SERUM LIPID PROFILE IN WELL AND POORLY CONTROLLED PATIENTS WITH DIABETES MELLITUS.

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Background: Abnormal secretion or action of insulin has been considered the main cause of changes in carbohydrate, lipid and protein metabolism during diabetes mellitus (DM).
Aim : To study the effect of metabolic control on lipid profile in patients with diabetes mellitus wether they are hypertensive or not matched with age ,sex and body mass index non- diabetics.
Method: This study presents lipid profile (measured by enzymatic method) in 99 diabetic patients (age range 14-74 y) as modulated by the type of disease and degree of metabolic control (reflected by HbA1c level).These patients are grouped into optimally controlled (11 with HbA1c <= 7.9 %); acceptably controlled (23 with HbA1c < = 10.1%) and poorly controlled (65 with HbA1c > 10.1%).
Forty-two of the diabetics belong to Type I and 57 to Type II diabetes mellitus. The results obtained from the diabetics are compared with those obtained from 40 non-diabetic subjects (age range 18 - 40y).
Both groups of diabetics and non-diabetics are subdivided into normo- and hypertensives. Other clinical parameters like proteinuria and body mass index (BMI) are considered in the study.
Results: Dyslipidemia of diabetes mellitus involves an increase in all serum lipids except the HDLc, which shows a significant reduction. Dyslipidemia of DM could be attributed to poor glycemic control that reflects insulin insufficiency or resistance with consequent increase in lipid mobilization and tissue protein breakdown.
Hypertriglyceridemia (high VLDL) is the predominant feature of DM (especially Type II) and is greatly affected by the metabolic control being significantly higher in the poorly controlled than the optimally and acceptably controlled groups.
The effects of hypertension on serum lipids included an increase in total cholesterol, triglycerides, low density lipoprotein cholesterol and low density to the high density lipoprotein cholesterol ratio with a reduction in the high density lipoprotein cholesterol in the hypertensives of both diabetics and non-diabetic subjects; when compared with their normotensive groups.
Conclusion : Diabetics wether they are controlled or not especially with hypertension need treatment for their lipid disorders and must change their live style.

LIPID PROFILE IN TYPE 1 AND TYPE 2 DIABETIC PATIENTS

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Aim: to evaluate the lipid profile in type 1 and type 2 diabetic patients
Materials and methods: A retrospective study was performed on 849 hospitalised diabetic patients (male 353, female 496) at Clinic of Endocrinology in Skopje in period January-December 2001. Demographic data, diabetes history, laboratory findings were analysed. Patients were divided in two groups: group 1, 214 patients with Diabetes Mellitus (DM) type 1, on age 31.2±10.0 years with duration of DM 9.8±7.5 years, and group 2, 635 patients with DM type 2, age 56.3±11.8 years, duration of DM 8.3±8.1 years on oral antidiabetic agents or insulin therapy.
Results:

	BMI (kg/m2)	*HbA1C (%)*	*Triglycerides (mmol/L)*	*HDL CHL (mmol/L)*	*LDL CHL (mmol/L)*
Gr. 1	23.4±3.6	10.2±2.6	2.1±1.8	1.5±1.2	3.2±1.7
Gr. 2	33.4±4.4	10.1±4.1	2.5±3.1	1.2±0.5	3.5±0.6

Patients in group 1 had lower lipid profile findings than patients in group II ($p<0.05$). Hypertension was found in 11.5% of patients in group 1, and 30.4% of patients in group 2 ($p<0.05$). Coronary artery disease was found in 3.1% in group 2, instead of 18.4% in group 2 ($p<0.05$).
Conclusion: Patients in both two groups had bad glycemic control. Patients with DM type 2 are obese and had higher lipid profile findings instead of patients with DM type 1. Hypertension and coronary artery disease was found in both groups, but more significant is in patients with DM type 2.

LIPID PROFILE IN TYPE 2 DIABETES

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We investigated 236 patients with type 2 diabetes (the average age of patients is 57,3 ± 7,9, the duration of DM is 8,5 ± 5,6 years). There were 66,1% female and 33,9% male patients; 67% of patients had BMI more then 30 kg/m². To divide the patients according to the degree of dyslipidaemia we used the criteria of the European Society of Cardiology, the European Atherosclerosis Society and the European Society of Hypertension (1993) – the normal level of cholesterol was below 5,2 mmol/l, the border level was between 5,2 and 6,5 mmol/l and hypercholesterolaemia was considered as a level higher than 6,5 mmol/l; the normal level of triglyceride was below 1,8 mmol/l, the border level was between 1,8 and 2,3 mmol/l and hypertriglyceridaemia was considered as a level of triglyceride higher than 2,3 mmol/l.

Among the patients with normal and high weight there were 49,9% and 53,8% of patients correspondingly with the border level of cholesterol. Hypercholesterolaemia was revealed in 16,4% of patients with the normal weight and in 30,8% of patients with the high weight. 30,7% of patients with the normal weight had the normal level of cholesterol. Only 15,1% of patients with the high weight had the normal level of cholesterol.

The same number of patients with the normal and high weight (30%) had the low level of HDL-cholesterol.

44,5% of patients with the high weight had hypertriglyceridaemia. At the same time only 10,7% of patients with the normal weight had hypertriglyceridaemia.

Conclusion: Patients with type 2 diabetes and high weight have been found to have more atherogenic lipid profile than the patients with the normal weight.

SERUM LIPOPROTEINS AND INSULIN SENSITIVITY IN TYPE 2 DIABETIC PATIENTS: INFLUENCE OF OBESITY

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Previous studies have shown that diabetes and obesity, as a part of metabolic syndrome X, are associated with high incidence of coronary heart disease (CHD). However, in obese patients with type 2 diabetes levels of oxidized LDL (oxLDL), lipoproteins, plasminogen activator inhibitor 1 (PAI-1) and insulin sensitivity correlates with development of CHD. The aim of this study was to analyze correlation between different lipoproteins and insulin sensitivity in obese and non obese diabetic patients. The study was performed in obese type 2 diabetics (group A: N= 28) and type 2 diabetics with normal BMI (group B: N=22) Lipids profile was estimated by measuring Total, HDL and LDL cholesterol (Ch), as well as, triglycerides (Tg) by enzymatic methods. Apolipoprotein (Apo) AI, AII, B100 and E as well as lipoprotein (Lp) (a) were measured by nephelometry. Oxidized (Ox) LDL was measured by ELISA methods (Mercodia). Insulin sensitivity was determined by model of homeostasis (HOMA).
In our study we found significant differences between these 2 groups in the levels of oxLDL (131,2 vs 98,9 IU/l,), Apo I (1,88 vs 1,68g/l), Apo II (333,9 vs 370,5g/l,) as well as, PAI-1(5,08 vs 3,6 g/l), (A vs B $p < 0,05$), respectively. In contrast, we didn't find significant differences considered conventional lipid profile. Insulin sensitivity was lower in group A than in group B (9,29 vs 7,94; A vs B; $p <0,05$). Also, in both groups there was significant correlation between levels of oxLDL and LDL-c ($r=0,72$; $p < 0,01$), as well as, levels of HDL-c and ratio Tg/HDL-c ($r= 0,793$, $p < 0,01$). Also, between oxLDL and HDL-c we found significant, but negative correlation ($r = -0,202$, $p <0,05$).
Our results signify that obesity amplify multiple changes of lipids and lipoproteins. Also, our results imply that obesity exerts decerased insulin sensitivity in diabetic patients.

DIABETES MELLITUS: CORONARY ARTERY DISEASE AND RISK FACTORS

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OBJECTIVE: To evaluate coronary artery disease (CAD) and risk factors in diabetic patients

METHODS: The Dispensery for diabetes at the Medical Center Negotino covers a territory of 23200 inhabitants with 640 registered diabetic patients, where type 1 are 11 and type 2 are 629. Age is between 10- 86 years. Duration of DM is 8.3±6.3 years. CAD is confirmed on a basis of clinical symptoms, laboratory tests, ECG, coronary stress test, echocardiography and coronary angiography.

RESULTS: From total number of diabetic patients, 164 had CAD (25,6%). Hypertension (>135/80mmHg) was found in 196 patietns (30.6%). BMI over 30kg/m2 was found in 135 patients (21.1%). The following lipid profile was notified: triglycerides 1,79±1,8mmol/l, total cholesterol 5.7±1,2 mmol/l, LDL cholesterol 3.1±0,6 mmol/L.

CONCLUSIONS: The prevalence of CAD in our study is comparable to macedonian and global data. 25.6% had CAD, 30.6% are hypertensive, 21.1% are obese. These patients are on risk for macrovascular diabetic complication, and are reason to underline the prevention of diabetes.

PREVALENCE AND ROUTINE CARE TREATMENT OF DIABETES MELLITUS IN GERMANY: FINDINGS FROM THE DETECT STUDY

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Cardiovascular disease accounts for more than two thirds of the deaths in patients with type 2 diabetes (DM) and three fourths of these deaths result from ischemic heart disease. Type 2 diabetes increases CHD risk two to three times in men and three to seven times in women. Yet, only a fraction of the patients needing therapy seem to be recognized and receive adequate antidiabetic and lipid-lowering treatment.

The epidemiological study DETECT (*D*iabetes Cardiovascular Risk *E*valuation: *T*argets and *E*ssential Data for *C*ommitment of *T*reatment) was launched to identify the reasons, the extent and the short-term consequences of unmet needs in patients with high cardiovascular risk. DETECT is a large multistage cross-sectional and prospective 12-month study of 70000 consecutive patients in over 3000 primary care offices, nationwide. Here, we report on the findings from a subset of approximately 7500 patients characterized by an extensive standardized laboratory program with focus on the treatment modalities in patients with type 2 diabetes mellitus.

According to the guidelines of the American Diabetes Association (fasting glucose > 126 mg/dl, no caloric intake for at least 8 h) or clinical history, 20 % of the patients were identified as diabetic. DM was more frequent in men (32,5 %) than in women (16,3 %). The prevalence of DM increased with advancing age of the patients (33,5 % at the age of 70 to 79 years). Surprisingly, only 62 % of the diabetic patients were previously recognized by the physicians, more than one third was newly identified by our screening program. Only 70 % of the known patients with DM received antidiabetic medication (34,2 % metformin, 16,7 % insulin, 9,5 % sulfonylurea, 9,6 % others) and about one third was treated with lipid-lowering medication. The majority of the diabetics did not achieve the treatment goals for fasting glucose (65 %), HbA1c (32 %), LDL cholesterol (76 %), and triglycerides (56 %).

Taken together, our results indicate that a significant proportion of the diabetic patients were not recognized by the physicians and the treatment of DM was often insufficient. Patients with DM are at high risk for CHD. However, mainly lipid-lowering therapy in these patients was inadequate.

PLASMA LIPIDS LEVEL MAY REFLECTED THE SEVERITY OF DIABETIC NEPHROPATHY

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Background and aim. Diabetes mellitus (DM) characterized with plasma lipid abnormalities which are risk factor both micro- and macrovascular complications. Measuring of plasma lipids content is important for diabetes control. The aim of our study is investigation of the plasma lipid contents at the several degree of diabetic nephropathy.

Material and methods. 68 patients with Diabetes Mellitus with disease duration from 2 months to 25 years and 21 non-diabetic subjects were observed. Among patients with DM 30 were with microalbuminuria, 28 - have proteinuria and 20 patients with end stage of renal disease. In patients were measure total cholesterol and beta lipoproteins level and compare with carbohydrate and renal function indexes.

Results. Blood glucose contents, HbA1c level were increased in all patients with DM and not different among the groups. Renal glomerular filtration rate (GFR) was significantly decreased (43.6±6.7 ml/min) and creatinine level was increased (in 2.38 time, P<0.05) only in ESRD group. Plasma total cholesterol and beta lipoproteins level were elevated in all group then non diabetic subjects. Total cholesterol level increased on 1.13 time (P<0.05) in patients with MAU, on 1.3 time in patients with PU and in 1.8 time in patients with ESRD. Plasma beta lipoproteins level were elevated on 1.51 time (ns), in PU patients on 2.14 time (P<0.05), in patients with ESRD on 1.81 time (P<0.05).

Conclusion. Plasma lipids level increased according to kidney damage at the DM and indicate of renal failure earlier than plasma creatinin and GFR level.

PREVALENCE OF URINARY TRACT INFECTIONS IN WOMEN WITH TYPE 2 DIABETES IN GEORGIA

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Background and Aims: Microvascular damage is the most wide-spread complication of diabetes, though patients (pts), especially with type 2 diabetes (T2DM), very often develop urinary tract infections (UTI). Not proper treatment of UTI and poor glycemia control result in their relapse; as a result UTI may turn to chronic disease. The aim of the present work was to study the prevalence of UTI in our pts, and their correlation with HbA1c levels. It was the first attempt to assess UTI prevalence and specificity in T2DM ever made in Georgia.**Materials and Methods**: In total 130 T2 DM pts (mean age - 55±14yrs., diabetes duration 10±3 yrs.) with UTI, but without overt kidney disease in anamnesis were enrolled in the study. According to the type of diabetes therapy they were devided into 3 groups(Gr.): Gr. 1 – 31 pts, treated with Sulfanylurea and Metformin, twice daily. Gr. 2 – 35 pts, treated with 2 daily injections of Actrapid/ Insulatard and Sulfanylurea and Metformin twive daily. Gr.3 – 27 pts, treated with intensive insulin therapy (Actrapid, 3 premeal injections and Insulatard at bedtime). Hypocaloric diet and mild-to-moderate physical activity were prescribed to all the patients. HbA1c levels were – 9.8±1.3% (Gr. 1); 9.4±1.4% (Gr. 2); 7.2±1.3% (Gr. 3). In hypertensive pts mean arterial BP was >140/90mmHg. At entry urinalysis, urine inculation and test to antibiotic sensitivity for proper treatment selection were performed. Urinalyses was repeated after two weeks of UTI treatment. **Results:** On the background of high HbA1c levels UTI were very frequent in Gr. 1 (19 out of 31 pts, 61.3%) and Gr. 2 (20 out of 35 pts, 57.1%). Besides, in Gr. 1 UTI relapse was registered the most frequent (in 11 pts out of 19, 57.9%) and in 3 cases pielonephritis developed; while in Gr. 3, where pts were on intensive insulin therapy, and HbA1c levels were the lowest, relapse was less frequent (9pts out of 27, 33.3%). **Conclusion:** Our observations revealed that poor DM control with elevated HbA1c levels is associated with high prevalence of UTI; the higher are the HbA1c levels, the higher is the prevalence of primary and repeated UTI. Complex therapy (especially when intensive insulin therapy is used) aimed at achiving good metabolic control and proper UTI treatment results in decrease in relapse frequency, and prevents UTI transition to chronic diseases.

DUTY - REGISTRY TO ASSESS AND IMPROVE LIPID THERAPY IN PATIENTS WITH DIABETES MELLITUS

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Patients with diabetes mellitus (DM) are at high risk for cardiovascular events. Therefore consistent risk management according to meets guidelines is needed to reduce the risk for complications such as coronary heart disease (CHD) or nephropathy. The aim of the DUTY registry (**D**iabetes mellitus needs **U**nrestricted evaluation of patient data **T**o **Y**ield treatment progress) was to determine whether guidelines are incorporated consistently into the daily practice of lipid management of patients with DM. For this purpose educational material, such as official guidelines and cardiovascular risk calculators, were provided to 3,213 general practitioners. 45,605 patients with pre-existing or newly diagnosed DM were recruited. Data were obtained at baseline and 9 months after the educational process of the physicians was initiated. 51% of the patients were women. The patients had a mean age of 64 years and a BMI of 29 kg/m². 92.4% had DM type 2, 0.4% were undergoing dialysis. The most common diagnoses and cardiovascular risk factors were, a positive family history of CHD (44.8%), CHD (26.9%), previous or current cigarette smoking (21.6%) and neuropathy (19.4%). At the baseline visit 25.6% of patients were receiving statins and 13.8% had LDL-cholesterol level below 100 mg/dl (Mean 136±37 mg/dl). During the observation period the percentage receiving statins increased to 41.1% but still only 16.6% of patients reached the LDL-cholesterol target values (Mean 125±32 mg/dl).. Fibrates were used in 4.5% of patients at baseline and 4.8% at 9 months. Mean triglyceride levels at the beginning of the study were 219 mg/dl and at the end 200 mg/dl. HDL-cholesterol was initially 48±14 mg/dl, at 9 months 50±13 mg/dl. From these results it may be concluded that too many patients with DM do not receive an adequate lipid lowering therapy according to recent guidelines and therefore do not reach target lipid values.

THE INFLUENCE OF OBESITY ON CARDIOVASCULAR RISK FACTORS IN PATIENTS WITH TYPE 2 DIABETES MELLITUS

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Patients with type 2 diabetes mellitus are at increased risk for development of cardiovascular diseases. Dyslipidemia, increased levels of fibrinogen, uric acid and proteinuria are common findings in this group of patients. Obesity represents cardiovascular risk factor per se but it is usually associated with disorders of other risk factors. The aim of our study was evaluate the influence of obesity on lipid levels and other risk factors. Therefore, in 350 patients with type 2 diabetes stratified by level of nutrition (Group 1 – normal weight: n=118, Group 2 – overweight: n=123, Group 3 – obese: n=109) total cholesterol, HDL, LDL, triglycerides, fibrinogen, uric acid and proteinuria (grams per 24 hours) were determinated. The levels of HbA1c were comparable between these groups of patients (Group 1 vs. Group 2: 9.61 ± 2.59 vs. 10.24 ± 1.91, $p > 0.05$; Group 1 vs. Group 3: 9.61 ± 2.59 vs. 9.65 ± 2.17, $p > 0.05$; Group 2 vs. Group 3: 10.24 ± 1.91 vs. 9.65 ± 2.17, $p > 0.05$). We observed significant difference between Groups 1 and 3 in HDL (1.17 ± 0.4 vs. 1.02 ± 0.3, $p < 0.05$), triglyceride (2.51 ± 2.85 vs. 3.27 ± 2.81, $p < 0.05$) and uric acid (266 ± 96.84 vs. 346 ± 136.59, $p < 0.05$) levels. Also there were difference in triglyceride levels between groups 1 and 2 (2.51 ± 2.85 vs. 3.48 ± 3.47, $p < 0.05$) and proteinuria levels between groups 2 and 3 (185.84 ± 23.60 vs. 383 ± 86.25, $p < 0.05$). In conclusion, obesity significantly increases the levels of cardiovascular risk factors in patients with type 2 diabetes.

