Evidences for primary hornblende could, therefore, not be discernible and its genesis is attributed to retrogression that has led to its formation at a temperature and pressure range of 550–625° C and 3–4 kb respectively, being the maximum in the facies transitional to granulites[5]; and the porphyroblastic growth by metamorphic differentiation.

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GROWTH OF BARLEY AND WHEAT ENDOSPERM IN CULTURES

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ENDOSPERM plays a very significant role in the nutrition and differentiation of embryo. In angiosperms it develops as a result of triple fusion and is mostly triploid. The tissue derived from the culture of endosperm is a homogeneous mass of parenchymatous cells and, therefore, offers a very suitable system for studies on growth and differentiation.

In recent years several attempts have been made to culture immature and mature endosperm, but success has been very limited. So far it has been possible to culture and induce differentiation in the endosperm of some dicotyledonous plants belonging to Euphorbiaceae, Loranthaceae and Santalaceae (*see* Johri[1]; Sehgal[2]).

In 1947, LaRue[3] succeeded in establishing cultures of maize endosperm. Since then several workers (Sehgal[4]; Straus[5]; Straus and LaRue[6]; Tamaoki and Ullstrup[7]) made futile attempts to get differentiation and organogenesis in maize endosperm callus. Norstog[8] established continuous cultures from the endosperm of English rye grass. However, he also could not get differentiation from the callus (*see also* Norstog *et al.*[9]). Trione *et al.*[10] failed to grow the endosperm of wheat. The present work was undertaken to study the morphogenetic potentialities of endosperm in two monocotyledonous plants; barley (*Hordeum vulgare* L.) and wheat (*Triticum aestivum* L.).

Ovaries of *Hordeum* and *Triticum* collected 8 days after pollination were surface-sterilized with chlorine water for 7–10 minutes, followed by rinsing in sterile distilled water. The chalazal part of the endosperm was scooped out and planted under aseptic conditions on modified White's basal medium containing 4% sucrose jelled with 0·8% Difco Bacto-agar (WM). The medium was also supplemented with various concentrations of adenine (Ad — 20, 40 ppm); autoclaved, coconut milk (CM — 10, 20%); casein hydrolysate (CH — 0·1, 0·25%); indole acetic acid (IAA — 1, 5 ppm); kinetin (Kn — 0·5, 1 ppm); yeast extract (YE — 0·1, 0·25%); zeatin (Ze — 0·5, 1 ppm) and 2,4-dichlorophenoxy acetic acid (2,4-D–1, 5 ppm) either singly or in various combinations. The pH of the medium was adjusted to 5·8 before autoclaving. For each treatment 48 cultures were maintained in diffuse daylight at 25 ± 1° C and 55 ± 5% relative humidity.

In the preliminary experiments endosperms were cultured 4, 6 and 8 days after pollination. The ones excised after 4 and 6 days failed to respond to any of the treatments. Therefore, in subsequent experiments only the endosperms collected from grains 8 days after pollination were inoculated.

In *Hordeum* the endosperm failed to grow on WM or WM supplemented with various concentrations of the above growth regulators, either singly

or in various combinations excepting CH + IAA. On WM + CH (0·25%) + IAA (1 ppm) the endosperm showed the initiation of callusing 10 days after inoculation (Fig. 1 A) in 68% cultures. The growth of callus was slow (Fig. 1 B). When this callus was transferred to WM + CM (10%) + 2, 4-D (1 ppm) it grew profusely (Fig. 1 C, D) and was yellowish-green in colour. This callus could be easily subcultured on the above medium. Though the callus was cultured continuously for 12 months

