Katoh, S. Josai Dental University, Sakado-machi, Saitama-ken, Japan. Two yellow eye pigments of D. melanogaster catalyzed by sepiapterin reductase.

Two yellow pigments have been isolated and identified from the eye of mutant sepia of D. melanogaster: sepiapterin (2-amino-4hydroxy-6-lactyl-7,8-dihydropteridine) and isosepiapterin (2-amino-4hydroxy-6-propionyl-7,8-dihydropteridine). Sepiapterin, the main yellow pigment in the eye, was found to be catalyzed to a non-color substance with the aid of NADPH by an extract of the fat body of adult Drosophila or by a homogenate of mammal liver of several species. This catalyzation of sepiapterin is, in fact, mediated by an enzyme named "sepiapterin reductase" as the following equation:

$$\text{sepiapterin} + \text{NADPH} + \text{H}^+ \rightleftharpoons \text{dihydrobiopterin} + \text{NADP}^+$$

Sepiapterin is reduced to dihydrobiopterin (2-amino-4-hydroxy-6-1',2'-dihydroxypropyonyl-7,8-dihydropteridine) by the enzyme, the C^6-substituted lactyl group of sepiapterin is converted to the 1',2'-dihydroxypropyl group[1].

Sepiapterin reductase has recently been purified 5000-fold from an extract of horse liver by protamine sulfate treatment, ammonium sulfate fractionation and column chromatography of hydroxylapatite and of DEAE-cellulose[2]. The pH optimum of the purified enzyme is 5.5 and Km values of sepiapterin and NADPH are 2.1×10^{-5} M and 1.4×10^{-5} M, respectively, at pH 6.4.

Isosepiapterin and xanthopterin B_2 (2,4-dihydroxy-6-lactyl-7,8-dihydropteridine, isolated from mutant lemon of silkworm) are also reduced by the purified enzyme at a very slow rate (2 and 3% of that of sepiapterin, respectively). All these yellow pteridines have a carbonyl group in $C^{1'}$ in C^6 - side chain and are 7,8-dihydroform.

When isosepiapterin is added to the reaction mixture of sepiapterin reductase with sepiapterin, the reduction of sepiapterin by the enzyme is strongly inhibited. Besides isosepiapterin, many other eye pigment pteridines such as 6-carboxypterin, biopterin, leucopterin and pterin also inhibit the enzyme reaction more or less. % inhibitions are 83, 77, 24, 21 and 18, respectively, when 5×10^{-5} M of each pteridine is added to the same concentration of sepiapterin in the reaction mixture.

The enzyme reaction is reversible as shown in the equation. The pH optimum of this reverse reaction is 10.5. Both reactions of reduction and oxidation are possible to proceed significantly at neutral pH, although the equilibrium lies in favor of reduction (formation of dihydrobiopterin) ($-\Delta G = -12$ Kcal per mole).

These above natures of sepiapterin reductase suggest the complexity of biosynthesis of eye pigment pteridine in Drosophila.

In mammal liver, sepiapterin is reduced to tetrahydrobiopterin by the functions of NADPH-dependent sepiapterin reductase and dihydrofolate reductase as below[3]:

$$\text{sepiapterin} \xrightarrow[\text{(sepiapterin reductase)}]{\text{NADPH} + \text{H}^+} \text{dihydrobiopterin} \xrightarrow[\text{(dihydrofolate reductase)}]{\text{NADPH} + \text{H}^+} \text{tetrahydrobiopterin}$$

Dihydrobioterin has been isolated from rat liver[4]. Tetrahydrobiopterin, thus reduced from sepiapterin, serves as the indispensable natural cofactor of liver phenylalanine hydroxylase, brain and adrenal medulla tyrosine hydroxylase and brain tryptphan 5-hydroxylase, and then, it controls the synthesis of noradrenaline, adrenaline, serotonine and melatonine.

Isosepiapterin can also be converted to an active cofactor of this system but with a very slow rate.

Therefore, sepiapterin and isosepiapterin are not only eye pigments of Drosophila but also very active substances in animals.

References: 1) Nagai, M. 1968, Archi. Biochem. Biophys. 126:426; 2) Katoh, S. 1971, Arch. Biochem. Biophys. 146:426; 3) Matsubara, M., S. Katoh, M. Akino and S. Kaufman 1966, Biochem. Biophys. Acta 122:202; 4) Kaufman, S. 1963, Proc. Natl. Acad. Sci. USA 50:1085.