Drosophila Enzyme-Genetics: A Table

Drosophila melanogaster surely ranks first among the higher organisms as a tool for genetic studies, although it has not matched the bacterial and viral systems in elucidating major genetical concepts. Its contributions at the biochemical level date from the classic studies of BEADLE and EPHRUSSI[1] on *Drosophila* eye pigments.

It is surprising that relatively little enzymological work followed those exciting experiments. Only in recent years has a more active interest in *Drosophila* enzymology become apparent, perhaps because it is becoming increasingly clear that we cannot explain all genetic phenomena in higher organisms in terms of models which are sufficient for bacteria[2,3]. The burden of bridging this phylogenetic gap now rests on the *Drosophila* geneticists, among others.

In the past 5 years, the number of mapped *Drosophila* enzymes has increased from 7[4] to 21 (Table I). Furthermore, a number of enzymes have been partially or totally purified and several enzymes have been localized histochemically (Table II). In addition, several complex systems are known in which non-structural modifier genes are active, e.g. tyrosinase[5,6] and the xanthine dehydrogenase, aldehyde oxidase, pyridoxal oxidase triplet[7]. It is anticipated that further study of the complex enzyme systems, modifier genes and 'biochemical mutations'[8] will begin to fill the phylogenetic gap.

Another approach which may benefit from enzymological work in *Drosophila* is that of correlating gene activity with the phenotypic expression of specific end products. BEERMANN[9], BERENDES[10], GROSSBACH[11] and SLYZIN-

Table I

Name of enzyme	Gene symbol[a]	Genetic locus[a]	Fold purification	Molecular weight
Acid phosphatase	Acph-1	3–101.4[28]		
Alcohol dehydrogenase	Adh	2– 50.1[29]	145[54]	44,000[54]
Aldolase			90[55]	
Aldehyde oxidase	Aldox	3– 56.6[30]	350[30]	250,000[30]
Alkaline phosphatase	Aph	3– 46.3[31–33]		
		3– 47.3[34]		
α-Amylase	Amy	2– 77.7[16,35–37]		
Esterase-6	Est-6	3– 36.8[38]		
Esterase-C	Est-C	3– 49[39]		
Glucose-6-phosphate dehydrogenase	Zw	1– 63[20]	242[56]	317,000[56]
α-Glycerophosphate dehydrogenase	α-Gpdh	2– 20.5[40]	55[57]	63,000[57]
	Gdh	2– 17.8[41]		
Isocitrate dehydrogenase	Idh-NADP	3– 27.1[21–23]	105[21]	81,000[21]
Lactate dehydrogenase			130[58]	149,000[58]
Leucine aminopeptidase-A	Lap-A	3– 98±[42]		
Leucine aminopeptidase-D	Lap-D	3– 98.3[42,43]		
Malate dehydrogenase	Mdh-1	2– 35.3[24]	120,180[25 c]	52,000[25]
	s-Mdh	2– 40±[25]		
	Mdh-2	2– 41.2[26]		
Octanol dehydrogenase	Odh	3– 49.2±[44]		
Phenol oxidase	lz+[b]	1– 27.7[45,46]	Partial[59,60]	Particulate[59,60]
Phosphoglucomutase	Pgm	3– 43.4[47]		
6-Phosphogluconate dehydrogenase	Pgd	1– 0.9[20]	Partial[61]	79,000±[61]
Pyridoxal oxidase	lpo	3– 57±[48]		225,000[64]
Tryptophan pyrrolase	v+	1– 33.0[18,49,50]	16[62]	
Tyrosinase	tyr-1	2– 52.4[5,6]		
	Tyr-2	2– 57[5,6]		
Xanthine dehydrogenase	ry+	3– 52.35[51–53]	529[63]	250,000[64,65]

[a] Several enzymes have been assigned gene symbols and genetic loci by more than one investigator. Differences in genetic loci are probably attributable to the use of different marker genes. [b] The lozenge gene may be the structural gene for a polypeptide common to the A components of the phenol oxidase complex[46]. [c] The two MDHs purified by ANDERSON[25] are the soluble (120 fold) and mitochondrial (180 fold) forms. Both have a molecular weight of 52,000.

Table II. Tissue distribution of enzymes in third instar larvae

Enzyme	Fat body	Intestine	Malpighian tubules	Salivary gland
Aldehyde oxidase[30]	±	+	+	—[a]
Alcohol dehydrogenase[66]	+	+	+	—
Alkaline phosphatase[67]	—	+	—	—
α-Amylase[68]	+	+	±	+
Deoxyribonuclease[69]	+	+	—	—
α-Glycerophosphate dehydrogenase[70]	+	±	N.D.	+
Isocitrate dehydrogenase[21]	+	+	±	±
Lactate dehydrogenase[70]	—	—	N.D.	±
Xanthine dehydrogenase[71–73]	+	+	+	N.D.

