g/kg/die). Di questi pazienti è stata raccolta la storia clinica, i parametri antropometrici e biochimici, in un follow-up di 7 anni per valutare la progressione di CKD (ingresso in dialisi).
Risultati. La valutazione del regime dietetico ha permesso di individuare 22 pazienti con DL, 87 con VLPD e 31 con LPD. La GFR basale non era significativamente differente tra I tre gruppi (16,2+6,1 vs 16,8+6,8 vs 20,9+9,8 mL/min/1,73 m^2, per dieta libera, VLPD e LPD rispettivamente, P=0,066). Dopo il periodo di follow-up, 64 sono entrati in dialisi. Di questi, 33 (51.5%) erano con DL, 27 erano con VLPD e 4 (6.2%) erano con LPD. In generale, il controllo dell'apporto proteico giornaliero era associato con una minore incidenza di ingresso in dialisi, se confrontata con DL (P<0,001). Infine, abbiamo osservato che l'effetto del controllo dell'apporto proteico era indipendente dalla terapia anti-ipertensiva.
Conclusioni. La dieta ipoproteica si associa a una ridotta progressione della CKD. Inoltre, questo effetto è risultato indipendente dalla terapia farmacologica anti-ipertensiva, suggerendo l'utilità clinica del regime alimentare ipoproteico sul controllo della CKD.

SOCIETÀ ITALIANA IPERTENSIONE ARTERIOSA (SIIA)
E SOCIETÀ ITALIANA PER LO STUDIO DELL'ARTERIOSCLEROSI (SISA)

Spring Meeting dei Giovani Ricercatori SIIA e SISA

IPERTENSIONE ARTERIOSA, DISLIPIDEMIE ED ATEROSCLEROSI: NUOVI LINK FISIOPATOLOGICI E TERAPEUTICI

Rimini, 11-12 Marzo 2016

COMUNICAZIONI GIOVANI RICERCATORI

Abstracts selezionati dalla Commissione Scientifica:
Rosa Maria Bruno, Arrigo F.G. Cicero, Elda Favari, Giuseppe Danilo Norata e Martina Rosticci

COMUNICAZIONI ORALI

THE EFFECT OF MELATONIN ON SMALL MESENTERIC ARTERIES AND AORTA OF AGING MICE

C. Agabiti Rosei, C. De Ciuceis, C. Rossini, E. Porteri, R. Rezzani[1], L.F. Rodella[1], G. Favero[1], A. Sarkar, E. Agabiti Rosei, D. Rizzoni
Clinica Medica and [1]Chair of Human Anatomy, Department of Clinical and Experimental Sciences, University of Brescia

Background. It has been previously demonstrated that inflammation in adipose tissue may be implicated in vascular dysfunction (Circulation 2009; 119: 1661-1670). A senescence-accelerated prone mouse (SAMP8) is a model of age-related cognitive decline and vascular dysfunction. Several studies demonstrated that SAMP8 suffers from increased oxidative stress and that accelerated senescence was associated with decreased eNOS and nNOS and increased O(2) synthesis. Aim of the study was to investigate contractile response and age-related vascular changes on the endothelium of small mesenteric arteries and aorta in SAMP8 in normal conditions and after chronic treatment with melatonin, an endogenous hormone with antioxidant and vasculoprotective properties.
Materials and methods. We investigated 7 SAMP8 and 7 SAMR1 normal controls. In each mice we analysed markers of aging on endothelium of aorta. Mesenteric small resistance arteries were dissected and mounted on a wire myograph (internal diameter 200 µm). A concentration-response to noradrenaline (NA, from 10-9 to 10-5 Mol/l) was evaluated in vessels with intact perivascular fat tissue (WF) and in vessels in which perivascular fat tissue was removed (NoF).
Investigations were repeated in 7 SAMP8 and 7 SAMR1 after 54 weeks of treatment with melatonin.
Results. In SAMP8 mice we have observed an overexpression of marker of oxidative stress. in SAMR1 control mice anticontractile effect of perivascular fat was present (WF vs. NoF: ANOVA p=0.04), while in aging SAMP8 mice the effect was less pronounced (WF vs NoF: ANOVA p=NS). Long-term treatment with melatonin had no effect in SAMR1, while in SAMP8 it was able to increase some vasculo-protective markers and to restore the anticontractile effect of perivascular adipose tissue (ANOVA p<0.001).
Conclusion. The anticontractile effect of perivascular fat is impaired in SAMP8, compared with controls. A long-term treatment with melatonin seems to reduce oxidative stress and to restore anticontractile effect of perivascular fat in SAMP8, maybe through its antioxidant properties.

EPCS BEFORE AND AFTER TREATMENT WITH METFORMIN IN PATIENTS WITH FCHL AND INSULIN RESISTANCE

C.O. Aragona[1], C. Conte[2], A. Lo Gullo[1], V. Cairo[1], F. Mamone[1], E. Imbalzano[1], M.A. Sardo[1], A. Saitta[1], G. Mandraffino[1]
[1]Department of Clinical and Experimental Medicine, University of Messina; [2]Division of Endocrinology and Metabolic Diseases, Catholic University of Sacred Heart, A. Gemelli Medical School of Rome

Aim. Familial combined hyperlipidemia (FCHL) is a disorder of metabolism, associated to increased cardiovascular (CV) risk, characterized by the overproduction of very low-density lipoprotein, by the altered remnants catabolism, and by an high prevalence of insulin resistance (IR). Endothelial progenitor cells (EPCs) are a heterogeneous population of cells with the ability to differentiate into cell types of different organs and systems. CD34+EPCs have already been evaluated in different clinical conditions: it was found associations between the severity of endothelial damage and cell count. IR was acknowledged as a strong prevalence predictor of CV disease. Several authors have suggested that metformin may increase EPC levels in diabetic patients, but to date there are no studies about CD34+EPCs in non-diabetic-IR patients.
Methods. We evaluated CD34+EPC count in newly diagnosed FCHL/IR patients at baseline (T0) and after 6-months metformin treatment (T1). We enrolled 29 patients (M:F 20:9; age 46±11) with no further CV or metabolic risk factor.
Results. We found that, after metformin treatment, there is a significant reduction of glucose (Δ=4.45%; p<0.05), plasmatic insulin (Δ=-18%; p<0.05), HOMA-IR (Δ=-19.9; p<0.05) and an increase in CD34 count (Δ=-69.9%; p<0.05). Moreover, we found a correlation between CD34 count with plasmatic insulin (r-0.571, p=0.01) and HOMA-IR (r-0.583, p=0.009). Dependence analysis showed an association between ΔHOMA and ΔCD34+ cells (t-2.961, p=0.009).
Conclusions. Our study suggest that in FCHL/IR patients a treatment with metformin, also by improving insulin-sensitivity, may increase CD34+EPC amount, likely influencing CV risk.

EFFECT OF STATIN TREATMENT ON ARTERIAL STIFFNESS IN INDIVIDUALS WITH NEWLY-DIAGNOSED FAMILIAL HYPERCHOLESTEROLEMIA

N. Artom[1], M. Canepa[2], A. Telchime[1], P. Ameri[2], C. Brunelli[2], F. Dallegri[1], A. Pende[1], L. Pisciotta[1]
[1]Clinic of Internal Medicine 1, University of Genoa, IRCCS AOU San Martino - IST, Genoa; [2]Cardiology Unit, University of Genoa, IRCCS AOU San Martino - IST, Genoa

Aim. Pulse wave velocity (PWV) is considered a major indicator of arterial stiffness, which quickly responds to therapeutic interventions, particularly antihypertensive medications. We sought instead to assess the short-term effect of HMG-CoA reductase inhibitors (statins) on PWV in individuals with newly diagnosed familial hypercholesterolemia.
Methods. Individuals fulfilling the inclusion criteria (i.e. statin-naïve with LDL cholesterol ≥190 mg/dl and a Dutch Lipid Clinic Network score ≥6) underwent physical examination, anthropometrics, blood lipids, and PWV at the screening visit and after a 3 month statin treatment. Carotid-femoral PWV was measured with the Sphygmocor CPV device, using 80% of the direct carotid-femoral distance and averaging three consecutive measurements.
Results. From November 2014, we screened more than 500 individuals and found 12 that fulfilled our strict inclusion criteria. At the time of the present analysis, 7 individuals (3 men, mean age 46±18 years, only one taking antihypertensive medications) who were all given 5-10 mg rosuvastatin completed the study. At 3 month follow-up we found a significant 37% reduction in total cholesterol (from 345±29 to 217±54 mg/dL, p=0.003) and 46% reduction in LDL cholesterol (from 259±21 to 141±46 mg/dL, p=0.002). Peripheral blood pressure also slightly decreased (systolic from 121±10 to 116±14 mmHg, p=0.18; diastolic from 72±8 to 67±10 mmHg, p=0.04), as well as heart rate (from 70±7 to 63±10 bpm, p=0.09). Concomitantly, we observed a significant 10% reduction in PWV (from 8.4±1.5 to 7.5±0.8 m/sec, p=0.036), which persisted after adjustments for mean blood pressure and heart rate. There was no significant change in anthropometrics and central-pressure parameters.
Conclusions. Our preliminary results suggest that in individuals with familial hypercholesterolemia, a 3-month treatment period with rosuvastatin determines a significant reduction in arterial stiffness, possibly through its pleiotropic vascular effect beyond lowering of plasma cholesterol.

DEVELOPMENT OF AN UHPLC-MS/MS METHOD FOR THE THERAPEUTIC DRUG MONITORING OF ANTIHYPERTENSIVE DRUGS IN HUMAN PLASMA: VALIDATION ON PATIENTS WITH "RESISTANT" HYPERTENSION

V. Avataneo*[1], A. De Nicolò*[1], G. Bonifacio[1], F. Rabbia[2], E. Perlo[2], P. Mulatero[2], F. Veglio[2], G. Di Perri[1], A. D'Avolio[1]
[1]Unit of Infectious Diseases[#], University of Turin, Department of Medical Sciences, Amedeo di Savoia Hospital, Turin; [2]Division of Internal Medicine and Hypertension Unit, University of Turin, Department of Medical Sciences, AOU "Città della Salute e della Scienza", Turin; [#]Laboratory of Clinical Pharmacology and Pharmacogenetic. UNI EN ISO 9001:2008 Certificate Laboratory; Certificate No. IT-64386; Certification for: "Design, development and application of determination methods for anti-infective drugs. Pharmacogenetic analyses." www.tdm-torino.org
**Both authors contributed equally to this work.*

Aim. The management of resistant hypertension (RH) is a critical health problem. One of the main issues is to discriminate "true" RH patients from cases of poor medication adherence, as called pseudoresistant hypertensives. A solution to this problem is Therapeutic Drug Monitoring (TDM), based on the measurement of drugs concentrations during treatment, of doubtful cases: however, all the existing quantification methods are good only for a few drugs and/or too expensive for a routine use.
The aim of this work was to validate and test on real-life patient samples an UHPLC-MS/MS method capable of simultaneously monitoring plasma concentrations of ten currently used antihypertensive drugs: amlodipine, atenolol, clonidine, chlortalidone, doxazosin, hydrochlorothiazide, nifedipine, olmesartan, ramipril and telmisartan.
Methods. Each sample was added with internal standard (IS, 6,7-dimethyl-2,3-di (2-pyridyl)quinoxaline) and underwent a protein precipitation protocol with acetonitrile. After a drying step, extracts were resuspended in water:acetonitrile 90:10 (v:v) and then analyzed through a Shimadzu Nexera X2 UHPLC system coupled with a LCMS-8050 tandem mass detector. The validated method was tested on real samples from patients with RH/pseudo-RH.

Results. Accuracy, intra-day and inter-day precision of the method fitted FDA guidelines for all analytes. 13 samples from 13 patients, all giving informed consent, were analyzed. On the basis of preliminary data, out of a total of 13 patients, 2 resulted partially non-adherent and 3 candidates to renal denervation were totally non-adherent. Continuing the TDM of those patients, after a period of tight control, two of them became adherent.
Conclusions. The simple, fast and cheap extraction procedure makes this method eligible for a clinical routine use.
From the clinical point of view we obtained encouraging results: we managed to discriminate some cases of poor adherence to the therapy and preserve those patients from an invasive and expensive therapeutic approach.

SYSTEMIC LUPUS ERYTHEMATOSUS FLARE-UP IS ASSOCIATED WITH INCREASED 5-YEARS CAROTID INTIMA-MEDIA THICKNESS PROGRESSION

A. Baragetti[1,2], M. Magnoni[6], K. Garlaschelli[2], L. Grigore[2], M. Berteotti[6], I. Scotti[6], E. Bozzolo[4], A. Berti[4], A.A. Manfredi[4], E. Ammirati[5], A.L. Catapano[1,3], G.D. Norata[1,2]
[1]Department of Pharmacological and Biomolecular Sciences, University of Milan; [2]Center for the Study of Atherosclerosis, Bassini Hospital, Cinisello Balsamo (Milano); [3]IRCCS, Multimedica Hospital, Sesto San Giovanni (Milano); [4]Unit of Medicine and Clinical Immunology, IRCCS San Raffaele Scientific Institute, Milan; [5]Niguarda Ca' Granda Hospital, Milan; [6]Department of Thoracic and Cardiovascular Surgery, University Vita-Salute San Raffaele Scientific Institute, Milan

Aim. Patients with Systemic Lupus Erythematosus (SLE) present increased carotid Intima-Media Thickness (c-IMT) and cardiovascular mortality compared to general population. Our aim was to investigate in SLE patients the association of immunoinflammatory disease activity and of classical cardiovascular risk (CVR) factors with five years c-IMT progression (Δc-IMT).
Methods. Clinical and pathological history, anthropometric, biochemical parameters were collected at baseline and after 5 years in 50 SLE patients (42+9 years-old) and 50 age and gender matched controls. SLEDAI score and disease flare-up during follow-up, disease activity indexes, were reported according to guidelines. Ultrasound c-IMT was measured to evaluate progression.
Results. SLE patients within the highest tertile of basal SLEDAI score presented a faster increase in c-IMT versus age and gender matched controls (0.007 (0.006) mm/year vs 0.003 (0.001) mm/year respectively, P=0.026) while, Δc-IMT was not associated nor with CVR factors neither with basal lupus serology (C3, C4 and anti-sDNA). During the 5 years of follow up, Disease flare-ups were more frequent with high SLEDAI (P= 0.037) and in those with faster Δc-IMT compared to those with low disease activity (0.008 (0.004) mm/year vs -0.006 (0.004) mm/year, P=0.021).
Elevated LDL-C levels were the only CVR factor associated with increased disease flare-up during the follow-up; this association might be the consequence of the aggressive immunosuppressant therapy in these patients.
Conclusions. SLE patients present increased c-IMT progression, which is associated more to disease activity rather than classical CVR factors, supporting a key role for the inflammatory response during vascular disease progression in patients with autoimmune diseases.

DIFFERENT IMPACT OF ALCOHOL CONSUMPTION ON THE REVERSE CHOLESTEROL TRANSPORT IN VIVO

S. Battista[1], D. Greco[1], L. Mele[2], A. Piemontese[1], A. Poli[3], F. Bernini[1], I. Zanotti[1]
[1]Dept of Pharmacy, University of Parma; [2]Dept of Food Science, University of Parma; [3]Nutrition Foundation of Italy, Milan

Epidemiological studies revealed that moderate and binge alcohol consumption exerts opposite effect on cardiovascular disease. Atherosclerotic cardiovascular disease is inversely correlated with reverse cholesterol (RCT), the process promoting the removal of excess cholesterol from arterial wall.
We aim to evaluate whether moderate and binge alcohol consumption may differently impact RCT in an animal model of atherosclerosis-prone mice. RCT was measured through a standardized, radiolabeled technique in apolipoprotein E knockout mice: placebo group (n=9) received water, mimicking the abstainers; moderate group (n=10) received 0.8 g/kg alcohol/day for 28 days, mimicking a moderate alcohol consumption; binge group (n=10) received 0.8 g/kg alcohol/day for 5 days, followed by the administration of 2.8 g/kg alcohol/day for 2 days/week, mimicking a binge alcohol consumption. Binge alcohol consumption caused an increase of 37.2% in plasma total cholesterol and an increase of 44.35% in HDL-C levels versus placebo group. Binge group also showed an increase of 30.95% in LDL-C and an increase of 23% in triglycerides compared to placebo group. Conversely, moderate consumption does not affect plasma lipoprotein profile. The removal of radioactivity from macrophages along RCT pathway was higher in the moderate group (12.2%+3.1, 15.1%+3.7, 13.3%+2.4 in placebo, moderate and binge group respectively). In conclusion, moderate alcohol consumption promotes the removal of cholesterol from macrophages along RCT pathway. Conversely binge alcohol consumption exerts deleterious effects on lipoprotein profile, but it does not seem to significantly affect RCT process.

