

### Research & Reviews in

ISSN: 0974 - 7532

# **BioSciences**



RRBS, 8(11), 2014 [422-428]

### Profiles of lactose and galactose in dairy products consumed by **Cameroonian populations**

Esther Ngah1\*, Valentin D.Guiama1, Robert G.Béka1, Catherine J.E.Ingram2, Bryony Leigh Jones<sup>3</sup>, David Zeitlyn<sup>4</sup>, Neil Bradman<sup>2</sup>, Dallas Swallow<sup>2</sup> <sup>1</sup>Department of Food Science and Nutrition, National Advanced School of Agro-Industrial Sciences, University of Ngaoundere, P B 455, (CAMEROON) <sup>2</sup>Department of Biology, Galton Laboratory, University College London, Wolfson House, 4 Stephenson Way, London NW1 2HE, (UK) <sup>3</sup>The Centre for Genetic Anthropology, University College London, London, (UK) <sup>4</sup>Centre for Social Anthropology and Computing, Department of Anthropology, University of Kent, Canterbury, (UK) E-mail: hadassa.ngah@gmail.com

#### **ABSTRACT KEYWORDS**

Lactose intolerance limits the consumption of milk and dairy products. This study was carried out to provide information concerning the levels of lactose and galactose in dairy products widely consumed in Cameroon. Lactose and galactose were determined in a total of 23 dairy products consumed by Cameroonian populations, using Megazyme kit. Results indicated that lactic acid fermentations result in a decrease in lactose content associated with the reduction of pH, and an augmentation in free galactose. The values of pH were ranged from 6.43 Biradam (Cameroon style unfermented dairy products) to 3.50 Pendidam Cameroon-style fermented milk foods. The values of lactose content increased from fermented to unfermented products, and ranged as follows: Pendidam 1.16 – 2.23 %; *Kindirmou* Cameroon style fermented milk 1.39 – 3.31 %; Biradam 4.39 - 5.0 %; imported dairy products 3.09 - 6.58 %. The concentration range of free galactose was 0.03 - 0.93% (Kindirmou), 0.06-0.42% (*Pendidam*), 0.0-0.35% (imported dairy products). In conclusion, lactic acid fermentation reduces significantly the levels of lactose and minimizes resulted free galactose in Cameroon style fermented dairy products. This suggests that traditional technology used to produce fermented milk is effective to provide dairy products to the lactose maldigesters. © 2014 Trade Science Inc. - INDIA

Lactose; Galactose: Dairy products; Cameroon-style fermented milk.

#### **INTRODUCTION**

Lactose is a milk sugar (4-0-β-D-galactopyranosyl-D-glucopyranose); it requires digestion by the intestinal enzyme lactase before it can be absorbed in the small intestine, and thus used as a food source<sup>[1]</sup>. Lactase is present in all healthy human babies but only persists into adulthood in some people<sup>[2]</sup>. The frequency of this trait varies dramatically in different populations of the world. It is for example very frequent in Northern Eu-

ropeans and very rare in southern China, while in Africa the distribution is patchy, with an observed correlation with pastoralism and drinking of fresh milk. In Sub-Saharan Africa, milk production is the trade of nomad people, who drink large quantities of fresh milk, and lactase persistence is frequent<sup>[3,4]</sup>.

The consumption of fresh milk by lactase non-persistent adults and even children can lead to symptoms of severe flatulence and diarrhoea<sup>[2,5]</sup>. However, fermentation of milk reduces its lactose content to variable extents, and making it more tolerable. During this process, free galactose which has been involved in the aetiology of senile cataract increases in the final dairy product<sup>[1]</sup>. Many people throughout Africa enjoy soured dairy product. In these products, lactic bacteria play an essential role in preserving a highly nutritious food product. Fermented dairy products are also of a great significance for their therapeutic values; for alleviating lactose intolerance<sup>[6]</sup>; social value and as a mean of generating income. In locally fermented milk (Pendidam) manufactured by Mbororo, no starter is added; fermentation is based on the spontaneous fermentation by natural microflora present in raw milk<sup>[7]</sup>. Sometimes, the milk is seeded with an old *Pendidam*, and the cream skimmed off after fermentation. Pendidam is mainly used as an ingredient for other dishes such as pap. The pasteurized milk is seeded with old Kindirmou or commercial starters at temperature of between 40 – 45°C for 6 hours. According to demand or variation of raw milk production, the fresh milk is increasingly replaced with imported powdered milk. Nowadays, locally fermented dairy products are both made of raw milk and imported powdered milk. The physicochemical and microbiological characteristics of fresh and Cameroonian style fermented milk have been evaluated[8-10]. These authors referred that the microbiological quality of those products is very changing, and often poor. However, there is very few information on lactose content in dairy foods consumed by Cameroonian populations. In certain cases, manufacturers give just total sugar concentration, but not lactose and galactose contents which are intolerant for some categories of consumers.

