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THE RELATIONSHIP BETWEEN MILK AND MILK PRODUCTS CONSUMPTION AND BLOOD LIPID PROFILE IN WOMEN

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Abstract

Evidence from observational studies indicate that the consumption of milk products may have a neutral effect or may be inversely associated with the risk of cardiovascular disease, despite a high content of saturated fat of milk products. The association of milk and milk products intake with serum lipid profiles including serum triglycerides, total cholesterol, HDL cholesterol and LDL cholesterol was examined.

Intake of milk and milk product (yogurt, hard cheese, semi-hard cheese, soft cheese, cottage cheese) was assessed in 386 women aged 21 - 93 using food frequency questionnaire. Body height (BH), body weight (BW), % body fat (%BF) and waist circumference (WC) were measured. HDL cholesterol, total cholesterol, and serum triglycerides, were determined enzymatically (Olympus System Reagent) with a clinical chemistry analyser Olympus AU400. LDL-cholesterol was calculated by using Friedewald formula: LDL-cholesterol = total cholesterol - HDL-cholesterol - triglycerides/2.2 (mmol/L). All statistical analyses were performed using SPSS v. 17.0. The selected significance level was 95% ($p < 0.05$).

The results showed %BF and waist to hip ratio (WHR) were significantly lower in participants who consumed cheese (hard cheese, semi-hard cheese, soft cheese) vs. cheese non-consumers. HDL cholesterol concentrations were significantly higher in participants consuming hard cheese and soft cheese ($p < 0.01$). Participants who consumed more than 71.4 g (median value) of yogurt daily had significantly higher HDL cholesterol concentrations and lower triglycerides concentration ($p = 0.05$) than participants who consumed less than 71.4g of yogurt per day. Additionally, values for several anthropometric parameters were significantly lower in participants who consumed more than 71.4 g of yogurt per day (WHR - $p = 0.013$, WC - $p = 0.007$ and body mass index (BMI) - $p = 0.040$).

This study added to the body of evidence suggesting that milk products consumption, especially yogurt may have favourable effect on blood lipid profile and anthropometric parameters such as WHR/WC and BMI.

***Key words**: Serum HDL, Serum triglycerides, Milk and milk products consumption, Waist to hip ratio, Body mass index.*

1. Introduction

Cardiovascular diseases (CVDs) are still the biggest cause of deaths worldwide. Statistical data shows that more people die annually from CVDs than from any other cause and it is estimated that until year 2030 this number will grow significantly [1]. It is widely accepted that CVDs are, among others, associated with elevated blood levels of total cholesterol, low-density lipoprotein (LDL) and triglycerides as well as with low level of high-density lipoprotein (HDL) [2]. Serum lipid profile abnormalities are associated with a higher prevalence of CVDs (especially when low HDL and/or abnormal triglycerides levels added to abnormal LDL levels) and this condition is frequent in the elderly [3].

Because of the so high extent of mortality caused by CVDs and other metabolic diseases large amount of data and papers are published discussing concerns of fat intake, especially saturated, and its effect on the lipid profile.

One of the foodstuff that is stated as a rich source of saturated fat is milk [4]. Milk and milk products are complex foodstuffs containing a large number of bioactive compounds and are the rich source of protein, calcium and added nutrients, but milk fat has the largest impact on plasma lipids which depend on fat amount and concentration, same as on consumer characteristic (gender, age, metabolic capacity) [5].

Over past two decades there was a lot of study examining relationship between milk and dairy products intake and the risk of CVDs and stroke, because of the high contribution to the intake of saturated fat from dairy products, but data published are still not consistent [6, 7]. Most of the results published show no relationship or inverse association between CVDs and milk and dairy products intake [6]. One of the earlier prospective case-control study that investigated this relationship, and use food questionnaire as the method for estimation of milk intake, reported an inverse association between dairy fat intake and risk factors for CVDs including total cholesterol and triglycerides as well as body mass index (BMI) [8]. Interesting are the data of National Health and Nutrition Examination Surveys (NHANES) III study where was found that in women, more frequent cheese consumption was associated with higher HDL and lower LDL. Same study, but in men, established positive correlation between cheese consumption and both HDL and LDL same as with BMI and waist circumference (WC) [9].

Despite inconsistency within results it is recommended in Dietary guidelines that milk and dairy products consumption is important part of well-balanced diet and indicates higher consumption of dairy foods (including milk, cheese and yogurt) may help reduce the risk for CVDs, coronary heart disease (CHD) and stroke in a variety of populations.

