PRE-EMERGENCE ROTTING OF PEAS IN SOUTH AUSTRALIA

I. FACTORS ASSOCIATED WITH THE SEED

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Summary

Laboratory tests have shown that individual wrinkle-seeded pea seeds vary in their ability to continue vigorous growth after germination. After the radicle has broken through the testa, some produce normal seedlings, some fail to develop further, while others are intermediate between these extremes and produce stunted seedlings. Thus some seeds capable of germination are incapable of emerging when planted in the field. Poor growth after germination is accompanied by prolific growth of moulds and bacteria on the seeds.

Smooth-seeded peas do not vary markedly in vigour of growth after germination and are generally free of moulds and bacteria.

I. INTRODUCTION

In South Australia wrinkle-seeded peas, such as William Massey and Greenfeast, are grown for green peas, canning, or seed. An important problem in growing satisfactory crops is the low percentage of emergence which often occurs in the field and results in thin stands and low yields. On the other hand this trouble rarely occurs with smooth-seeded peas such as White Brunswick, where good stands are regularly obtained.

This problem, investigated at the Waite Institute, Adelaide, between 1944 and 1953, has attracted considerable attention both in Australia and other countries. While several workers have shown that soil-borne fungi may cause seed rotting, failures in field emergence of wrinkle-seeded peas have occurred in South Australia under conditions where it was improbable that soil-borne fungi were responsible and the possibility of seed-borne factors was investigated.

Factors associated with the seed have been investigated less commonly than soil-borne factors. McNew (1943) showed that, in steamed soil, there were differences in emergence between different seed samples. Hull (1937), Padwick (1938), and other workers discussed "vigour" of seed samples, and Hynes and Wilson (1939) attributed much of the poor emergence of peas in New South Wales to low vigour. Crosier and Patrick (1939) and Crosier (1946) suggested that low vigour of seed was largely due to seed-borne moulds and bacteria and attempted to eliminate them in laboratory tests by using fungicidal dusts. Jones (1927) and Hickman (1941) showed that Aschochyta infection of seed influenced emergence. Hulbert and Whitney (1934) suggested that physical injury to the seed was an important cause of rotting. Wellington (1962) has emphasized that quality of pea seed has received little attention.

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II. EXPERIMENTAL

Germination tests were carried out in accordance with the current rules^{*} of the International Seed Testing Association. Seeds were placed separately in wet paper towelling or on moist sand at 20° C to germinate. The first count was made at the end of 3 days, and germinated seeds were then removed; the final count was made at the end of 7 days.

The purpose of the test was "to determine the ability of the seed to produce normal seedlings which should be capable of continued growth in the soil under favourable conditions. The final basis for judging normal seedlings is an intimate first-hand knowledge based on continued comparative study of seedlings, produced under artificial laboratory conditions and in the soil." As a guide the rules indicated which types of seedlings may not be expected to develop plants in a soil test.

Comparison of conditions for germination in the laboratory under the above rules, and emergence in the field, however, revealed two major differences:

- (1) In the laboratory test, approximately 80% of seeds had germinated and were removed at the end of 3 days. At this stage the radicles were only 0.5-1.5 cm long and the plumules were just showing. In the field the seeds were sown about 4 cm deep and thus had to produce radicles 5-6 cm long and shoots 4-5 cm long before they emerged and were counted.
- (2) Whereas seedlings in soil took 7–15 days to emerge, depending on soil moisture and temperature, comparable seedlings developed in the laboratory in $4\frac{1}{2}$ –5 days.

Either or both of these differences could have contributed to the discrepancies between laboratory germination and field emergence in wrinkle-seeded peas. To investigate these differences, standard and modified laboratory germination tests and emergence tests in sand and soil were carried out.

(a) Germination and Emergence Tests

Standard germination tests were made as directed by the International Rules. The values so obtained were referred to as "standard" germinations.

