

serious threat to its production. A lot of work is being done for the improvement of this crop. Recently, during April, 1977 a severe attack of stem rust was noticed on indigenous (TL-29 and TL-193) and Mexican (CIMMYT) material in the Experimental Farm of Punjab Agricultural University, Ludhiana. The disease incidence in the material was as high as 80 to 100%.

The characteristic symptoms observed on triticale were small to large, brown pustules on the stem, leaf sheath (Fig. 1) and leaves. In the highly



FIG. 1. Rust pustules on triticale.

susceptible material, ears were also attacked. Uredospores were variable in shape ranging from round, oblong, ellipsoid or elongated, thick walled, golden brown, strongly echinulated, $24-53.5 \times 14.5-26.8 \mu$. Teleutospores ellipsoid, oblong, bicelled, constricted at the septum, wall thickened at the apex, smooth, dark brown, $15-24.5 \times 35-60 \mu$. The fungus is identified as *Puccinia graminis* f. sp. *tritici* (Pers) Eriks & Henn.

The triticale (TL-29) and wheat (agra local) were inoculated with freshly collected uredospores from triticale. Ten-day old plants after inoculation were kept in a humidity chamber for 24 hrs. and were transferred to glass house at 25° C. The

characteristic symptoms were observed both on triticale as well as on wheat plants after 15 days of inoculations.

Stem rust has already been reported on *Triticale* in Mexico¹ and Czechoslovakia², but from India it has not been reported so far. Its occurrence and the future potential threat, in India, should also be taken into consideration in the breeding programmes.

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ON THE OCCURRENCE OF ALTERNATIVE GERMINATION IN THE GENUS *PHASEOLUS* L (PAPILIONACEAE)

THE importance of seedling characters have lately been realized and it is only relatively recently that information on seedling characters began to be used for identification purposes^{1,4,5} and even for genetic studies².

Seedlings characters in the Leguminosae have been studied by Burger¹ and Kalyansundaram¹. The latter author surveyed 58 genera and 200 species for the germination character and has shown the occurrence of both types of germination in Papilionaceae and Caesalpiniae. In the former, hypogeal germination being more common than the epigeal, and the reverse in the latter, while Mimosae shows epigeal type of germination only.

In this communication, the authors report the occurrence of alternative germination in the genus *Phaseolus* whose epigeal germination is well known.

In Table I data pertaining to different species of *Phaseolus* investigated with regard to the type of germination are listed. Out of the 9 species investigated *P. angularis* and *P. multiflorus* show hypogeal germination and *Phaseolus vulgaris* (Fig. 1-3) as also the six other species show epigeal germination. It may be further noted that even the large number of seedlings tested under the different varieties of *P. mungo*, *P. vulgaris* and *P. aureus* included in the present study show only epigeal type of germination though transitional forms are known to occur³. To find out whether the epigeal

