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ORIGINAL ARTICLE

Content of fatty acids in a diet and the homocysteine levels in women with fertility disorders

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Abstract

OBJECTIVES: Assessment of the effects of consumption of fatty acids on the homocysteine levels in women with fertility disorders.
METHODS AND RESULTS: The study included 286 women at the age between 23 and 46 years (the mean 33.13±4.21 years) with a fertility disorders. We measured: levels of homocysteine [μmol/L] (n=171), body weight and height (n=286). Body mass index (BMI) was calculated. A diet was assessed by 3-day dietary food records method including one day of the weekend. The mean homocysteine (Hcy) levels were 10.02±2.98 μmol/L. Body weight excess was observed in 29.3% of subjects. The percentage of fatty acids in the total energy [%E] it was demonstrated that the mean rate of MUFAs and PUFAs was statistically significantly lower in diets of women with the homocysteine levels >15 μmol/L compared to the group with the Hcy <10 μmol/L. Based on the results of a correlation between the homocysteine levels and consumption of n-6 fatty acids and n-3 fatty acids it is possible to conclude that there are negative correlations observed indicating that higher consumption of these acids is associated with lower levels of homocysteine.
CONCLUSION: Increased consumption of PUFAs, including α-linoleic acid, in a diet seems to be an important factor preventing from hyperhomocysteinemia.

Abbreviations:

ALA	- α-linolenic Acids	MUFA	- Monounsaturated Fatty Acids
DHA	- Docosahexaenoic Acid	PAI-1	- Plasminogen Activator Inhibitor 1
EPA	- Eicosapentaenoic Acid	PCOS	- Polycystic Ovary Syndrome
FPIA	- Fluorescence Polarization Immunoassay	PPAR-γ receptor	- Peroxisome proliferator-activated receptor gamma
HDL	- High Density Lipoprotein	PUFA	- Polyunsaturated Fatty Acids
Hcy	- Homocysteine	RDA	- Recommended Dietary Allowance
IR	- Insulin Resistance	SFA	- Saturated Fatty Acids
LA	- Linoleic Acid	WHO	- World Health Organization

INTRODUCTION

The normal levels of homocysteine (Hcy) in the blood plasma are 5–15 µmol/L; however, reports have demonstrated that even levels of 10–13 µmol/L have a detrimental effect on the vascular endothelium (Gąsiorowska *et al.* 2008; Winczewska-Wiktor *et al.* 2012). A group of patients who are at risk of increased homocysteine levels includes the following: diagnosed with a cardiovascular disease (e.g. myocardial infarction, coronary heart disease, atherosclerotic lesions in the carotid arteries), with increased risk of vascular diseases (e.g. arterial hypertension, cigarette smoking, diabetes mellitus, history of cardiovascular diseases) and those who may be at risk of vitamin group B deficiency (e.g. vegetarians, elderly patients, those with alcohol problem, with an unbalanced diet) (Kraczkowska *et al.* 2005).

A healthy lifestyle, including a healthy diet, has a significant effect on the normal levels of homocysteine in the body. A diet low in vitamins B_6, B_{12} and folic acid with high levels of protein has been assumed to play a special role in the aetiology of hyperhomocysteinemia (Kapka-Skrzypczak *et al.* 2012). Group B vitamins are necessary elements of the process aimed to remove homocysteine that involves its transformation into cysteine or methionine. Folic acid is especially important as its deficiency can be observed the most frequently because it cannot be synthesised endogenously and its absorption is incomplete (Łubińska *et al.* 2006).

It has been proven that high levels of homocysteine in the follicular fluid in the ovary may result in impaired interactions between the egg cell and sperm, resulting in a lower chance of fertilisation. Prolonged hyperhomocysteinemia also leads to impaired implantation of a fertilised egg and placental blood supply. The effects of vitamin B_6 on the normal course of pregnancy and a reduced number of miscarriages are associated with maintaining appropriate levels of progesterone and low levels of homocysteine. Additionally, a diet rich in vitamin B_{12} and folic acid reduces the risk of infertility due to ovulation disorders (Gąsiorowska *et al.* 2008; Kraczkowska *et al.* 2005; Łubińska *et al.* 2006).

