Atherosclerosis, including lipids and other risk markers

M088

SERUM OSTEOCALCIN FORMS AND OSTEOCALCIN-EXPRESSING ENDOTHELIAL PROGENITOR CELLS ARE INDEPENDENT BIOMARKERS OF CORONARY ATHEROSCLEROTIC DISEASE SEVERITY

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BACKGROUND-AIM

Osteocalcin (OC), an osteoblast-derived regulator of metabolic processes, and circulating early endothelial progenitor cells (EPC, CD34−/CD133+/KDR+) expressing OC (OC+) may link bone metabolism and the vasculature and be involved in the pathophysiology of vascular atherosclerotic calcification. This study aimed at assessing the association of circulating levels of different OC forms and of EPCs count with the severity of disease in patients with documented coronary atherosclerosis (CAD)

METHODS

Patients (n=59) undergoing coronary angiography were divided according to stenosis severity into 2 groups: 1. early coronary atherosclerosis (ECA) (n=22), and 2. late coronary atherosclerosis (LCA) (n=37). Total OC (TOC), carboxylated OC (cOC), undercarboxylated OC (unOC) were quantified by ELISA. EPC OC+ counts, assessed by flow cytometry.

RESULTS

EPC OC+ counts showed significant differences between ECA and LCA groups. unOC and unOC/TOC ratio were inversely correlated with EPC OC+ count. A significant decrease in TOC and unOC plasma levels was associated with greater cardiovascular risk factors (CVRFs) number. EPC OC+ count positively correlated with LDL-C, total cholesterol, and triglycerides, with a greater significance in the LCA group. No association between the different forms of circulating OC (TOC, ucOC, cOC) and severity of CAD was found.

CONCLUSIONS

In this study, a significant association between EPCs (CD34−/CD133+/KDR+/OC+), CAD severity and CVRFs was observed, suggesting an active role for EPC OC+ in the pathogenesis of CAD. An inverse correlation between TOC, ucOC, and number of CVRFs was also observed, suggesting that OC, regardless of its carboxylation status, may be relevant to improve metabolic profile and lower CV risk.

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CORDOVA FORMULA: A BETTER CLASSIFICATION TOOL FOR PATIENTS REQUIRING LIPID-LOWERING TREATMENT

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BACKGROUND-AIM

LDL Cholesterol (LDL-C), being an important risk factor for atherosclerotic cardiovascular disease, is targeted for the initiation of lipid-lowering therapies. The guidelines such as the 2013 American College of Cardiology/American Heart Association Guideline, 2011 European Society of Cardiology/European Atherosclerosis Society Guidelines, and 2012 Canadian Cardiovascular Society Guidelines have recommended varying cut-offs of LDL-C levels for starting the initiation of lipid-lowering therapy. Different formulae had been derived to calculate the LDL-C from other lipid profile parameters to supplant the need for direct estimation. Cordova's, Martin Hopkins', and Sampson's are the newly derived formulae proposed to replace the older calculation methods. We aimed to assess the ability of these formulae to correctly classify the patients who require lipid-lowering therapy when compared with the prevailing Friedewald formula.

METHODS

The analytical cross-sectional study collected lipid profile data from 4096 patients retrospectively. Receiver operating characteristic (ROC) curve, with LDL-C from direct estimation used as classification variable, was plotted at various LDL-C cut-offs such as 70 mg/dL, 75 mg/dL, 100 mg/dL, 115 mg/dL, 130 mg/dL and 190 mg/dL.

RESULTS

The comparison of area under the curve (AUC) of different formulae demonstrated Sampsons, Martin Hopkins, and Cordova to have a significant advantage over the Friedewald formula in classifying patients who require lipid-lowering therapy. Further, a significantly increased AUC was observed for Cordova's formula, when compared with Martin Hopkins', at LDL-C levels of 75 mg/dL (p value=0.0219), 100 mg/dL (p value=0.0038), 115 mg/dL (p value=0.0219), and 130 mg/dL (p value=0.0097). However, at LDL-C of 70 mg/dL, the increased AUC observed for Cordova when compared with Martin Hopkins was statistically insignificant (p-value =0.4901).