The aim of this work is to assess the content of the milk sugar (lactose) in dairy products widely consumed by people in Cameroon. In addition, galactose content of these dairy foods was determined, while pH was

only measured in the traditional milk products. Three categories of products were tested: fresh milk types (*Biradam*), traditional fermented milks (*Kindirmou*, *Pendidam*), and imported dairy products (powdered milk, condensed milk and long life milk).

#### MATERIALS AND METHODS

#### **Collection of samples**

Twenty three (23) samples group in three categories were collected within the period extending from November 2007 to February 2008 in the Ngaoundéré, Cameroon. Three samples of fresh milk were obtained from the nearby dairy farmers group (three groups). Twelve samples of traditional fermented milks were obtained from individual households (*Mbororo* women) in rural areas and small dairies so-called "dairies bar" in urban areas. Eight samples of imported milk products (four powdered milk, three condensed milk and one long life milk) were purchased from the local market. Samples were analysed within two days of purchase at least in triplicate.

### **Determination of pH**

The pH of traditional dairy samples was measured as soon as they were received in laboratory by direct probing of the glass electrode of pH-meter (Hanna Instruments, HI 9124, USA). Six replicates of each measurement were performed for each given sample. Dairy samples were stored at 4°C until lactose and galactose assay procedure.

#### **Determination of lactose and galactose content**

The lactose and galactose contents were determined according to the Association of Official Analytical Chemist methods<sup>[11]</sup>, for the enzymatic analysis of sugars, using Megazyme kit (lactose and galactose assay procedure K-LACGAR 08/05, Ireland). The method involved were the detection of galactose using galactose dehydrogenase and measuring the conversion of NAD+ to NADH by increase in absorbance at 340 nm, and of indirect determination of lactose by enzymatic conversion of lactose to galactose by  $\beta$ -galactosidase.

The powdered and condensed milks were prepared prior following the manufacturer's instructions. 26 g of NIDO samples were dissolved in 180 ml of warmed water. 22.5 g of INCOLAC samples were mixed with

150 ml of boiled but cooled water [room temperature]. 22 g of PEAK samples were mixed with 170 ml of boiled but cooled water [room temperature] and same for TOP MILK samples. For condensed milk samples the content was diluted in twice the volume of the container.

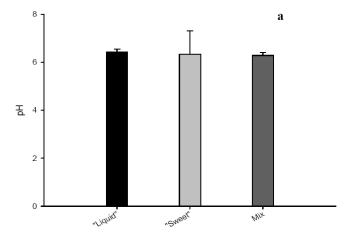
Each sample was weighed (about 0.5g) in triplicate into 50-ml volumetric flask to which water and Carrez reagents (1ml, 85 mM potassium hexacyanoferrate II and 1ml 250 mM zinc sulphate) were added to precipitate proteins. The pH of samples was adjusted by adding 100mM of NaOH/lactic acid and water was added to cool samples and make up the volume to 50 ml. Samples were filtered prior to analysis. Blanks were run with each sample containing the same ingredients. Assays for lactose and galactose involved enzymatic hydrolysis of lactose by \( \beta\)-galactosidase and oxidation of galactose by galactose dehydrogenase with the formation of stoichiometric amounts of NADH. The NADH produced was quantified by the increase in absorbance at 340 nm. The concentrations of NADH were calculated directly from its extinction coefficient but lactose standard curves were plotted using solutions of concentrations ranging between 8µg - 160 µg. The smallest differential of the absorbance for the assay is 0.010 absorbance units. Results were expressed as percentage (% w/w).

