RESPONSE OF YOUNG INFLORESCENCES OF *ANETHUM GRAVEOLENS* L. TO GROWTH SUBSTANCES

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THE inflorescence of *Anethum graveolens* contains potentially hermaphrodite, male and underdeveloped coral buds (*see* Sehgal).[1] Young inflorescences (6 mm. × 1·25 mm.) were surface-sterilized with chlorine water for 4-6 min., thoroughly washed in sterile, distilled water, and cultured under aseptic conditions on modified White's medium (WM) containing 4% sucrose. The medium was jelled with 0·8% Difco bacto-agar and supplemented variously with adenine (Ad), casein hydrolysate (CH), coconut milk (CM), indoleacetic acid (IAA), kinetin (Kn), yeast extract (YE), and 2,4-dichlorophenoxyacetic acid (2,4-D). The pH of the medium was adjusted to 5·8, and 15 ml. of the medium was dispensed in each culture tube. The medium was autoclaved at 15 lb./sq. inch for 15 min. The cultures were maintained in diffuse daylight, at a temperature of 25 ± 2° C. and 55 ± 5% RH. For each experiment 72 cultures were raised and the experiments were repeated once.

At the time of culturing, the inflorescences bore floral buds at stages ranging from a convex protuberance (without the differentiation of floral organs) to buds showing primordia of corolla, androecium and gynoecium. On WM the inflorescences shrivelled within 2 weeks. On WM + IAA (0·5, 1·0 ppm.) the inflorescence axis as well as the pedicels of flowers callused within 3 weeks. The growth of callus was slow and in another 2 weeks a number of roots differentiated from it (Fig. 1, A, B). In the above medium only 5–10 floral buds developed into hermaphrodite flowers (Fig. 1, B) as compared to nearly 400 such flowers in nature. However, even these finally collapsed due to lack of cross-pollination and fertilization.

On WM + CM (10%) the inflorescence axis as well as pedicels of flowers produced brownish-white fluffy mass of friable callus within 3 weeks. Addition of 2,4-D (1 ppm.) to the above medium improved growth of the callus, and it was successfully subcultured. Profuse callusing was obtained on WM + CH (500 ppm.) + 2,4-D (0·5 ppm.) + Kn (0·5 ppm.). However, the callus failed to differentiate into organs when left *in situ* or on transfer to WM with or without YE (1000 ppm.). If a portion of the callus was transferred to WM + KNO_8 (5 mM/1), roots developed all over the explant in 24% cultures. When transferred to WM + $(NH_4)_2SO_4$ (5 mM/1), numerous whitish nodules developed in 36% cultures within 2 weeks. In another week these nodules developed shoot buds (Fig. 1, C), the growth of which was slow and they did not grow beyond 1·5 cm. On WM + Ad (10 ppm.) the callus produced both roots and shoots in 56% cases, 4 weeks after subculture.

On WM + CH (500, 1000 ppm.) and WM + YE 8–14 floral buds developed into hermaphrodite flowers, while the remaining floral buds collapsed and the inflorescence axis as well as pedicels of flowers produced callus 3 weeks after culture. The callus showed slow growth and in another week produced several embryoids (Fig. 1, D). The ontogeny of the embryoids did not conform to any of the conventional type of embryogeny,[2] but the globular and heart-shaped stages were gone through as usual. The embryoids showed 2-4 cotyledons and developed into plantlets *in situ* (Fig. 1, E).

Young floral buds of *Anagallis arvensis*,[3] *Viscaria candida* and *V. cardinalis*[4] have been successfully reared to mature flowers in cultures. However, those of *Kalanchœ pinnata*,[5] *Phlox drummondii*[6] and *Ranunculus sceleratus*[7] failed to do so. All the floral buds in the young inflorescences of *Anethum graveolens* (present work) did not produce hermaphrodite flowers, but it has been possible to induce differentiation of the callus derived from the inflorescence axis and the pedicel of floral buds. This was achieved by appropriate balance of growth adjuvants in the medium.

