of diabetes: 10.03±4.91 years and duration of hypertension: 11.32±5.38 years). Patients were assigned to antidiabetic and antihypertensive treatment aiming to attain pre-defined level of glycemic (HbA1c < 7%) and blood pressure control (<130/85 mmHg). Nephropathy progression parameters, average annual increment of microalbuminuria (Δ UAE) and deterioration of glomerular filtration rate (Δ GFR), were prospectively followed-up for a mean period of 6.28 (0.88) years. Statistical analysis was performed using the STATISTICA 4.5 program (StatSoft, Tulsa, OK, USA). The significance of differences was evaluated using the Kruskal-Wallis test. A value of p<0.05 was accepted as statistically significant.

**Results:** A total of 62 (49.2%) patients achieved good glycaemic control while tight blood pressure control was achieved and maintained in 76 (60.3%) patients. Choice of antidiabetic treatment did not significantly influence the quality of glycaemic control (P=0.4233), while choice of antihypertensive drugs significantly influenced the quality of achieved blood pressure control (P=0.0357), speaking in favor of ACEI.

Choice of antidiabetic treatment did not significantly influence the rate of microalbuminuria progression as well as the rate of deterioration of glomerular filtration both in patients with good (P=0.641 and P=0.837, respectively) and those with poor glycaemic control (P=0.305 and P=0.256, respectively). However, choice of antihypertensive treatment significantly influenced the rate of microalbuminuria progression (P=0.011) as well as the rate of deterioration of glomerular filtration (P=0.0013), but only in those with tight blood pressure control.

**Conclusion:** Independently from the quality of achieved glycaemic control, choice of the antidiabetic treatment did not significantly influence the rate of nephropathy progression in hypertensive type 2 diabetic patients. However, choice of antihypertensive treatment significantly influenced the rate of progression of microalbuminuria and the rate of deterioration of glomerular filtration but only in those patients in whom good control of blood pressure was achieved.

**P 8: Peripheral insulin resistance influence the presence of hypertension in overweight type 2 diabetic patients**


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**Backgrounds and Aims:** Increased obesity is closely related to insulin resistance (IR) promoting the development of type 2 diabetes (T2D) and cardiovascular disease. IR represents a well established risk factor for development of hypertension, but the significance of the associations between IR and obesity in the development of hypertension in overweight patients with type 2 diabetes (T2D) still remains unclarified. We analyzed the levels of insulin resistance in: overweight T2D patients with hypertension (group A, n=48, 30±6 BMI ≥25kg/m²), overweight T2D patients without hypertension (group B, n=43, 30±6 BMI ≥25kg/m²) and overweight nondiabetics without hypertension (group C, n=40, 30±6 BMI ≥25kg/m²).

**Materials and Methods:** Patients were aged 40-70 years, matched by gender, duration of diabetes, with optimal metabolic control (HbA1c=6.5±0.6%). Hypertension was defined as systolic BP ≥140 and diastolic BP ≥90mmHg measured by sphygmomanometer, or by established use of antihypertensive drugs. Assessment of IR was done with two complementary indexes of insulin sensitivity: (1) oral glucose insulin sensitivity (OGIS) index derived from 75 g oral glucose tolerance test and (2) homeostasis model assessment of insulin resistance (HOMA-IR) determined from fasting glucose and plasma insulin (PI) levels. PI and levels were measured by RIA method.

**Results:** We have found significantly higher PI levels in group A (A: 38.05+/−4.24; B: 29.33+/−3.13; C: 21.34+/−2.23mlU/ml, A vs B: A vs C and B vs C p<0.05). Simultaneously, there was no significant difference in the level of HOMA-IR among diabetics (A: 9.43+/−2.69; B: 7.52+/−2.08; C: 5.73+/−1.83; A vs B p=NS; A vs C and B vs C p<0.05), reflecting hepatic insulin sensitivity. However, lowest OGIS were found in group A, being significantly lower in group A in comparison to group B (A: 287.67±35.42; B: 359.22±52.0; C: 494.29±23.81; A vs B p<0.05; A vs C p<0.01 and B vs C p<0.05) as measurement of peripheral IR. We have not found significant correlation between PI and presence of hypertension (r=0.177, p=NS). Also, there was no significant correlation between HOMA-IR and presence of hypertension among diabetics (r=0.218, p=NS). However, presence of hypertension among diabetics negatively correlate with OGIS index (r=−0.384, p<0.01), and in the model of linear regression analysis level of OGIS predicting the presence of hypertension irrespectively of the obesity (β= −0.334, p=0.07).