ARMOLIPID PLUS SIGNIFICANTLY REDUCED LEFT VENTRICULAR MASS IN SUBJECTS WITH METABOLIC SYNDROME. A MULTICENTRIC, DOUBLE-BLIND, RANDOMIZED, PLACEBO-CONTROLLED TRIAL

Francesca Battista[1], G. Pucci[1], F. Battista[1], G. Bosso[2], V. Fazio[3], V. Mercurio[2], M. Sarnataro[2], N. Brambilla[4], C. Vitalini[4], M. D'Amato[4], G. Giacovelli[4], F. Galletti[3], D. Bonaduce[2], G. Schillaci[1]
[1]Dipartimento di Medicina, Università di Perugia, S.C. Medicina Interna A.O.U. Santa Maria Terni; [2]Dipartimento di Scienze Mediche Traslazionali, Università Federico II di Napoli; [3]Dipartimento di Medicina e Chirurgia, Università Federico II di Napoli; [4]Rottapharm Biotech, Monza

Introduction. Subjects with metabolic syndrome (MS) frequently show left ventricular hypertrophy (LVH). The role of nutraceuticals with associated lipid-lowering properties in LVH regression is unknown. We evaluated the effects of 24 weeks of treatment with Armolipid Plus™ (ARMP, berberine 500 mg, red yeast rice 200 mg, and policosanol 10 mg, Rottapharm Madaus - Monza, Italy) vs placebo on LVH regression, in subjects with MS in a multicentric, double-blind, randomized, placebo-controlled study (www.clinicaltrias.gov: NCT02295176).
Methods. 150 subjects with MS, aged between 28 and 70 years, and with LVH at echocardiography (LV mass/height 2,7>44 in females,

>48 in males), were evaluated. Subjects were excluded if diabetics, if had a BMI>35 Kg/m^2, GFR<30 mL/min/1,73 m^2, heart failure and uncontrolled arterial hypertension (>140/90 mmHg despite treatment). The primary end-point was the number of subjects with regression of LVH; The secondary end-points was the rate of reduction in LV mass compared to baseline. Sensitivity analyses aimed to evaluate the impact on confounding factors on the main results. The analyses were intention-to-treat and on-treatment.
Results. 140 subjects (93%) completed the study. 72 were randomized to ARMP and 68 to placebo. There were no differencies in terms of sex, age, BMI, waist circumference, anti-hypertensive treatment (previous or new-onset) and LV mass between groups (all p=ns). LVH regression occurred in 10 subjects (7%) randomized to ARMP and in 3 subjects (4%) in the placebo group (p=0.22). Compared to baseline, the relative reduction in LV mass was significantly higher in subjects randomized to ARMP than in subjects randomized to placebo (-3% vs +1%, p<0.0001). The number of subjects showing LV mass reduction was significantly higher in the ARMP group than in the placebo group (56% vs 44%, p<0.0001). These differencies remained significant even after adjustment for age, sex, previous or new-onset anti-hyertensive or lipid-lowering treatment, baseline values of SBP/DBP, BMI, waist circumference, HDL-cholesterol, tryglicerides, serum glucose, HOMA, HbA1c, and their relative changes during treatment (p=0.003).
Conclusions. After 24 weeks of treatment, ARMP significantly and independently reduces LV mass in subjects with MS, as compared to the placebo group and independently from concomitant changes in BP, BMI, and other confounding factors. Treatment with ARMP may contribute to reduce negative consequences of an increased LV mass and the associated increased CV risk in subjects with MS.

CARDIOVASCULAR DAMAGE IN PATIENTS WITH PRIMARY ALDOSTERONISM OR ESSENTIAL HYPERTENSION

F. Bertacchini, A. Paini, C. Agabiti Rosei, C. Aggiusti, M. Salvetti, C. Donini, E. Casella, E. Agabiti Rosei, M.L. Muiesan
Università degli Studi di Brescia

Background. Primary aldosteronism is a relatively common condition in hypertensive patients. Only few studies, in small groups of patients, have evaluated large arteries alterations. In some, but not in all studies, positive relationship with vascular damage was observed.
Aim of the study. To compare the prevalence of cardiac and large arteries vascular organ damage in patients with essential hypertension (EH) or primary aldosteronism (PA).
Design and method. In 243 consecutive patients with no interfering therapy (147 M, mean age 48±11 years) a routine blood sample, including measurement of aldosterone/renin ratio (ARR) and saline load if ARR>30, was obtained. Echocardiography, carotid ultrasound and measurement of pulse wave velocity (PWV) were performed. We considered 3 groups: 48 patients with EH (ARR <30); 122 patients with positive ARR screening but negative saline load (indeterminate aldosteronism, IA); 73 patients with PA (positive ARR and post saline aldosterone >100 ng/ml) (51% with adrenal adenoma).
No differences between groups were observed in age, gender, BMI, BP values (clinic and 24 hours), glucose, lipids and renal function. LVMI was greater in PA vs both IA and EH (PA 45±18, IA 39±12, EH 39±10 gr/m 2.7, p<0,05). Left atrial volume/BSA was significantly greater in PA vs EH (PA 27±10, IA 24±8, EH 23±6 ml/m^2, p<0,05 for PA vs EH). A positive correlation was observed between ARR and LVMI (r=0,20 p=0,002), left atrium volume (r=0,201, p<0,001) and relative wall thickness (r=0,394, p<0,005). Indices of vascular damage did not differ between groups Aldosterone levels and ARR were not significantly correlated with indices of vascular damage.
Conclusions. A greater prevalence of cardiac, but not of large arteries damage is observed in PA as compared to EH when a simultaneous assessment of cardiac and vascular OD is performed.

INDOLEAMINE 2,3-DIOXYGENASE INTERFERES WITH THE INFLAMMATION-INDUCED CIRCULATING ENDOTHELIAL PROGENITOR CELL DEPLETION

V. Bianconi[1], F. Fallarino[2], M.R. Mannarino[1], M. Gargaro[2], A. Turco[2], G. Scalisi[2], M. Pirro[1]
[1]Unit of Internal Medicine, Department of Medicine. University of Perugia; [2]Unit of Pharmacology, Department of Experimental Medicine, University of Perugia

Aim. Systemic low-grade inflammation secondary to exposure to cardiovascular risk factors and the concomitant reduced bioavailability of endothelial progenitor cells (EPCs) are associated with an increased cardiovascular (CV) risk. Indoleamine 2,3-dioxygenase (IDO), an immunosuppressive enzyme involved in tryptophan degradation into kynurenine, has been identified as a pathology mediator in the context of inflammation. We investigated whether IDO activity is associated with the availability of circulating EPCs in the presence of variable exposure to low-grade systemic inflammation.
Methods. The ratio of tryptophan to kynurenine (a measure of IDO activity), high sensitivity C-reactive protein (hsCRP) and the number of EPCs were measured in 250 patients with either hsCRP ≤3 mg/L or >3 mg/L. Moreover, the effect of IDO deficiency and kynurenine supplementation on the number of EPCs has been investigated in C57BL/6 and IDO-knockout (KO) mice in the presence or absence of lipopolysaccharide (LPS)-induced low-grade systemic inflammation.
Results. Patients with low-grade inflammation had reduced EPCs levels than those without inflammation. The number of EPCs was inversely associated with hsCRP levels (r=-0.13, p=0.036) and positively with IDO activity (r=0.16, p=0.008). The presence of high CRP levels was paralleled by a significant reduced EPC number in patients with low IDO activity (r=-0.21, p=0.010)., but not in those with high IDO activity (r=-0.04, p=0.692). IDO-KO mice had lower EPC levels than C57BL/6 mice. Oral treatment with kynurenine (1 mg every other day) was paralleled by a significant increase in the number of EPCs in C57BL/6 mice and prevented the LPS-induced EPC decline.
Conclusions. Low-grade systemic inflammation reduces EPC levels; however, increased IDO activity and kynurenine supplementation seem to improve EPC bioavailability and to limit the inflammation-induced circulating EPC loss.

LIPOPROTEIN PHENOTYPE IN NAÏVE PATIENTS WITH KLINEFELTER SYNDROME

P. Bigolin, A. Antonucci, A. Bonaldo, P. Scarinzi, M. Barichello, N. Caretta, M. Ghezzi, R. Selice, M. Terrin, D. Vianello, R. Marin, A. Ferlin, C. Foresta, F. Fabris, E. Manzato, S. Bertocco, S. Zambon, A. Zambon
Department of Medicine, University of Padova

Aim. Klinefelter Syndrome (KS) is associated with increased cardiovascular disease morbidity and mortality. The study aims to in-

vestigate quantitative and qualitative lipoprotein abnormalities, including LDL density and oxidation, in naïve KS patients, focusing on lipoprotein phenotypes as compared to age-matched.
Methods. Anthropometric data, fasting blood samples (glycaemia, HbA1c, HOMA, LH, FSH, testosterone, SHBG, TSH), lipid profile, qualitative lipoprotein analysis by density gradient ultracentrifugation (DGUC), and oxidized LDL (ox-LDL) were analyzed in 30 naïve KS patients, 30 male and 40 age-matched female controls.
Results. KS patients are characterized by significantly higher total and HDL-cholesterol, and triglycerides than male controls, and significantly higher total, LDL-cholesterol, and triglycerides than female controls. By DGUC, KS patients show increased HDL and VLDL-cholesterol, and reduced dense LDL particles vs male (all $p<0.05$), and increased VLDL-cholesterol vs female controls. KS patients in the two upper tertiles of waist circumference (WC>91.3 cm) had a proatherogenic lipid profile with higher triglycerides, lower HDL, increased dense LDL (Rf=0.369 vs 0.392; $p<0.05$), and ox-LDL (61.1±16 vs 46.8±10 U/L, $p<0.05$) vs. the lower WC tertile group. We found a significant correlation between ox-LDL and dense LDL fractions by DGUC (all with $p<0,05$). By multivariate analysis, low testosterone levels were associated with increased WC (OR=0.73, 95%CI 0.54-0.97; p=0.029).
Conclusions. KS patients are characterized by a peculiar lipoprotein profile as compared to age-matched controls. In KS an increased WC is associated with a highly atherogenic lipid profile (increased triglycerides, dense and ox-LDL, reduced HDL). Low testosterone levels independently and significantly contribute to an increased WC.

SYMPATHETIC NEUROTRANSMISSION DURING ATHEROSCLEROSIS DEVELOPMENT: AN UNRECOGNIZED TARGET OF DYSLIPIDEMIA?

M. Busnelli[1], S. Manzini[1], D.S. Horner[2], M. Chiara[2], G.S. Ganzetti[1], F. Dellera[1], C.R. Sirtori[1], C. Parolini[1], G. Chiesa[1]
[1]Department of Pharmacological and Biomolecular Sciences, Università degli Studi di Milano; [2]Department of Biosciences, Università degli Studi di Milano

Aim. With the aim of discovering new genes/pathways involved in dyslipidemia-driven atherosclerosis, transcriptomic analysis was conducted on aortas of several transgenic mouse lines, with different lipid/lipoprotein profiles and different susceptibility to atherosclerosis.
Methods. C57Bl/6, apoE-deficient (EKO), apoE/apoA-I deficient (EKO/A-IKO) and apoE/apoA-I-deficient mice overexpressing human apoA-I (EKO/A-IKO/hA-I) were studied. Mice were fed chow or Western diet, starting from 8 weeks of age and were analyzed after 22 weeks of diet for plasma lipoprotein profile by FPLC and for aortic atherosclerosis by en-face analysis. The entire gene expression profile of murine aortas was investigated by a high-throughput sequencing approach (transcriptomics).
Results. On chow diet, plaques were visible only in the aortic arch of EKO and EKO/A-IKO mice, characterized by low or absent HDL, respectively, and cholesterol accumulation in VLDL-LDL. Western diet worsened hyperlipidemia and plaque formation in the aortic arch of EKO and EKO/A-IKO mice and led to modest atherosclerosis development in EKO/A-IKO/hA-I mice, characterized by elevated VLDL-LDL cholesterol levels and a large HDL cholesterol peak. Out of a total of 23,000 genes, about 2,300 genes were identified as differentially expressed in at least one condition. In the athero-prone genotypes, Western diet dramatically lowered the expression of genes coding for key enzymes of catecholamine synthesis and synaptic vesicular structure. A similar down-regulation was found in the low-HDL phenotype (EKO/A-IKO) compared with the high-HDL phenotype (EKO/A-IKO/hA-I), in both dietary conditions.
Conclusions. The sympathetic nervous system plays an established role in regulating vascular tone. In addition, some studies indicated that sympathetic neurotransmission deficiency may affect plasma cholesterol levels and lead to aortic cholesterol accumulation. Our data suggest that dyslipidemic conditions predisposing to atherosclerosis development (i.e. hyperlipidemia; low HDL levels) may interfere with the arterial sympathetic innervation by down-regulating the expression of genes involved in catecholamine biosynthesis, as well as in synaptic plasticity and transmission.

RENAL SODIUM HANDLING AND RISK TO DEVELOP HYPERTENSION AFTER 8-YEAR FOLLOW-UP: RESULTS OF THE OLIVETTI HEART STUDY

L. D'Elia, G. Rossi, I. Savino, M. Schiano di Cola, R. Iacone, O. Russo, E. La Fata, F. Galletti, P. Strazzullo
Department of Clinical Medicine and Surgery, ESH Excellence Center of Hypertension, "Federico II" University of Naples Medical School, Naples

Aim. The aim of this investigation was to estimate the predictive role of the renal sodium handling on the risk to develop HP and BP changes in an 8 year follow-up of a sample of men (The Olivetti Heart Study).
Methods. This study included 294 untreated normotensive non diabetics men, with normal renal function evaluated twice in 1994-95 and in 2002-04. Renal tubular sodium handling was estimated by exogenous lithium clearance and for this analysis FPRNa and distal sodium reabsorption (FDRNa) were considered.
Results. At baseline, higher tertile of FPRNa was associated with lower diastolic BP (DBP) and higher prevalence of smokers than lower tertile. Prevalence of baseline smoking habit was also greater in the last tertile of FDRNa. After 8-year of follow-up, there was a significant increase in BP from first to third tertile of FPRNa (SBP: I=9.8±11.0, II=14.9±14.2, III=16.4±15.2 mmHg; DBP: I=6.9±8.8, II=8.1±9.1, III=11.2±8.9 mmHg), confirmed also accounting for main potential confounders. While, there was not difference in BP changes across tertiles of FDRNa. In consideration of HP incidence of 52% in this sample, a significant difference was found across tertiles of FPRNa (p for trend =0.02), but not across FDRNa tertiles. In addition, multivariate analysis supported that baseline FPRNa was a significant predictor of HP, independently of potential confounders (OR: 1.63, 95%CI: 1.15 -2.33).
Conclusions. The results of this investigation indicated a predictive role of FPRNa on the changes in BP over time and on the risk to develop HP in a sample of healthy adult men.

