sitol, 0.5 mg/l nicotinic acid, 0.5 mg/l pyridoxin HCl, 1 mg/l thiamine HCl, 30,000 mg/l sucrose. Growth regulators 2,4-dichlorophenoxyacetic acid, α-naphthalene acetic acid (NAA) and kinetin (Kn) were added to the media in various concentrations according to the experimental objectives. After 120 days of culture, 1000 mg/l KNO_3 was added to the media. For solidification 7000 mg/l agar was added and the pH adjusted to 5.7.

The explants were placed in culture tubes each containing 20 ml medium. The cultures were kept under 16/8 h light/dark period at 25±2°C and 50–60% relative humidity. Subculturing was done after every 40 days.

The MS basal media containing 1 mg/l NAA+ 1 mg/l Kn. Within 15 days, the explants became swollen, expanded in size and turned yellowish brown. After 30 days of inoculation calli were initiated from cut ends and from adaxial surface of the explants. The initial callus was compact and smooth. Subculturing was done after 40 days when the full explant with the callus was transferred on the fresh medium. After 120 days, when the third subculturing was done, 1000 mg/l KNO_3 was added to the media.

After 15 days of the third subculture, callus became smooth, glossy, nodular and yellowish green. Gradually, globular nodule-like structures appeared on the calli (figures 1, 2). The nodules consisted of small spherical cells with thick cytoplasm and large nucleus from which embryos derived. Further, the development of these nodular structures gave rise to a cluster of embryoids of different stages such as globular, banana-like and torpedo-like (figures 3–5).

The present study demonstrate that somatic embryo can be induced from callus in *A. cooperi* following KNO_3 treatment as reported earlier[7,8]. Further study is in progress to secure large scale production of regenerants and to explore the possibility of securing variant, if any, often noted in regenerants from callus.

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ORGANOGENESIS IN ANTHER-DERIVED CALLUS CULTURE OF COWPEA [*VIGNA UNGUICULATA* (L.) WALP]

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THE culture of the whole anther and subsequent development of plants was successfully demonstrated with *Datura innoxia*[1]. Raising of anther callus as reported in cultured anthers of *Cajanus cajan*[2,3], *Phaseolus aureus*[4] and in *Winged bean*[5], and subsequent attempts for plant regeneration from the callus is a significant step towards legume improvement programme[6,7]. A similar attempt was made in *Vigna unguiculata*, an economically important drought-resistant food legume.

Flower buds of cowpea of different sizes were taken and anthers at several stages of development were excised and cultured. Callusing was observed only in young anthers containing thin-walled microspores at late uninucleate stage. Cultured anthers kept under weak light (250 lux) or continuous dark conditions for initial 20 days only callused while no callusing occurred in continuous light condition (250 lux). Further, callusing was obtained in the culture chamber at a relatively lower temperature (23 ± 1°C).

Kinetin or BAP in combination with IAA or IBA gave best results towards anther callusing. 1 mg/l Kn/BAP+0.5 mg/l IAA/IBA gave 85% response to anther callusing. A good response (50–60%) was also obtained on MS medium supplemented with 2 mg/l Kn/BAP+1 mg/l IAA/IBA. NAA and 2,4-D did not elicit any anther response when used in place of IAA/IBA. Cytokinins and auxins (0.5–5 mg/l) when used singly in the agar MS medium failed to induce anther callusing. Similarly

