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Association Between Serum Ceruloplasmin And Lipid Peroxidation Markers in Patients with Rheumatoid Arthritis

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Abstract

Background: Rheumatoid arthritis (RA) is a chronic systemic autoimmune disease with chronic synovial inflammation, immune system overstimulation and oxidative stress. **Objective**: To evaluate the serum levels of ceruloplasmin and some lipid peroxidation parameters (MDA, 4-HNE and 8-isoprostab) and to assess their correlation with severity of disease and between them in patients with rheumatoid arthritis (RA). **Methods**: This study is a case–control cross-sectional study that included 60 patients who have clinically diagnosed RA and 60 apparently healthy controls recruited from Al—Hilla Teaching Hospital. Patients were classified as having mild, moderate, and severe disease activity groups based on clinical severity assessment. Assessment of serum ceruloplasmin was carried out by enzyme linked immunosorbent assay (ELISA). As described in the Serum MDA, 4-HNE and 8-isoprostane levels were measured with quantitative biochemical immunoassay methodologies. Results: Compared with the health controls, the serum ceruloplasmin, MDA, 4-HNE, and 8-isoprostane levels were significantly higher in RA patients ($P < 0.01$). All studied biomarkers showed a stepwise increase in their concentrations from mild to moderate to severe RA; the highest concentrations were detected in the severe RA group. Ceruloplasmin had was a moderate positive and statistically significant correlation between ceruloplasmin and 8-isoprostane ($r = 0.368$, $P = 0.002$). **Conclusion**: Serum ceruloplasmin and lipoperoxidation biomarkers were significantly increased in RA patients and correlated directly with disease severity. The presence of a significant relationship between ceruloplasmin and the concentration of 8-isoprostane can suggest a nexus between the antioxidant response and lipid peroxidation in the pathophysiology of RA.

Keywords: RA, ceruloplasmin, MDA, 4-HNE, Isoprostanes

Introduction

Rheumatoid arthritis (RA) is a systemic autoimmune disorder characterized by inflammation of joints. It also includes destructive and progressive cartilage destruction, bone erosion and multiple extra-articular manifestations affecting quality of life and functional status. Rheumatoid arthritis (RA) is a systemic autoimmune disease characterized by inflammatory arthritis and extra-articular involvement (Chauhan et al., 2023). It is a chronic inflammatory disorder caused in many cases by the interaction between genes and environmental factors, including tobacco, that primarily involves synovial joints. It typically starts in small peripheral joints, is usually symmetric, and progresses to involve proximal joints if left untreated. Joint inflammation over time leads to the destruction of the joint with loss of cartilage and bone erosions, it affects about 0.5–1% of the global population with a higher prevalence in women, and causing significant morbidity, disability and increased cardiovascular mortality (Smolen et al., 2020). In the inflamed tissues, excessive production of reactive oxygen species (ROS) is mainly contributed by activated neutrophils, macrophages, and other inflammatory cells, whereas mitochondrial dysfunction and activation of ROS-producing enzymes further augment the oxidative burden. Therefore, oxidative stress may not only be involved in synovial inflammation but also may lead to tissue damage and the maintenance of the autoimmune response (Kondo et al., 2023; Staal et al., 2023).

Lipid peroxidation is a well-known biochemical consequence of pathological ROS production. Membranes from cells and lipoproteins that are high in polyunsaturated fatty acids are extremely sensitive to oxidative damage that produces unstable lipid hydroperoxides and secondary reactive aldehydes. Products that have been most widely studied in relation to oxidative damage are malondialdehyde (MDA), 4-hydroxynonenal (4-HNE) and F2-isoprostanes (isoprostanes). Different compounds are generated at different points of lipid oxidation and via different pathways, and thus provide complementary information regarding oxidative injury. MDA is the end product of the oxidative degradation of polyunsaturated fatty acids (PUFAs) peroxidation with low reactivity, while 4-HNE is a highly reactive α,β-unsaturated aldehyde which passively bonds to proteins, nucleic acids, and other cellular molecules. Isoprostanes, a collection of prostaglandin-like substances generated from the scratch of free-radical-mediated peroxidation of membrane phospholipids, are respected as fairly unique being in vivo indicators of lipid peroxidation (Kondo et al., 2023).

