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RESEARCH ARTICLE

# The relationship between serum zinc levels and rheumatoid arthritis activity

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**OBJECTIVES:** Rheumatoid arthritis (RA) is one of the most prevalent chronic autoimmune diseases; it typically involves the hands, wrists, ankles, and eventually all joints. Some studies have reported that zinc serum levels are lower in patients with RA than in healthy individuals.
**MATERIALS and METHODS:** Seventy-nine patients with RA were selected. The patients were all less than 75 years old and were diagnosed by a rheumatologist. Serum zinc levels were measured using the flame photometry method with a blood sample of 5 mL. The relationship between the average serum zinc level and disease activity was analyzed based on the DAS28 scoring scale for different RA groups. The significance threshold was set at $p < 0.05$. Data analyses were implemented in SPSS 22.0.
**RESULTS:** There was a significant inverse relationship between the serum zinc levels and disease activity. Chi-square tests were used to compare zinc serum levels with respect to disease activity. There were significant differences in zinc levels among three groups of patients with different levels of disease severity, such that disease activity increased as the serum zinc level decreased ($p < 0.001$).
**CONCLUSION:** There was a significant inverse relationship between the serum zinc level and RA activity based on the DAS28 score. Therefore, it is recommended that mineral deficiencies should be addressed by optimizing the zinc supply along with other standard medications in order to reduce RA activity.

**Keywords** serum zinc level, rheumatoid arthritis, disease activity score, DAS28

## Introduction

Rheumatoid arthritis (RA) is a chronic systemic inflammatory disease characterized by inflammation of the synovial membrane and progressive destruction of the articular cartilage and bone (Salgado and Maneiro, 2014). The most prominent feature is symmetric joint swelling of the feet, hands, and knees (Huizinga and Pincus, 2010). The progressive disability seriously impacts quality of life (Uhlig et al., 2014). Furthermore, many risk factors have not been established. Some researchers have focused on the role of trace elements, including zinc (Zn) (Onal et al., 2011; Afridi et al., 2012). Zinc is essential for many biological processes in humans. It is an integral functional component of many enzymes and transcriptional regulatory proteins, which play important roles in biochemical processes (Chan et al., 1998). Zinc is a component of nearly 250 human proteins, including angiotensin-converting enzyme inhibitors, alkaline phosphatase, carbonic anhydrase, metallothionein, superoxide dismutase, and DNA and RNA polymerases. It also plays important roles in cell division and apoptosis (Wang et al., 2011, Bonaventura et al., 2015). Zn is influenced by chronic inflammatory responses (Taneja and Mandal, 2009).

Several recent studies have examined the roles of trace elements in the etiology and pathogenesis of RA. Studies have identified decreased plasma zinc levels in patients with RA (Tuncer et al., 1999; Strecker et al., 2013). However, other studies have reported normal or elevated serum or plasma levels of zinc in patients with RA compared to those

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of a control group (Yazar et al., 2005; Mierzecki et al., 2011). In several studies, a relationship between serum levels of zinc and disease activity has not been detected (Li et al., 2014). Owing to the contradictory results regarding the relationship between serum zinc levels and RA activity, the present study aimed to investigate this issue.

## Materials and methods

This cross-sectional study was approved by the Ethics Committee of Ahvaz Jundishapur University of Medical Sciences (AJUMS), Iran (*ajums*.REC.1396.67). The study was carried out from April 2017 to June 2017 and included patients with RA who were out-patients at the Rheumatology Clinic of the Golestan Hospital, Ahvaz, Khuzestan province, Iran. Sampling was performed using a non-randomized sequential method. All participants provided consent for the trial.

### Inclusion criteria

Seventy-nine patients with RA were recruited during their visit to the Rheumatology Clinic of Golestan Hospital and were divided into three groups according to disease stage. Diagnosis of RA was based on ACR 2010 criteria, and all patients were younger than 75 years. All participants were residents of Khuzestan province, Iran.