Based on the current knowledge the quality and amount of lipids consumed may be a risk factor of numerous diseases including, among others, cardiovascular diseases, obesity, cancer and diabetes mellitus (Lottenberg *et al.* 2012). Monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs) are assumed to have beneficial effects on health. The effects of other fatty acids are also thought to be significant with regard to women fertility (Chavarro *et al.* 2007a, Chavarro *et al.* 2007b). A lipotoxic theory presenting development of insulin resistance that has been formed within the last dozen years and reduced activity of the peroxisome proliferator-activated receptor gamma (PPAR-γ receptor) seem to be important for this relationship. PUFAs are suggested to reduce the risk of infertility and to improve fertility as they promote insulin sensitivity and reduce inflammation (Chavarro *et al.* 2007a). Moreover, polyunsaturated fatty acids control development and metabolism of the fatty tissue probably via eicosanoides, especially prostaglandins. n-3 acids are thought to have antiadipogenic properties, whereas n-6 acids promote development of the fatty tissue (Grabiec *et al.* 2012). Moreover, the role of PUFAs as ligands of receptors activated by peroxisome proliferators such as PPAR-γ seems to be important (Chavarro *et al.* 2007a). Natural ligands of PPAR-γ receptors include arachidonic acid, linoleic acid, EPA and DHA, with decreasing affinity (Skoczyńska 2011).

Moreover, it seems that diet supplementation with n-3 PUFAs in women with fertility disorders diagnosed with polycystic ovary syndrome (PCOS) may prevent from cardiovascular diseases and metabolic diseases that often coexist with PCOS (Kilicdag *et al.* 2005). A Mediterranean diet with predominance of monounsaturated fatty acids (MUFAs) may also be beneficial in women with PCOS. This hypothesis is based on observations that obesity and insulin resistance are observed more rarely in Italian women compared to American women diagnosed with PCOS (Kowalik & Rachoń 2014).

The aim of the study was assessment of the effects of consumption of fatty acids on the homocysteine levels in women with fertility disorders.

METHODS

The study included 286 women at the age between 23 and 46 years (the mean 33.13±4.21 years) with a medical history of miscarriages, recurrent miscarriages (≥2 subsequent miscarriages), polycystic ovary syndrome and/or problems with getting pregnant (no pregnancy after at least a year of spontaneous sexual intercourses without contraception).

In all women, the body weight and height were measured and the body mass index (BMI) was calculated [kg/m^2]. The results were interpreted according to the guidelines of the World Health Organization (WHO) (WHO 2016).

A diet was assessed by 3-day dietary food records method including one day of the weekend. Consumption of nutrients (namely energy, macronutrients – including a profile of fatty acids, vitamins B_6, B_{12}, folic acid) was assessed with the following software: Dieta 5.0.

Moreover, the homocysteine levels were also assessed (n=171) with the fluorescence polarisation immunoassay (FPIA – Fluorescence Polarization Immunoassay). Patients were divided into 3 groups depending on their homocysteine levels: group 1: <10 µmol/L; group 2: 10–15 µmol/L; group 3: >15 µmol/L.

The statistical analysis was performed with IBM SPSS Statistics. For continuous variables descriptive statistics (minimum, maximum, mean and standard deviation) were calculated, and the categorical variables were presented as the frequency and percentage of individual categories per variable. The analysis included 95% confidence intervals for means and the r-Pearson parametric correlation coefficient. The p-value <0.05 was assumed as the level of statistical significance.

These observational studies are a part of the study approved by the Research Ethics Committee of Military Institute of Medicine in Warsaw (No. 12 / WIM / 2012).