Except saturated fat contain and its influence on serum lipid profile, preventive role of milk and milk products against different CVDs risk factors may be due to various mechanisms, because of essential nutrients content, such as calcium, potassium, phosphorus, vitamin D, various fatty acids and proteins, which play a role in the prevention [10, and 11].

The Statistical Yearbook of the Republic of Croatia for Year 2013 contains information about annual average quantities of food consumed in households per household member, and for food group Milk and milk products it is 75.7 L of milk, 7.3 kg of all types of cheese and 16.0 kg of other dairy products, what is right after the food group of cereals and cereal products [12]. Accordingly to amount of milk and milk products consumed among Croatian population it would be interesting to examine the correlation between risk factors (lipid profile and anthropometric characteristics) for CVDs and intake of the milk and milk products especially because mortality rate from CVDs among Croatian population is high (48.32% of total deaths in 2012 was caused by CVDs) [13] and because of missing data for our population.

This paper establishes the potential favourable effects of milk products, especially yoghurt, on blood lipid profile and anthropometric parameters such as WC and BMI.

2. Materials and Methods

The study was conducted at the University of Zagreb, in the Laboratory for Nutrition science, between years 2008 - 2011. A total of 386 women aged 21 - 93 were recruited trough public flyers, advertisements on health portal websites, and through personal acquaintances of researchers involved in the project. Dietary method used for assessment of milk and milk products (yoghurt and different types of cheese) intake was validated quantitative self-administered food frequency questionnaire (FFQ), composed of 15 food items with servings listed in three quantitative groups identified as small, medium, large represented with grams of food containing. Subjects were also asked to indicate the relative weekly frequency of use of each item. FFQ was previously validated in Italian population of adult women details previously described [14]. Servings of the dairy products were defined according to the US Food Guide Pyramid. The amounts of yogurt, milk, hard cheese, semi-hard cheese and soft cheese that count as a serving were 8 ounces (240 g), 1 cup (240 mL), 30 g, 45 g and 60 g, respectively [15].

Blood samples were collected from antecubital vein in the morning following an overnight 12 hours fasting for lipid profile analysis. Samples were collected into tubes without anticoagulant, centrifuged for 10 minutes at 3500 x g after what serums were separated and divided in three separate aliquots and stored at -20 ^{0}C until analysis were done. Measurements of serum cholesterol, HDL-cholesterol and triglycerides were measured at Olympus AU 400 (Olympus Life and Material Science Europa GmbH, Lisemeehan, Ireland) clinical chemistry analyser according to the manufacturer's instructions. Cholesterol was analysed directly by homogeneous enzymatic process, HDL-cholesterol by standard enzymatic method and triglycerides by colour enzymatic standard method using commercial reagents (Olympus system reagent 400, reagent kit, control serum and calibrator). LDL-choloesterol concentration was calculated from obtained data using the Friedewald equation: [LDL-cholesterol (mmol/L)] = [Total cholesterol] - [HDL-cholesterol] - ([Triglycerides]/2.2) [16].

Anthropometric measurements of subjects were performed at the time of blood drawing. Height and weight were measured according to standard instructions [17] at scale with attached stadiometer (Seca, Type 710 - 220, Vogel & Halke Gmbh & Co., Germany) where subjects were barefoot with minimal clothing. The height was measured with precision 0.1 cm, and body weight with precision 0.1 kg. BMI was calculated from obtained measures for each subject. Waist and hip circumference were measured with flexible, inelastic measuring tape with precision 0.1 cm. Measures were obtained while subjects were standing erectly, muscles relaxed at the level of the natural waist (horizontal plane at the top of right iliac crest) for WC and horizontal plane at the maximum circumference of the buttocks for the hip circumference. The waist-to-hip ratio (WHR) was calculated by dividing waist by hip circumference. Proportion and weight of body fat (BF) was measured with OMRON analyser model BF-300 (Omron Healthcare, Vernon Hills, IL, USA), whose operation principle is bioimpedance. All measurements were performed by trained personnel.