RETINOPATHY AND NEUROPATHY IN TYPE 2 DIABETES WITH AND WITHOUT METABOLIC SYNDROME.

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The purpose of this research was: to study the diabetic neuropathy (DN) and diabetic retinopathy (DR) development in metabolic syndrome (MS).
Materials and methods. 48 diabetic patients with MS (23 men and 25 women) – MS Group (MSG) and 24 diabetic patients without MS (16 men and 8 women) – Control Group (CG) were studied at primary receipt. Distinctions in sexual structure between two groups were statistically insignificant. The degree of DN was defined by scale of Yang. Degree of DR expression was defined by mark system.
Results. The CG characteristics: Age 44.8±8.58 year; BMI 24.4±1.30 kg/m²; Duration of diabetes 6.8 ± 5.50 year; Systolic BP 121.5 ±9.26 mmHg; Diastolic BP 77.5±4.66 mmHg; HbA1c 9.8±2.74%; TC 203.5±46.61 mg/dl; HDL-C 41.9±6.40 mg/dl; LDL-C 130.±36.07 mg/dl; VLDL-C 45.1±15.75 mg/dl; TG 239.2±69.02 mg/dl.
The MSG characteristics: Age 53.1±8.77 year ($P<0.001$); BMI 32.1±3.56 kg/m² ($P<0.001$); Duration of Diabetes 6.3±6.22 year ($P>0.05$); Systolic BP 152.4 ±15.05 mmHg ($P<0.001$); Diastolic BP 90.6 ±7.69 mmHg ($P<0.001$); HbA1c 9.6±2.46 ($P>0.05$); TC 233.3±67.62 mg/dl ($P<0.05$); HDL-C 44.3±15.10 mg/dl ($P>0.05$); LDL-C 148.0±60.47 ($P>0.05$); VLDL-C 58.0±39.71 ($P>0.05$); TG 279.6±191.90 ($P>0.05$). The degree of DN was higher at MSG (5.79±2.58 in MSG and 4.29±2.56 in CG; $P<0.05$). DR expressiveness was higher in MSG, but difference was not statistically significant (1.46±2.48 and 1.08±1.93 correspondingly). At MSG there was a positive correlation between degree of DR and degree of DN ($r= +0.32$; $P<0.05$). At the same time in CG group correlation between degree of DR and DN was absent.
Conclusion. Presence of a metabolic syndrome promotes heavier current of DN and DR.

CORONARY HEART DISEASE, RISK FACTORS & THE METABOLIC SYNDROME IN AFRICAN & WHITE MEN WITH TYPE 2 DIABETES

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Coronary heart disease (CHD) is rare in the African but common in the white population of South Africa. We evaluated the prevalence of CHD and the Metabolic Syndrome (MS, ATP III criteria) and risk factors in 278 consecutive men with type 2 DM newly attending the Diabetes Clinic, Johannesburg Hospital, South Africa – 206 African (A) & 172 white (W). A were ~5 yrs younger than W ($p<0.0001$), but duration of DM was similar. Overt CHD was present in 3.6% of the A and 26.2% of the W men ($p<0.0001$). Two or more features of the MS, in addition to the diabetes, were present 51.2% of A and 65.5% of W (NS). Compared to W men, the A were leaner with smaller waist circumferences (A, 95.6 vs W, 105.4cm), had lower levels of total and LDL cholesterol (4.98 vs 5.76mmol/l; 3.01 vs 3.51mmol/l respectively) and of triglycerides (1.60 vs 2.10mmol/l) ($p<0.003$ for each), but were more likely to be hypertensive (50.2% vs 37.8%, $p=0.018$). The prevalence of smoking and micro-albuminuria and HbA1c levels were similar. High levels of small dense LDL particles (assessed as Triglyceride:HDL-C ratio) were more frequent in W (W 60.1%, A 42.1%; $p=0.039$). 28.4% of A and 52.9% of W had 10 yr CHD (Framingham) risk >20% ($p<0.0001$). Among the A and W men **with** CHD, age and lipid profiles were similar. Conclusion: in comparison with W men, CHD remains rare in African men with Type 2 diabetes, associated with better lipid profiles, and probably less insulin resistance, despite a high prevalence of the MS. However, a large group of African men are at high risk. Once CHD is overt, risk profiles are similar in diabetic A and W men.

THE ATHEROGENICITY OF THE VARIOUS STIGMATA OF DIABETIC DYSLIPIDEMIA

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Background: It is not clear which components of dyslipidemia predict atherosclerotic disease in patients with diabetes type 2 (DM2) because data on lipid values in patients with both DM2 and coronary artery disease (CAD) are scarce. We hypothesized that the low HDL-C / high triglyceride pattern of diabetic dyslipidemia drives diabetic atherosclerosis.
Methods: We enrolled 756 consecutive patients undergoing coronary angiography for the evaluation of suspected myocardial ischemia. The incidence of vascular events was recorded during a follow-up period of 2.3 ± 0.4 years. LDL-C was measured directly with QuantolipLDL (Roche, Switzerland).
Results: After exclusion of patients receiving lipid lowering drugs (n = 251), coronary patients with DM2 (n = 96) had significantly higher triglycerides (191 ± 127 vs. 146 ± 86 mg/dl; p <0.001), lower HDL-C (44 ± 13 vs. 50 ± 14 mg/dl; p <0.001), a smaller LDL particle size (257 ± 7 vs. 260 ± 7 Å; p = 0.048), and, interestingly, lower LDL-C (124 ± 38 vs. 138 ± 33; p <0.001) than non-diabetic patients. Factorial analysis revealed two factors in the lipid profiles of our patients: triglycerides, HDL-C, Apo A1 and LDL peak particle diameter loaded high on factor 2 (resembling the low HDL-C / high triglyceride pattern of diabetic dylipidemia), and total cholesterol, LDL-C, and Apo B loaded high on factor 1 (resembling hypercholesterolemia). In Cox regression analysis DM2 was associated with an increased risk of vascular events (n = 95), with an adjusted OR of 1.84 (1.20 - 2.82; p = 0.005). Factor 2 was significantly predictive for vascular events both in the total study population and among patients with DM2 (standardized adjusted ORs 0.74 [0.60-0.93; p = 0.009] and 1.06 [0.86-1.31; p = 0.565], respectively). Factor 1 was not significantly associated with the incidence of vascular events.
Conclusions: In addition to the classical pattern of diabetic dyslipidemia coronary patients with DM2 exhibit low serum levels of LDL-C. Among patients requiring coronary angiography the low HDL-C / high triglyceride pattern, but not hypercholesterolemia is predictive for vascular events. In particular, this pattern of dyslipidemia has a strong impact on the incidence of vascular events among patients with DM2.

INSULIN RESISTANCE IS AN INDEPENDENT PREDICTOR OF VASCULAR EVENTS IN DIABETIC PATIENTS WITH CORONARY ARTERY DISEASE

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Background: Patients with both diabetes and established coronary artery disease are at a high risk of cardiovascular events. Insulin resistance (IR) is a central feature of diabetes mellitus type 2 (DM2). Therefore, the impact of IR on the incidence of vascular events in diabetic patients with established CAD is of particular interest.
Methods: We estimated insulin resistance by the HOMA index in 495 patients with angiographically proven CAD and recorded the incidence of vascular events over a mean follow-up time of 2.3 ± 0.4 years.
Results: The HOMA index was higher in coronary patients with DM2 (n = 127) than in nondiabetic coronary patients (6.5 ± 5.9 vs. 3.0 ± 4.2; p <0.001). Thirty-one (23.8%) patients with DM 2 and 60 nondiabetic patients (14.5%) experienced at least 1 vascular event. In Cox regression analysis adjusting for age, gender, and baseline extent of coronary artery disease (number of angiographic stenoses 50%) diabetes was an independent predictor for the incidence of vascular events (OR = 1.725 [1.116 – 2.667]; p = 0.014). Equally, the HOMA index proved independently predictive for the incidence of vascular events in the total study cohort: the standardized OR adjusted for age, gender, and baseline extent of CAD was 1.178 [1.026-1.351]; p = 0.010. In subgroup analyses with respect to diabetes status, the HOMA index was significantly predictive for vascular events in patients with diabetes (OR = 1.354 [1.083 – 1.694; p = 0.008]), but not among nondiabetic patients (OR = 1.022 [0.729 – 1.432]; p = 0.901).
Conclusions: In the setting of secondary prevention, IR is a strong and independent predictor of vascular events among patients with DM2. Thus, the degree of IR significantly contributes to the adverse effects of diabetes on the prognosis in coronary patients.

CRP, FIBRINOGEN, AND WHITE BLOOD CELL COUNT IN PATIENTS WITH STABLE CORONARY ARTERY DISEASE AND IMPAIRED GLUCOSE TOLERANCE

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Background: Inflammatory markers are elevated in patients with diabetes mellitus and in patients with coronary artery disease (CAD). However, data are scarce on inflammatory markers in patients with both CAD and diabetes. As impaired glucose tolerance (IGT) frequently precedes diabetes type 2 (DM2), inflammatory markers in patients with IGT are of particular interest.
Methods: We measured CRP, fibrinogen, and white blood cell count (WBC) in 468 patients with clinically stable, angiographically proven CAD. Patients with diabetes type 1 (n = 3) were excluded from the analyses. Oral glucose tolerance tests (oGTT) were performed in patients without established diabetes.
Results: DM2 had previously been established in 100 patients. In oGTT another 34 patients proved diabetic, 69 had IGT, and glucose tolerance was normal in 262 patients. CRP, fibrinogen, and WBC were similar in patients with established diabetes and in patients with newly diagnosed diabetes; these patients were thus pooled to a single diabetic group. Between patients with normal glucose tolerance and with IGT, CRP (0.30 ± 0.53 vs. 0.30 ± 0.60 mg/dl; p = 0.822), fibrinogen (377 ± 68 vs. 386 ± 65 mg/dl; p = 0.206), and white blood cell count (6.5 ± 1.8 vs. 6.8 ± 2.2 G/l; p = 0.741) were not significantly different. However, all these inflammatory markers were significantly elevated in patients with DM2 (0.49 ± 0.68 mg/dl, 412 ± 83 mg/dl, 7.5 ± 2.1 G/l; p values were 0.001 for the comparisons vs. normal glucose tolerance and 0.014, 0.102, and 0.007 for the comparisons vs. IGT). In leukocyte subtype analyses only neutrophils were significantly higher in diabetic patients than in patients with normal glucose tolerance and IGT (4.5 ± 1.4 vs 3.7 ± 1.1 and 4.0 ± 1.7 G/l; p <0.001 and p = 0.015, respectively).
Conclusions: Among patients with clinically stable and angiographically proven CAD, CRP, fibrinogen, and white blood cell count are significantly higher in diabetic than in non-diabetic patients, but patients with IGT do not significantly differ from non-diabetic individuals. Specifically, an increase of neutrophils accounts for the overall increase of white blood cell count in diabetic patients with stable CAD.

LIPID ABNORMALITIES IN WOMEN WITH INSULINRESISTANCE AND PCOS

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Dislipidimia, insulinresistance, hyperandrogenia and hyperaestrogenia at PCOS are risk factors for development cardiovascular deseases. The aim of the study was to defines dislipidimia and insulinresistance in women with and PCOS, and their relationship. 16 obese women with PCOS were at the age of (29,6±2,58) years (BMI>27kg/m2) и 21 women without obesity at age (25,05±1,95) years (BMI<27kg/m2). As a result of carrying out oral glucose tolerance test (OGTT) diabetes mellitus (DM) type 2 – at 2/16, impaired glucose tolerance (IGT) – at 3/16, impaired fasting glucose (IFG) – at 2/16 has been revealed. With increase of BMI grew both abdominal obesity WHR (0,87±0,04) (0,88±0,01) p<0,05 and arterial pressure. The systolic arterial pressure is more sensitive (p<0,05) to increase of BMI. The data of lipid parameters specify the possible tendency to development of atherosclerosis in all groups. Insulin correlates in group with obesity with the years (r=0,49 t<0,05), with systolic arterial pressure (r=0,49 t<0,05), with postprandial insulin (r=0,49 t<0,05), with ratio of fasting glucose/insulin (r=-0,77 t<0,01), with aestradiol (r=-0,57 t<0,05). Insulinresistance is defined in all groups, but postprandial hyperinsulinimia accures only at BMI increase. As insulinresistance, dyslipidimia, hyperglycemia are obsereved in all groups at a different degree, and in group with obesity and postprandial hyperinsulinimia, it is possible to make the assumption of probable big risk of development of cardiovascular deseases and DM type 2 at PCOS not only at women with obesity, but also without obesity.

INSULIN RESISTANCE AND ALTERATIONS IN LIPID PROFILE IN WOMEN WITH POLYCYSTIC OVARY SUNDROME

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Objective: To evaluate the insulin resistance (IR) and alterations in the lipid profile in women with polycystic ovary syndrome (PCOS).
Material and methods: Analysed were 38 women with PCOS presented with chronic anovulation and hyperandrogenism. Initial evaluation included measurement of basal serum concentrations of follicle stimulating hormone (FSH), luteinizing hormone (LH), testosterone (T) and dehydroepiandrosterone sulphate (DHEAS), total cholesterol (CH), triglycerides (TG), high density lipoprotein cholesterol (HDL) and low density lipoprotein cholesterol (LDL). All patients underwent 75 gr.oral glucose tolerance test (OGTT), during which fasting and stimulated levels of glucose and insulin were measured at 0', 60' and 120'. Insulin resistance (IR) was derived using the homeostasis model assessment as an index of insulin resistance (HOMA-IR) formula: [fasting insulin (μU/ml) * fasting glucose (mmol/l)]/22,5.
Results: The cohort included 38 women with hyperinsulinemic PCOS, mean age of 24,4 ± 5,95 years, BMI of 30,7 ± 6,7 kg/m2, average duration of menstrual cycle (MNZ) of 69 ± 50 days, testosterone (T) levels of 1,78 ± 1,08 nmol/l, fasting insulinemia of 20,50 ± 7,67 μU/ml, stimulated insulinemia at 60'of 154,08 ± 80,66 μU/ml and stimulated insulinemia at 120' of 140,15 ± 90,1 μU/ml. BMI greater than 27 kg/m2 was found in 58% of the patients. The mean value of HOMA-IR was 4,38 ± 1.63. Thirty eight percent of the patients had serum concentrations of HOL>4,8 mmol/L, 45% had serum concentrations of HDL<1,2 mmol/L and 34% had values for LDL>3 mmol/L. Univariate regression analysis disclosed a strong significant correlation between HDL and T levels (p=0,010)
Conclusion: Women with PCOS are at high risk for dyslipidemia due to hyperandrogenism and frequent obesity. On the other hand, hyperinsulinemia increases the risk of developing dyslipidemia due to insulin resistance. Our results indicate that women with with PCOS show multiple lipoprotein changes and that despite hyperinsulinemia, hyperandrogenism and obesity, these lipoprotein alterations may contribute to an increased cardiovascular risk in women with PCOS.

LIPID PROFILE IN WOMEN WITH POLYCYSTIC OVARY SYNDROME BEFORE AND AFTER METFORMIN THERAPY

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Objective: To evaluate the therapeutic effect of metrofmin on lipid profile, BMI, hormonal and metabolic parameters in women with hyperinsulinemic polycysitc ovary syndrome (PCOS).
Material and methods: Analysed were 25 women, of mean age 27,28 ± 7,85 years, BMI of 34,42 ± 6,61 kg/m^2, Ferriman-Gallwey (F/G) score of 17,43 ± 5,45 and duration of menstrual cycle of 79,2 ± 54,5 days. Basal serum concentrations of lipids were measured before and after metformin therapy. Basal hormonal measurements included: FSH, LH, Testosterone, 17 OH Progesterone, DHEAS, E_2 and Progesterone. All patients underwent 75 gr oral glucose tolerance test (OGTT), during which fasting and stimulated levels of glucose and insulin were measured at 0', 60' and 120'. Both basal and stimulated parameters were examined before and after treatment with metformin (500 mg orally, three times daily for 9 months).
Results: After metformin treatment we observed statistically significant decrease in the metabolic parameters, both basal insulinemia (22,18 ± 5,76 μU/ml *vs.* 17,19 ± 6,67 μU/ml, $P < 0.01$), stimulated insulinemia after 60' (179,18 ± 88,96 μU/ml *vs.* 136,38 ± 75,43 μU/ml, $P = 0,04$), stimulated insulinemia after 120'(163,25 ± 89,2 μU/ml *vs.* 88,46 ± 61,5 μU/ml, $P < 0,01$) and glucemic response to OGTT on 120' (7,07 ± 1,82 mmol/L *vs.* 6,15 ± 1,52 mmol/L, $P = 0,04$). Improvement in the levels of HDL cholesterol were also observed (0,94 ± 0,16 mmol/L *vs.* 1,13 ± 0,19 mmol/L, $P = 0,02$). The mean frequency of menstruation has significantly improved (79,2 ± 54,5 days *vs.* 31,61 ± 7,7days, $P < 0.01$), mean testosterone has significantly decreased (2,78 ± 1,23 nmol/l *vs.* 1,72 ± 0,95 nmol/l, $P < 0.01$). No changes were noted in hirsutism, serum LH/FSH ratio and other sex steroids.
Conclusion: In women with PCOS treatment with metformin is effective in lowering of hyperinsulinemia and hyperandrogenemia. Metformin treatment leads to increasement of HDL cholesterol lipid fraction. No changes were observed in the levels of LDL, trygliucerides and total cholesterol, which may be due to the short period of follow-up of the patients.