RESULTS

The age of patients was 23–46 years. Women at the age of 35–40 years accounted for 30.1% of all subjects, and above the age of 40 years – 4.5%. The mean homocysteine (Hcy) levels were 10.02±2.98 µmol/L. The majority (n=252) of patients declared they did not smoke. The mean body weight index was 23.86±4.55 kg/m^2 (Table 1). Body weight excess was observed in 29.3% of subjects, and obesity in 10.1% of women. Almost half of subjects (47.1%) experienced at least two miscarriages. Polycystic ovary syndrome was diagnosed in 57% of women (Table 2).

The mean energetic value of a diet was 1822.68±531.6 kcal. The mean consumption of folates was 66.6% RDA (RDA=400 µg/d), and consumption of vitamins B_6 and B_{12} was above the norm for this age group (RDA=1.3 mg and 2.4 µg/d, respectively). Additionally, the mean levels of animal protein were higher than of the plant protein. The mean proportion of n-6 and n-3 fatty acids was 5.51±3.09 (Table 1).

The rate of women with the Hcy levels >10 µmol/L was 26.5% (Tab. 3). There were no statistically significant differences ($t(135)=0.645$; $p=0.52$) between the levels of homocysteine among women with PCOS (mean: 10.23 µmol/L; 95%CI: 9.61–10.84 µmol/L) and without this syndrome (mean: 9.93 µmol/L; 95% CI:

Tab. 1. Descriptive statistics of the study group: anthropometric details, plasma homocysteine levels, consumption of selected nutrients.

Descriptive statistics	N	Minimum	Maximum	Mean	Standard deviation
Age [years]	286	23	46	33.13	4.21
BMI [kg/m^2]	286	16.6	41.2	23.86	4.55
Hcy [µmol/L]	175	4.40	18.70	10.02	2.98
Cigarette smoking [pcs/week]	285	0	140	3.49	16.75
		DIET			
Energy value [kcal]	280	751.69	4330.68	1822.68	531.60
Folates [µg]	279	79.04	739.27	266.47	93.18
Vitamin B_6 [mg]	279	0.66	3.96	1.99	0.65
Vitamin B_{12} [µg]	279	0.25	80.98	3.55	5.21
Total protein [g]	280	28.34	153.58	79.64	22.52
Animal protein [g]	279	0.16	130.92	54.21	20.87
Plant protein [g]	279	3.19	70.84	24.15	8.22
Methionine [mg]	279	504.67	3986.35	1878.77	593.87
Fat [g]	280	15.21	230.40	69.04	30.08
SFA [g]	279	3.95	98.36	27.04	14.72
MUFAs [g]	279	4.52	98.92	26.82	12.41
PUFAs [g]	279	1.17	36.77	9.86	5.16
Linoleic acid (18:2)	279	0.78	30.97	7.82	4.28
Arachidonic acid (20:4)	279	0.00	0.59	0.10	0.10
Dihomogammalinoleic acid (20:3)	279	0.00	0.00	0.00	0.00
Linolenic acid (18:3)	279	0.31	12.56	1.65	1.35
Eicosapentaenoic acid (20:5)	279	0.00	1.43	0.06	0.16
Docosahexaenoic acid (22:6)	279	0.00	3.14	0.13	0.37
n-6 : n-3	279	0.59	26.19	5.51	3.09
Cholesterol [mg]	279	0.00	767.13	241.64	118.20
Total carbohydrates [g]	280	63.58	549.08	237.07	80.37

BMI – body mass index; Hcy – homocystein; SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids

Tab. 2. Number and percent of women depending on the interpretation of BMI [kg/m^2], number of miscarriages and diagnosis of PCOS.

Variable		Frequency	Percentage
BMI [kg/m^2]	underweight	11	3.8
	normal weight	191	66.8
	overweight	55	19.2
	obese class I	19	6.6
	obese class II	8	2.8
	obese class III	2	0.7
	Total	286	100.0
Number of miscarriages	0	93	32.5
	1	55	19.2
	2	64	22.4
	3	51	17.8
	4	15	5.2
	5	2	0.7
	No data	6	2.1
	Total	280	97.9
PCOS	YES	163	57
	NO	123	43
	Total	286	100.0

BMI – Body Mass Index; PCOS – Polycystic Ovary Syndrome

9.24–10.62 μmol/L). There was no linear correlation between the number of cigarettes smoked and the Hcy levels ($r(173)=0.041$, $p=0.59$).