±, very low level of enzyme; +, enzyme present; —, enzyme absent; N.D., no data. [a] Aldox is present in the stalk, but not the body of the salivaries.

SKY[12] have already made significant progress in this direction by correlating the presence or absence of specific salivary gland products with the presence or absence of specific puffs on the salivary polytene chromosomes of *Chironomus* (*Camptochironomus*) and *Drosophila*.

This same technique may be further exploited with *Drosophila melanogaster* using one or more of the many enzymes of known genetic loci (Table I). This information, together with the known puffing patterns for salivary gland chromosomes[13–15], should allow a correlation between specific puffs and the activity of the mapped enzymes. The loci for α-amylase[16], alcohol dehydrogenase[17], tryptophan pyrrolase[18] and xanthine dehydrogenase[19] have already been localized to fairly restricted regions on the cytogenetic map. These 4 enzymes would then be logical first choices for such a study. In addition, since it is known that fat body, malpighian tubules and gut as well as salivary glands possess giant chromosomes, enzymes can be used which differ in their organ distribution (Table II).

At a more general level of organization it would be interesting to know if the loci of metabolically related *Drosophila* enzymes are clustered or distributed at random throughout the genome. Some relevant data are already available in Table I: there is loose linkage between the genes for glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase[20]; the soluble forms of isocitrate dehydrogenase and malate dehydrogenase are coded for by genes on separate chromosomes[21–26]. Of immediate interest, then, would be the genetic loci for other members of the glycolytic pathway as well as those of the soluble citric acid cycle.

It is worth noting that HUTTON and RODERICK[27] have shown in mice that the loci of several enzymes of the glycolytic pathway are on separate chromosomes. ELSTON and GLASSMAN[3] have shown, however, that there is a tendency for functionally related (according to the body part affected) genes to be found on a particular chromosome, but that intra-chromosomal clustering may be accounted for by the known clustering of all genes within chromosomes. While the present data for *Drosophila* enzymes are too scanty to make any generalizations, it is expected that in the near future more data will be available.

Since this manuscript was submitted, the following additional information has become available: 1. The molecular weight of alcohol dehydrogenase[54] by a somewhat