ASOCIATION OF VISCERAL OBESITY WITH HYPERLIPIDEMIA

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Objective: To evaluate the lipid profile in patients with visceral type of obesity
Material and methods: The basal serum lipid concentrations were analyzed in 90 females, mean age of 33 ± 12 years, with BMI > 25 kg/m^2, without cardiovascular complications (CVC). According to BMI the patients were divided into 4 groups, and according to the visceral obesity assessed by waist/hip ratio (WHR) the patients were divided into three groups: A with normal fat distribution, B with moderate visceral fat distribution and C with extreme visceral fat distribution.
Results: The patients age shows positive correlation with the atherogenic index (cholesterol/HDL cholesterol) (p<0,04) and with LDL cholesterol/HDL cholesterol (p<0,01). BMI showed significant correlation with tryglicerides (TG) (p<0,016) and cholesterol/HDL cholesterol (p<0,038). The increasement in the degree of visceral fat distribution is characterized with significant elevation of TG (p<0,0001), HDL cholesterol (p<0,001), LDL cholesterol/HDL cholesterol (p<0,002) and significant decrease in HDL cholesterol (p<0,032).
Conclusion: Dyslipidemia in visceral obesity is characterized with increased TG values, lower levels of HDL cholesterol and high atherogenic index.

ASSOCIATION OF LEPTIN RESISTANCE WITH METABOLIC DISORDERS IN ELDERLY JAPANESE PATIENTS

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Although the role of leptin in the regulation of body weight by inhibiting food intake and stimulating energy expenditure, its role in insulin resistance or lipid abnormalities remains unknown. To study the role of leptin resistance in the development of metabolic disorders, we evaluated the association with serum leptin levels with such metabolic parameters as serum lipids, apoproteins, insulin levels and HOMA-IR. The subjects employed were 51 outpatients (23 males and 28 females with an average age 73±10). Those receiving hypoglycemic or lipid lowering drugs were not included. Blood samples were taken in the fasting state for measurement of serum lipids (total cholesterol, LDL-cholesterol, RLP-cholesterol, triglycerides, HDL-cholesterol), free fatty acids, apoproteins (A-I, A-II, B, C-II, C-III, E), glucose, insulin and leptin. Both male and female groups were divided into 3 groups according to the serum leptin levels. 1) Female group had a higher leptin level than that of male group (10.8±5.4 ng/ml vs. 3.8±1.4 ng/ml, p<0.01), while serum lipid levels and insulin, HOMA-IR levels did not differ between the genders. 2) Serum levels of leptin correlated with BMI in both male and female groups (r=0.576, p<0.005, r=0.479, p<0.001, respectively). 3) Serum levels of triglycerides and RLP-cholesterol increased proportionally with leptin levels in female group, but no correlations with leptin levels in male group. 4) Fasting insulin levels and HOMA-IR had positive correlations with serum leptin levels (r=0.533, r=0.547, p<0.005) in female group, but no correlations with leptin levels in male group. These results indicated that lipid abnormalities and insulin resistance in females were more closely related with elevation of serum leptin than in males.

BODY FAT AND C-REACTIVE PROTEIN LEVELS IN HEALTHY NON-OBESE MEN.

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The relationships between C-Reactive Protein (CRP) levels, adipose tissue and metabolic alterations have not been clearly established in healthy non-obese subjects. We investigated the relationships between body fatness, CRP levels and metabolic variables in healthy, non obese sons of patients affected from the Metabolic Syndrome (MS). Age, CRP and Interleukine 6 (IL-6) levels, anthropometric measures (Body Mass Index, BMI; Waist circumference and Waist-to-Hip Ratio, WHR), total and regional fat content (determined by dual X-ray absorptiometry, DXA), total and LDL cholesterol and metabolic variables related with MS (HDL-cholesterol, triglycerides, glucose and insulin levels, Fasting Insulin Resistance Index, FIRI; blood pressure) were evaluated in 85 healthy non-obese sons of patients affected from the MS. Linear and multiple regression analyses were performed to evaluate the associations between body fat, metabolic variables and CRP levels, and whether the association between body fat content and metabolic variables persist after adjustment for CRP levels. Body fat was associated with all of the variables investigated. CRP levels were associated with total and regional body fat, with anthropometric index of weight, with age and with some metabolic alterations (HDL-cholesterol, triglycerides, systolic blood pressure, fasting insulin and LDL-cholesterol). The associations between total body fat and metabolic variables were not modified after adjustment for CRP levels. Total body fat was the best predictor of CRP levels ($p<0\cdot 0001$). In healthy, non-obese sons of patients affected from the MS, total body fat is the best predictor of CRP levels and it remains strongly associated with metabolic abnormalities after adjustment for CRP levels. These findings strongly support the hypothesis that at least in healthy subjects, body fat is the main determinant of metabolic abnormalities and low inflammatory state.

BODY FAT IS THE MAIN PREDICTOR OF FIBRINOGEN LEVELS IN HEALTHY NON-OBESE MEN.

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Previous studies have demonstrated that circulating levels of C-Reactive Protein (CRP), a marker of cardiovascular risk, are strictly related with body fatness. Elevated fibrinogen levels are also predictive of future cardiovascular events. The metabolic background of this relationship and the predictors of fibrinogen levels have not been well established. We aimed to evaluate whether fibrinogen levels are associated with body fat content and distribution and which are the independent predictors of fibrinogen levels in a sample of healthy, non-obese, non-smoking young-adult men. Age, anthropometric measures (Body Mass Index, BMI; Waist-to-Hip Ratio, WHR), total and regional fat content (determined by dual X-ray absorptiometry, DXA), metabolic variables (total, LDL and HDL-cholesterol; triglycerides; glucose and insulin levels; Fasting Insulin Resistance Index, FIRI; blood pressure), Interleukin-6 (IL-6) and acute phase reactants levels (Fibrinogen, highly sensitive C-Reactive Protein, hs-CRP) were determined in 87 healthy non-smoking, non-obese subjects. Linear regression analysis was used to evaluate association between body fat, fibrinogen and metabolic variables, and multiple regression model analysis was used to examine the independent predictors of fibrinogen levels. Eighty-seven (30.5±3.5 years) non-obese (mean BMI 24.1±3.5) men were studied. Fibrinogen levels were strongly associated with measures of body fat and with metabolic variables. Total body fat ($p<0.0001$) and LDL-cholesterol ($p<0.01$) were the independent predictors of Fibrinogen levels, accounting for 29.5% and 10.9% of its variance, respectively. Total body fat was the best independent predictor of hs-CRP levels, accounting for 32.5 % of its variance. We conclude that in healthy, non-obese subjects, body fat content is the main predictor of fibrinogen levels, as well of hs-CRP levels. These findings support the speculation that there exists a direct mechanism by which adipose tissue might regulate the levels of circulating acute-phase reactants.

DISTURBANCES OF PHOSPHATE METABOLISM: ANOTHER FEATURE OF THE METABOLIC SYNDROME

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Despite the important recent advances in the understanding of the consequences of the metabolic syndrome, its pathophysiology remains unclear. It has been proposed that disturbances of phosphate metabolism may contribute to the development of this constellation of cardiovascular risk factors. However, so far there is only little clinical data supporting this hypothesis. The aim of our study was to confirm the presence of hypophosphatemia in patients with metabolic syndrome as well as to investigate the mechanisms that may underlie the disturbances of phosphate metabolism in this patients group. A total of 255 individuals were enrolled. The diagnosis of the metabolic syndrome was based on the recently released ATP III guidelines. Subjects with less than three criteria for the diagnosis of the metabolic syndrome were served as controls. Patients with metabolic syndrome exhibited significantly lower phosphate and magnesium concentrations as compared to control individuals. Since the fractional excretion values of phosphate was similar in both groups, we assume that hypophosphatemia in patients with the metabolic syndrome can be attributed to the decreased dietary intake as well as to the internal redistribution of this element. Lower magnesium values in the patient group may result from the same mechanisms as lower phosphate levels. In addition, the hyperinsulinemia-induced renal magnesium wasting may also be a contributory factor. In conclusion, patients with the metabolic syndrome exhibit significantly lower phosphate and magnesium levels as compared to healthy individuals. The clinical significance of these disturbances as well as their importance as targets for preventive or therapeutic interventions remains to be established.

PLASMA PROCARBOXYPEPTIDAZE U: A NOVEL COMPONENT OF THE METABOLIC SYNDROME

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Impairment of fibrinolytic function is a predisposing factor to coronary heart disease (CHD) and other atherothrombotic conditions. Carboxypeptidaze U or activated thrombin activatable fibrinolysis inhibitior (TAFIa) is a novel inhibitor of fibrinolysis, the clinical significance of which remains unknown.
The relationship of plasma procarboxypeptidaze U (proCPU) concentration to clinical and metabolic risk indicators for CHD were examined in 78 middle-aged healthy men with an apolipoprotein E3/E3 genotype. Plasma proCPU was determined by a method based on quantitative activation of the zymogen by thrombin-thrombomodulin complex, followed by determination of total enzymatic activity of CPU with the substrate hippuryl-L-arginine, using high performance liquid chromatography-based determination of the released hippuric acid.
Results. Among clinical risk indicators, body mass index BMI ($p<0.001$) and waist-to-hip circumference ratio ($p<0.05$) were significantly correlated with plasma proCPU concentration. Correlation analysis with metabolic risk indicators determined in the fasting state revealed a strong positive relation of proCPU with VLDL triglycerides ($p<0.001$). Other metabolic markers that are perturbed in the insulin resistance syndrome, such as LDL particle size ($p<0.05$), blood glucose ($p<0.05$), HOMA estimate of insulin resistance ($p<0.05$), plasma proinsulin ($p<0.05$), and LDL triglyceride content ($p<0.01$) were also significantly correlated with the plasma proCPU concentration. ProCPU also showed consistent positive correlations with plasma concentrations of large VLDL, large and small chylomicron remnants in the fasting state and postprandially after intake of a mixed meal.
The final multivariate model showed BMI to be strongest determinant of proCPU concentration, accounting for 26% of the variation, whereas fasting plasma triglycerides proved to be the strongest metabolic correlate, contributing another 12% of the variation.
Conclusion. ProCPU can be considered as a component of the metabolic syndrome.

ADIPONECTIN AS A BIOMARKER OF THE METABOLIC SYNDROME

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Adiponectin possesses anti-atherogenic and insulin-sensitizing activities. Significance of adiponectin in the metabolic syndrome was investigated in 661 middle-aged adults (479 men, 182 women). Plasma adiponectin levels correlated negatively with waist circumference, visceral fat area, serum triglyceride level, fasting plasma glucose, fasting plasma insulin, and systolic and diastolic blood pressure in both sexes. Positive correlation was found between plasma adiponectin and HDL cholesterol levels in both sexes. The mean number of components of the metabolic syndrome increased with the decrease of plasma adiponectin level. The number was 2.57 ±1.34 for men and 2.00 ±1.51 for women with adiponectin levels <4.0 μg/ml. In all, 52.3% of men and 37.5% of women with adiponectin levels <4.0 μg/ml fulfilled the criteria for the metabolic syndrome. Our findings indicate that hypoadiponectinemia is closely associated with the clinical phenotype of the metabolic syndrome and suggest that measurement of plasma adiponectin may be useful for management of the metabolic syndrome.

APOLIPOPROTEIN B48 IS A CRUCIAL MARKER OF METABOLIC SYNDROME.

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Postprandial hyperlipidemia is known to be a risk factor of atherosclerotic diseases. However, we have difficulties to define the postprandial hyperlipidemia, because we do not have a good assay system to make the diagnosis. Recently, we developed the assay system of apolipoprotein B48 (apo B48), which is a specific apolipoprotein of chylomicron in human. On the other hand, metabolic syndrome is focused recently as an atherogenic disease entity. However, we do not have a crucial criterion for the metabolic syndrome. According to the criteria of WHO, glucose intolerance is one of major clinical criteria. The subjects with glucose intolerance are known to show postprandial hyperlipidemia, and the postprandial hyperlipidemia is reversed by the treatment of glucose intolerance. The purpose of our study is to determine whether it is possible to estimate the metabolic syndrome by fasting plasma level of apo B48. Methods: A monoclonal antibody (4C8) was raised against apo B48 C-terminal specific decapeptide. We developed the assay system of plasma apo B48 by ELISA with use of the antibody, 4C8. No cross-reactivity was found with apo B100. Intra- and interassay CVs were 4.4% and 3.0%, respectively. We measured plasma apo B48 level and LDL size and other lipid parameters of continuous 112 subjects who were received coronary angiography in our hospital. We found 84 subjects with prominent coronary stenosis (more than 75%). We divided the subjects into two groups with or without metabolic syndrome according to the criteria of NCEP-ATP III except for waist circumference. Results: The subjects with metabolic syndrome were about 20% of those with coronary stenosis. Although there were no significant differences between the groups with and without metabolic syndrome in total cholesterol and LDL-cholesterol levels, LDL size was smaller (p=0.0003) and fasting apo B 48 level was higher (p=0.0007) significantly in those with metabolic syndrome. Discussion: With use of assay of apo B48, we may be able to estimate metabolic syndrome, and it is possible to find the new drugs to reduce apo B48, as a drug for metabolic syndrome.

ENDOTHELIAL FUNCTION AND SERUM LEVELS OF INFLAMMATORY MARKERS IN METABOLIC SYNDROME PATIENTS WITH HYPERCHOLESTEROLAEMIA

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Background: The interaction between metabolic syndrome (MS) and hypercholesterolaemia (HC) in terms of inflammation and endothelial dysfunction is less clear. We compared the serum levels of inflammatory markers and endothelial function in patients with HC and normocholesterolaemia (NC), with and without MS. ***Materials and Methods:*** A total of 316 subjects with primary HC (n=119) and NC (n=197) were studied. HC and NC were defined as total cholesterol levels of ≥6.5 and <5.2mmol/L respectively. MS was defined according to the Adult Treatment Panel III National Cholesterol Education Program (NCEP-ATPIII) criteria but with lower cut-offs for waist circumference (>80cm for females, >90cm for males). Subjects were divided into 4 groups: HCMS+, HCMS-, NCMS+, NCMS-. Anthropometric indices, blood pressure, fasting plasma glucose, serum levels of lipid profile, highly sensitive C-reactive protein (hsCRP), interleukin 6 (IL6) and soluble intercellular adhesion molecule-1 (sICAM-1) were determined. Endothelial function was assessed by brachial artery flow mediated dilatation (FMD). ***Results:*** The HCMS+ had higher hsCRP ($p<0.02$) and IL6 ($p<0.05$) levels compared to the HCMS- group. The NCMS+ had higher hsCRP ($p<0.001$) and lower FMD ($p<0.005$) compared to the NCMS- group. sICAM-1 levels were higher in the HCMS+ than the NCMS+ ($p<0.0001$). The HCMS- compared to the NCMS- had higher levels of IL6 ($p<0.005$) and sICAM-1 ($p<0.0001$), but lower FMD ($p<0.0001$). ***Conclusion:*** Both HC and MS are associated with increased inflammatory state and endothelial dysfunction. There is further enhancement of inflammation by MS in HC patients. In NC patients, MS is associated with increased inflammatory state and endothelial dysfunction. MS may have an important role in accelerating atherogenesis in both HC and NC patients.

SERUM LEVELS OF FIBRONECTIN AND ENDOTHELIN-1 IN PATIENTS WITH METABOLIC SYNDROM

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Endothelial dysfunction is implicated in the pathogenesis of metabolic syndrome. Cellular fibronectin is an endothelium-derived protein involved in subendothelial matrix assembly. Endothelin-1 is the potent endothelium-derived vasoconstrictive and proliferative factor. However, an association between fibronectin and endothelin-1 production, and metabolic syndrom remains not fully understood. Therefore, the aim of the study was to investigate serum levels of fibronectin and endothelin-1 in patients with metabolic syndrom, find also interrelation between these factors. We studied 53 patients with metabolic syndrom (age: 56.8+2.2 years, BMI: 35.3+0.8 kg/m2; waist/hip ratio (WHR) - 0.98+0.03; data everywhere are presented as mean+SEM), 16 control subjects (age:47.4+1.4 years, BMI: 24.0+0.6 kg/m2; WHR - 0.84+0.02). Serum fibronectin, endothelin-1 levels were determined by immunoenzyme assay. Statistical analysis was performed by Student's paired and Pearson's correlation tests. We found an increase of serum fibronectin, endothelin-1 levels in patients with metabolic syndrom compared to control subjects. In patients with metabolic syndrom and control subjects fibronectin serum level were – 356.8+19.54 and 226.81+37.46 pmol/L($p<0.001$), endothelin-1 - 8.09+0.51 and 4.38+0.31 pmol/L($p<0.001$) respectively. Moreover, it was a significant elevation of serum fibronectin and endothelin-1 levels in in patients with metabolic syndrom compared to to control subjects. In obese patients with diabetes it was significant correlation between fasting glucose, BMI and fibronectin levels($r=0.62$, $p<0.05$ and $r=0.78$, $p<0.05$, respectively) and endothelin-1 levels ($r=0.51$, $p<0.05$ and $r=0.68$, $p<0.05$, respectively); between fibronectin and endothelin-1($r=0.48$, $p<0.05$). The revealed changes of fibronectin, endothelin-1 serum levels could reflect an endothelial dysfunction in patients with metabolic syndrom, which is the most pronounced in those with diabetes. Hyperglycemia and obesity appear to be significant factors contributing to elevation of fibronectin, endothelin-1 production in patients with metabolic syndrom.

ASSOCIATION OF METABOLIC SYNDROME WITH NOVEL CARDIOVASCULAR RISK FACTORS IN WOMEN WITH PREVIOUS GESTATIONAL DIABETES MELLITUS.