In groups that varied with regard to the homocysteine levels the degree of consumption of total fat, saturated fatty acids, monounsaturated fatty acids and polyunsaturated fatty acids, including n-6 and n-3, was compared. The mean consumption of total fat was significantly lower in the group with the homocysteine levels >15 μmol/L compared to two other groups. Similar results were obtained for monounsaturated fatty acids (the mean consumption in the group with high homocysteine levels was significantly lower compared to two other groups). With regard to polyunsaturated fatty acids all groups under comparison were significantly different – higher levels of homocysteine were associated with lower consumption of this type of fat. No differences were observed for saturated fatty acids (SFA) only. With regard to percentage of fatty acids in the total energy [%E] it was demonstrated that the mean rate of MUFAs and PUFAs was statistically significantly lower in diets of women with the homocysteine levels >15 μmol/L compared to the group with the lowest levels of Hcy (<10 μmol/L), as follows: 10.68% vs. 13.81% and 3.36% vs. 5.34%. The percentage of fatty acids belonging to n-6 and n-3 fatty acids in the energy value of a given diet was significantly higher in diets of women with the lowest Hcy levels compared to other groups (1.03% vs. 0.86% and 0.47%) (Table 4).

A correlation between the homocysteine levels and the content of individual fatty acids in a diet was studied. Significant negative correlations were observed between the Hcy levels and the content of polyunsaturated fatty acids ($r(169)=-0.253$; $p<0.05$) and monounsaturated fatty acids ($r(169)=-0.127$; $p<0.05$) in a diet (Table 5).

Additionally, a correlation indicating a significant, negative relationship between the homocysteine levels and consumption of 18:02 fatty acid (linoleic acid, LA) was demonstrated ($r(169)=-0.218$; $p<0.05$). Higher consumption of this acid was associated with lover levels of homocysteine. A similar correlation was observed for the consumption of 18:3 acid (linolenic acid) ($r(169)=-0.216$; $p<0.05$). Based on the results of a correlation between the homocysteine levels and consumption of n-6 fatty acids ($r(169)=-0.219$; $p<0.05$) and n-3 fatty acids ($r(169)=-0.225$; $p<0.05$) it is possible to conclude that there are negative correlations observed indicating that higher consumption of these acids is associated with lower levels of homocysteine (Table 6).

DISCUSSION

Homocysteine (Hcy) is an amino acid and its abnormal levels are associated with many diseases. Its elevated levels are observed in the course of the following conditions: hepatic and renal failure, diabetes, hypothyroiditis, or Cushing's disease. The effects of homocysteine on development of mental disorders (depression, parkinsonism, schizophrenia) are probably due to its direct neurotoxic properties and direct effects including promotion of vascular lesions. The majority of reports confirm a correlation between homocysteine and development of cardiovascular diseases. It has been emphasised that it is an independent risk factor for such diseases. Hyperhomocysteinemia has a negative effect on reproduction (Łubińska *et al.* 2006). Results of many studies also indicate a correlation between elevated levels of Hcy, deficiency of folic acid and complications during pregnancy. Its high levels in the ovarian follicular fluid may impair interactions between sperm and egg cells resulting in reduced chances of fertilisation. Moreover, it affects an early period of implantation and embryogenesis and it impairs blood supply to the placenta and its functions leading to pregnancy loss, inhibited foetal development or congenital defects in a child. It has been demonstrated that homocysteine has direct teratogenic properties and it is a risk factor for recurrent miscarriages and early deliveries (Kraczkowska *et al.* 2005; Magnucki *et al.* 2009). A correlation between recurrent pregnancy loss and PCOS has been suspected but the study results are not unanimous (Amer *et al.* 2002; Ford & Schust 2009; Chakraborty *et al.* 2013). A high incidence of obesity and hyperinsulinemia in women with polycystic ovary syndrome is

Tab. 3. Number and rate of women assigned to groups depending on the homocysteine levels [µmol/L].

