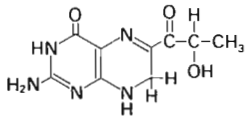
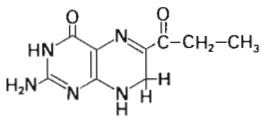
Takikawa, S. Kitasato University, Sagamihara, Japan. Eye pigments of D. melanogaster.

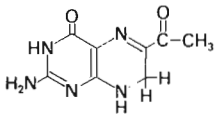
It is well known that wild type flies of D. melanogaster have red and yellow eye pigments, and the structures of the pigments are interesting from the viewpoints of genetics and pteridine metabolism. The mutant sepia of D. melanogaster contains only yellow pigments (Ziegler and Hadorn, 1958). One of them is sepiapterin, whose structure is 2-amino-4-hydroxy-6-lactyl-7,8-dihydropteridine (Nawa, 1960). Another pigment is isosepiapterin (Viscontini and Möhlmann, 1959), whose structure is 2-amino-4-hydroxy-6-propionyl-7,8-dihydropteridine (Forrest et al., 1959). In this communication, the isolation of the third yellow pigment from mutant sepia and the determination of its structure. We call the pigment "neosepiapterin", simply because this particular pigment accumulates in the sepia mutant. The structures of these yellow pteridines are shown in Figure 1.



Sepiapterin



Isosepiapterin



Neosepiapterin

The technique used for column chromatographic separations followed that of Tsusue and Akino (1965) and that of Fukushima and Akino (1968). In order to separate neosepiapterin from isosepiapterin, cellulose powder was used as an adsorbent and it was eluted with the solvent, n-butanol, ethanol, water (2:1:1 v/v). Flies were reared on standard yeast medium at 25°C and were harvested on the 1st, 5th and 9th days after eclosion. The contents of the pigments relative to the ages of the flies is shown in Table 1. Since the eye color of the sepia flies tends to become darker after eclosion, it has been supposed that the content of the pigments increases as the flies grow older. But these data show that the quantity of sepiapterin and neosepiapterin does not increase after 5 days eclosion.

Table 1. Quantity of yellow pteridines related to the ages after eclosion. Starting material was 50 g of sepia mutant of D. melanogaster respectively.

	Quantity (mg)		
Days after eclosion	Sepiapterin	Isosepiapterin	Neosepiapterin
1	19.8	0.36	0.27
5	34.4	1.33	0.36
9	34.4	1.87	0.37

On the other hand, isosepiapterin continues to increase.

The Rf values of the yellow pteridines are shown in Table 2. The molecular formula of neosepiapterin was determined to be $C_8H_9O_2N_5$ by mass spectrometry (molecular ion peak, m/e 207.0774). This pigment is thought to be C-6 substituted pterin because its alkaline potassium permanganate oxidation gives 2-amino-4-hydroxypteridine-6-carboxylic acid. The formation

Table 2. Rf values of yellow pteridines

	Solvents				
Pteridines	1	2	3	4	5
Sepiapterin	0.29	0.34	0.47	0.27	0.36
Isosepiapterin	0.21	0.44	0.52	0.46	0.53
Neosepiapterin	0.21	0.32	0.42	0.30	0.36

Solvents: 1. 3% ammonium chloride
2. isopropanol, 1% ammonia (2:1)
3. isopropanol, 2% ammonium acetate (1:1)
4. n-butanol, acetic acid, water (4:1:1)
5. n-propanol, ethylacetate, water (7:1:?)

of 2,4-dinitrophenyl-hydrazone shows carbonyl group at the sidechain. And the ultraviolet absorption spectrum of neosepiapterin resembles that of isosepiapterin. These data indicate that the structure of neosepiapterin is 2-amino-4-hydroxy-6-acetyl-7,8-dihydropteridine.

The distribution of neosepiapterin in sepia fly remains unknown, but it is supposed to be contained in the eye since sepiapterin and isosepiapterin are the eye pigments. Neosepiapterin was not detected when pterins were extracted from a large amount (1.2 kg) of wild type flies. Many derivatives of pteridine, which have three carbons at the sidechain on C-6 of the pteridine ring, have been isolated from natural sources. But those with two carbons have never been isolated before. Therefore, the occurrence of reduced 6-acetyl pterin suggests an unknown pathway of pteridine metabolism. Studies on the origin of a two carbon sidechain of the pigment is under investigation.

References: Forrest, H.S., C. Van Baalen and J. Myers 1959, Arch. Biochem. Biophys. 83: 508; Fukushima, T. and M. Akino 1968, Arch. Biochem. Biophys. 128:1; Nawa, S. 1960, Bull. Chem. Soc. Japan 33:1555; Tsusue, M. and M. Akino 1965, Zool. Mag. Tokyo 74:91; Viscontini, M. and E. Möhlmann 1959, Helv. Chim. Acta 42:836; Ziegler-Günder, I. and E. Hadorn 1958, Z. Vererbungslehre 89:235.

Bächli, G. University of Zürich, Switzerland. Drosophilidae of Kanha National Park, M.P., India.

Drosophilidae were collected by net sweeping over banana baits during the period of August 26 to September 18, 1972. The 8 collection sites are located in the center of the Kanha National Park, about 200 km northeast of Nagpur. The altitude is around 600 m above sea level. The species collected and the number of specimens are listed in Table 1. Drosophila (Scaptodrosophila) sp. and Hypselothyrea sp. are new species, while Leucophenga flavicosta, Lissocephala sabroskyi and Drosophila minima are reported for the first time from India. Domestic and cosmopolitan species were mostly absent. In the collection period (end of monsoon season), the ecological conditions of the center of the Park are therefore considered non-domestic.

Table 1. List of species collected, in order of frequency.

Species	Number of Specimens	Species	Number of Specimens
D. malerkotliana	10,643	** D. (Scaptodros.) sp.	6
D. jambulina	4,115	Leuc. albicincta	4
D. paratriangulata	1,349	* Leuc. flavicosta	4
D. albomicans	387	** Hypselothyrea sp.	2
D. takahashii	373	* Lissocephala sabroskyi	2
D. latifshahi	62	Leuc. guttiventris	1
D. kikkawai	26	D. silvalineata	1
D. chandraprabhiana	9	D. bipectinata	1
		* D. minima	1

* first record from India
** new species

Hardy, R.W., Le Ngoc Anh and Ng. H. Xuong University of California, San Diego, La Jolla, California. Three dimensional measurements of spermatid nuclei in Drosophila melanogaster from electron micrographs of serial cross sections.

We have developed a method for measuring the volume and morphology of spermatid nuclei. Testes are prepared for electron microscopy by the method of Tokuyasu, Peacock and Hardy (Z. Zellforsch. 124:479-506, 1972). Serial sections of an entire bundle of nuclei of spermatids in the early coiling stage are cut and the thickness of each is estimated from its interference color. Electron micrographs of selected sections are scanned with a digital densitometer connected to an IBM 1800 computer (Xuong, J. Physics E. 2:485, 1969). A program written in