



# CDR FoodLab® method for the determination of Lactose content in lactose-free dairy products.

This study demonstrates the great accuracy and repeatability of CDR FoodLab® method for the determination of total Lactose content in milk and dairy samples. It allows the producers to improve at best the execution of this analysis in the QC laboratory or even directly at the production line thanks to its quickness and easiness.

#### **Abstract**

Nowadays lactose-free diary products are gaining an ever bigger stake in the market, due to the increase in lactose intolerance in adult population. For this reason lactose determination in lactose free products appears more and more as an important analisys for producers, both at line and as a final quality control on the finished dairy product . In this work a new CDR FoodLab® improvement for lactose determination is assessed in terms of reproducibility, accuracy and recovery. CDR FoodLab® method and traditional enzymatic method are also taken into comparison on adding determined aliquotes of lactose in real lactose-free matrices. CDR FoodLab® method appears to be much quicker and with a friendly use for everyone if it is compared to the tradional enzymatic kit.

#### Introduction

Lactose is the main sugar molecule that can be found into milk. It is a disaccharide, formed by D-galactose and D-Glucose. The two monosaccharides can be liberated during digestion by lactase (b-galactosidase), an enzyme found in gastric juice that can promote lactose hydrolisys. The lactase is the enzyme added in the delactozation process that removes the lactose from milk. The monitoring of this

process and the final lactose concentration are two important steps that have to be carefully controlled. Also the concentration in the finished



dairy products such as for instance cream, yogurt or cheese has an high importance for the quality control. Then, lactose determination seems to be a crucial analysis for the internal quality control laboratories and for the monitoring in real-time of the delactosation process in dairy industry.

#### The aim of CDR FoodLab®

CDR FoodLab® is a food analysis system designed to perform quality controls during all the production phases. In the CDR laboratories was developed a new method to quantify total lactose content in lactose-free products. This new method is much faster than traditional procedures to determine lactose content and it can easily be





used in the internal QC laboratory or even directly at production line.

# **Traditional enzymatic method**

The traditional enzymatic method for the determination of lactose in food matrices is based on photometric measurements of the evolution of NADH after lactose digestion by means of lactase enzymes. The sample preparation and analysis consists in various steps and could be considered very time consuming for the ordinary quality assessment. First of all the samples have to be treated using a basic sodium borohydride solution at controlled temperature (40°C) for about 30 minutes and then the sample has to be filtered or centrifuged (depending on the fat/protein content in it). For the analysis step two different cuvettes for each sample have to be prepared, in order to quantify precisely the lactose content. On both cuvettes three different reagents have to be added and an incubation time of 2h (for freelactose samples) is mandatory. The final lactose content is determined photometrically at a wavelength of 340 nm by NADH evolution after two stages of enzyme digestion. Prior and during the assay various sample dilution steps are needed to obtain a certain lactose content in the cuvette. This can force the user to perform various subsequent tests, in order to optimize the procedure (expecially for unknown lactose For all these reasons, the lactose determination with the common commercial kits appears to be very time consuming and furthermore the analysis must be carried out from people with high chemical skill.

# CDR FoodLab® method



CDR FoodLab® analyzer is based on photometric technology, equipped with LED emitters, reading cells and 37°C

thermostated incubation cells. The using of this specific emitting technology allows to perform analysis on milk or dairy process without the long clarification process used by normal photomethers equipped with standard tungsten lamps.

CDR FoodLab® method for lactose determination uses a reagent in pre-filled cuvette. 10 microL of diluted sample (1 to 10 with water) and 50  $\mu$ l of reagent R1a are added to the cuvette and after 300 seconds the reaction is blanked. 15  $\mu$ l of reagent R2 are then added and the final reading is performed after another 300 seconds of incubation time. Various samples can also be analyzed at the same time due to the versatility of the CDR methodology.

The reaction is measured at the wavelength of 505nm. All reagents are supplied ready to use, then operators do not have to handle toxic or carcinogenic compounds. There is no need of a dedicated laboratory or to wash either containers or traditional glassware because CDR FoodLab® analysis system allows to perform test on samples in total autonomy. Thanks to its easiness also people without chemical skill are able to carry out this test in total autonomy.





