For determinations, 4 ml of the PDAB reagent is added to 1 ml of an aqueous solution containing indole acetic acid and the intensity of the coloured complex measured at 565 mμ. The method is superior to the conventional salkowsky[5] in rapidity, but is non-specific for indole acetic acid. The reagent forms colour complex with indole (λ_{max} 430 mμ), indole acetic acid (λ_{max} 565 mμ), indole pyruvic acid (λ_{max} 560 mμ), indole propionic acid (λ_{max} 585 mμ), indole butyric acid (λ_{max} 570 mμ) and indole lactic acid (λ_{max} 580 mμ).

Bacterial metabolism of tryptophan is characteristic of its degradation to indole and pyruvic acid with the liberation of ammonia. Micro-organisms can as well breakdown the side chain of tryptophan forming indole pyruvic acid and indole acetic acid. The classical method employed for qualitative detection of indole is the Kovac's test[6], employed for the classification of coliform bacteria[7]. The indole acetic acid and indole pyruvic acid however do not respond to Kovac's reagent[6]. The modified PDAB reagent in perchloric acid may be used for the qualitative detection of the different substituted indole compounds including indole acetic acid and indole pyruvic acid. Detection of indole acetic acid with the Salkowsky reagent[5] is time consuming while indole pyruvic acid poorly responds to the reagent.

CPCRI, Regional Station, V. P. POTTY.
Krishnapuram 690 533. K. V. JOSEPH.
Kayangulam, Kerala, N. P. JAYASANKAR.
October 15, 1977.

1. Ehrlich, P., *The Chemistry of Heterocyclic Compounds*. McGraw-Hill, New York, 1946, p. 68.
2. Jayasankar, N. P. and Bhat, J. V., *Anal. Biochem.*, 1966, **23**, 454.
3. — and Towers, G. H. N., *Ibid.*, 1968, **25**, 565.
4. Joseph, K. V., Potty, V. P. and Jayasankar, N. P., *J. Plant. Crops*, 1974, **2**, 1.
5. Gordon, S. A. and Palag, L. A. *Physiol. Plantarum*, 1957, **10**, 39.
6. Kovacs, N., *Z. Immun, Forsch.*, 1928, **55**, 311.
7. Burrows, W., *Text Book of Microbiology*, Toppan Company, Tokyo, 1968, p. 485.

STIGMATIC EXTRACTS OF *CHLOROPHYTUM HEYNEANUM* ENHANCE *IN VITRO* GERMINATION OF *C. MALABARICUM* POLLEN GRAINS

In vitro germination requirements of pollen grains vary. The pollen of some species are able to germinate in distilled water while a number of others require simple or mineral supplemented sugar solutions. In other cases, pollen grains easily germinate *in vivo* but fail to do so in culture media. Addition of stigmas to the culture medium may initiate pollen germination under such circumstances[1]. As for *in vivo* germination, stigmas generally support development of functional pollen of the same species but not of alien species[2]. However, we found that the pollen of *Chlorophytum malabaricum*, although inert on its own stigma, germinated easily on the stigma of *C. heyneanum*. Furthermore, stigmatic extracts of *C. heyneanum*, added to culture medium, brought about marked improvement in the rate of pollen germination of the other species.

C. heyneanum is male fertile with an average *in vitro* germination of 90%. The other species, *C. malabaricum*, bears short styled totally male sterile as well as long styled partially sterile flowers with an average of 30% germination *in vitro*. Only the latter type of flowers of this species was used in this study. As in other plants, *Chlorophytum* also shows flower to flower and even anther to anther variations in pollen germination. Discrepancies on this account have been minimised by mixing pollen grains from 20–25 flowers and taking samples from the same mixture.

In vivo germination was studied under laboratory and also field conditions. Germination was checked 2–3 hr after pollination by viewing unstained and cotton blue-stained stigmas under the microscope. Post-pollinated stigmas were also observed periodically up to 12 hr to study the development of pollen tubes.

The standard medium containing sugar, calcium and boric acid was used as controls and for trials ethanolic or distilled water extracts of twenty stigmas prepared according to the method of Namboodiri and Tara[3] were added to the medium. Two-dimensional chromatograms of the stigmatic extracts with BAW and Acetic Acid as solvent systems were developed and spot tests were conducted according to standard techniques.

A summary of pollen germination percentages under various conditions is given in Table I. *C. heyneanum* pollen germinate in standard medium in the range of 80–96%. Addition of alcohol to the medium inhibits the rate of germination by as much as 30%. No significant effect on the rate of *in vitro* germination occurred with the addition of stigmatic extracts — either of its own or that of *C. malabaricum*—to the medium.