Modified tests were made as above, but after the first count at 3 days germinated seeds were retained in the germinator tray and re-examined after 5 days. It was then possible to distinguish five "germination groups" in the wrinkle-seeded Greenfeast and William Massey peas used:

- Group 1: Seedlings which had continued vigorous growth after the first count and showed no sign of rotting of cotyledons or seedling axis.
- Group 2: Similar to group 1 but showing slight rotting of cotyledons.
- Group 3: Seedlings which showed poor growth after the initial count at 3 days. In these seedlings the root or shoot or both were partly rotted; the cotyledons were severely rotted.

 \ast These rules were modified in 1953 along lines similar to the procedure developed in this investigation.

- Group 4: Seedlings which had germinated but failed to develop further and in which the cotyledons and radicle were entirely rotted.
- Group 5: Seeds which had failed to germinate. Some of these were rotted, but in others the embryo had been damaged mechanically or by insect attack.

Seedlings in groups 1 and 2 were regarded as being "normal" under the seed testing rules and a count of these was made. This count is referred to as "vigorous" germination to distinguish it from "standard" germination.

Smooth-seeded White Brunswick peas showed no sign of rotting in any of the samples tested. All seeds which germinated continued growing vigorously and only seeds which were broken or showed damaged embryos failed to germinate.

Emergence was tested either in coarse sand or in "Waite Institute" loam. This is a heavy red-brown silt loam, pH 6.5, field capacity 20-22%, and wilting point 6-8% moisture.

(i) *Emergence in Coarse Sand.*—Seeds were planted 4 cm deep in sterilized sand at a moisture level of $10\%^*$ and incubated at 20°C. Emergence was complete after 5 days when counts were made. All seeds and seedlings were then removed and examined.

(ii) Emergence in Waite Institute Loam.—Seeds were planted 4 cm deep in Waite Institute loam at a moisture level of 12% and kept in the glasshouse at 15-18°C. Other investigations (Flentje 1964) showed that no attack by soil-borne fungi occurred under these conditions. After 15 days when emergence was complete counts were made and all seeds and seedlings were removed for examination.

(b) Seed Samples Used

More than 50 seed samples obtained from different areas in Australia and New Zealand were used in the investigation. The results are presented for five representative samples of each of the wrinkle-seeded varieties William Massey and Greenfeast and of the smooth-seeded variety White Brunswick from the following areas:

 Origin:
 New Zealand
 Victoria
 South Australia

 Variety:
 Greenfeast 1, 3
 Greenfeast 4
 Greenfeast 2, 5

 William Massey 2
 William Massey 3
 William Massey 1, 4, 5

III. RESULTS

The results obtained in the germination and emergence tests are given in Table 1. Statistical analysis of the data after angular transformation shows there was no significant difference between laboratory germination percentage and soil or sand emergence percentage for White Brunswick peas.

With Greenfeast and William Massey peas, however, the standard germination percentage was significantly higher than the soil emergence percentage for all but 2 of the 10 seed samples. Percentage vigorous germination and percentage emergence

* Moisture levels are expressed as percentage of oven dry weight of soil or sand.

TABLE 1

PERCENTAGE GERMENATION AND EMERGENCE OF 10 SAMPLES OF WEINKLE-SEEDED PEAS (GEBENFEAST, WILLIAM MASSEY) AND FIVE SAMPLES OF SMOOTH-SEEDED PEAS (WHITE BRUNSWICK)

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T.		Ċ	Greenfeast	ŧ			Will	William Massey	yey			Whi	White Brunswick	wick	
n cont	F	2	er	4	ũ		5	ო	4	ι¢	1	લ્ય	en	4	2
Standard germination	93.8	93 • 4	84 · 0	0.96	0.96	92.6	92.6 89.2	73-6	73-6 79-4 93-8	93-8	98-0	93.4	98.0 93.4 83.2	96 • 0	0-86
Vigorous germination	82.4	86-2	75-4	87.2	86.6	86.4	85.0	64.8	58.4	80-2	98•0	93.6	84.4	96.2	0.86
Sand emergence	84.0	88•4	76.4	84.4	85.2	87.6	86.8	63 · 2	56.4	81.6	97.4	94.2	83 • 4	95.6	97•4
Soil emergence	84.0	84 · 0	73.6	84-8	84•0	84.8	82.8	59.2	$61 \cdot 2$	80.8	80.8 98.0 93.6	93.6	85.2	95.8 97.6	97-6

in sand did not differ significantly from percentage soil emergence for any sample, but they were significantly lower than percentage standard germination for all except Greenfeast 2 and Massey 2, where only soil emergence was significantly lower than the standard germination.

