Viviparus ater Hemocyanin: Investigation of the Dioxygen-Binding Site and Stability of the Oxy- and Apo-Forms

Dessislava Nikolova Georgieva^a, Stanka Stoeva^b, Wolfgang Voelter^b and Nicolay Genov^a*

^a Institute of Organic Chemistry, Bulgarian Academy of Sciences, Acad. G. Bonchev-Str.
bl. 9, Sofia 1113, Bulgaria. Fax: 00359-2-700 225. E-mail: genov—n@yahoo.com
^b Abteilung für Physikalische Biochemie, Physiologisch-chemisches Institut der Universität Tübingen, Hoppe-Seyler-Straße 4, D-72076 Tübingen, Germany

*Author for correspondence and reprint requests

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The active site of *Viviparus ater* (mollusc) hemocyanin was investigated using the fact that the binding of dioxygen to the binuclear copper-containing sites of hemocyanins is connected with the appearance of specific dichroic bands which are very sensitive to changes in the structrure and polarity of the environment. Oxy-*Viviparus ater* hemocyanin exhibits near UV and visible circular dichroism spectra different from those of other molluscan and arthropodan hemocyanins. These differences are due probably to variations in the geometry or charge distribution in the dioxygen binding sites of the compared proteins.

The thermostability of *Viviparus ater* hemocyanin and the significance of the copper-dioxygen system for the stability were also investigated. "Melting" temperatures, $T_{\rm m}$, of 77 °C for the oxy-hemocyanin and 57 °C for the apo-protein were calculated from the denaturation curves which demonstrates the considerable role of the binuclear active site for the thermostability. *Viviparus ater* hemocyanin is more thermostable than other hemocyanins for which data are published.