CONCLUSIONS

Our study depicts Cordova's formula to be superior to the recently derived Martin Hopkins and Sampson's formulae in classifying patients at LDL-C cut-offs of 75 mg/dL, 100 mg/dL, 115 mg/dL, and 130 mg/dL. Cordova's formula, which is the simplest of all formulae, can better classify patients for lipid-lowering therapy than other formulae.

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INTERACTION BETWEEN HOMOCYSTEINE, SOME ANTIOXIDANTS, LIPIDS AND D-DIMER IN PATIENTS WITH CAROTID ATHEROSCLEROSIS

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BACKGROUND-AIM

: Antioxidants, homocystein, lipids and D dimer are an important risk factors for atherosclerotic diseases . The purpose of this study were find relationship between homocysteine , total antioxidant status(TAS), superoxide dismutase(SOD), glutationperoxidase(GPX) , lipids and D- dimer levels in patients with carotid atherosclerosis.

METHODS

89 patients with ACA (age 59.4±1.7 years) were divided into 2 groups. Group I comprised 51 patients, with hemodynamically insignificant stenosis of CA (<50%) and IMT 0.95±0.04mm. Group II 38 patients with hemodynamically significant stenosis (>50%), IMT was 1.1±0.1mm. Control group 49 healthy patients
Biochemical parameters such are homocystein, TAS, GPX, SOD ,total cholesterol, HDL. And LDL were done on biochemical analyzer COBAS c 311(Roche Diagnostics). D dimer was done on STACompact Max(Stago). Carotid arteries were investigared by extracranial ultrasound on TOSHIBA APLIO 500 (8-12 mgh).

RESULTS

: In group I Correlation between these parameters was positive(from r=0.500 to r=0.628). Homocysteine increased up to 17.0±0.3 mkmol/l. TAS increased up to 2.20±0.03mmol/l, SOD also increased 280.2±0.3E/L. Tchol, LDL were 6.24±0.14 and 4.8±0.15 mmol/l. D dimer 0.9±0.05 mkg/ml. The relation between these parameters and CA stenosis were positive(from r = 0.473, to r = 0.533). HDL was 1.11±0.04 mmol/l and GPX 3423.2±1.1E/L. The correlation between HDL, GPX , CA stenosis and IMT was negative (r= - 0.433,r = - 0.401, r = - 0.452).
In Group II, The homocystein was 24.0±0.3 mkmol/l, TAS level 2.4 ±0.03mmol/l SOD 293.1±0.4E/L, Tchol 6.97±0.04 and LDL 5.63±0.05 mmol/l. D dimer was 1100.9±3.0 ng/ml, Correlation between these parameters and CA stenosis was positive(r= 0.505, r = 0.621). HDL was 0.84±0.02mmol/l, GPX293.1±0.7E/L. Correlation between HDL, CA stenosis and IMT was negative(r = -0.531, r = -0.491).

CONCLUSIONS

Taking into consideration the result obtained, we think it is possible to use positive correlation between lipids, D dimer, homocystein, TAS and SOD, also negative correlation between HDL,GPX and the degree of CA stenosis, as the markers of development of carotid atherosclerosis in patients with carotid atherosclerosis.

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ANALYTICAL ISSUES IN LIPOPROTEIN (A) DETERMINATION AND THEIR IMPACT ON ASSESSING THE THERAPEUTIC EFFECT

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BACKGROUND-AIM

Lp(a) is recognized by professional associations such as ESC/EAS, HEART UK, EAS/EFLM (for LDLC correction) as a cardiovascular disease risk factor. Although Lp(a) molecules have a highly heterogeneous structure and the ratio of mass to molar concentration varies between individuals, recommendations present Lp(a) levels in different units and some additionally suggest arbitrary conversion coefficients. Furthermore, the analytical method and measuring system used can alter the follow-up of the applied therapeutic approach such as apheresis or innovative medications that selectively target Lp(a). The aim of this study was to compare Lp(a) results on two different analytical platforms.

METHODS

This evaluation was carried out as part of the Abbott Alinity CI analyzer's verification process. Results in mg/dl were compared to results obtained by routinely used test calibrated in nmol/l, traceable to the IFCC reference material and performed by Roche Cobas6000cee. Only serum samples with the requested Lp(a) were used. Precision of Alinity CI was tested analyzing two levels of commercial control during five days in pentaplicate.