## CDR FoodLab® to the test:

First of all CDR FoodLab® method was tested to estimate the quantity of the analyte (total lactose) in milks whith different lactose concentration. Samples with a content of 0.05, 0.10, 0.20, 0.50 and 1.00 grams of lactose on 100 grams of product were prepared mixing a  $\beta$ -lactose standard solution to a delactosate milk (lactose concentration below 0.01 g/100g). The delactosate milk was boiled before the additions to deactivate the

	Lactose concentration (g/100g)		
	Calculated	CDR FoodLab	
Addition 1	0.05	0.06	
Addition 2	0.09	0.10	
Addition 3	0.20	0.21	
Addition 4	0.45	0.50	
Addition 5	0.90	1.05	

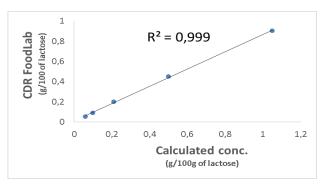
**Table 1** Calculated and measured lactose concentrations with CDR FoodLab® on the spiked free lactose milk samples.

excess lactase enzyme (in every one of the following tests). All samples were measured as triplicates. In Table 1 are showed the calculated and measured lactose concentration in the created samples.

The results were also plotted in Figure 1, in which the theoretical sample concentrations and the measured concentrations obtained using CDR FoodLab $^{\otimes}$  method are correlated, with a remarkable R $^2$  of 0,9993.

At the bottom of these results, a series of commercial low lactose milk and dairy products were tested using the CDR method.

After the assay, on different kind of dairy products such as cream, cheese, yoghurt etc., specific amounts of lactose were added, to obtain an increase in lactose concentration of 0.10 g/100g each. Before and after each addition,



**Figure 1** Correlation between CDR FoodLab® and theoretical calculated concentrations of lactose in spiked samples.

lactose concentrations were determined using CDR FoodLab® method in order to evaluate the percentage of recovery, Table 2.

10 tests using CDR FoodLab® method were performed to obtain and average value and the standard deviation, Table 3.

	Initial	Final	%
	conc.	conc.	Recovery
	with CDR	with CDR	
	FoodLab	FoodLab	
Lactose-free milk	<0.01	0.11	110
Cheese	0.01	0.12	109
Cream 1	<0.01	0.10	100
Cream 2	<0.01	0.12	120
Yogurt	0.06	0.18	112

**Table 2** Recovery tests on different kind of dairy products.

products.		
	Lactose (g/100g)	
Repetition 1	0.14	
Repetition 2	0.16	
Repetition 3	0.16	
Repetition 4	0.17	
Repetition 5	0.18	
Repetition 6	0.17	
Repetition 7	0.17	
Repetition 8	0.16	
Repetition 9	0.15	
Repetition 10	0.16	
AVERAGE	0.16	
STANDARD DEV.	0.01	
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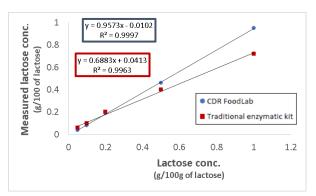
**Table 3** Recovery tests on different kind of dairy products.





# Comparison between CDR FoodLab® method and a commercial enzymatic kit

Both the CDR method and the traditional method with enzymatic kit were employed to measure lactose content in a series of samples obtained cutting delactosate milk with different amounts of undelactosated milk. Five different lactose concentrations were again obtained: 0.05, 0.1, 0.2, 0.5 and 1.0 lactose grams on 100 grams of product. In Figure 2 the theoretical concentrations are plotted vs. the measured concentration for both methods.



**Figure 2** Correlation between CDR FoodLab<sup>®</sup> , traditional enzymatic kit results and theoretical concentrations.

It can be seen how in the traditional method, despite showing a good correlation, displays lower lactose concentrations in respect to the calculated ones. This can be probably attributed to the errors introduced by the various diluition steps needed to perform the analisys.

The results obtained demonstrate that CDR FoodLab® is a method able to quantify total lactose content in real samples in a fastest but yet accurate way in respect to the traditional method.

#### **Conclusions**

CDR FoodLab® is a versatile system specifically developed by CDR to respond to the needs of milk and dairy products producers. CDR FoodLab® analysis system allows to perform tests with one single instrument, more rapidly and easily than traditional methods.

## References

J.A. Sánchez-Manzanares , M.R. Fernández-Villacañas, F.Marín-Iniesta, J.Laencina. Determination of lactose by an enzymatic method. Food Chemistry Volume 46, Issue 4, 1993, Pages 425-427.

Beutler, H. O. (1988). Lactose and D-Galactose.

In Methods of Enzymatic Analysis
(Bergmeyer, H. U., ed.), 3rd ed., Vol. VI,
pp. 104-112, VCH Publishers (UK) Ltd,
Cambridge, UK.





