## MILK COMPONENTS IN WHEY PREPARATIONS - CHARACTERISTICS AND UTILISATION

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Neutralised whey obtained after lactic fermentation and acidic coagulation of milk proteins carried out during tvarog (white cheese) production was concentrated selectively 3 times by means of membrane nanofiltration. The aim of this research was to identify properties obtained in this way in the industrial liquid whey preparation of milk components (WPMC) and utilisation of its natural water phase as a component of wheat dough. The main principles were that the use of the whey preparation will limit the amount of the technological water as a result of inclusion of this whey water phase and that the elimination or reduction of salt added during wheat dough formation will be possible. It was established that the acceptance of a slight salty taste in bakery products (rolls or bread) is possible by the addition of WPMC using both types of wheat flour (type 500 or type 750) except for the technological salt in the dough formation process. Wheat products with WPMC had a tendency to form the crumb characterised with small porosity and diminished volume. It was clearly noticed that incorporation of lactose doubled the content of disaccharides in crumb of experimental products. That phenomenon affords the opportunity for lactose supply as a substrate for the intestinal microflora both for children and adults. The participation of WPMC preparation, with the exception of technological salt, influenced changes in the proportion of individual elements, especially the proportion between sodium and potassium which accounted for 1:4, compared to the control sample where it was 3:1. The content of calcium and magnesium, whose active transport in a human body may proceed with hydrolysed lactose, could be increased significantly. This afforded the possibility for obtaining low-sodium products, being attractive special or prophylaxis food to be used in obesity, heart diseases, hypertension and as the support of the intestinal ecosystem.

#### INTRODUCTION

The acidic coagulation of milk proteins during the technological process of white cheese (tvarog) production leads to the output of the so-called "acid whey". Besides proteins, mainly whey protein, it contains a significant amount of lactose and lactic acid being products of lactic fermentation, and an increased amount of elements, especially of calcium phosphate. It is released from the casein micelles as a result of solubilisation and dissociation at pH 4.6, which constitutes the isoelectric point of coagulated casein proteins in the technological process of white cheese (tvarog) production. Thanks to the new selective separation technologies utilised under the industrial conditions, in recent years it has become possible to obtain a preparation of milk components as a result of concentrated "acid whey" due to the application of the modern technology of tvarog production. Thus, after the neutralisation of "acid whey" and its selective membrane separation - nanofiltration - a new halfproduct was obtained, avoiding the expensive dehydration. The application of membrane technologies allows decreasing the content of water in a whey preparation by 2/3, influences positively its stability and diminishes the costs of transportation. But the most important is the fact that partially deacidification and selectively demineralisation of whey characterised by a beneficial concentration of milk compounds and resembling its pH, could be an interesting material to be utilised in other branches of the food industry, *e.g.* baking.

The aim of the research was to characterise milk compounds in the form of condensed but liquid whey preparation and to explain whether the utilisation of valuable nutritional components: whey proteins, lactose and micro- and macroelements present in the water phase of milk, as wheat dough components is possible.

It was assumed that that the use of liquid whey preparation will permit to: (1) limit the amount of the technological water necessary to obtain the appropriate consistency of wheat dough, replacing it by the water phase of milk; and (2) eliminate or reduce the amount of added salt during technological wheat dough formation.

### MATERIAL AND METHODS

**Experimental material.** Material to be analysed comprised: milk components in the whey preparation commercial wheat flour type 500 and commercial wheat flour type 750.

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Analysis of chemical composition. The dry mass, contents of ash, proteins and lactose in liquid whey material were determined according to AOAC [1990] and PN-A--86364 [1996] methods.