The lack of any significant difference in percentage emergence in soil and sand indicates that the factors governing rate of growth of germinating seedlings in the field are not responsible for the differences between standard germination and field emergence: rate of development in sand was as rapid as in the germinator trays, and much more rapid than in the soil.

The varying ability of seeds in any sample to continue vigorous growth after germination, as revealed in the vigorous germination test, offers a more satisfactory explanation for the difference between standard germination and soil emergence.

TABLE 2 PERCENTAGE OF GERMINATED BUT WEAK SEEDLINGS IN THE MODIFIED GERMINATION TEST AND IN THE SOIL AND SAND EMERGENCE TESTS, COMPARED WITH THE DIFFERENCE BETWEEN STANDARD GERMINATION AND SOIL EMERGENCE PERCENTAGES

	Gre	eenfea	st: Sa	mple]	No.	Willi	am Ma	issey :	Sampl	e No.
	1	2	3	4	5	1	. 2	3	4	5
Difference between standard germination and soil emergence percentages	9.8	9.4	10.4	11.2	12.0	7.8	6-4	14.4	18.2	13.8
Weak and rotted seedlings in: Modified germination test Sand emergence test Soil emergence test	$9 \cdot 2$ 10 \cdot 0 $9 \cdot 0$	6.0	11.0 12.0 11.0	9.6	9·0 10·0 10·0	$5 \cdot 8$	$5 \cdot 0$ 5 \cdot 6 4 \cdot 8	12.8	$20 \cdot 0$ $20 \cdot 0$ $20 \cdot 0$	14 • 4

This suggestion is supported by examination of seeds and seedlings recovered from sand and soil after the emergence tests. These seeds and seedlings could be separated into five groups corresponding in vigour of growth and degree of rotting with those listed under the vigorous germination procedure. Seedlings corresponding to those in the germination groups 1 and 2 had, with few exceptions, emerged in both the sand and soil tests. Those in groups 3 and 4, however, had failed to emerge and as far as could be judged were incapable of making satisfactory plants. Immediately after examination these unemerged seedlings were replanted in moist soil in pots in the glasshouse, but less than 5% of them continued growth for more than 14 days, and even these produced unsatisfactory plants; the remainder died within 14 days. The numbers of such seedlings determined by the modified laboratory germination procedure and the sand and soil emergence tests are compared with the difference between standard laboratory germination and percentage emergence in soil in Table 2.

The difference between standard germination and soil emergence was closely correlated with the number of weak and rotted seedlings, except with Massey 1 where the difference between standard germination and soil emergence percentages was significantly higher at the 5% level than the number of weak and rotted seedlings

recorded in the germination and emergence tests. This appeared to be due to a number of seedlings which, although included in germination group 2, were just on the point of emergence in the soil and therefore had not been counted in percentage emergence.

(a) Organisms associated with Seed

The reason for the rotting and weak growth after germination was further investigated. It was not connected with the presence of recognized seed-borne pathogens such as *Ascochyta pisi* or *Pseudomonas pisi*. Duplicate 200-seed lots from each seed sample were examined for these organisms by two methods. Seeds were surface sterilized in 1 : 1500 mercuric chloride and after washing were planted on potato-dextrose-agar to detect fungal pathogens. Similarly treated seeds were placed in sterile tap water for 24 hr which was then streaked on nutrient broth plates to detect the presence of *Ps. pisi*. No trace of recognized pathogens was found, but *Rhizopus, Botrytis, Aspergillus, Mucor, Penicillium*, and many different bacteria were consistently isolated.