RESPONSE TO TREATMENT AND OCCURRENCE OF CARDIOVASCULAR (CV) COMPLICATIONS IN PATIENTS WITH AUTOSOMAL RECESSIVE HYPERCHOLESTEROLEMIA (ARH): A RETROSPECTIVE ANALYSIS

L. D'Erasmo[1], I. Minicocci[1], L. Masana[2], S. Muntoni[3], P. Pintus[4], F. Bertolini[5], C.R. Sirtori[6], L. Calabresi[6], C. Pavanello[6], M.R. Averna[7], AB. Cefalù[7], D. Noto[7], A. Pacifico[8], G. Pes[9],

M. Harada-Shiba[10], J.R. Roeters Van Lennep[11], R. Fellin[12], E. Manzato[13], S. Zambon[13], M. Arca[1]
[1]Department of Internal Medicine and Allied Sciences, Sapienza University of Rome, Rome; [2]Unitat de Recerca en Lípids i Arteriosclerosi, Hospital Universitari de Sant Joan, Universitat Rovira i Virgili, CIBERDEM, Reus, Spain; [3]Department of Biomedical Sciences, University of Cagliari and Centre for Metabolic Diseases and Atherosclerosis, The ME.DI.CO. Association, Cagliari; [4]Centro per le Malattie Dismetaboliche e l'Arteriosclerosi, Dipartimento Internistico, Cagliari; [5]Department of Internal Medicine, University of Genova, Italy; [6]Università degli Studi di Milano; [7]Biomedical Department of Internal Medicine and Specialistics (DIBIMIS), University of Palermo; [8]Unità Operativa Diabetologia e Malattie Metaboliche, Azienda Ospedaliero Universitaria, Sassari; [9]Department of Clinical and Experimental Medicine, University of Sassari; [10]Department of Molecular Innovation in Lipidology, National Cardiovascular Center Research Institute, Suita, Osaka, Japan; [11]Department of Internal Medicine, Erasmus University Medical Center, Rotterdam, The Netherlands; [12]Department of Medical Sciences, University of Ferrara; [13]University of Padova

Objectives. ARH is a rare, severe form of recessive hypercholesterolemia due to mutations in the LDLRAP1 gene. As a few data on clinical management and outcome are available, we have carried out a retrospective analysis of reported ARH cases.
Methods. All published ARH cases were identified by electronic search through PubMed and Medline. Corresponding authors were invited to provide published and unpublished information of patients according to a pre-specified database. The major endpoints of retrospective analysis were:
1) proportion of ARH patients under pharmacological and/or LDL apheresis (LA) treatment;
2) LDL-C lowering effect of treatments;
3) incidence of CV outcomes such as death, coronary and peripheral artery disease, aortic stenosis.

Results. We were able to collect complete data for 39 ARH patients (22 females/17 males; mean age 40.4, range 9-80 yrs). The mean observational period from diagnosis was 10.8±6.4 yrs. At follow-up, 61.5% of patients were treated with LA combined with lipid lowering drugs, while 38.5% were on drug therapy alone. Average maximum LDL-C reduction was 67±17.2% (from 535 mg/dl to 169.2 mg/dl) and only 14.3% of ARH patients reached the LDL-C goal of <100 mg/dl. The cumulative incidence of major CV outcomes were: CV death: 9.7%, coronary heart disease: 27.7%, severe aortic stenosis: 40%, peripheral artery disease: 20%.
Conclusions. Despite standard care including LA, LDL-C levels remain high and residual CV risk significant. These data confirm that an effective treatment for ARH is still lacking.

IS FATTY LIVER AN INDEPENDENT RISK FACTOR FOR SUBCLINICAL ATHEROSCLEROSIS?

A. Di Costanzo*[1], L. D'Erasmo*[1], L. Polimeni[1], L. Loffredo[1], P. Coletta[1], M. Del Ben[1], F. Angelico[2], A. Montali[1], G. Girelli[3], B. De Masi[3], M. Maranghi[1], M. Arca[1]
*[1]Department of Internal Medicine and Medical Specialties, Sapienza University, Rome; [2]Department of Public Health and Infectious Disease, Sapienza University, Rome; [3]Department of Immunohematology and Transfusion Medicine Unit, Policlinico Umberto I, Rome *Equal contribution*

Background. Non-Alcoholic Fatty Liver Disease (NAFLD) has been reported to be a risk factor for atherosclerosis. However, this association might be biased by the high prevalence of pro-atherogenic metabolic abnormalities in NAFLD. It has been reported that carriers of the M148M variant in the patatin like phospholipase 3 (PNPLA3) gene develop NAFLD without metabolic abnormalities.
Aims and methods. To clarify whether fatty liver itself promotes vascular damage, we compared subclinical atherosclerosis in 3 groups:
1) subjects with NAFLD due to metabolic syndrome, but not carrying the PNPLA3 variant (metabolic NAFLD group) (n=100);
2) subjects with NAFLD due to the PNPLA3 M148M (genetic NAFLD) (n=35);
3) normal controls (n 58). Fatty liver was demonstrated by ultrasounds.

Carotid intima-media thickness (CIMT) was measured as marker of subclinical atherosclerosis. Comparisons were adjusted for age, gender, smoke and grade of fatty liver by GLM.
Results. Age and gender were not different among groups. Subjects with metabolic NAFLD had significantly higher values of anthropometric variables, lipids, glucose and transaminases as compared to those with genetic NAFLD and controls ($p<0.05$). Overall, CIMT of subjects with metabolic NAFLD (0,84±0,17 mm) was significantly higher than that of subjects with genetic NAFLD (0,72±0,19 mm), which in turn was similar to controls (0,73±0,14 mm) [adjusted $P<0.001$]. These differences persisted even after adjustments for age, sex, smoke and grade of fatty liver ($P=0.003$).
Conclusions. We showed that subjects with metabolic NAFLD have increased subclinical vascular damage as compared to those with genetic NAFLD, thus suggesting that fatty liver per se might not be a risk factor for atherosclerosis.

AGE-RELATED CHANGES OF ULTRASOUND VASCULAR BIOMARKERS IN HEALTHY MICE

N. Di Lascio[1,2], C. Kusmic[2], F. Stea[2,3], F. Lenzarini[2], F. Faita[2]
[1]Institute of Life Science, Scuola Superiore Sant'Anna; [2]Institute of Clinical Physiology, National Research Council; [3]Department of Clinical and Experimental Medicine, University of Pisa

Ultrasound (US) can provide arterial stiffness parameters and hemodynamic indexes of cardio-vascular interaction characterizing the vascular aging process, associated with an increased cardiovascular risk. Aim of this study was to characterize the vascular aging in mice investigating changes in US-derived arterial stiffness biomarkers and Wave Intensity Analysis (WIA) indexes of abdominal aorta and carotid artery. Nine young (2 mo), eleven adult (5.5 mo) and nine old (15.5 mo) wild type male mice were imaged with high-resolution US (Vevo 2100). Relative distension, pulse wave velocity and distensibility were obtained for abdominal aorta and carotid (relDabd and relDcar, PWVabd and PWVcar, Dsabd and Dscar) using the diameter-velocity loop. Wave intensity (WI) signals were achieved performing WIA; first and second positive peaks intensities and negative part area were calculated (W1_abd and W1_car, W2_abd and W2_abd, NAabd and NAcar).
relDabd (young: 24.4±3.9%, adult: 22.1±6%, old: 15.5±3.5%) and relDcar (young: 29.4±6.6%, adult: 21.5±6.7%, old: 19±3.7%) were different among age groups ($p<0.01$ for both). Same results were found for PWV (PWVabd: young: 1.69±0.45 m/s, adult: 1.89±0.42 m/s, old: 2.62±0.66 m/s, $p<0.05$; PWVcar: young: 1.15±0.29 m/s, adult: 1.41±0.32 m/s, old: 1.66±1.09 m/s, $p<0.01$) and Ds (Dsabd: young:

0.39±0.21 kPa-1, adult: 0.30±0.14 kPa-1, old: 0.17±0.10 kPa-1, p<0.05; Dscar: young: 0.88±0.61 kPa-1, adult: 0.53±0.24 kPa-1, old: 0.20±0.16 kPa-1, p<0.01). W1 abd was lower in older animals (young: 10.7±4.2 10-7 m^2/s, adult: 6.87±5.08 10-7m^2/s, old: 5.38±2.62 10-7 m^2/s, p<0.05) while NAcar was higher in younger mice (young: 6.24±3.38 10-10 m^2, adult: 3.66±2.62 10-10 m^2, old: 3.14±1.10 10-10 m^2, p=0.052). Arterial stiffness biomarkers showed that both the abdominal aorta and the carotid artery were affected by the aging process; in addition, aging is associated with a decrease in the amplitude of the first peak of the abdominal aorta WI signal and in the negative area of the carotid WI waveforms. This US-based approach can provide stiffness biomarkers and hemodynamic indexes of cardio-vascular interaction and is suitable for studies with mouse models of cardiovascular disease.

IMPACT OF SEASONALITY AND AIR POLLUTANTS ON CAROTID-FEMORAL PULSE WAVE VELOCITY IN HYPERTENSIVE PATIENTS

M. Di Pilla[1], R.M. Bruno[1], L. Ghiadoni[1], F. Stea[2], L. Massetti[3], I. Bertolozzi[3], S. Taddei[1], P.A. Modesti[3]
[1]Department of Clinical and Experimental Medicine, University of Pisa; [2]Institute of Clinical Physiology, CNR, Pisa; [3]Department of Experimental and Clinical Medicine, University of Florence

Objective. The effects of seasonality on blood pressure (BP) and cardiovascular events are well established. However, the influence of seasonality and other environmental factors on arterial stiffness and wave reflection, key parameters for cardiovascular risk stratification in hypertensive patients, have never been analyzed. This cross-sectional study aimed at investigating whether seasonality (daily number of hours of light) and acute variations in outdoor temperature and air pollutants may affect carotid-femoral pulse wave velocity (PWV) and wave reflection, measured as Augmentation Index (AIx) and Augmentation Pressure (AP).
Design and method. 731 hypertensive patients (30-88 years), 417 treated with antihypertensive drugs (241 men) seeking medical consultation in our outpatient clinics during a 5 year period (2006-2011) were enrolled. PWV, central BP, AIx and AP were measured noninvasively in a quiet, temperature-controlled (22-24°C) room. Data of the local office of the National Climatic Data Observatory were used to estimate meteorological conditions and air pollutants (PM10, O3, CO, N2O) exposure the same day of the vascular examination.
Results. PWV (mean value 8.5±1.8 m/s) was related to age (r=0.467, p<0.001), body mass index (BMI) (r=0.132, p<0.001), central systolic (r=0.414, p<0.001) and diastolic BP (r=0.093, p=0.013), daylight hours (r=-0.176, p<0.001), mean outdoor temperature (r=-0.082, p=0.027), O3 (r=-0.135, p<0.001), CO (r=0.096, p=0.012), N2O (r=0.087, p=0.022) in the univariate analysis. Moreover PWV was higher in individuals with chronic kidney disease, impaired fasting glucose and hypertriglyceridemia. In multiple linear regression analysis, adjusted for confounders, PWV remained independently associated only with daylight hours (β=-0.170; 95% CI: -0.273 to -0.067, p=0.001). No significant correlation was found between wave reflection parameters and daylight hours, mean temperature and air pollutants.
Conclusions. Seasonality, expressed by the daily number of hours of light, independently affects PWV but does not influence either AP or AIx75. Thus, seasonality should be taken into account when assessing arterial stiffness for cardiovascular risk stratification or in clinical trials.

MACROPHAGES DIFFERENTATED IN VITRO ARE HETEROGENEOUS: MORPHOLOGICAL AND FUNCTIONAL PROFILE IN PATIENTS WITH CORONARY DISEASE

S. Fiorelli, S. Eligini, N. Cosentino, G. Marenzi, E. Tremoli
Centro Cardiologico Monzino IRCCS, Milano

Aim. Monocytes and tissue macrophages, cells involved in the inflammatory process, play a crucial role at any stage of coronary artery disease (CAD). Macrophages are hallmarked by morpho/phenotypic heterogeneity described also in atherosclerotic plaque where the presence of a particular macrophage phenotype may have harmful or beneficial functions on CAD development. Tissue macrophages are not easily obtained and monocyte-derived macrophages (MDMs) are accepted as a good surrogate. We previously reported that in healthy subjects MDMs spontaneously differentiated in vitro show two dominant morphotypes, spindle and round, with pro- and anti-inflammatory proprieties respectively. This study is conceived to delineate the morphological and functional profile of MDMs obtained from CAD patients compared to those of healthy subjects.
Methods. Monocytes were isolated from venous blood of healthy subjects and from CAD patients and differentiated for 7 days in medium supplemented with 10% autologous serum. The uptake of apoptotic Jurkat T cells, for efferocytosis assay, was detected by flow cytometry. Transglutaminase 2 (TG2) and tissue factor (TF) were determined by immunofluorescence and western blotting. Thrombin generation was evaluate using a thrombinoscope.
Results. Morphologically, MDMs of CAD patients show a prevalence of round morphotype respect to spindle. Nevertheless, these MDMs displayed less efferocytic capacity compared to control. Impaired efferocytosis may be due to the reduces levels of TG2, protein involved in phagosome formation. Moreover, CAD MDMs present higher TF levels that are associated with a quickly thrombin generation.
Conclusions. MDMs of CAD patients show a pro-inflammatory and a pro-thrombotic profile characterized by reduced efferocytic capacity and increased of TF levels. MDMs of CAD patients can contribute to plaque progression and activation besides that to thrombus formation. Drug handling of different macrophage phenotypes may provide a basis for new therapeutic strategies able to limit the progression of atherosclerosis.

ASSOCIATION BETWEEN PLASMA LIPOPROTEIN(A) LEVELS AND LONG-TERM MORTALITY: DATA FROM THE BRISIGHELLA HEART STUDY

F. Fogacci, S. D'Addato, A.F.G. Cicero, E. Bertagnin, L. D'Agostini, S. Palmisano, E. Grandi, M. Giovannini, M. Rosticci, C. Borghi
Alma Mater Studiorum University of Bologna, Department of Medical and Surgical Sciences, Bologna

Aim. Our aim was to assess whether serum Lp(a) levels can significantly influence long-term survival in subjects with an equal general cardiovascular risk profile.
Patients and methods. This study prospectively involved a Brisighella Heart Study cohort sample of 1172 adult subjects (M:573; F:599; aged 40-69) who had no cardiovascular diseases at enrollment. According to the CUORE project risk-charts (Italian-specific risk-charts), individuals were stratified into a low- (n=865), an intermediate - (n=232) and a high - (n=75) cardiovascular-risk groups by age,

gender, diabetes, smoking-status, total cholesterol and systolic blood pressure. Kaplan-Meier 25 year survival analysis was carried out examining each class of risk. The log-rank statistic was used to estimate the survival of subjects with and without elevated serum Lp(a) levels compared to the median of the population. Survival functions were age-adjusted considering the subjects' starting age (if it was higher or lower than the middle study-population's age at enrollment, which was 56). Finally, for each cardiovascular-risk group, we constructed a ROC curve using plasma Lp(a) levels as test-variable and death as state-variable. In this way we evaluated whether serum Lp(a) concentration was an independent long-term mortality prognosticator.
Results. In the low-cardiovascular-risk group, no significantly difference was observed in the 25-year survival for what concerns increased serum Lp(a) levels (compared to the median of the population, which was 18 mg/dl). According to the Mantel-Cox test, subjects at intermediate-cardiovascular-risk, aged 56-69 and with elevated serum Lp(a) levels, showed a significantly higher survival-time-estimate rather than the people with the same age but lower serum Lp(a) levels (15,1±0,8 vs 12,8±1 years, P=0,01). However, in the first group the mortality-rate was 1,5 times greater compared to the second one (60,2% vs 39,8%). In the high-cardiovascular-risk group, since we had only older subjects than the average of the population we were prevented from making any statement. Furthermore, in this group, dosing serum Lp(a) appeared a mildly accurate, though predictive, test of long-term mortality (AUC=0,63, CI[0,50-0,76], P=0,05, with 17,5 mg/dl best cut-off value), losing any kind of predictive power in subjects at low or intermediate cardiovascular risk.
Conclusion. A tight control of modifiable cardiovascular-risk factors is advisable in subjects with high serum Lp(a) levels.

ASSOCIATION BETWEEN URIC ACID AND RENAL FUNCTION IN HYPERTENSIVE PATIENTS: WHICH ROLE FOR SYSTEMIC VASCULAR INVOLVEMENT?

G. Geraci, G. Mulè, M. Morreale, D. Altieri, A. Castiglia, F. Gervasi, F. Zotta, C. Geraci, S. Cottone
Dipartimento Biomedico di Medicina Interna e Specialistica (DIBIMIS), Unit of Nephrology and Hypertension. European Society of Hypertension Excellence Center, University of Palermo

Aim. The role of systemic vascular involvement in mediating the association between serum uric acid (SUA) and renal function in hypertension and chronic kidney disease (CKD) has not been fully explored. The purposes of our study were:
1) to investigate the relationship between SUA with both carotid intima-media thickness (cIMT) and aortic pulse wave velocity (aPWV) in hypertensive subjects;
2) to assess the influence of renal function on these relationships;
3) to study whether systemic vascular changes may mediate the association between SUA and renal function in this population.