Recent evidence supports the increased lipid peroxidation in RA. Quiñonez-Flores et al Detecting attention with GSR: A systematic review. All the studies assessing the circulating concentrations of MDA in RA have shown elevation in concentrations as compared to controls. Notably, the MDA concentrations reported in several studies were also positively related to disease activity, which may suggest that oxidative damage represents a measure of inflammatory burden. Both reviews recognized that F2-isoprostanes rank amongst the top lipid peroxidation products studied in RA, and stressed the possible utility of oxidative biomarkers for disease pathogenesis, evolution and maybe for identifying effectiveness of anti-inflammatory therapy. Recent evidence still supports this relationship. Merino de Paz et al. (2024) conducted a large sequential multicenter cohort study of 430 patients with RA, especially investigated serum MDA, another indicator of lipid peroxidation, in relation to clinical manifestation of RA.

Of particular consideration is 4-HNE because, unlike MDA, this aldehyde is more than just a marker of oxidative stress, it can influence cellular signaling and inflammatory processes. Specifically, it can accept proteins and change structural and functional characteristics, which may facilitate the generation or modification of immunogenic molecules. Akgöl et al (2016) conducted a clinical study ans reported that 4-Hydroxy-2-nonenal (4-HNE) is a lipid peroxidation product and serves as a highly reactive endogenous oxidative stress marker. reported that the serum 4-HNE levels were markedly higher in RA patients than in controls. In addition, 4-HNE was greater in patients with established disease and associated with radiographic structural damage, but did not correlate with EULAR-based or conventional measures of disease activity. These results suggest that lipid-derived aldehydes may be related to tissue damage over time rather than merely markers of ongoing inflammation. Moreover, experimental evidence supports the ability of 4-HNE also to conjugate to human serum albumin, which may exist in folded and unfolded conformations after exposure and contribute to altered structural properties, processes involved in immunopathological processes relevant to RA (Khan et al., 2018).

They are another qualitative assessment of lipid peroxidation. Differential from some traditional lipid oxidation assays, F2-isoprostanes are generated directly due to free-radical-mediated oxidation of arachidonic acid, thus regarded as accurate markers of in vivo oxidative stress. In RA, substantially increased 8-isoprostane concentrations in plasma and urine were reported, as well as an association between urinary isoprostanes and disease progression. Additionally, their study showed increased levels of several lipid-derived aldehydes with MDA and 4-HNE, confirming the hypothesis that RA is a global lipid oxidative injury. Therefore, the concurrent measurement of MDA, 4-HNE, and isoprostanes could reflect lipid peroxidation more

comprehensively than just one oxidative marker (Luczaj et al., 2014).

Among the proteins that exist in this oxidizing content, ceruloplasmin (Cp) is particularly well-positioned as an attractive protein candidate due both to its well-characterized antioxidant and acute-phase functions. Ceruloplasmin is a glycoprotein with ruby red hue that carries copper and is the major ferroxidase in the bloodstream. As a synthetase, Holo can facilitate the binding of Fe through iron binding or RT, and inhibits the formation of redox-active iron: by oxidizing Fe^{2+} to Fe^{3+}, Holo prevents Fe^{3+} from participating in oxidation-reduction reaction, in which Fe will act as an ROS generator. This also has many different antioxidant activities but may have opposing roles and is also dependent on its chemical structure and inflammatory microenvironment. Thus, the upregulated ceruloplasmin seen during chronic inflammation could be indicative of a necessary adaptive response for defense against oxidative stress, though in some cases ceruloplasmin may also function as a pro-oxidant (Hellman & Gitlin, 2002; Fox et al., 1995).

This is particularly relevant to RA with regard to ceruloplasmin and lipid peroxida-tion. Initial clinical investigations signaled the significantly higher plasma ceruloplasmin in addition to the higher MDA in the patients with RA marked by the positive correlation between these two measurements (Gambhir et al., 1997). Similarly, Özgüneş et al. Novack et al. (1995) also demonstrated a relationship between plasma MDA and ceruloplasmin activity in RA, suggesting an interaction between the acute-phase antioxidant response and lipid oxidative injury. More generally, these studies of proteins related to trace elements have highlighted that in RA, ceruloplasmin and other antioxidant systems may be modulated and may suggest an imbalance between inflammation, oxidative stress and antioxidant defense (Asemi et al., 2015).

