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Original Research Article

AN EVALUATION OF SERUM HIGH-DENSITY LIPOPROTEIN CHOLESTEROL LEVELS AND THEIR ASSOCIATION WITH CLINICAL OUTCOMES IN PATIENTS WITH SEPSIS

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ABSTRACT

Background: Sepsis remains one of the leading causes of mortality in the intensive care units (ICUS) worldwide, despite advances in antimicrobial therapy and critical care management. The wide range of clinical manifestations and prognosis emphasizes the need to identify helpful and affordable prognostic biomarkers. Beyond transport into lipid, the high-density lipoprotein cholesterol (HDL-C) possesses anti-inflammatory, anti-oxidant and endotoxin-inactivating qualities, which can lead to evident interest in foretelling sepsis. The objective was to determine the association between serum HDL cholesterol levels and the disease severity, organ dysfunction, and the clinical outcomes in patients with sepsis

Materials and Methods: This was a prospective observational study done for 12 months in the wards and intensive care units of General Medicine of a tertiary care teaching hospital in Central Kerala. The participants were adults (>18 years) and satisfying the Sepsis-3 criteria (SOFA score>2). The serum HDL-C levels were measured on Day 1 (24 hrs of admission) and Day 5. The clinical outcomes like mortality rate in the hospitals, mechanical ventilation, inotropic support, renal replacement therapy (RRT), and days of stay in the ICUs were recorded. Statistical analysis included student t -test, Chi-square test, Pearson correlation and receiver operating characteristic (ROC) curve.

Results: 194 patients with sepsis were included with mean age 58.6 ±11.6 years. The result showed significant difference between non-survivors and survivors at the time of admission in the mean value of HDL-C level (27.52 ± 3.53 mg/dL versus 41.47±7.65mg/dl respectively; $p < 0.001$). On Day 5, HDL-C levels in non-survivors decreased to 16.27±5.26mg/dL whilst the level increased in survivors 47.92± 6.13mg/dL. The HDL-C and SOFA scores at the admission ($r = -0.67$) and at Day 5 ($r = -0.76$) had a significant negative correlation between them. An HDL-C cut off value of 29.5mg/dL was identified to relate survival with sensitivity of 95.5% and specificity of 80% (AUC =0.965).

Conclusion: Lower serum HDL-C has a strong relationship with critical sepsis, organ support, prolonged ICU stay and in-hospital mortality. The HDL-C is a free and readily available clinical useful prognostic biomarker of sepsis.

Keywords: Sepsis; HDL cholesterol; Prognostic biomarker; SOFA score; Critical care.

INTRODUCTION

Sepsis is one of the life-threatening syndromes resulting from a huge dysregulated host response to various infection, leading to the acute organ dysfunction as well as high mortality. An excessive death rate caused by sepsis is not acceptable, despite the standardized care regimens and early goal-

oriented therapy, and more so in countries with low and middle-income. There is a need to have the at-risk patients identified at an earlier stage.
Such current prognostic tools as SOFA and APACHE II indices are practical, still, they are laboratory-sensitive and might not readily be applied on a board-rundown basis.[1]