The amount of lactose in bread crumb was determined according to Bagdach and Laskowski methods [Rutkowska, 1981] as follows:

The extraction of sugars was carried out from 10 g of the fragmented sample of bread in 150 mL of distilled water shaken for 5 min. The clarifying reagent (5 mL of 1 mol/L NaOH and 2.5 mL of 15% CuSO<sub>4</sub>) was added, the sample was supplemented to 200 mL and after shaking it was filtered. 50 mL of clarified filtrate was taken, 5 mL of HCl  $(d=1.19 \text{ g/cm}^3)$  was added and the sample was heated for 5 min at 70°C in order to carry out the inversion. After cooling, the sample was neutralised with 2.5 mol/L NaOH in the presence of methyl orange and it was supplemented with water to 100 mL. In accordance with sugar content of the sample, the appropriate volume of a solution was taken (when sugars ranged: 2-5%, 25 mL of solution was taken; 5.5–7% – 20 mL; 7.5–10% – 15 mL; 10.5–15% – 10 mL), and then the sample was supplemented to 25 mL. In the clarified filtrate at 20°C, an angle of rotation of polarised light was measured in a polarimeter. The content of lactose was calculated from the equation:

$$X \% = (a \times 1000)/(l \times 52.5)$$

where: a - an angle of rotation of polarised light; l - the length of polarimeter tube (dm); and 52.5 - an angle of rotation of polarised light for lactose.

The content of elements was determined by atomic absorption spectrometry with electrothermal atomization. Most of the elements were determined with the flame method using an Atomic Absorption Spectrometer Unicam 939 coupled with date disk ADAX, background correction and appropriate cathodic lamps. For the validation of calcium measurement, the solution of lanthanum chloride was added to all samples in the amounts assuring 0.5% concentration of La<sup>3+</sup>. Potassium and sodium were assayed with the photometric flame method (Flame Photometer 'Flapho 4'). The content of phosphorus was investigated with the colorimetric method by molybdate with hydroquinonate and sodium sulfate (IV).

Test laboratory baking. Dough for test baking was

obtained with the direct method from two types of commercial wheat flour (type 500 or type 750) with 10% of liquid whey preparation added (expressed in dry mass), and its water phase was used to obtain dough consistency of 350 B.U. An addition of 1% of technological salt was used only to control dough, while the experimental dough was without salt. After 1-h fermentation at 30–32°C, the baking was carried out at 200°C for about 20–30 min. The properties of bread were estimated according to PN-A-74108 [1996], and the experimental product was tested with an organoleptic test by 6-person pannel acc. to PN-89/A-74108.

#### **RESULTS AND DISCUSSION**

In the baking industry, liquid milk or milk powder were used, and even dry whey called "sweet" obtained during rennet cheese production, have been used for years as an additional component improving the nutritive value and sensory properties of bakery products. Due to nearly neutral pH of milk its use in the bakery technology does not rise any objections, whereas whey, especially that obtained during tvarog production as a result of lactic fermentation, is characterised by pH ranging from 4.4 to 4.6, a high level of lactic acid (0.4-0.6%) and mineral salts, mainly calcium phosphate (0.20-0.26%), which has a great technological significance [Oziemkowski, 1993]. About 50-60% of the dry mass of milk permeates to whey, including 20-25% of milk proteins and almost all "whey proteins" ( $\alpha$ -lactalbumins and  $\beta$ -lactoglobulins), the majority of lactose and mineral compounds [McIntosh et al., 1998]. The biological and nutritive value of these components may be improved by the functional properties of a whey preparation used during bread making [Soral-Smietana et al., 2005] or in confectionery production.

Because of the high acidity and low pH value the use of whey obtained during cheese production has been onerous so far. However, the development of new technologies allowed for neutralisation and selective membrane filtration of whey, which provides the opportunity for obtaining a preparation of modified chemical composition constituting about 17–18% d.m. at pH 6.3 (Table 1). The physicochemical analysis of the examined whey preparation of milk components (WPMC) showed that at density of ~1.08 g/cm<sup>3</sup> it contained nearly 14% d.m. of proteins, above 75% d.m. of lactose and nearly 5.5% d.m. of total mineral compounds. The high degree of concentration of these components

TABLE 1. The physicochemical properties of concentrated whey preparation (WPMC) after nanofiltration.

