Increased heat stability of α-chymotrypsin through its confinement in liposomes

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For applications of enzymes in confined space, for example inside liposomes (lipid vesicles), the enzyme stability is a critical issue [1]. During the course of our investigations on the entrapment of enzymes inside submicrometer-sized liposomes, we found that the confinement of α-chymotrypsin in liposomes formed from POPC (1-palmitoyl-2-oleoyl-*sn*-glycero-3-phosphocholine) leads to a significantly increased thermostability of the enzyme. Since through the method used – dispersing a dried POPC layer with an aqueous enzyme solution, followed by polycarbonate membrane extrusion – the enzyme entrapment in the liposomes occurs during liposome formation, a stochastic enzyme distribution among the liposomes is obtained. Heat stability experiments showed that a considerable fraction of liposomal α-chymotrypsin is still active after being treated at 80 °C for 30 min, whereas the free enzyme is completely deactivated. For liposome-confined α-chymotrypsin, the heat stability increases as the average number of enzyme molecules per liposome decreases. This high heat tolerance can be explained by a decrease in interactions between partially unfolded enzyme molecules as a result of a decrease in the number of enzyme molecules per liposome compartment. In the extreme case, there is no opportunity for the irreversible formation of enzyme aggregates – which leads to enzyme deactivation – in the case of single enzyme molecule confinement. Whether this finding also holds for other monomeric enzymes is currently under investigation.

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