inoculation with low numbers. (72) Truesdell (84) found that phosphate markedly increased the growth of Rhizobia meliloti in the soil.

CHAPTER IX.

ROOT STUDIES, OBJECTIVES, LAYOUT, MATERIALS AND METHOD

OBJECTIVES

The advantages of pelleting are thought due to an early extensive root development, and ease and rapidity of phosphate absorption by the roots. Root characteristics and distribution were studied to try to understand in what manner the plant respo- ed to the localized fertiliser placement and why there was that response. A pilot trial of duplicate blocks was laid down on 3rd April 1950 to try to explain the results from pelleting.

There were three objectives.

(i) To study the root/shoot ratio of pelleted, broadcast and control plants. Conflicting views have been presented that phosphate fertiliser increases top growth in proportion to root growth and vice versa.

(ii) To study treatment effects on nodulation; the number, distribution and colour of the nodules. A side issue was the application of the data to test current nodulation theories.

(iii) To see whether there was a concentration of surface roots with the localized placement of phosphate fertiliser. A surface distribution of roots may be harmful in a following dry period.

LAYOUT

The trial consisted of duplicate blocks of white clover plot

The treatments were

O - control

b - broadcast fertiliser equivalent to 0.02 gm. of superphosphate per seed, and

p - 0.02 gm. of superphosphate pelleted per seed.

These were allotted at random. Four white clover plants per square foot were sown. Border effect was minimized with the sowing of every alternate row as a buffer row (sample rows wer "lifted" at four weekly intervals), likewise the end plant in row acted as a buffer plant and was not sampled.

This simple layout was used because the number of plants able to be handled in root studies is limited, and also a simp layout would be sufficient for a pilot trial. The plots were laid down as follows:

A	O	C	b	E	P		O	-	Control
							p	-	Pelleted
B	P	D	O	F	b		b	-	Broadcast

MATERIALS

The fertilisers, pellets, seed and soil used were those used in the pot trial. Sufficient soil was left over from the pot experiments to use in this small trial. Results from the pot trial and root studies should be comparable, as the same materials and soil were used in both experiments.

METHOD

Laying down the plots -- The plots were 4 ft. long by 2 ft. 6 inches long, and 8 inches deep. Plots this size were excavat and hollow frames of 8 X 1 inch *Pinus radiata* fitted. These lined, empty plots were then filled to a depth of 8 inches with equal volumes of the silty sand ex glasshouse and pot supply.

Each plot was well firmed by trampling, and then thoroughly watered.

The white clover was sown 6 inches apart. Three seeds were sown at each plant position (to allow for losses) and later thinned to give 4 plants per square foot - the same density as the pot trial.

The broadcast plots received a 1 : 1 mixture of superphosphate and Bentonite at an equivalent rate to the pelleting, i.e. 0.02 gm. superphosphate/36 sq. inches. This amounted to 0.8 gm. of superphosphate on a 10 sq. foot plot.

The plots were sown on 3rd April 1950.

0.39 inches of rain fell on 5th April 1950.

<u>Lifting</u>: A technique of soaking, washing and sieving was used to separate the root system from the soil. The individual plant within the treatments were sampled as a core 7 inches in length and 5 inches in diameter. The plants were spaced 6 inches apart in the plots and this core size allowed 1 inch margin for errors slight departures from the vertical, etc.

The cores were removed by placing a thin tin cylinder of th right dimensions over the clover plant to be sampled, and drivi it to its full depth into the soil. This was then cut away with a trowel and the small encased core removed. (Figure 13 shows method of core extraction) The soil was carefully washed free from the roots using a hose and a gentle stream of water. This separation was relatively easy because of the sandy nature of the soil. Washing commenced at the bottom of the core and when most of the soil had been removed, the tin, plus clover plant and top few inches of soil, was inverted into a fine sieve. The

sieve was agitated to and fro under water and finally the plan
and roots were washed free on the sieve with a gentle stream o
water. The freed plants were placed in water in labelled petr
dishes and removed to the laboratory for further study.

The inner plants only were sampled neglecting those at th edges which might show a possible "border effect" and leaving the front and rear guard rows. During the early samplings whi the root systems were still small and restricted, and the guar rows were performing no function, these plant positions in the B and C plots were sampled also. Thus normally the three inne plants of each second row were sampled at varying dates. The first liftings were 2 and 4 weeks after sowing and thence at approximately four-weekly intervals. The final sampling on th 27th August 1950 was after a double interval-period of eight weeks.

In the laboratory the individual plants were placed in water in large dishes, the organic debris removed by forceps and the roots teased out and freed from clinging soil. At the earlier liftings the root systems were examined under a low power binocular microscope to check on the efficiency of the method and to ensure the roots were intact, i.e. the root tips and caps were present. The lifted series were placed in a Frigidaire and stored at 22°F while the individual plant root systems were studied in detail.

<u>Root studies</u>: The plants were removed singly from the Frigida for detailed study. The individual plants were numbered for e of recording and three different factor-aspects were considere

FIGURE 13. Plot E p showing method of extracting root systems. Guard rows on either side of sample plants. Note tin, encasing sample plant core removed for root separation.

(i) The shoot/root system - the tops were dissected from the root system; the number of trifoliate leaves counted and the separate portions oven-dried and weighed.

(ii) Nodule relationships. The number distribution and colo of the nodules were recorded. Counting was done with the root separated under water in glass dishes. Total nodule numbers, the proportion of red nodules, i.e. (actively fixing nitrogen) and the numbers of nodules along the tap root, were counted. Colouration was a subjective estimate, a pink tinge showing against a grey background; while the large nodules had a distinct red colouration. During the earlier liftings, the nodules in cases of doubt, were inspected and counted under a low-power, vinocular microscope.

(iii) The distribution and morphology of the root system - Th root systems were drawn directly from an image onto graphed drawing paper. The sectioned root system was teased out and t individual roots separated until all the roots were in the one plane on a square glass plate. A similar plate, hinged to the first was then lifted over and the roots remained mounted be-tween the two glass plates. This mounted root system was then placed in a lantern slide projector and the image projected on graph paper pinned to the wall. With focusing, a clear, defin outline was obtained that could easily be traced directly onto the paper. Any desired magnification can be obtained, X 4.5 being considered quite suitable for the purpose desired. A centimetre scale was drawn on celluloid and projected onto the screen for scale comparison. At the final liftings the root systems, especially of the pelleted plants, were quite extensiv and sometimes half the root system at a time was projected.

Routine care and notes - The plots were weeded at intervals. Spurrey (*Spergula arvensis*) and mouse-eared chickweed (*Cerastium glomeratum*) were the principal weeds. There was no need to water the plots.

A visual assessment of establishment and early growth was made four weeks after sowing and the plants classified into three classes.

Observations were made as to whether the pellet traces remained on the number of stolon roots, and the difficulty or otherwise of disentangling the roots from organic debris.

CHAPTER X.

RESULTS - ROOT STUDIES

ESTABLISHMENT

The treatment effects showed up within a short time of sowing. The pelleted clovers were obviously more developed than adjoining control plants. Figure 14 shows the contrast between pelleted and control rows.

The establishment and development of the sown plants were assessed visibly four weeks after sowing. Three stages of development were recognized and the clovers placed in one of the three classes. (Table XI gives the establishment assessment)

TABLE XI. Establishment assessment. Date: 1-5-50

Key to symbols - f = first true leaf
ts = first trifoliate leaf, unopened or partly opened.
t_2 = second trifoliate leaf, opened.

TOTAL COUNTS

Plot No.		f	ts	t2
Bp	Pelleted	-	1	9
	Guard plants	12	10	2
Ep	Pelleted	-	1	8
	Guard plants	6	19	-
Cb	Total plants	7	27	-
Fb	" "	11	24	-
Ao	" "	8	25	-
Do	" "	16	18	-

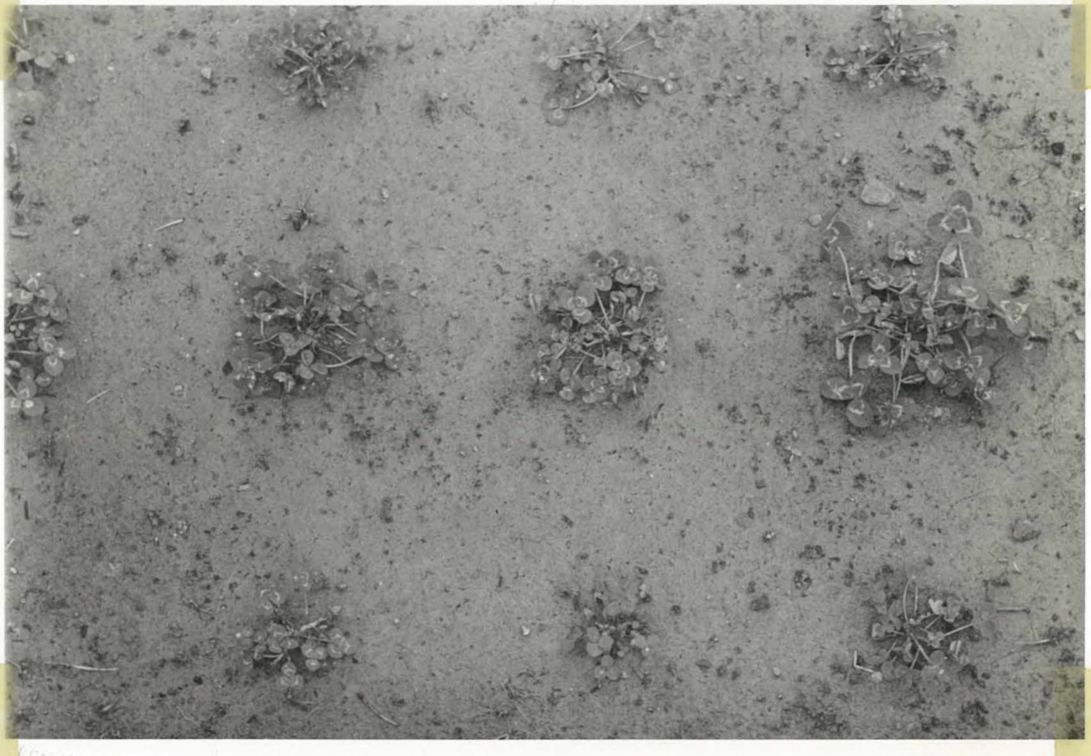


FIGURE 14. Plot Ep showing contrast between the pelleted plants (centre row) and much less vigorous control plants (front and back rows)
28/5/50

Classes as a percentage of the Total

Plot No.		%f.	%ts.	%t2
Bp	Pelleted	-	10	90
	Guard plants	50	42	8
Ep	Pelleted	-	11	89
	Guard plants	24	76	-
Cb	Total plants	21	79	-
Fb	" "	31	69	-
Ao	" "	24	76	-
Do	" "	47	53	-

The results are more striking when the counts are expressed as percentages of the total. In the two pelleted plots, 90% of the pelleted plants were in the 2nd trifoliate leaf stage, while untreated control plants in the same plots still had only the 1st trifoliate leaf. The broadcast and control plots had no plants i the 2nd trifoliate leaf stage - approximately 70% of the plants had the 1st trifoliate leaf partly opened, and the remainder had still the first leaf. The earlier growth and establishment of th pelleted plants is clearly illustrated, and is in striking contra to the results from the pot trials where the treatments had littl effect. (Compare with Table IV - Establishment Assessment of Pot Trials)

HERBAGE YIELDS

The herbage yields from the three treatments obviously diffe ed. (Detailed result figures are recorded in Appendix VI)

The last liftings of the two replicates were analysed as two simple experiments with differing numbers in the sub-classes.

Replicate 1 comprised plots Ao, Bp and Cb, and Replicate 2 plot Do, Ep and Fb. This same form of analysis was used in testing treatment effects on root weights and nodule numbers.

TABLE XII. Root Studies - Analysis of Variance of Herbage Yie

Rep.	Source of Variance	Degrees of Freedom	Sum of Squares	Mean Square	F Variance ratio	F 5% ratio reqd.	Significance
Rep. 1	Tr	2	31877	15939	10.87	4.46 (8.65)	ss
	Error	8	11733	1466.6			
	Total	10	43610				
Rep. 2	Tr.	2	192627	96314	37.61	4.74 (9.55)	ss
	Error	7	17924	2560.6			
	Total	9	210551				

ss = significant at the 1% level

Coefficients of variation - Replication 1 = 34.2%

Replication 2 = 26.8%

The variance due to treatments was highly significant in both replications. The differences between means were tested using Student's 't' test (Snedecor, 1946)

TABLE XIII. Root Studies - Table of differences between means of herbage yields.

Block	Treatment	Mean Yields (mg.s)	Tr. differences	Differences reqd. d.05	Actual differences between means	Significance
Rep. 1	O	62.0	P vs.B	72.1 (104.91)	78.0	s
	P	192.3	O vs P or B	64.5 (93.8)	130.3 (P - O)	ss
	B	114.3			52.3 (B - O)	-

TABLE XIII (Cont'd.)

Block	Treat-ment	Mean Yields (mg.s)	Tr. differences	Differences reqd. d .05	Actual differences between means	Signific
Rep. 2	O	93.8	P vs.B	97.7 (145.2)	296.7	ss
	P	401.0	O vs.P or B	91.4 (135.2)	307.2(P-O)	ss
	B	104.3			10.5(B-O)	

Figures in brackets refer to 1% level

s = significant at 5% level

ss = significant at 1% level

Results derived from the table of means are -

(i) Pelleting gives significantly greater herbage yields than broadcasting or control at the 1% level.

(ii) Broadcasting is significantly better than control at the 10% level, i.e. the probability of getting the above means for B and C on the hypothesis that there is no effect over O, is 90%

Figure 15 compares the average herbage weights of the treatments over the experimental period.

The graphed results show little difference between control and broadcast treatments with pelleting much superior.

When the logarithms of herbage yields are plotted against days from sowing, the rates of growth (as shown by the slope of the graphed results) are roughly constant over the three harvests for all three treatments. The three curves in Figure 16 are virtually the same, except that they have been shifted slightly along the horizontal axis. (The joining of the points by straight lines tends to hide this fact)

Although it is difficult to draw very definite conclusions

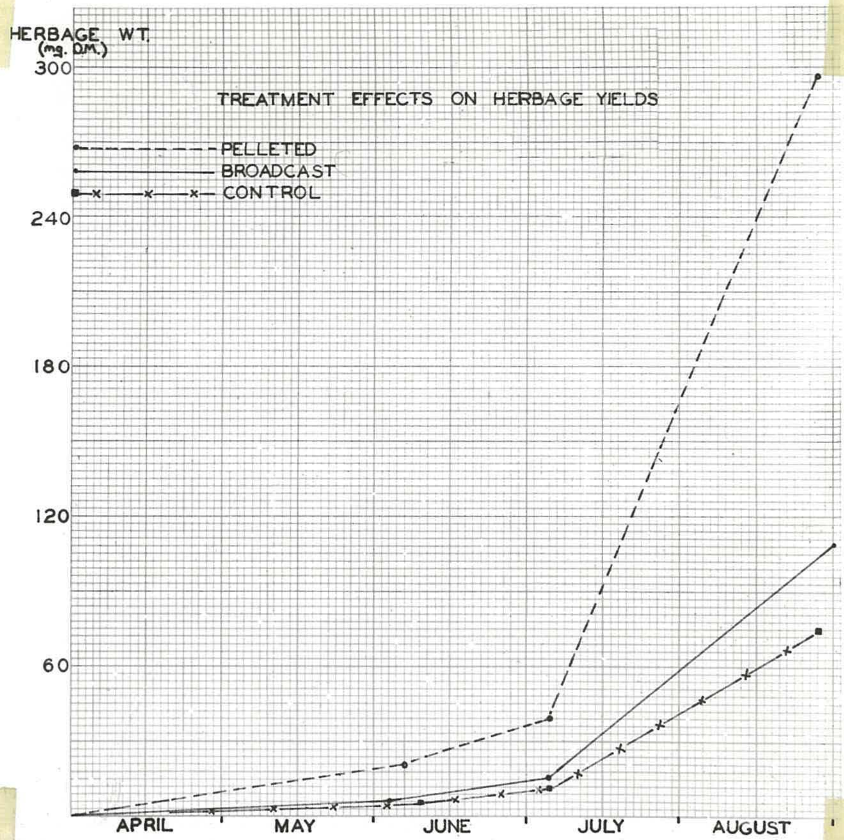


FIGURE 15. Graph of average herbage dry matter yields of the three treatments.

from only three harvests, the results are tentatively interpretated as follows:-

The pelleting, and to a lesser extent the half pelleted - half broadcast treatments, have given the seedlings an initial boost to growth, as shown by plant size at the first harvest. Thereafter the differences in growth are largely due to this initial boost.

ROOT WEIGHTS

The treatment effects on root weights and herbage yields were very similar.

The root weights at the last lifting were analysed in the same way as the herbage yields.

TABLE XIV. Root studies - Analysis of variance of root weights

Block	Source of variance	Degrees of Freedom	Sums of Squares	Mean Square	F variance ratio	F ratio reqd.	Si ni ce
Rep. 1	Tr.	2	3467	1734	8.9	4.46	[illegible]
	Error	8	1564	196			
	Total	10	5031				
Rep. 2	Tr.	2	27411	13706	11.4	4.74	[illegible]
	Error	7	8421	1205		9.55	
	Total	9	35832				

ss = significant at the 1% level

Coefficient of variation Rep. 1 = 27.3%

Rep. 2 = 45.8%

The variance due to treatments was highly significant in both replications. The differences between means were tested using Students' 't' test.

TABLE XV. Root Studies - Table of differences between means of root weights.

Block	Treatment	Mean root weights [illegible]	Treatment differences	Differences reqd. $d_{.05}$	Actual differ- ences between means	Sig ic- anc
Rep. 1	O	56	P vs. B	26.5	29.0	S
	P	79	O vs. P or B	23.5(34.2)	43(P-O)	SS
	B	50			14(B-O)	-
Rep. 2	O	39	P vs. B	67.1(99.0)	111	SS
	P	156	O vs. P or B	62.8(92.7)	117(P-O)	SS
	B	45			6(B-O)	.