Methods. We enrolled 523 hypertensive subjects with (n=263) or without CKD (n=260). The study was also conducted in the population divided into tertiles of SUA based on sex-specific cut-off values. cIMT was assessed by Duplex-Doppler ultrasonography and aPWV through oscillometric device.
Results. cIMT and aPWV were higher, and eGFR lower, in uppermost SUA-tertile patients when compared to those in the lowest ones (all p<0.001).
Uricemia strongly correlated with eGFR, cIMT and aPWV at univariate analysis (p<0.001) in all subjects, and with eGFR and cIMT after adjustment for confounders (p<0.001). The relationships between SUA and cIMT were significant in both patients with or without CKD, in absence of significant differences between groups. Moreover, the adjustment of SUA for cIMT attenuated the relationship between SUA and eGFR (p=0.019).
Conclusion. Systemic vascular changes related with SUA seem in part to mediate the association between SUA and renal function in hypertensive patients, regardless of kidney function.

ARTERIAL TONOMETRY AND CAROTID ATHEROSCLEROSIS: INFLUENCE OF SIDE SELECTION AND CAROTID ATHEROSCLEROTIC PLAQUES IN CENTRAL BLOOD PRESSURE AND PULSE WAVE VELOCITY MEASUREMENT

A. Grillo[1], G. Simon[3], M. Rovina[3], R. Carretta[3], P. Salvi[2], G. Parati[1,2]
[1]Department of Health Sciences, University of Milano-Bicocca, Milan; [2]Department of Cardiovascular Neural and Metabolic Sciences, San Luca Hospital, Istituto Auxologico Italiano, Milan; [3]Department of Medical, Surgical and Health Sciences, University of Trieste

Aim. To assess whether central blood pressure (CBP) and carotid-femoral pulse wave velocity (PWV) measurements by applanation tonometry are influenced by the presence of stenosing atherosclerotic carotid plaques in the site of measurements or by the side of measurement.
Methods. Twenty-nine healthy young subjects (CTRL group) and ninety-two patients affected by carotid atherosclerosis were recruited. These were divided into 3 groups according to carotid ultrasound findings (Toshiba Aplio XG): 27 patients had increased in intimal media thickness bilaterally (S group), 30 patients had bilateral stenosis (stenosis equal or superior to 30%) (SP group), 26 patients had unilateral stenosis (difference in stenosing grade equal or superior to 40% between the two carotid) (A group). CBP and PWV were measured by applanation tonometry (PulsePen, DiaTecne), on both common carotid artery.
Results. No significant differences in CBP (mmHg, mean value ± standard deviation m/s;) were found between the two side of measurements for all groups, both for systolic: (CTRL: right 110±15, left 110±15 P=ns). (S: right 140±26, left 138±23 P=ns). (SP: right 141±26, left 140±24 P=ns). (A: right 143± 28, left 142±28 P=ns), and diastolic CBP: (CTRL: right 67± 8, left 67±7 P=ns). (S: right 78±10, left 80±10 P=ns). (SP: right 76±7, left 76±8 P=ns). (A: right 77±11, left 78±11 P=ns). Neither significant differences in c-f PWV were found, between the two side of measurements (m/s, mean value±standard deviation): (CTRL: right 5,55±1,12 P=ns, left 5,65±1,04 P=ns). (S: right 11,27±5,26 P=ns, left 9,98±3,83 P=ns). (SP: right 11,99±4,32 P=ns, left 11,98±4,75 P=ns). (A: right 12,58±4,65 P=ns, left 12,59±4,96 P=ns).
Conclusions. The side (right vs left) of the measurements, neither the presence of carotid atherosclerosis affect CBP and PWV measurement, performed by applanation tonometry.

HELPER-DEPENDENT ADENOVIRUS-MEDIATED GENE TRANSFER OF AN LDL RECEPTOR/TRANSFERRIN CHIMERIC PROTEIN REDUCES AORTIC ATHEROSCLEROSIS IN LDL RECEPTOR-DEFICIENT MICE

E. Leggiero[1], G. Labruna[2], M. Esposito[1,3], D. Savoia[1,3], L. Sacchetti[1], L. Pastore[1,3]

[1]CEINGE-Biotecnologie Avanzate, Napoli; [2]Fondazione IRCCS SDN, Istituto di Ricerca Diagnostica e Nucleare, Napoli; [3]Dipartimento di Medicina Molecolare e Biotecnologie Mediche, Università di Napoli 'Federico II', Napoli

Aim. FH is a well-characterized hyperlipidemia due in most of the cases to mutations in the LDLR gene and characterized by elevated concentration of plasma LDL-C. Statins can lower cholesterol levels but are not effective in all patients. In the past, we have developed safe and effective gene-therapy strategies for hepatocytes transduction and consequent expression of anti-atherogenic proteins using PEGylated helperdependent adenoviral (HD-Ad) vectors. We have recently devised a therapeutic strategy for reducing LDL using a secreted protein that can potentially be expressed in non-hepatic tissues used as bioreactors. At this aim, we developed an HD-Ad vector for the expression of the soluble form of the extracellular portion of the human LDLR fused with transferrin (LDLR/Tf).
Methods and results. We evaluated the efficacy of LDLR/Tf in cellular models such as CHOldla7 in which we restored the cell ability to uptake of labeled LDL; we administered intravenously 1X10E11 vp/kg of the HD-Ad vector expressing LDLR/Tf in LDLR-deficient mice and demonstrated the efficacy of the vector in reducing total and LDL cholesterol levels; in addition, expression of LDLR/Tf significantly reduced aortic atherosclerotic lesions in treated LDLR-deficient mice compared to controls. We demonstrated the efficacy of serum secretion of LDLR/Tf in reducing aortic atherosclerosis in FH mice.
Conclusions. These results will allow the evaluation of HD-Ad vector-mediated expression of LDLR/Tf in non-hepatic tissues using alternative routes of administration in order to develop safer gene transfer protocol more compatible with clinical applications.

EXPRESSION AND CHANGE IN MIRS 145, 221 AND 222 EXPRESSION IN HYPERTENSIVE SUBJECTS TREATED WITH ENALAPRIL, LOSARTAN OR OLMESARTAN

G. Mandraffino[1], C.O. Aragona[1], V. Cairo[1], F. Mamone[1], A. D'Ascola[2], A. Alibrandi[3], M.A. Sardo[1], A. Lo Gullo[1], E. Imbalzano[1], A. Saitta[1]
[1]Department of Clinical and Experimental Medicine, Internal Medicine; [2]Department of Biochemical, Physiological and Nutritional Sciences; [3]Department of Statistics, University of Messina

Aim. We evaluated whether the anti-hypertensive drugs enalapril, losartan or olmesartan may have effects on monocyte miRs 145, 221 and 222 in hypertensives without organ involvement.
Methods. Sixty-four essential hypertensives without additional risk factor for CVD and 42 controls were included; we evaluated blood pressure (SBP/DBP), lipid profile, glucose, C-reactive protein (CRP), fibrinogen, arterial stiffness (AS) indices (pulse way velocity: PWV; augmentation index: AIx) and carotid intima-media thickness (cIMT) at baseline (T0) and after 24 weeks treatment (T1). Subjects with plasma levels of LDL-C ≥160 mg/dl, TG ≥200 mg/dl, BMI ≥30, alcohol consumption, a personal or familial history of CVD, T2DM, or thyroid, liver or kidney diseases were excluded. Patients were randomly assigned to receive once a day enalapril 20 mg, losartan 100 mg or olmesartan 20 mg.
Results. At T1, we found a significant improvement of both SBP and DBP (SBP: -19.03%, p<0.001; DBP: -14.41%, p<0.001), lipid profile (HDL-C: +4.4%, and LDL-C: -6.4%; both p<0.001), glucose (-2.5%, p<0.001), BMI (-3.1%, p<0.001), fibrinogen (-6.2%, p<0.001), CRP (-9.2%, p<0.005), AS indices (AIx: -19.1%; PWV: -14.4%; both p<0.001), and monocyte miR expression (miR221: -29.8%; miR222: -39.7%; miR145: +41.1%; all p<0.001). We have analyzed separately each arm of treatment: Olmesartan appeared the most effective in reducing CRP levels (-9.48%), and miRs221/222 (-32.9% and -42.4%, respectively); losartan reduced PWV (-37.6%) and improved HDL-C levels (+7.9%) and miR145 (+51.5%) more than olmesartan and enalapril; enalapril appeared more effective on fibrinogen reduction (-9%); no differences were found as regards BMI, glucose, LDL-C, SBP, DBP, AIx, and cIMT.
Conclusion. Enalapril, losartan and olmesartan are effective in improving mechanical and humoral factors associated to AS and atherogenesis; these drugs restored in untreated hypertensives the deregulated connection between miRs221/222 and miR145, thus contributing to slow the progression of vascular damage already shown in the clinical studies.

PCSK9 IMPROVES THE MIGRATION AND PROLIFERATION OF SMOOTH MUSCLE CELLS IN RESPONSE TO PDGF-BB

S. Marchianò[1], G. Tibolla[1], A. Corsini[1,2], A.L. Catapano[1,2], N. Ferri[3]
[1]Dipartimento di Scienze Farmacologiche e Biomolecolari, Università degli Studi di Milano; [2]Multimedica IRCCS, Milano; [3]Università degli Studi di Padova

Proprotein Convertase Subtilisin/Kexin type 9 (PCSK9) is an important regulator of hepatic low-density lipoprotein (LDL)-cholesterol levels. We have previously shown that PCSK9 is expressed in cultured smooth muscle cells (SMCs) and it is detectable in human carotid atherosclerotic plaques. In the present study, we investigated the role of PCSK9 on proliferation, migration and dedifferentiation of mouse SMCs, pivotal features of atherosclerosis and restenosis processes. Freshly isolated PCSK9-/- mSMCs showed higher levels of the contractile marker smooth muscle α-actin (α-SMA; 2.24±0.36 fold; P<0.01) and myosin heavy chain II (MHC-II; 8.65±1.55 fold; P<0.01), otherwise decreased levels of synthetic markers l-caldesmon (-54±14%; P<0.01) and Col1a1 mRNA levels (-31±11%; P<0.05) compared to PCSK9+/+ cells. The absence of PCSK9 also affected the proliferation rate and capability, as determined by cell counting, iCelligene system (doubling time 106.3±4.5 h vs 57.3±2.1 h) and flow-cytometric analysis. The mSMC PCSK9-/- displayed a transition to the S phase, after the stimulation with Platelet-Derived Growth Factor (PDGF-BB), significantly less pronounced compared to mSMC PCSK9+/+. The reconstitution of PCSK9 expression, by retroviral infection of PCSK9-/- mSMCs, led to a downregulation of contractile markers (α-SMA) and induction of synthetic markers (l-caldesmon and Col1a1). Proliferation rate of mSMCs PCSK9-/- was significantly lower compared to PCSK9 reconstituted cells (doubling time 41.2±1.9 h v. 32.2±3.1 h). Finally, morphological changes in response to PDGF-BB of PCSK9-/- mSMCs, measured by iCelligence monitoring and cytoskeletal staining, was significantly impaired compared to PCSK9+/+ cells. These differences were associated with lower activation of Rac1 small G protein and reduced PDGF receptor β expression. Taken together, the present results suggest that the absence of PCSK9 could have a protective role in the vascular restenosis process, potentially through an impairment of PDGF receptor signaling.
The present work was supported by the grant 2012-0549 from Fondazione Cariplo, Italy.

INTERACTION OF LCAT AND APOA-I ON LIPID METABOLISM IN HUMAN LCAT X HUMAN APOA-I DOUBLE TRANSGENIC MICE

A. Ossoli[1], B. Vaisman[2], S. Gordon[2], M. Amar[2], L. Calabresi[1], A.T. Remaley[2]
[1]Center E. Grossi Paoletti, Department of Pharmacological and Biomolecular Sciences, Università degli Studi di Milano; [2]Lipoprotein Metabolism Section, National Heart, Lung and Blood Institute National Institutes of Health, Bethesda, MD, United States

Aim. Lecithin: cholesterol acyltransferase (LCAT) is a key enzyme that catalyzes the esterification of free cholesterol in plasma high-density lipoproteins (HDL) but its role in atherosclerosis has not been clearly established.
Methods. We recently developed a novel mouse model of human LCAT x human apoA-I double transgenic mice and compared lipid metabolism in these mice with normal C57Bl/6N, hLCAT transgenic and apoE-KO mice.
Results. Double transgenic mice on normal chow diet had highest level of plasma total cholesterol, which was significantly higher than in apoE-KO animals and reached more than 1000 mg/dL. FPLC analysis showed that in contrast with apoE-KO mice in double transgenic mice most of lipids were located in HDL fraction. Despite their higher total cholesterol than apoE-KO mice LCAT x apoA-I transgenic mice were protected against development of atherosclerosis. Expression analysis of the different tissues of the mice showed that in double transgenic mice had induction of genes involved in the reverse cholesterol transport (RCT) pathway. Genes related to inflammation were suppressed in the aorta of double transgenic mice compared to control mice and apoE-KO mice on HFHC diet.
Conclusion. The obtained results marked significance of HDL and efficient RCT for atheroprotection even in situation of very high total plasma cholesterol. The interaction between LCAT and ApoA-I is able radically enhance RCT and HDL. These data can be helpful in development of new prospective treatments of atherosclerosis and acute coronary syndrome with combination of LCAT and ApoAI or ApoA-I mimetic peptides.

METABOLIC AND CATHECOLAMINERGIC SYSTEM CHANGES DURING DRUG-INDUCED SLEEP ENDOSCOPY IN PATIENTS WITH OBSTRUCTIVE SLEEP APNOEA

M.F. Pengo, V. Bisogni, E. Casiraghi, G. Ceolotto, G. Maiolino, G.P. Rossi
Department of Medicine (DIMED), University of Padua

Obstructive sleep apnea syndrome (OSAS) is a well established risk factor for hypertension. It is associated with an enhanced sympathetic activation and with increased catecholamine levels. Catechol-O-methyltransferase (COMT) is one of the major mammalian enzymes involved in the metabolic degradation of catecholamines.
We hypothesised that COMT activity, measured during drug induced sleep endoscopy (DISE), is altered during intermittent hypoxia in patients with OSAS.
We enrolled 15 patients, 6 of which met all inclusion criteria (diagnosis of OSAS by means of polysomnography with an AHI >5 events/hour), and underwent medical assessment, blood tests and ambulatory blood pressure monitoring. All 6 patients underwent DISE during which 7 blood samples were drown for the analysis of COMT activity and catecholamines concentration before, during and after an obstructive event.
The enrolled patients were 5 males and one female, aged 48 years, body mass index was 27.71 (3.42) kg/m^2. All patients had hypertension either treated or untreated.
COMT analysis showed an increase of the enzyme activity after propofol administration, which was more marked when propofol-induced central apnea occurred compared to baseline (181.2 (12.8) pmol/min/mg vs 99.4 (11.1) pmol/min/mg, $p<0.05$).
A less marked increase in COMT activity was observed during the obstructive apnea and COMT activity did not seem to be influenced by propofol when red blood cells were incubated with the anaesthetic.
In this preliminary analysis we showed that the catecholaminergic system is stimulated during DISE after apneic events. This might be due to an altered activity of COMT during hypoxia therefore suggesting a novel pathophysiological pathway responsible of hypertension in patients with OSAS.

EFFECTS OF BIFIDOBACTERIUM BIFIDUM PRL2010 ON LIPID PROFILE OF MICE AND IN VITRO INVESTIGATIONS OF CHOLESTEROL LOWERING CAPACITY

A. Piemontese[1], F. Turroni[2], F. Bernini[1], L. Elviri[1], M. Ventura[3], I. Zanotti[1]
[1]Department of Pharmacy, University of Parma; [2]Alimentary Pharmabiotic Centre, University of Ireland, Cork; [3]Laboratory of Probiogenomics

Aim. In the recent years, the use of probiotics bacteria is proposed as a new approach to decrease plasma cholesterol levels. Probiotics are live micro-organisms which, when administered in adequate amounts, confer a health benefit on the host. We screened several gut bifidobacterial strains in vitro, in order to find strains with high cholesterol uptake. Furthermore, we characterized the mechanism of cholesterol uptake and the lipid metabolism of a strain with significant uptake capacity. Then we evaluated the hypolipidemic effects in animal model after probiotic administration.
Methods. We detected the cholesterol uptake of 17 Bifidobacteria strains, after 3h incubation with 3H-cholesterol into bacterial cultures. The cholesterol uptake mechanism was analysed, incubating alive and thermally inactivated bacteria with 3H-cholesterol for 28 h. The quantification of free fatty acids, cholesterol, coprostanol after 3 h of incubation with cholesterol in medium was performed by liquid chromatography-mass spectrometry. Afterwards we analysed the change of lipid profile in ApoE-/- mice after daily administration of 109 cells of PRL2010 for 6 weeks.
Results. A significant uptake capacity was observed for BbPRL2010. The values of 3H-cholesterol found in alive bacteria were between 0,101 µCi (t=3 h) and 0,131 µCi (t=28 h), whereas those one of thermic inactivated were between 0,066 µCi (t=3 h) and 0,093 µCi (t=28 h). Upon bacterial exposition with cholesterol, the coprostanol, a key metabolite of cholesterol, was increased (~60%), as well as all saturated and unsaturated fatty, compared with untreated samples. In addition, in the ApoE-/- mice, we observed a decrease of total cholesterol (-27%) in the treated group.
Discussion. A formulation containing the Bifidobacterium bifidum PRL2010 could be a potential nutraceutical tool to modify the cholesterol levels in patients with mild or moderate hyperlipidemia.