In this regard, for the first time, this study assessed the correlation of serum ceruloplasmin and lipid peroxidation markers such as MDA, 4-HNE and isoprostanes with RA emergence, which may help to shed light on the oxidative pathways behind RA pathophysiology.

Methods

Patients and data collection

This multicenter cross-sectional case–control study was carried out at the Al-Hilla Teaching Hospital in Babylon, Iraq, from 9 September 2025 to 1 April 2026. We enrolled a total of 120 participants, consisting of 60 patients with clinically diagnosed rheumatoid arthritis (RA) and 60 healthy controls. Diagnosis of RA was established based on clinical manifestations, serological findings, inflammatory indices and imaging findings when appropriate according to 2010 American College of Rheumatology/European League Against Rheumatism (ACR/EULAR) classification criteria.

The controls were healthy subjects' frequency-matched to the patient group for age and sex. Controls had no history of autoimmune disease, chronic inflammatory disorders, recent infection, malignancy, renal impairment, or cardiovascular disease and received no antioxidant supplementation in the three months prior to enrollment. The 28-joint Disease Activity Score (DAS28) was used to evaluate disease activity among RA patients and patients were clustered into mild, moderate and severe disease-activity groups corresponding to the result of DAS28 in order to evaluate the level of detection of certain serum biomarkers based on the disease activity.

Inclusion Criteria

Participants were diagnosed with RA per the 2010 ACR/EULAR classification criteria, aged 18–50 years, provided written informed consent, and had no active bacterial or viral infection within four weeks prior to the study. Patients with chronic systemic diseases; malignancy; acute infection or sepsis; other autoimmune or chronic inflammatory disorders; pregnancy or lactation; severe renal or hepatic dysfunction or having used antioxidant supplements in the previous three months were excluded. Clinical and laboratory data (age, sex, BMI, disease duration, treatment history, RF, anti-CCP antibodies, ESR, CRP, and DAS28 score) were collected by a standard data form and from the medical records as close as possible to blood sampling.

Blood Collection and Serum Preparation

After an overnight fast, 5 mL of peripheral venous blood was drawn from each study subject under sterile conditions. The blood samples were then collected into sterile plain tubes and allowed to clot at room temperature for 20–30 minutes. Samples were then centrifuged at 3000 rpm for 10 minutes to obtain the serum.

The separated serum was carefully transferred into sterile Eppendorf and stored at −20°C until used for biochemical analysis. In all cases, serum samples were aliquoted as needed to reduce repeated freeze–thaw cycles and maintain the stability of the markers of interest.

Measurement of Serum Biomarkers

Serum Ceruloplasmin

Quantification of serum ceruloplasmin levels was performed using a Human Ceruloplasmin SimpleStep ELISA® Kit (Abcam, Cambridge, UK; Cat. Merck, [No. ab236718) according to the guidelines of the manufacturer. The pre-coated microplate wells were loaded with serum samples and standards, the appropriate assay reagents, followed by incubation steps. The colorimetric reaction was developed by adding the substrate solution to the wells following washing to remove unbound components. Absorbance was read at 450 nm using a microplate reader. Serum concentrations of ceruloplasmin were derived from a calibration curve and measured in ng/mL

Serum Malondialdehyde (MDA)

Serum MDA levels used to characterize lipid peroxidation were measured via the TBARS Assay Kit (Cayman Chemical, Ann Arbor, MI, USA; Item No. 10009055) following the guidelines provided by the manufacturer. In this assay, the malondialdehyde (MDA) reacts with thiobarbituric acid (TBA) under acidic and high temperature to form a red MDA–TBA adduct. Absorbance was recorded at around 530–540 nm using a microplate reader after completing the reaction. MDA concentrations were determined from the standard curve and expressed as nmol/mL.

Serum 4-Hydroxynonenal (4-HNE)

Serum levels of 4-hydroxy-nonenal (4-HNE) were assessed using a Human 4-Hydroxynonenal ELISA Kit (Abcam, Cambridge, UK; Cat. No. ab287803). The same was carried out using an assay kit as per the manufacturer's instructions. The serum was added to the appropriate wells (standard or sample), along with the correct antibody and assay reagents. Substrate was then added after the incubation and wash steps and absorbance was measured at 450 nm. 4-HNE concentrations were determined from the standard curve, and expressed in pg / mL.