Sepsis is one of the complex and life-threatening clinical syndrome that mainly arises when the body's immune response to the infection becomes dysregulated, leading to that for the widespread inflammation, tissue injury, as well as the organ dysfunction.[2] It is a major morbidity and mortality load all across the globe particularly in the critically ill patients. Septic shock is the most perilous form of sepsis, it is marked with severe cellular and metabolic dysfunctions as well as circulatory collapse which contributes extensively to death. Sepsis-2 definitions of 2001 incorporated additional clinical and laboratory criteria such as hypoxemia, oliguria, altered mental state, high-level and lactate, and coagulation dysfunction.[3]
Organ dysfunction operationalization and Sequential Organ Failure Assessment score with two or more points was adopted in Sepsis 3 in 2016; a change associated with a high probability of death.[4]
Preliminary assumptions established sepsis as the effects of excessive inflammatory response or otherwise known as the cytotoxic storm which was largely the activity of cytotoxic mediators and they comprised tumor necrosis factor-alpha and interleukin 1.[5] Subsequent research indicated that sepsis go along with the existence of inflammatory pathways, anti-inflammatory pathways, metabolic pathways, and coagulation pathways. The neutrophils, in sepsis, belong to the centre of the innate immune. The extreme effects in emergency granulopoiesis in severe infections is the neutrophil release in circulation as immature neutrophils.[6]
Sepsis is frequently well associated with the profound coagulation abnormalities and highly associated with bad results.[7] Immunosuppression is a significant component of sepsis, and potentially can be comorbid to systemic inflammation at the onset of the disease. Sepsis is associated with an increase in the rate of immune cells death, reduction in antigen deliverance, and cytokine synthesis defects.[8]
The common cardiac dysfunction associated with sepsis is typically reversible myocardial depression, dysfunction of the diastole and arrhythmias.[9] Respiratory failure (typically acute respiratory distress syndrome) is the most frequent outcome of sepsis that is characterized by the damage of the alveoli, a conspicuous rate of pulmonary permeability, and severe hypoxemia .[10]
The role of high-density lipoprotein cholesterol is beyond the lipid and cardiovascular protection. Reverse transportation of cholesterol, anti-oxidant effect, inflammatory homeostasis, neutralization of endotoxins, and immune/regulation are the functions of HDL.
The drop in HDL cholesterol level is extremely rapid in sepsis which unveils the alterations in hepatic production, degradation, and redistribution of cholesterol to the acute-phase reactants. The reduced balances of HDL level are effective in inhibiting the clearance process of endotoxins and increasing the inflammatory response and improving disease progression.[11] A growing body of evidence exists to show that low HDL-C is associated with high severity, dysfunction of sepsis, and mortality hence it may prove suitable prognostic biomarker.
Experimental and clinical researches have also established that there is a drastic lowering of levels of HDL-C during acute inflammatory processes especially sepsis. However, the Indian populations lack prospective information of the same. The association between the severity of diseases, malfunctioning of the organs, and mortality of sepsis patients with the level of serum HDL-C is evaluated in this study.

MATERIALS AND METHODS

This was a prospective observational study done in General Medicine and Intensive Care Unit of a tertiary care teaching hospital in Central Kerala for a period of one year. The Institutional Review Board approval was obtained before initiating the study. All adult patients diagnosed with sepsis aged eighteen years and above who were hospitalised the study site during the study period were included Sepsis was ascertained in regard to the official Third International Consensus Definitions of Sepsis and Septic Shock also referred to as Sepsis-3. Based on this definition, sepsis may be described as life-threatening dysfunction of the organ functions as a result of dysregulatory host response to bacteria whereby dysfunction of the organs is determined by acutely escalating the Sequential Organ Failure Assessment score by more than two points compared to their baseline.[4]
A consecutive strategy was used in the recruitment to eliminate the selection bias as well as providing the sample in the study as an estimate of the actual population in a real-life clinical practice. Patients who had different levels of illness severity and different sources of infection were included.Patients receiving statin, cancer patients, those with chronic liver disease or chronic kidney disease, those with diabetes mellitus and thyroid conditions, pregnant women, those with malabsorption syndromes and chronic gastrointestinal illness were excluded. A standardized and structured pro forma was used to conduct the data collection. At admission demographic data such as age and sex were captured. The clinical history, comorbid conditions like chronic respiratory disease, High blood pressure, and previous lung-tuberculosis, alcohol use, and neurological disorders and other chronic illnesses were recorded. Laboratory findings, signs of microbiology examination and imaging studies were