HIGH DENSITY LIPOPROTEIN (HDL)-ASSOCIATED SPHINGOSINE 1-PHOSPHATE (S1P) INHIBITS MACROPHAGE APOPTOSIS BY STIMULATING SIGNAL TRANSDUCER AND ACTIVATOR OF TRANSCRIPTION 3 (STAT3) ACTIVITY AND SURVIVIN EXPRESSION

F. Poti[1,2], R. Feuerborn[3], M. Brodde[4], H. Schmidt[3], J.-R. Nofer[3]
[1]Department of Neurosciences - Unit of Pharmacology, University of Modena Parma, Parma (Italy); [2]Azienda USL of Modena; [3]Center for Laboratory Medicine, University Hospital Münster, Münster (Germany); [4]Experimental and Clinical Haemostasis, Department of Anaesthesiology and Intensive Care, University Hospital Münster, Münster (Germany)

Background and Aims. Macrophage apoptosis is a critical process involved in atherosclerosis. High density lipoprotein (HDL) carries biologically active lipids such as sphingosine-1-phosphate (S1P) that may contribute to its atheroprotective capacity. We here examined the effect of S1P and HDL on the apoptosis of RAW264.7 murine macrophages.
Methods and results. Mitochondrial or endoplasmic reticulum-dependent apoptosis was induced by exposure of RAW264.7 cells to etoposide or thapsigargin, respectively. Cell death induced by each of these compounds was inhibited by S1P as assessed by annexin V binding and TUNEL staining. In addition, activation of both terminal caspase 3 and upstream caspases 9 and 12 were inhibited in RAW264.7 cells pretreated with S1P. S1P induced expression of the inhibitor of apoptosis protein (IAP) family proteins cIAP1, cIAP2 and survivin, which inhibit caspase activity, but only the suppressant of survivin expression YM155 and not the cIAP1/2 blocker GDC0152 reversed the inhibitory effect of S1P on macrophage apoptosis. We further observed that S1P activated signal transducer and activator of transcription 3 (STAT3) and Janus kinase 2 (JAK2) and the stimulatory effect of S1P on survivin expression and its inhibitory effect on apoptosis were attenuated by STAT3 or JAK2 inhibitors, S3I-201 or AG 490, respectively. The effects of S1P on STAT3 activation, survivin expression and apoptosis were emulated by HDL and HDL lipids, but not by HDL deprived of S1P by incubation with charcoal. In addition, JTE013 and CAY10444, S1P receptor 2 and 3 antagonists, respectively, compromised the capacity of S1P and HDL to stimulate STAT3 activation and survivin expression, and to inhibit apoptosis.
Conclusions. HDL-associated S1P inhibits macrophage apoptosis by stimulating STAT3 activity and survivin expression. The suppression of macrophage apoptosis may represent a novel mechanism utilized by HDL to exert its anti-atherogenic potential.

LDL APHERESIS IMPROVES CORONARY FLOW RESERVE ON THE LEFT ANTERIOR DESCENDING ARTERY IN PATIENTS WITH FAMILIAL HYPERCHOLESTEROLEMIA AND CHRONIC ISCHEMIC HEART DISEASE

F. Sbrana, E.M. Pasanisi, F. Bigazzi, C. Petersen, M. Coceani, B. Dal Pino, A. Ripoli, M. Pianelli, R. Luciani, T. Sampietro
Fondazione Toscana Gabriele Monasterio, Pisa

Aim. LDL apheresis (LA) influences the microcirculation, endothelial function and cardiovascular homeostasis. The aim of our study was to analyse temporal variations of coronary flow reserve (CFR) on the left anterior descending artery obtained during dipyridamole stress echocardiograpy in patients receiving LA for severe familial hypercholesterolemia and in a control group of hypercholesterolemic patients with coronary heart disease.
Methods. The LDL apheresis group consisted in 10 patients (mean age 65±7 years, male 70%) with Familial Hypercholesterolemia (6/10) or Familial Combined Hypercholesterolemia (4/10) and chronic ischemic heart disease on maximally tolerated lipid lowering therapy and chronic LA treatment (median 7 years, interquartile range 6-14 years). Associated hyperlipoproteinemia (a) was present in 6/10 subjects. LA was performed biweekly by dextran-sulfate or heparin-induced LDL precipitation technique. Coronary heart disease was diagnosed at a mean age of 44±8 years. No relevant comorbidities were present.
The control group was matched for age, sex and follow-up period. CFR was calculated as the ratio between diastolic velocity sampled at peak stress with dipyridamole and baseline diastolic velocity (normal value >2,0).
Results. During a median follow-up of 27 months (interquartile range 23-50 months), a significant increase in CFR (from 1.86 ±0.47 to 2.25±0.35; p<0.001) was observed following LA. During this period, no patients modified their anti-ischemic therapy and no cardiovascular events were reported. In the control group, during the study time (24 months - interquartile range 14-57 months) no significant variation in CRF vas observed (from 2.08±0.39 to 1.92±0.26; p=0.283).
Conclusion. CFR measured at dipyridamole stress echocardiography is a reliable marker of therapeutic efficacy in patients with severe familial hypercholesterolemia on LA. The observed increase in CFR points to an improvement in myocardial perfusion which represents a primary target of LA.

POSTER MODERATI

NUTRACEUTICAL MODULATION OF HDL FUNCTION

M.P. Adorni[1], F. Zimetti[1], V. Trimarco[2], R. Izzo[3], F. Rozza[4], M.V. Manzi[3], F. Bernini[1]
[1]Department of Pharmacy, University of Parma; [2]Hypertension Research Center & Department of Neurosciences, Federico II University, Napoli; [3]Hypertension Research Center & Department of Translational Medical Sciences, Federico II University; [4]Department of Advanced Biomedical Sciences, Federico II University, Napoli

Cholesterol efflux capacity (CEC) is the main cardioprotective HDL function, representing their ability to accept excess cholesterol from macrophages and it has been inversely related to the incidence of cardiovascular events, independently of HDL plasma levels. Cholesterol efflux to HDL mainly occurs through the transporters Scavenger Receptor class B type I (SR-BI), ATP Binding Cassette A1 (ABCA1) and G1 (ABCG1). Each transporter recognizes specific HDL subclasses. Several nutraceuticals (NUTRA) have been shown to modulate HDL CEC. For example HDL CEC significantly improved after polyphenol-rich intervention or walnuts assumption, a rich source of α-linolenic acid, γ-tocopherol and phytosterols. Also anthocyanin supplementation improved HDL CEC in subjects with hypercholesterolemia.
Aim. Our study aimed to evaluate the effect of a NUTRA combina-

tion of Berberine, Red yeast rice powder and Morus Alba on HDL CEC and on serum cholesterol loading capacity (CLC), an index of the overall serum ability to load macrophages with cholesterol and thus to induce foam cell formation.
Methods. 9 healthy volunteers have been treated 4 weeks with the above NUTRA combination. HDL CEC was measured using specific cell-based radio isotopic assays. Serum CLC was measured fluorimetrically in human macrophages.
Results. The NUTRA combination significantly reduced total, LDL cholesterol (-14 % and -17%, p<0.01 and <0.05, respectively) and HDL cholesterol (-11.7%; p<0.01). The treatment significantly increased CEC through SR-BI and ABCG1 (+27% and +13%, respectively; p<0.05) and reduced serum CLC (-23.2%; p<0.05).
Conclusions. Despite the effect on HDL-C plasma level, treatment with the NUTRA combination improved CEC selectively trough SR-BI and ABCG1 pathways, suggesting a redistribution of HDL towards particles specific for these transporters. Both the improved HDL functionality and the decreased plasma LDL cholesterol levels may contribute to the reduction of the serum pro-atherogenic potential (CLC) observed after treatment.

ABNORMAL LEFT VENTRICULAR GLOBAL STRAIN DURING EXERCISE-TEST IN YOUNG HEALTHY SMOKERS

C.O. Aragona, G. Mandraffino, V. Cairo, S. Quartuccio, F. Mamone, M.A. Sardo, A. Saitta, E. Imbalzano
Department of Clinical and Experimental Medicine, University of Messina

Aim. Cigarette smoking (CS) may induce alterations in cardiovascular (CV) system at any age, although how precociously CS may impact on cardiac structure and function is unknown. Myocardial function may be explored by conventional echocardiography, but it allows to detect abnormalities only when a remodelling occurred. Strain imaging might overcome these limitations. In the present study we evaluated whether young smokers with normal echocardiographic pattern may present abnormal myocardial deformation at rest, or during physical effort.
Methods. Fifty young smokers without additional risk factors for atherosclerosis and 60 non-smokers underwent to a standardized exercise-test. The cardiovascular response to exercise was evaluated by standard echocardiography, and deepened by speckle-tracking analysis (2D-STE).
Results. No difference between smokers and controls as regards anthropometric and baseline characteristics, except for HDL-cholesterol (smokers lower, p<0.005), fibrinogen, C-reactive protein (CRP), interleukin-6 (IL-6) (smokers higher). By conventional echocardiography we found no difference, while a different behavior of global longitudinal strain (GLS) and twist during exercise-test was detected by 2D-STE. An association between CS, inflammation and GLS change during exercise was found.
Conclusions. GLS and twist behave differently in smokers and in controls during a standardized exercise-test. Regression analysis suggested that smoking duration, fibrinogen and HDL-C plasma levels should be taken into account to understand how GLS behaves differently, but not twist. Strain imaging is confirmed as a useful tool to identify early changes of cardiac dynamics, as delayed adaptation to physical effort; this finding may represent a very precocious functional abnormality, probably long before that structural damages occur.

RENAL DENERVATION RAPIDLY RESTORES CIRCULATING PROANGIOGENIC HEMATOPOIETIC CELLS IN PATIENTS AFFECTED BY DRUG-RESISTANT HYPERTENSION

C.O. Aragona, G. Mandraffino, V. Cairo, S. Quartuccio, Mamone, M.A. Sardo, Antonino Saitta, E. Imbalzano
Department of Clinical and Experimental Medicine, University of Messina

Aim. To investigate whether blood pressure (BP) lowering after renal sympathetic denervation (RSD) affects CD34+ cell number in drug-resistant hypertension (R-HTN)
Methods-We enrolled 11 patients with R-HTN, already treated with at least 6 antihypertensive drugs, including a diuretic, at full dosages; patients with office BP of >160 mmHg (>150 mmHg for type 2 diabetes) were considered eligible for the procedure. Adherence to drug treatment was accurately checked by patient's general practitioners.
Results. Mean age was 61±7.9 years; M:F 8:5. We measured clinic (sphygmomanometer) and ambulatory (Tonoport V GE-Healthcare) BP, and heart rate (HR; electrocardiogram), at baseline and 30 days after RSD procedure (Symplicity; Medtronic). 24 h BP recordings and home BP protocols were consulted in addition to office BP measurements at the hospital before enrollment. At T0: SBP: 179.1±9.3mmHg; DBP: 101.2±5.5 mmHg; HR 79.9±9.4; CD34+ cells: 1.66 ±0.51. At T1 SBP values were reduced on the average of 40.2 mmHg (138.9±7.3, Δ=-22.5%, p<0.001) DBP of 18 mmHg (83.2±3.2, Δ=-17.7%, p<0.001), and HR of 10.4 bpm (67.3±6.0, Δ=-17.7%, p<0.005), and CD34+cell number increased on an average of 0.34 cells/μL (2.0± 0.51, Δ=+21.2%, p<0.001).
Conclusion. To our knowledge, this is the first observation that RSD is able to rapidly restores CD34+cell number in patients affected by true R-HTN; if these results will be confirmed on a larger scale, they could provide new insights about CD34+ cells and pathophysiological aspects of arterial hypertension.

PCSK9 (PROPROTEIN CONVERTASE SUBTILISIN/KEXIN TYPE 9) AND GLUCOSE METABOLISM: WHICH CONNECTION?

G. Balzarotti[1], G. Tibolla[1], M.Ruscica[1], M. Stramba-Badiale[1], A.L. Catapano[1,2], G.D. Norata[1]
[1]Department of Pharmacological Sciences, University of Milan; [2]IRCCS MultiMedica Hospital, Milan

Objective. Hepatic low density lipoprotein (LDL) receptor is a key target of PCSK9, however, circulating and/or locally produced PCSK9 has extrahepatic effects which are associated with systemic metabolic derangements. The aim of this work is to specifically investigate whether PCSK9 deficiency affect glucose metabolism.
Methods. 2-months old PCSK9 KO and WT mice were fed for 20 weeks with a HFD (High Fat Diet) (n=12) or SFD (Standard Fat Diet) (n=12). To assess the glucose metabolism we performed a Glucose Tolerance Test (GTT) and an Insulin Tolerance Test (ITT). Moreover, we measured the glucose and insulin plasma levels after a fasting and refeeding experiment and the insulin content in the pancreas.
Results. PCSK9 KO mice presented a significant increased glycemia under fasting and refeeding condition, compared to WT mice

(+25%±7%; p<0,05). Furthermore PCSK9 deficiency resulted in impaired glucose tolerance compared to control mice (GTT-AUC +25%±11% SFD; +40%±9% HFD, p<0,05), both after 12 and 20 weeks, while ITT was similar among the two groups. Insulin levels were significantly lower in plasma from PCSK9 KO mice (-35%±13%; p<0,05) while resulted increased in the pancreas of KO mice compared to WT.
Conclusion. Our data suggest that the impaired glucose tolerance observed in PCSK9 mice is not due to insulin resistance, but it seems associated with a dysfunction in insulin secretion from the pancreas.

Acknowledgments. This work was supported by grants from Telethon Foundation (GGP13002), Ministero della Salute WFR GR-2011-02346974 to GDN, from Fondazione Cariplo (2012-054).

PENTRAXIN 3 DEFICIENCY IS ASSOCIATED WITH INCREASED ARTERIAL THROMBOSIS IN ANIMAL MODELS

F. Bonacina[1*], S.S. Barbieri[2*], L. Cutuli[1*], P. Amadio[2], A. Doni[3], M. Sironi[3], S. Tartari[3], B. Bottazzi[3], C. Garlanda[3], E. Tremoli[1,2], A. Mantovani[3], A.L. Catapano[1,4], G.D. Norata[1,5,6]
*[1]Department of Pharmacological and Biomolecular Sciences, Università degli Studi di Milano; [2]IRCCS, Centro Cardiologico Monzino, Milano; [3]IRCCS, Humanitas Research Foundation, Rozzano, (Milan); [4]IRCCS Multimedica, Milano; [5]SISA centre for the Study of Atherosclerosis, Bassini Hospital, Cinisello Balsamo (Milan); [6]William Harvey Research Institute, Barts and The London School of Medicine & Dentistry, Queen Mary University, London (UK) *These authors equally contributed to the work*

PTX3 is a biomarker of cardiovascular diseases and exerts protective functions in acute myocardial infarction and atherosclerosis. Here we investigated the role of PTX3 during FeCl3-induced arterial thrombosis.
PTX3 KO mice showed a 60% reduction in carotid artery blood flow with a greater thrombus formation compared to 20% of WT mice (p<0.01) following arterial thrombosis, an effect mediated by PTX3 derived from non-hematopoietic cells: indeed, PTX3 KO mice transplanted with bone marrow from WT or PTX3 KO mice presented a significant increased carotid occlusion compared to WT mice transplanted with bone marrow from WT or PTX3 KO mice (p<0.01). PTX3 plasma levels were not increased after arterial thrombosis and the protein co-localized with fibrin within the border of the damaged artery and the thrombus. The pro-thrombotic phenotype observed in PTX3 KO mice was independent on altered hemostatic properties, impaired platelet activation (in terms of P-selectin and integrin αIIbβIII expression) and aggregation, modulation of P-selectin activity as P-selectin KO/PTX3 KO mice showed a significant reduction in carotid artery blood flow and increased arterial thrombus formation compared to P-selectin KO (p<0.01). Platelet aggregation induced by collagen and fibrinogen incubated with PTX3 was significantly decreased (p>0.01), an effect depended mainly on the C-terminal and N-terminal domain respectively. Finally, exogenous administration of hrPTX3 reverted the pro-thrombotic phenotype in PTX3 KO mice and improves the outcomes in WT (p<0.01) after arterial thrombosis.
In conclusion, PTX3 deficiency is associated with increased arterial thrombosis via modulation of collagen and fibrinogen thrombogenicity.