Serum 8-Isoprostane (8-iso-PGF2α)

Serum concentrations of 8-isoprostane (8-iso-PGF2α) were determined with the 8-Isoprostane Express ELISA Kit (Cayman Chemical, Ann Arbor, MI, USA; Item No 516360) following the manufacturer's instructions. As an indicator of free-radical-mediated lipid peroxidation, 8-iso-PGF2α was quantified using the assay. The samples of serum and standards were added to the wells coated with the antibody and performed as per the specified incubation and washing protocol of the manufacturer. Absorbance at 412 nm was measured in the microplate reader following addition of the substrate. 8-Isoprostane concentration is determined from the standard curve and is expressed in pg/mL.

Ethical Considerations

Before conducting the study, the study protocol has been reviewed and approved by the relevant Research Ethics Committee of Al-Hilla Teaching Hospital/Babylon Health Directorate. Prior to enrolment, all participants were informed of the study's purpose and procedures received and signed a written informed consent. All methods were carried out in accordance with the Declaration of Helsinki. Data including all study participants' personal and clinical information were recruited and identified only for research purpose.

Statistical Analysis

Data were analyzed using IBM SPSS Statistics. All continuous variables are expressed as mean ± standard deviation (SD) and categorical variables as frequencies and percentages. An independent-samples t-test was performed to compare mean serum concentrations of ceruloplasmin, MDA, 4-HNE and 8-isoprostane between the RA patients and healthy controls. For categorical variables, the chi-square (χ^2) test was used. Data were further utilized. One-way analysis of variance (ANOVA) was used to compare biomarker concentrations between RA patients grouped by disease activity (mild, moderate, or severe). For the case of statistically significant overall ANOVA, least significant difference (LSD) test was performed to compare each pair of groups (Al-fahham, 2018). Pearson's r was used to evaluate correlations between ceruloplasmin and markers of lipid peroxidation (MDA, 4-HNE, and 8-isoprostane). Statistical significance was defined as a P value of < 0.05 with highly significant defined as < 0.001.

The Results

The baseline demographic and clinical characteristics of RA and control subjects are summarized in Table 1. The biggest percentage of participants belonging to the 36–45-year-old age group was also found among RA patients (33.3%) and controls (31.7%). There was no significant difference of age distribution between the two groups compared ($P = 0.564$). Likewise, females presented as primary gender in both groups, with 68.3% among patients living with RA and 61.7% among controls ($P = 0.341$). With respect to residence, similar proportions of both RA patients (63.3%) and controls (68.3%) resided in urban areas, and the difference was non-significant ($P = 0.548$).

Table 1. Comparison of age, gender and residence between RA patients and control

Items		RA Patients (N= 60)		Control (N= 60)		(P value)
		Freq.	%	Freq.	%	
Age	26-35	14	23.3	16	26.7	0.564
	36-45	20	33.3	19	31.7	
	46-55	16	26.7	15	25	
	> 55	10	16.7	10	16.7	
Gender	Male	19	31.7	23	38.3	0.341
	Female	41	68.3	37	61.7	
Residence	Rural	22	36.7	19	31.7	0.548
	Urban	38	63.3	41	68.3	

The distribution of patients according to disease severity demonstrates that moderate rheumatoid arthritis was accounting for 55% (n=33) of the studied cases, whereas mild disease activity represented 30%, 9 (n=18) and severe disease constituted only 15% (n=9), as shown in figure 1.