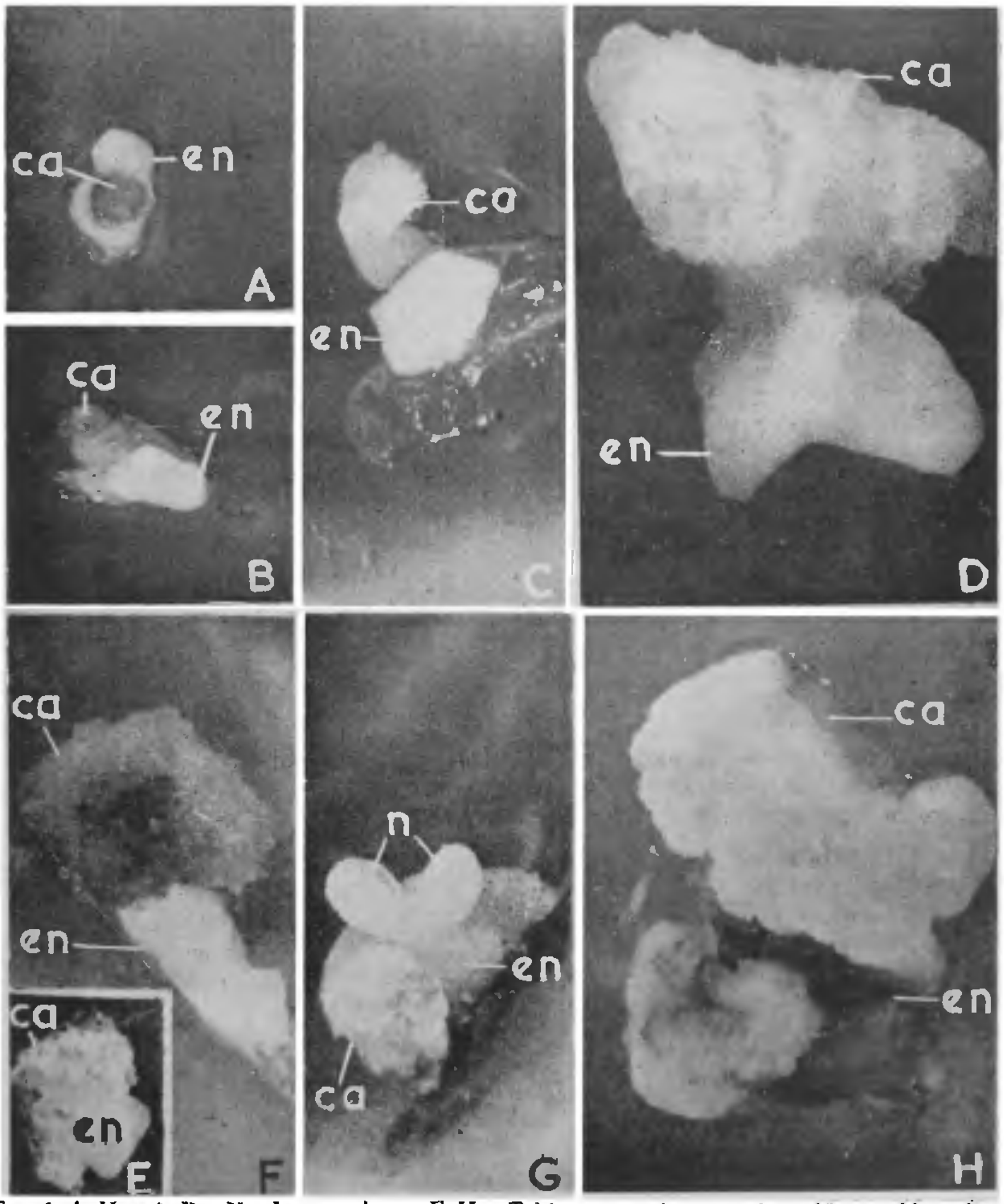

FIG. 1 A–H. A–D. *Hordeum vulgare*, E–H. *Triticum aestivum*. A. 10-day-old culture on WM + CH (0·25%) + IAA (1 ppm), showing initiation of callus. × 4. B. Same, 4-week-old. × 4. C, D. Endosperm raised for 4 weeks on WM + CH (0·25%) + IAA (1 ppm) subsequently transferred on WM + CM (10%) + 2, 4-D (1 ppm) and grown for 1 and 2 weeks, respectively. × 4. E. 1-week-old culture on WM + CM (10%) + Kn (0·5 ppm) + 2, 4-D (1 ppm). × 3. F. Same, 2-week-old; note profuse callusing. × 5. G. Endosperm raised for 1 week on WM + CM (10%) + Kn (0·5 ppm) + 2, 4-D (1 ppm) and subsequently transferred on WM + Ad (20 ppm); note two nodular outgrowths which appeared one week after transfer. × 5. H. Same, 2 weeks after transfer. × 5. (*ca*, callus; *en*, endosperm.)

and subjected to different treatments, so far it has not produced any root or shoot.

In *Triticum* the endosperm failed to grow on WM as well as WM supplemented with Ad (20, 40 ppm); CH (0·1, 0·25%); IAA (1, 5 ppm); Kn (0·5, 1 ppm); YE (0·1, 0·25%); Ze (0·5, 1 ppm) and 2, 4-D (1, 5 ppm) individually or in various combinations. However, an actively growing callus was obtained on the following combinations: (*a*) WM + CM (10%) + IAA (1 ppm); and (*b*) WM + CM (10%) + Kn (0·5 ppm) + 2, 4-D (1 ppm). Of these two combinations, the latter proved to be better because on this medium 84% cultures showed callusing as compared to 62% on WM + CM (10%) + IAA (1 ppm). Callus was initiated one week after culture (Fig. 1 E). It grew rapidly to form a whitish-yellow friable tissue in another week (Fig. 1 F). With the passage of time, proliferation continued but the callus failed to differentiate into plantlets. When the above callus was transferred to WM + Ad (20 ppm) it formed nodular outgrowths in 56% cultures (Fig. 1 G). These outgrowths did not differentiate but callused further (Fig. 1 H)., Trione *et al.*[10] tried as many as 20 different media to culture wheat endosperm, but all their attempts failed.

In the present study I have been able to culture barley and wheat endosperm. However, it has not been possible to get differentiation from the callus. It is concluded that though differentiation from the endosperm of some dicotyledonous plants has been achieved, the production of callus and its differentiation into plantlets from the endosperm of monocotyledonous plants is yet a challenging problem.

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