[1] G. W. BEADLE and B. EPHRUSSI, Genetics *22*, 76 (1937).
[2] G. M. TOMKINS and B. N. AMES, Natn. Cancer Inst. Monogr. *27*, 221 (1967).
[3] R. C. ELSTON and E. GLASSMAN, Genet. Res., Camb. *9*, 141 (1967).
[4] M. S. FUCHS, Drosoph. Inf. Serv. *40*, 105 (1965).
[5] H. W. LEWIS and H. S. LEWIS, Proc. natn. Acad. Sci. *47*, 78 (1961).
[6] H. W. LEWIS and H. S. LEWIS, Ann. N.Y. Acad. Sci. *100*, 827 (1963).
[7] E. GLASSMAN, J. COLLINS, E. J. DUKE, T. SHINODA and D. M. JACKSON, Genetics *56*, 561 (Abstr.) (1967).
[8] E. NOVITSKI, Drosoph. Inf. Serv. *37*, 51 (1963).
[9] W. BEERMANN, Chromosoma *12*, 1 (1961).
[10] H. D. BERENDES, Chromosoma *17*, 35 (1965).
[11] U. GROSSBACH, Chromosoma *28*, 136 (1969).
[12] B. M. SLYZINSKY, *Genetics Today*, Proc. XI. Int. Congr. Genet., The Hague *1*, 108 (1963).
[13] M. ASHBURNER, Chromosoma *21*, 398 (1967).
[14] M. ASHBURNER, Chromosoma *27*, 47 (1969).
[15] M. ASHBURNER, Chromosoma *27*, 64 (1969).
[16] W. W. DOANE, J. exp. Zool. *171*, 321 (1969).
[17] E. H. GRELL, in *Genetic Variations of Drosophila melanogaster* (Eds. D. L. LINDSLEY and E. H. GRELL; Carnegie Institution of Washington Publication No. 627, 1968), p. 11.
[18] M. M. GREEN, Proc. natn. Acad. Sci., USA *40*, 92 (1954).
[19] E. H. GRELL, Z. Vererbungsl. *93*, 371 (1962).
[20] W. J. YOUNG, J. Hered. *57*, 58 (1966).
[21] D. J. FOX, Ph.D. Thesis, The Johns Hopkins University (1969).
[22] D. J. FOX, Drosoph. Inf. Serv. *45*, 35 (1970).
[23] D. J. FOX, Biochem. Genet., in print.
[24] S. J. O'BRIEN, Drosoph. Inf. Serv. *44*, 113 (1969).
[25] M. ANDERSON, Ph.D. Thesis, The Johns Hopkins University (1969).
[26] E. H. GRELL, Drosoph. Inf. Serv. *44*, 47 (1969).
[27] J. J. HUTTON and T. H. RODERICK, Biochem. Genet. *4*, 339 (1970).
[28] R. J. MACINTYRE, Genetics *53*, 461 (1966).
[29] E. H. GRELL, K. B. JACOBSON and J. B. MURPHY, Science *149*, 80 (1965).
[30] W. J. DICKINSON, Ph.D. Thesis, The Johns Hopkins University (1969).
[31] L. BECKMAN and F. M. JOHNSON, Genetics *49*, 829 (1964).
[32] F. M. JOHNSON, Science *152*, 361 (1966).
[33] R. J. MACINTYRE, Drosoph. Inf. Serv. *41*, 62 (1966).
[34] B. B. WALLIS, Biochem. Genet. *2*, 141 (1968).
[35] W. W. DOANE, Drosoph. Inf. Serv. *38*, 32 (1963).
[36] H. KIKKAWA, Jap. J. Genet. *39*, 401 (1964).
[37] E. BAHN, Hereditas *58*, 1 (1967).
[38] T. R. F. WRIGHT, Genetics *48*, 787 (1963).
[39] L. BECKMAN and F. M. JOHNSON, Hereditas *51*, 212 (1964).
[40] S. J. O'BRIEN and R. J. MACINTYRE, Drosoph. Inf. Serv. *43*, 60 (1968).
[41] E. H. GRELL, Science *158*, 1319 (1967).
[42] L. BECKMAN and F. M. JOHNSON, Hereditas *51*, 221 (1964).
[43] E. V. FALKE and R. J. MACINTYRE, Drosoph. Inf. Serv. *41*, 165 (1966).
[44] J. B. COURTRIGHT, R. B. IMBERSKI and H. URSPRUNG, Genetics *54*, 1251 (1966).
[45] *Genetic Variations of Drosophila melanogaster* (Eds. D. L. LINDSLEY and E. H. GRELL; Carnegie Institution of Washington Publication No. 627, 1968), p. 146.
[46] E. E. PEEPLES, A. GEISLER, C. J. WHITCRAFT and C. P. OLIVER, Biochem. Genet. *3*, 563 (1969).
[47] P. HIJORTH, Hereditas *64*, 146 (1970).
[48] J. F. COLLINS and E. GLASSMAN, Genetics *61*, 833 (1969).
[49] G. A. MARZLUF, Genetics *52*, 503 (1965).
[50] K. D. TARTOF, Genetics *62*, 781 (1969).
[51] E. GLASSMAN and P. SAVERANCE, J. Elisha Mitchell Sci. Soc. *79*, 139 (1963).
[52] A. CHOVNICK, A. SCHALET, R. P. KERNAGHAN and M. KRAUSS, Genetics *50*, 1245 (1964).
[53] T. T. T. YEN and E. GLASSMAN, Genetics *52*, 977 (1965).
[54] W. H. SOFER and H. URSPRUNG, J. biol. Chem. *243*, 3110 (1968).
[55] O. BRENNER-HOLZACH and F. LEUTHART, Helv. chim. Acta *51*, 1130 (1968).
[56] M. W. STEELE, W. J. YOUNG and B. CHILDS, Biochem. Genet. *2*, 159 (1968).
[57] M. C. RECHSTEINER, Ph.D. Thesis, The Johns Hopkins University (1969).
[58] M. C. RECHSTEINER, J. Insect Physiol. *16*, 957 (1970).
[59] H. K. MITCHELL, U. M. WEBER and G. SCHAAR, Genetics *57*, 357 (1967).
[60] H. K. MITCHELL and U. M. WEBER, Science *148*, 964 (1965).
[61] H. H. KAZAZIAN JR., Nature, Lond. *212*, 197 (1966).
[62] G. A. MARZLUF, Z. Vererbungsl. *97*, 10 (1965).
[63] S. D. PARZEN and A. S. FOX, Biochim. biophys. Acta *92*, 465 (1964).
[64] H. M. MOON, Ph.D. Thesis, University of N. Carolina (1967).
[65] E. GLASSMANN, T. SHINODA, H. M. MOON and J. D. KARAM, J. molec. Biol. *20*, 419 (1966).
[66] H. URSPRUNG, W. H. SOFER and N. BURROUGHS, Wilhelm Roux' Archiv EntwMech. Org. *164*, 201 (1970).
[67] H. SCHNEIDERMAN, W. J. YOUNG and B. CHILDS, Science *151*, 461 (1966).
[68] W. W. DOANE, Proc. XII Int. Congr. Entomol., London (1964), p. 233.
[69] J. B. BOYD, Biochim. biophys. Acta *171*, 103 (1969).
[70] M. C. RECHSTEINER, J. Insect Physiol. *16*, 1179 (1970).
[71] E. HADORN and H. URSPRUNG, Drosoph. Inf. Serv. *34*, 83 (1960).
[72] H. URSPRUNG and E. HADORN, Experientia *17*, 230 (1961).
[73] P. MUNZ, Z. Vererbungl. *95*, 195 (1964).

different procedure has been estimated to be 60,000 (K. B. JACOBSON and P. PFUDERER, J. biol. Chem. *245*, 3938, 1970). 2. The locus of another glycolytic enzyme, hexokinase, has been mapped to 2–79 ± (D. J. FOX, K. MADHAVAN, and H. URSPRUNG, unpublished). Note the proximity of this locus to that of α-Amylase (Table I).