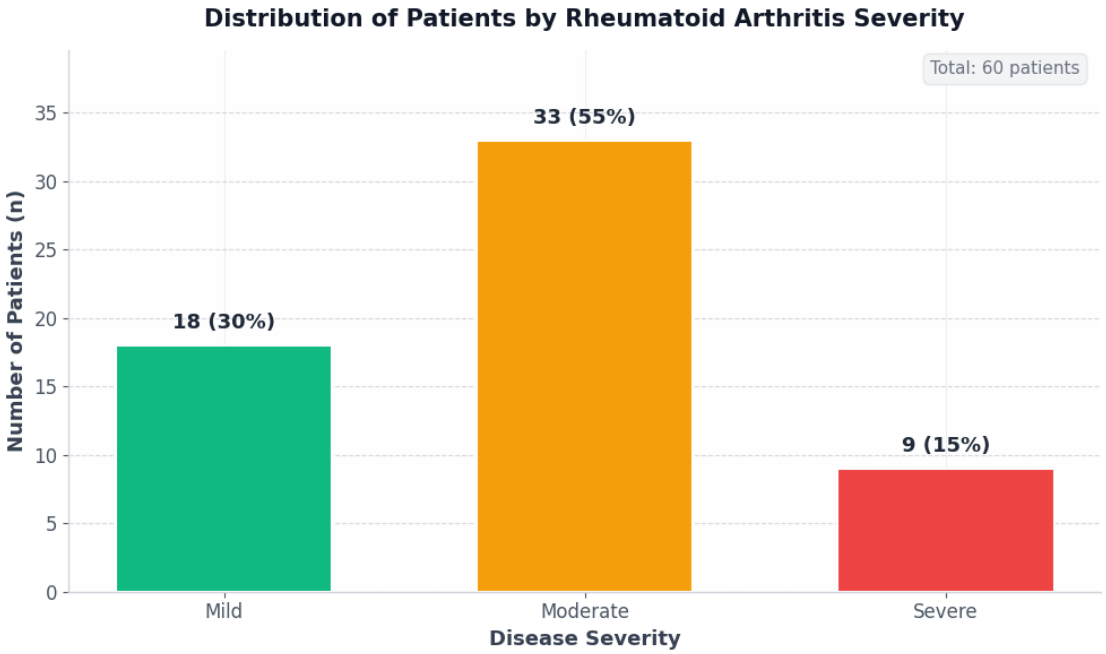

Figure 1. Distribution of RA patients according to the severity of the disease

As shown in Table 2, levels of serum ceruloplasmin and lipid peroxidation biomarkers were significantly elevated among those with Rheumatoid Arthritis compared to healthy controls. It shows that serum ceruloplasmin was higher in RA patients than controls (426.8 ± 76.4 vs. 318.5 ± 61.7 ng/mL; P < 0.002). Similarly, MDA (3.84 ± 0.72 vs. 2.17 ± 0.48 nmol/mL; P < 0.002), 4-HNE (684.6 ± 118.3 vs. 421.7 ± 92.5 pg/mL; P < 0.003), and 8-isoprostane (386.5 ± 74.2 vs. 241.8 ± 51.6 pg/mL; P < 0.002) were significantly elevated in RA patients.

Table 2. Comparison of procalcitonin and oxidative stress between RA patients and healthy control

Biomarkers	RA Patients (N= 60)		Control (N= 60)		(P value)
	Mean	SD	Mean	SD	
Ceruloplasmin (ng/mL)	426.8	76.4	318.5	61.7	< 0.002*
MDA (nmol/mL)	3.84	0.72	2.17	0.48	< 0.002*
4-HNE (pg/mL)	684.6	118.3	421.7	92.5	<0.003
8-Isoprostane (pg/mL)	386.5	74.2	241.8	51.6	< 0.002*

*** High Significant at P value <0.01**

Serum concentrations of all studied biomarkers were progressively increased along with RA disease severity (table 3). In mild disease, ceruloplasmin was 365.4 ± 58.7 ng/mL and was 502.8 ± 71.5 ng/mL in severe disease (P < 0.001). In a like manner, MDA changed from 3.21 ± 0.54 to 4.36 ± 0.68 nmol/mL (P = 0.014), and 4-HNE from 596.3 ± 91.5 to 758.4 ± 119.6 pg/mL (P = 0.012) respectively. The increasing levels of 8-Isoprostane from 318.6 ± 54.8 to 441.7 ± 72.3 pg/mL (P < 0.003).

