Effects of Ecdysterone on the *in Vitro*
Development of Wing Discs of Rice
Stem Borer, *Chilo suppressalis*
(Lepidoptera : Pyralidae)¹

Recently OBERLANDER and FULCO (1967) reported that wing discs from mature larvae of wax moth, *Galleria mellonella* partially metamorphosed *in vitro* after the addition of ecdysone to the culture medium. Subsequently it was demonstrated that even very small wing discs from the final instar larvae were competent to respond to ecdysone and its analogues, including ecdysterone, *in vitro* (OBERLANDER, 1969).

In the present paper we intend to make sure how the moulting hormone ecdysterone affects the growth and differentiation of the wing discs taken from the different stages of the last instar larvae of the rice stem borer, *Chilo suppressalis in vitro*.

The culture methods and the medium used for this experiment were the same as those mentioned previously (MITSUHASHI and MARAMOROSCH, 1964). Wing discs taken from diapausing larvae, or non-diapausing larvae within 1 day after the final moulting did not show any morphological changes during cultivation in the medium containing ecdysterone, where the dosage was 3 µg per ml of the medium. On the other hand, more developed wing discs from non-diapausing larvae of 2-3 day stage of the final instar which completed the formation of channel and the distribution of tracheoles into the wing tissue, distinctly metamorphosed about 48 hr after cultivation. More precisely, epidermal cells on both sides of the basement membrane enlarged much, and new cuticle formation was observed. It may be added that when cultivated without ecdysterone, wing discs of this stage did not develop as a whole, and furthermore partial degeneration of epidermal cells occasionally occurred (Fig. 1). The most remarkable changes of the wing discs caused by ecdysterone were seen when the larvae reached to the 3-4 day stage of the final instar. Namely the wing tissue rapidly stretched out, breaking the hypertrophied wing pouch. Nevertheless, the wing discs at this stage could not develop *in vitro* without ecdysterone. In the wing discs of the later stage, about 1-2 day before pupation, it was proved

Fig. 1. A : Longitudinal section of *Chilo* wing disc, 48 hr after onset of cultivation, showing the degeneration of wing bud in the medium lacking ecdysterone ($\times 600$). B : Longitudinal section of *Chilo* wing disc, 48 hr after onset of cultivation, showing the multiplication and enlargement of the epidermal cells and formation of the new cuticle in the presence of ecdysterone. Wing disc was fixed in BOUIN's fluid, embedded in paraffin, sectioned at 6 µ and stained with DELAFIELD's hematoxylin-eosin. Cu : cuticle; EpC : epidermal cell; BM : basement membrane.

that they were able to grow and differentiate *in vitro* even in the absence of ecdysterone. Therefore, at this stage, the wing disc tissues may already have received hormonal action or have had the capability to develop by themselves.

In conclusion, ecdysterone affects neither the less developed wing discs of the diapausing larvae, nor those of non-diapausing larvae at the early stage of final instar. However there seems to be a critical stage in which they are highly susceptible to this hormone. An interesting fact is that probably the wing discs which have passed over this critical stage, did not require any control of the moulting hormone to develop and extend the

¹ Appl. Ent. Zool. 4(3) : 158—159 (1969)

wing bud.

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