Statistical analysis was performed using SPSS statistical package (version 17.0, SPSS Inc., Chicago, IL, USA) and Microsoft Office Excel 2013 (Microsoft, Seattle, WA, USA). All subject was first divided in two group (consumer vs. non-consumers), and after that into two groups according to consumption of milk and milk products, using the median intake as the cut-off point for each product. Difference in mean values of selected parameters was tested by Student's t test, with p values < 0.05 were considered as significant. Chi-square test was used to detect any significant differences in the distribution of subjects across quartiles of dairy intake with regard to anthropometric and biochemical variables. All data were presented as a mean ± SD or percentage.

3. Results and Discussion

Subjects included in final analysis sample were 386 Croatian women, mean age 54.8 ± 16.3 years. Mean BMI of the specified subjects was 26.5 ± 4.9 kg/m^2, with 57% of overweight or obese subjects. Waist circumference higher than 88 cm (defined as high-risk waist circumference [18]) was found in 43% of all subjects. Figure 1 shows presence of lipid profile parameters as a selected cardiovascular risk factors among observed women. Mean age of subjects in "no risk" group was 46.0 ± 16.4 years, while the mean age of the subjects in group with inadequate status of all observed lipid profile parameters was 56.2 ± 15.8 years. The difference in the mean age between risk groups (p<0.001) was expected because the risk of CVD shows a linear increase with age [19].

The highest consumption rate of all observed dairy products was found for yogurt and similar fermented dairy products, i.e. 90% of subjects reported consuming yogurt (Figure 2).

Among consumers average daily intake of milk was 182.7 ± 134.9 g, while average yogurt intake was 103.7 ± 76.9 g.

Subjects were divided into two groups according to consumption of milk and dairy products (consumers and non-consumers). There were no observed significant differences between groups according to anthropometric and biochemical parameters for yoghurt and milk consumption but were among consumption of all cheese subgroups what is shown in Table 1. In this study body fat percentage and waist to hip ratio (WHR) were significantly lower in subjects who consumed cheese (hard cheese, semi-hard cheese, soft cheese) compared to non-consumers. HDL cholesterol concentrations were significantly higher in subjects consuming hard cheese and soft cheese (p < 0.01) compared to non-consumers (Table 1) what is in accordance to study of Houston *et al.* [9] which found that cheese consumption was associated with a more favourable cardiovascular lipid profile in women.

The mean intake of cheese among consuming group of subjects was 25.0 ± 20.8 g or 0.5 ± 0.4 servings per day, but when analysing the data we need to take into account that there are differences in quantity intakes according to type of the cheese consumed. Still, it is not clear weather consuming cheese, regardless of the fat content, may be associated with other food choices that may either enhance or neglect the relationship of the food and CVD risk factors, or more important are other cheese constituents, such as calcium and conjugated linoleic acid, which may modulate cheese effect on CVD risk factors [9]. Huth *et al.* recently concluded that further work should be conducted regarding the mechanism behind the blood lipid neutrality of cheese [6].

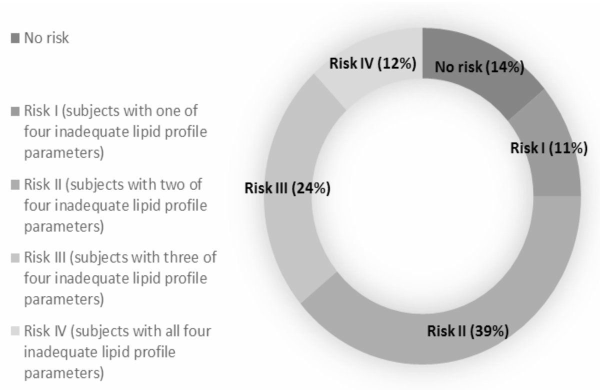

Figure 1. Distribution of subjects (n = 386) according to inadequate lipid profile parameters (total cholesterol, triglycerides, HDL and LDL cholesterol)

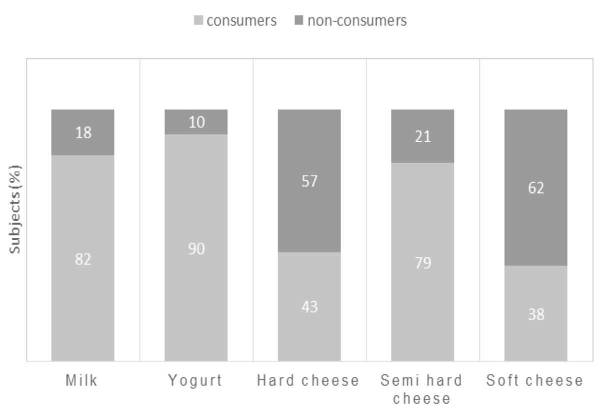

Figure 2. Distribution of subjects (n=386) according to consumption of milk and milk products

