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Studies on the Mericlinal Protocorm of Orchid (IV)

Protocorm development from seed embryo

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洋蘭의 生長點培養에 關한 研究

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ABSTRACT

Several days after culture, the parts around suspensor turned brown. In about 10 days the embryo started to form protocorm sending out hairs through seed coat. Around 20 days after culture, most of the protocorms emerged out of seed coat and some of them began to take green color. When observed two months after culture, the protocorm took the characteristic top-shape feature.

INTRODUCTION

Thanks to Morel's pioneer works on the mericlinal propagation (Morel 1960, 1964, 1965, 1966, Champagnat et al. 1966) now mass production of orchid by means of vegetatively increasing protocorm-like body has become routine work practiced by many commercial orchid grower. However, the identity of protocorm-like body and its initial development from cultured explant such as shoot apex, whole bud, or sliced protocorm-like body have hardly been investigated (Champagnat et al. 1966).

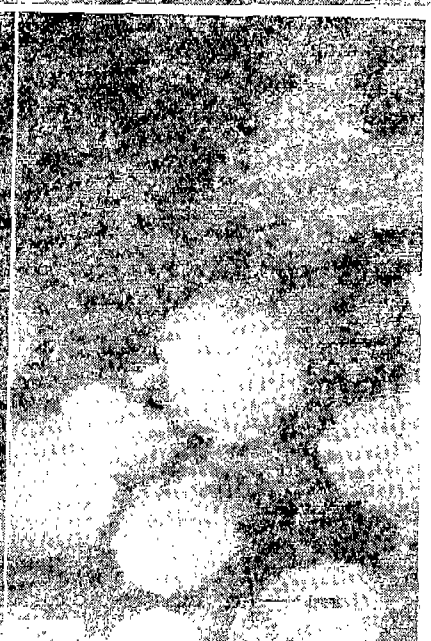
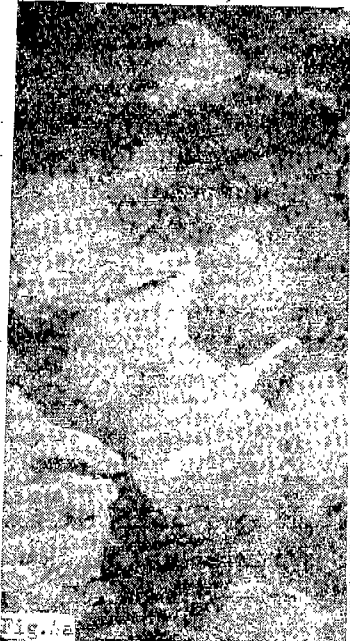
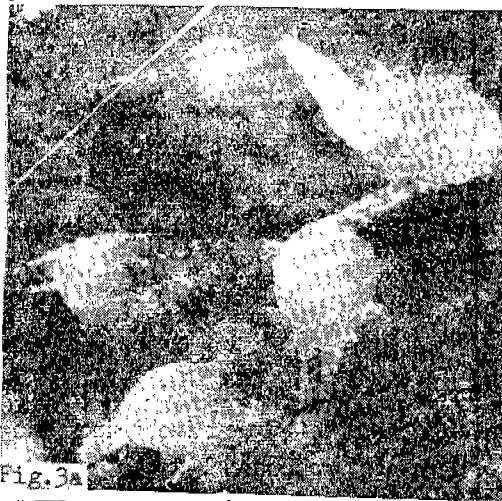
The author, in his series of studies on mericlinal protocorm (Harn 1968 a,b,c,d, Hamaya et al. 1968), has been interested, in the question of whether the mericlinal protocorm (protocorm-like body derived from meristem culture) is the same one as the embryonal protocorm (protocorm developed from embryo).

Many works have been reported already by many botanists on the development of protocorm and plantlet from seed embryo, using various orchid species (Kano 1965, Kohl 1962, Torikata 1965).

Present experiment was designed to compare the identities of two kinds of protocorms, embryonal and mericlinal. Observation on the mericlinal protocorm, however, will be reported in different paper.

MATERIALS AND METHODS

Seed embryo of *Cymbidium* whose varietal name was unknown were sown on Hyponex-peptone medium or Knudson C supplemented with 1 ppm NAA and 0.1 ppm 2,4-D, with pH adjusted at 5.2 after autoclaving. No clear difference was observed between the two kinds of medium in



