Comparative study of different lipases acting on milk fat globule membrane monolayers.

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Lipases are enzymes that specifically act on aggregated lipids in an aqueous environment and more accurately at interfaces. In order to explain the pathway of lipolysis, several investigators have proposed a reversible adsorption or penetration of the enzyme into the interface. This first step is supposed to precede the formation of the enzyme-substrate complex (*1*). Studies using monolayers have shown that activity of lipolytic enzymes will be greatly influenced by "the interfacial quality" (*1*) (*2*). For reasons of simplification, most studies of model interfaces have involved only one or two type of lipid and/or protein. Generally all biological interfacial regions are however composed of a complex mixture of lipids and proteins. An example of action of lipases which causes real problems is lipolysis of milk (*3*). In milk and dairy cream, the interfacial region between milk fat and the aqueous phase where lipases may be present is the milk fat globule membrane (MFGM). This MFGM is a complex combination of proteins and phospholipids (*4*).

The main purpose of this work was to describe the interactions between a MFGM monolayer and three kind of lipases in order to investigate the mechanism of lipolysis in milk.

The monolayer technique used in this work has the advantage that the arrangement of the molecules can be controlled by changing the molecular area and the surface pressure of the monolayer. So this monolayer technique was used to provide a model system where the quality of the interface can be controlled (*5*) (*1*).

The lipases studied in this work are representative of those which are responsible for lipolysis in milk products. We used porcine pancreatic lipase (PPL) and milk lipoprotein lipase (MLPL); these two enzymes are close in term of structure and activity. We also studied a lipase from *Pseudomonas fluorescens* (PFL) because this is a typical bacterium of refrigerated milk (*3*) (*6*).

So the interactions of porcine pancreatic lipase, milk lipoprotein lipase and lipase from *Pseudomonas fluorescens* with milk fat globule membrane (MFGM) monolayers have been compared using a Langmuir film-balance.

MFGM was first extracted from raw cream and compositional analyses were performed to confirm that material was representative of that which is really present around milk fat globules.

Interaction of these three lipases with MFGM depends on surface pressure of MFGM monolayer, and thus on its organisation.

Lipase from *Pseudomonas fluorescens* was able to interact with MFGM monolayer compressed at surface pressure up to 30 mN/m while porcine pancreatic lipase and milk lipoprotein lipase are unable to interact with MFGM monolayer compressed at surface pressure higher than 25mN/m (Figure 1a-b).