Yolk Proteins in the Turnip Sawfly, *Athalia rosae ruficornis* Jakovlev (Tenthredinidae, Hymenoptera)

Masatsugu HATAKEYAMA[1)], Masami SAWA[2)] and Kugao OISHI[1,2)]

[1)]*Department of Biology, Faculty of Science, Kobe University, Nada, Kobe, Hyogo 657, Japan*
[2)]*Division of Science of Biological Resources, Graduate School of Science and Technology, Kobe University*

Sex determination in the turnip sawfly, *Athalia rosae*, is apparently regulated by the sigle-locus multiple-allele system: homozygotes and hemizygotes develop to males and heterozygotes to females (Naito and Suzuki, 1985). Diploid males can be obtained easily by repeated brother-sister matings. It should be interesting to examine the pattern of sex-specific expression of genes such as those for yolk proteins. If the sex determination gene is not the only regulatory element, there may be some experimentally detectable differences between diploid and haploid males as to the regulation of sex-specific gene expression. We report here the results of our preliminary examinations on the yolk proteins. A brief description of ovarian development has been given previously (Sawa *et al.*, 1987).

Yolk proteins (vitellins) and their precursors (vitellogenins) have been studied in several insect species, especially in higher Diptera (Bianchi, 1985 ; Bownes, 1986). Vitellogenins are synthesized in the fat body of females, secreted into hemolymph, and then taken up by the growing oocyte. Vitellogenins are also synthesized in the follicle cell and transported to the oocyte.

We first used SDS polyacrylamide gel electrophoresis for the analysis of the hemolymph proteins and the proteins extracted from mature unfertilized eggs, and from embryos of various ages (Fig. 1). Comparison of female and male hemolymph proteins revealed a female-specific band(↗)among abundantly present proteins. The most prominent band(↗) in both unfertilized eggs and in embryos had the same apparent molecular weight as the female-specific band seen in the hemolymph. There was another prominent band(↘), although much less in quantity, in eggs and embryos. The quantity of these two most prominent proteins decreased with the embryo age. We conclude that these two bands in eggs and embryos represent major vitellins and may also conclude that the female-specific hemolymph protein is the precursor protein (vitellogenin) for the most abundant vitellin protein.

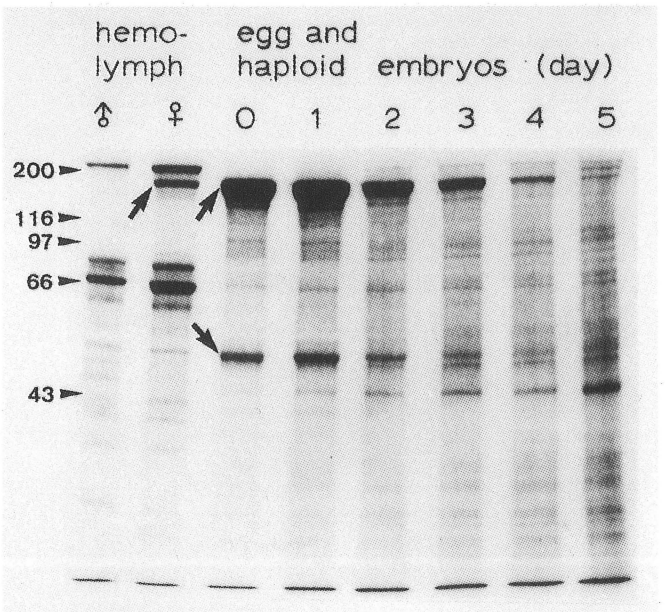

Fig. 1 SDS-PAGE of hemolymph proteins and of proteins extracted from unfertilized eggs and from embryos of various ages. Arrowheads along the left side indicate approximate positions of molecular weight (in kd) markers. Arrows indicate a female-specific hemolymph protein band (↗), and most prominent (↗) and second-most prominent (↘) egg protein bands.

Developing ovaries become distinguishable from testes in gross morphology in the third instar larvae. Individual ovarioles can be recognized clearly in sixth (last) instar larvae. The overall ovarian morphology becomes much the same as that in adults by the first day of pupal development (PCF5: the fifth day of post-cocoon formation), although even the most developed oocyte is still quite immature and is smaller than the nurse cell cluster connected to the oocyte. The oocyte is smaller than the nurse cell cluster until PCF 8, becoming about the same size on PCF 9 and larger on PCF 10 when the pupa ecloses. As shown in Table 1, the female hemolymph begins to show the presence of the female-specific protein (vitellogenin) only on PCF 9. Clearly the secretion of the protein from (and probably the synthesis in) the fat body is well regulated and corresponds to the development of oocytes.

Table 1 Appearance of female-specific protein in hemolymph.

Femmale-specific protein	Number examined			
	PCF 8	PCF 9 early	PCF 9 late	PCF 10
Present	0	0	14	8
Absent	10	10	6	0

We have found that immature ovaries dissected from a female can easily be transplanted into the abdomen of another female. In the hope that one could use inter-specific transplantation to obtain information on species-specific regulatory mechanisms in vitellogenin synthesis and vitellin deposition, we examined hemolymph proteins and the extracts from unfertilized eggs of three closely-related species, *A. Iugens infumata* Marlatt, *A. japonica* Klug, and *A. Kashmirensis* Benson. To our disappointment, no significant differences were detected in the electrophoretic patterns.

References

Bianchi, de A. G. (1985) *Insect Biochem.*, **15**, 77-84.
Bownes, M. (1986) *Annu. Rev. Entomol.*, **31**, 507-531.
Naito, T. and H. Suzuki (1985) *Jpn. J. Genet.*, **60**, 646.
Sawa, M., M. Hatakeyama and K. Oishi (1987) *Jpn. J. Genet.*, **62**, 550.