BIOCHEMICAL STUDIES ON THE ACCESSORY

REPRODUCTIVE GLAND SECRETIONS OF THE

MALE DESERT LOCUST, Schistocerca gregaria Forskäl

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SUMMARY

The two initial objectives of this research project were

(1) to obtain a detailed chemical description of secretions from

the accessory reproductive glands (ARGs) of the male desert locust,

Schistocerca gregaria Forskal, and (2) to identify the chemical

nature of the spermatophore and seminal fluids in order to obtain

a better understanding of the roles of individual ARG secretions

in their formation.

The secretions of the 16 pairs of ARGs were subjected to disc gel electrophoresis and the gels were stained for proteins, glycoproteins and lipoproteins. Although a number of common protein bands were found in the secretions of some glands, the electrophoresed, secretions of each gland showed distinct multiple protein and in some cases, multiple glycoprotein patterns. The number of protein fractions ranged from 10 in the seminal vesicle secretions to 26 in those from the homogenous glands. On the basis of histochemical and ultrastructural studies, Odhiambo (1969a, 1969b) indicated that the secretions of glands 3 and 5 were similar and were the only secretions containing lipoproteins. In this study, however, the electrophoresed secretions of these two glands showed distinct protein and glycoprotein patterns and

lipoproteins could not be detected with Sudan black B staining.

Odhiambo (1969a) also reported that secretions of gland 2 and the homogenous glands were rich in mucopolysaccharides, but after electrophoresis the secretions from these glands produced single, narrow and slightly stained glycoprotein bands.

Immunochemical studies on the secretions of the various ARGs confirmed some of the conclusions drawn from the results of electrophoretic investigations. The secretions of glands 1,11 and 12 and the haemolymph contained one common antigen. Another common antigen was found in the secretions of glands 3,4,5 and 6. On the basis of similar electrophoretic mobilities, there appear to be more common proteins in the various gland secretions than were demonstrated by the immunochemical experiments. Some of these proteins may have been present in concentrations which were too low for antibody production or they may not have been antigenic in the guinea-pig immune system.

Eighteen free amino acids were detected in methanolic extracts of the various ARGs, the paragonadal fat body and the haemolymph by two dimensional ascending paper chromatography. Of these, α -alanine, methionine, phenylalanine, serine and aspartic acid were of quantitative importance. In descending order the highest concentrations of α -alanine were present in the secretions

of glands 3,5,6,1 and 2.

Trehalose and/or inositol, an amino sugar and an unidentified compound were demonstrated by descending paper chromatography in the secretions of all ARGs. Glucose was demonstrated in the secretions of gland 12, while fructose was present in the secretions of all other ARGs. The secretions of glands 3,4,11 and the seminal vesicle also contained sucrose. Spot tests for purines and pyrimidines and the Sakaguchi test on concentrated methanolic extracts of the unidentified compound indicated that it is probably a guanine derivative.

The neutral and phospholipid composition of the various ARG secretions, as revealed by thin-layer chromatography of chloroform-methanol (2:1 v/v) extracts were generally similar, but the secretions of gland 4 were distinguished by the absence of all neutral lipids. The phospholipid compounds of ethanolamine, serine, inositol and choline were also demonstrated in chromatographed secretions of the various ARGs. The phospholipid composition of secretions from glands 5,6,12 and the homogenous glands appeared to be identical.

Secretions of the various ARGs were tested for the presence of 8 enzymes. The levels of alkaline phosphatases were high in

secretions from glands 1-6 as compared to levels in secretions from glands 11,12, the homogenous glands and the seminal vesicle. Low acid phosphatase activity was detected in the electrophoresed secretions of glands 1 and 3-6. The secretions of all ARGs could be differentiated on the basis of electrophoretic patterns for alkaline phosphatases, acid phosphatases, malic dehydrogenase, α -glycerophosphate dehydrogenase and non-specific esterases. Malic enzyme, xanthine dehydrogenase and iso-citrate dehydrogenase activities were not detected in any of the ARG secretions.

Protein and glycoprotein patterns were determined for the electrophoresed washings of unevacuated spermatophores and supernatents of homogenized evacuated spermatophores. The distinct electrophoretic pattern of unevacuated spermatophores contained 20 protein bands, 2 of which were glycoproteins. Two precipitin arcs were obtained when guinea-pig anti-serum to homogenous glands was cross-reacted with the washings of unevacuated spermatophores, but no reactions were obtained when guinea-pig anti-sera to various ARG secretions were reacted against the supernatents of evacuated spermatophores. The electrophoretic pattern of evacuated spermatophores showed 14 protein bands, 4 of which were glycoproteins. Protein patterns of evacuated spermatophores appear to represent the polymerization products of ARG secretions which are used in spermatophore construction, while protein patterns from

evacuated spermatophore washings may come primarily from accessory seminal fluids which also appear to be polymerization products

The third objective of this research project was to determine whether the injection of antibodies raised against ARG secretions could be used to disrupt the normal growth and/or function of the ARG complex in sexually maturing adults of the male desert locust. When the injection of anti-serum against ARG secretions began as early as the third day of adult development a marked inhibition of ARG growth was accompanied by suppression of secondary sexual characteristics (yellow body colouration) and normal mating behaviour. Inhibition of ARG growth, while significant, was less pronounced when anti-serum injections began on the seventh day of adult development, but on day 19 the injected males did not exhibit normal mating behaviour when placed with virgin females. When single anti-serum injections were administered on day 10 of adult development the males had obtained their yellow body colouration by day 19 and copulated normally with virgin females, although their ARGs were only equivalent in size to those of 13 day old males. Since certain antigens were common to both the haemolymph and accessory gland secretions it is possible that ARG growth is slowed when these antigens are precipitated in the haemolymph prior to their transport to the ARGs. Another mechanism of inhibition might involve

the penetration of antibodies into the ARGs where they might precipitate gland antigens. These data indicate the great potential of immunological approaches to studies on arthropod reproductive processes.