Figures in brackets refer to 1% level

S = significant at the 5% level

SS = significant at the 1% level

Results derived from the above table are -

(i) Pelleting gives significantly greater root weights than broadcasting or control at the 1% level.

(ii) Broadcasting is not significantly superior to control.

Root weights and herbage yields respond similarly to the treatments. This would be expected from the interdependence o roots and shoots. (Figure 16 shows treatment effects on top an root growth)

Figure 17 compares the average root weights of the treatm over the experimental period. The plotted root weights compar closely with the herbage yields, i.e. little difference betwee broadcasting and control, with pelleting much superior.

The results can be interpreted in the same way as the

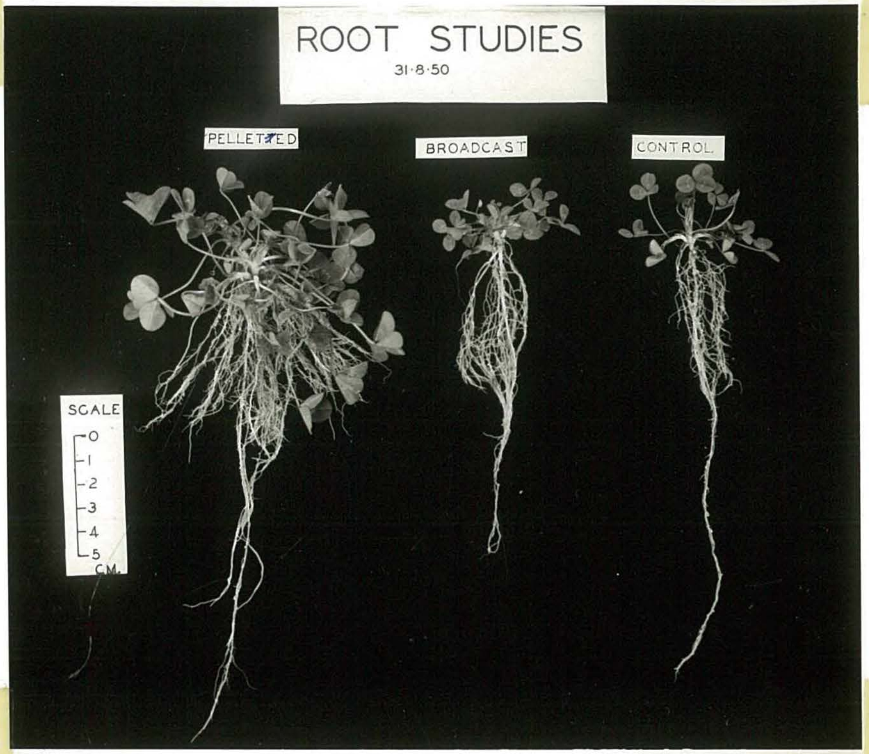


FIGURE 16. Treatment effects on top growth and root yields, 21 weeks after sowing.

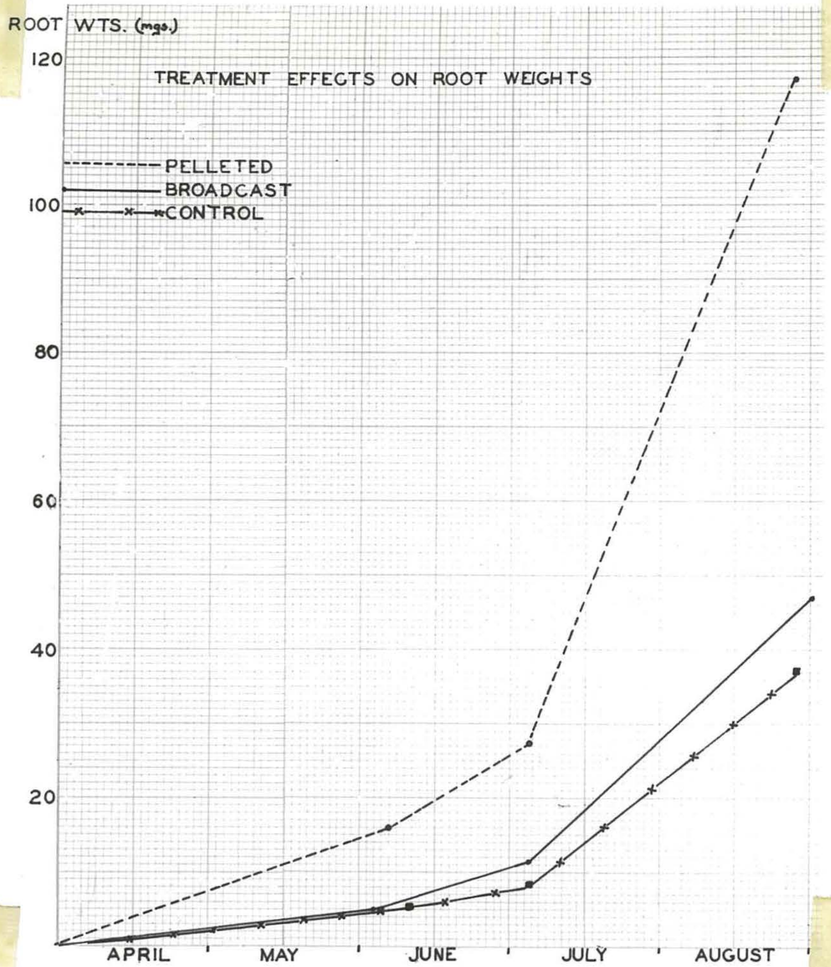


FIGURE 17. Graph of the average root weights (mg.s D.M.) of the three treatments.

herbage yields, i.e. the pelleting, and to a lesser extent the broadcasting treatments have given the seedlings an initial boost to top growth and root weight. The differences in root weights at later dates are largely due to this initial advantage.

There was no observable difference between treatments at two weeks.(see figures 18 and 19) At four weeks there were marked differences in the root systems - the pelleted plants had larger and more branched roots. (see Appendix VII) Thus, root response to pelleted superphosphate became apparent between two and four weeks from sowing.

There is this initial lag while seedling growth is dependent upon stored seed reserves, until the seedling roots have sufficiently developed and the plant is able to absorb soil nutrients. The herbage response is similar - observable differences only becoming apparent at four weeks. The uptake of phosphate by the pelleted plants may have been earlier than this, a delay occurring between the uptake of phosphate and a visible response.

(Appendix VII contains scale drawings of the root systems and shows the treatment differences at the various lifting dates.)

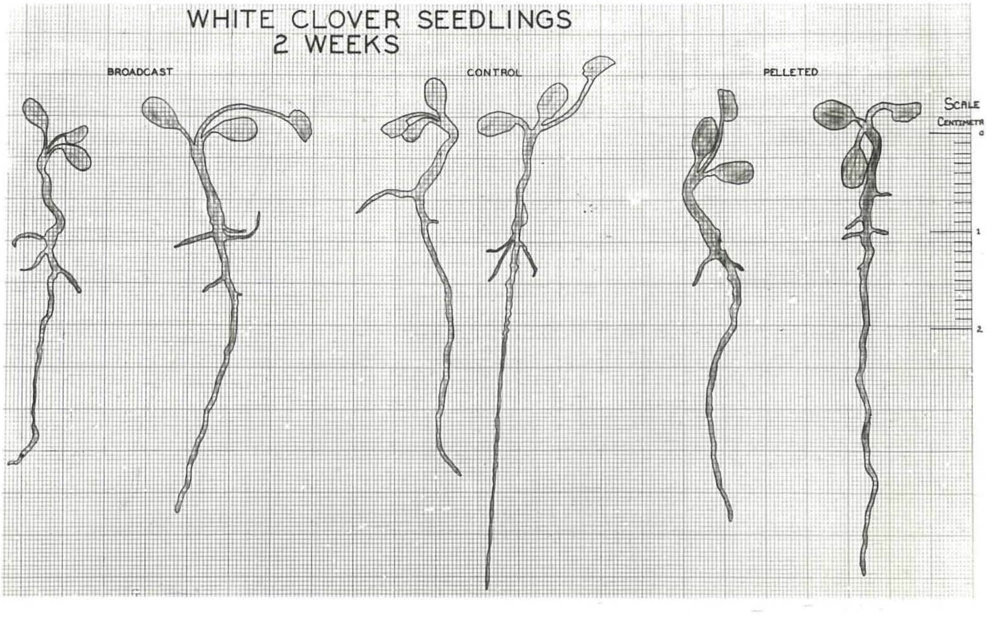


FIGURE 18. Drawings of pelleted, broadcast and control plants at 2 weeks. At this time there were no differences between treatments.

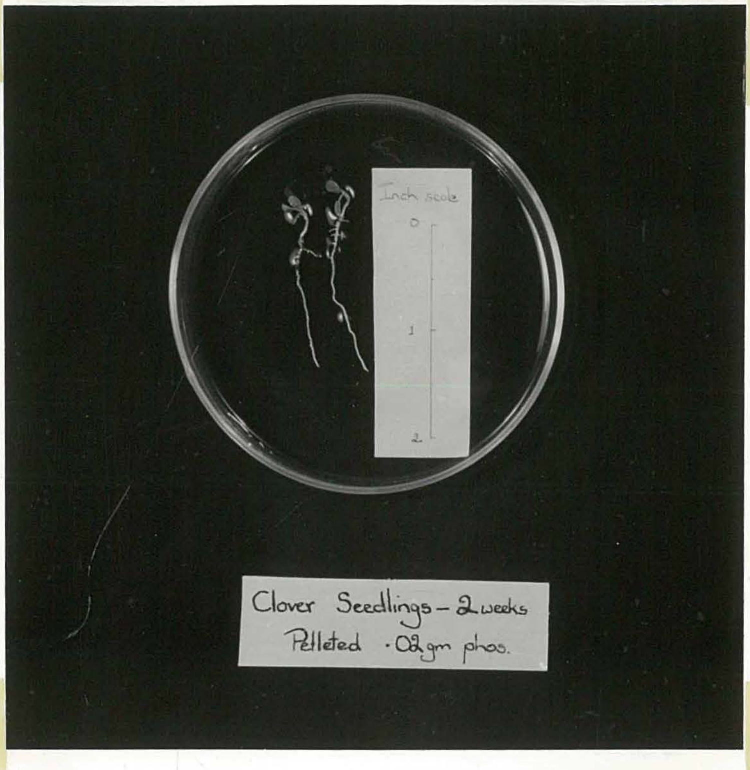


FIGURE 19. Pelleted seedlings at 2 weeks X 0.6 magnification.

ROOT/SHOOT RATIO

Conflicting views have been presented that phosphate fertiliser increases top growth in proportion to root growth, and vice versa. The root/shoot ratios of the lifted plants were examined to see whether there was any treatment effect on this ratio.

The pelleted, broadcast and control values fell at random along a line of general slope and formed a single population distributed as in Figure 20. The individual treatments did not deviate from this mean. The small quantity of fertiliser pelleted (0.02 gm. per seed) does not cause a differential response in either top growth or roots.

The correlation between shoot weight and root weight was calculated. Figure 20 shows considerable deviation of the young seedlings at low shoot and root weights. Apparently this characterises the transition from seedling growth and the use of nutrients stored in the seed, to a balanced development of root and shoot in the growing plant. The radicle emerges first during germination. Development of the leaves then follow, as the photosynthetic organs are necessary to manufacture the carbohydrates required for further growth. After the initial period the roots and shoots apparently develop in balance.

This initial divergence is shown in comparing the correlation coefficient for all root and shoot weights, with the correlation coefficient from data with shoot weight less than 20 mg.s.

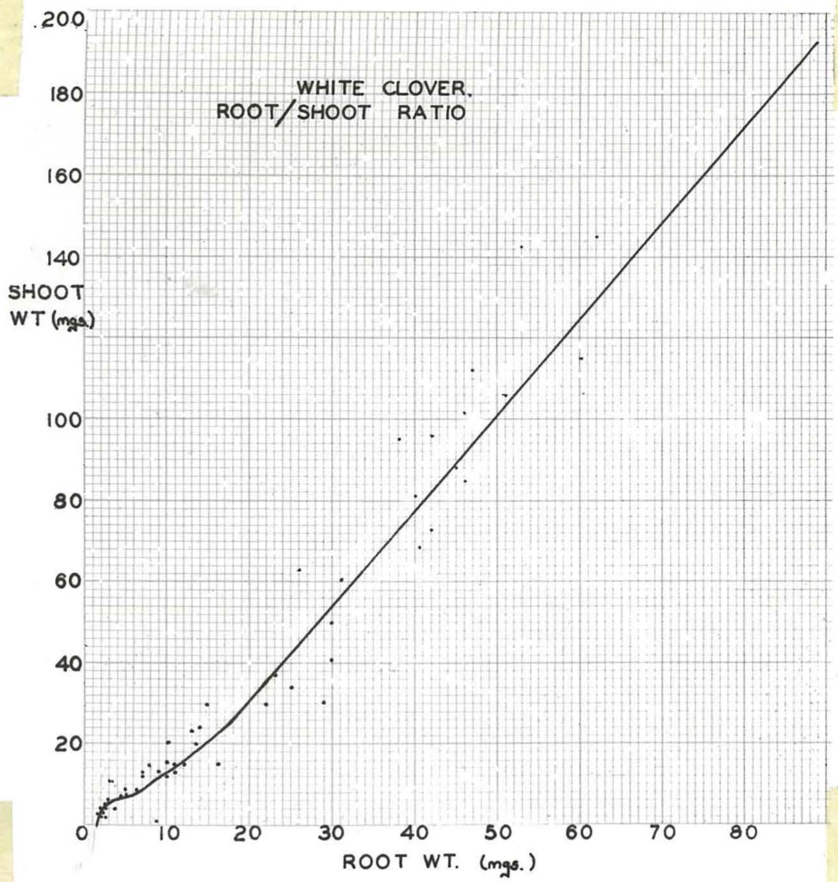


FIGURE 20. Graph showing relationship of white clover root and shoot dry weights.

$r = + 0.978$

Note deviation of young seedlings at low root and shoot weights.

r (roots and shoots) = + 0.973 SS.

r (shoot wts. less than 20 mg.s) = + 0.773 SS.

SS = significant at the 1% level. That is, over th total range, roots and shoots are highly correlated, with the very small seedlings the degree of correlation is less.

The treatments are compared in Figure 21. The three plan although differing greatly in size, have the same proportion of top and root growth.

LATERAL ROOT MORPHOLOGY

The root systems of the lifted plants were studied to see whether the localized placement of superphosphate resulted in surface concentration of roots, or stimulated lateral root initiation.

The root systems were drawn on paper as described in the section on "Methods". The treatments could then be compared later at leisure. Appendix VII contains scale drawings of the root systems. The roots of the pelleted plants are obviously larger and more developed and appear to have greater lateral r branching.

The treatment effects on lateral root distribution were compared. It was felt that any response due to the pelleting of the superphosphate would be evident in the surface layers. (The fertiliser pellets were placed on top of the soil) Hence measurements were restricted to the area within 1 centimetre radius from the junction of the 1st lateral root and tap root. The numbers of lateral roots in the top centimetre of the root

FIGURE 21. Treatment effects at 10 weeks after sowing. Illustrates greater growth and development of pelleted plant, but the root/shoot ratio is constant for all three plants.

system were used to measure root initiation.

The treatments were compared only between plants of equal root weight, otherwise the differences in root numbers between the large pelleted plants and the smaller broadcast and control plants, would merely reflect differences in root size.

A preliminary count was taken of the lateral roots of plant of equal root weights within the treatments to determine the variation when there is no treatment effect. (See Table XVI)

TABLE XVI. Lateral root numbers of plants of equal root weights

Treatment	Root Weight (mg.s)	(i) No.of lateral roots along tap root	(ii) No.of branc lateral roo
Pelleted	10.8	8	3
	10.7	9	18
	13.5	9	2
	13.5	11	5
	14.4	7	1
	14.7	4	11
Broadcast	4.9	5	1
	4.9	6	3
	16.3	6	4
	16.8	7	6
	4.5	4	7
	4.6	4	3
Control	4.4	5	0
	4.3	7	4
	3.7	6	0
	3.7	6	2
	3.9	5	0
	11.7	6	1
	11.1	7	3

Key - (i) Number of lateral roots along the top cm. of the

tap root (taken from the junction of the 1st lateral root and t[illegible]
root).

(ii) Number of secondary branch roots arising from the lateral[illegible]
in (i).

The table shows there is good general agreement of the primary lateral roots, (i), between plants of equal root weight, and fair agreement of the branch, lateral roots. The pelleted plants show the greatest divergence in both cases. The samplin[illegible] variation is high.

Bearing in mind this expected sampling variation, the treat- ment effects on root distribution were compared using plants of equal root weights. (see Table XVII)

<u>TABLE XVII</u>. Treatment effects on lateral root numbers of plants of equal root weights.

Treatment	Root weight (mg.s)	No.of lateral roots (i)	(ii) No.of bran[illegible] lateral ro[illegible]
P	12.1	6	15
B	12.6	7	5
O	12.6	4	11
P	14.7	5	12
B	15.0	5	2
P	19.5	8	23
B	19.0	5	1
P	22.6	5	11
B	23.5	8	2
P	29.3	8	43
O	30.0	8	2
P	25.1	7	32
O	26.0	7	7

Key = (i) Number of lateral roots along the top cm. of the tap root.
(ii) Number of secondary branch roots arising from the laterals in (i).

The results derived from the above table are (a) the treatme do not alter the number of primary lateral roots in the surface layers,

(b) the pelleti of superphosphate results in increased branching of the primary lateral roots.

The increased number of absorbing rootlets in the concentrat fertilized zone is probably one reason for the superiority of the pelleted plants.

NODULATION

The nodulation data is separated into four sections:

(i) nodule numbers

(ii) colouration

(iii) distribution

(iv) current theories on nodulation.

Nodule numbers - It was noticed at the earlier liftings that the pelleted plants were nodulated while the broadcast and control pla had none. The nodule numbers were counted at each lifting and differentiated into total number of nodules, number along the tap root and number of red nodules.