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Density(g/cm <sup>3</sup> )	pH	Dry mass	Proteins(Nx6.38)	Lactose	Ash
		(%)	(% d.m.)	(g/100 g) / (% d.m.)	(g/100 g) / (% d.m.)
$1.076 \pm 0.001$	6.30	$17.66 \pm 0.01$	$13.92 \pm 0.04$	$13.52 \pm 0.07$ 76.56 $\pm 0.07$	$0.96 \pm 0.01$ $5.42 \pm 0.01$

	TABLE 2.	Technological an	d qualitative	properties o	f experimental	baking products.
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Sample	Productivity of dough (%)	Crumb mass (g/cm <sup>3</sup> )	Volume (mL) 250 g 100 g		Porosity coefficient acc. to Dallman	
Control bread of wheat flour type 500	50.1	0.48	755	302	70	
Control bread of wheat flour type 500 with WPMC	50.3	0.46	645	258	80	
Control bread of wheat flour type 750	50.3	0.46	710	284	90	
Control bread of wheat flour type 750 with WPMC	50.2	0.48	605	242	80	

together with the appropriate level of mineral salts and pH creates the possibility of multidisciplinary application.

The studies were focused on the recognition and estimation of possibilities of the selectively concentrated milk components in a liquid form, as the components protecting mainly the water sorption capacity of both types of commercial wheat flour, type 500 or type 750.

In comparison with the control samples from both wheat flour types the experimental baking products, obtained from 250 g of dough, differed in the volume and in the porosity coefficient acc. to Dallman (Table 2). Although in general products obtained from flour type 500 reached larger volume, in the analysed flour the same tendency was kept when WPMC preparation was added. About 15% smaller volume of bread loaf was noticed in comparison with the control sample of both wheat flour types. Type independent and comparable was the crumb mass and porosity of the experimental samples, while in the laboratory research the influence of the whey preparation added on bread efficiency was not observed. The experimental products obtained were subjected to the organoleptic test executed by 6 persons. In a 5-degree scale both experimental breads analysed reached 4 points, demonstrating slight salty taste.

In the baking experiment it was established that the use of the WPMC preparation as a water phase in the technological process of wheat dough formation (flour type 500 or 750) allowed to arrange the added compounds through interactions and/or complex formation with wheat starch and gluten proteins (Table 3), during yeast fermentation and baking process. Although the content of proteins was not changed, the influence of milk proteins was manifested as deterioration of the gluten net structure, causing the reduction of the total or unit volume and small-porosity in crumb structure of the products with WPMC (Table 2). Similarly, changes in the total ash content caused by the WPMC presence in experimental bread crumb were indistinct, as compared with the control sample (Table 3). Established differences were provoked by the content of mineral compounds in both wheat flour types 500 and 750, *i.e.* 0.50% d.m. or 0.75% d.m., respectively. Therefore about 0.40% d.m. of both flour types resulted from the addition of salt (NaCl) supplemented for technological and taste-improving purposes. It should be noted that the total content of mineral compounds of the crumb with WPMC was similar, but about 0.40% of ash content was provided by elements occurrence in the WPMC preparation. Analysing the composition of individual elements from experimental material it was ascertained that the wheat flour type 750 contained significant amounts of potassium, phosphorus and magnesium (Table 4). While WPMC preparation, besides potassium, was characterised by almost physiological proportion of calcium to phosphorus. The elements characteristic for the crumb with the WPMC preparation showed a 5-fold increase in calcium content, as compared to the control crumb. Also the magnesium content was observed to increase by about 25%. But the most interesting was the sodium/potassium ratio which accounted for 1:4, compared to 3:1 reported for the control crumb.

Analysing the chemical composition of crumb with the addition of the WPMC preparation, in comparison with the control crumb, the content of disaccharides was determined (Table 3), that resulted from lactose incorporation, whose rich source in milk sugar was the whey preparation (Table 1). The sweetness of lactose reaches only 20% towards the saccharose, what is more, it is an important component of a well-balanced diet of humans. According to Piotrowska-Jastrzębska & Kierus-Jankowska [2005], lactose has a positive influence on the correct development of the sense of taste and appetite regulation in children. These properties constitute an important mechanism of overfeeding and obesity prevention in the early stage of organism development, a significant aspect of many civilization diseases, including decay. It is an important information towards the so-called "obesity epidemic" occurring also among children from many European countries. Taking into account numerous biological properties of lactose as well as its metabolic benefits for health and development Piotrowska-Jastrzębska & Kierus-Jankowska [2005] suggested that, according to current knowledge, lactose elimination from a healthy child's diet is unjustified.