#### Statistical analysis

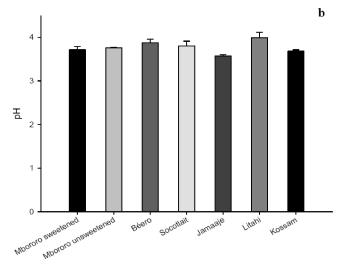
Statistical analysis was performed using Statgraphics® Plus version 5.0. Data were analyzed by one way ANOVA (analysis of variance) at 95 % level of confidence. Multiple comparisons were performed through 95 % least significant difference intervals. Values represent mean of three or more replicates, in addition to standard error of the mean.

#### RESULTS AND DISCUSSION

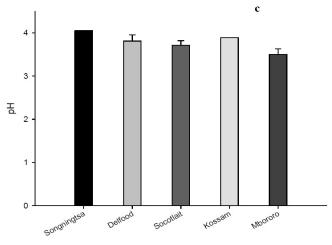
The list of dairy products tested in this study and their characteristics in terms of pH, lactose and galactose contents are shown in TABLE 1, Figure 1&2. *Biradam*, unfermented dairy products exhibited the greatest pH ranging from 6.28 (mix) - 6.43 (liquid). There was no difference (P>0.05) between *Biradam* dairy products in terms of pH (Figure.1a). The value of pH in traditional fermented dairy products (*Pendidam* and *Kindirmou*) was variable. It ranged from 3.57 (from



Biradam dairy products group



Kindirmou dairy products group



Pendidam dairy products group

Figure 1: pH of artisanal dairy product group found consumed by Cameroonians populations. (a) Biradam; (b) Kindirmou; (c) Pendidam. Values represent the mean  $\pm$  standard deviation of three or more samples.

*Jamaaje*, small dairy) to 3.99 (from *Litahi*, small dairy) for *Kindirmou* fermented milk group, and from 3.50

(Pendidam from Mbororo dairy farm) to 4.05 (Pendidam from Songningtsa, small dairy) for Pendidam fermented dairy group products. Concerning Kindirmou dairy group products, fermented milks from Jamaaje and Kossam, small dairies were more acidic than the other products of this group (Figure. 1b). The pH values of Pendidam from Delfood, Socotlait small dairies and Mbororo farm were not different (P>0.05), while they were lower (P<0.05) than those of Pendidam from Kossam and Songningtsa

small dairies (Figure 1c). *Pendidam* from *Mbororo* dairy farm was more acidic than *Kindirmou* from the same farm. There was positive, high and significant polynomial relationship (R<sup>2</sup>= 0.85; P< 0.001) between lactose content and pH of milk (Figure 3). It is obvious that the decrease in pH value is essentially due to the fermentation of lactose. However, the value of pH may be modified also through fermentation of amino acids derived from milk proteins or fermentation of fatty acids produced by hydrolysis of milk fat by some micro-

TABLE 1: Variation of lactose and galactose contents in traditional and imported dairy products consumed by Cameroonian populations

Product group	Description	Galactose (% w/w)		Lactose (% w/w)	
		Mean	SEM	Mean	SEM*
	Biradam/fresh or pasteurized milk				
Artisan raw/ pasteurized	"Liquid"	ND	-	4.39 <sup>b</sup>	_
	"Sweet"	ND	-	$5.0^{b}$	0.01
	Mix	ND	-	$4.50^{b}$	-
Artisan fermented	Kindirmou				
	Mbororo sweetened	0.23 <sup>e</sup>	-	1.90 <sup>de</sup>	0.68
	Mbororo unsweetened	$0.03^{g}$	-	$2.87^{c}$	0.01
Small-dairies fermented	Béero	0.93 <sup>a</sup>	0.11	3.31°	0.21
	Socotlait	$0.52^{b}$	0.07	3.15°	0.44
	Jamaaje	$0.84^{a}$	0.03	1.39 <sup>e</sup>	0.36
	Litahi	$0.04^{\mathrm{fg}}$	-	2.84°	0.02
	Kossam	$0.32^{d}$	0.02	$2.23^{cd}$	0.23
Artisan fermented	Pendidam				
	Mbororo*	$0.06^{\mathrm{fg}}$	-	1.16 <sup>e</sup>	0.08
	Songningtsa	$0.42^{c}$	0,01	$2.11^{d}$	0.54
	Delfood	$0.36^{\rm c}$	0,06	$2.19^{d}$	0.62
	Socotlait	$0.27^{d}$	0,02	$2.49^{cd}$	0.11
	Kossam	$0.12^{f}$	-	2.23 <sup>cd</sup>	-
Imported dairy products	Whole milk powder				
	Top milk	ND	-	3.56 <sup>bc</sup>	0.97
	Nido	$0.02^{g}$	-	5.03 <sup>b</sup>	1.15
	Incolac	$0.03^{g}$	-	$3.10^{\rm c}$	0.96
	Peak	$0.02^{g}$	-	$3.09^{c}$	0.55
	Sweetened evaporated milk				
	Peak	ND	-	6.58 <sup>a</sup>	0.88
	Nestlé	ND	-	5.28 <sup>ab</sup>	0.48
	Unsweetened evaporated milk				
	Olympic	0.35 <sup>d</sup>	-	3.62 <sup>bc</sup>	0.37
	Long life milk				
	Bridel	$0.04^{g}$	-	$4.78^{b}$	0.06