Figure 22 compares the average nodule numbers of the treatme over the experimental period. There are large obvious differenc between treatments, and the results are similar to the herbage yields and root weights. There is a difference however in that th treatment curves for nodule numbers, especially pelleting, flatte out slightly in June. This flattening of the curve is not correl- ated with root or shoot growth, and may be due to the effect of th

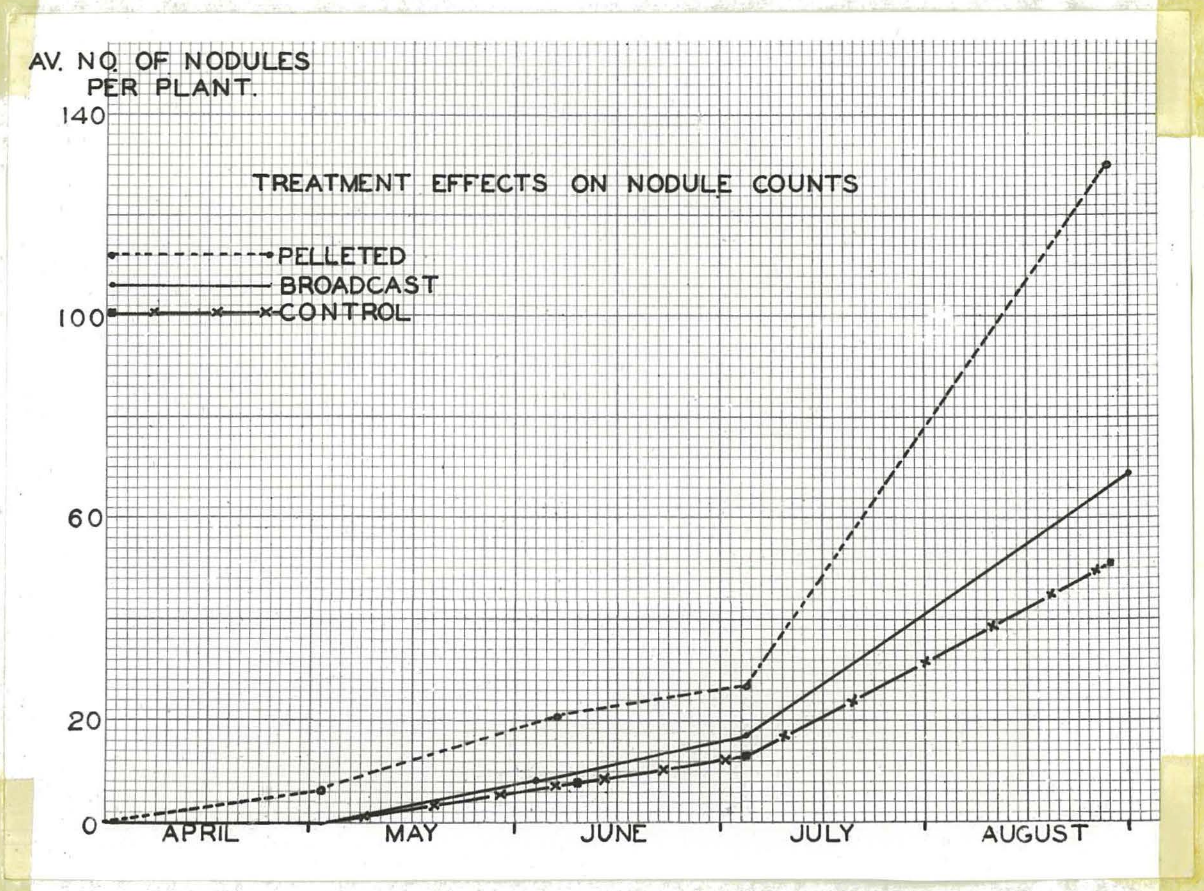


FIGURE 22. Graph comparing treatment effects on nodule numbers. Note slight flattening of graph in June, probably associated with the effects of the low temperatures on the Rhizobia organisms.

lower temperatures on the Rhizobia organisms.

The nodule numbers at the last lifting were analysed in a similar way to herbage yields and root weights.

TABLE XVIII. Root studies - Analysis of variance of nodule numbers.

	Source of variance	Degrees of freedom	Sums of Squares	Mean Square	F variance ratio	F 5% ratio Reqd.	Signi: ifance
Rep. 1	Tr.	2	4912	2456	14.2	4.46 (8.65)	SS.
	Error	8	1385	173			
	Total	10	6297				
Rep. 2	Tr.	2	41169	20585	38.5	4.74 (9.55)	SS.
	Error	7	3748	535			
	Total	9	44917				

Figures in brackets refer to the 1% level.

SS = significant at the 1% level

Coefficients of variation - Replication 1 = 20.6%

Replication 2 = 21.8%

The variance due to treatments was highly significant in both replications (see Table XVIII). The differences between means were tested using Students 't' test.

TABLE XIX. Root studies - Table of differences between means of nodule numbers.

Block	Treat-ment	Mean Nodule numbers	Treatment differences	Differences required. d.05	Actual diff. between means	Signif icance
Rep. 1	O	43	P vs.B	24.7	25	S
	P	94	O vs.P or B	32.1(32.2)	51(P-O)	SS
	B	69			26(B-O)	S
Rep. 2	O	61	P vs.B	44.8(66.1)	134	SS
	P	203	O vs.P or B	41.8(61.9)	142(P-O)	SS
	B	69			8 (B-O)	-

Figures in brackets refer to 1% level

S = significant at 5% level

SS = significant at 1% level

Results derived from the table of means are -

(i) Pelleting gives highly significant increases in nodule numbers over broadcast and control treatments.

(ii) Broadcasting gave a significant increase in nodule number over control only in Replication i.

These results follow the same pattern as the herbage yiel and root weights. Nodulation occurs within a two phase syster bacteria and plant, and plant growth and nodule numbers are inter-related. The correlation between nodule numbers and roc weight was calculated;

$$r = + 0.933$$

i.e. nodule numbers and root weight are highly correlated. Figure 23 shows the correlation between nodule numbers and roc weight.

Whether phosphate fertilisation had a direct effect on nodulation was not apparent from the review of the literature. The pelleted plants were noticeably nodulated four weeks after sowing while most of the control and broadcast plant had no nodules.

Average number of nodules/plant - 4 weeks.

Control	0.03
Broadcast	0.03
Pelleted	5.80

The root systems did not differ greatly between treatments at this time, yet the nodule numbers differed as above. (Figure 2 compares a nodulated, pelleted plant at 4 weeks with a poorly nodulated broadcast plant.)

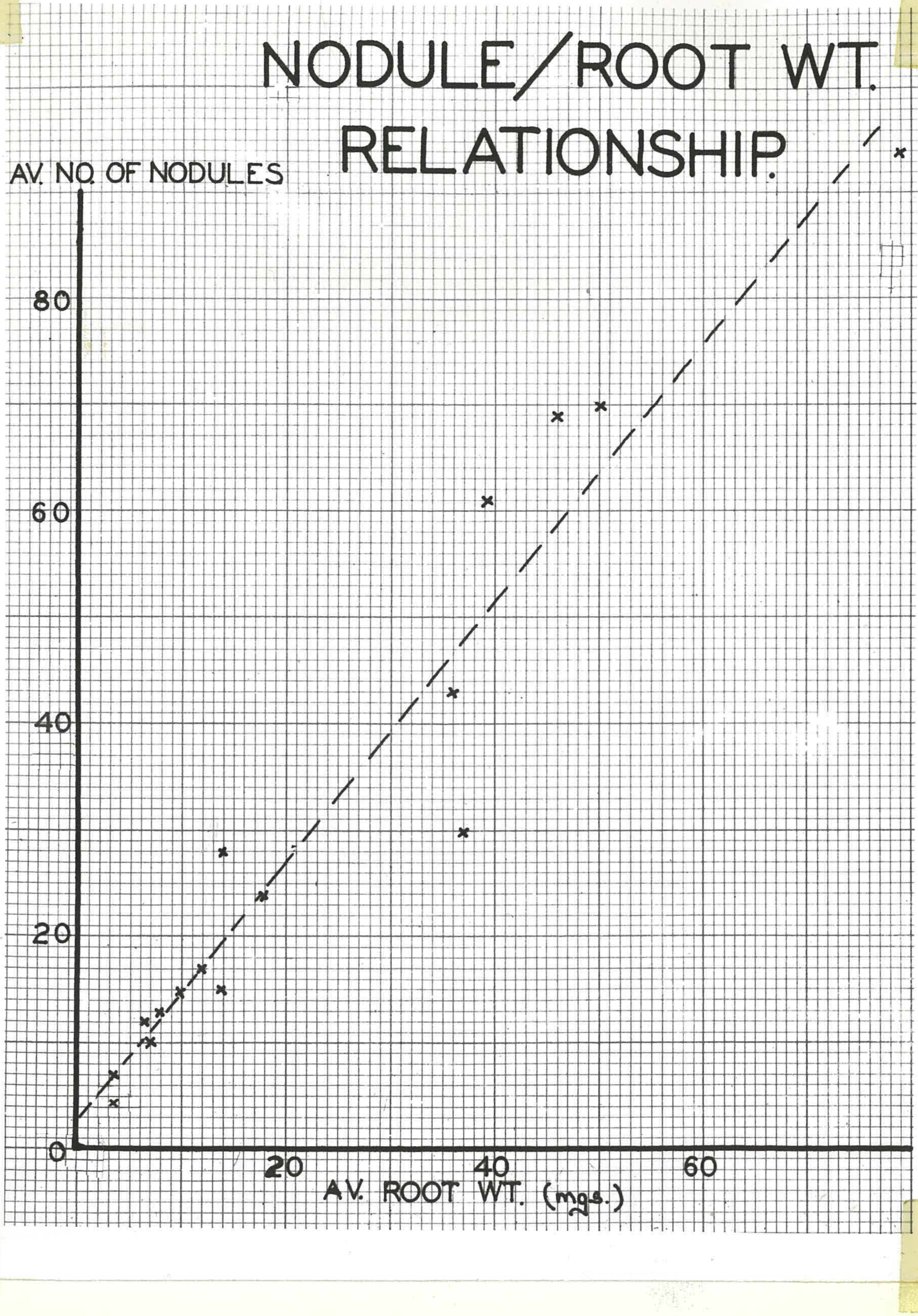


FIGURE 23. Showing correlation between nodule numbers and root weights.

$r = + 0.933$

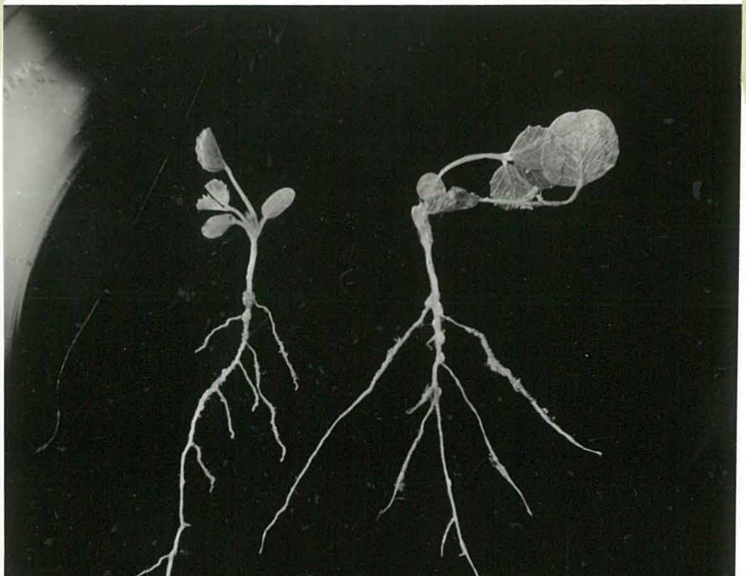


FIGURE 24. Broadcast plant on left, pelleted plant on the right at 4 weeks. Note conspicuous nodules on the pelleted plant, and only one nodule on the broadcast.

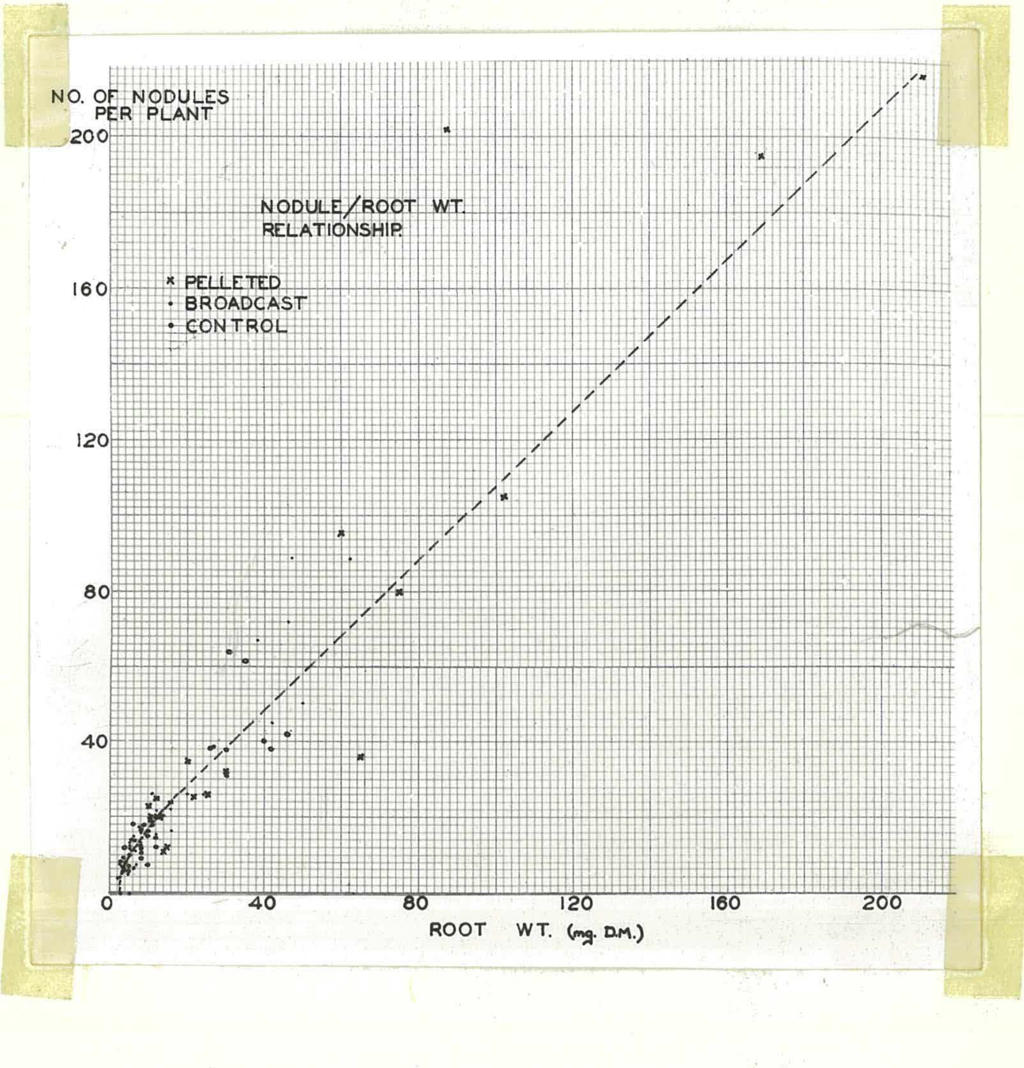


FIGURE 25. Showing nodule/root weight relationship of the treatments - no treatment differences.

Although pelleting of superphosphate increased nodulation of the young seedlings, and at the final lifting pelleting gave a highly significant increase in nodule numbers; comparisons betwee plants of equal root weights over the experimental period show no treatment effect on degree of nodulation. Figure 25 compares the nodule-root weight relationship of the treated plants, and shows that the three treatments form a single population randomly distributed round a general mean. The average number of nodules pe mg. of root over the experimental period showed no treatment effe on degree of nodulation, viz:

Pelleted	plants	averaged	1.1	nodules/mg.root		
Broadcast	"	"	1.5	"	"	"
Control	"	"	1.5	"	"	"

In conclusion, the effect of pelleted superphosphate on nodule numbers appears correlated with treatment effect on root growth, $\Upsilon = 0.933$. The degree of nodulation is unaffected by the treatments except at the very early stages, when the pelleted plants were more nodulated. This early nodulation may be a partial explanation of the early growth increase of the pelleted plants.

NODULE DISTRIBUTION

Trumble in "Blades of Grass" states that with effective organisms the nodules are clustered near the upper portions of th tap root - with ineffective organisms the nodules are more widely scattered over the secondary branches of the root system.

It is obvious that during early growth all nodulation will occur on the tap root. As the root system develops and branches, the tap root will decline in importance. This is shown in the nodule counts. At the early liftings most of the nodules occur along the tap root. The pelleted plants had a lower percentage of tap root nodules, showing a more developed and extensive root system.

TABLE XX. Nodules along tap-root as a % of total nodules.

	Pelleted	Broadcast	Control
6/6/50	72	86	84
27/8/50	16	31	35

At the last lifting, all treatments had much branched root systems (shown by low figures for tap root nodules %s) but the relative positions were the same, i.e. pelleted plants had a lower % of tap-root nodules.

NODULE COLORATION

It is considered that the reddish coloured nodules are those actively fixing nitrogen. (See review of literature) The number of red nodules per plant were counted at each lifting.

Except for the first lifting the pelleted plants showed a higher percentage of red nodules.

TABLE XXI. Red nodules as a % of total nodules.

Date	P	B	C
2/6/50	24	35	29
4/7/50	61	40	56
30/8/50	79	68	67

This could indicate that the placement of superphosphate stimulat nodule activity and efficiency. However, the examination of the data on the basis of comparing plants of equal root weights show that this treatment difference was largely due to differently sized root systems.

Comparing pelleting and broadcasting for 5 pairs of plants of equal root weights, the red nodules as a percentage of total nodules were:

P 59%

B 53%

Similarly comparing pelleting and control for three pairs of plants of equal root weights, the percentage red nodules were:

P 61%

O 72%

It is seen from Table XXI that the proportion of red nodul increases with length of time from sowing, i.e. plants with bigger root systems have a higher proportion of active nodules. Appendix VII shows that the pelleted plants as a series had larger root systems than the control or broadcast plots.