RESULTS

Coefficients of variation for repeatability 0.97/0.77% and reproducibility 1.25/1.16% meet the eligibility criteria and are below 3.8% which is desirable imprecision for Lp(a) according to EFLM database. In the measurement range of 7-358 nmol/l and 3.1-141.7 mg/dl, excellent comparison (R=0.993; p=0.001) was observed with routine method for 35 patient sera, and Passing Bablok analyses revealed regression equation y=2.55(1.42-3.70)+0.38(0.36-0.39)x, indicating the presence of constant and proportional difference in Lp(a) values. The application of an arbitrary coefficient (2.5) did not improve the regression y=2.53(1.39-3.69)+0.95(0.86-0.98)x, as expected. In four patients, the Lp(a) reduction in percentage after apheresis ranged from 5-70% in nmol/l to 31-67% in mg/dl.

CONCLUSIONS

As is well known, methods that are not standardised should not be used interchangeably. Due to the polymorphic structure of Lp(a), it is not possible to recalculate the results from mg/dl to nmol/l or vice versa. The expected therapeutic effect cannot be easily assessed in all patients with the analytical options available today.

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LP(A) IN PERSONS OVER 40

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BACKGROUND-AIM

Lp (a) is a cholesterol-containing low-density lipoprotein. Recent Epidemiologic studies demonstrated that increasing Lp(a) levels are a heritable, independent risk factor of atherosclerotic cardiovascular disease (ASCVD) and calcific aortic stenosis through its proatherogenic, proinflammatory, and potentially prothrombotic properties. ECS/EAS guidelines recommend Lp(a) measurement at least once in all adults. Current guidelines recommend Lp(a) monitoring in patients with: risk of ASCVD due to familial hypercholesterolemia, a family history of early ASCVD, elevated Lp(a) in the past, and progressive ASCVD despite receiving optimal therapy. This study aimed to analyze serum Lp(a) concentration in patients who are at least 40 years old concerning LDL cholesterol and statin treatment.

METHODS

We analyze serum Lp (a) concentration using immunoturbidometric method (Cobas 6000 analyzer, Test LPA2 II generation, (Roche Diagnostics, Switzerland)) in 225 patients; 156 women (mean age 59 ±9) and 69 men (mean age 59±12).

RESULTS

Average serum Lp(a) concentration in studied group was 27,7 ± 41,3 ng/ml. 76,4% had <30mg/ml which is required value of Lp(a), 4,8 % had Lp(a) concentration between 30-50 mg/dl (moderate risk of ASCVD), 17,3% had 50-180 mg/dl (high risk) and 1,3% of studied persons – only woman - had Lp(a) >180mg/dl, recognized as very high risk of ASCVD. 29,4% (61 pers.) were under statins therapy, and in this group 41,7% have Lp(a)>30mg/dl, and 20% of them have also total cholesterol above 5 mmol/l, 25% in this group have LDL-CH >3.0 mmol/l, and 50 % of population who take statin and have Lp(a) > 30 mg/dl have LDL-CH > 2,6 mmol/l.

CONCLUSIONS

We concluded that the percentage of people with elevated Lp(a) is high in the general population. The high percentage of people with elevated Lp(a) under statin therapy and simultaneously elevated levels of total cholesterol and/or LDL-CH, indicate that there is a need for determination of this particle, which helps to identify people with increased cardiovascular risk.

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RED BLOOD CELL FATTY ACID PROFILE AND BIOMARKERS OF DYSLIPIDEMIA AMONG TESTICULAR CANCER SURVIVORS

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BACKGROUND-AIM

Although testicular cancer (TC) is considered to be a highly curable malignancy, treatment may be associated with non-negligible adverse effects and subsequent risk of cardiometabolic morbidity. The fatty acid (FA) composition of red blood cells (RBCs) is acknowledged as a robust biomarker of long-term FA dietary intake, endogenous metabolic pathways, and tissue status. Unbalanced levels of membrane FAs and dysregulation of desaturase enzymatic activity represent predictive data for cardiovascular risk assessment. This study aimed to characterize the RBC FA composition among TC survivors and to ascertain potential association with serum lipid profile parameters.

