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Isolation and amino acid composition of Pigeon (*Columba livia*) egg white and egg yolk Riboflavin carrier protein

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Riboflavin carrier protein (RCP) from pigeon egg white and egg yolk have been isolated. The molecular weight of purified RCP of pigeon egg white and yolk has been found to be nearly same as that of previously isolated hen egg white and yolk RCPs. The amino acid composition of pigeon RCPs were observed to be almost similar to that of hen RCPs.

Key words : Riboflavin Carrier Protein, Amino acid composition, Purification,*Columba livia.*

INTRODUCTION

Riboflavin carrier protein (RCP) is present in both avian egg yolk and white, in contrast reptilian eggs it is only present in the yolk (Rhodes et al 1959 and White and Merrill 1988). RCP has an important role in the uptake of riboflavin to the developing oocyte. In mutants of the domestic fowl lacking RCP, the developing embryos die of riboflavin deficiency at around 13 days of incubation (Winter et al 1967). At this stage a rapid increase occurs in flavin kinase activity, requires for the synthesis of FMN and FAD17. RCPs from hen egg white, yolk and plasma have been purified and sequenced (Norioka *et al.,* 1985). All the three proteins are the products of the same gene, although plasma and egg yolk RCPs are synthesized in the liver and egg white RCP is in the oviduct. All the three show polymorphism at 14th position. Egg yolk RCP differs from the other two having 11-13 fewer amino acid residues. When the plasma RCP is taken up into the egg yolk the C-terminal peptide is cleaved. There is also some difference in the glycosylation pattern, although in all the three the oligosaccharides are linked by asparagines 36 and 147 (Miller *et al.,* 1982).

Little detailed characterization has been carried out on any RCPs from other avian sources. The purified RCP from duck egg white has a quite distinct amino acid composition from that of domestic fowl (Muniyappa and Adiga1980). Because of the difference in amino acid composition of egg white RCPs reported for duck and domestic fowl, a wide range of species needs to be investigated in order to establish the common features and differences.

In the present study RCPs from egg white and yolks of pigeon and hen are isolated and pigeon egg white yolk amino acid compositions are analyzed.

MATERIALS AND METHODS

Pigeon eggs were obtained from the local source. DEAE-Sephadex A–50 used in the present study was obtained from Pharmacia Fine Chemicals, Uppasala, Sweden; Sephadex G - 100 from Sigma Chemical Company, St. Louis, USA. Bovine serum albumin, acrylamide N, N, N^1, N^1-Tetramethylethylenediamine, N, N^1-methylene-bis-acrylamide, SDS were procured from Loba Chemical Industrial Company, Bombay, India. All other reagents used were of analytical grade.

Pigeon egg whites and yolks were carefully separated and used immediately. RCP from pigeon egg white was isolated following the methods (Farrell *et al.,* 1969, Hamazume 1984 and Rhodes *et al.,* 1959) with a few modifications as described below. Pigeon egg white was homogenized with an equal volume of 0.1M sodium acetate buffer, pH 4.5; the precipitated protein was removed by centrifugation. To the clear supernatant, DEAE sephadex previously equilibrated with 0.1M sodium acetate buffer pH 4.5 was added. The mixture was stirred for 12 hours at 4 ^{0}C and then suction filtered. The DEAE sephadex with bound protein was washed with excess of 0.1M sodium acetate buffer pH 4.5. Bound proteins were eluted with same buffer containing 0.5M sodium chloride by suction filtration. The eluted protein fraction was dialyzed against water.

Fresh DEAE sephadex, previously equilibrated with

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