Eliminating differences due to root size, the localized pla ment of fertiliser did not affect the efficiency of nitrogen fixation as measured by the number of red nodules.

CURRENT NODULATION THEORIES

The data on nodule numbers and lateral roots was used to test Nutman's theory of nodulation, i.e. that lateral roots and nodules are physiologically homologous and that infection by th bacteria can only take place at predetermined foci on the root corresponding to the sites of initiation of lateral roots.

Appendix I considers the results from the root studies in the light of this theory of nodulation.

CHAPTER XI

DISCUSSION

General:

The need to check the progressive deterioration of much of the New Zealand hill-country is the purpose behind these trials evaluating the pelleting of clover seed and fertilisers. This new technique - the pelleting of seed and fertiliser, may be valuable as a method of introducing the essential clovers into the hill country pastures (possibly by aerial sowing).

Much of the 10½ million acres of North Island hill country has the problem of low or declining stock carrying-capacity. T present situation is that the original store of fertility has long since been spent. Many of the pasture plants that can respond to increased fertility and produce the feed needed for higher stock-carrying have died out. The pastures are uniforml poor and deficient in clovers.

Trials (87) have shown that the chief factor in increasing pasture production is the clover plant. There is little respon from phosphate topdressing unless clovers are present in the sward.

The first phase in improving the hill country is to establ clovers in the pasture, mainly through surface-sowing. Over-sowing trials with clovers have resulted in variable success - "in all cases, establishment compared with number of seeds sown is extremely poor." (7) One objective was to see whether by pelleting the seed and fertiliser, economy in seeding rates could be effected.

The pelleting of seed and fertiliser may be considered a special case of fertiliser placement. The fertiliser surrounds the seed in coats of varying thickness. According to the metho of manufacture, the shape and size of the pellets, quantities and kinds of fertiliser added, and the inclusion of other materials, may vary.

Pelleting of seed with fertiliser or other inert materials is still new. Commercial firms in the U.S.A. have incorporated fungicides, insecticides, growth regulating substances etc., as well as fertiliser in the pelleting of vegetable, crop and gras seeds.

The advantages of the pelleting of seed and fertiliser wer thought to be:

(i) more efficient use of fertiliser

(ii) earlier and better establishment of seedlings

(iii) economy in seeding rates, and

(iv) greater ease of aerial sowing

Disadvantages would be:

(i) the technical costs of pelleting

(ii) reduced germination due to fertiliser injury, and

(iii) possible "burn" of the seedling roots due to localized concentration of the fertiliser

The unsatisfactory distribution of seeds sown from the air (22, 23) results from the differing ballistic properties of the seeds. The different densities, sizes and shapes of clover see are reflected in a band distribution. (21) Uniform pellets would overcome this difficulty. The aerial sowing of pelleted seed then is advantageous. If it could be shown that pelleted seed resulted in more effective use of the fertiliser and super ior seedling growth, then this technique would merit serious consideration in the restorative treatment of the unploughable hill country.

No critical work evaluating the pelleting of seed and fertiliser has been published. That was the objective of this research - to determine the advantages and disadvantages of the pelleting of clover seed and fertiliser; to compare pelleting fertiliser and broadcasting experimentally, and to study the plant's response.

PRELIMINARY PELLETING TRIALS

Because of the lack of published data, the first need was to find the maximum amounts of fertiliser which could be pelleted with clover seed without seriously impairing germin- ation. The germination injury and extent of radicle deformati due to the concentration of fertiliser were determined, as wel as the effects of the different fertilisers on the physical properties of the pellets.

The pellets were made by hand. This permitted absolute control of the amount of fertiliser per pellet but was slow ar laborious. The various large-scale commercial methods of pelleting were impractical, and the alternative methods listed by Saxby (9) were discarded as they did not control the quanti of fertiliser per individual pellet, i.e. single plant (or pel et) comparisons would not be valid. In the early trials some seed damage was caused by too great a compaction.

The pellets contained equal quantities of fertiliser and bentonite clay. This dilution of the fertiliser aided in the measurement of the small quantities required, and helped reduce the injury due to the localized concentration of fertiliser.

Points kept in mind were:-

(i) An advantage of pelleting might be the supplying of the clover seedlings' phosphate requirements for the initial year in pelleted form. This would save the additional topdressing otherwise required. Thus the quantities of fertiliser per pellet were the maximum possible without causing serious germination loss.

(ii) The amount of inert clay was kept to a minimum, otherwise any advantage resulting from the saving in quantity of fertiliser required in aerial sowing would be counter-balanced by the amount of inert material carried.

Saxby (9) was concerned in his paper with the physical characteristics of the pellets. The qualities required are;-

1. Resistant to breakage from shock and abrasion.
2. Addition of adequate fertiliser to supply the seedlings' requirements.
3. Ability to disintegrate under weathering to allow germinati
4. Local concentration of fertiliser salts round the seed to b low enough to avoid germination injury.
5. The pellet to be mainly phosphatic with a minimum of inert material.

The method used determines the choice of materials and the ease and practability of making pellets with the above characte istics. Commercial firms in the U.S.A. using a positive binder such as methylcellulose in the coating are able to apply successive layers of different materials. There is more flexib ity in the choice of raw materials in the extrusion or compress ion methods of pelleting compared with tumbling in a rotating drum.

The method of pelleting used in this work, simple compression, formed pellets of satisfactory physical properties. They were fairly shock-proof (could be dropped onto concrete without breaking) and disintegrated fairly readily under weathering.

Solubility tests showed that the sulphate of ammonia in t N pellets was completely soluble. The superphosphate in the P pellets was 72% soluble (running extraction), and the pellets containing 50% sulphate of ammonia and 50% superphosphate had intermediate solubility.

The stability of the pellets appears related to the proportions of soluble constituents. There is variability among pellets due to variations in compaction, but the east of disintegration was N > PN > P. This trend had been noted in the field.

The major defect of pelleting seed and fertiliser is the resultant lowering in germination. This hazard has been recog nized by other workers, but apparently the physiological natur of this damage is not definitely known. In this trial germin ation injury ranged from 10-50%, while approximately 10% of th germinated seed had deformed radicles due to fertiliser "burn"

The amounts of fertiliser used per seed, were far greater than any recorded in the literature. The fertiliser levels chosen for pelleting were 0.01 and 0.02 gms. of fertiliser per seed. These gave reasonable germination counts, 60-90%, depending on the particular fertiliser and quantity used. It was believed that these levels of fertiliser would give a measurable response in yield trials.

American reports on pelleting (10) limit the quantities of fertiliser to very low levels in the avoidance of fertiliser injury - "With ammonium nitrate, 0.25% based on the weight of the seed will kill most plant germs. However ammonium sulphate has proved a safe nitrogen source for some seeds in concentrati up to 30% of the weight of the seed." (10)

Allowing 700,000 white clover seeds per pound, these level of fertiliser would result in 0.0002 gm. of sulphate of ammonia pelleted per seed. The rates of phosphate fertiliser used are similarly very low - equivalent to 0.00006 gm. superphosphate per pelleted white clover seed, (10% of seed weight). This supply of nutrients would be rapidly exhausted, but is said to exert a beneficial effect on growth and maturity rates during the 12-15 days after planting.

The levels of fertiliser (mixed with equal quantities of bentonite) used in this experiment were:

White clover - 31 times weight of seed

Subterranean Clover 3.3 times weight of seed

Saxby (9) found that serpentine superphosphate and basic slag caused little germination injury when pelleted with white and subterranean clovers. This was in contrast to pelleted sulphate of ammonia and superphosphate which reduced germinatio Details of his results are :-

<u>Saxby - pelleting tests</u>

White Clover (Serpentine superphosphate, 0.012 gm/seed = 76%
(germination.
(Serp. Super. + sulphate of ammonia, 0.004 gm/seed
(= 61% germination.

Subterranean Clover (Serpentine superphosphate, 0.078 gm/seed = 85% germination.
(Serp.Super. + sulphate of ammonia, 0.013 gm/seed (= 79% germination

Saxby's figures compare well with those recorded in this experim except that his nitrogen levels are much lower and in this case comparison is possible. The results obtained in this trial with white clover are:

P (superphosphate) 0.02 gm/seed = 90% germination

PN (0.01 gm.superphosphate + 0.01 gm sulphate of ammonia)/see (= 60% germination.

N (sulphate of ammonia) 0.02 gm/seed = 30% germination

It seems that the American figures for pelleted levels of superphosphate (0.00006 gm/seed) are well below permissible amounts, for white and subterranean clovers at any rate. Pellet superphosphate 400 times the U.S. quoted rates gave 90% germination in this experiment; and Saxby using 200 times the U.S.rate with white clover recorded 76% germination. The American figure however, may refer to more susceptible species. Legumes, as a group, are considered more tolerant to salt injury than cereals (16, 17, 18).

The germination injury increased progressively from P, N to PN pelleted, and agrees with U.S. reports. The germination loss would presumably be much greater if nitrates instead of sulphate of ammonia had been used as the nitrogen carrier. Two ways in which localized fertiliser concentration may lower germination are postulated:

(i) osmotic pressure retards or prevents absorption of water.

(ii) certain salts or ions may be toxic to the embryo or seedli

As the germination tests were carried out on moist filter paper in a saturated atmosphere, it is unlikely that there was osmotic effect of any magnitude. Toxicity of the localized concentration of fertiliser is probably the main cause of the reduced germination and abnormalities in the seedlings. A further factor may be a pH effect, or H ion toxicity.

The fertiliser effect varies with the soil moisture content. Germination injury would probably be greater on drier soils under field conditions than the values recorded in these germination tests under optimal moisture conditions.

Approximately 10% of the total germinated seedlings had deformed radicles and root injury. (Routine germination counts usually exclude any deformed or abnormal seedlings) Root "burn" may result from a lower fertiliser concentration than that which is capable of causing serious germination injury. The seedling radicle becomes stunted and malformed.

The retarding of germination by the pelleted fertiliser has an indirect effect on germination losses. The delay in germination allows the growth of saprophytic fungi which cause a secondary germination loss. This secondary loss may be considerable in the field. The incidence of fungal growth varied in the trials, and no exact estimate of its importance and occurrence could be made.

An unsatisfactory feature of the germination tests if the variability in the germination counts. Superphosphate, 0.02 gm. per seed, pelleted, varied from 35-90% germination in two tests 15 months apart. Uncontrolled factors have influenced the viability of the pelleted seed. One factor may be the incidence of saprophytic fungi, another cause of variability may be differences in the moisture status of the germination pods.

It appears then, that much work could be done on seed injur resulting from localized concentration of fertiliser, and the factors involved.

POT TRIALS

The aim of this trial was to determine the efficiency of th pelleting of seed and fertiliser as compared with broadcasting.

Increased yields due to the placement of fertiliser result from increased uptake of the localized fertiliser by the plant, minimised losses by leaching and fixation. (75-90% of the phosph ic fertiliser added to soils is "fixed" in forms unavailable to plants.)

American work has shown that band placement of fertiliser i superior in many cases to broadcasting. A striking placement effect resulted from soaking grain seed in a phosphate solution. Utilisation of the phosphate was 60 times as efficient as when superphosphate was applied to the soil. Granulation of phosphat fertilisers is recommended by many workers. Granulation minimis fixation by reducing the surface area of fertiliser in contact with the soil.

Pelleting combines placement and granulation.

An experimental standard of four plants per square foot was used. This figure was the maximum establishment obtained in any of the oversowing trials. Four pelleted plants/sq.foot (0.02 gm fertiliser/plant) represents a dressing of only 7.7 lbs. of

superphosphate per acre. Even if the quantities of seed recommen
(7) for oversowing are pelleted, viz.

1 lb. white clover)
2 lbs. red clover) per acre
2 lbs. subterranean clover)

only 61 lbs. of superphosphate/acre is used, i.e. approximately quarter the usual dressing of 2 cwt./acre. The lowered quantity fertiliser required would save topdressing and transport charges, especially if the seed and fertiliser is sown by air; i.e. a plan with an equal load of pelleted seed could cover at least four tim the area sown broadcast with the usual quantities of fertiliser.

The effectiveness of pelleted superphosphate compared with broadcast fertiliser, increases with fewer plants per square foot (providing pelleting is not injurious). The broadcasting handic will result from the same quantity of fertiliser per plant being spread more lightly over a wider area. Thus the treatment result may differ at different plant densities, but the choice of four plants/square foot appears reasonable and practical.

Loss of data due to death of plants, accidents and abnormal causes is a normal experimental hazard. The usual precaution is replication of treatments. In these trials 7% of the total yield were missing at the completion of the experiment. This figure is high and emphasises the vulnerability of using single plant pots sown from seed as the treatment unit. Half of the 7% missing plo values were deleted because of abnormal yields due to virus disea This disease was presumably transmitted from infected neighbourin experimental plots. When working with a susceptible species, it

would be a wise precaution to be suitably isolated from other a
of that species.

The statistical examination of the experimental data showe that the only significant variation was that due to varieties. It is known that large varietal differences occur. The only interesting feature is that competition from the four ryegrass plants in the M series caused the white clover plants in this series to differ significantly from the white clover plants sow alone.

The treatment effects were more marked and showed less variability in the first yield cuts. There was a significant i crease in yield with each increase in level of superphosphate. Sulphate of ammonia had the opposite result. There was a trend for the highest N level to have a depressing effect. Many expe ments have shown the stimulating effect of phosphate fertiliser on clovers. The higher N level however, must retard or depress early seedling growth.

With surface-sowing of clovers it is doubtful whether a dressing of superphosphate sown at seeding benefits the young clover seedlings. It is probable that the established grasses with well-developed, extensive roots are able to use most of th available phosphate before the seedling clovers are established This is shown by the treatment differences which are only signi ficant in the M series 1st cut. In this case, the competition from the ryegrass seedlings emphasises the advantages of pellet ing and the quicker establishing grasses compete for and take up most of broadcast fertiliser, depriving the clover plant.

The highest yielding treatment was 0.02 gm. of superphospl /seed, half pelleted and half broadcast. 0.02 gm. of superphosphate pelleted/seed yielded less than the lower level, 0.01 gm/seed, pelleted. This relative depression in yield is probal the reason for the superiority of the P_2 pb treatment, 0.01 gm. of superphosphate per seed pelleted and 0.01 gm. broadcast.

The general trend of results were as expected, i.e. pellet and half pelleted - half broadcast treatments appeared superior to broadcasting, except with the highest level of sulphate of ammonia. In this case pelleting yielded the least.

The results from the root study plots do not agree with these trends. There pelleting was much superior, and there was little difference between pb and control. The pot trial was so a month earlier than these plots and the drier conditions may have handicapped pelleting at the higher fertiliser levels.

The question arises as to why results from the pot trial generally were non-significant, whereas data from the 1st cuts of the Ac and M series gave significant results, and the root studies showed treatment differences.

There are two reasons why the 1st cuts show significant results:

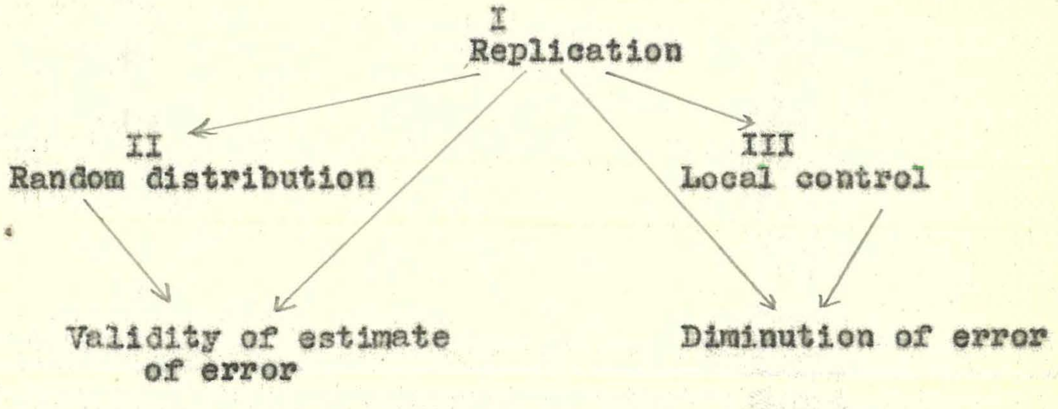
(i) the yields are small, and heredital and environmental variation does not mask the treatment effects.

(ii) the pelleting and half pelleted - half broadcast treatment give an initial boost to seedling growth. Thereafter differences in growth are largely due to this initial boost. Thus treatment effects are more pronounced at the early stages.

There may be four explanations of the general non-significance of the pot trial results.

(i) Poor layout and design. The experiment was limited to 200 eight inch pots, the total number available. The design o: the trial therefore was a 3 X 3 X 3 X 5 factorial with one replication. It is now realized that a better layout would ha been to simplify the experiment, - to replicate certain treatments at the expense of some of the desired information. The layout used may have been sufficient had the uncontrolled vari ion been smaller and treatment differences more marked. (The precaution was taken of calculating the coefficient of variati of a previous thesis (85) using the same pots, before this experiment was laid down).

Fisher (86) gives a diagram to show experimental design relationships.



Concerning error control he states: "the differences between plots of a single treatment in a replicated experiment are par due to experimental error and partly to the average difference between replicates. The variance due to replicates is general removed from the error. The precision of the experiment becom greater when a large amount of the total variability can be removed in this way."

(ii) Poor experimental technique - Careless or inaccurate wo may be one cause of a large error variability. In this trial all the influencing factors were kept constant as far as possible; the pots were filled with a uniform weighed quantity of soil, the amount of fertiliser in the pellets were accurate to 0.001 gm., watering of the pots were uniform etc. The 7% missing values figure is high but as pointed out previously, half of the missing values represent abnormal results due to virus infection. This could not be foreseen. The remaining missing yields may reflect injurious treatment effects on single seedlings in pots.