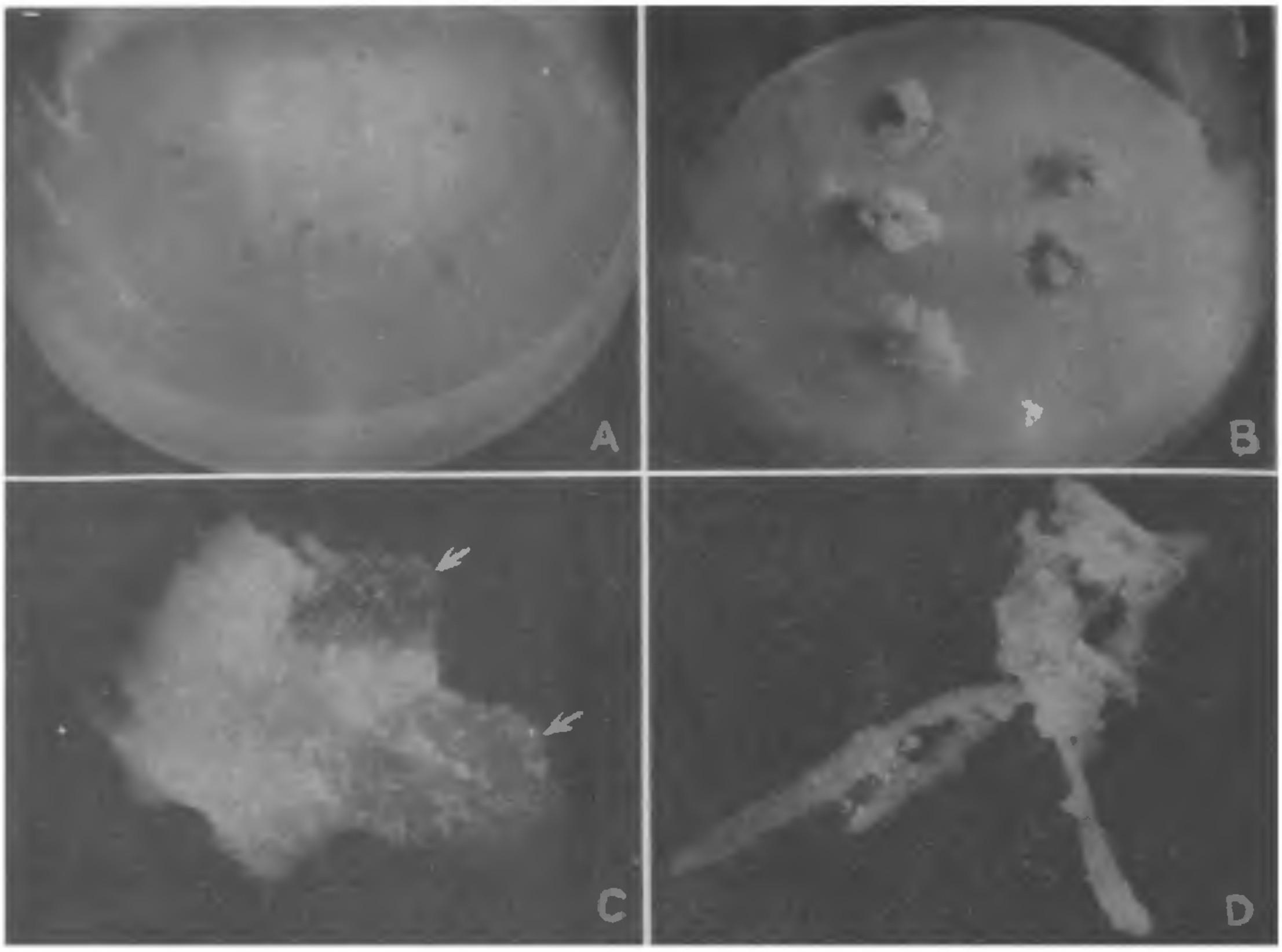

Figure 1A–D. **A.** Anthers of *Vigna unguiculata* at the time of inoculation; **B.** Anther callusing on MS + 1 mg/l BAP + 0.5 mg/l IBA; **C.** Organogenesis (leaf-like structures) in suspension cultures, and **D.** Rhizogenesis from the callus mass.

no response was noted in liquid medium with varied combination.