Table 3. Comparison of biomarkers between RA patients and healthy control

Biomarkers	Mild (N= 18)		Moderate (N= 33)		Severe (N= 9)		(P value)
	Mean	SD	Mean	SD	Mean	SD	
Ceruloplasmin (ng/mL)	365.4 ± 58.7	431.6 ± 63.9	502.8 ± 71.5	365.4 ± 58.7	431.6 ± 63.9	502.8 ± 71.5	< 0.001*
MDA (nmol/mL)	3.21 ± 0.54	3.78 ± 0.61	4.36 ± 0.68	3.21 ± 0.54	3.78 ± 0.61	4.36 ± 0.68	< 0.014*
4-HNE (pg/mL)	596.3 ± 91.5	671.8 ± 105.7	758.4 ± 119.6	596.3 ± 91.5	671.8 ± 105.7	758.4 ± 119.6	<0.012*
8-Isoprostane (pg/mL)	318.6 ± 54.8	379.5 ± 63.7	441.7 ± 72.3	318.6 ± 54.8	379.5 ± 63.7	441.7 ± 72.3	< 0.003*

*** High Significant at P value <0.01**

Table 4 shows that ceruloplasmin had no significant correlation with MDA (r = 0.056, P = 0.182) or 4-HNE (r = 0.046, P = 0.116). In contrast, ceruloplasmin demonstrated a moderate positive and statistically significant correlation with 8-isoprostane (r = 0.368, P = 0.002).

Table 4. Pearson correlation coefficient between ceruloplasmin and lipid peroxidation markers

Markers	Ceruloplasmin (ng/mL)	
	r	P value
MDA (nmol/mL)	0.056	0.182 (NS)
4-HNE (pg/mL)	0.046	0.116 (NS)
8-Isoprostane (pg/mL)	0.368	0.002*

*** High Significant at P value <0.01**

Discussion

This study directly shows the anticipated link between RA and systemic oxidative stress as suggestd by significantly raised serum concentrations of ceruloplasmin and products of lipid peroxidation (malondialdehyde, 4-hydroxynonenal and 8-isoprostane). RA patients had significantly higher levels of ceruloplasmin (426.8 ± 76.4 vs. 318.5 ± 61.7 ng/mL), MDA (3.84 ± 0.72 vs. 2.17 ± 0.48 nmol/mL), 4-HNE (684.6 ± 118.3 vs. 421.7 ± 92.5 pg/mL), and 8-isoprostane (386.5 ± 74.2 vs. 241.8 ± 51.6 pg/mL), compared with healthy counterparts. Together, these results support the notion that oxidative processes are increased during the chronic inflammatory state of RA.

In RA patients, MDA presented significantly higher levels than in other diseases, confirming the role of lipid peroxidation in the pathogenesis of RA. MDA is a significant by-product of polyunsaturated fatty-acid oxidation and is a typical marker of oxidative injury to cellular membranes. Such higher serum or plasma MDA has been previously described in RA patients when compared with controls. For instance, Khadim and Al-Fartusie (2023) demonstrated significantly higher MDA concentrations in 120 women with RA compared to healthy controls and proposed that the oxidative-stress measurements may be relevant for monitoring the severity of RA. More recently, Nabih et al. (2024) found that serum MDA was significantly higher in RA and in addition displayed significant positive associations with ESR, CRP, disease activity score, and morning stiffness, furthering the possible link between indices of oxidative stress and indices of inflammatory disease activity. Similarly, we observed a positive association between serum median values of MDA with ESR and DASH28-ESR in a large cohort study of 430 RA patients, but the associations of MDA with other clinical characteristics were inconsistent (Ruiz et al., 2023).

Increased levels of 4-HNE in the current study further corroborate the increased lipid peroxidation that occurs in RA. In contrast to MDA, 4-HNE is an extremely reactive aldehyde that is mainly produced via the autoxidation of omega-6 polyunsaturated fatty acids and is capable of directly affecting proteins, enzymes and cell signaling pathways. Thus, its accumulation might cause not only oxidative damage but also prolonged inflammation. In agreement with our finding, previous studies reported that serum 4-HNE levels in RA patients were significantly higher than those in healthy controls. In particular, Aydin et al. (2016) reported increased serum 4-HNE in RA and observed higher concentrations in established RA compared to early RA. Notably serum 4-HNE was associated with the modified Larsen score suggesting a link between systemic lipid peroxidation and structural damage in the joint. Similarly, Kędziora-Kornatowska et al. (2015) also found that many lipid-peroxidation aldehydes such as 4-HNE and MDA are elevated in RA, suggesting that lipids are extensively oxidatively modified in the disease.