SEM, standard error of the mean; ND, not detected; \*low fat yoghurt live, Means in the same column followed by different letters in superscript are significantly different at probability level 0.05.

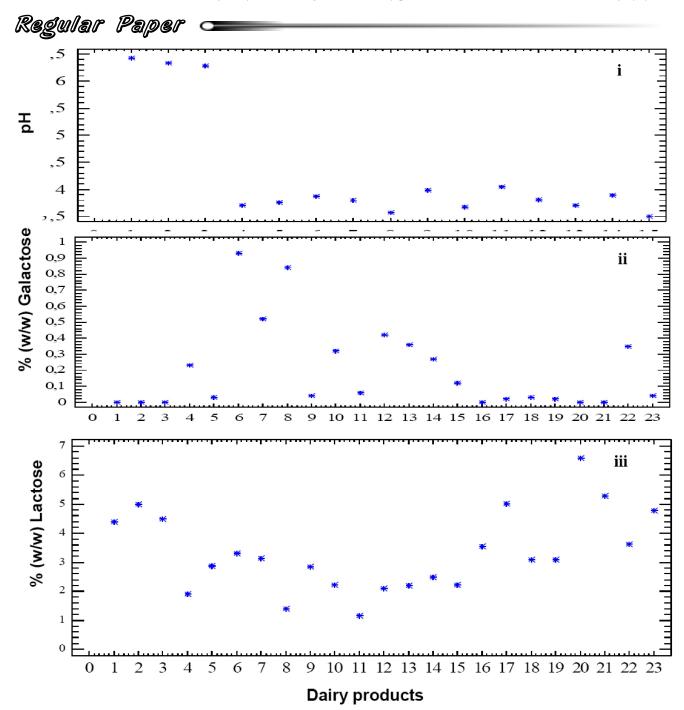


Figure 2: Profiles of pH (i), galactose (ii) and lactose (iii) in dairy products consumed by Cameroonian populations: 1, fresh milk without treatment; 2, pasteurized and sweetened milk; 3, mix of the two later samples; 4, pasteurized, fermented and sweetened milk from *Mbororo* farm; 5, pasteurized, unsweetened and fermented milk from *Mbororo* farm; 6, fermented milk from *Mbéero* small-dairy; 7, fermented milk from *Socotlait* small-dairy; 8, fermented milk from *Jamaaje* small-dairy; 9, fermented milk from *Litahi* small-dairy; 10, fermented milk from *Kossam* small-dairy; 11, *Mbororo* yoghurt like from farm (low fat); 12, Cameroon style yoghurt (with zebu milk or milk powder) from small-dairy (*Songningtsa*); 13, Cameroon style yoghurt from small-dairy (*Mossam*); 14, Cameroon style yoghurt from small-dairy (*Socotlait*); 15, Cameroon style yoghurt from small-dairy (*Hossam*); 16, imported milk powder (Topmilk); 17, imported milk powder (NIDO); 18, imported milk powder (Incolac); 19, imported milk powder (Peak); 20, sweetened concentrated milk (Peak); 21, sweetened concentrated milk (Nestlé); 22, unsweetened concentrated milk (Olympic); 23, long life milk (Bridel). Values represent the mean of three or more samples.

organisms<sup>[12]</sup>. Like other milk technological processes, it modifies the sugar profiles in final products<sup>[5]</sup>. However, Lactic acid fermentation improves minerals utili-

zation and conservation of milk<sup>[13]</sup>.

