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Modification of Separated α and β Subunits of Human Hemoglobin by Iron Tetrasulfonated Phthalocyanine

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In previous papers [1–3] we have shown that it is possible to obtain artificial hemoglobins with metal tetrasulfonated phthalocyanine in the place of heme.

The results presented demonstrate that combination of apoproteins of isolated α and β subunits (apoα and apoβ) with iron tetrasulfonated phthalocyanine (Fe(III)L) leads to the formation of close analogs of the native α and β-chains, Fe(III)L(apoα) and Fe(III)L(apoβ). Both model complexes exhibit characteristic absorption bands at 650 and 678 nm, for the oxidized and the reduced form, respectively. Incorporation of Fe(III)L into α-globin results in a decrease of its molecular weight to the value corresponding to the monomer (21500). Free α-globin in the same conditions occurs as a dimer. Combination of Fe(III)L with β-globin brings about an increase in molecular size from dimeric to tetrameric (64500). The same is observed in the reaction of heme with α and β-globins. Interaction of Fe(III)L(apoα) with Fe(III)L(apoβ) leads to the formation of tetrameric compound $[Fe(III)L(apo\alpha)–Fe(III)L(apo\beta)]_2$.

Combination of α and β-globins with Fe(III)L gives a significant increase of helicity in both proteins, as shown by CD spectra (Fig. 1). In contrast to heme-containing α and β-chains both phthalocyanine derivatives differ in helicity content, that of Fe(III)L(apoβ) being higher. Reconstituted phthalocyanine hemoglobin $[Fe(III)L(apo\alpha)–Fe(III)L(apo\beta)]_2$ exhibits a far UV CD spectrum different from the sum of the individual CD spectra of its subunits. This fact suggests that combination of the modified α and β-chains results in a change in their conformations due to chain–chain interaction, to produce a more ordered or differently ordered structure.

Like native α and β-chains, their phthalocyanine analogs are not equivalent in their properties. It is shown that reduction of the modified β-chain by ascorbic acid occurs at a markedly greater rate than that of the α-chain. Likewise, the rate of oxygen binding is higher in the case of the β-chain. Reconstituted phthalocyanine hemoglobin displays a much higher stability than its subunits.

Recombination of the phthalocyanine modified α and β-subunits with the alternate heme-containing subunits gives hybrid hemoglobins $[Fe(III)L(apo\alpha)–heme(apo\beta)]_2$ and $[Fe(III)L(apo\beta)–heme(apo\alpha)]_2$. These compounds are stable in their ferric forms. Reduced with dithionite they combine with molecular oxygen. Deoxygenation by argon bubbling leads to the formation of inactive species which finally undergo irreversible denaturation.

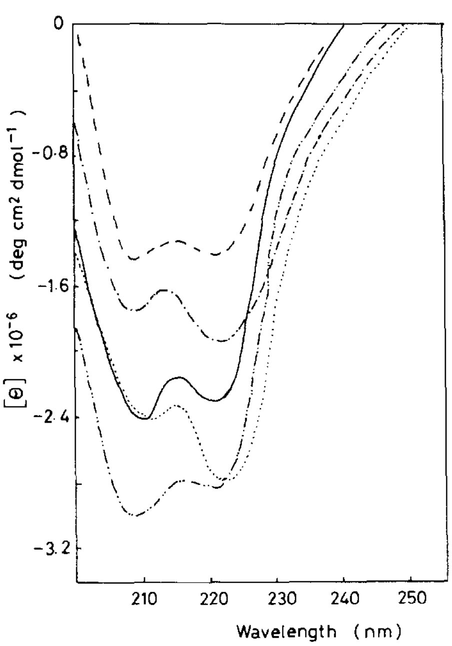

Fig. 1. CD spectra of Fe(III)L(apoα) (———), Fe(III)L(apoβ) (—··—) and $[Fe(III)L(apo\alpha)–Fe(III)L(apo\beta)]_2$, compared to α-globin (-----) and β-globin (-·-·-).

Further studies are in progress.

1 H. Przywarska-Boniecka, L. Trynda and E. Antonini, *Eur. J. Biochem., 52,* 567 (1975).
2 H. Przywarska-Boniecka and L. Trynda, *Eur. J. Biochem., 87,* 569 (1978).
3 H. Przywarska-Boniecka and H. Swirska, *J. Inorg. Biochem., 13,* 283 (1980).

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Infrared Spectroscopy of Carbonmonoxide Ligation to the Coelomic Hemoglobins of *Glycera Dibranchiata*

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Glycera dibranchiata is a marine annelid which possesses both a high molecular weight hemoglobin fraction and a monomer hemoglobin fraction of molecular weight 15,590D [1–8]. NMR studies along with companion isoelectric focusing and column chromatography work have established the extensive microheterogeneity of the monomer hemoglobin