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Women with previous Gestational Diabetes Mellitus (pGDM) are at risk for developing type 2 diabetes and frequently show components of the insulin resistance syndrome that could contribute to cardiovascular risk. Aim of this study was to evaluate the rate of MS and associated novel cardiovascular risk factors in pGDM women.
We studied 166 pGDM women and 98 controls (CON) 16 months after delivery. Fasting plasma glucose (FPG), insulin, lipid profile, fibrinogen, high sensitive C-reactive protein (CRP), omocystein, serum uric acid, blood pressure and anthropometric parameters were determined. HOMA-R was used to estimate insulin resistance and metabolic syndrome (MS) was defined by NECP-ATP III criteria.
The two groups were comparable for age (34.7±4.2 vs 33.9±3.9 years), parity (56.2 vs 51.4%), and BMI (25.4±5.4 vs 24.2±4.2 kg/m^2). pGDM compared to CON were more insulin-resistant (HOMA-R 2.18±2 vs 1.35±0.6; p<0.05). The prevalence of MS determinants was higher in pGDM than in CON: high FPG 7.8 vs 0% (p<0.005), low HDL 37.3 vs 17.3% (p<0.01), hypertriglyceridemia (9.63 vs 2%; p<0.01), abdominal obesity (34.3 vs 18.3% (p<0.01), while high blood pressure values did not differ (5.4 vs 7.1%; ns). The prevalence of MS was 9.03% (n=15) in pGDM and 1.02% (n=1) in CON (p<0.01). Women with MS (MS+, n=16) compared to those without MS (MS-, n=248) showed significantly higher fibrinogen (348.7±54 vs 311.4±70 mg/dl p<0.05), serum uric acid (4.7±1.2 vs 4±0.8mg/dl p<0.01), CRP (6.1±5.03 vs 1.7±2.3 mg/dl p<0.001), HOMA-R (4.1±2.3 vs 1.8±1.1; p<0.001), while omocystein levels were not different (10±5.8 vs 8.03±2.9 mmol/L p=0.08). Moreover among subjects without any determinant of MS, those with pGDM (n.70) showed higher levels of CRP compared to CON (n.62) (0.99±1.1 mg/dl vs 0.39±0.3 mg/dl; p=0.05). In the whole group CRP concentrations increased progressively according to the number of the determinants of MS: no criteria 1.03±1 mg/dl; 1-2 criteria 2.4±2.8 mg/dl; 3 criteria 6.1±5 mg/dl (p<0.0001). In addition CRP levels were positively correlated to FPG (r^2=0.041, p<0.01) and insulin (r^2=0.21, p <0.001). This study shows that MS is present in a sizable proportion in pGDM women and is associated with novel cardiovascular risk factors (fibrinogen, serum uric acid and CRP). Levels of CRP tend to increase accordingly to the number of determinants of MS. The identification of MS in women with pGDM should be extensively sought in order to reduce the impact of cardiovascular risk.

SULFATIDES AS MARKERS OF DEVELOPMENT DIABETIC PERYPHERIC NEUROPATHY

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Sulfatides are one of representatives of glycosphingolipids presenting on external surface of cell plasma membranes of human organism. Together with glycoproteins they participate in processes of cell-cell recognition, differentiation, cell adhesion and immunochemical events. In latest publications on question discuss possibility to using of sulfatides as markers dynamics of development vascular complications of diabetes mellitus. The aim of this investigation is study dynamics of alteration content of sulfatides in blood serum healthy individuals and patients with different stage diabetic neuropathy. Obtained data show that in blood serum of patients with diabetes mellitus observed increase level of sulfatides. While this glycolipid is not detectable in blood serum of healthy individuals. Containing of sulfatides in blood serum of diabetic patients is correlated with indexes of glycated hemoglobin and coagulation. Conducted investigations permit to make conclusion that containing of sulfatides in blood serum upon diabetes mellitus has direct correlation with stage of development diabetic neuropathy and propose using of sulfatides as biochemical markers of development diabetic neuropathy.

CONCENTRATION OF GLUCATED PROTEINS IN URINE AS A PARAMETER FOR RENAL DAMAGE IN DIABETIC PATIENTS

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Glycated proteins (GP) are found in the blood and as well as in the urine of patients with Diabetes Mellitus (DM). The clinical significance of their concentrations are not enough explored. Beside HgA_{1C}, other glycated proteins, especially proteins with small molecular mass (albumins) easaly pass the glomerular membrane. In conditions of reduced reabsorption they should be found in an increased quantity in urine. In our study, we determined the concentration of glycated proteins (without HgA_{1C}) in urine in the patients with Chronicall Renal Disease and DM with increased markers of damage of the glomerular membrane and tubular reabsorption (microalbuminuria and α_1 microglobulin). Concentrations of the GP in the urine of these patients are significantly higher (p<0,05) in compare with the control group. But, there is no correlation between their concentration and the concentrations of albumins and α_1 microglobulins in the urine. We determined GP in urine with our modification of photometric method with nitrotetrazolium-blue (NBT) with reagents HOFMAN LA ROCHE (fructosamine), microalbuminuria and α_1 microglobulimuria with DAKO tests

ARE THE VALUES OF APOLIPOPROTEIN /APO A-I / AND HDL CHOLESTEROL SIGNIFICANT PREDICTORS FOR DEVELOPMENT OF PERIFER VASCULAR DISEASE IN NIDDM PATIENTS

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It is known that dislipidemia in non-insulin dependent diabetes mellitus /NIDDM/ is one of the leading factors for development of atherosclerosis but it is less known whether dislipidemia is the only cause and to what extend it influences PVD . The aim of this study is to estimate if there is any difference in degree of serum lipids and lipoproteins fractions and if there is any these should be compared within the following groups A /NIDDM patients with lower limb occlusive disease/, group B / NIDDM without macroangiopathy/ and control group C of non-diabetic patients with lipid disorder but without macroangiopathy
We have examined lipid parameters in 41 patients of group A , 38 of group B and 43 of group C. The examined groups included only men of the same age /56-60/, non-smokers, without arterial hypertension. T he patients of groups A and B had suffered from diabetes for almost the same period of time / 8-10 years/, were under constant medical control /HbA1c 6,8+/-0,3%/ and without any significant disorder of lipids. Lipid parameters were estimated by measuring total HDL and LDL cholesterol /Ch/ and triglycerides by enzymatic methods. ApoA1 and lipoprotein /Lpa/ were measured by nephelometry.
Comparing lipid and lipoprotein values these three groups we have concluded that there was a significant decrease in HDL cholesterol / A 1.2057+/-0.2596, B 0.9714+/-0.3132, C 1.3650+/-0.2143/ and ApoA1 serum levels / A 1.3081+/-0.3272, B 1.4401+/-0.2973, C 1.9809+/-0.927 / in group A and B when compared to control group C p<0.01, but there was no significant difference between groups A and B p>0.05. The values if cholesterol / A 5.639+/-1.346 B 5.73+/- 1.2113, C 6.83+/-2.11 / and triglycerides / A2.013+/-0.123, B 1.99+/-0.023, C 3.61+/-0.945/ are lowered in groups A and B in comparison to group C <0.01 and there was no significant difference between group A and B p > 0.05. The values of cholesterol / HDL, LDL , ApoA1 and Lpa p>0.05 did not differ within these three groups
Based on the achieved results we can ask the same question whether ApoA1 and HDL cholesterol are the significant predictors for development of PVD because we have found such low values of these parameters in groups A and B

DETERMINATION OF HAPTOGLOBIN AT WAGNER CLASSIFICATION OF DIABETIC'S ULCERATIONS ON THE FOOT (DUF)

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Goal: To estimate the effect when we use the Wagner classification (W-C) in the treatment of diabetic's ulcerations on the foot, with following the haptoglobin values. Haptoglobin is a sérum protein that functions as an antioxidant by virtue of its ability to bind to haemoglobin and thereby to prevent the oxidative tissue damage that may be mediated by free haemoglobin. It has been recently found that haptoglobin phenotype is a predictor of the risk of micro vascular and macro vascular complications in diabetes.

62 patients (58±3.1), 30 male and 32 female with diabetes mellitus (NIDDM treated with insulin) and diabetic foot disease were classified under the 5- stage of Wagner classification, which is based on lesions deepness, gradient of infection and gangrene extension. 0- stage: Without open lesion-10 patients (18,5%); 1-stage: Superficial ulcers –18 patients (33,3%); 2-stage: Deep ulcerations-12 patients (22,2%); 3-stage: Deep ulceration with abscess and osteomyelitis-10 patients (18.5%); 4-stage: Limited gangrene- 9 patients (16,6%); 5- stage: Expanded gangrene-3 patients (5,5%). All of them were examined with routine laboratory analyses, Doppler sonografy, and Colour duplex scanning and haptoglobin values measured by Automatic analyser Mira-Cobas ROCHE diagnostic.
We found that the level of haptoglobin is elevated in all cases, significantly associated with progression of diabetic foot disease. The obtained haptoglobulin values suggest that an increased oxidative stress in diabetic patients with macro vascular complications exists and helps in determination of the treatment protocol.

SOLUBLE CELLULAR ADHESION MOLECULES AND SMOKING IN PATIENTS WITH DIABETES MELLITUS

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Cellular cellular adhesion molecules (CAMs) may be involved in the development of atherosclerosis. The aim of the present study was to investigate the relation of serum CAMs to smoking habits in patients with diabetes mellitus (DM). Forty-three patients with type 1 DM and 38 patients with type 2 DM had fasting serum levels of soluble ICAM-1, VCAM-1 and P-selectin determined. Patients were divided into current smokers and non-smokers. No significant differences between mean CAMs in type 1 DM and type 2 DM were observed. Smokers had significant higher ICAM-1 (309±67 vs. 271±69, $p<0.05$) and P-selectin (96±32 vs. 77±24) than non-smokers. In a regression analysis, smoking was the only independent correlate of ICAM-1, whereas P-selectin was independently correlated to both smoking and haemoglobin A1C. VCAM-1 was only independently correlated to body mass index (beta=0.308, $p<0.01$). Smoking is significantly positively associated with both sICAM-1 and P-selectin in patients with DM. Thus, soluble CAMs may represent a biomarker of the accelerated atherogenesis associated with smoking.

EFFECTS OF PARAOXONASE POLYMORPHISMS AND ACTIVITY ON OXIDIZED LDL IN TYPE 2 DIABETES.

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Among mechanisms that may explain the anti-atherogenic properties of HDL, paraoxonase (PON1) has raised particular interest. PON1 inhibits the oxidation of LDL (oxLDL) in vitro. We determined serum PON1 activity and two PON1 polymorphisms (L/M, Leu55Met and Q/R, Gln192Arg) in 317 patients type 2 diabetes (DM2) and 106 controls (C) and related them to oxLDL. PON1 activity did not differ in DM2 and C (66±41 vs 58±44 nmol/min/ml, $p=0.09$). PON1 activity does not change with age, DD or BMI, while inversely correlates with HbA1c ($r=-0.152$, $p=0.018$). Subjects with PON activity in the two higher quartiles have HbA1c lower than those in the first quartile ($p=0.03$). In DM2 the L55 allele frequency was 0.57 (M55: 0.43) and that of the Q192 allele 0.66 (R192: 0.34), similar to those of C. As far PON1-192, the QR genotype was the most common (50.8%) followed by QQ (41.0%) and RR (8.2%); for PON1-55, the LM genotype was the most common (67.6%) followed by LL (23.4%) and MM (9.0%). PON1 activity by genotype was the same in both groups; for PON1-55, LL>LM>MM and for PON1-192, RR>RQ>QQ with an addictive effects of the two polymorphisms. OxLDL did not differ in C (49.0±11.1) and DM2 (55.6±25.3 U/L, $p=0.16$). PON1 activity does not correlate with lipids other than ApoA1 ($r=0.17$, $p<0.005$), and was not related to oxLDL ($r=0.05$, $p=0.35$). No differences in oxLDL were observed by quartiles of PON1 activity. The PON1-192 polymorphism did not affect lipid profile, while carriers of the M allele of PON1-55 showed lower HDL ($p<0.05$) and ApoA1 ($p<0.03$). Neither PON1-192, nor PON1-55 affect oxLDL levels. In conclusion, our data suggest that, in type 2 diabetes, variations in PON1 activity and PON1 gene polymorphisms (that regulate PON1 activity) does not affect oxidation of circulating LDL.

CHARACTERIZATION OF ANTIOXIDANTS AND ELUCIDATION OF OXIDATIVE MECHANISMS OF TISSUE DAMAGE IN DIABETES MELLITUS

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Oxidative stress in humans and animals is defined as the imbalance between free radicals and antioxidants. This condition can be found in diabetic patients. Free radical reactions play the important role of initiating immune system response, but if overproduced, cause tissue injury and cell death (Roesen 2002). Diabetes can be treated, but patients may suffer from long-term complications, such as disease of the eyes, nervous system, heart, blood vessels and kidneys. Research of the role of antioxidants in diabetic tissue and plasma is still of great interest and will be undertaken by the use of cyclic voltammetry (CV), high performance liquid chromatography and mass spectrometry (HPLC, HPLC-MS) methods.
Water soluble (vitamin C and uric acid) and lipid soluble (vitamin E and ß-carotene) low molecular weight antioxidants are objectives of further investigation by CV and HPLC-MS measurements.
A range of analytical techniques will be presented relevant to characterizing oxidative stress in Diabetes Mellitus. These will be applied to drug trials, including tissue from animal models with samples representing control, induced-diabetes, drug-applied prior to diabetes, and drug applied once diabetes has been induced. The information gained will be added into the evaluation of the effectiveness of the treatments being developed in the field of heart disease.

Roesen, P et al. Handbook of Antioxidants 2002. "25 Vascular Complications in Diabetes: Mechanism and the Influence of Antioxidants". 511-533

PLASMA LIPIDS AND OXIDIZED LDL LEVELS BY APO-E POLYMORPHISM IN TYPE 2 DIABETES MELLITUS.

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Variation in the ApoE gene, coding for the three isoforms E2, E3 and E4 affects plasma lipids. Both primary (cysteine content) and tertiary structure of Apo E might account for different antioxidant capacity (E2>E3>E4). We evaluated ApoE genotypes and alleles in 605 type 2 diabetics (age 61±8, age at diagnosis of diabetes 48±12, duration of diabetes (DD) 13±10 years, BMI 28.7±5.3 kg/m^2, HbA1c 8.7±1.8 %) and the contribution of ApoE polymorphism on lipid profile and oxidized LDL (ox-LDL). Genotype distribution E2E2-E2E3-E3E3-E2E4-E3E4-E4E4 was 0.5/11.9/76.8/0.8/9.7/0.3% and allele frequency 0.876, 0.068 and 0.056 for E3, E2 and E4, respectively. Patients features, lipid profile and ox-LDL were evaluated in E3 carriers (E3c = E3E3, n. 466) versus E2 carriers (E2c = E2E3 and E2E2, n. 74) and E4 carriers (E4c = E4E3, E4E4 and E2E4, n. 65). The three groups were similar in age, DD, BMI, HbA1c, and blood pressure. E2c patients showed lower total (E2c: 190±42, E3c: 205±42 and E4c: 200±43 mg/dl, p<0.04), LDL (E2c: 111±32, E3c: 127±35 and E4c: 128±34 mg/dl, p<0.005) and non-HDL-Ch levels, while E4c had the lowest HDL (E2c: 48±12, E3c: 47±14 and E4c: 42±10 mg/dl, p<0.05). No differences were found in triglyceride levels (E2c: 154±91, E3c: 152±86 and E4c: 158±90 mg/dl, p=0.92). Ox-LDL were significantly different in the three groups (E2c: 46±22, E3c: 55±25 and E4c: 60±27 U/L, p=0.006), and correlated with total-, LDL-, non-LDL-cholesterol and triglycerides. Logistic regression analysis performed after ox-LDL stratification in quartiles did not highlight any independent role of Apo-E polymorphism on LDL oxidation. In conclusion, in type 2 diabetes ApoE polymorphism might induce either protective (E2c: low LDL-Ch and low ox-LDL) and high risk (E4c: high LDL-Ch and high ox-LDL) lipide profile. In type 2 diabetes, ApoE polymorphism does not exert any independent effect on LDL oxidation.

LDL OXIDABILITY IN OBESITY WITH AND WITHOUT DIABETES : A POSSIBLE ROLE FOR INSULIN-RESISTANCE?

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LDL modified by oxidative processes have been found in the course of arteriosclerosis and type 2 diabetes, conditions often associated to obesity. The susceptibility of LDL to oxidation depends on several factors, among which insulin-resistance may also play a role.
Aim of the study was to investigate differences in oxidability of LDL isolated from plasma of obese patients, with or without diabetes, compared to healthy control subjects. Seven obese type 2 diabetic patients (age 44±5; BMI 34.1±2.7 Kg/m^2; plasma cholesterol 175.6±26.9 mg/dl; triglycerides 119.8±58.1 mg/dl; HDL-cholesterol 35±6.3 mg/dl) and nine obese non diabetic patients (age 44±9; BMI 35.4±2.6 Kg/m^2, plasma cholesterol 190.1±33 mg/dl, triglycerides 101,7±39 mg/dl, HDL-cholesterol 43.9±12.6 mg/dl) were compared to five age-matched, normal-weight, healthy subjects (age 41±3; BMI 23.9±1.7 Kg/m^2; plasma cholesterol 163.2±28.7 mg/dl; triglycerides 102.0±73.2 mg/dl; HDL-cholesterol 41.6±11.4 mg/dl) as regards LDL oxidability (lag-time and TBARs production) after exposure to copper ions. The two groups of patients were comparable for what concerns insulin-resistance as shown by the glucose infusion rate during hyperinsulinemic euglycemic clamp (M value 4.7±1.4 and 4.0±1.4 mg/Kg/min), while they were significantly more insulin resistant than control subjects (M value 7.5±2.6 mg/Kg/min, p<0.05).
Lag-phase values, calculated by monitoring conjugated dienes formation in LDL oxidized by 10 μM $CuSO_4$, did not show any differences between the two patient groups, while they were significantly lower in patients with respect to the controls (60.1±7.8 vs 87.0±11.4 min; p<0.0001).
TBARs production after LDL oxidation by metal ions was significantly higher in obese patients than in controls (64.6±5.1 vs 42.4±4.1 nM/mg of LDL protein, p<0.05), but no differences were observed between diabetic and non-diabetic obese patients.
These results provide evidence that LDL particles are highly susceptible to oxidation in conditions characterized by insulin-resistance, suggesting the presence of an in vivo relationship between obesity, insulin- resistance and LDL oxidized state, while no independent effect of diabetes is observed.