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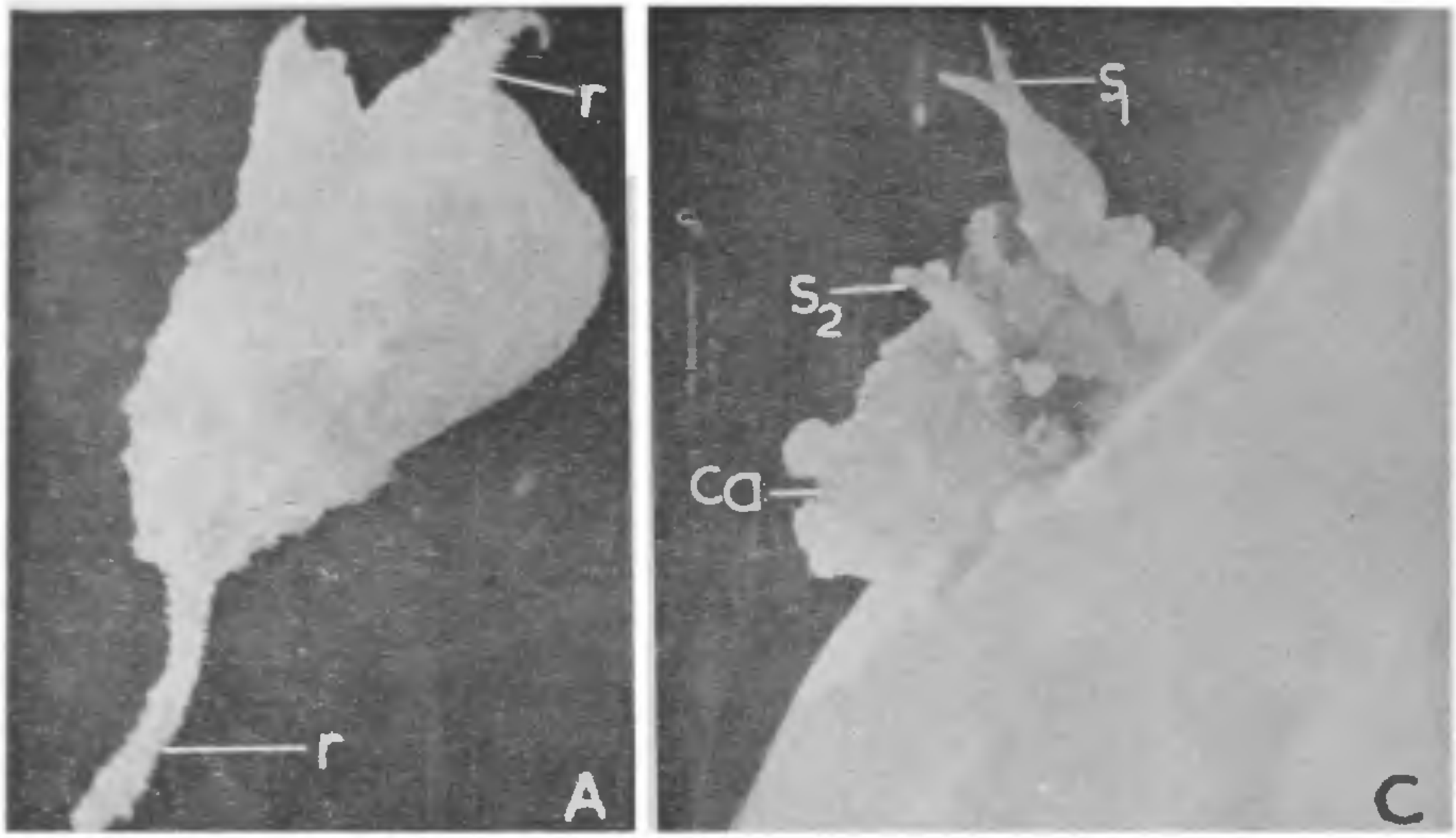