Anther callus so obtained on the solid medium was subcultured after 40 days of growth in liquid MS medium with 1 mg/l BAP + 0.5 mg/l IBA. Flasks inoculated each with about 200 mg callus were maintained on a rotary shaker (100 rpm) under weak light conditions. Greening of the callus followed by the emergence of leaf-like structures was observed in 2–3 weeks (figure 1C) after inoculation. In the second experiment anther callus obtained on the solid MS medium when subcultured after 40 days of callus growth on the solid MS medium supplemented with 1 mg/l Kn/BAP + 0.5 mg/l IBA gave extensive rooting in 25–30 days of subculture under continuous light conditions (figure 1D). No shoot formation was observed in organogenic/rhizogenic callus cultures so obtained, even when subcultured on MS with varied concentration of growth regulators. The exact nature of anther callus and its origin in the present study could not be traced out. Further attempts for complete plant regeneration are in progress.

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SURVIVAL OF *COLLETOTRICHUM CAPSICI* (SYD.) BUTLER AND BISBY WITH URD BEAN (*PHASEOLUS MUNGO* L.) SEEDS

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SEED plays a vital role in the production of healthy crop. Seed-borne disease of urd bean like anthracnose, can reduce the yield to a considerable extent. Anthracnose caused by *Colletotrichum capsici* (Syd.) Butler & Bisby was first observed[1] on *Phaseolus mungo* L. Urd bean is one of the major pulse crops of Northern India, and not much work has been done on *C. capsici*, so it was thought desirable to study the role of seed in perpetuation of anthracnose disease in the prevailing Pantnagar conditions. The present work comprises the aspects of survival of *C. capsici*, germination percentage of infected seeds and comparison between blotter paper and agar plate methods. This study confirms the above mentioned aspects of *C. capsici*, unknown till to date on urd bean.

During September 1983, seeds from severely infected pods of urd bean (variety Pant U-19) were collected, dried and stored at 5 C in a polythene bag. The same seed lot was used throughout the experiment each month starting from September, 1983 to August 1984. Twelve hundred seeds were tested by blotter and agar plate methods, each, as recommended by Neergaard and Saad[2], and International Seed Testing Association[3]. Potato dextrose agar medium was used in the agar plate method for plating the sterilized seeds. All petri plates were incubated at 25 ± 1 C for 7 days in an incubator. Subsequently, the fungal infection was observed after 7 days by microscope and sometimes with the help of stereobinocular.

Infected seeds were soaked overnight (for 16 h) in distilled water and then placed in plastic petri plates lined with moist filter paper (Whatman No. 1) under laboratory conditions (25 ± 1°C). Germination was recorded after 10 days for each month.

The data reveal that the fungus was readily isolated from seeds, immediately after harvest, i.e. from September 1983 to as late as August 1984 (table 1). The percentage of seeds that yielded the fungus varied (from 11 to 28%) in unsterilized and (8–24%) in sterilized seeds by the blotter paper method. Agar plate method, on the contrary reported a lower (3–19%) fungal population. Germination range of infected seeds was 46–76%. However, the difference is significant between the initial and last stages of the experiment.

Unsterilized seeds exhibited 20.25% pathogen (table 2). In contrast to unsterilized seeds, 16.25% of

Table 1 *Isolation frequency of C. capsici and germination percentage of stored urd bean seed by monthly interval*

Months	Percentage of *C. capsici* yielded from seeds: Blotter paper: Unsterilized seeds	Blotter paper: Sterilized seeds	Agar plate method	Per cent germination of seeds
Sept (1983)	28	22	19	46
Oct.	27	23	16	47
Nov.	27	24	16	53
Dec.	28	24	15	56
Jan. (1984)	22	17	15	54
Feb.	21	18	12	63
Mar.	18	15	12	62
April	17	13	8	72
May	17	13	7	71
June	14	10	8	68
July	13	8	4	74
Aug.	11	8	3	76
C.D. at 5%	3.91	3.68	3.02	5.85

Table 2 *Per cent recovery of Collectotrichum capsici with urd bean seeds (No. of seeds planted: 1200)*

Treatments	No. of seeds yielded *C. capsici*
Blotter paper method	
(a) Unsterilized seeds	243 (20.25%)
(b) Sterilized seeds	195 (16.25%)
Agar plate method (sterilized seeds)	135 (11.25%)