Table 1. Anthropometric and biochemical parameters according to cheese consumption

Parameters	Hard cheese			Semi hard cheese			Soft cheese		
	Consumers	Non-consumers	P*	Consumers	Non-consumers	P*	Consumers	Non-consumers	P*
Triglycerides (mmol/L)	1.25 ± 0.74	1.41 ± 0.93	ns	1.32 ± 0.86	1.42 ± 0.84	ns	1.29 ± 1.04	1.38 ± 0.72	ns
Total cholesterol (mmol/L)	5.67 ± 1.27	5.64 ± 1.11	ns	5.67 ± 1.18	5.62 ± 1.15	ns	5.59 ± 1.34	5.70 ± 1.06	ns
HDL cholesterol (mmol/L)	1.38 ± 0.34	1.25 ± 0.38	<0.001	1.32 ± 0.37	1.27 ± 0.37	ns	1.37 ± 0.33	1.27 ± 0.38	0.006
LDL cholesterol (mmol/L)	3.76 ± 1.08	3.75 ± 1.04	ns	3.75 ± 1.07	3.79 ± 1.03	ns	3.68 ± 1.14	3.80 ± 1.01	ns
Body weight (kg)	71.01 ± 13.34	71.54 ± 12.63	ns	71.49 ± 13.06	70.64 ± 12.47	ns	71.77 ± 13.97	71.03 ± 12.27	ns
Body fat (%)	31.68 ± 7.49	36.77 ± 8.14	<0.001	34.06 ± 8.18	36.34 ± 8.32	0.030	32.29 ± 7.90	35.93 ± 8.17	<0.001
Body fat (kg)	23.30 ± 9.37	26.89 ± 9.15	<0.001	25.05 ± 9.44	26.28 ± 9.29	ns	24.04 ± 9.92	26.10 ± 9.00	0.039
Waist (cm)	83.22 ± 12.17	89.43 ± 13.10	<0.001	86.12 ± 13.03	89.11 ± 12.98	ns	84.08 ± 13.08	88.37 ± 12.81	0.002
Hips (cm)	105.11 ± 9.68	107.73 ± 9.90	0.010	106.52 ± 9.81	106.93 ± 10.20	ns	105.76 ± 9.99	107.11 ± 9.80	ns
Body mass index (kg/m²)	25.54 ± 4.86	27.28 ± 4.78	<0.001	26.39 ± 4.89	27.01 ± 4.89	ns	26.01 ± 5.15	26.84 ± 4.70	ns
Waist to hip ratio	0.79 ± 0.08	0.83 ± 0.08	<0.001	0.81 ± 0.08	0.83 ± 0.08	0.011	0.79 ± 0.08	0.82 ± 0.08	<0.001

*ns - not significant, statistically significant at $p < 0.05$.

As there was no difference between anthropometric and biochemical parameters among groups according to the consumption of the milk and yoghurt, subjects were divided according to median consumption of milk (100 g/day). There were neither significant difference in the concentration of the all observed blood lipid parameters nor in the selected anthropometric parameters beside body fat percentage ($p = 0.037$) (Table 2). Increased dairy products consumption in the diet without energy restriction might not lead to a significant change in weight or body composition; whereas inclusion of dairy products in energy-restricted weight loss diets can significantly affects weight, body fat mass, lean mass and WC compared to the usual weight loss diets [20].

In our study milk consumption was not considered according to fat content, and more detailed classification might have influence on the results because many studies showed negative relationships between milk with reduced fat content and serum triglycerides [8, 21 to 23]. Still there is no consistent evidence that higher intake of milk or dairy products, regardless of milk fat levels, are associated with an increased risk of cardiovascular disease [24 to 26]. Recent meta-analysis of randomized studies with healthy adults randomized to increased dairy food more than one month without additional interventions, showed no significant changes in waist circumference, LDL-cholesterol, HDL-cholesterol and with no or minor effects on other cardio-metabolic risk factors [27].