During the fermentation process, lactose is hydrolyzed by  $\beta$ -D-galactoside galactohydrolase or  $\beta$ -D-

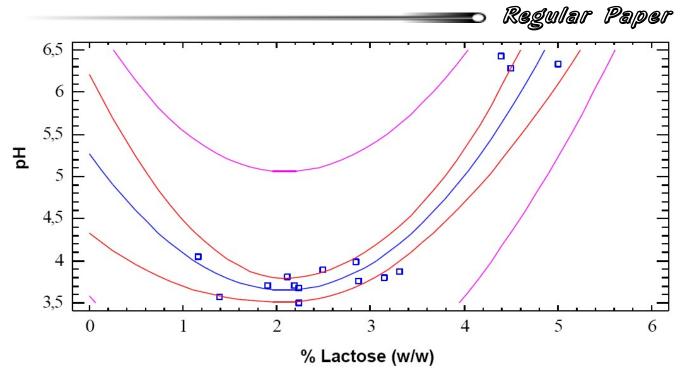


Figure 3 : Polynomial relationship between lactose levels and pH values of Cameroon artisanal dairy products ( $R^2 = 0.85$ ; P< 0.001).

phosphogalactoside galactohydrolase from microorganisms to produce galactose and glucose. In these conditions, growth media contains lactose, galactose, and glucose, in favour of catabolite inhibition, microorganisms use lactose and glucose simultaneously; while galactose remains until the other sugars have disappeared<sup>[12]</sup>. That explains the significant increase in free galactose in all cultured products compared with unfermented milk (TABLE 1, Figure 2ii). The greatest values found in Kindirmou from Béero (0.93 %) and Jamaaje (0.84 %) mini-dairies. The values of free galactose in fermented milks from dairy farm (0.23 % and 0.03 %, sweetened and unsweetened Kindirmou from Mbororo; 0.06 % *Pendidam* from *Mbororo*) was lower (P < 0.05) than the ones of cultured milks from small dairies, excepted for Pendidam from Kossam (0.12 %) and Kindirmou from Litahi (0.04 %). Surprisingly, unsweetened condensed milk (Olympic) showed higher (P<0.05) value of free galactose than other imported dairy products. Free galactose was not detected in all Biradam groups of products, sweetened condensed milks and Top Milk whole powdered milk (TABLE 1). The values of free galactose in dairy products (range 0-0.93) consumed by Cameroonian populations are lower to those reported by<sup>[5]</sup> for cultured dairy products found on UK market (range 0-1.97%). The fermentation used in the manufacture of Cameroon style fermented dairy foods minimizes the levels of free galactose; due to either commer-

cial starters or spontaneous microflora.

Consumption of milk and dairy foods is growing in interest among the Cameroonians populations. So, digestion disturbances of milk (lactose intolerance) which can occur due to lactase deficiency or intestine injury could become a major interest to health professionals and the public<sup>[1,13]</sup>. The values reported in the present study for lactose in dairy products widely consumed by Cameroonian populations were ranged from 1.16 (Pendidam from Mbororo) – 6.58 % (Peak – sweetened condensed milk) (TABLE 1, Figure 2iii). The levels of lactose were lower (P < 0.05) in the traditional fermented dairy products than Biradam group and imported dairy foods. Traditional technology used to make Cameroon-style fermented milk lead up to the decrease in lactose from 34.6 % (Kindirmou) - 73.6 % (Pendidam). Pendidam group of products showed the lowest values of lactose (range 1.16 – 2.23 %) than those of *Kindirmou* (1.39-3.31%). This is probably a consequence of long period of fermentation during production of *Pendidam* (12 hours), while *Kindirmou* production lasts 06 hours. The values of lactose in Cameroon-style fermented milk were similar to those reported by O'brien<sup>[5]</sup> for fermented milk found on the UK market, excepted fermented buttermilk (5.86 %) and, Yoghurts – plain low fat and full fat (4.17 % and 3.90 % respectively). The high levels of lactose in fermented dairy products could be due to the addition of

the powdered milk and sucrose during manufacture<sup>[5]</sup>. Lactic acid fermentation may take place using either free glucose or glucose and fructose from sucrose as substrate instead of glucose and galactose from lactose; maintaining the high levels of lactose in fermented dairy foods. However, there is evidence that fermented milks improve lactose digestion and tolerance in lactose intolerant individuals<sup>[2]</sup>.