The *in vitro* rate of germination of *C. malabaricum* is relatively low (20–35%). As in *C. heyneanum*, alcohol has an inhibitory effect on *in vitro* pollen germination of this species. Pollen of *C. malabaricum*

TABLE I

In-fluence of total stigmatic extracts of Chlorophytum *on* invitro *pollen germination. In trials, the standard medium uas supplemented with an equal volume of aqueous or ethanolic stiu'matic extractss In contiols, the extracts uere substituted with distilled water or ethanol respectiiely. The percentage of germination given is the average of 20 counts.*

Trials	Percentage of pollen germination in Controls		
		C. heyneanum	*C. malabaricum*
I. *Germination of* C. m·l:bzrcium *pollen in standard medium* plus *stigmatic extracts of*			
(*a*) *C. heyneanum* in distilled water	69·4	86·7	36·2
(*b*) *C. heyneanum* in ethanol	37·8	57·0	15·5
(*c*) *C. malabaricum* in distilled water	39·6	96·2	34·4
(*d*) *C. malabaricum* in ethanol	22·3	53·3	17·7
II. *Germination of* C. heyneanum *pollen in standard medium* plus *stigmatic extracts of*			
(*a*) *C. malabaricum* in distilled water	83·6	86·1	28·9
(*b*) *C. malabaricum* in ethanol	50·1	53·0	17·2
(*c*) *C. heyneanum* in distilled water	82·9	83·0	26·8
(*d*) *C. heyneanum* in ethanol	49·9	50·7	14·3

shows no germination on its own stigma though the pollen germinate and develop pollen tubes on the stigma of *C. heyneanum*. Stigmatic extracts of *C. heyneanum* (aqueous or alcoholic) increase the rate of *in vitro* pollen germination of this species by 20–30%. Addition of stigmatic extracts of *C. malabaricum* to the culture medium produces only marginal effect on the rate of pollen germination. The slight inhibitory effect of *C. malabaricum* extract on the germination of the pollen of both species is not considered significant in the background of natural variations exhibited in the rate of pollen germination.

These results indicate that the stigmatic extracts of C. heyneanum stimulate the *in vitro* pollen germination of *C. malabaricum*. It is therefore possible that the constituents of the extracts of *C. heyneanum* contain some factor or factors that promote pollen germination. A study of the chromatograms of the stigmatic extracts of both species show that there are three unique spots in the extracts of *C. heyneanum*. Two of these spots respond to colour tests for phenolics.

The promotional effects of stigmatic extracts of *C. heyneanum* in pollen germination focus attention on the role of stigmatic exudates in pollen development. Konar and Linskens[4] reported that stigmatic exudates of *Petunia* did not seem to contribute anything to pollen germination. In contrast, Labarca and Loewus[5] have shown that in *Lilium*, stigmatic exudates are used by the pollen for tube wall formation. Further, the need for some of the constituents of the stigmatic exudates for pollen germination was demonstrated in the colour mutants of *Impatiens sultani*[6]. Obviously no common role can be attributed to the stigmatic exudates in pollen development. However, present data shows that at least in *C. heyneanum* the stigmatic exudates have a direct influence in enhancing the germination rate of pollen grains of partially sterile species. Also, the preliminary identification of phenolics as unique spots in the stigmatic exudates of the fertile species is in line with the reported role of these chemicals in influencing growth processes[7].

We are grateful to Prof. C. A. Ninan for facilities and to the CSIR for the award of a Fellowship to one of us (M. B.).

Department of Botany,
University of Kerala,
Kariyavattom 695 581,
November 19, 1977.

MEERA BHASKAR.
A. N. NAMBOODIRI.

Mailing address: Dr. A. N. Namboodiri, Professor of Botany, University of Kerala, Kariyavattom 695 581, Kerala.

1. Johri, B. M. and Vasil, I. K. *Bot. Rev.*, 1961, 27, 325.
2. Martin, F. W., *Bull. Torrey Bot. Club*, 1970, 97, 1.
3. Namboodiri, A. N. and Tara, C. P., *Ind. J. Exptl. Biol.*, 1973, 11, 110.
4. Konar, R. N. and Linskens, H. F., *Planta*, 1966, 71, 372.
4. Laparca, C. and Loewus, F., *Plant Physiol*,. 1972. 50, 7.
6. Tara, C. P. and Namboodiri, A. N., *Am. J. Bot.*, 1974, 61, 585.
7. Galsto, A. W. and Davies, P. V., *Science*, 1969, 163, 1288.