TABLE I
List of *Phaseolus* species showing type of germination

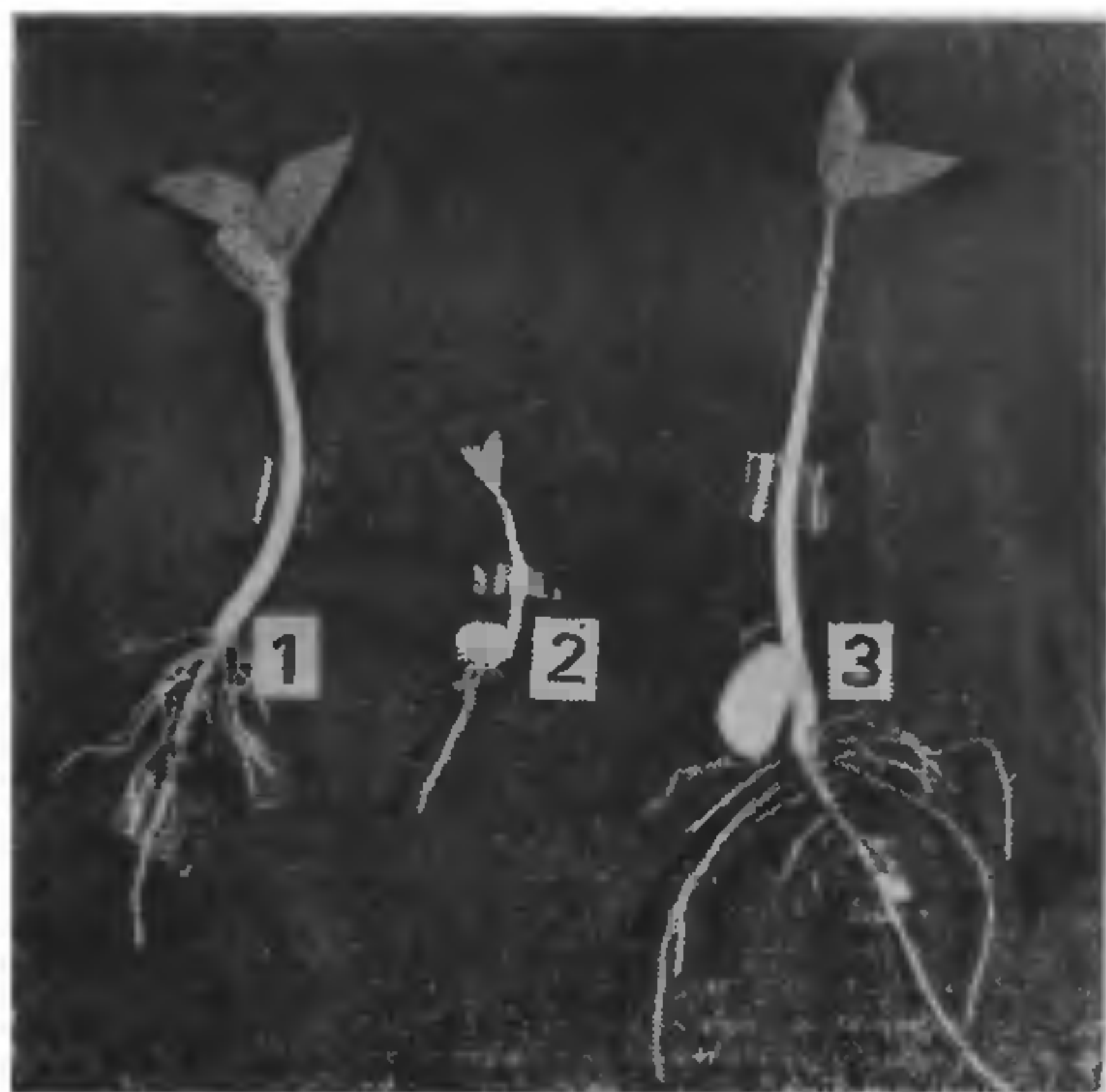
Sl. No.	Species/Variety	Type of germination	Number of seedlings tested	Source of material
1.	<i>Phaseolus angularis</i> EC 87897	Hypogeal	113	N.B.P.I., Reg. Stn., Simla
2.	<i>Phaseolus multiflorus</i> PLB 190-2	do.	99	do.
3.	<i>Phaseolus lunatus</i> EC 18199	Epigeal	102	do.
4.	<i>Phaseolus vulgaris</i> PLB 14-1	do.	83	do.
	<i>Phaseolus vulgaris</i> PLB K 1	do.	78	do.
	<i>Phaseolus vulgaris</i> (Local)	do.	109	Agric. Market, Warangal
	<i>Phaseolus vulgaris</i> Premier	do.	59	Commercial, Hyderabad
5.	<i>Phaseolus aureus</i> (Local)	do.	2142	Agric. Res. Stn., Warangal
	<i>Phaseolus aureus</i> PLM-629	do.	35	D.P.I., IARI, New Delhi
	<i>Phaseolus aureus</i> Pusa Baisakhi 100	do.	102	Commercial, Hyderabad
	<i>Phaseolus aureus</i> - Von G 65	do.	63	Agric. Res. Inst., Hyderabad
	<i>Phaseolus aureus</i> PR-16	do.	131	Commercial, Hyderabad
6.	<i>Phaseolus mungo</i> PLM-221	do.	28	D.P.I., IARI, New Delhi
	<i>Phaseolus mungo</i> (Local)	do.	2980	Agric. Market, Warangal
	<i>Phaseolus mungo</i> U-30	do.	45	A.R.I., Hyderabad
	<i>Phaseolus mungo</i> US-132	do.	64	do.
	<i>Phaseolus mungo</i> Krishna-11	do.	102	do.
7.	<i>Phaseolus atropurpureus</i> EC 276115	do.	16	D.P.I., IARI, New Delhi
8.	<i>Phaseolus aconitifolius</i> (Local)	do.	22	Kakatiya University Campus, Warangal
9.	<i>Phaseolus trilobus</i> (Local)	do.	23	do.