(iii) Uncontrolled variation - The large experimental error masked any differences due to treatments. This large variation may be constantly associated with single plants grown from seed, but the dry conditions and sandy pots accentuated this variation. This is shown by comparing the 62% coefficient of variation figure in the pot trial with the results from the root study plots. In the latter case the seeds were sown one month later in plots of the same soil as used in the pot trial, but the site was damp and there was well distributed rainfall. The coefficient of variation

for Replication 1 = 34%

and Replication 2 = 27%

i.e. just half the error variation of the pot trials. In the root studies a minimum of three plants per treatment were samples in each replication at each harvesting. The value of replication and optimum conditions for seedling growth in reducing uncontrolled variation is thus strikingly illustrated.

The use of clonal material is one method of lessening heredital variation. Edmond (88) used clones of grass species and found a coefficient of variation of 38%. McNeur (85) using white clover in the same pots had a coefficient of variation of 30%. These values compare well with the variation in the root study plots, but are lower than the error variance of the pot tri

(iv) The fourth possible reason for non-significance of results is that the treatments had no effect. Pelleting and half pellete - half broadcast only resulted in a 20% yield increase over broad casting. The root studies and 1st cut in the M series however, showed pelleting superior. Pelleting yielded 100% more than broadcasting.

As significant treatment differences have been shown in supplementary trials, it is concluded that insufficient replicati and the large plant variation resulted in non-significance of the pot trial data.

It is worth while comparing the pros and cons of pot experiments. The advantage of using pots is one of scale. By the use of "identical" pots much of the soil heterogeneity can be removed and small-scale easily handled trials can be laid down - a far greater number of treatments and interactions can be compared. The use of pots allows greater experimental control - the plants can be grown in a glasshouse under controlled conditions, watered when necessary. Plots require a greater area and larger quantiti of seed, fertiliser and pellets. Data from pot trials and field experiments have been found to agree fairly well in fertiliser experiments. Small-scale pot trials, in preference to plots, were decided upon to compare the large number of treatments becau of the slow, tedious manufacture of the pellets.

The disadvantage of pot trials is the artificial conditions imposed - temperature, moisture and aeration differ from natural conditions and root growth in pots is abnormal. Results from po trials need to be applied in the field with caution and should be checked with field plot experiments.

Pot trials probably have a useful purpose as small-scale pilot trials.

ROOT STUDIES

The advantages of pelleting were thought due to increased seedling uptake of the localized fertiliser. Root characteristi and distribution were studied to try to understand in what manne the plant responded to the localized fertiliser placement, and w there was that response.

Plant root systems become modified with placement of fertil er. Jacquès (53) has shown experimentally that grass roots accumulate in the top fertile zone, resulting in reduced root penetration. The root penetration of white clover however, was little affected by superphosphate placement. Reports on granul- ation of superphosphate (32, 33) state that bunches of fine roo develop round the granules, and plant roots concentrate in thes zones of steady phosphate supply.

The distribution of lateral roots was therefore investigat to see whether pelleting caused localized root concentration.

The same methods, materials and soil was used as in the po trial so that results from the two sections should be comparabl

The treatment effects showed up within a short time of sow ing. There was no observable difference between treatments at two weeks. At four weeks there was marked differences between treatments in both leaf and root growth. The pelleted plants h larger and more branched roots, and greater leaf development. Thus the plant response to pelleted superphosphate was during early seedling growth, and because apparent between two and fou weeks from sowing. The uptake of phosphate by the pelleted plants may have been earlier than this, a delay occurring betwe phosphate uptake and a visible response.

The statistical analysis of the final harvest showed that pelleting gave significantly greater herbage yields and root weights than broadcasting or control. These trends were eviden over the whole experimental period, i.e. there was little diffe ence between control and broadcasting treatments, with pelletin much superior.

When yields are plotted against time, the three treatment curves appear virtually the same, except that they have been shifted along the time axis varying distances. The rates of growth of the three treatments (after an initial period) are roughly constant. The conclusion then, is that pelleting, and a lesser extent the half pelleted - half broadcast treatment, have given the seedlings an initial boost to growth, as shown plant size at the first harvest. Thereafter, the treatment differences are largely due to this initial boost. Each pellet contains only 0.02 gm. of superphosphate/seed. This small quantity of fertiliser may have been largely used by the first harvest.

The question arises, why there is this early boost at the seedling stage? Phosphate starvation is usually most pronounc during the early growth of plants. At that time, owing to reduced root development, the roots cannot take up broadcast fertiliser, or extend to and use the soil phosphate; and the immobile phosphate cannot move to the roots. This early phosph starvation is especially true if growing conditions at that tim are not ideal.

By pelleting 0.02 gm. superphosphate per seed, a fertilise concentration equal to 2½ cwt. of superphosphate/acre is obtain in the cubic inch around the seed. If, as is likely, the pelle ing effect is concentrated within a smaller area than 1 cubic inch, then the localized concentration of fertiliser over this restricted area will be more than 2½ cwt./acre. It is this small quantity of fertiliser, readily available to the seedling and equal (within a restricted area) to a heavy superphosphate dressing that gives the initial boost to the pelleted plants.

When given supplementary phosphate, legume seedlings rapid reach the stage when they are able to obtain their mineral need from the soil reserves.

The placement of the fertiliser in relation to the legume seedling is apparently very important. With band seeding of lucerne, Haynes and Thatcher (29) found that seedlings as littl as half an inch to either side of the fertiliser bands perform unfertilised plants.

It is interesting to compare the results obtained in these trials with work reported by Jacques (53), even although his placement positions differed from those used in these experimen Jacques found that white clover herbage yields showed little difference between surface placement (broadcasting of superphosph and mixing the fertiliser in the top six inches of soil, but fertiliser placed in a layer at six inches considerably increased yield. These results refer to nearly one year old plants and do not differentiate an early seedling response. However, a decided early growth response was recorded from surface application of superphosphate on grass, but this was not maintained for long and the total yield was lower than from sub-surface placement.

Conflicting views have been presented that phosphate fertilis ers increase top growth in proportion to root growth, and vice versa. The root/shoot ratios of the lifted plants were examined see whether there was any treatment effect.

In general it can be stated that the application of fertilis increases the weight of roots, but that the weight of the tops generally increases more in proportion, so that the top/root rati is generally higher in a fertile than in an infertile soil. Jacques (53) found that root weights differed from herbage weight in that there was less distinction between the fertilised and unfertilised series. High herbage yield was not necessarily associ ed with high root yields.

In this trial there was no treatment effect on root/shoot ratio. The small quantity of superphosphate pelleted, 0.02 gm/ seed, did not cause a differential response in either top growth or roots. Root and shoot weights were highly correlated, $r = + 0.978$. The degree of correlation was less for small seedlings, r (shoot weights less than 20 mg.) $= + 0.733$. The divergence at low values apparently represents the transition from seedling

growth dependent on stored seed reserves, to a balanced developmen
of roots and shoots.

The root systems of the lifted plants were studied to see whether the localized fertilizer placement had caused a surface co
centration of roots, or stimulated lateral root initiation.

It is known that roots tend to congregate in highly fertile regions at the expense of penetration and side growth. A surface concentration of roots in response to pelleted superphosphate then may prove disadvantageous when the placement effect of the fertili
has been exhausted, especially in dry conditions.

The technique of projecting and tracing the root systems is swift and accurate due to the X 4.5 magnification. It allows the handling of a considerable number of plants within a reasonable ti
The difficulty and laboriousness of studying root systems has previously helped in restricting root investigations - the small numb
handled and the considerable plant variation made the interpretation of reliable results difficult.

The treatment effects on lateral root distribution were compa
ed only between plants of equal root weight, otherwise differences in numbers of lateral roots would merely reflect differences in root size. The number of lateral roots in the top centimetre of t
root system were used to measure root initiation. Any response to the pelleting of superphosphate would be evident in the top layers (the pellets were placed on the soil surface), hence measurements were restricted to within 1 cm. radius of the junction of the firs
lateral root and tap root. The treatments did not alter the numbe
of primary lateral roots in the surface layers, but pelleting resu

ed in increased secondary branching of the primary lateral roots This increased number of absorbing rootlets in the concentrated fertiliser zone is probably one reason for the superiority of tl pelleted plants.

Jacques concluded that white clover differed from ryegrass and cocksfoot, in that surface fertility did not tend to cause root accumulation in the fertilized zone. (53) These trials however, have shown that localized placement of a small quantit of superphosphate tended to cause increased branching in the fertilized area. As the plants develop, this effect would prob be masked by general root growth, and it is unlikely that surfa concentration of roots in response to pelleting would ever pres a serious problem.

It was noticed at the earlier liftings that the pelleted clovers were nodulated while the broadcast and control plants h none. As the numbers and activity of the nodules are an index of nitrogen fixation, the numbers of nodules, coloration and di tribution were studied to see whether treatment effects on nodulation were a cause of yield differences.

The differences in nodule numbers were largely correlated with treatment effects on root weight, $r = + 0.933$. This mutual dependence results from the symbiotic relationship of ho plant and rhizobia bacteria. Whether phosphate fertilising had direct effect on nodulation was not apparent from the literatur The pelleted plants were noticeably nodulated at four weeks, wh the broadcast and control plants had no nodules, yet the root systems of the three treatments did not greatly differ. Howeve the average number of nodules per mg. of root over the whole experimental period showed that the degree of nodulation was unaffected by the treatments.

Many workers report that phosphate fertiliser increases nodulation (73, 74, 75, 76), but Anderson (72) claims that clove low in phosphate are usually well nodulated. It is difficult to separate the indirect effect of phosphate on plant growth (inclu ing root development and C/N ratio), when trying to evaluate the effect of phosphate fertilisation on nodulation. The calcium contained in the superphosphate may stimulate nodulation, but it questionable whether the small amount contained in the pellets h an appreciable effect.

The degree of nodulation of white clover seedlings in this experiment was unaffected by the treatments, except at the very early stages when the pelleted plants were move nodulated. This early nodulation may be a partial explanation of the early growt increase of the pelleted plants.

Assuming that the reddish coloured nodules are those active fixing nitrogen, it was found that the proportion of active nodu increases with length of time. This is expected, as a larger proportion of the nodules during early seedling growth would be newly formed and immature. Eliminating differences due to root size, the treatments did not affect the proportion of red nodule i.e. the pelleting of superphosphate did not affect the efficien of nitrogen fixation.

The pelleting of 0.02 gm. of superphosphate per clover seed this experiment did not directly effect either the degree of nodulation or the activity of nitrogen fixation. The treatment effects on nodulation appeared correlated with the general growth response of plants. This correlation reflects the symbiotic relationship of host plant and nodule bacteria.

SUMMARY OF RESULTS

SECTION A.

The pellets containing equal quantities of fertiliser and bentonite clay were formed by pressure. Their physical propertie were satisfactory. The sulphate of ammonia in the N pellets was completely soluble. The superphosphate in the P pellets was 72% soluble, and the pellets containing 50% sulphate of ammonia and 5 superphosphate had intermediate solubility.

The stability of the pellets appears related to the proporti of soluble constituents. Ease of disintegration was N > NP > P.

The germination injury caused by the concentration of fertil er round the seed, increased from P, PN to N. As the quantity o fertiliser per seed increased, there was both a lowering and a retarding of germination (P 0.02 gm/seed pelleted = 90% germinat
(PN " " " = 60% "
(N " " " = 30% "

Approximately 10% of the total germinated seedlings had injured radicles.

Saprophytic fungi caused losses when germination was long delayed.

SECTION B.

The analysis of variance of the pot trial as a 3 X 3 X 3 X 5 factorial, only gave significant effects due to varieties. The competition, from the ryegrass plants in the M series caused the M white clover plants to differ significantly from the Ac series.

There was a phosphate response shown in the first Ac cut, P2 > P1 > Po.

Pelleting and half pelleted - half broadcast treatments, were significantly better than broadcasting in the first M cut. The methods had differing effects within the levels of phosphate. At the higher phosphate level, pelleting had a depressing effect and half pelleted - half broadcast treatment was superior.

SECTION C.

The treatment differences showed up within a short time of sowing. The plant response to pelleted superphosphate was during early seedling growth, and became apparent between 2-4 weeks from sowing.

Pelleting gave significantly greater herbage yields and root weights than broadcasting or control. There was little difference between broadcasting and control.

The rates of growth of the three treatments (after an initial period) are approximately constant. The pelleting, and to a lesse extent the half pelleted - half broadcast treatments, have given the seedlings an initial boost to growth. Thereafter differences in growth are largely due to this initial advantage.

There was no treatment effect on root/shoot ratio, i.e. 0.02 gm. superphosphate pelleted per seed did not cause a differential response in either top growth or roots. Root weights and herbage yields were highly correlated, $r = + 0.978$.

Pelleting of superphosphate stimulated root initiation. The localized placement of the small quantity of fertiliser (0.02 gm.) on the soil surface, resulted in a marked increase in lateral root branching. There was no treatment effect on the number of primary lateral roots in the surface layers, but apparently the initiation of the secondary branch roots was stimulated. The increased numbe of absorbing rootlets in the concentrated fertiliser zone is proba one reason for the superiority of the pelleted plants.

Pelleting gave highly significant increases in nodule numbers over broadcasting and control. (Broadcasting and control did not differ greatly)

The effect of pelleted superphosphate on nodule numbers appea correlated with treatment effect on root growth, $r = +0.993$. The degree of nodulation was unaffected by the treatments except a the early stages, when the pelleted plants were more nodulated. This early nodulation may be a partial explanation of the early growth increase of the pelleted plants.

Pelleting does not affect the efficiency of nitrogen fixation as measured by the numbers of red nodules.

CONCLUSIONS

The object of these experiments was to evaluate the pelleting of seed and fertiliser - to determine the advantages and disadvant es likely to accrue, and to study the plant's response.

These objects have been accomplished in part, with the limitation that the results apply to the particular conditions of these trials.

The disadvantages of pelleting are shown clearly by the considerable germination injury, depending on the kind and quantity c fertiliser used. Variable advantages in growth and development of pelleted plants were recorded in two separate sections of the experiment, - (environment factors dominated treatment effects); but normally pelleting of clover seed and phosphatic fertiliser results in increased growth and development of the treated plants.

The superiority of the pelleted plants, compared with broadcast fertiliser or control, no fertiliser, was largely due to increased early seedling growth. The pelleting of 0.02 gm. of fertiliser per seed is equivalent to a broadcast dressing of $2\frac{1}{2}$ cw per acre within the cubic inch round the seed. It is this small quantity of fertiliser, readily available to the seedling, and equ (within a restricted area) to large superphosphate dressing, that gives the initial boost to the pelleted plants.

Earlier nodulation, and an increased number of absorbing, branch rootlets in the zone of concentrated fertiliser may be a partial explanation of the advantages of pelleting.

Pelleting of seed and fertiliser solves ballistic problems in the aerial sowing of differently sized seed, and also ensures that the applied fertiliser benefits the clover seedling and is not dissipated by competition from the established grasses.

If the hazard of germination injury can be overcome, th pelleting of clover seed and phosphatic fertiliser shows promise as an improved and more efficient method of introduc clovers into unploughable hill country.

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APPENDIX I. NODULATION STUDIES

The results from the root studies were examined to see how far they fitted in with Nutman's theory of nodulation.

Put forward in 1948, this hypothesis was based on results obtained from a study of lateral roots and nodules on red clove grown in test-tubes on slopes of an agar medium; the plant mate consisting of the progeny of parent plants selected for high ar low nodule number. The seedlings were inoculated with effectiv and ineffective strains of bacteria.

A brief review of Nutman's papers is presented.

PHYSIOLOGY OF NODULE FORMATION.

Nutman (63, 64) investigated the physiology of nodulation attempting to analyse the factors concerned in determining the number and site of nodules in the clover plant. The limitatio of the number of nodules on the root can be explained in terms of a physiological homology between roots and nodules and it wa also suggested that inhibitory factors may be concerned in the development. Two major aspects of the physiological developme of nodulation are concerned:

1. The morphogenic factors controlling the development of the root system, and
2. The factors contributed by the nodule in such control of root development and the interaction between these two sets of factors.

Evidence has been presented (63) to show that nodules and lateral roots originate from the same meristematic centres, th these are limited in number and predetermined in position. Th sum total of roots and nodules has been shown to be almost constant in plants inoculated with either effective or ineffective strains of bacteria. The rate of formation of nodules and lateral roots was determined by the same hereditary factors in the plant.

Plant roots are exposed to mass infection, nevertheless the number of nodules produced on the plant is very small compared wit the number of points of invasion. A method by means of which such restriction of nodulation might be controlled has been suggested, namely, an inhibiting effect of mature nodules on initiation of subsequent nodule development. This may account for the higher ra of nodule formation in plants inoculated with ineffective bacteria strains as compared with effective strains.

Chen and Thornton (89) have shown that with ineffective strai the development of the nodule is never complete but is arrested by cessation of growth of the apical meristem accompanied by degeneration of the bacterial tissues. This strongly suggests that the inhibiting substance originates either in the apical meristem or in the bacterial tissues.

The varying size of the nodules is a result of this differential behaviour of the meristem. There is an overall inverse correlation between the numbers of nodules produced and their average size. (64)

Preliminary work has supported the hypothesis that inhibitory substances are released during meristematic activity and are translocated round the root. The apex of the root is never occupi by bacteria and it has been shown that the roots of clover seedlin produce a substance that has an inhibitory effect on inoculation (51).

It may be worth while to review and interpret as far as is possible the whole process of infection in terms of root morphogenesis, and in the diagram (Fig.A) an attempt has been made to distinguish and illustrate its various stages.

1. The growth of the bacteria outside the root leads to the production of a secretion, having the properties of indolylacetic acid and which causes root hair deformation.

11. A proportion of the curled root hairs are infected, and the bacteria develop within the root-hair, the infection thread.

III. A few of these root-hair infection threads grow into the cortex and the formation of the nodule follows.

An infected root hair has in all probability only a limited period of life, and it is well known that formation of nodules on the older parts of the root system does not take place. Thus infection is limited to the zones A in the diagram on the younger part of the root and particularly to the points F lying within these zones which can be regarded as indeterminate incipient meristems.

The number of nodules appearing on the root will depend upon the number of the points F arising within Zone A and upon their conversion into nodule rudiments which, with any given set of external factors is determined by -

(i) the genetic constitution of the plant, and

(ii) the production of inhibitory substances in the root and nodule apices (M).

