retained after the reaction of 9 mol of thiol groups/mol of enzyme with 5,5'-dithiobis-(2-nitrobenzoic acid). With *p*-mercuribenzoate there was a fall of 40% in activity at this extent of reaction.

Titration of the more slowly reacting thiol groups (up to 14–18 mol of thiol groups/mol of enzyme) with *p*-mercuribenzoate or 5,5'-dithiobis-(2-nitrobenzoic acid) led to complete inhibition. Under the conditions used 1 mM-[¹⁴C]N-ethylmaleimide failed to react with this class of thiol groups. On reduction with 2-mercaptoethanol the activity of *p*-mercuribenzoate-treated RNA polymerase was completely restored, but with the 5,5'-dithiobis-(2-nitrobenzoic acid)-treated enzyme recovery depended on the groups/mol of enzyme that had reacted.

The observed loss of activity during the reaction of the slowly reacting thiol groups was interpreted as being due to a change in the tertiary structure, which might be entailed by the reaction of *p*-mercuribenzoate or 5,5'-dithiobis-(2-nitrobenzoic acid) with partially buried groups. This hypothesis was tested by titrating a complex of RNA polymerase and 8-anilino-naphthalene-1-sulphonate with *p*-mercuribenzoate, the fluorescence of the bound chromophore being used as a probe of conformation (Dodd & Radda, 1969). As *p*-mercuribenzoate was added a 20% increase in the fluorescence intensity coincided with loss of enzymic activity, and both recovered their initial values on reversal of the reaction with 2-mercaptoethanol.

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Different Ribonucleic Acid Polymerase Activities in Larval and Adult Blowflies

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RNA synthesis has been reported in nuclei isolated from larval blowfly tissues (Congote *et al.*, 1969; Shaaya & Sekeris, 1971), but RNA polymerase activities in nuclei from adult blowflies were not examined. The finding that the exotoxin from *Bacillus thuringiensis*, a compound believed to act by inhibiting RNA polymerase (Sebesta & Horska, 1968), was 100-fold less toxic to adult insects than to

larval forms (Bond *et al.*, 1971) prompted us to investigate whether the altered toxicity reflected a change in the RNA polymerases at metamorphosis.

Nuclei isolated from fat-bodies of larval blowflies (*Sarcophaga bullata*) exhibited RNA polymerase activities similar to those of mammalian nuclei. There was an activity manifest at low ionic strength that was preferentially stimulated by Mg²⁺ and largely resistant to the inhibitor α -amanitin; another activity, mainly sensitive to α -amanitin and with a preferential requirement for Mn²⁺, was manifest when the ionic strength was raised (Novello & Stirpe, 1969). Both activities were inhibited by exotoxin. The α -amanitin-resistant enzyme was the more sensitive to the compound, 0.8 μ M-exotoxin inhibiting activity by 50%. The α -amanitin-sensitive enzyme was 50% inhibited by 20 μ M-exotoxin.

Nuclei were prepared from total tissue homogenates of adult *Sarcophaga*. RNA polymerase activities were present but differed from those of larval nuclei. No activity was observed at low ionic strength in the presence of rifamycin and α -amanitin, conditions that did not inhibit the larval low-ionic-strength enzyme. Activity was stimulated at high ionic strength in the presence of Mn²⁺ or Mg²⁺. Mg²⁺-dependent activity was both rifamycin- and α -amanitin-sensitive, whereas Mn²⁺-dependent activity was resistant to rifamycin and less sensitive to α -amanitin. Both activities demonstrated different responses to exotoxin from those observed with the larval enzymes, requiring 5 μ M-exotoxin with Mg²⁺ or 200 μ M-exotoxin with Mn²⁺ for 50% inhibition of activity. The physiological significance of the adult high-ionic-strength RNA polymerase activities remains unknown.

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The Use of [5-³H]Uridine for Pulse-Labeling Ribonucleic Acid of Mouse Ehrlich Ascites-Tumour Cells

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To achieve pulse-labelling of RNA, a necessary condition for investigating the kinetics of RNA synthesis and migration, it is advantageous to use a