noted. Routine infections such as respiratory tract infections, urinary tract infections, soft tissue and skin infections and intra-abdominal infections were mentioned. A proper physical examination was performed and this included examination of vital signs, systemic examination. Estimation of neurological condition assisted by use of the Glasgow Coma Scale and renal function was observed by urine output. Complete blood count, testing of renal functions, testing of liver functions, and analysis of the arterial blood gas were included in the investigation as the baseline. Serum levels of high-density lipoprotein cholesterol were determined by standardized enzymatic methods The Sequential Organ Failure Assessment score was used to rate each patient on Day 1 and Day 5 according to the standard criteria applied to six organ systems, respiratory, cardiovascular, neurological, renal, hepatic, and coagulation systems. The follow-up of the patients was till discharge or in-hospital death. The clinical outcomes that were measured included survival, intensive care unit hospitalization, mechanical ventilation, inotropic or vasopressor therapy, renal replacement therapy and ICU stay.

Statistical Analysis: All the data available was keyed into the Microsoft Excel and the analysis was performed with the Statistical Package of Social Sciences 20. Continuous variables were summarized by use of mean plus standard deviation and frequency and percentages used to summarize categorical variables.

A comparison was conducted in the form of survivor and non-survivor groups by the independent Student t-test of continuous variables and Chi-squares of categorical variables. To find out the correlation that the level of serum HDL cholesterol has with the severity of the disease, Pearson correlation analysis was employed to identify how the HDL-C level correlates with SOFA scores on Day 1 and Day 5.The receiver operating characteristic curves of the analysis were conducted in order to determine the optimal cut-off of the prognostic HDL-C to determine the survival as well as the adverse prognosis. The level of statistical significance at which the cut-off was established was 0.05.

RESULTS

Baseline Characteristics: A total of 194 patients diagnosed with the sepsis were included in the study of which 107(55.2%) were female patients and 87 (44.8%) males. The mean age was 58.6±11.6 years and age range of 18 and 79 years. The patients mainly belong to older age groups with most of the patients being between the 61-70-year of age bracket as older patients are highly vulnerable to sepsis and other associated complications.[12] The demographic data indicate that the vulnerable patients to sepsis are the aged patients which is predictable considering the reduced immunological functions and the rise in the comorbidity rate with age.

Respiratory tract infection was the most common etiology of sepsis of 59.3 % followed by urinary tract infections with 24.2 %. Intra-abdominal infections and skin and soft tissue infections had lower proportions. This distribution emphasizes the situation of prominent prevalence of pulmonary and urinary infections among the causes of sepsis among patients under hospital admission, particularly tertiary care units. This is attributed to the fact that the prevalence of respiratory infections is very high which can be attributed to old age, underlying pulmonary diseases and presentation in the health care facility at an advanced age.

The comorbidities included chronic respiratory disease, systemic hypertension, history of pulmonary TB, and alcohol consumption. Despite the fact that patients who had diabetes mellitus, chronic liver, and chronic kidney disease were excluded, which reduced the confounding impact on lipid metabolism, the other comorbid conditions patients presented with sepsis with a complex clinical picture. Such base features provide an important background in terms of interpreting the clinical outcomes and biochemical changes which occur within the framework of the study.[13]

The overall rate of in-hospital mortality was 31.4% thus suggesting high mortality rate despite routine care. The level of pathology in the patients was revealed by a significant percentage of patients being placed in the intensive care unit, mechanical ventilation, inotropic support, and renal replacement therapy. The prevalence rate of the baseline variables indicates that the study group is representative of patients with moderate to severe sepsis who are admitted in a tertiary care facility.

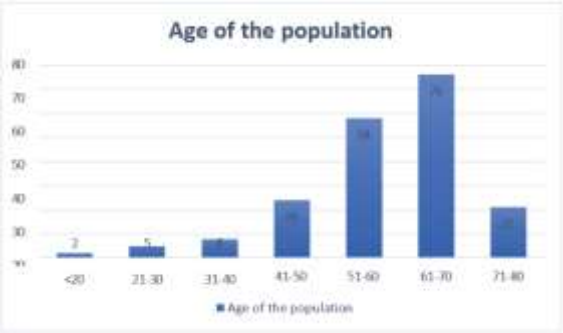

Figure 1: Distribution of age among the study population

HDL-C Levels and Survival: Serum high-density lipoprotein cholesterol levels showed marked differences between survivors as well as the non-survivors at both measured time form of points. The non-survivors had a substantial lower amount of HDL-C at the time of admission compared to survivors. The individual level of HDL-C in the survivors was maintained in comparison to the non-survivors at the point of admission rate, suggesting an initial interaction between the low levels of HDL-C and the adverse occurrence.