**Conclusion:** Our results imply that increased peripheral insulin resistance compared to hepatic insulin resistance stronger influence the presence of hypertension in overweight type 2 diabetic patients. Moreover, this association was not depending of obesity.

**P 9: Taurine protects against impairment of mitochondrial function and oxidative stress in the heart of rats with fructose-induced insulin resistance**

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**Background and aims:** Taurine (2-aminoethanesulfonic acid) is a free amino acid found in substantial amounts in all mammalian tissues. Recently, data is accumulating that show the effectiveness of T against diabetes mellitus, insulin resistance and its complications, including retinopathy, nephropathy, neuropathy, atherosclerosis and cardiomyopathy, independent of hypoglycemic effect. Numerous experimental and several clinical studies demonstrated that T helps the cardiovascular system through a variety of mechanisms including modulation of intracellular calcium concentration, antagonism of angiotensin II action, membrane-stabilizing, antioxidant, and lipid-lowering effects. The aim of the study was to assess the effects of T on mitochondrial respiratory chain activity and lipid peroxidation in the heart of prediabetic rats fed a high-fructose diet.

**Materials and Methods:** Male Wistar rats were divided into three groups: the control group fed on a regular diet (C, n=8), the high fructose-fed group (F, n=8), which had free access to 250 g/L solutions of fructose for 8 weeks and the fructose-fed group treated with taurine (F+T, n=8) for 8 weeks (100 mg/kg/day per os). Mitochondria were isolated by differential centrifugation from the hearts of fed rats. Oxygen consumption rate was measured polarographically at 37°C using a Clark-type oxygen electrode with either glutamate/malate or succinate as energy substrates of complex I or II, respectively. Levels of lipid hydroperoxides, reduced glutathione (GSH) and glutathione reductase (GR) activity were determined in mitochondrial preparations. Result: We found reduction of NAD-dependent substrate oxidation rate in metabolic state 3 in the heart mitochondria of rats with fructose-induced insulin resistance by 30% in comparison with intact control (p<0.02). However, FAD-dependent substrate oxidation rate was not changed in all experimental groups. Administration of T normalised the rate of glutamate/ malate oxidation in the state 3 (F+T: 79.04±3.97 vs F: 66.1±5.00; C: 82.60±7.48 nmol/min/mg of protein, p<0.05) and the values of respiration control (F+T: 5.38±0.32 vs F: 4.63±0.25 ; C: 5.83±0.35, p<0.02). Improvement of mitochondrial function in the
heart of insulin resistant rats treated with T accompanied by reduction in mitochondrial lipid hydroperoxides contents (p<0.05), enhanced GR activity (F+T: 35.21±2.52 vs F: 22.69±1.31; C:38.43±2.16 nmol NADPH/min/mg of protein, p=0.02) and GSH level (F+T: 3.67±0.29 ±0.60 vs F: 2.10±0.27; C:3.84±0.28 nmol/mg of protein, p<0.01).

Conclusion: These results demonstrate that taurine protects against impairment of respiratory chain activity complex I and attenuates oxidative stress in the heart mitochondria of rats with fructose-induced insulin resistance. Thus, this action of taurine could be an important mechanism for providing benefits to the cardiovascular system in patients with insulin resistance and type 2 diabetes.

P 10: Enhancement of antioxidant defence against oxidative stress by timolol-treatment prevents age-/diabetes-related cardiac Ca2+ handling

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Objectives: We investigated whether defective intracellular Ca2+ handling can be controlled by timolol in age- as well as in diabetes-related cardiac dysfunction.

Background: It has been shown that the defects observed in mechanical activity of heart from both aging and diabetic subjects include alteration of intracellular Ca2+ handling via changes in critical processes responsible for its regulation, in part due to increased oxidative stress. Furthermore, increasing evidence shows a marked beneficial effect with beta-blockers, additional to their blocker actions, in heart dysfunction via scavenging free radicals and/or acting as an antioxidant.

Methods: To explore the antioxidant role of chronic timolol treatment (25 mg/kg, daily, 12 weeks) on either age- or diabetes-related changes in heart function, we used Langendorff-perfused rat hearts to determine hemodynamic parameters, patch-clamp technique to monitor L-type Ca2+-current (ICaL), and confocal microscopy to study intracellular both global Ca2+ transients evoked by electrical stimulation and local Ca2+ changes (sparks) in quiescent cardiomyocytes loaded with fluorescence Ca2+ dye fluo-3AM.