Homocysteine levels [µmol/L]		Frequency (%)				
		All	PCOS	No PCOS	Cigarette smokers	Non-smokers
Important	<10	99 (34.6)	58	41	14	85
	10–15	67 (23.4)	40	27	7	60
	>15	9 (3.1)	7	2	0	9
	Total	175 (61.2)	105	70	21	154
No data		111 (38.8)				
Total		286 (100)				

PCOS – Polycystic Ovary Syndrome

Tab. 4. Means and confidence intervals (95% CI) for total fat, SFAs, MUFAs, PUFAs depending on the Hcy levels.

Hcy levels [µmol/L] (n=171)	Energy [kcal]	Mean (confidence interval (95%CI))					
		Total fat [g] [%E]	SFA [g] [%E]	MUFA [g] [%E]	PUFA [g] [%E]	n-6 [g] [%E]	n-3[g] [%E]
<10 (n=96)	**1800.79** (1703.28–1898.29)	**70.93** (64.99–76.87)	**26.56** (23.72–29.40)	**27.84** (25.45–30.23)	**10.67** (9.55–11.78)	**8.52** (7.62–9.41)	**2.09** (1.73–2.46)
		34.47 (32.58–36.37)	**13.39** (12.24–14.55)	**13.81** (12.93–14.70)	**5.34** (4.87–5.81)	**4.27** (3.89–4.66)	**1.03** (0.87–1.19)
10–15 (n=66)	**1857.38** (1716.78–1997.97)	**69.87** (62.36–77.38)	**28.21** (24.29–32.13)	**26.84** (23.79–29.88)	**9.1** (7.99–10.21)	**7.36** (6.47–8.25)	**1.70** (1.41-1.99)
		32.75 (30.74–34.76)	**13.25** (12.21–14.29)	**12.99** (11.93–14.06)	**4.48** (4.02–4.95)	**3.60** (3.23–3.96)	**0.86** (0.70–1.02)
>15 (n=9)	**1713.82** (1348.30–2079.34)	**52.72** (39.49–65.95)	**23.16** (15.88–30.45)	**19.57** (14.05–25.09)	**5.87** (4.58–7.16)	**5.03** (3.81–6.25**)**	**0.84** (0.67–1.00**)**
		28.26 (21.31–35.20)	**12.44** (9.01–15.87)	**10.68** (7.64–13.72)	**3.36** (2.21–4.51)	**2.89** (1.85–3.92)	**0.47** (0.32–0.62)

E – energy; Hcy – homocystein; SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids

Tab. 5. Correlation coefficients for the relationship: the blood homocysteine levels and the content of SFAs, MUFAs and PUFAs in a diet.

		SFA [g]	MUFA [g]	PUFA [g]
Homocysteine	Pearson's correlation	0.032	–0.127	–0.253
	Significance (unilateral)	0.341	0.049	0.000
	N	171	171	171

SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids

Tab. 6. Correlation coefficients for the relationship: the blood homocysteine levels and the content of individual polyunsaturated fatty acids.

		18:2 [g]	18:3 [g]	20:3 [g]	20:5 [g]	22:6 [g]	n-6	n-3
Homocysteine	Pearson's correlation	–0.218	–0.216	–0.108	–0.072	–0.092	–0.219	–0.225
	Significance (unilateral)	0.002	0.002	0.079	0.176	0.117	0.002	0.002
	N	171	171	171	171	171	171	171

more often indicated as a risk factor for spontaneous miscarriages (Wang *et al.* 2002). Hyperinsulinemia is a reason for some fertility disorders probably due to its effects on the production of androgens. However, many reports indicate that insulin resistance (IR) is an element linking PCOS/obesity and recurrent pregnancy loss (Tian *et al.* 2007; Maryam *et al.* 2012). Moreover, many reports emphasise a possible correlation between IR and hyperhomocysteinemia, and it is more and more often observed in women with PCOS (Schachter *et al.* 2003; Wijeyarante *et al.* 2004). The latest reports confirm that reduced processes of fibrinolysis (hypofibrinolysis) associated with plasminogen activator inhibitor-1 (PAI-1) in women with PCOS may result in pregnancy loss (Sun *et al.* 2010; Gosman *et al.* 2006). Increased levels of PAI-1 may also affect increased values of homocysteine, finally leading to thrombosis. Additionally, the serum PAI-1 levels are associated with dyslipidaemia, hyperinsulinemia and arterial hypertension that are associated with high levels of Hcy (Chakraborty *et al.* 2013; Bastard *et al.* 2000).