METHODS

A sample of patients attending survivorship care after curative treatment for TC was recruited at the Clinic of Urology, University Clinical Center of Serbia. RBC FA panel was determined by gas chromatography and product-to-precursor FA ratios were used as surrogate measures of desaturase activity. Serum levels of total cholesterol (TC), HDL-C, and triglycerides (TG) were directly determined by an automatic biochemical analyzer, while the LDL-C concentration was calculated using the Friedewald formula.

RESULTS

Among participants (n=24, mean age x#=38.54±8.18 years) two-thirds had dyslipidemia and achieved low (<4%) Omega-3 Index (calculated as the sum of 2 prominent long-chain n−3 fatty acids: 20:5n-3 and 22:6n-3) suggesting high cardiovascular risk. Association was determined between TC concentration and RBC C20:3n-6 ($r=0.432$, $p<0.05$), as well as between TG and estimated stearoyl–CoA desaturase-18 activity ($r=0.406$, $p<0.05$). LDL-C correlated positively with C18:0, and inversely with C20:5n-3, Omega-3 Index, and delta-5-desaturase ($r=0.472$, $r=-0.455$, $r=-0.455$, $r=-0.427$, $r=-0.430$, all $p<0.05$, respectively). Higher HDL-C was associated with higher total monounsaturated FA content ($r=0.534$, $p<0.05$), and lower delta-6-desaturase activity ($r=-0.450$, $p<0.05$).

CONCLUSIONS

RBC membrane FA panel provides valuable insight into the pathology of lipid metabolism, in conjunction with serum lipoprotein profile, offering the possibility to monitor dietary intake, membrane incorporation, and biochemical transformations of FAs with an aim to personalize interventions for the prevention and management of lipid disturbances among TC survivors.

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EVALUATING THE CLINICAL IMPACT OF USE NON-FASTING LIPID PROFILE

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BACKGROUND-AIM

Lipid profile is conventionally measured in serum or plasma after fasting. However there are advantages in using non-fasting samples and it is endorsed by several societies and clinical guidelines.
Non-fasting state predominates over a 24 hours period, while fasting state only occurs a few hours. For this reason, fasting lipid profile not reflects the daily average lipid concentrations and cardiovascular risk associated.
The aim of this study was to evaluate the use of non-fasting rather than fasting lipid profile and its clinical impact.

METHODS

Serum was obtained from 52 volunteers. For each one we collected fasting serum and serum at 4 hours after high-fat meal. Previous studies were observed that postprandial serum TG concentration reaches the maximum value at 4 hours after a high-fat meal.
In all fasting and non-fasting samples total cholesterol (TC), high-density lipoprotein cholesterol (HDLC), triglycerides (TG), apolipoprotein AI (ApoAI), apolipoprotein B (ApoB) and lipoprotein(a) [Lp(a)] were measured in AU 5800 (Beckman Coulter). Low-density lipoprotein cholesterol (LDL-C) was calculated by the Fridewald equation.
For each parameter the percentage change (PC%) and the reference change value (RCV) were calculated and compared.
VRC=1.65*CVi/2, being CVi/2 the desirable intra-individual coefficient of variation.
PC%>RCV represents a variation clinically significant (95% statistical confidence).
Estimation of false positive (FP) and false negative (FN) was calculated to evaluate the clinical impact of non-fasting lipid profile.

RESULTS

Comparing non-fasting with fasting lipid profiles, increases in TG (75%), LDL-C (12%), ApoAI (13%) and ApoB (12%) were observed, with no differences in TC, HDL-C and Lp(a).
Concentrations of TG, LDL-C, ApoAI and ApoB showed clinically significant differences (PC%>VRC).
FP was 21,2% for TG, 5,8% for ApoAI and 1,9% for ApoB; FN was 23,1% for ApoAI, 9,6% for LDL-C and 1,90% for ApoB.

CONCLUSIONS

Lipid profile shows differences between fasting and non-fasting states. It is necessary to establishment adequate cut-off values for non-fasting lipid profile in order to minimize FN or FP results.
Published studies comparing fasting and non-fasting populations, while this study is performed in the same individual before and after high-fat meal.