Thus normal nodule formation leads to a lowering of the meristematic potentiality of the root by the metamorphosis of lateral meristems into nodules which are not self-reproducing. The amount of inhibitor produced is related to the activity of the meristem, a root apex producing as much or more of the inhibitory substance than the meristem of an effective nodule, and

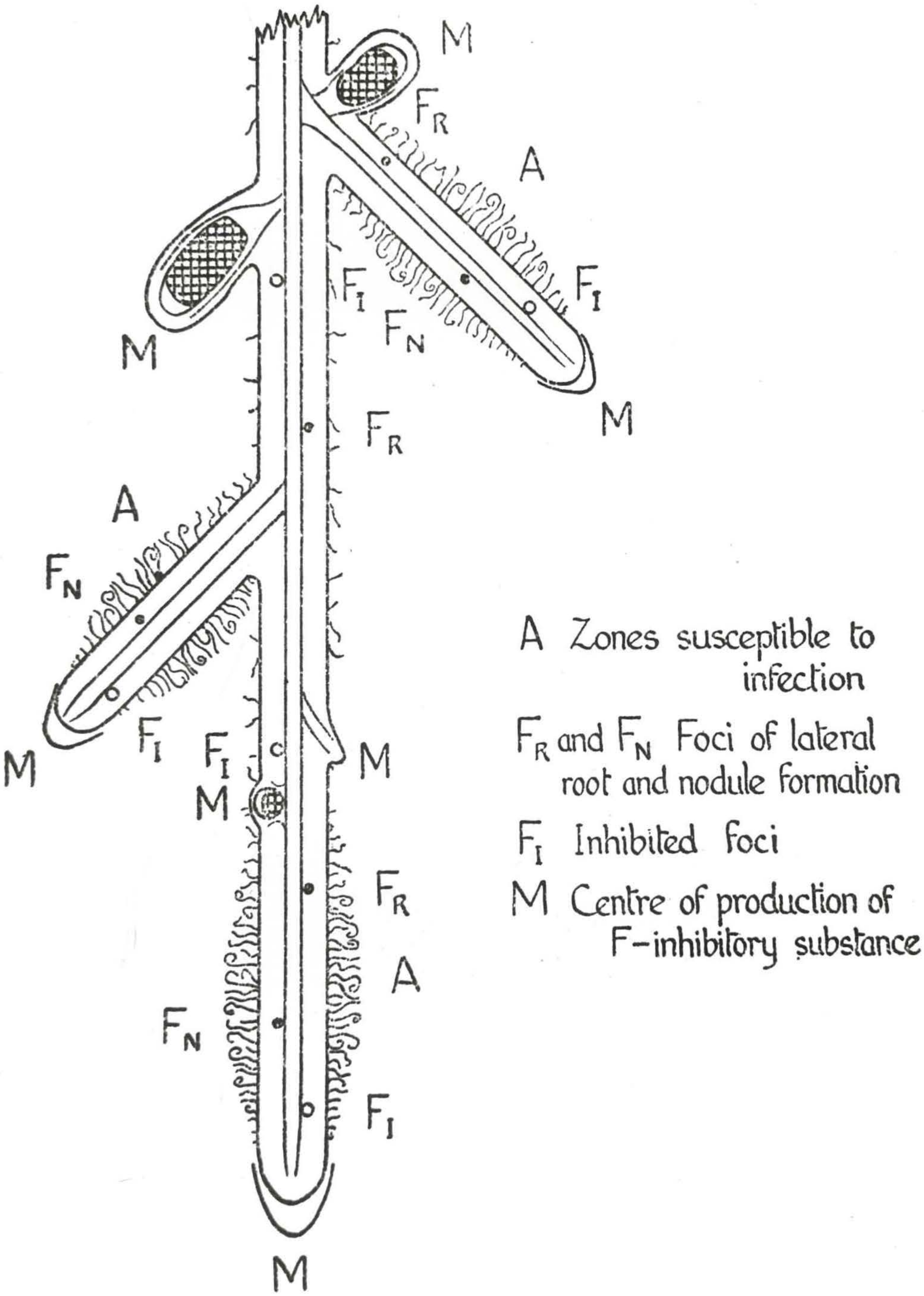


Diagram to illustrate the manner in which sites of nodule and lateral root development may be determined.

FIGURE A. Diagrammatic illustration of physiology of nodule formation; (after Nutman, Ann.Bot.N.S.Vol.12 pp.81-96) an ineffective nodule meristem producing an inappreciable quantity.

The numbers of nodules and lateral roots along the tap root were used to check Nutman's theory of nodulation. To save time, only the nodules and laterals along the tap root were counted.

The data examined on a plot basis at each lifting did not show any inverse correlation between numbers of lateral roots and nodules. The totals of nodules and laterals (along the tap root) were not constant for plants treated alike. The totals of nodules + laterals of paired plants of equal root weights, did not agree. (These findings are at variance with Nutman's results)

The totals of nodules + laterals plotted against the logarithm of time from sowing, did show a linear relationship. (See figures B, C and D) However, both lateral root numbers and nodules plotted separately, showed a linear relationship with the logarithm of time. This could indicate that both nodulation and lateral root formation are dependent on some general factor; which is the co-ordinated development of the plant.

This selected data does not agree with the conclusions drawn from the laboratory studies of Nutman. It has already been shown that total nodule numbers are strongly correlated with root weights. It appears that lateral root numbers and nodulation in the field, merely reflect the growth and development of the plant.

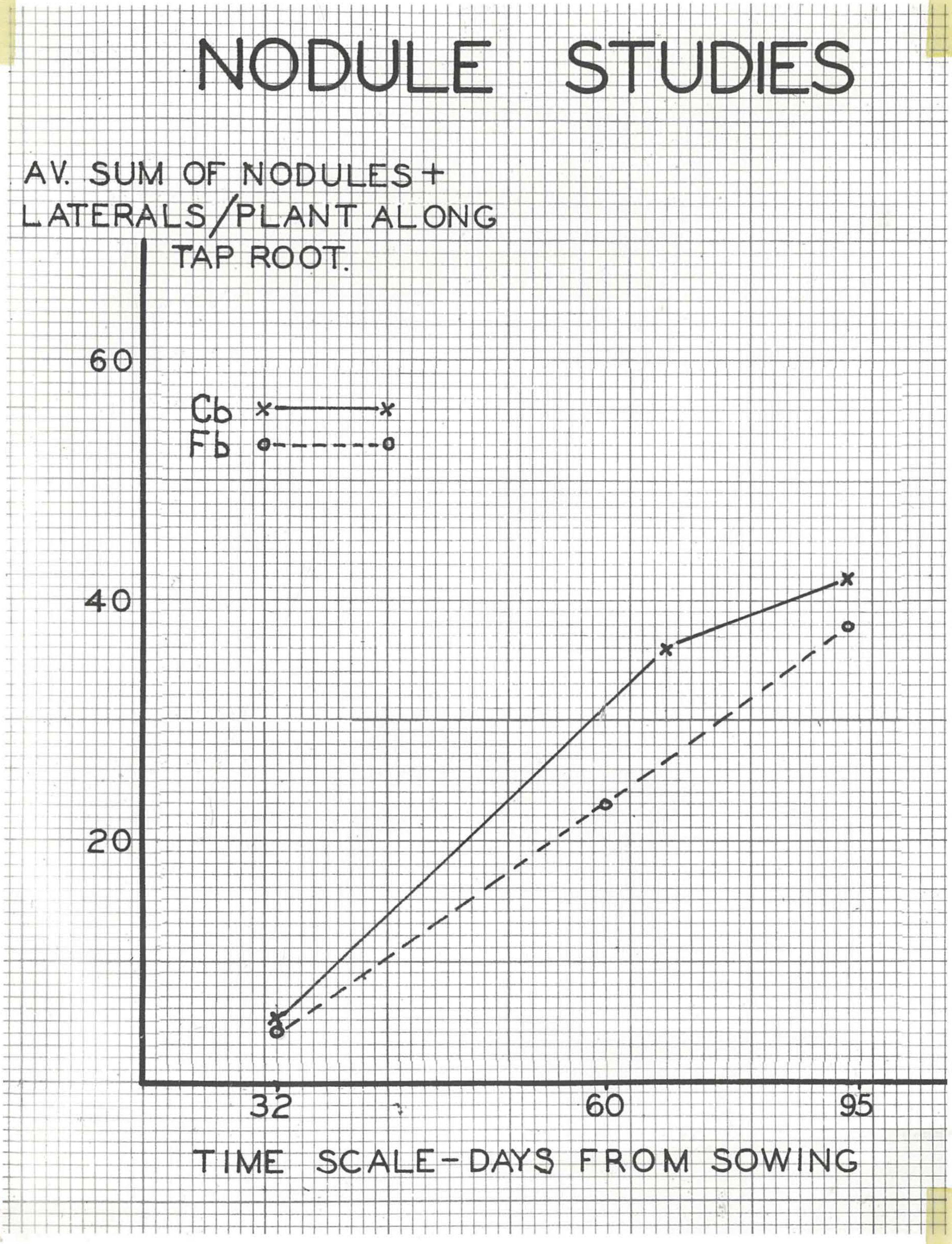


FIGURE B. Graph of nodules + lateral roots of the pelleted plots.

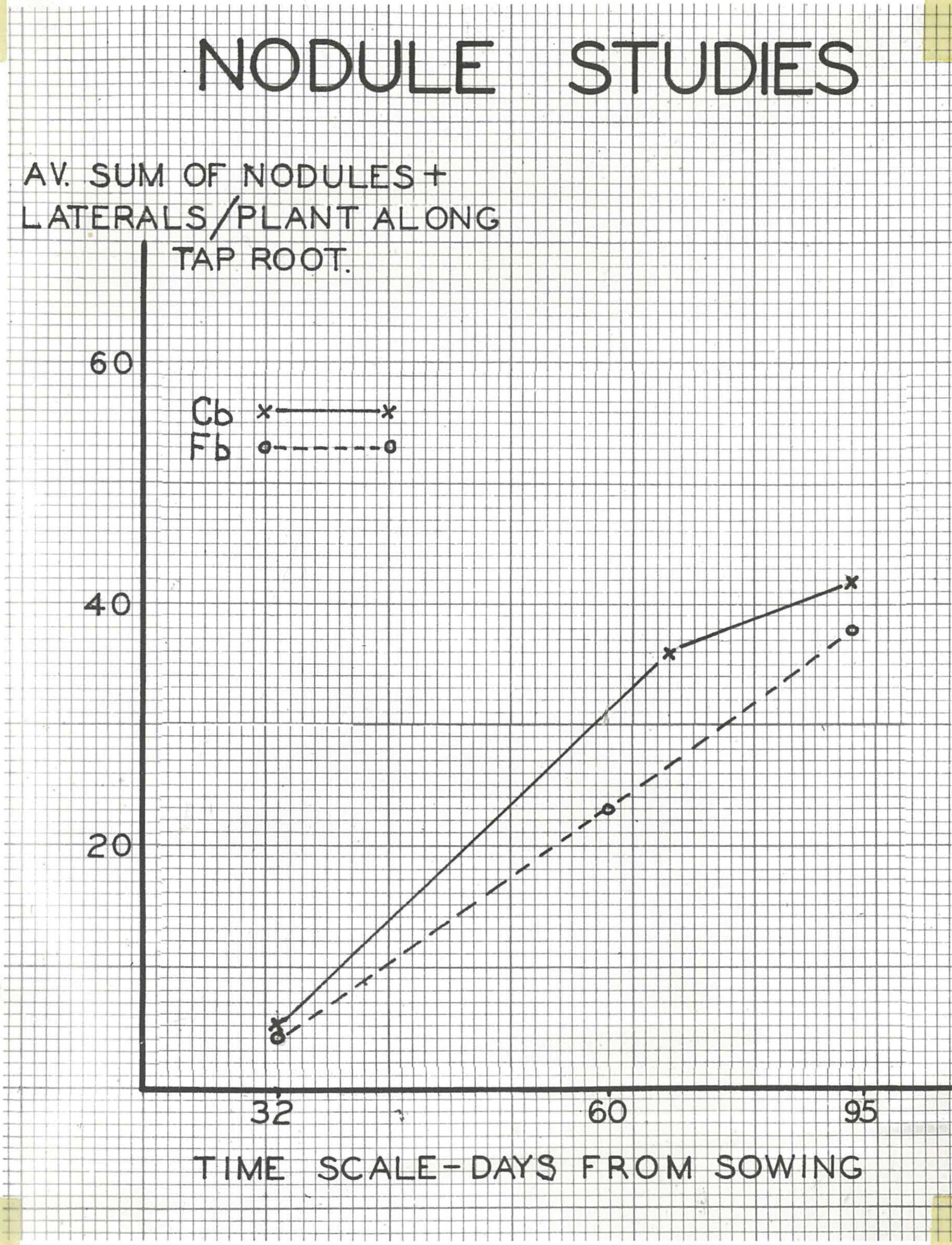


FIGURE C. Graph of nodules + lateral roots of broadcast plots

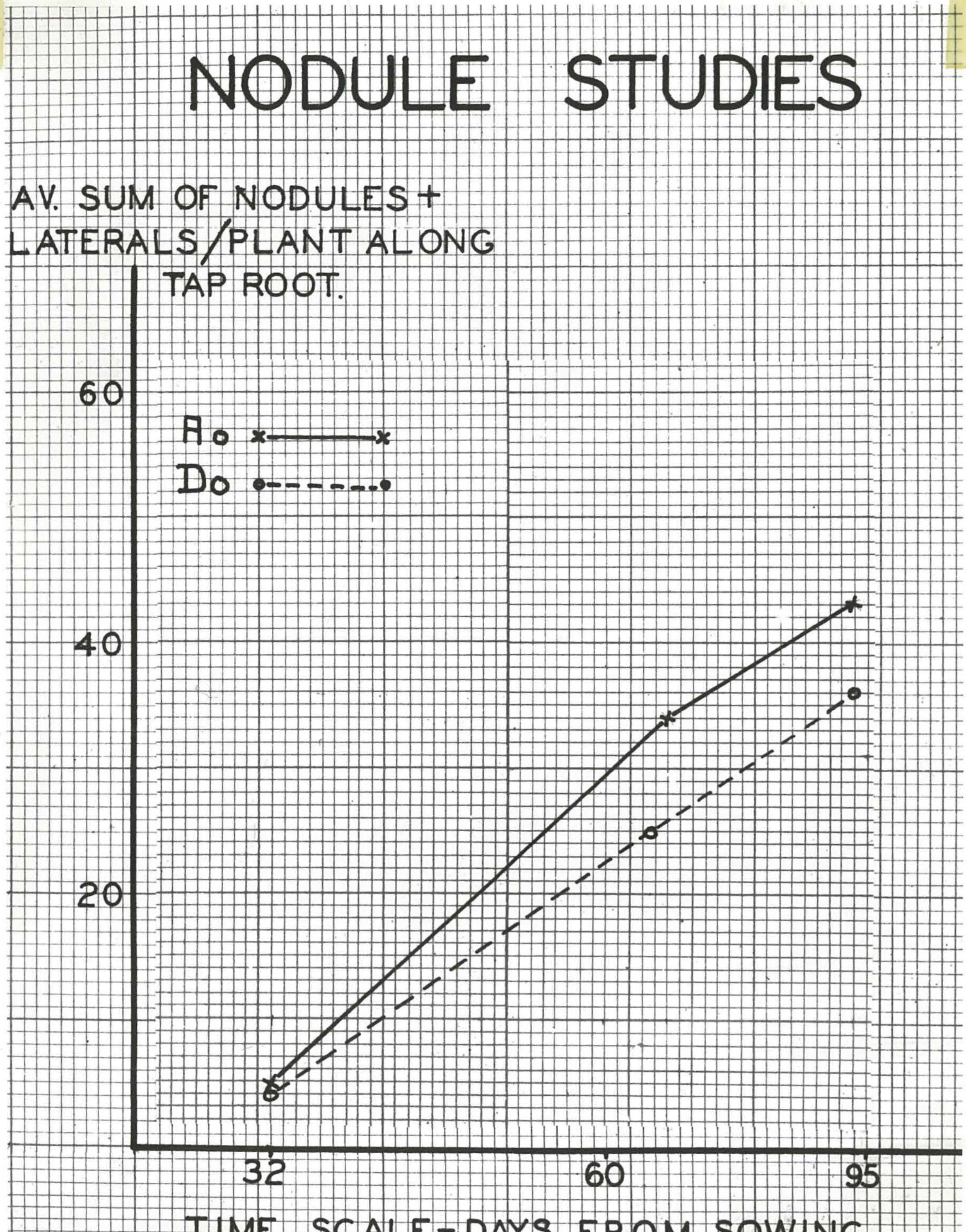


FIGURE D. Graph of nodules + lateral roots of control plots.

