# KINETIC DETERMINATION IN REAL TIME OF THE STARCH HYDROLYSIS REACTION USING LASER INTERFEROMETRY TECHNIQUES

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#### Abstract

This paper presents a study about kinetic determination in real time of the influence of some food additives (lecithin, cholesterol and oleic acid) over the starch hydrolysis reaction using laser interferometer techniques.

The principle of this method consists in determination in real time of refractive index changes of amylase-starch biochemical system using a laser interferometer. A C.C.D. camera acquire in real time the fringes of interference modification and the image processing and analysis and also the drawing of the graphics are realised by using a computer program made by the authors.

The lecitine, cholesterol and oleic acid act as inhibitors for amylase activity and decrease the rate of the enzymatic reaction so, these lipids could be utilised for the amelioration of the hydrolytically process of the starch.

#### INTRODUCTION

The starch hydrolysis with amylases is a very important reaction in the bakery industry. In bakery, they usually use food lipids as lecithin, cholesterol and oleic acid to improve the quality of bakery products. These lipids seem to influence the starch hydrolysis reaction. To determine the rate of the amylase activity, the authors had created and realised a device which function principle is based on the determination of the refractive index variations at the starch hydrolysis with amylase [1]. At each hydrolysis step it takes place a modification of the refractive index, variation that is determined in real time.

The system realised is one complete integrating. So, using a Michelson interferometer is determined the modification in real time of the interference fringes due to the refractive index variations of the biochemical starch-amylase system, modification that is acquired and processed in real time by a computer which had attached a CCD camera [2].

The Michelson interferometer used provides interference fringes, which are formed on a screen made by a white sheet of paper, and so, the visual sensor that is located on the opposite side of the screen, acquires the image in optimum conditions. On the screen appear successive images with interference fringes. The solution refractive index is changing in time, in the same way that the hydrolysis reaction occurs, so on the screen appear new interference maxims that correspond to the different hydrolysis steps. The CCD visual sensor acquires the image formed on the screen and sends it to the data acquisition board. The CCD sensor used had a density of 10000 receptors/mm<sup>2</sup> uniform distribute, and the total number of the receptor is 640x480, that determine a high resolution of the system. The program realised and elaborated in C++ language offers the possibility of acquiring and processing images, processing which consists in the determination of the number of changes in the interference fringes [3, 4, 5].

In figure 1 is presented the principle scheme of the conceptual and realised device.



Figure 1. The principle scheme of the device

### **MATERIALS-METHOD**

### Reagents:

- 1. Soluble starch supplied by Merck, Darmstadt was used in 1% concentration in aqua's solution.
- 2. It was used an enzymatic extract of alpha and beta amylase from wheat flour prepared by extracting 10 g wheat flour in 100 ml distillate water for 30 minute using a magnetic stirring, than centrifuging at 6000 rpm

for 10 minutes. The supernatant obtain was dilute 10 times in distillate water.

3. Lipids were used in 0,1% concentration as solutions in methanol: chloroform: water (1:1:1)

Interferometer analysis:

In the recipient located on the interferometer it was introduced 5 ml soluble starch, 2 ml distilled water, 0,5 ml lecithin 0,1% (sample 1), 0,5 ml cholesterol 0,1% (sample 2) and 0,5 ml acid oleic 0,1% (sample 3) and 1 ml amylase extract as shown in table 1

Table 1

	Lab techniques			
	Control	Sample 1	Sample 2	Sample 3
Starch	5ml	5ml	5ml	5ml
Amylase	1ml	1ml	1ml	1 ml
Distillate water	2ml	2ml	2ml	2ml
Lipid 1%	0,5ml	0,5ml	0,5ml	0,5ml

For the samples was made a control, identically with the tests, except that in the control there was no lipid in solution. When the amylase is introduced in tube, the hydrolysis reaction started and the rate of hydrolysis is expressed as the numbers of changes of refractive index in time.

### **RESULTS AND DISCUSSIONS**

The graphics (numbers of changes vs. time) were realised by using a computer program made by the authors and are shown in figure 2.



Figure 2. The influence of the lecithin, cholesterol and oleic acid over the hydrolysis reaction of starch with amylase

## CONCLUSIONS

The lecitine, cholesterol and oleic acid are inhibitors for amylase activity and decrease the rate of the enzymatic reaction. These results indicated that these lipids could be utilised for the amelioration of the hydrolytically process of the starch with low or great amylasic activity.

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