Due to its multidirectional properties Hcy affects many processes in the body, therefore it is an interesting subject of numerous studies. This paper analysed the levels of this amino acid in women with fertility disorders such as recurrent miscarriages, polycystic ovarian syndrome, and a correlation of its levels with consumption of selected nutrients, including especially unsaturated fatty acids. Recurrent pregnancy loss and PCOS were observed in approximately half of patients (in 46.1% and 57%, respectively).

Normal Hcy levels are 5–15 mmol/L; however, according to some reports levels as low as 10–13 mmol/L might already have adverse effects on the vascular endothelium, therefore the reference value range should be determined individually for various populations according to expert recommendations (Gąsiorowska *et al.* 2008). Pregnant women usually have lowered Hcy levels that rarely exceed 10 mmol/L; nonetheless, even when the levels are slightly increased this increase is associated with an increased risk of complications presented earlier (early miscarriage, early delivery, eclampsia, placental circulatory disturbances, low birth weight) (Łubińska *et al.* 2006; Magnucki *et al.* 2009). In the own paper the Hcy levels of <10 mmol/L were considered to be low and desired. These criteria were also determined based on the fact that levels of this amino acid might have been affected by body saturation with folic acid and vitamins B_6 and B_{12} resulting from previous intake of supplements recommended in the preconception period, although there were no cases where an attending physician recommended to take such supplements in the diagnostic period.

Methionine is a source of homocysteine, and it mainly comes from animal-derived products (animal protein), therefore increased consumption of such products may cause a temporary increase in its blood levels. However, there is no evidence to prove that high consumption of methionine causes chronic hyperhomocysteinemia in subjects with normal supply of folates and vitamins B_6 and B_{12} (Gąsiorowska *et al.* 2008). The results of the own paper confirm this hypothesis. No correlations have been observed between the consumption of animal protein and the methionine levels in a diet with the homocysteine levels. This was accompanied by sufficient, moderate intake of vitamins B_6 and B_{12}.

Hyperhomocysteinemia may directly lead to early damage of the decidua or chorionic vessels leading to embryo implantation disturbances (Chakraborty *et al.* 2013). Reduced Hcy levels due to intake of folic acid do not; however, prevent from cardiovascular diseases (Clarke *et al.* 2010). Nonetheless, an important role of n-3 polyunsaturated fatty acids in this process has been emphasised. They may reduce the risk of coronary heart disease via various mechanisms: prevention of arrhythmias, reduction of blood serum levels of triglycerides and total cholesterol, reduction of blood platelet aggregation and reduction of inflammation (Demaison & Moreau 2002). Additionally, n-3 PUFAs have been shown to participate in regulation of mRNA expression of genes encoding key enzymes involved in the homocysteine metabolism (Huang *et al.* 2012). A meta-analysis of interventional studies suggests that supplementation with n-3 PUFAs may reduce the plasma homocysteine levels (Huang *et al.* 2011). The own study demonstrated a significant negative correlation between the blood plasma homocysteine levels and the content of polyunsaturated fatty acids ($r=-0.253$; $p<0.05$), n-3 PUFAs ($r=-0.225$; $p<0.05$) and n-6 ($r=-0.219$; $p<0.05$) in a diet. Also in papers of other authors higher consumption of n-3 PUFAs was associated with reduced plasma levels of homocysteine (Berstad *et al.* 2007).