LIPID METABOLISM, C-PEPTIDE AND IMMUNOREACTIVE INSULIN LEVELS IN FEMALES AFTER OVARIECTOMY

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Background. It was documented that in older females the rate of lipid metabolism disorders was increased. It was assumed that these disorders were related to menopause and might be to the development of atherosclerosis. Moreover, it was in older age that carbohydrate metabolism disorders were more frequently encountered. The relevance of these statements to young females after ovariectomy was the subject of this study.
Aim. The aim of the work was to study lipid metabolism, concentrations of C-peptide and immunoreactive insulin in females after ovariectomy.
Material and Methods. Twelve females with "surgical menopause" (after ovariectomy) aged 21 to 42 years were studied in on average 1.5 years after the operation. The control group was composed of 24 females of the same age. Cholesterol, triglycerides, lipoprotein fractions were investigated. Student's t-criterion was used to assess the significance of difference between the groups.
Results. In females after the surgery the content of triglycerides and cholesterol was significantly increased compared to the control group - 2.374±0.040 against 1.373±0.038 mmol/l and 5.92±0.12 against 4.45±0.03 pmol/l, respectively (P<0.01). Concentrations of low density lipoproteins (41.841±0.209 g/l) and very low density lipoproteins (36.018±0.731 g/l) in females of the test group was also higher than that in the control group 35.250±0.59 g/l and 26.145±1.770 g/l, respectively. At the same time the content of high density lipoproteins was significantly lower in the ovariectomied females (21.28±1.2 g/l) than that in the control group (33.8±2.3 g/l) (p<0.02). The level of C-peptide didn't significantly differ in the groups – 2.473±0.478 mkg/l and 2.238±0.90 mkg/l, respectively. The level of immunoreactive insulin was significantly higher in the test group – 224.69±30.066 against 123.255±20.616 pmol/l (P<0.01). The discrepancy between C-peptide and insulin parameters was probably related to the detection of proinsulin in blood and was characteristic of early stages of carbohydrate metabolism disorders.
Conclusion. Therefore, the findings testified the effect of artificial menopause on lipid metabolism disorders and prerequisites to the development of atherosclerosis in the females. Along with it metabolic situation arised with hyperinsulinism and a decrease of biological activity of the hormone.

THE RELATIONSHIP BETWEEN DIABETIC MICROANGIOPATHY AND SURFACE TEMERATURE OF LEGS MEASURED BY THERMOGRAPHY AT REST

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Backgrounds: Microvascular complications determine the quality of life and prognosis of the diabetic patients. Thus, the early detection method of diabetic microangiopathy would be needed. Thermography (ThG) is a quick and non-invasive method to measure the temperature of the skin.
Methods: In the current study, we enrolled 33 patients with diabetes mellitus diagnosed within 3 years and 12 control subjects. All subjects were measured the surface temperature of both toe and instep by ThG. Diabetic patients were divided into 2 groups by the differences of the temperature between toe and instep, namely group N (< 2 degree) and group C (2 degree). Diabetic triopathy were also estimated.
Results: The number of subjects who showed significant (2 degree) difference of the temperature between toe and instep was greater in diabetic patients compared with normal control (82% vs. 33%, p < 0.05). In diabetic patients, there was no significant difference between group N and group C in regard to HbA1c, body mass index, ankle-brachial pressure index, and pulse wave velocity. Furthermore, no distinction in the incidence of the microangiopathy was seen between these 2 groups.
Conclusion: The significant difference in the temperature (2 degree) between toe and instep in ThG were seen more frequently in patients with diabetes mellitus than in normal controls. However, the difference of skin temperature measured by ThG was not associated with the existence of diabetic microangiopathy. Some other factor may play a role in the lower toe temperature in diabetic patients.

WHAT ARE THE MAJOR RISK FACTORS FOR TYPE 2 DIABETES DEVELOPMENT IN THE HISTORICAL COHORT OF THE BRISIGHELLA HEART STUDY? A 16-YEARS FOLLOW-UP

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Aim: The aim of this study is to evaluate and quantify the relative role of different type 2 diabetes risk factors in the long-term development of type 2 diabetes mellitus in an Italian rural population sample. Methods: The Brisighella Heart Study (1972-2003) is a prospective, population-based longitudinal epidemiological cohort involving 2939 randomly selected subjects, aged 14 to 84 years, resident in the Northern Italian rural town of Brisighella. For this study, we selected 1442 age-matched adult subjects (M:712, F:730) consecutively visited during the last four-yearly BHS surveys (1988-2004), with comparable baseline BMI, fasting plasma glucose and smoking habit. Patients affected by type 1 diabetes were previously excluded form the study. The Cox regression analysis will be used to determine the independent prognostic significance of age, BMI, FPG, Blood Pressure, triglyceridemia, and HDL-C for type 2 diabetes development on an 16-year long follow-up. Results: In our population sample, after 8 years of follow-up, age appears to be a significant predictor of type 2 diabetes when inserted alone in the model (p=0.007), but it is completely not relevant when adjusted for baseline BMI and or FPG. Among subjects affected by IFG, the diabetes incidence/year has been estimated to be 5.3% for men and 3.9% for women (p=0.009). In the whole population sample, basal glycaemia values under 110mg/dL have not shown to be significant long-term predictors of diabetes development, while BMI was the best predictor, especially in men. Final results after 16 years of follow-up (survey yet on-going) will be presented at the meeting. Conclusion: Our preliminary findings confirm the relevant role of IFG and BMI as predicting parameters of type 2 diabetes development in the Brisighella population. The evaluation of the best diabetes predictors in each population should help to identify effective approaches for prevention.

PREVALENCE AND THE RELATIVE ROLE OF THE MAIN PREDICTORS OF METABOLIC SYNDROME DEVELOPMENT IN THE HISTORICAL COHORT OF THE BRISIGHELLA HEART STUDY: A 16-YEARS FOLLOW-UP

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Background: It is well known that metabolic syndrome is associated to a very high cardiovascular disease risk and that its prevalence is generically very high in Western countries, however the main predictors of metabolic syndrome development are not yet clearly known for the different populations. Aim: The aim of our study is to evaluate the relative role of different metabolic variable in metabolic syndrome development. Methods: We have selected 690 subjects (348 men and 342 women) aged more than 65 years from the 1988 survey list of the Brisighella Heart Study for which all sampled data were available. Then, carrying out a step-wise logistic regression method, we have estimated the relative role of BMI, systolic and diastolic blood pressure, TC, HDL.-C, TG, basal glycaemia and uricemia for the development of metabolic syndrome in the next 16 years. Results: Among studied variables, uricemia was associated to a higher risk to develop metabolic syndrome in men (OR 1.10; CI 1,00-1,11), while basal glycaemia in women (OR 1.10; CI 1.03-1.13). On the contrary high HDL-C plasma level was associated to a lower risk to develop metabolic syndrome both in men (OR 0.89; CI 0.84-0.94) and women (OR 0.95; 0.91-0.99). Conclusion: On the basis of the results obtained in our 16-year long follow-up, it appears that HDL-C has a protective effect s it regards the development of metabolic syndrome in both sex. Basal glycaemia is a significant predictor of metabolic syndrome in women, while uricemia in men.

A RANDOMISED, DOUBLE-BLIND CLINICAL TRIAL ON THE LONG TERM EFFICACY OF PSYLLIUM HUSK VS. GUAR GUM ON THE CLINICAL FEATURES OF METABOLIC SYNDROME

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Background: Increasing dietary fibre intake is widely recommended as a safe and practical approach for cholesterolemia reduction and glycaemia control in diabetic subjects. Among dietary fibres, the soluble ones appear to be the most effective, however there are no direct comparison trials between them. Aim: The aim of this study is to compare the metabolic effect of high supplementation of pyllium husk and guar gum on metabolic syndrome affected patients. Methods: Sixty-four age- and sex matched non-smoking patients who complied to the NCEP-ATP III criteria for the metabolic syndrome diagnosis were recruited and randomised to the psyllium group or the guar group at a doses of 3.5 gr t.i.d. before meals, and followed for 6 months. The following parameters have been considered: BMI, waist circumferences, TC, HDL-C, TG, basal glycaemia, systolic and diastolic blood pressure. Results: While body weight decreased significantly in both group (-1.4±0.7 kg), no difference has been found as it regards waist circumference. The psyllium group experienced a significant reduction in systolic blood pressure (-4.1±0.5 mmHg), TC (-12.2±3.4 mg/dL), TG (-10.9±4.7 mg/dL), and basal glycaemia (-6.2±2.9 mg/dL), while the guar group only of TC (-9.9±4.8 mg/dL) and basal glycaemia (-6.4±3.7 mg/dL). Diastolic blood pressure and HDL-C plasma level were not modified by both treatment regimens (p>0.05). At the end of the supplementation period, 12.5% of subjects treated with psyllium and 3.12% of those treated with guar did not comply with the criteria for the metabolic syndrome diagnosis (p<0.05). The drop out rate was 6.25% in the psyllium group and 12.5% in the guar group (p<0.05), mainly because of gastrointestinal side effect. Conclusion: In our patients, psyllium supplementation appeared to be more efficacious and safe in controlling component of the metabolic syndrome.

EFFECT OF FLUVASTATIN 80 MG ON LIPID PROFILE IN PATIENTS WITH CORONARY HEART DISEASE AND METABOLIC SYNDROME

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Resistant dyslipidemia, associated with metabolic syndrome (MS) contribute largely to increased cardiovascular risk in individuals with insulin resistance. The response to lipid-lowering therapy in different types of dyslipidemia may be variant.

Objective: to assess the effect of Fluvastatin 80 mg on the lipid profile in patients with the metabolic syndrome.

Study population: 55 male and 33 female, aged 39 to 89 (mean age 61.3±11,1), with a diagnosis of coronary heart disease and MS. The control group was their 68 counterparts without metabolic syndrome.

Methods: Fluvastatin 80 mg tablets was administered for the period of 6 month. Complete lipid profile – total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C) were calculated.

Results: after 6 weeks of treatment, Fluvastatin significantly decreased plasma lipid levels both in MS and control groups and this effect persisted over the period of 6 months. LDL-C has been decreased in MS and control group by 25.6% and 30.6% respectively, while similar decrease in 29% of non-HDL-C has been observed in both groups. The target concentrations of LDL-C and non-HDL-C over the period of 6 weeks have been achieved in 52.3% and 34.1% of patients respectively.

Conclusion: Fluvastatin 80 mg for the treatment of atherogenic dyslipidemia in patients with MS is as effective and well tolerated as in their counterparts without insulin resistance.

ROSUVASTATIN HAS GREATER BENEFICIAL EFFECTS THAN ATORVASTATIN ON LDL-C, HDL-C AND APOLIPOPROTEINS A-I AND B IN SUBJECTS WITH THE METABOLIC SYNDROME

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COMETS (4522IL/0069) compared the efficacy of rosuvastatin (RSV) with atorvastatin (ATV) and placebo in patients with the metabolic syndrome. The primary end point was LDL-C reduction; other lipid assessments included HDL-C, apolipoprotein (apo) A-I, apo B and the apo B/apo A-I ratio. After a 4-week dietary lead-in period, statin-naïve metabolic syndrome patients (NCEP ATP III definition), with LDL-C 3.36 mmol/l (130 mg/dl), additional multiple risk factors conferring a 10-year CHD risk >10% but no evidence of CHD or other atherosclerotic disease or overt diabetes, were randomised (2:2:1) to receive RSV 10 mg, ATV 10 mg or placebo for 6 weeks, subsequently followed by RSV 20 mg, ATV 20 mg or RSV 20 mg, respectively, for 6 weeks. At 6 and 12 weeks, % change from baseline in lipids were compared across treatment groups by ANOVA. RSV provided significantly greater LDL-C lowering and significantly improved HDL-C and the apolipoprotein profile compared with ATV and placebo. All treatments were well tolerated.

	Least-squares mean change from baseline (%)				
	6 weeks			12 weeks	
	RSV 10 mg (n=164)	ATV 10 mg (n=155)	Placebo (n=78)	RSV 10/20 mg & placebo/RSV 20 mg (n=242)	ATV 10/20 mg (n=155)
LDL-C	–41.7	–35.7***	–4.1***	–48.7	–42.7***
HDL-C	+9.3	+4.8**	+2.2***	+10.5	+5.7**
Apo A-I	+5.9	+1.2***	+1.8**	+6.2	+3.2*
Apo B	–34.1	–30.7*	–3.3***	–40.7	–36.0**
Apo B/apo A-I ratio	–37.1	–30.9***	–4.3***	–43.6	–37.5***

*p<0.05, **p<0.01, ***p<0.001 versus RSV at same time point

EFFECTS OF ROSUVASTATIN AND ATORVASTATIN ON LDL-C, AND APOLIPOPROTEINS A-I AND B IN PATIENTS WITH TYPE 2 DIABETES: RESULTS OF THE URANUS STUDY

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The URANUS study (4522SE/0001) compared the efficacy of rosuvastatin (RSV) 10–40 mg with atorvastatin (ATV) 10–80 mg in patients with type 2 diabetes. The primary end point was LDL-C reduction; other lipid assessments included apolipoprotein (apo) A-I, apo B, and the apo B/apo A-I ratio. After a 6-week dietary lead-in period, 469 adult patients with LDL-C ≥3.3 mmol/l were randomised (1:1 blinded) to RSV 10 mg or ATV 10 mg for 4 weeks; doses were then up-titrated at 4-weekly intervals for 12 weeks (up to RSV 40 mg or ATV 80 mg) to achieve the 1998 European LDL-C goal of <3.0 mmol/l for patients with diabetes. At 4 and 16 weeks, changes from baseline in lipid parameters were compared across treatment groups by ANCOVA. RSV provided significantly greater LDL-C lowering and improved the apolipoprotein profile significantly more than ATV at initial doses (4 weeks), and after titration (16 weeks). All treatments were well tolerated.

	Least-squares mean change from baseline (%)			
	4 weeks		16 weeks	
	RSV 10 mg (n=232)	ATV 10 mg (n=231)	RSV 10–40 mg (n=221)	ATV 10–80 mg (n=220)
LDL-C	–47.6***	–38.5[a]	–52.3***	–45.5
Apo A-I	+2.6*	+0.8	+2.6**	–0.2
Apo B	–42.9***	–35.3	–45.2***	–40.1
Apo B/apo A-I	–43.9***	–35.4	–46.3***	–39.6

*p=0.05, **p<0.01, ***p<0.001 vs ATV at same time point; [a]n=232

In conclusion, RSV improved LDL-C and the apolipoprotein profile significantly more than ATV in patients with diabetes and dyslipidaemia.

LIPID-LOWERING EFFECT OF SIMVASTATIN IN PATIENTS OF TYPE 2 DIABETES MELLITUS

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BACKGROUND: The role of simvastatin in the management of dyslipidemia in patients with type 2 diabetes mellitus is very often evaluated. This study examinated effect of simvastatin in age, sex and body mass index matched patients with type 2 diabetes mellitus with dyslipidemia. METHODS AND RESULTS: An open-label, prospective study was conducted on 86 patients with type 2 diabetes mellitus, who had modareta glycemic control with a glycated hemoglobin < 9,5%. The lipid profile was re-evaluated after 8 and 16 weeks. The patients in the group were advised behavioral modification and given simvastatin. The starting dose was 10 mg at bed time. After 8 weeks of simvastatin therapy, a lipid profile was done. If the goal of lipids level was not achieved, the dose of simvastatin was increased to 20 mg at bedtime for another 8 weeks. In the group, there was a significant alteration in all lipid fractions, and the target levels were achieved in 80% of patients after 12 weeks. There was no significant alteration in glycemic control and liver functions. CONCLUSIONS: In our study, pharmacological therapy with statins should be resorted to in patients with type 2 diabetes mellitus who carry a high risk of coronary heart disease. Simvastatin is a safe and efficacious lipid-lowering drug.

THE EFFECT OF BASAL-BOLUS INSULIN THERAPY ON LIPOPROTEIN LIPASE MASS IN PREHEPARIN SERUM

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There exists lipoprotein lipase mass in preheparin serum (prehaparin LPL mass), even though the activity is scarcely found. We reported that low serum preheparin LPL mass levels were one of risk factors of acute coronary syndrome[1]. Furthermore, serum preheparin LPL mass levels in diabetic patients were significantly low, and increased by conventional insulin therapy[2]. To clarify the effect of basal-bolus insulin therapy (B-B therapy) on serum preheparin LPL mass levels, thirty two type 1 diabetes mellitus patients treated by insulin conventional therapy were measured serum preheparin LPL mass level, body mass index (BMI), HbA1c, total cholesterol (TC), triglyceride (TG), high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C) and LDL particle size after exchange into B-B therapy. Preheparin LPL mass was measured by the sandwich enzyme-linked immunosorbent assay (ELISA) using specific monoclonal antibody (Daiichi pure chemicals, Japan). LDL paticle size was decided by relative mobility value of peak of LDL fraction (LDL-Rm) detected by lipoprotein polyacrylamide gel disc electrophoresis. After exchange into B-B therapy, HbA1c significantly decreased and no significant change was observed in BMI, TC and LDL-C. TG decreased and HDL-C increased. Preheparin LPL mass significantly increased from 48.5 to 57.2 ng/ml ($p<0.05$), and LDL-Rm significantly decreased from 0.379 to 0.368 ($p<0.05$). In 21 subjects, whose total insulin dose a day were not changed before and after B-B therapy, HbA1c significantly decreased and no significant change was observed in BMI, TC, TG, HDL-C and LDL-C. Preheparin LPL mass significantly increased from 46.5 to 54.5 ng/ml ($p<0.05$), and LDL-Rm significantly decreased from 0.384 to 0.373 ($p<0.05$). These results suggested that preheparin LPL mass was greatly regulated by insulin action, and spiked increase of serum insulin concentration might play an important role in LPL production.

