identification of the *X*-chromosome is readily apparent when chromosome pair 5 (underlined) in Figure b is examined. A large segment of constitutive heterochromatin is present near the centromere in one homologue effectively masking the secondary constriction in that chromosome in the conventional karyotype. The constriction is evident in both homologues in the G-banded karyotype (Figure a) if the chromosomes are not over contracted.

Further examination of Figure b reveals a considerable amount of constitutive heterochromatin in the karyotype of *Scalopus aquaticus*. The short arms of 4 pairs of autosomes are comprised almost entirely of constitutive heterochromatin. At present, we are unable to account for the apparent pattern of inheritence of the heteromorphic C-band regions wherein all individuals possess the heteromorphic pair of autosomes.

The secondary constriction region in this species corresponds to the nucleolar organizer region (NOR) as indicated by the AgAS techniques. We have not attempted in situ hybridization studies but in other species, the AgAS technique stains the sites of known 18s and 28s ribosomal cistrons. No difference in the size of the NOR region between homologues is indicated in our material.

Acid Phosphatases in *Drosophila auraria*

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Summary. A study was carried out dealing with the acid phosphatases, in larvae, pupae and adults of *Drosophila auraria*. The most interesting finding is that the acid phosphatases are observed in 3 different phenotypes appearing as groups of 3 bands of different mobilities in the homozygotes and giving a 5 band phenotype in the heterozygote. A hypothesis is discussed in an attempt to explain the phenotypes observed and their variation during the development of the species.

In the literature of the last decade, one finds many references to enzyme polymorphisms of various *Drosophila* and other species. These studies are quite often extended to cover developmental variations of the various enzymatic patterns as well as species comparisons[2]. Since Kastritsis and Grossfield[3] have described the existence of 2 Balbiani rings in one of the chromosomes of *D. auraria*, our laboratory has undertaken a study of various aspects of the development and the genetics of this species.

Materials and methods. The laboratory stocks of *D. auraria* used in this investigation were obtained from Dr. J. Grossfield of New York University. As a reference we used stocks of *D. melanogaster* and *D. simulans* kept in our laboratory. Larvae of 1st, 2nd or 3rd instar, prepupae and pupae (early and late), as well as adults (♂ and ♀) of 3 h, 1, 2, 3, 4, 6, 8, 10 and 22 days of age were used to perform horizontal starch gel electrophoresis. For the 1st and 2nd larval instars, 4 individuals per sample were homogenized on glass microscope slides, as described by Beckman and Johnson[4]. In subsequent experiments with animals of the remaining ages, single individuals were utilized. The electrophoresis was performed using a discontinuous buffer system[5] with a voltage gradient of 8–10 V per centimeter. Assays were performed by using the techniques of MacIntyre[6].

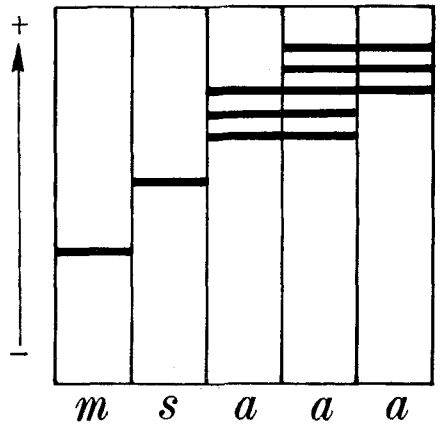

Fig. 1. Schematic representation of the relative positions of acid phosphatases in *D. melanogaster* (m. genotype Acph–$1^{1.00}$ / Acph–$1^{1.00}$), *D. simulans* (s, genotype Acph–$1^{1.51}$ / Acph–$1^{1.51}$), and the 3 *D. auraria* phenotypes (a, A/A, A/B and B/B). The lines represent only the position of each band and not their developmental variation.

In order to facilitate species comparisons, the allozyme controlled by the most common allele of *D. melanogaster* was assigned a mobility of 1.00; all other allozymes of the 3 species used were compared to that 'standard' and named accordingly. To make comparisons with other reports[6] possible, it should be mentioned that Acph-1^F of *D. melanogaster* is designated here as Acph-$1^{1.00}$ while the relative mobility of the Acph-1^F of *D. simulans* in reference to Acph-1^F of *D. melanogaster* is Acph-$1^{1.51}$.

Results and discussion. In the zymograms of *D. auraria*, 3 different phenotypes of Acph have been found, i.e. phenotype A (slow), AB, and B (fast) respectively, as shown in Figures 1 and 2. Phenotype A consists of 3 bands (1, 2, 3 in the direction of migration), phenotype AB of 5 bands (1, 2, 3, 4, 5) and phenotype B of 3 bands (3, 4, 5). The relative mobilities of these bands in reference to Acph-$1^{1.00}$ of *D. melanogaster* are Acph-$1^{1.90}$, Acph-$1^{2.06}$, Acph-$1^{2.22}$, Acph-$1^{2.38}$, and Acph-$1^{2.54}$.