PUFAs are especially important in women's diet and not only for reproductive reasons. A-linolenic acid (ALA, 18:3, n-3), docosahexaenoic acid (DHA, 22:6, n-3) and eicosapentaenoic acid (EPA, 20:5, n-3) are important as they are involved in the prevention of cardiovascular diseases in women from all age groups. n-3 fatty acids are also thought to play a role in the prevention of some neoplastic diseases like breast cancer or colon cancer, they may reduce the risk of post-partum depression, manic depressive disorder, dementia (Alzheimer's disease and other), arterial hypertension, diabetes and, to some degree, macular degeneration. They may play a positive role in the prevention of premenstrual stress syndrome, especially dysmenorrhoea and hot flushes associated with menopause. It is probably a result of reduced production of eicosanoides, pro-inflammatory molecules that result from an increased amount of n-6 fatty acids in a diet. Sufficient consumption of n-3 PUFAs may be beneficial for women with diabetes who are pregnant as they prevent from the development of delayed metabolic diseases associated with macrosomy in their offspring. Positive effects of n-3 fatty acids on the glucose metabolism and body weight have been also demonstrated (Bourre 2007).

Polyunsaturated fatty acids belonging to n-6 and n-3 families cannot be synthesised in the human body due to lack of enzymatic systems that are able to introduce double bonds in n-6 and n-3 positions in the chain. Therefore, linoleic acid (LA, 18:2 n-6) and α-linolenic (ALA, 18:3 n-3), precursors of long-chain polyunsaturated fatty acids (such as EPA, DHA), have to be provided with a diet and are defined as essential unsaturated fatty acids. In all organs, including the nervous system and heart, DHA comes directly from a diet or it is produced by ALA transformation in the liver via desaturases. There are some doubts whether optimum consumption of ALA may translate into sufficient body supply with DHA as it competes with LA as a substrate for the same enzymes. According to some authors the ratio between n-3 and n-6 acids is important (Simopoulos 2002). It has been demonstrated that the total levels of DHA, n-3 PUFAs in phospholipids and the n-3/n-6 ratio were significantly negatively correlated with the Hcy plasma levels (Li *et al.* 2006). In a different paper increased levels of n-3 PUFAs in plasma phospholipids and the n-3/n-6 ratio were associated with reduced Hcy levels in patients with hyperlipidaemia (Li *et al.* 2007). In this study, the ratio of n-3/n-6 fatty acids was 5.51±3.09 on average, and no significant correlation with the blood plasma levels of Hcy were demonstrated. Additionally, there were also no significant correlations between the EPA (20:5) and DHA (22:6) levels in a diet with homocysteine values; however, higher LA ($r=-0.218$; $p<0.05$) and ALA ($r=-0.216$; $p<0.05$) consumption was associated with significantly lower blood levels of this amino acid. Apart from total consumption of unsaturated fatty acids their percentage contribution to the energetic value of a diet seems to be important (Schwingshackl & Hoffmann 2012). The own study showed that women whose diets demonstrated high PUFA content in the total energy value had significantly lower Hcy levels compared to patients with a low percentage of these acids in the energy value of their daily nutritional portion (5.34% vs. 3.36%, respectively).

Elevated plasma Hcy levels are an independent risk factor for cardiovascular diseases (Huang *et al.* 2011). Therefore, it is important to change these levels using a diet as it potentially helps treatment. High consumption of n-3 polyunsaturated fatty acids, especially docosahexaenoic acid (22:6 n-3) and eicosapentaenoic acid (20:5 n-3), has beneficial effects on thromboembolic factors and it reduces the mortality rate due to cardiovascular incidents (Li *et al.* 2006).