These organisms appear to occur below the seed coat as surface sterilization with mercuric chloride failed to remove them. In germination tests seed treated with mercuric chloride showed no difference from untreated seed, either in the amount of rotting that occurred or in the aerial growth of *Mucor*, *Botrytis*, and *Rhizopus*.

As surface dusting of seeds with fungicides is permitted under the International Rules, seed treated with Spergon was compared with untreated seed in further germination trials carried out with the 10 wrinkle-seeded samples. At the end of the test seeds were divided into the various germination groups as previously. There was no significant difference between the two treatments for any sample in either the amount of rotting or the number of seeds in the different groups. The only effect of Spergon treatment was to inhibit the aerial development of fungi and this facilitated counting in samples where the amount of rotting was high.

(b) Association between Harvesting Conditions and Rotting

Conditions at harvest, particularly the occurrence of rain, appear to influence the percentage of vigorous germination and amount of rotting which occurs in any seed sample. The harvesting conditions for the Australian samples of Greenfeast and William Massey were known. In Massey 3, 4, and 5, where rotting was most severe, between 0.3 and 0.5 in. of rain fell a few days before harvest; with all other samples no rain was recorded within 7 days of harvesting. The importance of conditions at harvest was emphasized in the case of another crop of William Massey not discussed above. Half the crop was harvested on one day and brought in under cover. Approximately 0.5 in. of rain fell overnight and the remainder of the crop was harvested 2 days later. The results of germination and emergence tests carried out on the two lots of seed obtained are shown below and illustrated in Figure 1.

Sample	Standard Germination	Healthy Germination	Emergence in Sand	Emergence in Soil
	(%)	(%)	(%)	(%)
A (before rain)	95	87	83	84
B (after rain)	75	50	47	48

IV. DISCUSSION

Germination tests carried out with wrinkle-seeded and smooth-seeded peas have shown that in any sample there may be some seeds in which the embryo has been damaged during harvesting or by insect attack. These seeds may remain ungerminated or produce obviously distorted seedlings.

In the smooth-seeded peas all undamaged seeds were usually able to continue vigorous growth after germination. In no case was there stunting or rotting associated with substantial fungal and bacterial growth. This suggests that germination tests



Fig. 1.—Emergence in Waite Institute soil at 10-12% moisture content of two seed samples from the one crop, harvested before (*left*) and after (*right*) rain.

carried out according to the old International Rules would give a reliable indication of the number of seedlings which would emerge in the field under favourable soil conditions: experience in the field in South Australia confirms this.

In wrinkle-seeded peas which were capable of germination within 3 days, a percentage, even under favourable conditions, are unable to continue growing vigorously enough to emerge when planted 4 cm deep in soil. Such seed, which varied from 4 to 20% between different samples, would have been included as "germinated"

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under the old International Rules, prior to 1953. Such germination counts would then have been too high by this percentage. A more accurate measure of their ability to grow would be obtained in the modified germination procedure suggested, in which the seeds are counted after $4\frac{1}{2}$ -5 days and separated into different groups. The International Rules, which were altered in 1953 to give a first count after 5 days and a final count after 8 days, now cover the difficulties outlined above.

Inability to continue vigorous growth is associated with a prolific growth of bacteria and fungi usually regarded as saprophytic. These organisms bring about rapid rotting of the cotyledons. If, as has been suggested, rainfall at harvest time is an important factor in increasing the amount of rotting that occurs in the seeds, it is possible that it does so by promoting growth of various fungi and bacteria which establish themselves between the testa and cotyledons or within these structures. Their activity at this time would be curtailed by subsequent drying of the seed, but would be resumed with the germinator trays. More work, however, is needed to determine whether this rotting is the main cause of restricted growth or whether it is a secondary effect which occurs only when the physiological condition of the seeds is altered by conditions during growth and harvesting.

Seed samples of wrinkle-seeded peas obtained from New Zealand showed the same range from low to high quality as did the samples from crops grown in Australia.

V. ACKNOWLEDGMENT

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