There was an increasing trend in the levels of HDL-C over time amid the survivors. On the other hand, the non-survivors had further reduction of their HDL-C suggesting continued inflammatory reaction, worsening of organ functions and disruption of metabolic processes.[14]The dynamics of the HDL-C in sepsis and its potential use as the marker of the disease development and estimating the treatment results are illustrated by the non-convergence tendencies of the two groups.

The dynamicity of the HDL-C changes lies by the fact that low admission statistics may be employed to determine the category of patients that is possibly at risk of decline, and when the levels of the HDL-C levels do not rise throughout the stay, this may serve to indicate the persistence of the illness in the patient and adverse prognosis. The differences between the survivors and non-survivors in the concentrations of the HDL-C at each of the two periods are statistically significant, which demonstrates the hypothesis, that serum HDL-C is highly linked to sepsis responds. These findings are in favor of the likelihood of the HDL-C being a biomarker of sepsis.

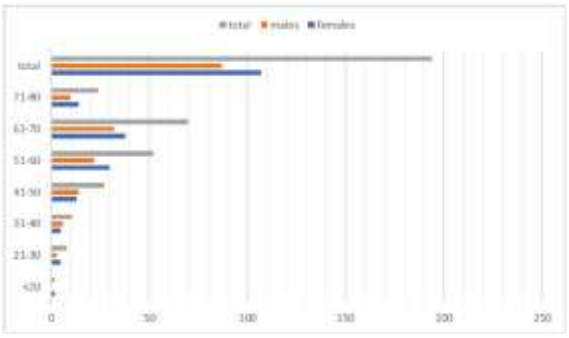

Figure 2: Distribution of Age & sex

Correlation with Disease Severity: The correlation of the HDL-C values with the Sequential Organ Failure Assessment scores was done. On Day 1, these data indicate that HDL-C has a significant baseline correlation with the SOFA scores with a correlation coefficient of -0.67 and that the correlation is found to be strong.It translates to the fact that the low degree of HDL-C on admission corresponded to the increase in the SOFA scores and the organic dysfunction. This relationship confirms a strong correlation between the decrease in the HDL-C and the amount of the physiological injury of sepsis.[15]

This association between the HDL-C levels and the SOFA scores is further exacerbated in Day 5 but the relationship is correlated with the coefficient of -0.76. The degree of correlation was more on Day 5 that indicates the prognostic value of serially collected HDL-C samples.

Poor clinical outcome with the need of mechanical ventilation, inotropic support or vasopressor and renal replacement therapy was correlated extensively with reduced levels of HDL-C. The correlation with inflammation was high and showed that the respiratory failure occurred due to hypoxemia and correlated with the HDL-C levels of the patients who were under ventilatory support in comparison to the patients who were treated without the respiratory support. Patients receiving inotropic management recorded low HDL-C which correlated to cardiovascular dysfunction and poor lipid metabolism. Renal malfunction during sepsis was associated with drastically reduced levels of the HDL-C providing that there was also a need of renal replacement therapy.

Lower levels of HDL-C and high disease burden have been correlated. All these results combined with each other point to the fact that the serum HDL-C level does not only correlate with the mortality but serves as the powerful indicator of the extent of organ dysfunction as well as the need to use rather complex forms of supportive treatment in sepsis.