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The developmental study has shown that 1st instar larvae give a negative Acph reaction, while 2nd instar larvae rarely give a positive reaction. This situation is identical in all 3 phenotypes. On the other hand, positive reaction (for all 3 phenotypes) was observed in all other stages studied. However, variations in the staining intensity of the various bands were found, and these variations appeared to be stage-specific. Thus, the A phenotype showed that band 1 of 3rd instar larvae (early and late) was more intense than bands 2 and 3 which stained identically. The situation in prepupae and pupae was the same but the difference between band 1, and bands 2 and 3 was less evident. In virgin individuals (3 h after emergence) bands 2 and 3 appeared more intense and band 1 continued staining as intensely as in pupae. Thereafter, the intensities of the 3 bands could be classified as $3 > 2 > 1$ with the intensity of band 1 declining as compared to earlier stages, and the intensity of band 3 increasing; band 2 remained about the same. The B phenotype showed that band 3 of 3rd instar larvae (early and late) immediately appeared more intense and remained more intense (gradually increasing) as the individuals became older. On the other hand, band 4 increased in intensity in 3rd instar larvae, prepupae and pupae, but remained constant thereafter. Finally, band 5 appeared at first in late 3rd instar larvae, and increased through the pupal and adult stage. Overall, the order of intensity was $3 > 4 > 5$.

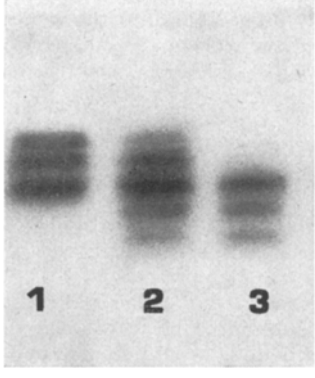

Fig. 2. Photograph of a zymogram of the 3 different acid phosphatase phenotypes of *D. auraria* (1 = phenotype B; 2 = phenotype AB; 3 = phenotype A). The zymogram was obtained from adult individuals.

Phenotype AB is produced from the cross of phenotypes A and B (Figures 1 and 2). The intensities of the 5 bands produced followed the relationship $3 > 4 > 2 > 5 \geq 1$ in adult individuals.

In order to determine the mode of inheritance of the genes controlling the Acph phenotypes, all possible crosses were performed and the results showed that the 3 phenotypes appear to be controlled by simple Mendelian inheritance. However, the question that needs to be answered is: what controls the appearance of the various Acph bands in each phenotype?

Various hypotheses could be proposed in attempting to explain the data presented in this report. The results of the crosses indicate that the acid phosphatases of *D. auraria* are controlled by 2 alleles of a single autosomal gene. This, however, is not sufficient to explain the existence of the 3 and the 5 bands that appear in the 2 homozygotes and the heterozygote respectively. This can be explained only if we were to assume that the enzymes are polymers consisting of 2 or 4 subunits and that they are controlled by 2 pairs of closely linked autosomal genes, Acph-A and Acph-B. Of these, gene Acph-A is represented by alleles Acph-A^1 and Acph-A^2 controlling enzymes with the mobility of bands 1 and 3 respectively, and gene Acph-B is represented by alleles Acph-B^1, and Acph-B^2 controlling enzymes with the mobility of the bands 3 and 5 respectively. The hypothesis requires that the one gene (Acph-A or Acph-B) is the product of a duplication of the other (as has already been described for amylases of *Drosophila*[7] and aspartate aminotransferase in fishes[8]) and that mutations created the alleles Acph-A^1 and Acph-B^2. Thus, genes Acph-A^2 and Acph-B^1 have the same origin and structure, and this explains the similar behavior of band 3 that they control in all phenotypes and in the various developmental stages.

According to this hypothesis, phenotypes A, B and AB are derived from genotypes $\frac{\text{Acph-A}^1\ \text{Acph-B}^1}{\text{Acph-A}^1\ \text{Acph-B}^1}$, $\frac{\text{Acph-A}^2\ \text{Acph-B}^2}{\text{Acph-A}^2\ \text{Acph-B}^2}$, and $\frac{\text{Acph-A}^1\ \text{Acph-B}^1}{\text{Acph-A}^2\ \text{Acph-B}^2}$ respectively.

The different activity of the Acph bands in the various developmental stages could be explained by the existence of regulatory mechanisms[7] determining the amount of enzyme to be produced by the action of each gene.

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Unstirred Layer Thickness in Perfused Rat Jejunum in vivo

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Summary. The effective unstirred layer thickness in the rat jejunum perfused in vivo amounts to at least 530 μm. This value has been estimated from the absorption increase of L-phenylalanine due to better mixing of the luminal solution by air bubbles.

The unstirred layer adjacent to the mucosal surface of the intestine may be the source of biased results: reduced permeability coefficients[1-3], raised K_m-values[4-7], shift of the pH-absorption curves to the right or the left[8-11]. When the effective thickness of the unstirred layer is known, corrections can be applied[1,2,5]. The unstirred layer thickness in the intestine can be determined in vitro by measuring the change of the potential difference after raising or lowering the osmolarity in the bulk phase[1,3,6,12,13]. In vitro the unstirred layer can be reduced by vigorous stirring[13,14] or shaking[7]. In vivo, these methods cannot be applied. By adapting the in