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CLINICAL SIGNIFICANCE OF RISK FACTORS ,BIOCHEMICAL AND LIPID PARAMETERS IN STUDENTS POPULATION

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BACKGROUND-AIM

The term "risk factors" is used to describe genetic, physical and biochemical characteristics, as well as lifestyle factors that could predict increased risk for cardiovascular disease (CVD).
The aim of this study was to perform continuous research of the impact of risk factors on biochemical parameters and lipid status in a population of students at the University of Novi Sad, from 2015 to 2021.

METHODS

1640 students of the University of Novi Sad took part in the study, 817 males and 823 females. These 540 participants were divided into two groups, the control group and the risk group, age matched. The criteria for the selection of participants for the control group were BMI < 25 kg/m^2 and the waist circumference (WC) lower than 94 cm for males and 80 cm for females.
The criteria for the risk group were an increased BMI and/or the waist circumference over these values.
Biochemical, hematological and lipid status analyses were performed in both groups.

RESULTS

The obtained results indicated that there was no statistically significant difference in the mean values for all the examined lipid parameters in the survey groups. The values of lipid status were fairly uniform in both student groups and were within normal limits, although there was an increasing trends in the parameter values in the risk group of students. By comparing hematological and biochemical parameters which were monitored in both groups, it was found that there were statistically significant differences in certain parameter values: Hemoglobin in whole blood, fibrinogen, creatinine and uric acid level, the activity of the enzymes ALT and GGT. All hematological and biochemical parameters were higher in the risk group, but still within normal limits. A further comparison of other biochemical parameters showed that there was significant difference between two groups in the level of SE, the concentration of fibrinogen in the blood plasma, the concentration of glucose, urea and total serum protein.
By comparing the lipid status of all participants in research, it was determined that only the HDL- cholesterol value was significantly higher in the new group of respondents during the testing time. By comparing the values of the parameters of lipid status between the control group and risk group, significantly increase of the total cholesterol, LDL-cholesterol, IA and FR was found.

CONCLUSIONS

After the continuous analysis of the impact of risk factors on biochemical parameters and lipid status of the respondents, it was concluded that the student population of the University of Novi Sad is at risk for cardiovascular disease later in life. All this indicates the importance of early diagnosis of all precursors of atherosclerosis, early prevention and modification of all risk factors.

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COMPARISON OF THREE EQUATIONS FOR CALCULATED LDL CHOLESTEROL

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BACKGROUND-AIM

Friedwald equation for estimation of LDL cholesterol (LDL-C) is inaccurate when triglyceride (TG) concentration is high. Martin equation introduced in 2013 and Sampson equation published in 2020 are more accurate in wider TG concentration range. We aimed to compare three equations for LDL-C with direct LDL-C (dLDL-C) measurements and to test if there is a difference in patient assessment according to recommended LDL-C values for cardiovascular disease (CVD) risk categories.

METHODS

We retrieved data collected in 2020 for total cholesterol (TC), HDL cholesterol (HDL-C), Friedwald calculated LDL-C (fLDL-C), triglycerides and dLDL-C if available, from laboratory information system of Clinical department of laboratory diagnostics, Clinical hospital centre Rijeka, Croatia. TC, TG and dLDL-C were measured on Roche cobas 6000 c501 (Roche diagnostics, Mannheim, Germany) and HDL-C was measured on Beckman Coulter AU480 (Beckman Coulter, Brea, USA). We calculated LDL-C using Martin (mLDL-C) and Sampson (sLDL-C) equation. Passing-Bablok regression analysis for LDL-C was done using data with available dLDL-C. All calculated LDL-C were categorized according to recommended values for CVD risk categories into three groups: LDL-C<1.8 mmol/L very high and high risk, 1.8-3.0 mmol/L moderate and low risk, >3.0 mmol/L patients with no risk. We used X2 test for testing the difference in risk patient assessment. We used MedCalc statistical software (MedCalc Software, Ostend, Belgium). P<0.05 was considered significant.

RESULTS

Out of 10784 patients with LDL-C data, 184 had dLDL-C. Of those, we retrieved 50 patients assuring wide TG range (0.6-9.5 mmol/L) for regression analysis. Regression equations were: fLDL-C vs dLDL-C Y=-0.885(-1.610-(-0.200))+1.197(1.000-1.400)X; mLDL-C vs dLDL-C Y=0.100(-0.191-0.463)+1.000(0.875-1.091)X; sLDL-C vs dLDL-C Y=-0.175(-0.740-0.094)+1.042(0.941-1.200)X. The difference in categorization of LDL-C using cut-off values for patients with CVD risk between three calculated methods was performed on all 10784 data (X2=53.932, P<0.001).