APPENDIX II. PLAN OF TRIAL

1 M 0 (p)	2 Ba P2 p
3 Ba N2P1 p	4 Ak N2P2 b
5 Ak N2 b	6 Ba 0 (p)
7 Ak P2 b	8 Ac N1P2 b
9 Ac 0 (p)	10 M N1 b
11 Aa N1P2 p	12 Ak N2 p
13 Ac N1P2 pb	14 Ac P2 p
15 Ak P2 p	16 Aa P1 p
17 Aa P1 pb	18 Ak P2 pb
19 Aa N2P1 pb	20 Ak P1 p
21 Ba N1P2 p	22 Ac N2P2 p
23 Ba N1P2 b	24 M P2 p
25 Ba P1 p	26 Aa P1 b
27 Aa N2 b	28 Ac P2 pb
29 Aa N1P1 pb	30 Aa 0 (p)
31 M N2P1 p	32 Ak N2P1 pb
33 Ba P2 b	34 Ak N2P1 b
35 Ba N2P2 p	36 Ak 0 (pb)
37 Ac N2P2 p	38 M N2 pb
39 Ac N1P1 pb	40 Ac P1 pb
41 M N2P1 b	42 Ba N2 pb
43 Ac P1 b	44 Ac 0 (b)
45 Aa N1P1 b	46 Aa N1P2 b
47 Aa N1 pb	48 Ak 0 (p)
49 Ba NaP1 pb	50 Ak N1 p
51 Ak 0 (b)	52 Aa P2 pb
53 M P1 p	54 Ba P2 pb
55 Ba N2P1 b	56 Ac P2 b

57 Ac P1 p	58 Aa N1P1 pb
59 Ac N1 p	60 Aa N2P1 p
61 M N1P2 b	62 Ak N1P2 b
63 Ak N2P1 p	64 Ac N1P1 p
65 M 0 (pb)	66 Ak N1P2 pb
67 Aa N2P1 b	68 Aa 0 (pb)
69 Ak N2P2 pb	70 Ac N2P1 b
71 Aa P2 b	72 M N1P1 pb
73 Aa N2P2 p	74 Aa N1P1 p
75 M N1P2 b	76 Aa N2P2 b
77 Ba P1 pb	78 Ba N1 p
79 Ac N2P1 pb	80 Ac N1 b
81 Ba P1 b	82 Ac N1P2 p
83 Ba N1 b	84 Ba N2 b
85 Ak N1 pb	86 Ac N2 pb
87 Ak N1P1 b	88 Ba N2P2 pb
89 M 0 (b)	90 Ak N1 b
91 Ak N2P2 p	92 Ac N2 p
93 M P1 pb	94 M N2P2 b
95 M N2P2 pb	96 Aa N1 p
97 Aa N1 b	98 Ba N2 p
99 Ba N1 pb	100 Ak N1P2 p
101 Ba N1P2 pb	102 M N1P1 p
103 Ba 0 (pb)	104 Ba N1P1 pb
105 Ak N2 pb	106 M N2P1 pb
107 Ak N1P1 p	108 Ak P1 b
109 M P2 b	110 Ac N2 b
111 Aa N2P2 pb	112 M N2 p

113 Ba N2 b	114 Ac 0 (pb)
115 Aa 0 (b)	116 Ac N1 pb
117 M N1P2 pb	118 Ac N2P2 pb
119 M N2P2 b	120 M N1 p
121 M N2 b	122 Aa N2 p
123 Ba N1P1 p	124 Ba N1P1 b
125 M N1P2 p	126 M P1 b
127 Aa N2 pb	128 Aa P2 p
129 Ac N1P1 b	130 Ak P1 pb
131 Ak N1P1 pb	132 M N1 pb
133 Ba 0 (b)	134 Ac N2P1 p
135 M P2 pb	

APPENDIX II. PLAN OF TRIAL

Plot	Treatment	Plot	Treatment
1	M O (p)	2	Ba P2 p
3	Ba N2P1 p	4	Ak N2P2 b
5	Ak N2 b	6	Ba O (p)
7	Ak P2 b	8	Ac N1P2 b
9	Ac O (p)	10	M N1 b
11	Aa N1P2 p	12	Ak N2 p
13	Ac N1P2 pb	14	Ac P2 p
15	Ak P2 p	16	Aa P1 p
17	Aa P1 pb	18	Ak P2 pb
19	Aa N2P1 pb	20	Ak P1 p
21	Ba N1P2 p	22	Ac N2P2 p
23	Ba N1P2 b	24	M P2 p
25	Ba P1 p	26	Aa P1 b
27	Aa N2 b	28	Ac P2 pb
29	Aa N1P1 pb	30	Aa O (p)
31	M N2P1 p	32	Ak N2P1 pb
33	Ba P2 b	34	Ak N2P1 b
35	Ba N2P2 p	36	Ak O (pb)
37	Ac N2P2 p	38	M N2 pb
39	Ac N1P1 pb	40	Ac P1 pb
41	M N2P1 b	42	Ba N2 pb
43	Ac P1 b	44	Ac O (b)
45	Aa N1P1 b	46	Aa N1P2 b
47	Aa N1 pb	48	Ak O (p)
49	Ba NaP1 pb	50	Ak N1 p
51	Ak O (b)	52	Aa P2 pb
53	M P1 p	54	Ba P2 pb
55	Ba N2P1 b	56	Ac P2 b

Plot	Treatment	Plot	Treatment
57	Ac P1 p	58	Aa N1P1 pb
59	Ac N1 p	60	Aa N2P1 p
61	M N1P2 b	62	Ak N1P2 b
63	Ak N2P1 p	64	Ac N1P1 p
65	M O (pb)	66	Ak N1P2 pb
67	Aa N2P1 b	68	Aa O (pb)
69	Ak N2P2 pb	70	Ac N2P1 b
71	Aa P2 b	72	M N1P1 pb
73	Aa N2P2 p	74	Aa N1P1 p
75	M N1P2 b	76	Aa N2P2 b
77	Ba P1 pb	78	Ba N1 p
79	Ac N2P1 pb	80	Ac N1 b
81	Ba P1 b	82	Ac N1P2 p
83	Ba N1 b	84	Ba N2 b
85	Ak N1 pb	86	Ac N2 pb
87	Ak N1P1 b	88	Ba N2P2 pb
89	M O (b)	90	Ak N1 b
91	Ak N2P2 p	92	Ac N2 p
93	M P1 pb	94	M N2P2 b
95	M N2P2 pb	96	Aa N1 p
97	Aa N1 b	98	Ba N2 p
99	Ba N1 pb	100	Ak N1P2 p
101	Ba N1P2 pb	102	M N1P1 p
103	Ba O (pb)	104	Ba N1P1 pb
105	Ak N2 pb	106	M N2P1 pb
107	Ak N1P1 p	108	Ak P1 b
109	M P2 b	110	Ac N2 b
111	Aa N2P2 pb	112	M N2 p

Plot	Treatment	Plot	Treatment
113	Ba N2 b	114	Ac O (pb)
115	Aa O (b)	116	Ac N1 pb
117	M N1P2 pb	118	Ac N2P2 pb
119	M N2P2 b	120	M N1 p
121	M N2 b	122	Aa N2 p
123	Ba N1P1 p	124	Ba N1P1 b
125	M N1P2 p	126	M P1 b
127	Aa N2 pb	128	Aa P2 p
129	Ac N1P1 b	130	Ak P1 pb
131	Ak N1P1 pb	132	M N1 pb
133	Ba O (b)	134	Ac N2P1 p
135	M P2 pb		

APPENDIX III.

SOIL ANALYSIS

THE PREPARATION OF SOIL SAMPLES

On arrival at the laboratory the soil is spread out to dry room temperature and is then passed through a 2 m.m. round-hole sieve, any lumps being previously gently disintegrated in a mort with a wooden pestle. The residue on the sieve, consisting of stones and fine gravel, before being discarded is weighed and expressed as a percentage of the total weight of the air-dry soi The material passing the sieve (the "fine earth") is the bulk sample from which separate sub-samples are uniformly taken. For mechanical analyses and physical determinations, it is advisable to pour the whole prepared soil sample from the sample bottle on to a sheet of paper and take with a spatula at least 10 small portions from different parts of the heap to make up the require weight.

MECHANICAL ANALYSIS.

Moisture. Place exactly 10 g. of air-dry soil in a weighed flat bottomed porcelain dish (7 cm.). Place the dish (uncovered) in oven at 105°C. and dry for 12-16 hours. Place the dish in a des ccator (cover with a watch-glass), cool and weigh. Multiply the loss in weight by 10 to obtain the percentage moisture in the ai dry soil.

Loss on Ignition. After determining the moisture, transfer the dish and soil to a cool muffle furnace. Raise the temperature a ignite for 30 minutes at a bright red heat. When excess heat ha passed off, cover with a watch-glass in a desiccator, cool and weigh. The further loss in weight corresponds to the loss on ignition. Multiply by 10 to obtain the loss on ignition expresse as a percentage of the air-dry soil.

Grading into Fractions: Place 10 gm. of air-dry soil in a dry ta 600 ml. beaker, add 50 ml. of 6% hydrogen peroxide and heat the contents on the water-bath taking care to avoid frothing over. Agitate frequently either by stirring or by shaking the beaker wi a rotatory motion. When vigorous frothing has subsided boil unti the volume is about 30 ml. If much organic matter is present, a further addition of peroxide may be necessary.

To the contents of the beaker, add 20 ml. of N.hydrochloric acid and make up the volume to about 100 ml. If much calcium carbonate is present, more acid should be added (2 ml. of N. hydrochloric acid for each 1% of calcium carbonate). Allow to stand for one hour with frequent stirring and then filter through an 11 c.m. Whatman No. 1 paper. When the filtrate has passed through wash the soil on the filter paper three times with 50 ml. of water. The acid filtrate is transferred to a beaker and boile with 5 ml. nitric acid to oxidise ferrous compounds; concentrate about 100 ml. and add concentrated ammonia in excess. Filter off the precipitate, ignite and weigh. This gives loss by solution.

Transfer the residue on the filter paper by means of a blunt spatula and a jet of hot water to a 500 ml. beaker. Add 4 ml. of normal sodium hydroxide solution and beat in the milk-shake machi for 10 minutes. Note the volume should be about 400 ml. Then transfer the contents to a 500 ml. cylinder without lip, make up to the mark, shake for one minute with repeated inversion and allow to stand away from sources of heat and out of direct sunlig to avoid convection currents in the suspension.

After standing for four minutes forty-eight seconds at 20°C. or the equivalent time at any other temperature, 20 ml. are withdrawn from a depth of 10 cm. by means of the special pipette whic is mounted on a stand provided with levelling screws and includes wheel and ratchet device by which the pipette can be raised or lowered to any desired depth. The pipette is lowered with the ta closed and the lowering is commenced about 20 seconds before the time is up. Care must be taken to avoid too rapid ingress of the liquid with consequent eddying in the suspension.

The contents of the pipette (20 ml.) are delivered into a weighed flat-bottomed porcelain dish (7 cm. in diameter) and evaporated to dryness on the water bath and afterwards dried over night in the electric oven at 105°C. cooled in a desiccator and weighed. The weight of the dried residue in mgm. divided by 4 is equal to the percentage of silt + clay.

The suspension is thoroughly shaken and again allowed to settle for the clay sampling which is carried out at 10 cm. below the surface after settling 8 hours at 20° or the corresponding ti at any other temperature. The contents of pipette are delivered into a weighed flat-bottomed dish, dried at 105°C, cooled in a desiccator and weighed. Divide this weight expressed in mg. by 4 and deduct the clay percentage thus obtained from the percentag of silt and clay to obtain the percentage of silt. Correct the clay percentage for the amount of sodium hydroxide used for dispersion by deducting 1.6 from the percentage of clay found above.

The greater part of the suspension in the measuring cylinde is then poured away and the sediment is washed into a 400 ml. beaker through a 70 I.M.M. sieve resting on the 400 ml. beaker a wash with a jet of hot water until clean sand remains; rubbing is generally unnecessary. Dry the coarse sand on the sieve at 105°C, transfer to a weighing bottle and weigh.

The filtrate is well stirred and made up to 10 cm. allowing for the thickness of sediment on the bottom. After standing 4 m 48 sec. at 20°C. or the corresponding time at any other temperature the turbid suspension is poured away. The beaker is again filled to the mark with water and the process is repeated until the super-natant liquid is quite clear after standing for the requisite period. The residue is fine sand which is collected, dried at 105°C. and weighed. On account of their small loss in weight on ignition, neither sand fraction need be ignited.

APPENDIX IV. POT TRIALS

HERBAGE WEIGHTS IN MILLIGRAMS

SPECIES		Red Clover (Aa)		White Clover (Ac)		Sub.Clover (Ak)		Per. ryegrass (Ba)			Per. rye + White Clover (M)				
											Per. rye.			White Clover	
Date of Cutting		29.10.50	4.12.50	24.6.50	5.12.50	20.6.50	13.9.5	20.4.50	20.6.50	14.10.50	20.6.50	1.10.50	21.10.50	21.10.50	15.12.50
N_1	p	747	901	25	6320	309	--	7	176	3887	936	4710	5266	900	9053
	b	2085	4219	0	2980	--	114 "	9	385	4958	621	3107	6523	260	2094
	pb	1251	1604	22	15005	416	7000	3	140	876	513	796	3318	697	4670
N_2	p	2000	8311	0	8100	107	1053	13	631	876	980	2362	3465	698	3786
	b	137	2472	15	17580	228	2501	9	203	754	617	2577	4578	1482	12270
	pb	1047	10696	-	-	-	152"	12	314	2386	686	1897	4111	1765	11424
P_1	p	-	-	15	15580	258	2696	8	189	1649	809	2670	4661	1463	9680
	b	2457	8063	63	15460	305	2714	6	607	1968	1841	2253	3536	273	1149
	pb	220	1322	90	16320	392	2579	-	-	333"	863	1917	3737	806	5237
P_2	p	3152	8098	144	10560	266	1541	15	282	560	701	2572	4525	1113	7752
	b	2120	9998	41	7000	258	1530	9	340	2071	832	520	869	1281	12844
	pb	2258	6943	52	10520	155	140"	15	223	679	941	4426	6298	2059	13952
N_1P_1	p	1345	2310	105	11870	370	2510	34	435	5200	545	3843	7181	1661	10362
	b	2307	7939	0	2550	171	2170	18	315	3200	765	1410	1270	183	2851
	pb	677	2886	8"	4650	777	3363	29	138	5355	890	4530	7252	943	5634
N_2P_2	p	1653	6109	28	14470	470	5241	10	285	3530	877	4021	6946	785	10416
	b	--	--	15"	6390	273	1593	0	473	3496	473	1744	2200	532	4165
	pb	1325	3435	53	5080	578	3818	0	188	893	700	3273	5810	1788	13650
N_2P_1	p	1501	1341	10	8630	120	5915	1	213	593	682	1442	2632	1498	9576
	b	621	1404	13	11470	475	2727	1	248	870	1296	4962	5354	498	2658
	pb	2448	8120	30	24890	283	2096	1	243	1560	559	2631	5008	0	39"
N_1P_2	p	1650	4542	230	13030	424	2745	18	335	487	768	7320	9523	1195	13388
	b	2065	8960	94	18300	539	760	0	297	2612	910	3350	7205	626	4814
	pb	1646	3292	85	14870	135	1822	12	591	4750	610	5048	10589	1183	15543
0		1372	5299	5	12240	228	1221	15	286	468	536		1091	697	6155
0		625	881	19	3410	384	1595	10	296	940	616	760	2517	1680	11974
0		1422	2312	0	5510	341	2018	10	231	637	908	2232	3737	253	4014

" Plants appearing small and stunted or otherwise abnormal on observation.

Key to Symbol

p - pellete

b - broadca

pl - ½ ferti pellete broadca

N_1 = total of .01 gm. of N fertiliser

N_2 = " " .02 gm. " " "

P_1 = " " .01 gm. of superphosphate

P_2 = " " .02 gm. " "

APPENDIX V. POT TRIALS - TOTALLED TREATMENT FIGURES

Fert.	Treatment	Aa	Ac	Ak	Ba	M (Ac)
N_1	p	1648	6345	(2233)	4070	9953
	b	6304	2980	(1961)	5352	2354
	pb	2855	15027	7416	1019	5367
N_2	p	10311	8100	1160	1520	4484
	b	2609	17595	2729	966	13752
	pb	11743	(15684)	(8863)	2712	13189
P_1	p	(8308)*	15595	2954	1846	11143
	b	10520	15523	3019	2581	1422
	pb	1542	16410	2971	(700)	6043
P_2	p	11250	16704	1807	857	8865
	b	12118	7041	1788	2420	14125
	pb	9201	10572	(3324)	917	16011
N_1P_1	p	3655	11975	2880	5669	12023
	b	10246	2550	2341	3533	3034
	pb	3563	4658	4140	5522	6577
N_2P_2	p	7762	14498	5711	3825	11201
	b	(6337)	6405	1866	3969	4697
	pb	4760	5133	4396	1081	15438
N_2P_1	p	2842	8640	6035	807	11074
	b	2025	11483	3202	1119	3156
	pb	10568	24920	2379	1804	(13508)
0	p	6671	12245	1449	769	6852
	b	1506	3429	1979	1246	13654
	pb	3734	3510	2359	878	4267

* Figures in brackets are calculated values.