Additionally, increased levels of HDL cholesterol and reduced levels of triglycerides were observed in case of a diet rich in monounsaturated fatty acids. Both in long-term and short-term studies the efficacy of increased MUFA consumption on reduced arterial blood pressure was shown. In type 2 diabetes MUFAs had hypoglycaemic properties and reduced the levels of glycated haemoglobin in a long term. However, studies regarding the effects of MUFA consumption on the risk of coronary heart disease do not provide unanimous results (Schwingshackl & Hoffmann 2012). This paper presents a significant negative correlation between the homocysteine levels in the blood plasma and the content of MUFAs in a diet ($r=-0.127$; $p<0.05$). Women whose diets demonstrated high MUFA content in the total energy value also had significantly lower Hcy levels compared to women with a low percentage of these acids in the energy value of their daily nutritional portion (13.81% vs. 10.68%, respectively). Although the literature documents relatively well the beneficial effects of high consumption of MUFAs on health there are no specific recommendations regarding their amount in a diet. However, due to significant importance of dietetic interventions in the prevention and treatment of numerous diseases such as cardiovascular diseases, monounsaturated fatty acids may be an important tool with regard to modification of nutritional behaviour. There is strong evidence that when SFAs and carbohydrates are replaced with monounsaturated fatty acids the effects of various cardiovascular risk factors are significantly reduced. Results of numerous meta-analyses emphasise that a diet rich in MUFAs has beneficial effects on the reduction of systolic and diastolic arterial blood pressure as well as parameters of glycaemia control, although its effects on lipid parameters are still controversial (Schwingshackl & Hoffmann 2012). However, taking into account women with fertility disorders – this group includes a high rate of women with PCOS and with accompanying carbohydrate metabolism disturbances – it seems important to assess the consumption of MUFAs and to introduce any necessary corrections of their consumption in order to prevent long-term effects of diseases, including fertility disturbances associated with high levels of Hcy.

Plant oils (mainly olive oil) and macadamia and hazel nuts are a good source of MUFAs, whereas rape seed oil and walnuts are a good source of ALA (18:3, n-3). On the other hand, EPA and DHA are almost exclusively found in fish and seafood. They contain even 20 times more of these acids compared to pork. In general, there is no risk of too high consumption of n-3 fatty acids. It is thought that possible side effects (namely bleeding due to coagulation disorders and blood platelet aggregation) would be associated with consumption that is 50–100 times higher than in a typical diet. Epidemiological studies in many countries have demonstrated the effects of a diet rich in fish on the prevention of cardiovascular diseases in all age groups and in both sexes; however, especially in women (Bourre 2007; Schwingshackl & Hoffmann 2012). The Nurses Heath Study demonstrated that consumption of fish five times daily reduced the risk of cardiovascular death by 50%. Even women who consumed fish only 1–3 times a month had 20% higher chances of survival. The effects of fish consumption were more important for diabetic women – high consumption reduced this risk by 60%, and low consumption by 30%. Beneficial effects were also observed in young women, including those being overweight (Oh *et al.* 2005). Study subjects who consumed the highest levels of omega-3 fatty acids had the lowest levels of inflammatory markers and markers responsible for endothelial cell activation (Lopez-Garcia *et al.* 2004).

Within the last two decades several interventional studies with low numbers of subjects and short duration documented beneficial effects of n-3 PUFA supplementation on the plasma Hcy levels (Huang *et al.* 2011). It is also emphasised that a diet rich in MUFAs plays an important role in the prevention and treatment of metabolic diseases. Results of the own work confirmed that high Hcy levels were significantly negatively correlated with the presence of both MUFAs and PUFAs, especially ALA, a precursor of long-chain n-3 polyunsaturated fatty acids (EPA and DHA), in a diet. Therefore, with regard to the prevention of infertility disorders in all women, especially those with PCOS, it seems reasonable to introduce the Mediterranean diet that is naturally rich in beneficial fat as it can be an element supporting treatment and preventing long-term complications associated with a disease. As the own paper assessed only correlations, it is necessary to perform an experimental study in order to confirm a causal relationship between the consumption of MUFAs and PUFAs and the homocysteine levels.

CONCLUSIONS

Increased consumption of PUFAs, including α-linoleic acid, in a diet seems to be an important factor preventing from hyperhomocysteinemia.

As the elevated levels of homocysteine have adverse effects on reproduction women with such problems, especially those with PCOS, should follow the Mediterranean diet which is rich in PUFAs and MUFAs as an element supporting treatment and preventing long-term complications associated with diseases.

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