CONCLUSIONS

Friedwald equation has a constant error and Martin and Sampson equation are in agreement with dLDL-C measurement. Different LDL-C calculation method can influence assessment of patients with CVD risk.

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TOWARDS STANDARDISATION OF LIPOPROTEIN (A) MEASUREMENTS: COMMUTABILITY ASSESSMENT OF CANDIDATE REFERENCE MATERIALS USING A LC-MRM-MS CANDIDATE REFERENCE MEASUREMENT PROCEDURE AND COMMERCIALLY AVAILABLE IMMUNOASSAYS

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BACKGROUND-AIM

The lack of global standardisation of current immunoassay-based measuring procedures (MPs) for lipoprotein(a) (Lp(a)) leads to confusion in the care of patients with elevated Lp(a) levels. Traceability to a common higher order reference would ensure high quality and long-term equivalence of measurement results obtained by different MPs. Towards this goal, a higher order reference measurement system is under development by the IFCC working group on quantification of apolipoproteins by mass spectrometry. This system will be based on a peptide-calibrated reference measurement procedure (RMP) measuring apolipoprotein(a) (apo(a)), the specific apo of Lp(a), and the development of secondary serum-based reference materials (RMs) with certified apo(a) content. Efficient implementation of measurement standardisation, based on the concept of metrological traceability, requires a sufficiently close correlation between results of the different MPs in the traceability chain, as well as a good commutability profile of the RM(s) intended for use as common calibrator(s).

METHODS

We have performed a correlation study between the MS-based candidate RMP (cRMP) and eight immunoassay-based MPs. A panel of 39 clinical samples (CS) from individual donors that cover the Lp(a) concentration range was measured with each MP. We have also investigated the commutability of 14 different candidate RMs that included unspiked human serum pools and serum pools spiked with recombinant apo(a) isoforms to select a suitable RM format for the future development of a Lp(a) certified RM.

RESULTS

Comparison of immunoassay-based MPs with the cRMP for measurements of CS in nmol/L demonstrated a good linear correlation, but showed significant measurement bias and sample specific differences. Upon analysis to determine the effect of apo(a) isoform sizes (different KIV2 numbers) on measurement bias, we found that samples with larger apo(a) isoforms tended to have a higher bias than samples carrying smaller isoforms. Five out of seven unspiked human serum pool candidate RMs were commutable for all combinations of the cRMP and MPs expressing results in nmol/L. In contrast, none of the seven candidate RMs based on recombinant apo(a) isoforms spiked into serum pools fulfilled the commutability criterion for all MP – cRMP combinations.

CONCLUSIONS

Correlation between the cRMP and current Lp(a) immunoassay-based MPs is sub-optimal due to, in part, the sensitivity of immunoassay-based MPs to KIV2 numbers resulting in sample specific differences. The results of this study demonstrate that unspiked human serum pools are good candidates for future use as certified RMs. The production of commutable certified RMs by pooling serum units with known apo(a) concentrations and specific apo(a) isoforms could greatly help Lp(a) MP standardization. However, to meet the clinical performance goals, it may still be necessary to redesign the current Lp(a) immunoassay-based MPs.

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ESTABLISHMENT OF AN SI-TRACEABLE REFERENCE MEASUREMENT SYSTEM FOR SERUM APOLIPOPROTEINS (A), A-I, B, C-I, C-II, C-III AND E

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BACKGROUND-AIM

Reference measurement systems (RMS) are essential to ensure IVD-manufacturers, clinicians and lab professionals of accurate test results. Apolipoproteins (apos) are increasingly recognized as relevant functional biomarkers for cardiovascular risk assessment, and new insights and therapies enhance their clinical measurement. Former WHO-IFCC RMS for apos A-I, B and Lp(a) are no longer available. Hence, there is an unmet need for a higher order, globally available, RMS for conventional and emerging apolipoproteins. Here we present the performance characteristics of the established candidate reference measurement procedure (cRMP).