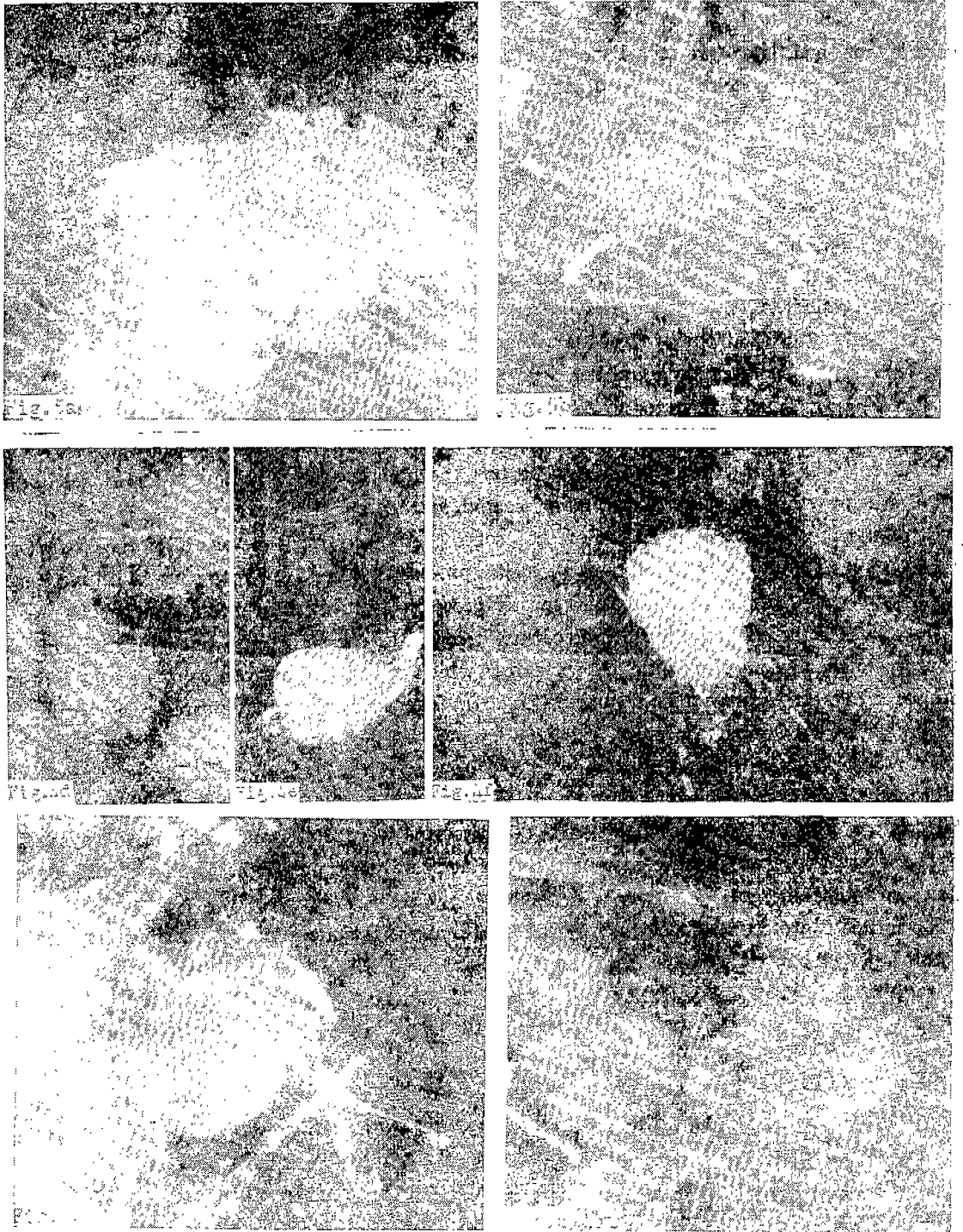


Fig. 1, seed with undifferentiated embryo encased in papery seed coat; Fig. 2, 9 days old embryo starts to grow; Fig. 3, embryo-protocorm 13 days old; Fig. 4, 23 days after culture; Fig. 5, long-haired protocorms emerge out of seed coats; Fig. 6, top-shape orientation of protocorm.

germination and later growth. Part of the materials was fixed in acetic alcohol at definite intervals for later histological investigation.

OBSERVATION

Cymbidium seed is an undifferentiated embryo consisting of mass of cells enveloped by papery seed coat(Fig. 1).

When observed several days after culture, the part around the suspensor of the embryo turned brown. 9 days after, the embryo already started to develop into protocorm, sending out hairs, the originally ellipsoidal embryo becoming swollen and turning rather roundish(Fig. 2). 13 days after culture, the number of hairs increased and the hairs became more elongated. Some of the swollen embryo finally emerged out of the thin papery one-layered seed coat(Fig. 3). The embryo-protocorm became larger, the color turned milky-white, and there was no sign of chloroplast formation. This variety seemed to be early germinating type. 19 days after, the embryo-protocorm grew still larger and the hair became longer, but the color remained milky-white. Most of the protocorms emerged out of seed coat. 23 days after culture, some of the protocorm started to get green color. The hitherto roundish protocorm turned to be top-shape with the centre of flat top concaved(Fig. 4). The hairs mostly grew on the underside of the protocorm.

One month after culture, all the protocorms were out of the seed coat, but still most of the protocorms remained milky-white except a few which turned to be green color(Fig. 5). Flat-top in most of the protocorms turned upward(Fig. 6). The hair might have some role in orienting the protocorm to a top-like posture.

When observed two months after culture, the protocorm took the characteristic feature, the flat-top concaving in the center and giving rise to projectiles. The top-flat as well as the lower tapering part turned dark green. When the protocorms were full grown around 70 days after culture, there appeared in the center of the top-flat some variegated light part from which shoot of new plantlet emerged.

The mode of protocorm development from seed embryo is quite similar to the development of protocorm-like body from cultured shoot apex.

摘 要

洋蘭의 生長點培養에 의한 增殖法の 開發로 洋蘭栽培에 莫大한 利得을 가져왔고, 最近 이 技術의 開發에 對해 더욱 많은 努力이 傾注되고 있다. 그러나 生長點培養에 對한 여러 基礎研究은 아직 거의 안되고 있는 實情이다.

本 研究은 生長點由來의 原塊體樣體(Protocorm-like body)와 胚發生時 形成되는 原塊體(Protocorm)의 를 比較키 爲한 一連의 實驗인바, 于先 胚起源의 原塊體의 形成過程을 報告한다.

培養 數日後에 벌써 Suspensor 附近이 褐變하고, 約 10日이 되면 胚는 原塊體化하고 種皮를 뚫고 毛茸을 낸다. 20餘日이 경과되면 原塊體는 種皮外로 거의 全部나오고 一部 原塊體는 綠變하기 始作한다. 約 2個月 後면 原塊體는 典型的인 倒圓錐形의 特徵을 갖게 되는데 外觀으로는 生長點由來의 原塊體樣體와 전혀 區別이 안된다.

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