References: 1) Hitumoto et al. atherosclerosis 2000; 153: 391-6
2) Miyashita et al. Diabetes Res Clin Pr 2002; 56: 181-7,

SERUM LEVELS OF APOLIPOPROTEINS AND LEPTIN IN PATIENTS WITH TYPE 2 DIABETES MELLITUS BEFORE AND AFTER TREATMENT WITH SIMVASTATIN

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There are convincing evidences of beneficial role of correction of dyslipidemia with statins in reduction of cardiovascular morbidity and mortality in patients with diabetes. However, not all mechanisms of positive action of statins are fully understood. Therefore, the aim of this study was to investigate the influence of treatment with simvastatin on serum levels of apolipoproteins A and B and leptin in patients with type 2 diabetes. We studied 20 patients with type diabetes mellitus (age 59.1±1.69 years old, BMI – 31.9±1.07 kg/m^2, diabetes duration - 5.6±1.25 years, data are presented as mean±SEM) who were treated simvastatin 20 mg once daily for 6 weeks. Serum cholesterol, LDL, triglycerides, apoipoprotein A and B and leptin levels were measured before and after treatment with simvastatin. Statistical analysis was performed by Student's paired test. Treatment with simvastatin resulted in significant reduction of total cholesterol (6.97±0.22 vs. 4.97±0.20 mmol/L, p<0.05), LDL (4.62±0.19 vs. 2.92±0.18 mmol/L, p<0.05), triglycerides serum levels (2.79±0.20 vs. 1.79±0.13 mmol/L, p<0.05), and an increase of HDL (1.08±0.06 vs. 1.24±0.05 mmol/L, p<0.05 before and after treatment, respectively). Moreover, treatment with simvastatin led to significant increase of serum apoliporotein A levels – 144.08±5.82 vs. 158.98±4.80, p<0.05 and decrease of apoliporotein B levels - 148.88±7.99 vs. 120.62±8.02, p<0.05 and apoB/apoA lipoproteins ratio - 1.06±0.06 vs. 0.77±0.06, p<0.05 before and after treatment, respectively. It was trend toward decrease of serum leptin levels as the result of simvastatin use - 7.74±1.52 vs. 6.61±1.18, p=0.07. We may conclude that the treatment with simvastatin resulted in improvement of dyslipidemia, positive changes of serum apolipoproteins levels with the trend toward reduction of serum leptin level. These changes could contribute to the positive effect of simvastatin on the reduction of cardiovascular risk in patients with type 2 diabetes mellitus.

METABOLIC EFFECTS OF FLUVASTATIN 80 mg AND ATORVASTATIN 20 mg IN TYPE 2 DIABETES MELLITUS AND LOW HIGH-DENSITY CHOLESTEROL. A 4-MONTH TRIAL

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The central role of decreased serum HDL-C level in diabetic cardiovascular disease has prompted the establishment of a target of 50 mg/dL in patients with diabetes mellitus (DM). Fluvastatin (XL) 80 mg (F) or atorvastatin 20 mg (A) were evaluated in 100 patients (45-71 years) with type 2 diabetes and low levels of serum HDL-C in 4-month trial (one month dietary run-in). F decreased mean (SD) LDL-C from 149 (33) to 95 (25) mg/dL (36%; $P < 0.01$), TG from 437 (287) to 261 (164) mg/dL (40%; $P < 0.01$), and increased HDL-C from 41 (7) to 46 (10) mg/dL (12%; $P < 0.05$). Apo A-I increased from 118 (18) to 124 (15) mg/dL (5%; $P < 0.05$) and apo B decreased from 139 (27) to 97 (19) mg/dL (30%; $P < 0.05$). A decreased LDL–C from 141 (25) to 84 (23) mg/dL (40%; $P < 0.01$) and TG from 411 (271) to 221 (87) mg/dL (46%; $P < 0.01$). Neither HDL-C (41 [7] vs 40 [6] mg/dL; 2%) nor apo A-I (117 [19] vs 114 [19] mg/dL; 3%) changed. Apo B decreased from 131 (20) to 92 (17) mg/dL (30%; $P < 0.05$). Mean changes in HDL-C (+5 [8] vs –1 [2] mg/dL; $P < 0.01$) and apo A-I (+6 [18] mg/dL vs –3 [21] mg/dL; $P < 0.01$) were significantly greater in F group. Both F and A achieved mean serum LDL-C (100 mg/dL) and apo B target levels (100 mg/dL) in the majority of patients with type 2 DM, but mean serum HDL-C level was increased significantly only with fluvastatin—16 patients (32%) in the fluvastatin group compared with none in the atorvastatin group achieved HDL-C levels 50 mg/dL. The increase in HDL-C in the fluvastatin-treated patients was associated with an increase in apo A-I, suggesting a potential pleiotropic and selective effect in patients with low HDL-C levels.

TWO YEAR STATIN THERAPY DOES NOT ALTER THE PROGRESSION OF INTIMA-MEDIA THICKNESS IN TYPE 2 DIABETES MELLITUS WITHOUT MANIFEST CARDIOVASCULAR DISEASE

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Background and Aims: Cardiovascular disease is the most important cause of mortality in patients with type 2 diabetes mellitus (DM2). We aimed to determine the effect of statin therapy versus placebo on the progression of carotid Intima-Media Thickness (IMT) in DM2 patients without manifest cardiovascular disease.
Materials and Methods: A randomised, placebo controlled, double blind clinical trial was performed in 250 patients with DM2. Patients were given either 0.4 mg cerivastatin daily, or placebo daily. At August 2001, when cerivastatin was withdrawn from the market, cerivastatin 0.4 mg was replaced by 20 mg simvastatin, without deblinding the study. The primary endpoint was the change of mean common carotid IMT, as measured by B-mode ultrasound, over 2 years.
Results: Baseline IMT and other characteristics were similar between study groups. Common carotid IMT in the placebo group was 0.780 mm at baseline and 0.774 mm at 2 years (p=0.50), in the statin group it was 0.763 mm at baseline and 0.765 mm at 2 years (p=0.78). There was no significant difference between the change in IMT in the placebo group and the statin group (p=0.48). LDL cholesterol was reduced with 25 % in the statin group and increased with 8% in the placebo group (p<0.001). The results remained unchanged after correction for duration of cerivastatin treatment. Cardiovascular events occurred in 12 patients in the placebo group and in 2 patients in the statin group (p=0.006).
Conclusions: We did not find any effect of 2 years statin therapy on mean common carotid IMT in DM2, in spite of a mean LDL cholesterol reduction of 25 %. The natural history of IMT in our patients was milder than anticipated. In contrast, we observed a significantly lower cardiovascular event rate on statin therapy. We hypothesize that although statins do not influence the irreversibly changed glycosylated extracellular matrix, it may still have a beneficial effect on outcome in DM2 patients by its influence on plaque vulnerability. Other prognostic tools than IMT must be explored in this patient group.

THE EFFECT OF INSULIN SENSITIZERS ON LIPOPROTEIN PARAMETERS IN OBESE PATIENTS WITH TYPE 2 DIABETES MELLITUS

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In relative excess body weight, more than 20% of the patients with type 2 Diabetes mellitus have the risk of atherosclerosis if the patients have misbalance of lipoprotein parameters, as well. The aim of this study is to prove the effect of insulin sensitizers on lipoprotein parameters in patients with type 2 Diabetes mellitus who have not been treated with antilipemics. The study includes 60 patients with body mass index between 30.8 and 37.4 kg/m^2, both sexes, 38 males from 32-65 of age and 22 females from 39-65 of age. The patients have been treated with Metformin 1700mg per day and Rosiglitasone (tiasolidindion) Aventis – 4mg per day during four months. The glucose, glycosides hemoglobin (HbA1c), total cholesterol (TCH), triglycerides (TG), HDL-cholesterol, LDL-cholesterol, apolipoproteins A-I and B and lipoprotein (a) [Lp(a)] have been determined by using standard methods before the therapy and four times during the four months therapy. The statistical processing of the studied parameters has shown the following results: There is no significant statistical difference of the values before and after the four months therapy for TCH, TG, LDL-c and Apo B (p>0.05), namely (p<0.362, p<0.232, p<0.187 and p<0.260) respectively. In 18 out of total 60 patients from both sexes, there have been higher values of 30mg/dl for Lp(a), which remained the same before and during the therapy. A significant statistical difference p<0.05 has been found for serum glucose and HbA1c. In addition, there has been no significant statistical difference in relation to the sex or age of the patients. The four months therapy has proven the expected effect on the level of glucose and HbA1c, while the lipoprotein parameters have remained almost unchanged, which points out to the need of additional therapy for regulation of lipoprotein metabolism.

METFORMIN MIGHT ENHANCE LPL PRODUCTION AND IMPROVE LDL PARTICLE SIZE IN TYPE 2 DIABETES MELLITUS PATIENTS

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Metformin, a member of the biganide class, has been reported to exert its effects primary on the liver, such as inhibiting gluconeogenesis and reducing hepatic glucose out put. Recently, it is reported that metformin has the direct action to adipocytes. Lipoprotein lipase, which is mainly produced from adipocytes and muscle cells, is merely recognized in preheparin serum (prehaparin LPL mass). We reported that prehaparin LPL mass might reflect total LPL production in a whole body[1,2]. In this study, to clarify the effect of metformin on prehaparin LPL mass and lipid metabolism, we administered metformin to 28 type 2 diabetes mellitus patients, who had already received sulfonyl urea agents, and observed the change of preheparin LPL mass, body mass index (BMI), HbA1c, total cholesterol (TC), triglyceride (TG), high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C) and LDL particle size. Preheparin LPL mass was measured by sandwich enzyme-linked immunosorbent assay (ELISA) using specific monoclonal antibody (Daiichi pure chemicals,Japan). LDL particle size was decided by relative mobility value of peak of LDL fraction (LDL-Rm) detected by lipoprotein polyacrylamide gel disc electrophoresis. After 3 months of metformin treatment, preheparin LPL mass significantly increased from 43.0 ng/ml to 51.7 ng/ml ($p<0.01$). HbA1c significantly decreased and no significant change was observed in BMI, TC and LDL-C. TG decreased and HDL-C increased. LDL-Rm significantly decreased from 0.355 to 0.337 ($p<0.05$). No significant correlation was observed between ΔHbA1c and Δpreheparin LPL mass or ΔLDL-Rm. These results suggested that metformin might improve lipid metabolism and enhance LPL production by the direct action to adipocytes or muscle cells.

References: 1) Totsuka et al. Atherosclerosis 2000; 153: 175-179,
2) Miyashita et al. Diabetes Res Clin Pr 2002; 56: 181-7,

EFFECT OF GOINO PROCEDURE ON BLOOD GLUCOSE AND LDL CHOLESTEROL IN PATIENTS WITH NON INSULIN DEPENDENT DIABETES MELLITUS

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Our previous study have shown insulin-like activity of GOINO Procedure (GP), which is combination mixture prescribed by specific ratio of Ganoderma Lucidum (GL), Coriolus Versicolor (CV) and Panax Ginseng (PG), in patients with Non Insulin Dependent Diabetes Mellitus (NIDDM) by its insulin-like activity rather than PG along as single therapy.

AIM: This study was to specify if protein was included in GP as one of compositions, through comparing GL and CV as single therapy with GP, given as extract to NIDDM by randomised, double-bind cross-over study method as the last.

Method: 8 patients were treated with GL alone while 8 patients were treated with CV alone. Then the group initially receiving the GL was changed to CV and vice versa for a final treatment period, followed by wash out. Protein contained in GP was measured by SDS polyacrylamide gel slab electrophoresis.

Result: GP showed lowering blood glucose 10.4%($p<0.02$), C-peptide10%($p<0.011$), LDL18.0%($p<0.002$). GL single treatment showed lowering C-peptide 17%($p<0.001$), Insulin 21%($p<0.014$), and LDL 2.5%, while blood glucose did not change significantly. CV showed lowering C-peptide 2.3%, LDL 4.7% while blood glucose did not change significantly. SDS resulted that one of compositions of GP was found in 14KDa-66KDa migration.

Conclusion: The present data shows that GP treatment reduced blood glucose, LDL and C-peptide with NIDDM, and it is specified that one of its compositions comprised low-molecular-weight protein.

INFLUENCE OF *PINUS PINASTER* EXTRACT (PYCNOGENOL) SUPLEMENTATION TO DIABETES RABBITS FODDER ON GLUKOSE PROFILE.

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The impact of *Pinus pinaster* extract (Pycnogenol) from the Mediterranean pine's bark on glucosic profile during experimental diabetes in rabbits was examined. Pycnogenol was purchased in pharmaceutics manufacture „Unia". Sixteen male rabbits were used in the presented study. The experiment lasted 3 months. Rabbits were divided on 2 group (8 in group): group I - diabetes control: animals on standard diet with experimentally evoked diabetes; group II – diabetes with Pycnogenol: animals on standard diet with experimentally evoked diabetes and with 300 mg of Pycnogenol added to water every day of the experiment. Diabetes was inducted by single, intravenous alloxan injection in dose 150 mg/kg b.m.

In rabbits with alloxan diabetes, both in I and II groups noted down characteristic increase in glucose concentration in plasma in comparison to value observed at the beginning experiment (before induction diabetes). After 3 months of experiment is was observed tendency to glucose decrease in studied group received Pycnogenol. Similar changes were noted down in HbA1c concentration. Summarizing, Pycnogenol characterizes positive influence on carbohydrate economy state in rabbits with experimental diabetes and that this preparation can find the potential use as the supplement in the therapy at men diabetes.

INFLUENCE OF *PINUS PINASTER* EXTRACT (PYCNOGENOL) ADMINISTRATION ON ANTIOXIDANT LEVEL IN BLOOD SERUM RABBITS WITH EXPERIMENTAL DIABETES.

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The aim of this work was study if extract from the Mediterranean pine's bark (*Pinus pinaster*) influence on antioxidant parameters such as: uric acid, levels of iron (Fe), copper (Cu), zinc (Zn); α-tocopherol, paraoxonase (PON with and without NaCl stimulation), conjugated dienes (CD), superoxide dismutase (SOD), glutathione peroxidase (POX), catalase (CAT), glutathione reductase (GR) and malondialdehyde (MDA) during experimental diabetes in rabbits. Pycnogenol - rich of natural, biological active components such as flavonoids - is recommended as a substance of strong antioxidant properties. It was purchased in pharmaceutics manufacture „Unia". Sixteen male rabbits were used in the presented study. The experiment lasted 3 months. Rabbits were divided on 2 group (8 in group): group I - diabetes control: animals on standard diet with experimentally evoked diabetes; group II – diabetes with Pycnogenol: animals on standard diet with experimentally evoked diabetes and with 300 mg of Pycnogenol added to water every day of the experiment. Diabetes was inducted by single, intravenous alloxan injection in dose 150 mg/kg b.m. The analysis of α-tocopherol levels in rabbits plasma was conducted by mean of HPLC. The another antioxidant parameters in rabbit's plasma was carried out by using spectrophotometric methods. Obtained results confirm antioxidant properties of using *Pinus pinaster* extract during experimental diabetes in rabbits.

EFFECT OF *PINUS PINASTER* EXTRACT (PYCNOGENOL) SUPLEMENTATION TO DIABETES RABBITS FODDER ON SERUM LIPIDS PROFILE.

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The impact of *Pinus pinaster* extract (Pycnogenol) from the Mediterranean pine's bark on lipid profile during experimental diabetes in rabbits was examined. Pycnogenol was purchased in pharmaceutics manufacture „Unia". It is recommended as a substance of strong antioxidant properties. Sixteen male rabbits were used in the presented study. The experiment lasted 3 months. Rabbits were divided on 2 group (8 in group): group I - diabetes control: animals on standard diet with experimentally evoked diabetes; group II – diabetes with Pycnogenol: animals on standard diet with experimentally evoked diabetes and with 300 mg of Pycnogenol added to water every day of the experiment. Diabetes was inducted by single, intravenous alloxan injection in dose 150 mg/kg b.m. Obtained results suggest favourable Pycnogenol influence on lipids parameters connected with the risk of arteriosclerosis (CH, HDL, LDL). Different observed, positive effect of Pycnogenol was slowing down lipolyse demonstrated by decreasing FFA level. The next positive effect of Pycnogenol administration, is demonstrated by slowing down the process of enzymatic cholesterol esterolysis in circulation to free cholesterol. Results of our research show protective and anti-atherosclerosis effect of used extract.

STUDIES ON ALLOXAN AND STREPTOZOTOCIN INDUCED DIABETES.

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The induction of experimental diabetes in the rat using chemicals which selectively destroy pancreatic B cells is very convenient and simple to use. The most usual substances to induce diabetes in the rat are alloxan and streptozotocin. The aim of our study was comparison between alloxan and streptozotocine induced diabetes and influence diabetes on glucosic profile, antioxidant enzymes activity and malondialdehyde concentration. Twenty male rats were used in the presented study. The experiment lasted 2 weeks. Rats were divided on 2 group (10 in group): group I – alloxan diabetes; group II – streptozotocin diabetes. Diabetes were inducted by single, intraperitoneal injection of streptozotocine in dose 50 mg/kg b.m. and three times (24 hours intervals) alloxan injection in dose 50 mg/kg b.m. Glucose level was controlled after 3, 6, 24 hours and after 1 and 2 weeks. At the beginning and the end of experiment there were determined antioxidant enzymes activity of superoxide dismutase (SOD), glutathione peroxidase (POX), catalase (CAT) and glutathione reductase (GR), concentration of HbA1c and malondialdehyde (MDA). Obtained results suggest that: 1. Experimentally diabetes disturbance prooxidative/ antioxidative profile; 2. Effect of streptozotocin induced diabetes is more distinct then alloxan.

BENEFITS OF SHORT - TERM MEDICAMENT THERAPY OF OBESITY – ONE YEAR FOLLOW-UP PERIOD

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Beside dietary management and physical activity, effective treatment of obesity also includes drug therapy. Orlistat is a lipase inhibitor which partially inhibits dietary fat absorption and has been shown to promote weight loss and weight maintenance when combined with behavioral treatment. The present study included 12 obese adult diabetics, 4 and 8 (BMI>30 kg/m²) followed during 6 months. They were treated with orlistat 120 mg three times daily with each meal and with a mildly low energy diet (1500 -1800 cal) during first 3 months and only with diet management during the next 3 months. Body weight (TT), body mass index (BMI), and serum levels of glucose, cholesterol and triglycerides at the beginning of the study and after 3 and 6 months follow-up period were determined. Examined diabetics during the period when they were treated with orlistat achieved a weight loss of 8.71±1.84 kg (120.86±15.75 vs 112.14±16.32) and they reduced BMI (37.57±4.79 vs 34.85±4.88 kg/m²). They also showed the changes in serum levels of glucose (9.88 ± 2,62 vs 8,42±2.94 nmol/l), cholesterol (7.83±1.41 vs 6.73±0.87 mmol/l) and triglycerides (4.20± 1.96 vs 3.32±1.53 mmol/l). Medicament therapy with orlistat had effects on the reduction of body weight, BMI, and serum's glucose and lipids levels. In following three months, patients treated with only diet management continued to lose weight for 5.28±2.32 kg (106.86±15.63) and to reduce their BMI (33.21± 4.70 kg/m²), glucose (7,60+2,44nmol/l), cholesterol (6.31±0.80 mmol/l) and triglycerides (2.66±1.10 mmol/l) . Results of our study showed that medicament therapy used for a short period of time had achieved a significant weight loss (7,2% of body weight). It is possible that medicament therapy can have educative effects and influence on behaviour changes which contribute to further weight loss and better metabolic control.