and hypogeal type of germination has any functional significance to the species, we looked for the presence of stomata on the flat, smooth, inner and outer convex surfaces of the cotyledons. The outer surface of the cotyledons of the epigeal seedlings was found to be stomatiferous while the stomata were totally absent in hypogeal cotyledons. The inner smooth surface in both the epi- and hypogeal seedlings was devoid of stomata and the epidermis was made of rectangular cells in the former and elongated to polygonal cells in the latter. The mature stomata are anomocytic with 5-6 surrounding cells and measure $14.5 \times 10.5 \mu$ with an elliptic stomatal pore. The guard cells are typically kidney shaped with their inner

walls thicker than the outer and with several chloroplasts in them and hence functional. Yet another interesting observation is that the epigeal cotyledons are rough and wrinkled with linear striations externally while the hypogeal cotyledons are smooth. We are unable to offer any explanation for this behaviour but it is probable that it may be functionally related to the stomatiferous epigeal cotyledons.

Kalyansundaram⁴ regards hypogeal germination in the Leguminosae as ancestral and the epigeal as derived from it. But according to Eames⁵ hypogeal germination is 'clearly advanced' and epigeal germination primitive. The occurrence of alternative type of germination just described, to

our knowledge, is not on record for the genus *Phaseolus*. In view of the controversial opinions expressed the present observations are significant and may serve as taxonomic pointer in delimiting species. The occurrence of alternative germination in the genus *Phaseolus* thus raises phyletic problems and until further work is carried out it would be premature to regard a given *Phaseolus* species as primitive or advanced merely on the germination criteria alone.



FIGS. 1-3. Fig. 1. *Phaseolus vulgaris* showing epigeal seedlings. Figs. 2-3. *P. angularis* and *P. multiflorus* showing hypogeal seedlings.

N.B. Longitudinal striations on cotyledons of epigeal cotyledon can be seen while the hypogeal cotyledons are smooth.

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AN IMPROVED AND SIMPLE LEUCOCYTE CULTURE TECHNIQUE FOR CHROMOSOMAL PREPARATIONS FROM ALBINO RAT

RODENTS have been widely used for many bio-medical investigations but their use for cytogenetic studies is not so extensive. One reason may be the lack of reliable tissue culture techniques, *in vitro*, for yielding a large number of cells at metaphase. The media for leucocyte cultures described by Nichols and Levan^{1,2} (1961, 1962) and Ford and Woollam³ (1963) have led to hemolytic reactions in our laboratory resulting in poor cell growth. The new method developed in this laboratory is a modification of the techniques described by William and Ray⁴ (1965) and Sankar and Giesler⁵ (1973). The technique reported here has certain advantages over existing ones. The medium, TC 199 has been employed in place of other media and Phytohemagglutinin-P, a more powerful agglutinating agent, has been added to the medium instead of Phytohemagglutinin-M. This has resulted in a considerable increase in the number of cells at metaphase and as a result, a minimum of 10 slides can be prepared from each tube. Furthermore the use of diluted foetal calf serum and very dilute sodium citrate, for hypotonic treatment⁵, has been omitted. Treatment with Hank's solution followed by a brief incubation in 1% sodium citrate has been used instead. Another advantage of this technique is its adaptability, by suitable modifications, for culturing leucocytes from other rodents as well. For culturing leucocytes, the whole blood can also be used without going through the procedure of separating leucocytes but we prefer leucocyte separation for obtaining optimum cultures. The minimum mitotic index obtained was 30.

The blood was drawn into a 10 ml sterilized syringe, by cardiac puncture, rinsed with heparin. 1 ml of plasma containing 1.2×10^6 leucocytes/ml was added to the culture tubes (Corning, 30 ml) having the prewarmed (37° C) culture medium. The medium included TC Medium 199 (Difco) 10 ml; 1 ml of a solution of Penicillin-G (1000 units) and Streptomycin sulfate (1000 mcg) (Sigma); 1.80 ml L-Glutamine (Sigma) 100 μ moles/ml in saline; 0.02 ml of Phytohemagglutinin-P (Difco) and 200 I.U. Heparin (Koch Light Laboratories). The cultures were incubated for