The so-called acidic whey used for nanofiltration and obtained after the tvarog production was characterised by features resulting from milk acidic coagulation mechanism and revealed a high content of ash and lactic acid, which

TABLE 3. Chemical composition of experimental baking crumb.

Sample	Moisture (%)	Ash (% d.m.)	Proteins N x 5.7 (% s.m.)	Saccharides after inversion /Lactose (% d.m.)
Control bread crumb of wheat flour type 500	8.07±0.07	$0.94 \pm 0.05$	12.67±0.15	$1.59 \pm 0.01$
Bread crumb with WPMC of wheat flour type 500	$6.89 \pm 0.04$	$0.98 \pm 0.03$	$12.71 \pm 0.05$	$2.73 \pm 0.02$
Control bread crumb of wheat flour type 750	$7.73 \pm 0.03$	$1.13 \pm 0.02$	$13.29 \pm 0.05$	$1.45 \pm 0.2$
Bread crumb with WPMC of wheat flour type 750	$6.79 \pm 0.07$	$1.15 \pm 0.04$	$13.36 \pm 0.17$	$2.96 \pm 0.01$

TABLE 4. Content of elements in the experimental material.

Sample		Macroelements (mg/kg)				Microelements (mg/kg)			
	Mg	Ca	Р	K	Na	Mn	Fe	Zn	Cu
Wheat flour type 750	292	198	1451	2060	6	7.76	14.65	11.05	1.76
Whey preparation of milk compounds (WPMC)		1655	1114	2080	464	0.01	0.30	0.27	0.04
Control bread crumb of wheat flour type 750	330	255	1558	2360	7030	8.38	15.40	13.40	1.97
Experimental bread crumb of wheat flour type 750 + WPMC		1213	2000	3260	844	7.53	14.02	12.80	1.84

made its industrial utilisation and further processing impossible. The application of new technologies in the nanofiltration process allowed, on the one hand to remove these inconveniences, and on the other hand to obtain a concentrated product (to about 18% d.m.). This, in turn, increases its appeal, whereas the membrane selectivity influences the removal of a part of elements and lactose [Hassan, 1998]. According to Barrantes & Morr [1997] and Cheryan [1998], the composition and quality of whey can be modified in this way. As expected, the neutralisation influenced a decrease in lactic acid content by about 60% and in acidity as well, which indicates a change of pH. However, the use of membrane technology enabled suitable demineralisation, thus the limitation of ash content of the ready-made product. Irrespective of the order used, neutralisation and nanofiltration, whey concentrate shows very beneficial composition and physicochemical properties, e.g. elimination of salty taste, similarly as in the case of the so-called sweet whey obtained after enzymatic coagulation of milk proteins during the production of maturating cheese [Hansen, 1997; Barrantes & Morr, 1997]. The physicochemical properties of milk components applied in the whey preparation (WPMC) caused that it was used as an additive of natural components in baking products subjected to yeast fermentation. The beneficial reduction, to a larger extent of monovalent ions (sodium, potassium) than multi-valent ions (calcium, phosphorus), the presence of nutritional whey proteins, lactose and lactic acid encourage its use in the food industry, including bakery [Barba et al., 1998; Ye & Yoshida, 2000]. The whey preparation of milk components in the dry form has not only a high nutritive value, but also functional properties having a decisive effect on its usefulness in bakery. It has good capacity for water binding, emulsification and regulation of rheological properties [Zydney, 1998; Bednarski, 2001] as well as more specific physiological and therapeutic activities [Dallas, 1999].

The inclusion of WPMC as an additional component of crumb bread structure through gluten proteins and wheat starch, as a result of a modifying effect of technological processes [Soral-Śmietana *et al.*, 2001], could support their bioavailability. The mineral components complex through gluten proteins or wheat starch can be released in the gastrointestinal tract in a sequence of digestibility of both biopolymers, including also amylase resistant starch fraction. Lactose appearing in experimental products obtained in these investigations can facilitate the intestinal absorption of such scarce elements as calcium, magnesium, zinc, necessary for correct metabolism and physiology.