REDUCTION OF VISCERAL ADIPOSITY AFTER OPERATION IN A SUBJECT WITH INSULINOMA

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Generally, the mechanisms of metabolic syndromes are considered that visceral fat brings insulin resistance and hyperinsulinemia. But, whether hyperinsulinemia bring about accumulation of visceral fat is not clear enough. We followed a case with insulinoma, which caused primary hyperinsulinemia, and measured the change of visceral fat and insulin resistance before and after surgical resection of the insulinoma. A 58 year-old woman was admitted to investigate the cause of spontaneous hypoglycemia. Oral glucose tolerance test (OGTT) showed hyperinsulinemia with high basal level and the glucagon infusion test showed abnormally high insulin level. Abdominal computed tomography (CT) scan showed accumulation of visceral fat. A selective celiac angiography showed a pancreatic tumor shadow. Under a diagnosis of insulinoma, the pancreatic body and tail were removed. At 3 months after the operation, visceral fat area decreased from 132.6 to 64.2 cm². The fasting serum total cholesterol and triglyceride were reduced. And, high-density lipoprotein cholesterol and preheparin serum lipoprotein lipase mass increased. The midband on polyacrylamide gel disc electrophoresis of lipoproteins, which appeared before operation, was disappeared completely. OGTT showed non-diabetic pattern after operation. These results suggest that hyperinsulinemia might be one of the factors, which enhanced visceral adiposity and insulin resistance.

LOCUS OF CONTROL IN PATIENTS WITH TYPE 2 DIABETES AFTER LONG-TERM MANAGEMENT BY GROUP CARE

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Background: The locus of control (LoC) describes the relationship between patients' behaviour and daily life situations. We developed a model to manage type 2 diabetes (T2DM) by systemic group education (Group Care = GC) which resulted in sustained body weight decrease, increased HDL-cholesterol and stabilization of HbA1c, along with improved health conducts, knowledge of diabetes and quality of life. **Aims:** To compare the LoC in patients with T2DM followed for 5-7 years by GC and controls followed by traditional one-to-one care. **Methods:** 56 GC patients and 51 controls comparable by age, sex, diabetes duration, glycaemia, insulinaemia, weight and socio-economic variables, were studied. Two questionnaires, Peyrot and Rubin (PR) specific for diabetes, and Wallston and Wallston (WW), generic for chronic diseases, were administered to assess: 1) the internal control (IC) of disease, 2) the role of chance in influencing the disease, and 3) trust in health operators. **Results:** GC patients had lower HbA1c (7.40±1.21) than controls (7.99±1.48), p=0.027. Both questionnaires showed lower scores for Chance in GC patients. The PR showed, in the GC patients, increased IC which, on multivariate analysis, was inversely related to insulin resistance (HOMA index) independently of BMI and HbA1c.

	PR IC	PR Chance	PR Trust	WW IC	WW Chance	WW Trust
GC	31.8 ± 4.1	15.0 ± 5.6	28.1 ± 5.1	29.1 ± 5.0	16.3 ± 5.5	28.9 ± 6.1
Contr	28.8 ± 6.5	28.2 ± 2.0	28.0 ± 4.6	29.2 ± 4.6	27.1 ± 5.8	28.9 ± 5.8
p	<0.001	<0.001	NS	NS	<0.001	NS

Conclusions: Group Care may reduce fatalism and increase Internal Control in patients with T2DM without modifying their trust in health operators. These changes may influence insulin resistance above and beyond their effects on body weight and metabolic control.

EVALUATION OF GROUP EDUCATION PROGRAM ON LIPID METABOLISM, AND OTHER METABOLIC PARAMETERS IN DIABETIC PATIENTS

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Background: To evaluate the clinical effectiveness of the diabetes group education program on lipid metabolism, and other metabolic parameters in patients with diabetes mellitus (DM) on insulin therapy. **Methods:** One hundred and sixty two patients with DM on insulin therapy who completed a structured 5-day group education program were evaluated every six months in the time period of two years when was determined lipid status (LS), HbA_{1c}, as well as Body Mass Index (BMI), Arterial Tension (AT), Insulin Dose (ID). **Results:** In 162 patients (predominating Type 2 diabetes – 73%) we observed significant decrease in HbA_{1c} (from 11,33% to 8,07% $p<0.01$). This improvement in the metabolic control did not positively influenced in the regulation of LS (triglicerides: 2.01 to 2.04mmol/l, total cholesterol: 5.73 to 5.62mmol/l, HDL: 1.1 to 1.1mmol/l and LDL: 1.0 to 0.97mmol/l), BMI (25.87 kg/m^2 to 26.13 kg/m^2), and AT (140 to 141mmHg for systolic tension and 87 to 88mmHg for diastolic tension). Also ID did not show any changes in the researched period (40 to 41.56 IE). **Conclusions:** A structured group education program as used in this study is able to improve metabolic control within period of two years. However this improvement in metabolic control did not have positive influence in the regulation of LS, BMI, AT, and ID, which stayed almost unchanged in this period.

CONJUGATED LINOLEIC ACID SUPPRESSES GENERATION OF PRO-INFLAMMATORY PRODUCTS IN MACROPHAGES

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The present study was designed to evaluate the role of CLA in macro-phages related to (a) ROS generation and (b) thromboxane B_2 (TXB_2) and prostaglandin E_2 (PGE_2) synthesis. Experiments were carried out in PMA -differentiated human THP-1 monocytic leukemia cells or in macropha-ges isolated from peripheral blood. THP-1 were cultured in RPMI 1640 medium supplemented with 10% fatty acid (FA) free (charcoal-stripped) fetal calf serum, penicillin (100 U/ml) and streptomycin (100 mg/mL), at 37 C and 5% CO_2. Mononuclear cells were separated using Gradisol-L density gradient centrifugation. Human macrophages were obtained by culturing monocytes for 3 days in RPMI-1640 with 20% autologous hu-man heat-inactivated serum at 37°C and humidified 5% CO_2. The cell po-pulation consisted of >90% monocytes as determined by surface expre-ssion of CD14. COX-2 expression in adherent monocytes and THP1 be-came evident upon cell exposure (for 48 h) to 40 nM phorbol myristate acetate (PMA). FAs were added as 4 mM stock solution dissolved in 1 mM FA free bovine serum. LA and two CLA isomers (c9 t11 and c10 t12) were used at concentrations of 30 μM. Cells were cultured with the FA for 48 h. For intracellular ROS detection, cells were pre-loaded (30 min. at 37°C) with 5 μM 2',7' dichlorofluorescein diacetate. TXB_2 and PGE_2 were measured in the supernatant by enzyme immunoassay. Macrophages were converted to foam cells by treatment with oxidized low-density lipoproteins (oxLDL). Native LDL were isolated from plas-ma by sequential ultracentrifugation and oxLDL were prepared by incu-bating LDL with endothelial cells. CLA inhibited the synthesis of eicosa-noids in macrophages. t10 c12 CLA suppressed intracellular ROS gene-ration by 20% and reduced the amount of PGE_2 by 12% in macrophages from THP1, by 47% in blood macrophages, and by 17% in foam cells. The respective reductions for c9 t11 CLA were 19%, 45%, and 17%. Both CLA isomers seem to reduce levels of pro-inflammatory products in human macrophages by acting on COX (chiefly COX-2) and thus modu-lating ROS generation.

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EFFECT OF GARLIC AND ONION EXTRACTS ON SUSCEPTIBILITY OF LDL TO OXIDATION

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Background: Onion and garlic have been used in cooking and for their medicinal properties for thousands of years. Studies relating to the onion and its antioxidant effects make this a particularly interesting vegetable. The objective of this study was to investigate the antioxidant effect of aqueous extracts of onion and garlic on the LDL oxidation.
Methods: Aqueous onion and garlic's extract were prepared from fresh onion and garlic. LDL (150μg protein/ml) in Phosphate Buffer Saline (PBS) was incubated at 37° C in the presence of Cuso4 (final concentration 5μM) and 0.2 ml of each extract separately. The peroxidation process was continuously monitored by the change in absorbance at 234 nm. The electrophoretic mobility of intact and oxidized LDL in the presence and absence of each extract also was performed separately.
Result: The results showed that onion and garlic's extract at the applied concentration completely inhibit LDL oxidation at the time of incubation. Also showed that electrophoretic mobility of oxidized LDL is faster than native LDL and LDL which incubated with aqueous extracts of onion and garlic.
Conclusion: This study showed that onion and garlic have high inhibitory effects on LDL oxidation and because of suggested that oxidation of LDL may represent an important event in atherogenesis, consumption of onion and garlic regularly could make a significant contribution to antioxidant defenses in the blood and help in protect against several different kinds disease like atherosclerosis.

PHENOLIC EXTRACT FORM ARGAN OIL, INHIBIT HUMAN LOW DENSITY LIPOPROTEINS OXIDATION AND ENHANCE CHOLESTEROL EFFLUX FROM HUMAN THP-1 MACROPHAGE.

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Argan oil obtained from *Argania spinosa* seeds is eaten raw in Southwest Morocco and its rich composition in mono- and -polyunsaturated fatty acids, tocopherol and phenolic compounds make interesting a further study of its action on cardiovascular diseases. Furthermore, in comparison to olive oil, no study had been conducted so far exploring the antioxidant activity of Argan oil towards lipid. Thus this study was conducted to evaluate the properties of Virgin Argan Oil Phenolic Extracts (VAO-PE): **A-** To protect human Low Density Lipoproteins (LDL) against lipid peroxidation and **B-** To promote HDL-mediated cholesterol efflux. LDL and HDL (high density lipoprotein) were isolated from blood plasma of healthy volunteers and LDL oxidation was induced by incubation with Cu^{2+} (10µM) in the presence of different concentrations of VAO-PE (20-320µg/ml). LDL oxidation was followed by the formation of conjugated dienes (CD), MDA formation and vitamin E disappearance. Production of CD was determined by the continuous monitoring of increased absorbency at (234 nm) while MDA formation and vitamin E disappearance was determined by HPLC. Incubation of LDL with VAO-PE prolonged the lag-phase and significantly lowered the progression rate ($P<0.01$) of CD and MDA formation and reduce the disappearance of vitamin E in a concentration-dependent manner. CD and MDA formation was reduced respectively by 93.66% and 98.70% after 4 h incubation at 320µg/ml of the VAO-PE. In an other hand, incubation of HDL with VA-PE, increase significantly their membrane fluidity ($P=0.0004$) and HDL-mediated cholesterol efflux from THP-1 macrophage. Theses results suggest that Virgin argan oil provides a source of dietary phenolic antioxidants that may have potential effects in the prevention of cardiovascular diseases.

THE EXPRESSION OF PRO-ATHEROGENIC MOLECULES AND cGMP-SPECIFIC PDE5 ACTIVITY ARE AFFECTED BY OLIVE OIL ANTIOXIDANTS

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Epidemiological studies demonstrated a correlation between nutrition and atherosclerosis; in particular, the "Mediterranean-style" diet, where olive oil is the main source of lipids, is associated with a low incidence of cardiovascular risk. Minor polar compounds of olive oil such as hydroxytyrosol (HT) and oleuropein aglycon (OleA) could be protective against atherosclerosis while the mechanism(s) are not completely established. In addition, adhesion molecules as well as cGMP play a pivotal role in modulating cell adhesion and smooth muscle cells proliferation and migration; the increase of cGMP levels is sustained by inhibition of phosphodiesterase-5 (PDE5). To explore the antiatherogenic mechanisms, the effect of OleA, HT and its metabolite homovanillyl alcohol (HVAlc) on the expression of molecules involved in inflammation was investigated. In HUVEC, HT significantly downregulated the gene expression of VCAM-1 and ICAM-1, also in the presence of TNFα. The *in vitro* human recombinant PDE5A1 activity was also evaluated. HT and phenolic extract from olive oil exerted a concentration-dependent inhibition on PDE5A1 activity, already significant at low concentrations, while the contribution of OleA and HVAlc was negligible. These data demonstrated that minor components of Mediterranean diet could contribute to protect against cardiovascular risk not only by its scavenging activity but also to their capacity to regulate the expression of proatherogenic/proinflammatory genes.

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SILYMARIN INHIBITS ABSORPTION OF CHOLESTEROL IN RAT

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The influence of silymarin, the standardised extract from the medicinal plant *Silybum marianum* L. containing flavonolignans and fraction of polymerised polyphenolics, on cholesterol absorption in rats was studied using the dual-isotope plasma ratio method (1). This method is based on determination of the ratio of $^{14}C/^{3}H$ in plasma after i.v. application of ^{3}H-cholesterol and after intragastrical application of ^{14}C-cholesterol. Animals were fed for 3 weeks on high-cholesterol (1%) high-fat (10% of saturated lard fat) diet, supplemented with 1% of silymarin or 0.003% of ezetimibe, a specific inhibitor of cholesterol absorption, as a positive control drug. Feeding of rats on 1% of silymarin caused: 1) a decrease ($p<0.05$) in absorption of ^{14}C-cholesterol from intestine, 2) an increase ($p<0.001$) in content of ^{14}C-cholesterol in HDL and 3) a decrease ($p<0.001$) in content of ^{14}C-cholesterol in the liver. Findings of a decrease in level of unlabeled cholesterol in liver ($p<0.001$), plasma ($p<0.01$), and VLDL ($p<0.001$), and an increase in HDL ($p<0.01$) support the suggested inhibitory effect of silymarin on cholesterol absorption.

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EFFECTS OF UNSAPONIFIABLE MATERIALS IN RICE BRAN OIL ON SERUM CHOLESTEROL LEVELS

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Rice bran oil lower plasma cholesterol levels more effectively than other commonly usual vegetable oils rich in linoleic acid. This effect can not be simply explained by fatty acid composition. Rice bran oil is characterized by a relatively high content of unsaponifiable materials such as sterols, γ-oryzanol and tocotrienols. These components have the hypocholesterolemic function such as several drugs, a part of the cholesterol-lowering ability of rice bran oil is seems to be due to the contents of unsaponifiable materials. In this study the effects of unsaponifiable materials in rice bran oil on serum cholesterol levels were studied.

Male Sprague-Dawley rats (Exp.1) and male Golden Syrian hamstars (Exp.2) were fed a hypercholesterolemic diet containing cholesterol and sodium cholate and were put down after 3 weeks. Dietary lipid sources were lard in the control group and normal rice bran oil (R) and unsaponifiable materials free rice bran oil (FR) in the experimental groups. Serum total cholesterol (TC) levels in the control group were highest in 2 experiments, respectively. In the Exp.1, serum TC in the R group clearly decreased compared to that of the control group. FR group also showed significant decrease in serum TC. Serum HDL cholesterol (HDL-C) levels was lowest in the control group. However HDL-C level in the R group was clearly higher than that of the control group. HDL-C in the FR group tended to elevate compared to the level of the control group. Serum TC in the R and FR groups significantly decreased compared to that of the control group in the Exp.2.

From these results it was suggested that the cholesterol lowering effect was partly attributed to the unsaponifiable materials in rice bran oil.

CARNITINE DEFICIENCY IN FETAL-NEONATAL LIFE: LONG-TERM CONSEQUENCES ON LIPID METABOLISM AND INSULIN-SENSITIVITY IN RAT

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In order to test the hypothesis that carnitine deficiency in fetal-neonatal life could represent a risk factor for the development of insulin- resistance status in adult life [1] we administered sodium pivalate (20 mM in drinking water) to female rats during pregnancy and lactation, and monitored their newborns till 6 months of age. Pivalate treatment is known to cause rapid serum and tissue carnitine depletion in females, and to lower carnitine levels in their milk [2]. A second group of pivalate-treated mothers was supplemented with carnitine (40 mM in drinking water), for the purpose of restoring carnitine levels in their young. Neonates from pivalate-treated mothers showed a significant reduction (60-70%) in serum, liver, muscle, heart and brain carnitine levels. Carnitine deficiency induced in early neonatal life was characterized by defects in lipid metabolism and reduction in liver palmitate β-oxidation. At 4 and 12 days of age an increase in triglyceride content in various tissues and elevated serum ketone body levels were also observed in pivalate-treated rats compared to controls. Carnitine supplementation in pivalate-treated females restored carnitine levels and counteracted triglyceride accumulation in various tissues, and restored palmitate β-oxidation in the livers of their offspring. After weaning, all rats were monitored for serum glucose, insulin and triglyceride levels. The expression and the activity of carnitine palmitoyltransferase (CPT1 L-M) in liver and heart were also measured. Progressive hyperinsulinemia and hypertriglyceridemia were observed in rats from pivalate group with respect to controls, and carnitine supplementation counteracted these effects. At 6 months of age, all groups were subjected to an oral glucose tolerance test. Rats from pivalate-treated mothers showed an insulin-resistance state whose development appeared to be partially prevented in the group from carnitine-supplemented mothers. Our data suggest that moderate carnitine deficiency in fetal and neonatal life may lead to profound alteration of glucose and lipid homeostasis in adult life and that these conditions may be successfully counteracted by carnitine treatment.