Results: Normal cardiac function was well preserved in timolol-treated either diabetic or 12-month old rats compared to their age-matched controls. Moreover, our data strongly point out that this reverse remodelling is associated with normalization of the diastolic Ca2+, ryanodine receptor Ca2+ release channel (RyR2) macromolecular complex, ICaL function, unbalanced oxidant/antioxidant-defence system as well as cellular redox state both in circulation and in heart. The antioxidant N-acetyl-L-cysteine showed an effect similar to that of timolol.

Conclusion: Timolol, at a low concentration that is sufficient to produce antioxidant effect, improves the intracellular Ca2+ handling and contractile dysfunction by preventing the protein-thiol oxidation in leaky-RyR2 from both aged and diabetic rat hearts.

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P 11: Role of herbal extracts in the prevention of oxidative stress and modulation of atherogenic risk factors in experimentally-induced DM

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Background and aims: Complementary and alternative medicine modalities of treating diabetes mellitus and cardiovascular diseases (D&CVD) have been used in various parts of the world. Chemoprevention and chemotherapies using the naturally occurring nontoxic natural compounds has been advanced as a feasible strategy for D&CVD management.

Diabetes and cardiovascular diseases are currently the leading cause of death globally. Diabetes has a distinctive association with CVD. Diabetes is well known to induce physiological stress response that involves activation of inflammatory, metabolic, and immunological mediators. Considerable data indicate that reactive oxygen species (ROS) and oxidative stress are important features of CVD including atherosclerosis, hypertension, and congestive heart failure.

Clinical research has confirmed the efficacy of phyotherapy in the modulation of oxidative stress-induced CVDs associated with DM. Increasing number of studies reported that an increase in plasma total antioxidant capacity (TAC) is associated with intake of herbs, fruits and vegetables which are rich in antioxidants. This is based on findings that Leaf extract components, especially flavonoids have the free radical scavenging activity, showing the potential of antioxidant properties. Consequently, the present study was designed to investigate the hypoglycemic, hypolipidemic and antioxidative effects of crude extract from leaves of Zizyphus spina-christi (ZS), Morus alba (MA) and Olea europeaea (OE) in experimentally induced DM.

Materials and Methods: Fresh leaves were dried, extracted by methanol, and then vacuum evaporated. Crude extracts from leaves of Zizyphus spina-christi (ZS), Morus alba (MA) and Olea europeaea (OE) were freshly dissolved in distilled water just before oral administration. Experimental rat model of T2DM was induced in male rats by a single ip injection of 50 mg/kg streptozotocin. All experimental groups received herbal extracts orally and individually at a dose of 0.1g/kg/day for 5 weeks. The body weight and fasting blood glucose level of all rats were measured before and after 5 weeks of treatment. In addition, HBA1c, serum insulin, lipid profile, and cardiac antioxidants as well as lipid and protein oxidation were evaluated.

Results: After a period of 5 weeks, the body weight of diabetic rats was significantly (p<0.05) decreased as compared to their initial ones. On the other hand, the body weight of diabetic rats treated with extracts of ZS, MA, or OE (0.1g/kg/day) were increased in the same way as normal control rats. Diabetic rats showed a significant (p<0.05) decrease in serum insulin, hyperglycemia, elevated HBA1c, altered lipid profile. Concurrent with these changes, there was an increase in the concentration of oxidative stress markers in the heart. This oxidative stress was related to decreased antioxidant levels in the heart of the diabetic rats. Application of crude extract of the three plants (ZS, MA &OE) given individually resulted in a significant (p<0.05) amelioration of the alteration of serum insulin, glucose, HBA1c, total lipid, LDL, HDL, total cholesterol and TG as well as cardiac malondialdehyde, protein carbonyl, superoxide dismutase, catalase, glutathione, glutathione-S-transferase, total antioxidant capacity, and nitric oxide. Extracts of ZS, MA, & OE on their own significantly increased (p< 0.05) the GSH level, CAT activity and TAC in heart of the control rats. The alterations of cardiac antioxidants were correlated with the observed atherogenic alterations in serum of the diabetic rats. However, treatment with the three extracts independently significantly retained cardiac redox state and the estimated atherogenic parameters in serum towards their normal level. This was attributed to the ability of these extracts to empower the antioxidant defense of the heart.

Conclusion: Our data indicate that experimental diabetes led to alteration in the atherogenic parameters, with an increasing incidence of oxidative stress in the heart, and the effects of the extract might be attributed to the hypoglycaemic and antioxidative potential of flavonoids, the major components of the plant extract. These results provide the first evidence for the efficacy of ZS, MA or OE in