### Exclusion criteria

Individuals that consumed any form of zinc in the preceding year; individuals < 18 years old or with a history of an autoimmune disease other than RA, malabsorption with clinically relevant symptoms (e.g., chronic diarrhea), malignancy, a glomerular filtration rate (GFR) below 80 mL/min per 1.73 m; pregnant or lactating women; and those residing outside of Khuzestan province were excluded from the study.

### Measurement of serum zinc levels

The serum zinc level was measured using the flame photometry method (Robert Rifle KG Photometer 5010 V5 + Germany) with the Grainer laboratory kit (Germany) by obtaining a blood sample of 5 mL. Zinc was analyzed at a wavelength of 560 nm. The reference range for the serum zinc level was 70–120 μg/mL for both men and women (Williamson et al., 2011). Patients with RA were divided into two groups based on the serum zinc level. The first group had a zinc deficiency (under 70 μg/mL) and the second group had a normal zinc level (between 70 and 120 μg/mL). In the next phase, the relationship between the serum zinc level and disease activity in different groups was analyzed by an expert and a statistician, based on the DAS28 scale.

### Disease activity scoring

RA disease activity was assessed using the DAS28 questionnaire at the time of blood sampling. DAS is a quantitative measure of disease activity used to monitor the treatment of RA. DAS28, i.e., the disease activity score, is calculated using a formula that includes the number of tender or swollen joints (Malaviya, 2003). Data were collected based on clinical examinations performed by a rheumatology consultant, taking into account the number of swollen and painful joints and the erythrocyte sedimentation rate (ESR). ESR is a simple and inexpensive laboratory test for assessing the inflammatory response (Thomas et al., 1993; Jou et al., 2011). The International Committee for Standardization in Hematology (ICSH) recommends the use of the Westergren method (Jou et al., 2011). Data were then analyzed to obtain the DAS28 score for every patient. According to the scores obtained on the DAS28 scale for disease activity, results are presented in Table 1.

### Data collection instrument

All data, including age, gender, medicine consumed by patients, and examination results, were collected using primary data tables. The required examinations were conducted at Golestan Hospital, Ahvaz, Iran, and the same kit was utilized for each examination.

### Statistical analysis

The results were analyzed using Statistical Package for Social Sciences (SPSS) version 22 (SPSS Inc, Chicago, IL, USA). Descriptive statistical methods, including frequency distribution tables, graphs, and central and distribution tendencies, were utilized. Chi square tests were employed to examine the relationships among qualitative variables. For the statistical analysis of normally distributed quantitative variables, Pearson's correlation tests were used. The significance level was set at $p < 0.05$.

## Results

Demographic parameters were evaluated for all participants, including age, gender, and disease activity (Table 2).

**Table 1** Rheumatoid arthritis activity reference

| Instrument | Score range | Remission threshold | Low threshold | Moderate threshold | High threshold |
|---|---|---|---|---|---|
| Disease activity score in 28 joints (DAS28) | 0–9.4 | ⩽2.6 | ⩽3.2 | > 3.2 and < 5.1 | > 5.1 |

**Table 2** Baseline characteristics of participants

| Variables | Frequency | Mean | Standard deviation | Minimum | Maximum |
|---|---|---|---|---|---|
| Age | 79 | 46.25 | 12.313 | 21 | 74 |
| Gender | - | - | - | - | - |
| Male | 12 (15.2%) | - | - | - | - |
| Female | 67 (84.8%) | - | - | - | - |
| DAS28 | 79 | 4.7423 | 1.35410 | 2.61 | 6.88 |
| Zinc level | 79 | 75.442 | 17.7246 | 47.3 | 119.0 |

We used the Chi-square test to compare zinc levels and disease activity among patient groups with mild, moderate, and severe activity. Our results suggested that there is an inverse correlation between serum zinc levels and disease activity; as disease activity increased, the zinc level decreased ($p < 0.001$). Pearson's correlation coefficients confirmed this correlation ($r = –0.741$, $p < 0.001$). Results are summarized in detail in Tables 3, 4, and 5.