Based on the characteristic of the experimental baking products, it can be assumed that for good sensory properties of groceries of a daily consumption, it is possible to obtain a cereal product supplemented with valuable nutritional components originating from milk, improving a daily balance of scarce bioelements, especially calcium, magnesium and zinc. These elements are absorbed on the way of active transport and can be contributed by hydrolysed lactose. However, lactose – as a disaccharide consumed in wheat products – can function as microflora substrate for children or adults' intestines. That product in a diet provides a chance for a sequential availability of individual components in the gastrointestinal tract on account of different places of proteins and starch digestibility, which will affect their bioavailability. Apart from that, these products can play a role of low-sodium food necessary in special diets and arterial hypertension.

### CONCLUSIONS

The neutralisation and nanofiltration of tvarog resulted in the production of whey which, in turn, enabled obtaining a 3-times condensed whey preparation of milk components (WPMC) inlcuding proteins, lactose and elements at pH 6.3

The use of the whey preparation WPMC in the liquid form in the process of wheat dough formation allows to incorporate the milk water phase as a carrier of whey protein, lactose and selectively concentrated elements to the everyday products, permits omitting the addition of technological salt and positively modifies the contents of calcium, magnesium and the sodium to potassium ratio.

In the presence of good taste it is possible to obtain a cereal product supplemented with valuable nutritional components. It could improve the daily balance of the loss--making elements, especially calcium and magnesium, and fulfill the role of low-sodium food necessary in special diets and hypertension disease. At the same time, the presence of lactose, both in the form of disaccharide or hydrolysed, is of physiological importance in the gastrointestinal tract.

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# SERWATKOWY PREPARAT SKŁADNIKÓW MLEKA – CHARAKTERYSTYKA I ZASTOSOWANIE

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Stosując nanofiltrację membranową, neutralizowaną serwatkę po fermentacji mlekowej i kwasowej koagulacji białek mleka przy produkcji serów twarogowych zagęszczono selektywnie 3-krotnie. Celem badań było poznanie właściwości uzyskanego w ten sposób przemysłowego płynnego serwatkowego preparatu składników mleka (WPMC) oraz zastosowanie jego naturalnej fazy wodnej w roli komponentu ciasta pszennego. Poczyniono założenie, że stosowanie serwatkowego preparatu ograniczy ilość wody technologicznej w wyniku włączenia fazy wodnej oraz możliwe będzie pominięcie bądź zmniejszenie dodatku soli stosowanej podczas formowania ciasta pszennego.

Ustalono, że przy akceptowalności słabo wyczuwalnego smaku słonego w produktach piekarskich (bułki lub chleb), możliwe jest stosowanie WPMC do mąki pszennej obu asortymentów (typ 500 lub typ 750) z pominięciem soli technologicznej w procesie formowania ciasta. Produkty pszenne z WPMC miały tendencję do drobnoporowatości miękiszu i zmniejszonej objętości jednostkowej. Najwyraźniej odnotowano włączenie laktozy, która spowodowała podwojenie ilości dwucukrów w miękiszu produktów doświadczalnych. Tworzy to szansę dostarczenia laktozy jako substratu dla mikroflory w ekosystemie jelita, zarówno u dzieci jak i u dorosłych. Udział preparatu WPMC z pominięciem soli technologicznej wpłynął na zmianę proporcji sodu do potasu, z 3:1 w próbie kontrolnej do 1:4 w doświadczalnej. Znacząco zwiększyła się ilość wapnia i magnezu, w których aktywnym transporcie w organizmie człowieka może uczestniczyć hydrolizowana laktoza. Zatem powstaje szansa uzyskania produktu niskosodowego, atrakcyjnego jako żywność specjalna lub profilaktyczna do stosowania m.in. w otyłości, chorobach serca, w nadciśnieniu tętniczym oraz wspomagania ekosystemu jelita.