Table 1: DistributionofpopulationbasedonHDLlevelatDay1andDay5

HDL Level (mg/dL)	Admission (n=194)	Percentage (%)	Day5(n=194)	Percentage (%)
<30	54	27.8%	57	29.38%
>30	140	72.1%	137	70.6%

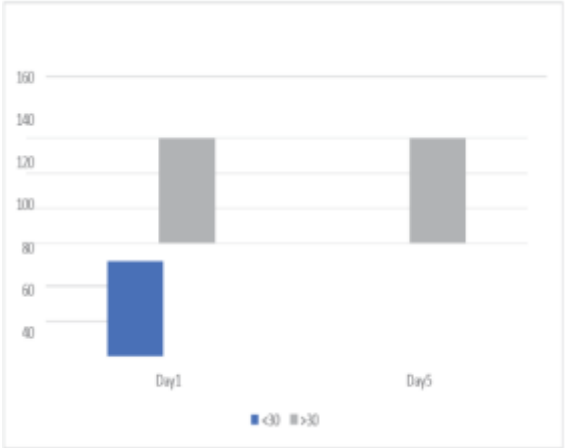

Figure 3: Distribution of Study Population Based on HDL Levels at Day 1 and Day 5

ROC Curve Analysis: The receiver operating characteristic curve analysis was performed for determining the predictive value of HDL-C levels to determine sepsis survivor in the patients. The analysis identified an optimal cut-off of HDL-C of 29.5 mg/dL, which is a predictor of in-hospital survival. This cut-off point had high sensitivity and specificity which have a high discriminatory ability.

The area of ROC curve was 0.965 with a good predictive performance. A substantial value of this reference implies that the admission values of HDL-C are extremely effective as the discriminating variable between survival and non-survival group.The excellent results of the HDL-C in ROC analysis provide strong focus to the possible clinical implications as a prediction biomarker.

The identified cut-off value enhances the generalizability of the HDL-C measurement in the clinical practice more so in the case whereby there are limited resources and the advanced process of scoring may not be readily achievable. These results suggest that HDL-C can be added to the list of the accepted clinical scoring tools to predict and take primary management decisions in the case of patients with sepsis.

DISCUSSION

Heterogeneous clinical presentation, and adverse mortality data in sepsis remains high which demands convincing, easily accessible prognostic biomarkers to help in the initial risk stratification and make clinical decisions.[16] According to the present study, the low serum levels of the high-density lipoprotein cholesterol are highly and significantly associated with a poor patient outcome in sepsis patients which support the idea that HDL-C is a promising prognostic variable.

Among the key findings, the level of HDL-C of both survivors and non-survivors, at admission, and during the hospitalization, differed significantly. The non-survivors had very low concentration of the HDL-C on Day 1 and lesser on Day 5 and conversely the survivors had increase in their HDL-C level with time. HDL-C represents the interplay of changing reactions of inflammation, immune inefficiency, and metabolic alterations during sepsis. The declining HDL-C levels in non-survivors is likely explained by ongoing systemic inflammation, hepatic synthesis arrest and lipoprotein breakdown and relocation of cholesterol to acute-phase products.[17]

The inverse correlation that appears to be extremely close between the HDL-C levels and the scores of the Sequential Organ Failure Assessment also has its contribution to the ongoing debate of the concept that the HDL-C is the marker of the disease severity.[18] The deterioration of organ dysfunction and the mortality risk have a positive association between SOFA scores. HDL-C may involve some additive prospective predictors compared to using a single baseline measure. The specified findings are more precisely applicable to clinical treatment of sepsis, in which the further course of the disease and the response to the treatment of the latter is often crucial as the disease manifestation in itself.

The clinical significance of HDL-C is also highlighted by the correlation between the low level of HDL-C and negative clinical outcomes which include the need for mechanical ventilation, vasopressor support, renal replacement, and prolonged ICU stays. These outcomes are signs of extreme impairment of the organs, and their association with low levels of HDL- C suggests that HDL- C not only predict the mortality but the aggregate morbidity. The patients requiring ventilatory support or inotropes are typically in an extreme respiratory and cardiovascular state and processes that are strongly associated with endothelial injury, systemic vascularity, and microvascular damage, and at which HDL could be a protective factor.