Such an increase in 8-isoprostane is a strong additional support for the hypothesis of oxidative lipid injury in RA. 8-Isoprostanes are stable products of free-radical-mediated in vivo oxidation of arachidonic acid and are thought to be relatively specific markers of in vivo lipid peroxidation. Studies have previously shown that 8-isoprostanes concentrations are increased in RA. Rho et al. (2009) conducted a study to compare oxidative stress with controls, the authors found that significantly higher 8-isoprostane correlated with increased cardiovascular risk in RA patients and postulated that oxidative stress is not only involved in the increased joint inflammation in RA but also in the increased

cardiovascular burden of the disease. Our earlier studies showed significantly increased levels of F2-isoprostanes in patients with rheumatic diseases such as RA as compared to the normal controls. As is well recognized, oxidative stress contributing to endothelial dysfunction and inflammatory signaling that underlie vascular injury, implicating a pathway between RA and its systemic complications (Mateen et al., 2001).

One of the most important results from this study was that the serum levels of all measurable biomarkers rose stepwise with increasing severity of RA. Specifically, ceruloplasmin increased from 365·4 ± 58·769 ng/mL at low stage disease to 502·8 ± 71·471 ng/mL at high stage disease (-MDA, -HNE and -FOP also showed a similar rise. This pattern indicates that there are increasingly greater oxidative dysfunctions as the disease progresses. The present finding is in agreement with previous work showing that oxidative biomarkers correlate with measures of RA activity. For example, Ruiz et al. Nabih et al (2024) found positive correlation of MDA and DAS28-ESR; MDA was positively associated with clinical indices of disease activity and damage (2024). In addition, the finding that 4-HNE is increased in chronic RA and correlates with structural damage. Nonetheless, inter-study differences indicate that the use of oxidative biomarkers should be interpreted as add-on information to conventional clinical disease-activity measures instead of being replacement markers (Aydin et al., 2016).

Particularly, the increase in ceruloplasmin needs to be discussed. Ceruloplasmin is an essential antioxidant and a copper carrying ferroxidase, an acute-phase protein. Its elevation in RA could be a compensatory response against increased production of reactive oxygen species and inflammatory stimulus. In RA, previous studies have repeatedly shown elevated circulating ceruloplasmin. Scudder et al. (1978) report significantly increased serum ceruloplasmin (S-Cp) and copper in rheumatoid arthritis (RA), while Cogalgil and Taysi (2002) also obtained increased serum Cp in RA patients compared to controls. Moreover, Rajeshwari et al. (1997) also had increased level of plasma ceruloplasmin along with increased MDA and cerulopasmin level had positive correlation with MDA and postulated that increased ceruloplasmin level indicates compensate antioxidant defense due to increased oxidative stress. Therefore, this higher ceruloplasmin observed in the current study can be interpreted as an adaptive increase in antioxidant protection and not as a direct mediator for oxidative injury.

In the same line, ceruloplasmin showed a weak but statistically non-significant correlation with either MDA or 4-HNE, but a moderate positive correlation with 8-isoprostane ($r = 0.368$, $P = 0.002$) (table 3). These selective associations may indicate that ceruloplasmin responds differently towards distinct pathways of lipid oxidation. MDA and 4-HNE are structurally reactive aldehydes depending on various lipid substrates and metabolic pathways while 8-isoprostane is predominantly generated by free-radical-mediated peroxidation of arachidonic acid. And thus our close association of ceruloplasmin and 8-isoprostane may reflect the fact that the handling of ceruloplasmin-related copper/iron and free-radical-mediated lipid oxidation are inextricably linked. However, the absence of robust correlations with MDA and 4-HNE findings show that ceruloplasmin cannot be applied generally as a surrogate marker of lipid peroxidation. This is consistent with previous observations that ceruloplasmin may be an acute phase reactant and an antioxidant response in RA (as opposed to elevations in ceruloplasmin merely paralleling markers of classical inflammatory activity) (Cogalgil & Taysi, 2002; Gutteridge, 1986).

Conclusion

This study showed elevated levels of serum ceruloplasmin and other lipid peroxidation markers (MDA, 4-HNE, and 8-isoprostane) among patients with rheumatoid arthritis, suggesting their involvement in the inflammatory process and the pathophysiology of rheumatoid arthritis. The levels of all studied biomarkers progressively increased with increasing RA disease severity, suggesting their potential association with disease progression. The correlation analysis showed that there was a moderate positive correlation between serum levels of ceruloplasmin and 8-isoprostane.

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