TELEMEDICINE IN PATIENTS WITH HEART FAILURE: IS THERE AN IMPROVEMENT IN THEIR QUALITY OF LIFE?

A. Caretta, C. Debenedetti, G. Ferrante, S. Persampieri, A. Pugno, L. Giugno, S. Carugo
U.O. Cardiologia/UTIC, Ospedale San Paolo, Milano

Introduction. Telemedicine in patients with heart failure represents an established opportunity to improve out-of-hospital assistance: it is therefore useful to investigate on outcomes that this kind of monitoring produces on patients' health.
Aim. Aim of the study is to evaluate if telemedicine improves quality of life in a group of heart failure patients (NYHA III-IV), both from the physical and the psychological point of view.
Materials and methods. A retrospective and observational study was carried out analysing data collected during the years 2014 and 2015. Patients were enrolled in the Cardiology Unit of "San Paolo" Hospital, after a discharge for heart failure and with a six months follow-up. Before discharge, patients were well trained to collect and report their blood pressure, heart rate, urinary output, body weight and to transmit their EKGs using a portable device.
In addition, during the study a "Short Form 12" questionnaire was given every month and a phone interview was made weekly, in order to evaluate patients' quality of life.
Results. 20 patients were enrolled (13 males and 7 females), aged 75±9 years.
At the end of the study: 2 patients were re-hospitalized (1 for ST-segment depression at the home-EKG and 1 for reported dyspnoea); 5 were referred to an out-of-hospital cardiological examination; in 9 patients a change in therapy was prescribed; 1 died for cardiac arrest of unknown cause.
From the "Short Form 12" questionnaire positive results were achieved regarding patients' quality of life, particularly in the physical scores.
Conclusions. Telemedicine shows encouraging results: re-hospitalization rate was low, in most cases clinical stabilization was possible without hospital admission and with a significant improvement in therapeutic adherence.
Quality of life proved statistically better, especially regarding patients' physical well-being.
However, because of a limited number of patients enrolled, larger researches would be advisable.

EVALUATION OF HDL CHOLESTEROL EFFLUX CAPACITY (CEC) AFTER CONSUMPTION OF AN INNOVATIVE PASTA ENRICHED WITH BIOACTIVE COMPONENTS AND FUNCTIONAL PROBIOTICS

E. Cipollari[1], F. Zimetti[1], M.P. Adorni[1], N. Ronda[1], D. Angelino[2], N. Pellegrini[2], F. Bernini[1], E. Favari[1]
[1]Department of Pharmacy, University of Parma; [2]Department of Food Science, University of Parma

Aim. Epidemiological evidence indicate that high intake of whole grain is associated with a reduced risk of cardiovascular and metabolic disease. This study aimed to investigate the effect of consumption of a new functional whole grain on serum HDL-CEC, a metric of HDL functionality, which has recently emerged as a new marker for cardiovascular risk evaluation.

Methods. 40 healthy volunteers were randomly assigned to two treatments such as experimental pasta made with whole-wheat flour enriched in β-glucan from barley and spores of B. coagulans GBI-30 and control pasta produced with the same technological process and with the same, but not integral, variety of wheat as the functional one. CEC measurement was performed ex vivo on whole plasma collected from subjects before and after three months of treatment. Individual cholesterol efflux pathways were evaluated by using specific, widely accepted cell-based radio assays.
Results. In our study, despite no change in HDL concentration, we did observe an improvement in ABCG1 CEC after treatment with the innovative pasta. Additionally, in treated subjects, but not in subjects treated with control pasta, ABCG1-mediated CEC inversely correlates with homocysteinemia, an independent risk factor for coronary disease, while a direct significant relation was found with plasmatic folic acid, which is considered a protective factor for cardiovascular disease.
Conclusions. Since HDL-CEC has been suggested as a new biomarker in CVD, our study is relevant to prove, despite preliminary, that treatment with a functional food modulates HDL functionality; correlations between plasmatic indices of cardiovascular disease and CEC provide new insight on its role as a biomarker.

HOME BLOOD PRESSURE MONITORING: CORRECT METHODS OF USE, RELIABILITY VALUE AND ITS UTILITY TO IMPROVE COMPLIANCE IN HYPERTENSIVE PATIENTS

V. Creato, C. De Simone, A. Cocco, A. D'Angelo, M. Bucci, F. Cipollone
Dip. Medicine and Aging Sciences, University of Chieti

Aim. Home-blood-pressure-monitoring (HBPM) is a cardiovascular (CV) risk predictor: it represents an instrument patients (pts) may use to cooperate with the physician to assess therapeutic effectiveness. Despite what, specific self-measurements recommendations for pts lack.
Objectives of the study was:
– to analyse how patients use blood-pressure measuring devices;
– to evaluate if a greater compliance to HBPM leads to better therapeutic effectiveness.
Methods. We selected 100 hypertensive pts who usually perform HBPM; their ambulatory pressures and arterial stiffness (PWV) through Vicorder were measured. Pts were then asked to answer a questionnaire about the devices used, the procedure of measurement and if they received or not instructions on how to use it.
They finally performed a self-measurement while observed and, accordingly to how accurately the steps were performed, were assigned to 4 credit groups.
Results.
– Almost half patients (45%) didn't follow an appropriate pattern, although 71% of pts had already received instructions on the devices;
– Previously instructed pts made fewer mistakes while performing self-measurements ($p<0,001$);
– Pts with long-standing hypertension performed self-measurement more accurately ($p=0,044$);
– Pts performing multiple self-measurements per occasion, made fewer mistakes ($p=0,038$);
– Pts less accurate in the procedures showed a greater number of risk factors ($p=0,039$) and PWV values >10m/s, index of sub-clinical organ damage ($p=0,003$; $rho=0,497$);
– Pts with target values of blood pressure for the most part belonged to the better credit groups in self-measurements.
Conclusions.
– HBPM cheaply provides BP values unaffected by the "white-coat effect";
– to obtain reliable measurements, pts must be adequately instructed about the devices;
– a better accuracy in self-measurements could be positively related to blood pressure treatment adherence and to a greater control of CV risk factors.

NON VALVULAR ATRIAL FIBRILLATION AND NOACS: OUR EXPERIENCE

A. D'Angelo, C. De Simone, V. Creato, A. Cocco, M. Bucci, F. Cipollone
Università degli Studi G. D'Annunzio, Chieti-Pescara

Atrial fibrillation (AF) is a significant problem due to population ageing and represents a major risk factor for stroke. Warfarin anticoagulation has been proven effective for stroke prevention in AF, but it can be difficult to manage and it requie intensive monitoring; this is why 30% of the population affected by AF was treated with antiplatelet therapy before the recent introduction of Novel Oral AntiCoagulants (NOACs). NOACs have been shown to be as effective as warfarin for stroke prevention in non valvular AF (NVAF) and are associated with a reduced risk of bleeding compared with warfarin. Since NOACs don't need frequent monitoring as warfarin, the number of AF patients treated by anticoagulation could increase but, in order to prevent patients from underestimating the risks connected to this new drug, it'simportant to accurately plan follow-up visits.
In the geriatric clinic of G. Bernabeo Hospital we established an ambulatory service dedicated to all patients affected by AF who wanted to begin or were already in treatment with a NOACs.
Our main purposes were to test efficacy and safety of NOACs in real life practice, to reduce the number of patients treated by antiplatelet therapy or with no anthitrombotic therapy at all and to provide a proper follow up in accordance with EHRA guidelines.
Between January 2013-2015, we started NOACs treatment in 156 NVAF patients; none of them showed a major embolic event and we observed only one case of cerebral bleeding (0,64%); 111patients (72%) were treated with antiplatelet drug or with no anthithrombotic therapy before switching to NOAc. We scheduled follow-up visits after 1, 3, 6 and 12 month sin the first year and then every 6 months and we observed that 98% of patients respected the scheduled visits. Our experience confirmed the safety and efficacy of NOACs in real life practice and allowed us to decrease the number of AF untreated patients in our region.

INSIGHTS ON PCSK9 TARGETING: A CASE REPORT

C. De Simone, V. Creato, A. D'Angelo, A. Cocco, M. Bucci, F. Cipollone
Università "G. d'Annunzio", Chieti

Familial hypercholesterolemia (FH), a co-dominantly inherited disease of lipid metabolism that markedly increases risk of premature CAD, is significantly under-diagnosed and available treatments options often reveal ineffective in achieving LDL-C goals or not toler-

ated. A 68 year old woman presented to our care for a reassessment of her lipid lowering therapy (rosuvastatin 40 mg and ezetimibe 10 mg). Two years earlier she had undergone CABG and PTA. She had a family history of CAD (AMI in her brother in early age). The patient had tried several treatment regimens in the past, all well tolerated, but not effective, including LDL apheresis, made difficult by unavailable venous accesses. Patient's LDL-C blood levels (LDL-C 177 mg/dl), performed during treatment, documented failure to reach the guideline-recommended LDL-C goal of <70 mg/dl. Physical examinations revealed no significant findings. Her DLCNS was 2 (unlikely phenotypic diagnosis of FH). Then, we calculated a hypothetical untreated LDL-C level, using the chart reporting the percentage reduction in LDL-C with statins. The value of LDL-C obtained (272 mg/dl) made the patient's DLCNS score increase from 2 to 6 thus defining a "probable" diagnosis of FH, as confirmed by the genetic test that detected a heterozygous mutation in LDL-R encoding gene. The patient was then enrolled in a Phase III study with Alirocumab, which led to achieve LDL-C therapeutic goals of LDL-C <70 mg/dl (-56% from baseline) at the end of study period, showing good clinical and laboratory tolerability profile. Detection of FH undiagnosed patients leads to a change in their CV risk profile and to more ambitious therapeutic goals, that often represent a hard challenge with conventional drugs available; the new classes of cholesterol-lowering drugs hold thus enormous potential in the management of dyslipidaemia in high risk patient.

ARYL HYDROCARBON RECEPTOR: A NOVEL TARGET FOR THE ANTI-INFLAMMATORY ACTIVITY OF STATIN THERAPY

V. Gandolfo[1], F. Fallarino[2], M.R. Mannarino[1], M. Gargaro[2], A. Turco[2], G. Scalisi[2], V. Bianconi[1], M. Pirro[1]
[1]Unit of Internal Medicine, Department of Medicine. University of Perugia; [2]Unit of Pharmacology, Department of Experimental Medicine, University of Perugia

Aim. Statin therapy is often associated with a significant attenuation of the systemic inflammation response. Aryl hydrocarbon receptor (AhR) is believed to play an active role in the control of inflammation. Interestingly, specific statins promote the transcription of CYP1A1, a cytochrome whose induction involves the classical activation cascade of AhR. We investigated whether AhR might be involved in the anti-inflammatory effect of statin treatment.
Methods. Luciferase assay was performed to test the response of murine hepatocytes overexpressing AhR and the xenobiotic responsive elements (XRE) to different statins. Moreover, lipopolysaccharide (LPS)-treated RAW 264.7 macrophage cells were tested for the expression of AhR and for their pro-inflammatory/anti-inflammatory response to different statins.
Results. Luciferase assay showed that both atorvastatin and pravastatin induced a significant activation of AhR at 100 μM; interestingly, atorvastatin was able to activate Ahr at 10 μM. LPS treatment of RAW 264.7 macrophage cell lines resulted in AhR induction and interleukin-6 (IL-6) secretion. Both pravastatin and atorvastatin reduced significantly the IL-6 response to LPS stimulation and increased the IL-10 secretion by LPS-stimulated macrophages.
Conclusions. Specific statins are able to activate AhR. Moreover, atorvastatin and pravastatin, possibly by binding AhR, attenuated the inflammation response induced by LPS, as evaluated by a reduction of the IL6/IL10 ratio in cultured LPS-stimulated macrophages. Therefore, AhR might represent a possible mediator of the anti-inflammatory effects of statin treatment.

VITAMIN D STATUS AND HDL FUNCTIONALITY IN HEALTHY PRE-MENOPAUSAL WOMEN.

D. Greco[1], D. Kocyigit[2], M.P. Adorni[1], N. Ronda[1], K.M. Gurses[3], S.H. Oguz[4], A. Gurlek[4], L. Tokgozoglu[2], F. Zimetti[1]
[1]Department of Pharmacy, University of Parma, Parma; [2]Department of Cardiology, Hacettepe University Faculty of Medicine, Ankara, Turkey; [3]Department of Cardiology, Konya Training and Research Hospital, Konya, Turkey; [4]Department of Internal Medicine, Section of Endocrinology and Metabolism, Hacettepe University Faculty of Medicine, Ankara, Turkey

Background. Low vitamin D (vitD) status has been linked to increased cardiovascular (CV) risk. Serum HDL cholesterol efflux capacity (CEC) is a metric of HDL functionality that is inversely correlated to CV risk. At present there is no data on the possible correlation between HDL CEC and vitD levels.
Aim. We evaluated whether impaired HDL functionality occurs in otherwise healthy vitD deficient pre-menopausal women.
Methods. 17 pre-menopausal vitD-deficient women and 14 age-matched women with normal vitD plasma levels were recruited. Serum HDL was isolated by precipitation with polyethylene glycole. HDL CEC was assessed by radioisotopic technique using specific cell models allowing to evaluate different cholesterol efflux pathways. Flow-mediated dilatation (FMD) and pulse wave velocity (PWV) were measured by standard techniques as markers of subclinical atherosclerosis.
Results. No differences were found between groups lipid profile, including HDL levels. ABCG1-mediated CEC was lower in the vitD deficient group (by 20%, $p<0.05$), in whom FMD was also significantly lower ($p<0.001$). Meanwhile, aqueous diffusion - and ABCA1 - mediated CEC did not differ between the two groups. PWV showed a non- statistical tendency to be higher in vitamin D- deficient women.
Conclusions. These preliminary data on a small number of subjects suggest a specific impairment in ABCG1-mediated HDL CEC in vitD deficient pre-menopausal women, despite no differences in HDL serum levels. This observation encourages to study the possible role of vitD deficiency in HDL functional impairment and verify the relative weight of the two metabolic defects for CV risk in larger cohorts.

MECHANISM INVOLVED IN THE PATHOGENESIS OF RENAL DISEASE IN LCAT DEFICIENCY

F. Lucca[1], A. Ossoli[1], E.B. Neufeld[2], C. Vitali[1], M. Locatelli[1], M. Abbate[3], C. Zoja[3], G. Franceschini[1], A.T. Remaley[2], L. Calabresi[1]
[1]Centro Grossi Paoletti, Dipartimento di Scienze Farmacologiche e Biomolecolari, Università degli Studi di Milano; [2]Lipoprotein Metabolism Section, Cardiovascular and Pulmonary Branch, National Heart, Lung and Blood Institute, National Institutes of Healt, Bethesda, Maryland, United States of America; [3]IRCCS-Istituto di Ricerche Farmacologiche Mario Negri, Centro Anna Maria Astori, Science and Technology Park Kilometro Rosso, Bergamo

Aim. Human familial lecithin:cholesterol acyltransferase (LCAT) deficiency (FLD) is an enzymatic disorder characterized by the presence in plasma of an abnormal cholesterol-rich multilamellar particle called lipoprotein-X (LpX), low levels of HDL and renal disease. The aim of this study is to investigate the role of LpX in FLD and its correlation with renal disease.
Methods. We created a synthetic LpX which is similar, in size and composition, of endogenous LpX found in FLD patients and we chronically administered it to WT and $LCAT^{-/-}$ mice. We also perform *in*

vitro studies to investigate the role of LpX in renal disease analyzing renal function, gene expression and metabolism of synthetic LpX.
Results. Our *in vitro* and *in vivo* studies demonstrated an apoA-I and LCAT-dependent pathway for LpX conversion to HDL-like particles, which likely mediates normal plasma clearance of LpX. Plasma clearance of exogenous LpX was markedly delayed in *Lcat*$^{-/-}$ mice, which have low HDL, and chronically administration of synthetic LpX induced proteinuria and nephrotoxic gene changes, as well as all of the hallmarks of FLD renal disease as assessed by histological analyses. *In vivo* EM studies revealed LpX uptake by macropinocytosis into mouse glomerular endothelial cells, mesangial cells and podocytes before to reach lysosomes where it was degraded. Endocytosed LpX appeared to be degraded by both human podocyte and mesangial cell lysosomal PLA_2 and induced podocyte secretion of pro-inflammatory IL-6 *in vitro*. It also increase the expression of nephrotoxic genes in *Lcat*$^{-/-}$ mice.
Conclusions. LpX seems to have a nephrotoxic role and, in absence of LCAT, induces all of the histological and functional hallmarks of FLD. In addition, our studies suggest that LpX-induced loss of endothelial barrier function and release of cytokines by renal glomerular cells likely plays a role in the initiation and progression of FLD nephrosis.