At the mechanical level, a variety of biological pathways can describe the association between the low levels of HDL-C and the unfavourable reaction to sepsis. It has also been understood that HDL contains anti-inflammatory aspects, anti-oxidant aspects, anti-apoptotic aspects and also anti-thrombotic aspects.[19] HDL re wraps endotoxins and transports them to the liver whereby they in turn are eliminated and activates toll-like receptors and downstream inflammatory cascades The reduction in the level of HDL-C influences the process of detoxification and, consequently, increases the duration of activation of immunity and the liberation of cytokines.[20]

HDL possesses the antioxidant property as it prevents the oxidation of low-density lipoproteins and the formation of reactive oxygen species. Oxidative stress is too much in sepsis and results in dysfunction of the mitochondria, endothelium and apoptosis. HDL plays an important role in the maintenance of endothelial integrity by promoting bioavailability of nitric oxide and preventing as well as inhibiting the endothelial activation. Reduction of the HDL-C levels is associated with enhancement of vascular porosity, capillary leak, or inadequate microcirculatory flow, the main components of a septic shock.[21]

HDL inhibits platelet proliferation and prompts fibrinolysis. Reduced levels of HDL-C can potentially cause the development of coagulopathy, microvascular thrombosis, and disseminated intravascular coagulation potentially leading to the further complications in the organ perfusion and functioning.[22] The correlation between low levels of HDL-C with renal replacement therapy, in its turn, can be seen as a consequence of the joint impact of microvascular thrombosis, endothelial dysfunction as well as inflammatory damage, which occur in the renal circulation.

The outcomes of the present research are comparable to the previous international and Indian research studies, which have detected lower concentrations of HDL-C in sepsis and its correlation with the extent of the disease and mortality.[17,23]These clinical observations are further supported by experimental research that demonstrates that HDL supplementation or recuperation of HDL functions can inhibit inflammatory response and increase the survival of inflammatory disease models of sepsis in animals.[24] The overall findings validate the biological feasibility and clinical exploitation of the HDL-C prognostic variable.

In contrast to other emerging biomarkers required to be assessed using special assays or even costly and time-consuming, HDL-C estimation is inexpensive, and could be readily located and performed in most of the hospital laboratories. This makes it especially helpful in resource limited situations whereby

availability of highly sophisticated biomarkers and elaborate scoring processes may be constrained.

Measurement of the HDL-C needs to be added to the sepsis evaluation but it should not replace the existing systems of clinical scoring. HDL-C is a simple biochemical index that is a reflection of many bio pathophysiological processes at the same time. Combination of HDL-C and conventional scoring systems is potentially significant, and could help in the relative prompt risk differentiation, speed in the care escalation, and distribution of care resources.

Limitations

It was conducted in a single tertiary care facility, and thus, it might not be very generalizable to those in another healthcare or a population. The associated differences are likely to impact the observed relationships because of possible variations in the patient characteristics, comorbidity patterns, and practice of treatments. The observational design does not allow the existence of causally related factors between low HDL-C level and adverse outcome.

Multivariate analysis of other inflammatory and metabolic biomarkers such as C-reactive protein, procalcitonin or lactate have not been performed. The long-term outcomes of the post-hospital discharge were not checked, and the prognostic ability of the HDL-C after the acute hospital stay has not been revealed.

CONCLUSION

In conclusion, the level of high density lipoprotein cholesterol in serum is closely related to the extent of the disease, the malfunction of the body organs, and the post-hospital death of sepsis patients. There is a high risk of adverse outcome in patients with low values of HDL-C having cut-off value of 29.5 mg/dl or below. Because of its affordability, high availability, and good prognostic value, HDL-C can be a good complement to the available clinical tools already present with obtaining results in the initial risk stratification, sepsis.

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