METHODS

A common mass spectrometry (MS) based cRMP was developed using bottom-up proteomics. Proteins in a small volume of serum are denatured, cysteine residues are reduced and alkylated prior to digestion with Lys-C and trypsin. Proteotypic peptides, representing their apolipoprotein, are quantified relative to stable isotope labelled synthetic peptides. Standardization was provisionally done with serum-based, commutable calibrators. The method is provisionally validated according to CLSI guidelines.

RESULTS

To increase analytical sensitivity, two to four peptides were selected per apo for quantitation and confirmation, and characteristic peptides were selected for apoE phenotyping. A 2-step digestion improved peptide yield and ideally results in equimolar digestion. Interpeptide agreement within individual apos showed Pearson's R ranging between 0.908 (apoC-I) to 0.991 (apoC-II). The quantitation of apo(a) was independent from size polymorphism and linear over a range of 3.4 to 450 nmol/L; the LoQ for apo(a) was determined to be 3.4 nmol/L. Total imprecision for apo(a) was 10.1, 8.5, 10.2, 10.4 and 9.7%, at concentrations of 49, 8.4, 16, 252 and 364 nmol/L. Average total imprecision for other apos was between 3.7 and 7.4%.

CONCLUSIONS

A next generation, 7-plex, proteomics-based cRMP has been developed that fulfils analytical performance requirements and allows molecular measurement of targeted apos. Peptide-based calibrators are developed to accomplish SI-traceability. Combining peptide based calibration and cRMP will provide a higher order RMS that enables SI-traceability of serum apo test results in the nearby future.

Atherosclerosis, including lipids and other risk markers

M099

EFFECT OF LIPOPROTEIN (A) IN LDL ANALYTICAL DETERMINATION AND ITS INFLUENCE IN THE FARMACOLOGICAL TREATMENT WITH ATORVASTATIN

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BACKGROUND-AIM

Low-density lipoprotein (LDLc) and lipoprotein (a) [Lp(a)] have been associated with the progression of aterosclerosis and related to traditional cardiovascular risk factors. Certain studies have shown that the LDLc concentration calculated from Friedewald equation is gradually overestimated as the Lp(a) concentration increases. On the other hand, statins have an LDL-lowering effect, however, its effect on Lp(a) metabolism is not well understood. There are few studies that reflect the effect of Lp(a) concentration on the effectiveness of statins on LDLc concentration.
The aim of this study is to examine the effect of serum Lp(a) concentration on the Friedewald equation for estimating LDLc and also analyze the influence of Lp(a) concentration on the efficacy os statin Atorvastatin LDLc-lowering effect.

METHODS

To study the effect of serum Lp(a) concentration on the Friedewald equation for estimating LDLc and to study the influence of Lp(a) concentration on the efficacy of statin Atorvastatin LDL-lowering effect we analyzed total cholesterol (TC), hig-density lipoprotein (HDLc), triglycerides (TG), Lp(a) and LDLc levels of 340 and 107 samples respectively. In the latter case we perform pre-tratment and post-treatment measurements, establishing 3 groups of patients according to the dose of Atorvastatin administered (10/20 mg/day, 40mg/day and 60/80mg/day).

RESULTS

We found a significant positive correlation between the plasma Lp(a) concentration and the percentage overestimation of LDLc ($r=0.960$, $p<0.001$) calculated according to the Friedewald formula. We found a significant negative correlation between the plasma Lp(a) concentration and percentage decrease in LDLc ($r=-0{,}500$, $p<0{,}001$); however, we found no correlation between Lp(a) concentration and percentage decrease in corrected LDLc ($r=0.186$, $p>0.05$), by subtracting the cholesterol portion of Lp(a) (estimated as 0.3xLp(a) according to the Dahlen ecuation), in patients treated with Atorvastatin.

CONCLUSIONS

We have verified that the LDLc concentration obtained from the Friedewald equation includes Lp(a) cholesterol, and the decrease of LDLc in patients receiving Atorvastatin depends only on the cholesterol present in LDLc and not cholesterol from Lp(a).