Zusammenfassung. Durch Verwendung elektrophoretischer Enzym-Mutanten ist es möglich, bei *Drosophila melanogaster* die chromosomale Position der entsprechenden Strukturgene zu bestimmen. Über 20 Gene sind auf diese Weise lokalisiert worden. Untersuchung der Gewebe- und Stadienspezifität dieser Enzyme, im Verband mit zytogenetischer Analyse der Riesenchromosomen, verspricht wertvolle Einblicke in das Problem der Genregulation. In den vorliegenden Tabellen sind die bereits erzielten Ergebnisse zusammengestellt.

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Karyotypes of Bats of the Subfamily Carolliinae (Mammalia; Phyllostomatidae) and Their Evolutionary Implications[1]

The North American leaf-nosed bats of the family Phyllostomatidae are a most complex group of animals. Some members are adapted to feeding on other vertebrates, insects, fruit, nectar, blood (Desmodontinae, see FORMAN et al.[2]), and we have observed *Carollia* feeding on tender stems of plants. Even though several extreme morphological modifications are found and form the basis for the several subfamilies, the phylogenetic affinities of most groups are not easily understood. Karyotypic characteristics were used by BAKER[3] to hypothesize phylogenetic affinities and to delineate several lines of evolution within the family. One line of evolution involved the Carolliinae and part of the Glossophaginae. At that time karyotypic data for the Carolliinae were based on two species of *Carollia*.

In this present communication we describe the chromosomes of another species of *Carollia* and two species of the other genus (*Rhinophylla*) of the Carolliinae.

Methods and Materials. Specimens were collected from natural populations with the aid of mistnets. Chromosomal preparations were made at nearby field stations. A 2 hour in vivo culture of bone marrow was followed by treatment with a 1% sodium citrate solution. Fixation was by methanol, acetic acid (3:1). Mitotic slides were blaze dried and stained with Giemsa's blood stain. Testicular meiotic preparations were by the aceto-orcein squash technique. Voucher specimens are deposited in the collection of mammals, Department of Biology, Texas Tech University. To measure the relative size of the *X* to the autosomes, microphotographs were made and the length of the chromosomes were measured with a pair of dial calipers.

The total length of the *X* chromosome was devided by the total length of the haploid autosomal complement. The *X* chromosome is easily determined in species of *Carollia* because of its secondary constriction[4,5]. The *X* of *Rhinophylla* cannot be positively determined by comparing the karyotypes of males and females, but the determination of the general size of the largest heteromorphic element in males is reasonably accurate. Fundamental number is considered to be the number of arms of the autosomal complement.

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[2] G. L. FORMAN, R. J. BAKER and J. D. GERBER, Syst. Zool. *17*, 417 (1968).

[3] R. J. BAKER, SWest. Nat. *12*, 407 (1967).

[4] T. C. HSU, R. J. BAKER and T. UTAKOJI, Cytogenetics *7*, 27 (1968).

[5] Y. YONENAGA, O. FROTA-PESSOA and K. R. LEWIS, Caryologia *22*, 63 (1969).

Size of the *X* chromosome to the autosomal genome

Karyotype Catalogue Number and Species		Sex	Locality in Colombia	Size of *X* in relationship to haploid autosomal size expressed as a percentage			No. of cells measured
				Low	Mean	High	
Z75	*Carollia castanea*	♀	Villavicencio	15.64	16.44	17.10	6
Z137	*Carollia subrufa*	♀	Leticia	14.94	17.32	18.71	6
Z3	*Carollia perspicillata*	♀	Restrepo	16.17	17.70	18.31	8
Z146	*Carollia perspicillata*	♀	Leticia	15.44	16.51	19.61	5
Z62	*Carollia perspicillata*	♂	Leticia	16.82	18.25	19.92	8
Z124	*Rhinophylla fischerae*	♂	Leticia	6.04	6.94	8.31	7
Z125	*Rhinophylla pumilio*	♀	Leticia	5.36	6.34	7.86	8
Z192	*Rhinophylla pumilio*	♂	Leticia	5.02	5.39	5.65	4

See text for method of determination.