Histopathology and Histochemistry of Insects Treated with Chemosterilants

II. On the studies of DNA in active testes of *Poicelocerus pictus* (Orthoptera: Acrididae) treated with apholate^{1,2}

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Successful sterilization of insects by chemosterilants has given an impetus to study their effect on reproductive organs in insects. Of the various chemicals tested as insect chemosterilants, alkylating agents, have given more spectacular results than any other. Though many papers are appearing on their effects on gonads (Cantwell and Henneberry 1963, Chamberlain 1961, Morgan and LaBrecque 1962, 1964, Schwartz 1964) little is known about their mode of action. In the present paper histochemical studies of the effect of apholate, an alkylating agent, on the DNA in the active testes of *Poicelocerus pictus* is discussed.

The adult *P. pictus* were caught in June and July 1967 and acclimatised in the laboratory. They were injected with 0.1 mg apholate per insect in glass distilled water. The animals were autopsied after 15, 24 and 30 days of treatment. The testes were taken out and fixed in Carnoy. After usual procedures they were subjected to Feulgen reaction for DNA studies.

The observations in the normal testes reveal a positive reaction in the chromatin of the spermatogonia and the spermatocytes. An intensely positive reaction in monosome is evident in the spermatids where as the rest of the nucleus is faintly positive as compared to spermatogonia and spermatocytes. The heads of the mature spermatozoa are intensely positive (Figs. 1 and 2).

15 days after the treatment the spermatogonia and the spermatocytes show an intensely Feulgen positive localised reaction in the form of a band in the middle region or on one side of the nucleus. The rest of the nucleus is negative. The spermatids show an intensely positive reaction through out the nucleus but for one pole (Figs. 3 and 4). After an interval of 24 days a few spermatogonia and spermatocytes are present. Majority of the spermatids show an intensely positive reaction throughout the nucleus. Some of the spermatids, however, are faintly positive (Fig. 5).

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² Apholate is supplied by Olin Research Centre, New Havens, U.S.A.

30 days after the treatment the spermatids do show a positive reaction throughout the nucleus but the intensity of the reaction has been found to be diminished. A few of them are totally negative. The mature spermatozoa remain unaffected throughout the period (Fig. 6).

Figs. 1-6. Photomicrographs of control and treated testes of *P. pictus*. 1, spermatogonia and spermatocytes. Control. Feulgen. $\times 320$. 2, spermatids and sperms. Control. Feulgen. $\times 320$. 3, spermatocytes 15 days after treatment. Feulgen. $\times 480$. 4, spermatids 15 days after treatment. Feulgen. $\times 320$. 5, spermatids 24 days after treatment. Feulgen. $\times 480$. 6, spermatids and sperms 30 days after treatment. Feulgen. $\times 480$.

The increase in the intensity and band like localization after 15 days of treatment may be due to the clumping of the chromatin material. Since the histological picture reveals a pycnotic condition of the nucleus (Saxena and Vikram Aditya 1968, in press) the increased intensity of the reaction may be due to the same condition. The diminished reaction for DNA at later stages (24 and 30 days) may be due to the fragmentation and diffusion of the chromatin material as a result of the effect of apholate and it can be the reason for the reaction coming in the whole of the nucleus. The negative

reaction in few of the spermatids may be due to the complete destruction of the DNA.

Since the Feulgen reaction is specific for DNA, the above results show that apholate can drastically effect the DNA content of the germ cells in the male grasshopper.

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