Atherosclerosis, including lipids and other risk markers

M100

GEOSTATISTICAL STUDY OF SEVERE DYSLIPIDEMIA FOR CARDIOVASCULAR PREVENTION IN PRIMARY CARE

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BACKGROUND-AIM

Geostatistical study of severe dyslipidemia for cardiovascular prevention in primary care.
Cardiovascular diseases are highly prevalent in our environment. Cardiovascular prevention is one of the main objectives in primary care. Geographic information studies can be of great help in primary prevention and even more so in secondary prevention.
The objective of this study is to quantify the prevalence of patients with severe dyslipidemia in selected health areas and their geolocation for the implementation of cardiovascular prevention strategies in primary care.

METHODS

We conducted a retrospective cohort study with laboratory data for triglyceride, LDL cholesterol and Lp(a) levels, subsequent to the incorporation of biochemical algorithms during 2019 and 2020. Geographic clustering clusters by zip codes for each parameter were studied along with chorolectic map representation.

RESULTS

The analytical data included in the study were Triglycerides (n = 301,069), LDL (n = 91316) and lipoprotein a Lp(a) Seville (n = 667). The areas with the highest and lowest percentage of cases for each parameter were identified. Two grouping clusters with statistical significance were detected, one for patients with TG levels>150 mg/dL of 16.47 km radius and another for patients with LDL>190 mg/dL of 6.23 km, with a relative risk of 1.08 and 1.23 respectively.

CONCLUSIONS

Studies using geolocation have been widely used in infectious diseases due to the need to know the epidemiology of diseases with a rapid capacity to spread. However, they have not been used as much in other pathologies that occur silently, such as those related to cardiovascular risk. These new tools can help control severe dyslipidemia and improve cardiovascular prevention in primary care.

Atherosclerosis, including lipids and other risk markers

M101

HIGH NEUTROPHIL-LYMPHOCYTE RATIO AND LOW LYMPHOCYTE-MONOCYTE RATIO COMBINATION AFTER THROMBOLYSIS IS A POTENTIAL PREDICTOR OF POOR FUNCTIONAL OUTCOME OF ACUTE ISCHEMIC STROKE

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BACKGROUND-AIM

Ischemic stroke is one of the leading causes of death and disability. An inflammatory response is observed in multiple stages of cerebral ischemia, particularly in the acute phase. Recent publications revealed that neutrophil-lymphocyte ratio (NLR) and lymphocyte-monocyte ratio (LMR) may be used to predict long term prognosis in acute ischemic stroke (AIS) after thrombolysis. To test whether there is a relationship between the combination of these parameters and long-term prognosis, we analyzed NLR-LMR combination in AIS patients treated with intravenous recombinant tissue plasminogen activator (rtPA).

METHODS

The study included 285 adults with diagnosis of AIS and rtPA treatment within 4.5h time window. Blood samples were obtained at admission and 24h after thrombolysis to calculate pre- and post-thrombolysis NLR and LMR. Clinical data including NIHSS was registered on admission and day 1. Long-term outcome was defined 90 days post-event by the modified Rankin Scale (mRS). Therapy-associated intracranial hemorrhage (ICH) was classified according to ECASS II. Receiver operating characteristic curve (ROC) analysis was performed to determine optimal cutoffs of NLR and LMR as predictors of therapy outcomes.

RESULTS

Patients were stratified by cutoffs of 5.73 for NLR and 2.08 for LMR. Multivariate logistic regression model including all possible confounders displayed no significant association of NLR or LMR with 3-months functional prognosis. The combination of high NLR-low LMR in patients vs. low NRL-high LMR in patients as obtained 24h after thrombolysis was found to be an independent predictor of poor 3-months functional outcome (mRS 2; OR 3.407, 95% CI 1.449 to 8.011, $p = 0.005$). The proportion of patients between low NLR-high LMR and high NLR-low LMR groups from admission to day 1 showed no significant change in the good outcome group. On the other hand, in the poor outcome group (mRS 2), low NLR-high LMR and high NLR-low LMR groups displayed a significant shift of patient proportions from 67% and 21% at admission ($p = 0.001$) to 36% and 49% at 24h after thrombolysis ($p < 0.001$), respectively.

CONCLUSIONS

Our study demonstrated for the first time that a high NLR-low LMR combination as observed at 24h after thrombolysis can serve as an independent predictor of 3-months poor outcome in AIS patients. This simple and readily available data may help clinicians to improve the prognostic estimation of patients and may provide guidance in selecting patients for intensified care post-thrombolysis.