CIRCULATING PROGENITOR CELLS IN HYPERTENSIVE SUBJECTS: EFFECTIVENESS OF A TREATMENT WITH OLMESARTAN IN IMPROVING CELL NUMBER AND MIRS PROFILE BESIDES EXPECTED PHARMACOLOGICAL EFFECTS

G. Mandraffino[1], C.O. Aragona[1], V. Cairo[1], F. Mamone[1], A. D'Ascola[2], M.A Sardo[1], A. Lo Gullo[1], E. Imbalzano[1], A. Saitta[1]
[1]Department of Clinical and Experimental Medicine, Internal Medicine; [2]Department of Biochemical, Physiological and Nutritional Sciences; University of Messina

Aim. CD34+ circulating progenitor cells (CD34+CPCs) are a population of multipotent cells able to delay the development of atherosclerosis and cardiovascular disease (CVD) in conditions of increased CV risk. Increased reactive oxygen species (ROS), a common feature of CV risk factors including hypertension, may be toxic for cells. MicroRNAs (miRs) 221 and 222 have been shown to participate in differentiation and proliferation of CD34+CPCs, inhibiting cell migration and homing; miR221/222 are increased and associated with cell number and ROS in CD34+CPCs from hypertensive patients without additional risk for CAD. Moreover, miR221/222 modulate different genes regulating angiogenesis and inflammation. Our aim was to evaluate whether in hypertensives a treatment with olmesartan may modify the number of CD34+CPCs and the levels of miR221/222 and ROS.
Methods. We evaluated CD34+CPC number, intracellular miR221/222 and ROS levels, arterial stiffness and echocardiographic indices at baseline (T0) and after a six-months treatment with olmesartan, 20 mg/die (T1) in 57 hypertensives with no additional risk factor for CAD, and in 29 healthy controls (baseline); fibrinogen, CRP, glucose and lipid profile were also evaluated.
Results. At T1, systolic and diastolic blood pressure, ROS and miR221/222 were significantly decreased (all $p<0.001$) with respect to T0, and cell number was increased ($p<0.001$). CRP and fibrinogen levels also were reduced ($p<0.001$), as were arterial stiffness indices.
Conclusion. Olmesartan is effective in reducing miRs and ROS levels in CD34+CPCs from hypertensives, as well as in increasing CD34+CPC number, besides its expected pharmacological effects.

AT1R BLOCKADE BY LOSARTAN AS WELL AS SMOKING CESSATION REDUCE BIGLYCAN EXPRESSION IN UNCOMPLICATED SUBJECTS

G. Mandraffino[1], C.O. Aragona[1], V. Cairo[1], F. Mamone[1], A. D'Ascola[2], M.A. Sardo[1], A. Lo Gullo[1], E. Imbalzano[1], A. Saitta[1]
[1]Department of Clinical and Experimental Medicine, Internal Medicine; [2]Department of Biochemical, Physiological and Nutritional Sciences; University of Messina

Aim. Whether the expression of the ECM proteoglycan Biglycan (BGN) may be modified by the administration of the AT1R-blocker Losartan in uncomplicated essential hypertensives and/or by smoking cessation in subjects with no risk factors for CVD.
Methods. 102 (out of 126 formerly enrolled) newly diagnosed uncomplicated essential hypertensives were treated with Losartan 100mg once a day for 6 months, and 71 healthy subjects (out of 251) who maintained long term abstinence (12 months) from cigarette smoking have been evaluated. BGN-mRNA expression was measured in monocytes isolated from peripheral blood at baseline (T0) and at T1. Inflammatory markers, lipid profile, cIMT and arterial stiffness indices were evaluated at each time-point.
Results. Both by Losartan administration (L) and smoking cessation (SC), several study parameters appeared to be improved: SBP (L:-14.5%; SC:-2%. $p<0.001$), DBP (L:-14%; SC:–5%. $p<0.001$), CRP (L:-75%; SC:-23.3. $p<0.001$), Fibrinogen (L:-15%; SC:–12%. $p<0.001$), Il-6 (L:-26%; SC:-9.5%. $p<0.001$), PWV (L:-14%; SC:-6.6%. $p<0.001$), AIx (L:-20%; SC:-20%. $p<0.001$), HDL-C (L:+8%; SC:+9.5%. $p<0.001$). As regards BGN, we observed both absolute (L:-17%; SC:-7.5%. $p<0.001$) and relative expression reduction (L:-31%; SC:-43%. $p<0.001$). Regression analyses suggested that BGN reduced expression is associated to treatment (L or SC), through inflammatory status improvement. In hypertensives, these data confirm that BGN expression may be sustained also via AT1R activity. Moreover, the improvement of PWV and AIx appeared to be not linearly associated with BP lowering.
Conclusions. BGN, an important player in atherogenesis in different clinical settings, is enhanced in essential hypertensives and in active smokers. Here we suggest that modulating RAAS by an AT1R blocker as Losartan, as well as removing the exposure to a well-known CV risk factor as cigarette smoking, BGN expression could be significantly reduced. Consistently, we can speculate that BGN under-regulation should be linked to slowing in the progression of vascular damage due to the offset of CV risk factors.

ASSOCIATION OF CAROTID ATHEROSCLEROSIS WITH CORONARY ARTERY DISEASE: A RETROSPECTIVE STUDY OF 589 PATIENTS

D. Migliorino, A. Mignano, S. Evola, G. Novo, E. Corrado, S. Novo
Chair and Division of Cardiology, Centre for the Early Diagnosis of Preclinical and Multifocal Atherosclerosis, Reference Regional Centre for the Diagnosis and Care of Heart Failure, Department of Internal Medicine and Cardiovascular Diseases, University Hospital "Paolo Giaccone" of Palermo

Background and Aim. Several studies have shown that the risk of cardiovascular events is higher in subjects with ultrasound evidence of subclinical carotid atherosclerosis. The aim of our study was to evaluate the correlation between carotid atherosclerosis evaluated by Doppler ultrasound and severity of coronary artery disease (CAD) in patients with typical chest pain undergoing diagnostic coronary angiography during hospitalization.

Methods. Wwe studied retrospectively 589 patients admitted to our Cardiology Unit for chest pain that underwent to coronary angiography. We evaluated carotid plaques according to ESC Guidelines. During the coronary angiography we considered one, two or three vessel disease if coronary vessels had stenosis >50%. Patients with a positive medical history for cardio and cerebrovascular disease and with positive biomarkers of myocardial infarction were excluded.
Results. The detection of carotid plaque was predominantly associated with the presence of angiographically diseased coronary arteries (p=0,009). Particularly, the presence of a carotid plaque with a diameter >2,1 mm (p=0,0001) was associated with a higher prevalence of coronary artery disease (sensitivity 61,3%; specificity 73,5%; PPV 97,4%). Dimensions of carotid plaques were significantly correlated with the complexity of coronary artery disease calculated by Syntax score (p<0,0001). Moreover bilateral carotid atherosclerosis was associated with coronary atherosclerosis too (p<0,0001). Besides the detection of carotid atherosclerosis was strongly correlated with the coronary artery disease itself (overall p=0,008).
Conclusions. Given the significant statistical correlation between the presence of carotid atherosclerosis and the severity of coronary artery disease (in terms of number of involved vessels), we believe that the evaluation of Doppler ultrasound of carotid arteries might provide the clinician additional informations about the global cardiovascular risk of the patients with typical chest pain and negative markers of acute coronary syndrome. Moreover, the presence of carotid atherosclerotic plaque may be predictive of coronary atherosclerosis.

A CASE OF TANGIER DISEASE WITH AN UNUSUAL CLINICAL PRESENTATION AND A NOVEL INTRONIC MUTATION

I. Minicocci[1], G. Truglio[1], E. Grieco[1], A. Gallo[1], M. Lucarelli[2], M. Arca[1], M. Maranghi[1]
[1]Departments of Internal Medicine and Allied Specialties; [2]Cellular Biotechnologies and Hematology, Sapienza, University of Rome

Tangier disease (TGD, OMIM #205400) is an autosomal recessive disorder characterized by severe reduction in HDL-cholesterol (HDL-C), peripheral lipid storage, and conduction block neuropathy. Tonsillectomy and hepatomegaly are reported in about 90% of TGD patients and abnormal splenic and bone-marrow ultrastructural features have been also associated to this rare condition. We describe a case carrying two intronic variants in ABCA1 gene who presented with spleen rupture.
A 40 years-old man was referred to our clinic for hypocholesterolemia and thrombocytopenia. The proband had very low levels of total-cholesterol (TC) 53 mg/dl, high-density-lipoprotein-cholesterol (HDL-c) 1 mg/dl, and mild hypertriglyceridemia (TG) 202 mg/dl, and a low platelets count (72000/µl). His clinical examination was normal. He reported tonsillectomy and splenectomy due to spontaneous spleen rupture. Liver ultrasonography showed hepatomegaly and steatosis. A bone biopsy and bone-marrow aspiration had been performed, showing 50% foamy cells infiltration and mild hypoplasia, respectively. Proband's father had thrombocytopenia and reduced HDL-C levels while proband's mother died at the age of 42 for cerebral aneurism rupture. Probands's sister and twin children showed splenomegaly and have performed tonsillectomy during childhood. All investigated family members presented reduced HDL-C levels.
Lipid levels and past clinical history were highly suggestive of a genetic HDL-deficiency condition. To confirm diagnosis, DNA extraction and sequencing of ABCA1, APOA-I, LCAT and LPL genes were performed. The proband was found to be compound heterozygous for two intronic ABCA1 variants, a novel c.1510-1G>A variation in intron 12 and a recently described TD-causing c.1195-27G>A variation in intron 10. All were considered damaging by in silico prediction. No mutations were found in APOA-I, LCAT and LPL genes.
To assess the effect of the novel 1510-1 G>A variation on mRNA splicing, cDNA derived from peripheral blood mononuclear cells of the proband and a control subject were analyzed. In proband two transcripts of 157 bp, corresponding to the wild-type allele, and 135 bp corresponding to the mutated allele were detectable. The sequence of mutant transcript showed that the lack of initial 22 bases of exon 13 with the presence of premature stop codon. This suggest the translation of a truncated, probably not functional, ABCA1 protein.
To our knowledge this is the second reported case of Tangier complicated with spleen rupture. This uncommon clinical presentation appear to be related to abnormal lipid accumulation into reticuloendothelial system due to defective ABCA1-mediated removal.

ASSOCIATION OF RENAL RESISTIVE INDEX WITH MARKERS OF EXTRARENAL VASCULAR CHANGES IN PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS

M. Morreale[1], G. Mulè[1], A. Ferrante[2], F. D'ignoto[1], S. Cottone[1]
[1]Dipartimento Biomedico di Medicina Interna e Specialistica (DIBIMIS), Unit of Renal Disease and Dyalisis, European Society of Hypertension Excellence Centre ; [2]Unit of Rheumatology, Università di Palermo

Aim. Recent data suggest that renal hemodynamic parameters obtained by duplex Doppler sonography, especially the intrarenal resistive index (RRI), may be associated with systemic vascular changes. We evaluated the relationships between RRI and arterial stiffness, assessed by aortic pulse wave velocity (aPWV), and between RRI and subclinical atherosclerosis, assessed by measuring carotid intima-media thickness (cIMT) in patients with Systemic Lupus Erythematosus (SLE).
Methods. We enrolled 39 SLE subjects (mean age 39 years) compared with 19 healthy controls, matched for age and sex. Each participant underwent 24 h ambulatory blood pressure (ABPM), aPWV, cIMT and RRI measurements.
Results. RRI correlated significantly with aPWV (r: 0.44; p=0.006), and with cIMT (r: 0.46; p=0.003). Both correlations held (p=0.01) even after correction for age, mean arterial pressure, and glomerular filtration rate.
Conclusions. Our results suggest that the RRI may be considered a marker of systemic vascular changes and probably a predictor of cardiovascular risk in SLE patients.

EARLY VASCULAR AGING IN NORMOTENSIVE PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS. COMPARISON WITH YOUNG HYPERTENSIVE PATIENTS

M. Morreale[1], G. Mulè[1], A. Ferrante[2], F. D'ignoto[1], S. Cottone[1]
[1]Dipartimento Biomedico di Medicina Interna e Specialistica (DIBIMIS); Unit of Renal Disease and Dyalisis, European Society of Hypertension Excellence Centre; [2]Unit of Rheumatology, Università di Palermo

Aim. It is well known that connective tissue diseases, like Systemic Lupus Erythematosus, are associated with early and accelerated

atherosclerosis. During the last years, it has well established the concept of "Early vascular aging" (EVA), whose the aortic stiffness represents one of the most important markers. The aim of this study was to evaluate EVA, assessed by measuring aortic pulse wave velocity (aPWV) and subclinical atherosclerosis, evaluated by carotid intima-media thickness (IMT) measurement, in a group of 50 normotensive patients with Systemic Lupus Erythematosus (SLE), (mean age 39±12 years). Then we compared these subjects with a group of 50 age - and sex - matched essential hypertensive (EH) individuals and with a control group of 20 healthy volunteers (CG).
Methods. Each patient underwent clinic and ambulatory blood pressure measurement (ABPM), routine blood chemistry, aPWV measurement and IMT evaluation.
Results. Despite clinic and 24-H BP values were significantly lower in the SLE patients and in CG when compared to those of the hypertensive subjects (all p<0.0001), aPWV and IMT were statistically lower in the CG when compared to those of SLE subjects and of EH individuals (all p<0.001). Overall, SLE and EH patients showed similar values of IMT (0.80±0.2 mm VS 0.77±0.2; p=0.31) and Apwv (8.8±2 m/sec VS 9.2±2 m/sec, p=0.47).
Conclusions. Our results seem to suggest that SLE has the same deleterious impact on vascular aging as well as high blood pressure. Probably this unfavorable effect of SLE may be mediated by chronic inflammation.

ISOLATED SYSTOLIC HYPERTENSION CHARACTERISTICS: DATA FROM THE BRISIGHELLA HEART STUDY (ITALY) AND THE ENAH STUDY (CROATIA)

A. Parini[1], V. Ivković[2], A.F.G. Cicero[1], S. Bacchelli[1], M. Cagnati[1], E. Grandi[1], D. degli Esposti[1], M. Rosticci[1], S. D'Addato[1], A. Vrdoljak[2], M. Abramović Barić[2], M. Fuček[2], E. Ivandić[2], S. Karanović[2], M. Laganović[2], I. Pećin[2], D. Rogić[2], B. Jelaković[2], C. Borghi[1]
[1]Medical and Surgical Sciences Department, University of Bologna; [2]University of Zagreb School of Medicine, Department of Nephrology, Hypertension, Dialysis and Transplantation, UHC Zagreb, Croatia

Aim. Isolated systolic hypertension (ISH) in elderly is associated with increased global risk. It could be considered as a rough biomarker of increased arterial stiffness and advanced biological aging. In our study, we evaluate the association between ISH and other cardio-metabolic risk factors in two rural European populations from South Europe (Italy and Croatia).
Methods. In this international prospective long-term follow up study data on 5162 subjects from BrEnah cohort formed from original cohorts of Brisighella Heart Study (Italy) and ENAH Study (Croatia) were analized. Out of them 2253 subjects (694 from Croatia, 1559 from Italy; 980 m, 1273 f) were eligible for further analyses. Fasting blood was analysed for glucose, lipids, uric acid, serum creatinine.
Results. In general rural population from South Europe prevalence of ISH is high. Difference between Croatian and Italian subgroups was found (28.8% vs 44.8%; p<0.001; no gender differences) what is concordance with difference in age between two cohorts. Beside age, ISH was significantly associated with eGFR and various metabolic parameters including visceral obesity glucose intolerance and uric acid
Conclusions. In this group of patients prevalence of ISH was high. Observed difference between Italian and Croatian subgroup is mostly due to difference in age. Metabolic disturbances are frequently associated with ISH additionally increasing global risk.