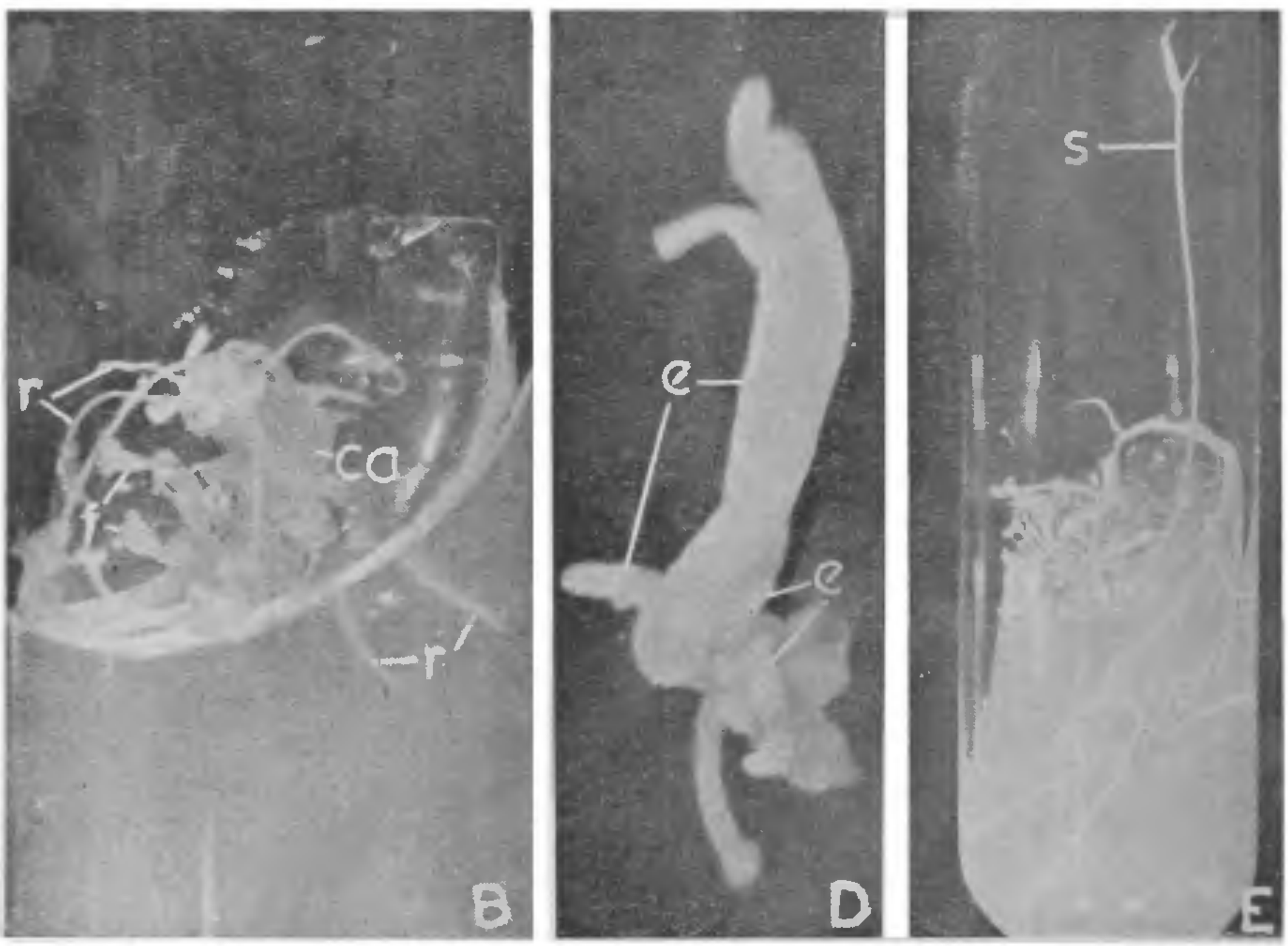

FIG. 1, A–E. Fig. A. A 5-week-old culture on WM + IAA (1 ppm.); note the presence of roots on the callus, × 8. Fig. B. Same, 11-week old; note profuse rooting and 5 hermaphrodite flowers (only 2 have been labelled), × 2·5. Fig. C. A portion of callus, raised on WM + CM (10%) + 2, 4–D (0·5 ppm.) subcultured for 10 weeks on WM + $(NH_4)_2SO_4$ (5 mM/l), showing shoot buds, × 3·25. Fig. D. Polyembryoid mass from WM + YE (500 ppm.) showing some developmental stages, × 35. Fig. E. 15 week-old culture on WM + YE (500 ppm.), showing normal plantlet, × 1·25. (*ca*, callus; *e*, embryoid; *f*, flower; *r*, root; *s*, shoot; s_1, s_2, shoot buds).