In this study subjects with yogurt consumption greater than 71.4 g/day had significantly lower waist circumference, WHR and BMI, same as marginally significantly lower concentration of triglycerides and higher concentration of HDL cholesterol, compared to subjects consuming less than specified amount (Table 2). Yogurt is a good source of several micronutrients and may help to improve diet quality and maintain metabolic well-being as part of a healthy, energy-balanced dietary pattern. In recent research, yogurt consumption was associated with lower levels of circulating triglycerides [28]. Favourable effect of yogurts on plasma lipids and lipoproteins appear to be strain specific [5].

Table 2. Anthropometric and biochemical parameters according to median consumption of milk and yogurt

Parameters	Milk consumption			Yogurt consumption		
	≤ 100 g/day (n = 210)	> 100 g/day (n = 176)	P*	≤ 71.4 g/day (n = 195)	> 71.4 g/day (n = 191)	P*
Triglycerides (mmol/L)	1.29 ± 0.67	1.42 ± 1.03	ns	1.42 ± 0.98	1.26 ± 0.70	0.053
Total cholesterol (mmol/L)	5.69 ± 1.20	5.62 ± 1.15	ns	5.59 ± 1.04	5.73 ± 1.30	ns
HDL cholesterol (mmol/L)	1.34 ± 0.37	1.27 ± 0.36	ns	1.27 ± 0.40	1.34 ± 0.33	0.050
LDL cholesterol (mmol/L)	3.77 ± 1.09	3.74 ± 1.04	ns	3.66 ± 0.97	3.85 ± 1.14	ns
Body weight (kg)	71.66 ± 14.14	70.89 ± 11.34	ns	72.16 ± 13.79	70.44 ± 11.96	ns
Body fat (%)	33.71 ± 7.90	35.48 ± 8.57	0.037	35.29 ± 8.36	33.73 ± 8.08	ns
Body fat (kg)	24.86 ± 9.78	25.83 ± 8.96	ns	26.13 ± 9.67	24.46 ± 9.07	ns
Waist (cm)	86.40 ± 13.33	87.19 ± 12.76	ns	88.52 ± 13.14	84.94 ± 12.76	0.007
Hips (cm)	106.68 ± 10.19	106.51 ± 9.53	ns	107.44 ± 10.30	105.74 ± 9.37	ns
Body mass index (kg/m²)	26.46 ± 5.14	26.60 ± 4.58	ns	27.03 ± 5.15	26.01 ± 4.56	0.040
Waist to hip ratio	0.81 ± 0.08	0.82 ± 0.08	ns	0.82 ± 0.08	0.80 ± 0.08	0.013

*ns - not significant, statistically significant at $p < 0.05$.

Distribution of subjects in quartiles according to consumption of dairy products showed greater percentage of subjects with enlarged waist circumference in the lowest quartile of milk consumption, and the lowest percentage of subjects with enlarged waist circumference in the highest quartile of milk consumption, although the difference was not significant. The similar association was found for the BMI, serum triglycerides, total serum cholesterol and serum LDL-cholesterol in relation to dairy intake distributed in quartiles (Table 3).

4. Conclusions

- This study added to the body of evidence suggesting that milk products consumption, especially yogurt may have favourable effect on blood lipid profile and anthropometric parameters.

- The future study should address questions whether the dairy products intake, taking into account milk fat content, would have same impact regardless baseline lipid profile levels and to clarify mechanism behind beneficial effect of specific dairy products.

Table 3. Distribution of subjects (%) with inadequate anthropometric and biochemical parameters across different quartile cut-offs of dairy intake[a]

	Quartile of dairy intake				
Parameters (% of subjects)	Q1 (n = 105)	Q2 (n = 94)	Q3 (n = 91)	Q4 (n = 96)	P[b]
Enlarged waist circumference[c]	48	39	47	38	0.341
Body mass index > 25 kg/m²	63	55	61	52	0.379
High serum triglycerides[d]	78	71	78	73	0.598
High serum total cholesterol[e]	29	21	18	18	0.214
Low serum HDL cholesterol[f]	47	36	49	39	0.190
High serum LDL cholesterol[g]	73	67	72	70	0.758

[a]Cutoffs were 0.8, 0.8 to 1.40, 1.41 to 1.90, and 1.91 servings/d for quartiles 1 - 4, respectively.
[b]P for differences among dairy quartiles (chi-square test).
[c]Enlarged waist circumference > 88 cm.
[d]Defined as > 1.7 mmol/L.
[e]Defined as > 5 mmol/L.
[f]Defined as < 1.2 mmol/L.
[g]Defined as > 3 moll/L.

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