APPENDIX VI. ROOT STUDIES

Date	Plot No.	Plant No.	Nodules Total	Red	No. on tap root	No. of trifoliate leaves	Wt. tops (mgs.)	Wt. r[oots] (mg.s)
30.5.50	Fb	A	0	0	0	3	6.9	4.5
		B	0	0	0	3	4.7	2.5
		J	7	-	7	3	5.1	3.1
		K	5	-	5	3	5.8	3.3
		L	4	-	4	2	2.3	2.0
		M	7	-	7	2	2.0	2.8
		N	4	-	4	2	3.5	2.2
		O	6	-	6	3	7.5	5.1
	Ep	C	12	-	12	5	29.8	14.7
		D	13	-	13	4	25.5	14.4
		E	20	-	20	4	24.4	13.5
	E	F	6	-	6	3	4.1	3.8
		G	3	-	3	2	2.6	1.7
		H	8	-	8	2	5.8	2.8
		I	7	-	7	3	4.1	2.3
7.6.50	Gb	P	26	9	18	5	13.0	11.0
		Q	7	3	7	3	5.2	7.3
		R	7	3	7	4	6.3	4.0
		S	13	6	13	4	8.4	8.7
		T	15	3	15	4	8.8	6.4
		U	11	4	8	4	6.4	4.9
		V	12	5	8	4	6.2	5.6
		W	13	6	9	5	9.3	4.9
		X	6	0	5	3	4.5	4.6

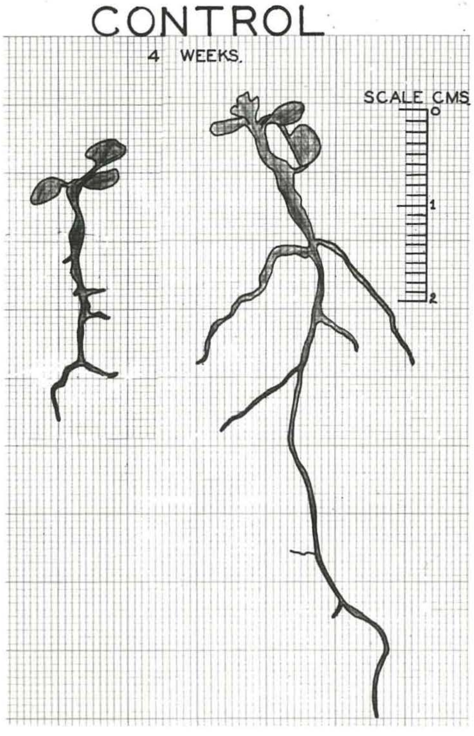
Date	Plot No.	Plant No.	Nodules Total	Red	No. on tap root	No. of trifoliate leaves	Wt. Tops (mg.s)	Wt. root (mg.
7.6.50	Do	1	7	-	7	3	5.8	5.0
		2	8	3	8	3	10.9	3.0
		3	6	2	5	3	5.1	4.4
		4	7	1	7	3	4.8	4.3
		5	7	-	3	3	3.1	3.7
		6	9	7	8	3	4.2	3.1
		7	5	3	5	3	5.3	3.7
		8	7	4	4	3	5.1	3.6
11.6.50	B	9	12	8	10	4	9.4	6.6
		10	9	4	7	3	5.4	5.6
	Bp	11	35	9	15	6	18.5	19.5
		12	25	5	16	5	16.2	12.1
		13	23	6	16	6	14.7	10.8
	Ao	14	11	2	11	3	4.6	3.9
		15	11	7	10	5	9.7	8.8
		16	7	4	5	4	8.1	9.5
		17	9	2	8	4	9.4	7.7
		18	9	1	9	3	5.0	3.3
		19	9	-	8	3	6.3	6.2
		20	11	-	7	4	8.1	8.6
4.7.50	B	21	15	7	12	5	12.7	7.7
		22	18	11	12	6	13.5	7.0
	Bp	23	32	23	13	9	31.8	29.3
		24	19	15	12	7	19.9	10.7
		25	20	13	14	4	20.0	13.5

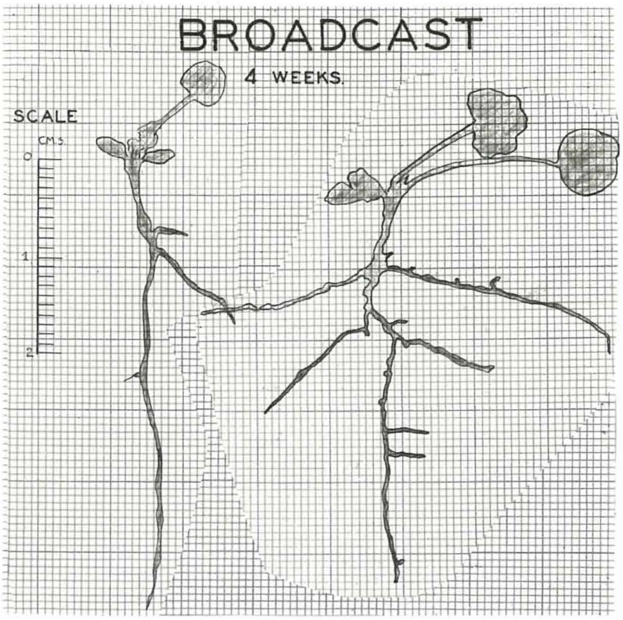
Date	Plot No.	Plant No.	Nodules Total	Red	No. on tap root	No. of trifoliate leaves	Wt. tops (mg.s)	Wt. ro (mg.s
4.7.50	Ao	26	10	6	7	3	6.7	4.7
		27	11	10	11	4	10.6	8.7
		28	14	9	11	3	10.0	6.0
		29	17	9	12	6	15.0	7.7
		30	14	11	11	5	10.9	6.5
		31	15	9	10	6	12.0	10.5
		32	12	7	10	5	15.3	11.7
	Cb	33	15	5	14	5	10.6	7.6
		34	27	13	20	8	37.3	23.5
		35	11	4	9	3	6.2	4.8
		36	16	6	14	4	12.4	7.6
		37	6	1	6	3	4.4	4.2
		38	24	6	16	7	15.1	16.3
	Do	39	16	8	11	5	13.6	8.9
		40	18	4	11	5	8.4	6.0
		41	9	4	4	4	3.7	4.2
		42	16	4	11	6	12.1	9.4
		43	13	8	13	6	15.0	12.6
		44	11	8	8	6	10.5	8.5
		45	20	12	12	6	14.8	9.8
		46	18	11	9	8	14.9	11.1
	Ep	47	25	14	17	12	31.5	22.6
		48	38	19	17	19	100.3	65.1
		49	26	14	17	10	33.7	25.1

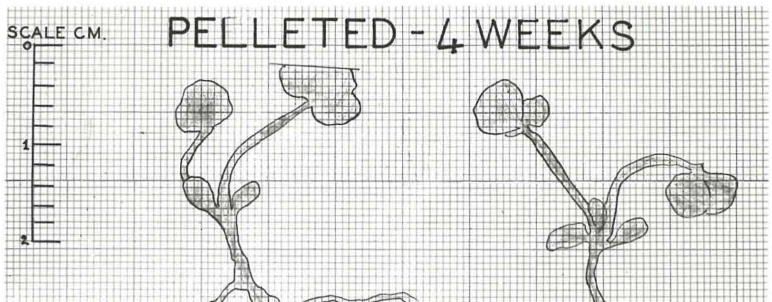
Date	Plot No.	Plant No.	Nodules Total	Red	No. on tap root	No. of trifoliate leaves	Wt. tops (mg.s)	Wt. roots (mg.s)
4.7.50	Fb	50	13	4	11	4	13.3	5.6
		51	11	5	7	4	5.5	6.5
		52	15	7	8	4	10.5	12.3
		53	22	6	13	6	16.5	15.6
		54	16	7	10	6	23.1	16.8
		55	21	12	11	8	21.0	12.6
		56	6	4	4	4	7.3	5.9
		57	24	9	13	8	14.9	15.0
		58 b	29	14	15	8	22.0	19.0
27.8.50	Bp	58 p	105	88	18	28	238	102
		59	96	73	25	18	115	60
		60	80	76	23	24	224	75
1.9.50	Ep	66	217	202	41	30	489	211
		67	202	155	29	26	321	87
		68	193	115	28	31	393	169
	Gb	70	89	50	20	20	145	62
		71	45	26	14	11	96	42
		72	72	64	27	17	102	46
	Fb	73	50	38	16	19	106	51
		74	67	35	20	13	95	38
		75	89	66	28	23	112	47
27.8.50	Ao	61	38	26	16	14	50	30
		62	42	26	11	14	85	46
		63	64	36	25	12	61	31
		64	32	17	15	7	41	29
		65	38	23	18	15	73	42
	Do	69	39	28	19	13	81	40

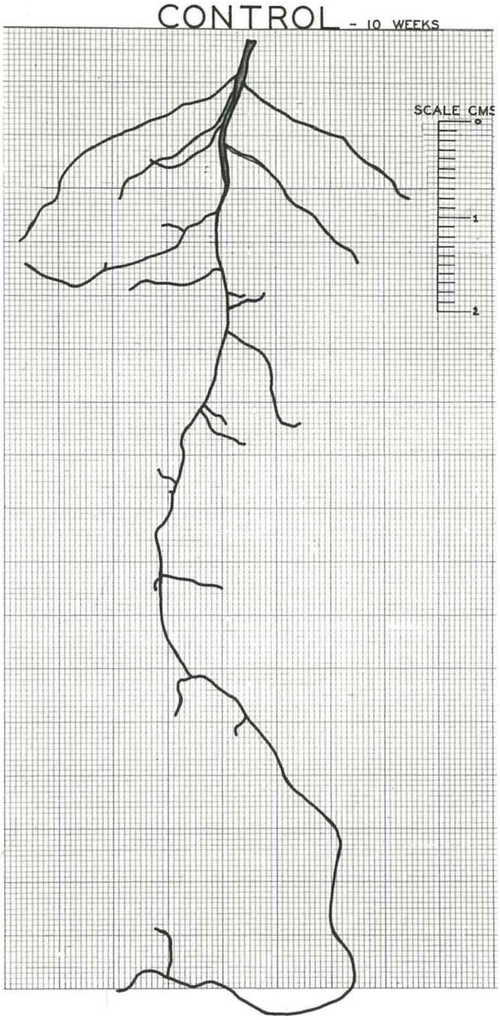
Date	Plot No.	Plant No.	Nodules Total	Red	No. on tap root	No. of trifoliate leaves	Wt. tops (mg.s)	Wt. roo (mg.s
		70o	104	72	20	18	143	54
		71o	62	46	19	18	88	35
		72o	38	31	17	11	63	26

APPENDIX VII.   Scale drawings of root systems of typical plants per treatment at each lifting date.




BROADCAST
4 WEEKS.
SCALE
CMS.
0
1
2


SCALE CM.
PELLETED - 4 WEEKS
0
1
2


CONTROL - 10 WEEKS
SCALE CMS
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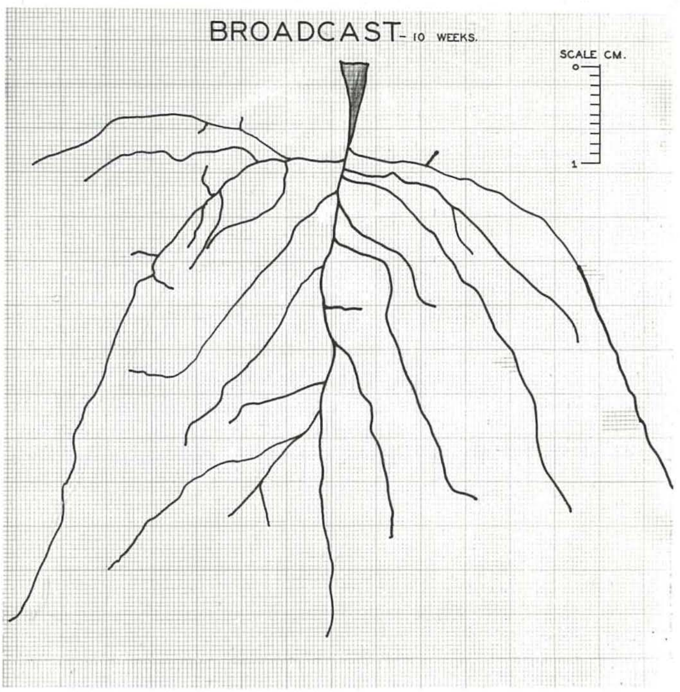
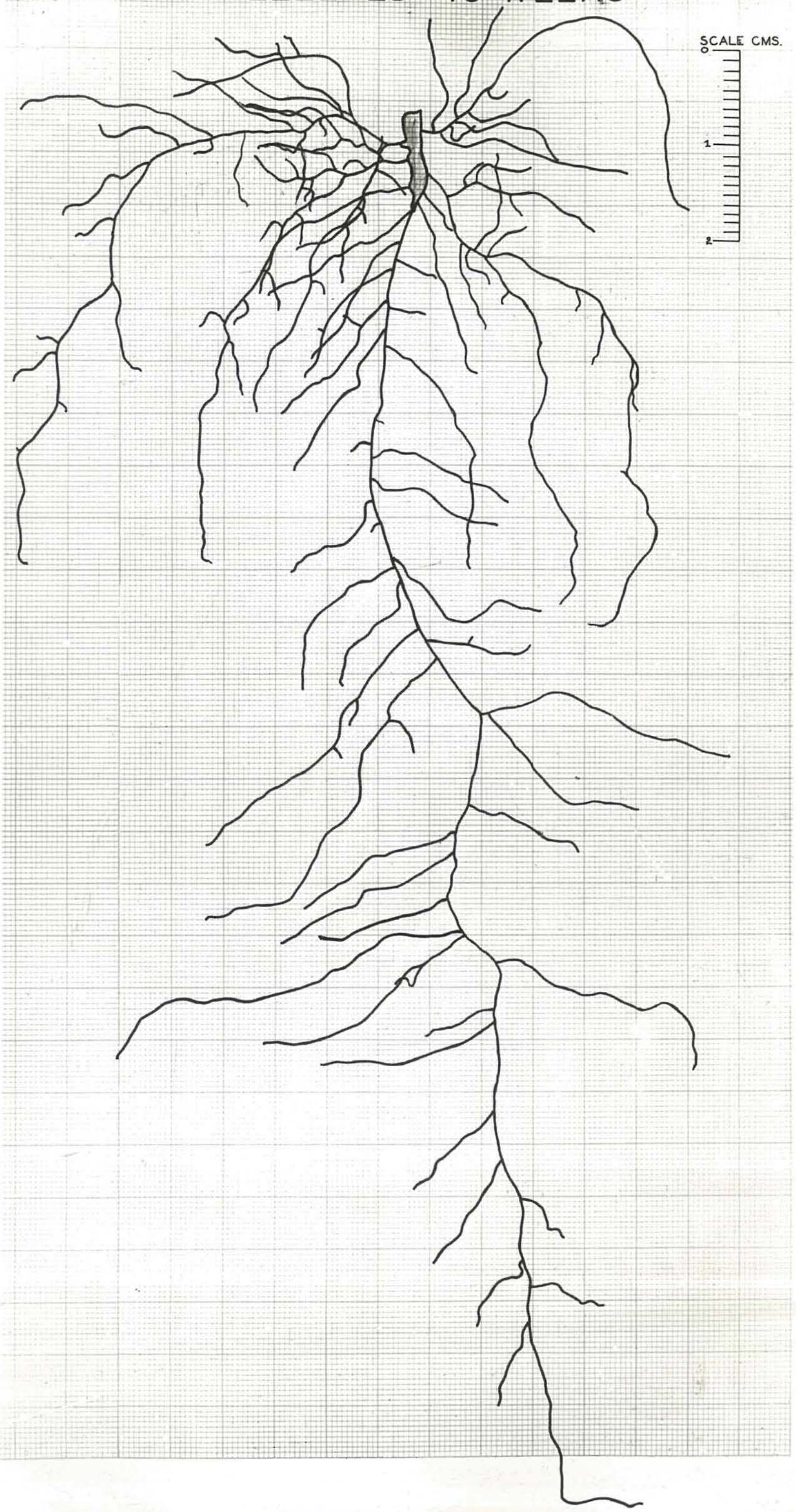
1
2

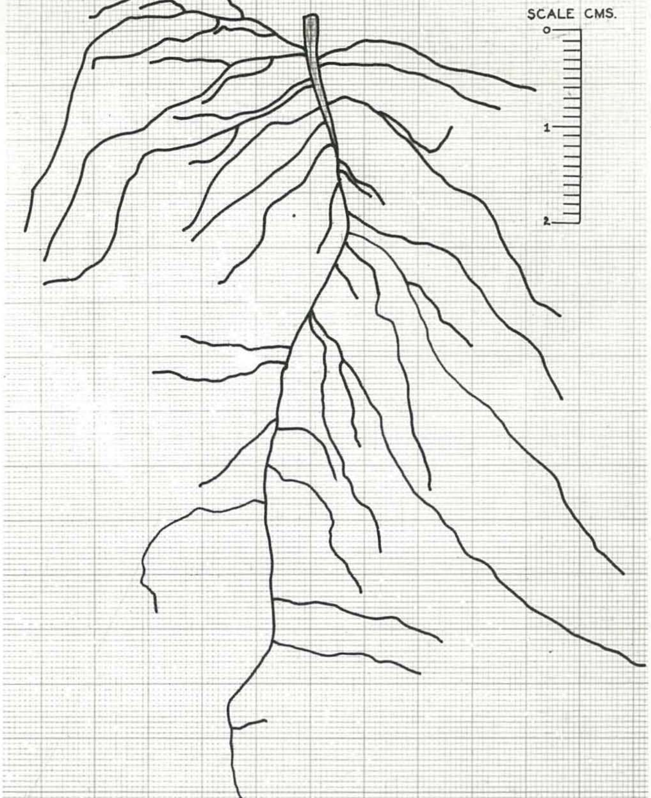


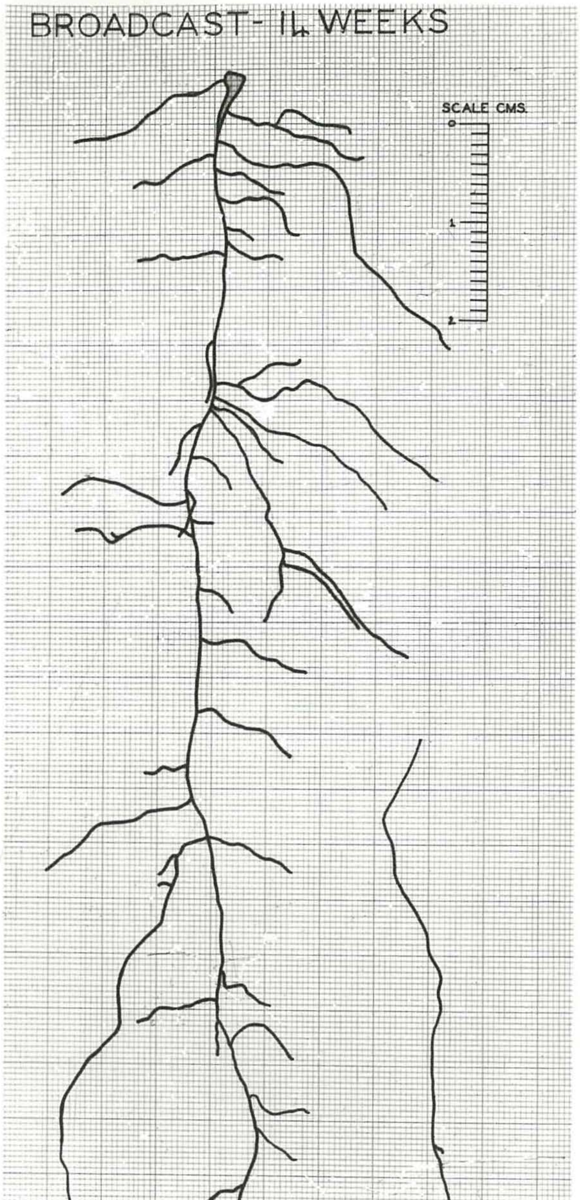
BROADCAST- 10 WEEKS.
SCALE CM.
0
1

PELLETED - 10 WEEKS



CONTROL 14 WEEKS.




BROADCAST - 14 WEEKS
SCALE CMS.
0
1
2

PELLETED 14 WEEKS.