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SOY PROTEIN- AND FIBER-CONTAINING DIETS REDUCE INTESTINAL UPTAKE OF STEROLS AND VLDL-CHOLESTEROL SECRETION IN APOE*3-LEIDEN TRANSGENIC MICE

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Dietary consumption of soy protein reduces hyperlipidemia and atherosclerosis in rodents. In this study, we investigated the effect of a soy diet versus a soy-fiber containing diet on 1) plasma lipid levels 2) mechanism of action in female ApoE*3-Leiden transgenic mice, a well-established model for hyperlipidemia and atherosclerosis. Five groups of 8 mice received for 4 weeks a diet containing 0.25% cholesterol and 20% or 40 % soy protein extract (Supro Soy (SS) 20/40) and 20% or 40% soy protein-soy fiber extract (Abacor New Formula (ANF) 20/40). ANF and SS differ by the type of fiber (cotyledon fiber and cellulose, respectively) and the amount of isoflavones (1.95 and 3.40 mg/g protein, respectively). Compared with a control 20% casein-containing diet, ANF40 and SS40 dose-dependently decreased plasma cholesterol (72% and 51%, resp. $p<0.001$) and triglycerides (52% and 42%, resp. $p<0.001$), mainly in VLDL-particles. Furthermore, fecal neutral sterols were increased in the ANF40 (97%; $p=0.03$) and SS40 (49%; $p<0.001$) groups compared to the control group, whereas there was no effect on fecal bile acid output. Hepatic free cholesterol, cholesteryl ester and triglyceride content were decreased in both groups, with the highest reduction in the ANF40 group (49%, 85%, 72%, resp. $p<0.05$). No differences were found in VLDL-triglyderide and VLDL-apoB production rates after soy consumption. However, soy feeding significantly decreased the amount of free cholesterol and cholesteryl esters incorporated in nascent VLDLs, whereas triglyceride content significantly increased. Again, the changes were most pronounced in the ANF40 group (-24%, -72% and +62%, resp. $p<0.05$).
In conclusion, in ApoE*3-Leiden transgenic mice, soy protein and soy protein-soy fiber feeding decreased plasma cholesterol levels as a result of a decreased intestinal uptake of neutral sterols, resulting in a reduced VLDL-cholesterol secretion. The effects were most pronounced in the soy protein-soy fiber group, indicating that cotyledon fiber has an additional cholesterol-lowering effect.

ABSENCE OF AN ATHEROPROTECTIVE EFFECT OF GARLIC POWDER PRINTANOR IN APOE*3-LEIDEN TRANSGENIC MICE

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Animal studies reported that garlic can protect against atherosclerosis. However, a comparable number of studies do not support this observation. This contradiction may result from differences in study design, use of different animal models, and use of different garlic formulations and preparations. Here, we investigated the effect of the chemically well-characterized and production-controlled garlic powder printanor on atherosclerosis in the APOE*3-Leiden transgenic mouse, a mouse model well-suited for evaluating anti-atherosclerotic properties of drugs and food components under human-like conditions. APOE*3-Leiden mice were fed a Western diet supplemented with 5 or 50 $g.kg^{-1}$ printanor. As a reference, the commercially available fermented garlic kyolic® was included (1.6 $g.kg^{-1}$ diet). Treatment with printanor (both doses) demonstrated reduced body weight, coinciding with increased feces production and fecal fatty acids excretion. Printanor (both doses) and kyolic® treatment did not affect plasma lipids, serum amyloid A, serum soluble intercellular adhesion molecule-1, plasma von Willebrand factor, and blood-leukocytes Tumor Necrosis Factor-α production. As analyzed after 28 weeks of treatment, printanor (both doses) and kyolic did not affect atherosclerotic lesion type, area or composition. Under conditions relevant to the human situation, the well-characterized and production-controlled garlic powder printanor does not display hypolipidemic, anti-inflammatory or anti-atherosclerotic properties.

DIETARY DIACYLGLYCEROL SUPPRESSES FATTY LIVER FORMATION ACCOMPANYING UP-REGULATION OF β-OXIDATION ENZYMES IN ZUCKER FATTY RATS

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Background & Aims*:** Obesity and diabetes mellitus associated with insulin resistance are the predominant cause of hepatic fat accumulation. We examined the effect of dietary diacylglycerol (DG), which is a minor component of edible oils, on lipid accumulation in the liver. ***Methods*:** Hepatic lipid content, β-oxidation activity, serum lipid levels were measured in Zucker fatty rats fed diets containing either 10% triacylglycerol (TG), 10% TG +4% α-linolenic acid-rich TG (ALATG), or 10% TG +4% ALA-rich diacylglycerol (ALADG) for 1 month. ***Results: Supplementation with ALADG resulted in significant reduction of triglyceride accumulation accompanied by up-regulation of β-oxidation activity and acyl-CoA oxidase, medium-chain acyl-CoA dehydrogenase, and L-fatty acid binding protein mRNA expression in the liver. However, there were no significant changes observed in PPARα and SREBP mRNA levels. ***Conclusion:*** ALADG feeding might be useful for prevention of fatty liver formation. Stimulation of lipid catabolism in the liver may be closely related to the beneficial effects of dietary DG. In addition, not only differences in the composition of fatty acids but also structural differences between DG and TG markedly affected the nutritional behavior of lipids.

REGRESSION STUDY OF *GRIFOLA FRONDOSA* AGAINST CHOLESTEROL-FED ATHEROSCLEROSIS IN RABBIT AORTA

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Purpose: Mushroom Maitake (*Grifola Frondosa)* which is beneficial foods and known as nutritional supplement, has several functions as reducing blood pressure in hypertensive rats. Powdered Maitake (SF-73, Santomi Sangyo Co., Ltd., Tokyo, Japan) was studied whether SF-73 has atherogenesis or not, utilizing cholesterol-fed rabbit. Methods: Thirty-two, male, Japanese white rabbit were fed with a 1.0% cholesterol and basal ration (100g/day) for 8 weeks, and then they were divided into 4 groups, 1) control was fed with 1.0% cholesterol ration, 2) administered with 1.5% SF-73 and 1.0% cholesterol ration, 3) administered with 3% SF-73 and 1.0% cholesterol ration, and 4) administered with 6% SF-73 and 1.0% cholesterol ration for more 8 weeks, n=8, in each group. Serum total cholesterol (TC) was measured through the experiment. At the end of experiment (total 16 weeks), thoracic aortas were removed and fixed with formalin. Surface involvement (SI) and Atherosclerotic index (AI) were determined for morphometrical evaluation of atherosclerosis. Components of the lesions were evaluated with scores as intimal thickness and foam cells. Results: There were no significant differences in body weight among groups. TC levels in the experimental groups (1600 mg/dl) tended to be lowered than in the control (2300 mg/dl) at 16 weeks, without significant difference. There were significant decreases in SI and AI of the 1.5% SF-73 group (SI:30.4±9.2%, AI:21.5±7.3) and 3% SF-73 group (SI:37.2±8.4%, AI:29.3±7.2) than those of the control (SI:59.9±3.8%, AI:51.6±3.1). Scores for intimal thickness and foam cell in the experimental groups (3.2, 2.8) were lowered than in the control (4.9, 4.7). Dietary-induced atherosclerotic lesions showed foam cell aggregation in thickened intima. SF-73 might decrease the numbers of foam cells in the intima and SF-73 may have anti-atherogenesis in rabbit aorta.

THE EFFECT OF LONG-TERM MODERATE ALCOHOL CONSUMPTION ON TISSUE-SPECIFIC LIPOPROTEIN LIPASE ACTIVITY AND EXPRESSION

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One of the positive effects of long-term moderate alcohol consumption is the increase in lipoprotein lipase (LPL) activity. The enzyme activity is known to be regulated in tissue-specific manner; however, it is not clear yet whether the effects of alcohol on LPL activity are also tissue-specific. To address such a question, we examined the influence of alcohol consumption on LPL activity and gene expression in different tissues of mice. Thirty C57Bl/6 mice (females) were fed a chow diet and given 5% ethanol (experimental group, n=15) or water (control group, n=15) for four weeks. Afterwards, the activity and expression of LPL were determined in heart and dorsolumbal and epididymal adipose tissue in ten animals per each group. The activity of LPL in postheparin plasma was measured in the remaining five mice. The alcohol consumption had no effect on lipid and lipoprotein levels (as determined by ultracentrifugation) and did not affect either postheparin LPL or hepatic triglyceride lipase activities. Moreover, it did not have any effect on LPL activity and expression in heart and both dorsolumbal and epididymal adipose tissues. Therefore, the alcohol consumption does not seem to influence lipoprotein metabolism and lipoprotein lipase activity in all the tissues under the study in mice fed a chow diet. However, it cannot be excluded that positive impact of alcohol on LPL activity could be observed on a high-fat diet that is commonly used in similar studies.
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EFFECT OF CLA RICH BUTTER, REGULAR BUTTER AND PALM OIL ON LDL PARTICLE SIZE AND PLASMA LIPIDS IN GROWING PIGS.

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Conjugated linoleic acid (CLA) is a group of polyunsaturated fatty acids found in beef, lamb and dairy products. Numerous health benefits have been attributed to CLA in experimental animal models. Since milk fat are related to a more favourable LDL particle size profile in humans, we decided to investigate LDL particles and plasma lipids in pigs given supplements of palm oil, regular butter or CLA rich butter (containing 0 g, 0.6 g and 2.1g c9,t11CLA/100 g fat, respectively). Thirty-eight weanling pigs from a commercial Norwegian crossbreed were randomly allotted to three groups that were matched according to weight, sex and litter. Diets were fed for 21 days, and the pigs were fed individually. Blood samples were taken at day 0 and at the end of the period (day 21). Fat represented 44 % of total energy intake in each diet, with 90% of the fat from the supplements. LDL particle size and distribution were characterized by nondenaturing 3-7,5 % polyacryl-amide gradient gel electrophoresis. Plasma lipids and serum fatty acid concentrations were determined.
Feeding three groups of pigs high fat diets containing palm oil or regular butter or CLA rich butter gave similar effect on LDL particle size and distribution of LDL particles. Also, no differences in plasma concentrations of unesterified fatty acids, triacylglycerol, cholesterol, HDL cholesterol, total antioxidant status, GPX and glucose were observed. The serum concentration of palmitic acid was significantly reduced and CLA, vaccenic acid and α-linolenic acid were increased in the pigs fed the CLA rich butter compared to regular butter or palm oil. These results demonstrate that CLA rich butter compared to regular butter or palm oil gave some improvements in serum fatty acids, but had no effect on LDL particle size and plasma lipids in pigs.

MONOUNSATURATED FAT DECREASE HEPATIC LIPID CONTENT IN NONALCOHOLIC FATTY LIVER DISEASE IN RATS

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Aim: To evaluate the effect of different types of dietary fats on the hepatic lipid content and oxidative stress in rat liver with experimental non-alcoholic fatty liver disease (NAFLD).
Method: Thirty five Sprague-Dawley rats were randomly divided into 5 groups. The animals in the control group (n=8) were on chow diet (Group 1), rats (n=6) on methionine-choline deficient diet (MCDD) (Group 2), rats (n=7) on MCDD enriched with olive oil (Group 3), rats (n=7) on MCDD with butter (Group 4) and rats (n=7) on MCDD with fish oil (Group 5). After 2 months, the rats were generally anesthesized and blood was sampled by heart puncture. Rats were sacrified and liver sections were examined.
Results: Rats and livers weights increased in Group 1 and decreased in groups 2-5. The fat content was normal in Group 1. There was a significant increase in lipid content in the liver of groups 2-5. Plasma lipid content did not changed in all groups. Severe fatty liver was seen in groups 4 and 5, but not in groups 2 and 3. Serum and hepatic triglycerides levels increased significantly in groups 2-5, but this increment was significantly less in Group 3. In group 3, there was 35% less triglyceride accumulation in the liver compared with group 2. Phospholipid levels in serum and content in liver decreased in Groups 2-5. Malonyl dialdehyde (MDA) hepatic levels increased in Groups 2-5 (p=0.003) in comparison to Group 1, and in Groups 3-5 in comparison to Group 2 (p=0.004). There was no difference between the groups in plasma concentrations of MDA. Paraoxonase activity decreased in the livers of Groups 2-5. α-tocopherol content in the liver decreased (p=0.01) in Group 3.
Conclusion:
Olive oil decreased the accumulation of triglycerides in the liver of rats with NASH.
Antioxidative liver enzymes failed to counteract the increased oxidative stress in livers of rats with NAFLD.

LIPIDS FROM NATURAL GAS-UTILIZING BACTERIA RICH IN PHOSPHATIDYLETHANOLAMINE REDUCE PLASMA CHOLESTEROL AND CLASSES OF PHOSPHOLIPIDS IN MINK COMPARED WITH SOYBEAN OIL

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We have compared the effects of three different high lipid diets, with 0%, 17% and 67% extracted lipids from natural gas-utilizing bacteria (LNGB), replacing soybean oil, on plasma lipoproteins and phospholipids in mink. Phospholipids are the main components in lipids from LNGB, consisting mainly of phosphatidyletanolamine (PE) with predominantly 16:0 and 16:1 fatty acids. The 18 mink studied were fed one of the three diets during a 25-day period in a parallel group design. Total cholesterol, LDL cholesterol, HDL cholesterol and unesterified cholesterol were significantly reduced when the animals consumed a diet with 67% LNGB compared with 100% from soybean oil (SB-diet). Plasma phosphatidylcholine (PC), total phospholipids, lysophosphatidylcholine and phosphatidylinositol were significantly lowered when the mink were fed 67%LNGB-diet compared with solely soybean oil. Plasma total cholesterol was correlated with total phosholipids as well as with PC ($R = 0.8$, $P < 0.001$). We conclude that phospholipids from LNGB decrease plasma lipoprotein and phospholipid level. Our findings indicate that the lowering effects on plasma cholesterol are mainly caused by a specific mixture of phosholipids, containing a high level of PE, and not the fatty acid composition.Thus plasma cholesterol is at least partly regulated by phospholipid methylation from PE to PC in the liver.

EFFECT OF CHOKEBERRY (ARONIA MELANOCARPA) ANTHOCYANINS ON THE ACTIVITY OF THE ANGIOTENSIN CONVERTING ENZYME (ACE) AND ON THE ADIPONECTIN LEVEL IN PATIENTS WITH CORONARY HEART DISEASE (CHD)

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As shown by in vitro experimental data, anthocyanins can regulate many cellular mechanisms *inter alia* through their effect on the activity of cyclooxygenase I and II and through increasing the expression of nitric oxide synthase. In fact, as we have found in our previous studies, supplementation of chokeberry anthocyanins in CHD patients leads to a significant reduction of systolic and diastolic blood pressure and of the CRP level.

The objective of the present studies was to evaluate the potential effect of chokeberry anthocyanins on ACE activity and the adiponectin level in patients with CHD.

The study was conducted as a double-blind placebo controlled parallel-group trial in 52 persons (39 men and 13 women) who underwent myocardial infarction at least 6 months before the study inclusion.

Patients were randomised into two groups: one receiving 3 x 85 mg/d of chokeberry anthocyanins, and the second receiving placebo for 6 weeks. The remaining treatment was left unchanged.

Under the effect of anthocyanins, a significant ($p<0.005$) drop of plasma ACE activity was noted, by 35% on average, which was parallel to the mean lowering of systolic blood pressure by 14 mmHg. At the same time, the adiponectin level increased significantly by 23% ($p<0.005$) and CRP was reduced by 17% ($p<0.001$). Such effects were not observed in the placebo group.

Our studies show that chokeberry anthocyanins can play a significant role in lowering the cardiovascular risk, particularly owing to their effect on blood pressure and limitation of the inflammation process.

PLASMA N-3 AND N-6 POLYUNSATURATED FATTY ACIDS ASSOCIATION WITH BLOOD PRESSURE LEVEL IN HEALTHY MALES.

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Background. Regular consumption of dietary fish and n-3 polyunsaturated fatty acids (PUFA) of marine origin can lower blood pressure levels and reduce cardiovascular risk. However the contribution of each n-3 and n-6 PUFA to blood pressure regulation is still controversial.

The aim of our study was to estimate the relationship between blood pressure level and proportions of n-3 and n-6 PUFA in plasma and erythrocytes in healthy males.

Material and method. We studied 31 male subjects (mean age 58.5±9.0 years) during their routine medical checkups. Systolic (SBP 137,2±24 mmHg), diastolic (DBP 84,3±9 mmHg) and mean (MBP 103,6±13 mmHg) blood pressure was calculated as the mean from two measurements performed six years apart. N-3 and n-6 PUFA in plasma and erythrocytes were determined by gas-liquid chromatography .

Results. Among whole erythrocyte fatty acids only C 20-5 n-3 PUFA was significantly inversely associated with mean DBP ($r = -0,44$; $p< 0.05$). Among plasma fatty acids we found an inverse association between SBP, DBP and MBP with the sum of n-3 PUFA; C18:4; C20:4; C20:5; C22:5; C22:6 ($r=-0,57$; $p<0,02$; $r=-0,65$ $p<0,01$ and $r=-0,62$, $p<0,01$ for SBP, DBP and MBP respectively) and the sum of n-6 PUFA ; C18:3; C20-3; C20:4; C22:4; C22:5 ($r=-0,63$ $p<0,01$; $r=-0,67$; $p<0,01$; $r=0,66$; $p<0,005$ for SBP, DBP and MBP respectively). The highest correlation between the individual PUFA concentration and blood pressure parameters was found between DBP and EPA C20-5 in plasma ($r=-0.51$, $p<0.05$) and erythrocytes ($r=-0.51$, $p<0.05$).

Conclusions. Our findings are consistent with the hypothesis that differences in plasma and erythrocyte n-3 and n-6 polyunsaturated fatty acids composition affect blood pressure level in healthy males.

OMEGA-3 FATTY ACIDS IN CARDIAC BIOPSIES FROM HEART TRANSPLANT PATIENTS: CORRELATION WITH ERYTHROCYTES AND RESPONSE TO SUPPLEMENTATION

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Background: Omega-3 fatty acids (FA) appear to reduce the risk for sudden death from myocardial infarction. This is believed to occur via the incorporation of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) into the myocardium itself, altering the dynamics of sodium and calcium channel function. The extent of incorporation has not been determined in humans, nor has a readily-available biomarker for cardiac omega-3 FA composition been identified.

Methods and Results: We first determined the correlation between red blood cell (RBC) EPA+DHA content (hereafter, the Omega-3 Index) and cardiac omega-3 FA levels in 20, medically-stable heart transplant recipients. We then examined the effects of 6 months of omega-3 FA supplementation (1 g EPA+DHA/d; Ocean Nutrition Canada) on the FA composition of human cardiac tissue and RBCs in 25 other transplant patients.

Results: Cardiac EPA+DHA levels were highly correlated with the Omega-3 Index ($r=0.82$, $p<0.001$). Supplementation increased EPA+DHA levels in cardiac tissue by 110% and in RBC by 101% ($p<0.005$ vs. baseline for both; responses between tissues were not significantly different).

Conclusions: The Omega-3 Index is highly correlated with cardiac EPA+DHA, and the FA response to supplementation is similar for the Omega-3 Index and cardiac muscle. Thus, the Omega-3 Index may serve as a validated surrogate for cardiac omega-3 FA status.