GENETIC AND PHENOTYPIC CHARACTERIZATION OF LDL-RECEPTOR IN PATIENTS WITH A CLINICAL DIAGNOSIS OF FAMILIAL HYPERCHOLESTEROLEMIA

C. Pavanello[1], C. Tarlarini[2], S. Penco[2], L. Calabresi[1], S. Castelnuovo[3], G. Mombelli[3], C.R. Sirtori[3]
[1]Centro E. Grossi Paoletti, Dipartimento di Scienze Farmacologiche e Centro E. Grossi Paoletti, Italy Biomolecolari, Università degli Studi di Milano; [2]S.C. Analisi Chimico Cliniche e Patologia Clinica Dipartimento di Medicina Laboratorio Niguarda Ca' Granda, Milano; [3]Centro Dislipidemie, Ospedale Niguarda Cà Granda, Milano

Familial hypercholesterolemia (FH) is an autosomal dominant disorder of lipoprotein metabolism. In the majority of patients, FH is caused by mutations in the gene encoding the low density lipoprotein receptor (LDLR).
We characterize the mutations of the LDLR gene in 91 patients (aged 4-77 years) with clinical definitions of possible (N=7), probable (N=13) or certain (N=71) FH. All patients were phenotypically characterized.
Plasma LDL-C level ranged from 168 to 526 mg/dl. LDLR defects were detected in 87 patients, in various regions of the gene with 1% in the signal sequence, 29% in the LDL binding domain (exons 2-6), 62% in the EGF precursor homology domain (exons 7-14) and 8% in the membrane spanning region (exons 16-17). Three patients were found homozygous for the mutations V523M, c.2312-3C>A and for the synonymous mutation G207G. One patient was compound heterozygous and 2 were double heterozygous. The most common mutations (G549D,G592E) were detected in 3 or more unrelated patients. Nonsense mutations accounted for 9.8%, missense for 51.7% and frame-shift mutations for 10.9%. We found one "in frame" deletion, 16 (18.4%) splicing mutations, and 6 (6.9%) large deletions.
Two heterozygous mutations were not described before. One of these is a duplication of the nucleotide in 1125 position which causes a frame-shift, ending in a stop at position 5 (p.K376Qfs*5) and one concerns a deletion of an adenine in intron 10 (c.1587-2delA).
All genotypes do not significantly differ in plasmatic LDL-C level. Tendon xanthomata were present in 72% of genetically defined FH.
Future analyses will be performed on genotype-negative patients to investigate other mutations involved in the development of the FH phenotype.

GENETIC DIAGNOSIS OF SUSPECTED FAMILIAL HYPERCHOLESTEROLEMIA (FH): EVALUATION OF CORONARY ATHEROSCLEROSIS BURDEN IN ASYMPTOMATIC PATIENTS

P. Pigna[1], F. Zaccagna[2], I. Carbone[2], M. Arca[1]
Dept of [1]Internal Medicine and Allied Sciences and [2]Radiology, Sapienza University of Rome, Policlinico Umberto I, Rome

Aim. Familial Hypercholesterolemia (FH) is a genetic lipid metabolism disorder that may cause severe cardiovascular disease both in adults and children. Performing genetic diagnosis may contribute to early and intensive lipid lowering therapy. Moreover the evaluation of coronary atherosclerosis burden in asymptomatic FH patients represents a very promising tool to assess cardiovascular risk.
Methods. In this observational study 41 asymptomatic patients (m.a 47.6±13.3 yrs) with heterozygous FH determined according to the criteria of the Dutch Lipid Clinic Network were enrolled. They were

under treatment with the maximum tolerated dose of statins (with or without ezetimibe) by 8.7±7 yrs; 95% of them experienced LDL-C reduction >50%, but only 18.3 % showed LDL-C <100 mg/dl. Thirty seven subjects (mean age 59±8 yrs) with 1-2 cardiovascular risk factors, no history of CHD and no lipid-lowering therapy were selected as a reference group. Both groups were subjected to coronary computed tomography and the overall prevalence (CAD), the severity (obstructive or non-obstructive defined as > or <50% stenosis), the composition (predominantly calcified, soft or mixed) of coronary plaques and the calcium score (Agatston score) were evaluated.
Results. The age - and sex - adjusted prevalence of CAD was significantly higher in FH than in controls (64% vs 34% respectively; p<0.005). FH women have a risk of CAD 4-fold higher than that among control women (75% vs 18.75%, p=0.019). The average number of plaques per patient was 2-fold higher in FH compared to controls (2.82±2.64 vs 1.37±2.07, p=0.021). Among FH, 41% of plaques were soft while 10.5% among controls (p=0.029). Compared to controls, the ATH-score was higher in FH group (4.72±4.51 vs 2.20±3.64, p=0.015), given the predominantly proximal location of plaques in the coronary vessel.
Conclusions. In FH patients, genetically determined, the development of CAD is accelerated despite intensive lipid-lowering treatment. To obtain a further reduction of atherosclerosis burden in FH, an earlier onset as well as the implementation of a more or late aggressive therapy is highly advisable, such as ab-antiPCSK9 or Lomitapide.

POSSIBLE PCSK9 PRO-INFLAMMATORY ACTION IN THP-1-DERIVED MACROPHAGES

C. Ricci[1], A. Corsini[1,2], N. Ferri[3]
[1]Dipartimento di Scienze Farmacologiche e Biomolecolari, Università degli Studi di Milano; [2]Multimedica IRCCS, Milan; [3]Università degli Studi di Padova

Chronic inflammation is directly linked to several metabolic diseases, such as atherosclerosis, and obesity, where inflammatory cells infiltrate adipose tissue and release pro-inflammatory mediators, such as TNF-α, IL-6, leptin and resistin, contributing to generate a state of chronic low-grade sub-clinical inflammation. PCSK9 and resistin share a common structural domain in the cysteine-rich portion of their carboxyl-terminal region and express some common pathophysiological activities. Indeed, both PCSK9 and resistin are positively correlated with increased hepatic VLDL and apoB100 production and with fasting serum triglycerides and both regulates LDLR degradation. In the present study, we will investigate the possible resistin-like activity of PCSK9 on macrophages.
We treated THP-1-derived macrophages for 24 h with increasing concentrations of PCSK9 (0.25, 0.5, 1.0 and 2.5 μg/ml) and resistin (20 and 50 ng/ml) and we evaluated pro-inflammatory cytokines mRNA levels. We observed a dose dependent increase in IL1β mRNA levels but, more surprisingly, IL-6, TNF-α, MCP-1 and MIP-2α mRNA levels show a very strong increase in response to the 2.5 μg/ml of PCSK9 (~65 fold for IL6, ~160 fold for TNF-a, ~17 fold for MCP-1, and ~42 fold for MIP2α). Moreover, through ELISA assay, we observed a significantly higher amount of IL-6 protein in the conditioned media, after treatment with 2.5 μg/ml of PCSK9. These results support a possible pro-inflammatory activity of PCSK9, potentially with a resistin-like activity. In the future, it will be determined the involvement of either the LDL receptor or the resistin receptor adenylyl cyclase-associated protein 1(CAP1), both expressed in macrophages, on the pro-inflammatory response.

HOW TO IMPROVE THE CHOLESTEROL-LOWERING EFFICACY OF EZETIMIBE IN STATIN-INTOLERANT PATIENTS IN CLINICAL PRACTICE: A RETROSPECTIVE STUDY

M. Rosticci, A.F.G. Cicero, M. Morbini, S. D'Addato, F. Fogacci, C. Borghi
Department of Medicine and Surgery Sciences, University of Bologna

Aim. Statin interruption is a relevant cause of prevention failure and increased costs. The main aim of our study was to evaluate the tolerability and efficacy of alternative approaches to reduce hypercholesterolemia in patients affected from statin-related myalgia.
Methods. We retrospectively evaluated 3534 CRFs filled in the period January 2012-December 2014 for first visits by the lipid clinic. We selected 252 CRFs based on the following criteria: hypercholesterolemia requiring pharmacological treatment, statin-related myalgia, previous failed treatment with at least two statins at low dosage, well-tolerated baseline treatment with Ezetimibe 10 mg 1 tablet/day. Then, the following lipid-lowering treatments were added in order to improve the ezetimibe LDL-lowering efficacy, based on clinical judgment: Fenofibrate 145 mg, Rosuvastatin 5 mg 1 tablet/week, Rosuvastatin 5 mg 2 tablets/week, Red Yeast Rice (standardized in Monacolin K 3 mg) + Berberine 500 mg, Berberine 500 mg b.i.d., Phytosterols 900 mg + Psyllium fiber 3.5 gr b.i.d. Patients continuing to claim a tolerable myalgia were then treated with Coenzyme Q10 nanoemulsions 200 mg per day.
Results. Almost 50% of the patients treated with Fenofibrate 145 mg/day, Rosuvastatin 5 mg 2 tablets/week, Red Yeast Rice + Berberine 500 mg, and Berberine 500 mg b.i.d. associated to Ezetimibe reached the LDL-C target foreseen for their class of cardiovascular disease risk, while the percentage was much lower for those treated with Rosuvastatin 5 mg once a week or Phytosterols 900 mg + Psyllium fiber 3.5 gr b.i.d. 11% of the patients treated with fenofibrate required treatment modification because of myalgia reappraisal, while the percentage was negligible for the other tested treatments. In patients with residual tolerable myalgia (N. 52), the treatment with Coenzyme Q10 for 8 weeks determined a mean improvement of the graduated myalgia score from 4,8±1,9 to 2,9±1,3 (p=0.013), without significant difference among different lipid-lowering drug groups. Myalgia ceased in 18 of the 52 subjects.
Conclusion. Patients suspending statins because of myalgia are largely heterogeneous in term of response to the alternative treatments, but some nutraceuticals seems to be effective and well tolerated, thus improving the Ezetimibe effect on cholesterolemia. Coenzyme Q10 nanoemulsions seems to improve the tolerability of myalgia in a large part of patients.

CAROTID ATHEROSCLEROSIS IN A VERY ELDERLY POPULATION: ROLE OF AGEING AND OTHER CARDIOVASCULAR RISK FACTORS

F. Spannella, F. Giulietti, G. Cocci, P. Giordano, M. Ricci, E. Espinosa, R. Sarzani
Internal Medicine and Geriatrics, "Hypertension Excellence Centre" of the European Society of Hypertension, Department of Clinical and Molecular Sciences, UNIVPM, IRCCS-INRCA, Ancona

Aim. Age seems to play a central role in the onset of atherosclerosis and cardiovascular (CV) disease. We compare the prevalence of carotid plaque in a very elderly population without history of PAD and major adverse cardiovascular events, with the prevalence in a population of hypertensive adults.

Methods. We studied 179 patients: 69 hospitalized very elderly (mean age: 88.5±5.5 years) admitted to our Internal Medicine and Geriatrics Department, and 110 hypertensive adults (mean age: 53.3±7.3 years) referred to our Hypertension Centre, affected by essential hypertension with or without other CV risk factors (gender, smoking, diabetes, dyslipidemia).
Results. There was no significant difference in the prevalence of carotid plaque between the two populations (51.8% in the hypertensive adults vs 58.0% in the very elderly). In the hypertensive adults smoking was the main CV risk factor associated with the prevalence of carotid plaque (OR 2.41; p=0.024), while in the very elderly the presence of hypertension had the strongest association (OR =10.5; p<0.001). Indeed, excluding the hypertensives from the very elderly, the prevalence of carotid plaque resulted significantly higher in the younger (51.8% vs 27.6%; p=0.020).
Conclusions. Our results show that CV risk factors, especially hypertension, play a key role in atherosclerosis, not only in the adult population, but also in the very elderly. Ageing is not necessarily synonymous with atherosclerosis but the real determinants of arterial "bad aging" are the superimposed CV risk factors. Prevention and treatment of CV risk factors is probably the only way to obtain a healthy longevity.

SERUM URIC ACID CHANGE AND MODIFICATION OF BLOOD PRESSURE AND FASTING PLASMA GLUCOSE IN A PHARMACOLOGICALLY UNTREATED POPOLATION SAMPLE: DATA FROM THE BRISIGHELLA HEART STUDY

R. Uso, A.F.G. Cicero, M. Rosticci, F. Fogacci, A. Parini, R. Urso, E. Grandi, S. D'Addato, C. Borghi for the Brisighella Heart Study Group
Medical and Surgical Sciences Dept., University of Bologna

Background. Serum uric acid (SUA) is an emerging risk factor for incident hypertension and type-2 diabetes. It is less clear if changes in SUA are associated to different incidence in these main cardiovascular risk factors.
Methods. For the purpose of this study we selected from the historical cohort of the Brisighella Heart Study, a longitudinal epidemiological study, subjects without diabetes, not treated in 2008 with uric acid lowering drugs nor antihypertensive ones, and that in the following 4 years did not suffer from gout, stage IV-V chronic kidney disease, neoplasms or used in non continuous way uric acid lowering drugs. Then we divided the subjects in four main groups based on SUA levels during the next 4 year: unchanged; increased by >1 mg/dL without treatment; reduced by >1 mg/dL without treatment; reduced by >1 mg/dL with continuous allopurinol therapy (150-300 mg/day).
Results. As expected, in the whole population, BMI, BP, FPG, Glucose, total and LDL-cholesterol, and TG had the tendence to increase with age between the two consecutive population surveys. However, evaluating the trend in the various studied parameters among the different subgroups, we observed specific differences only for SBP and FPG. No significant changes have been registered for anthropometric measurements, lipid pattern, liver and renal function in the different subgroups. Compared to 2008, SBP significantly increased in subjects with increased (and untreated) SUA, while improved in subjects treated with allopurinol. In 2012, subjects with increased (and untreated) SUA level had a significantly higher SBP compared with subjects with unchanged SUA and those with allopurinol treatment and lowered SUA (p<0.05). An identical trend has been observed for FPG.
Conclusion. Based on our data, obtained from a cohort of overall healthy subjects, it seems that SUA control could positively influence the age-related worsening of SBP and FPG in the general population.

CHOLESTEROL EFFLUX CAPACITY, HDL-C LEVELS AND CORONARY CALCIUM SCORE AMONG THE VERY ELDERLY SUBJECTS

F. Zimetti[1], L.A. Quaglia[2], A.M. Campos[2], W.M. Freitas[2], E. Favari[1], F. Bernini[1], A.C. Sposito[2], I. Zanotti[1]
[1]Department of Pharmacy, University of Parma;
[2]Cardiology Division, Faculty of Medical Sciences, State University of Campinas (UNICAMP), Cidade Universitária, Campinas, Brazil

Aim. Cholesterol efflux capacity (CEC) is a functional property of HDL inversely associated to prevalent atherosclerosis and with incident cardiovascular events. The aim of this study was to evaluate serum CEC in very elderly individuals free of cardiovascular events and to correlate it with calcium score as index of subclinical atherosclerosis.
Methods. Healthy individuals (n=60) aged 80 years or more, at the 50 th higher (69,5±8,1 mg/dL) and 50 th lower (41,4±9.7 mg/dL) HDL-C levels of the entire healthy cohort (n=208) were selected from The Brazilian Study on Healthy Aging. Serum CEC was measured with radioisotopic techniques by using J774 macrophages treated with a cAMP analogue and exposed to serum of subjects. The Me Multidetector-row cardiac CT for coronary artery calcium score (CACs) was used to detect subclinical atherosclerosis. The presence of not of vulnerable features in the coronary plaques was evaluated by using the Motoyama criteria.
Results. The groups at lower and higher HDL-C levels were of similar age and BMI. The higher HDL-C group was characterized by higher apoA-I, lower triglycerides and lower apoB content. Serum CEC was significantly higher in the whole elders group compared to a cohort of 169 healthy subjects of 35-82 years of age.
Conclusions. In these healthy very elders individuals serum CEC did not significantly correlate with traditional risk factors such as HDL-C, confirming independence of such functional parameter of HDL-C plasma concentrations. Serum CEC did not either associate with CACs as well as with vulnerable features in the coronary plaques. In addition, serum CEC was preserved and even ameliorated when compared with values obtained in younger subjects. These results may be explained by the characteristics of participants that reached old age without manifesting cardiovascular disease.