Elliot et al.<sup>[15]</sup> found that 95 % of "Bantou" people are lactase non-persistent persons. Majority of inhabitants in Cameroon are Bantou origin, so that lactose maldigestion is public health issue. Despite of this situation, the levels of lactose and galactose in Cameroon-style fermented milks can be tolerated by lactose non-digester. However, the consumption of these products induces symptoms lactose intolerance. This is often been attributed to the poor hygienic quality of fermented Cameroon-style fermented dairy products<sup>[10]</sup>.

#### **CONCLUSIONS**

The consumption of milk and dairy products is growing in interest in Cameroon. The pH values of Cameroon style fermented milks (Kindirmou and Pendidam) are significantly lower than unfermented milks (*Biradam*). Pendidam is more acidic than Kindirmou. The levels of lactose follow these tendencies; therefore lactic acid produced from lactose fermentation improves milk conservation and its nutritional properties, such as mineral utilization and lowering lactose. Contrary to the lactose, results showed that free galactose increase in fermented milks compared to unfermented milk products. This study suggests that technology used to produce Cameroon-style fermented dairy foods reduces significantly the levels of lactose and minimizes resulted free galactose. However, these fermented milks may be contaminated with E. coli, mould and yeast which may cause the same symptoms as for the lactose intolerance. We noted that process is more variable, and need standardization which must eliminate all the source of harmful microorganisms.

#### **ACKNOWLEDGEMENTS**

The authors thank Melford Charitable Trust for their funding contribution.

#### REFERENCES

- [1] V.H.Holsinger; Fundamentals dairy chemistry, N.P.Wong, R.Jenness, M.Keeney, E.H.Marth 3<sup>th</sup> Editionn. A Chapman & Hall Food Science Book, Gaithersburg, Maryland, 279–342 (**1999**).
- [2] G.D.Miller, J.K.Jarvis, L.D.McBean; Lactose digestion, in Taylors and Francis Group, Eds; Handbook of Dairy Foods and Nutrition, National Dairy Council, New York, 299-331 (2007).
- [3] C.A.Mulcare, M.E.Weale, A.L.Jones, B.Connell, D.Zeitlyn, A.Tarekegn, D.M.Swallow, N.Bradman, M.G.Thomas; American Journal of Human Genetic, 74, 1102–1110 (2004).
- [4] C.J.E.Ingram, M.F.Elamin, C.A.Mulcare, M.E.Weale, A.Tarekegn, T.O.Raga, E.Bekele, F.M.Elamin, M.G.Thomas, N.Bradman, D.M.Swallow; Human Genetic, **120**, 779–788 (2007)
- [5] J.O'Brien; Journal of human Nutrition and Dietetics, 12, 245-250 (1999).
- [6] S.R.Hertzler, S.M.Clancy; Journal of American Dietetic Association, **103**, 582-607 (**2003**)
- [7] N.F.Zambou, Z.El Desouky, S.Adb El Arazek, T.F.Mbiapo, M.El Soda; Eg.Journal Dairy Science, 32, 201-220 (2004).
- [8] P.Tiku-Kamga, P.Kamga, H.Imele; Journal of Food technology in Africa, 4, 48-50 (1987).
- [9] D.G.Libouga, C.N.Jiwoua Ngounou, C.P.Kouebou; Journal of Cameroon Academy of Science, 1, 15-20 (2001).
- [10] D.G.Libouga, J.J.Essia Ngang, H.Halilou; Sciences des aliments, 25, 53-66 (2005).
- [11] AOAC; Association of Official Analytical Chemists, 16<sup>th</sup> Edition, (1995).
- [12] J.F.Frank, E.H.Marth; Fermentations, N.P.Wong, R.Jenness, M.Keeney, E.H.Marth (Eds); Fundamentals dairy chemistry. 3<sup>th</sup> Edition. A Chapman & Hall Food Science Book, Gaithersburg, Maryland, USA, 655–738 (1999).
- [13] E.Renner; Milk and Dairy Products in Human Nutrition, W.Gmbh. Volkswirtschaftlicher Verlag, Munich, (1983).
- [14] A.Dahlqvist; Digestion of lactose, J.Delmont, S.Karger(Eds); Basel, Milk Intolerances and Rejection. New York, 11-16 (1983).
- [15] F.G.Elliot, J.Cox, B.L.Nyomba; Annales de la Société Belge de Médecine Tropicale, 53, 113–132 (1973).