## Discussion

Previous reports have indicated that the serum zinc concentration is lower in patients with RA than in healthy individuals (Farid et al., 2000; Wanchu et al., 2002; Ala et al., 2009; Taneja and Mandal, 2009; Afridi et al., 2011; Mierzecki et al., 2011; Ali and Al-Zubaidi, 2012; Afridi et al., 2013; Li et al., 2014; Sahebari et al., 2016; Ullah et al., 2016). However, Yazar et al. (2005) did not observe a significant difference in serum zinc concentrations in patients with RA compared with healthy subjects. A study conducted by Ala et al. (2009) showed that the average serum zinc level was lower in patients with RA than in a control group. Another study conducted by Wanchu et al. showed that serum zinc levels were lower in patients with active RA than in patients with inactive RA (Wanchu et al., 2002). Similar results have been reported in studies carried out in Iraq, Pakistan, and India (Farid et al., 2000; Taneja and Mandal, 2009; Ullah et al., 2016). The relationship between serum zinc levels and disease activity has only been examined in two studies by Mierzecki et al. (2011) and Sahebari et al. (2016). The results of a case-control study performed by Mierzecki et al. (2011) showed that serum, red blood cell, and hair zinc levels in patients with RA were lower than those in the control group, but there was not a significant relationship between the serum zinc level and RA activity. In another study, Sahebari et al.

**Table 3** Relationship between serum zinc levels and disease activity

| | | | DAS28 score category | | | Total |
|---|---|---|---|---|---|---|
| | | | Low | Moderate | High | |
| Zn level ($p < 0.001$) | Low | Count | 1 | 10 | 24 | 35 |
| | | % | 2.9% | 28.6% | 68.6% | 100.0% |
| | Normal | Count | 21 | 18 | 5 | 44 |
| | | % | 47.7% | 40.9% | 11.4% | 100.0% |
| Total | | Count | 22 | 28 | 29 | 79 |
| | | % | 27.8% | 35.4% | 36.7% | 100.0% |

**Table 4** Chi-square test

| | Value | *df* | Asymp. Sig.(2-sided) |
|---|---|---|---|
| Pearson Chi-square | 32.310[a] | 2 | 0.000 |
| Likelihood ratio | 37.193 | 2 | 0.000 |
| Linear-by-linear association | 31.448 | 1 | 0.000 |
| N of valid cases | 79 | | |

a: 0 cells (0%) have expected count less than 5. The minimum expected count is 9.75.

**Table 5** Pearson correlation coefficients

| | | Zn | DAS28 score |
|---|---|---|---|
| Zn | Pearson correlation | 1 | –0.741** |
| | Sig. (2-tailed) | | 0.000 |
| | N | 79 | 79 |

Correlation is significant at the 0.01 level, 2-tailed.

(2016) showed that the serum zinc level in patients with RA was lower than that of healthy group, and there was no significant relationship between disease activity and serum zinc levels. Evidence suggests that the zinc distribution in the body is influenced by inflammation (Yazar et al., 2005). Previous studies have suggested that there is a correlation between the extent of inflammation and serum zinc depletion (Leone et al., 2006; Knoell et al., 2009). Some studies have indicated a protective role of zinc in RA (Leone et al., 2006). Several studies have reported that serum zinc concentrations decrease during the aging process (Ala et al., 2009). All of the above studies compared serum zinc levels in patients with RA and a control group and had smaller sample sizes than that of our study (79 patients) (except for the study by Sahebari et al. (2016), which had a larger sample size) and this could affect the study results.

## Conclusion

In this study, serum zinc concentrations exhibited an inverse correlation with disease activity. Moreover, a significant inverse relationship was observed between the patients that did not report yet. In our study, however, confounding factors that can affect serum zinc levels, e.g., diet and the consumption of zinc supplements, were omitted. The results of our study indicate that there is a significant inverse relationship between the serum zinc level and RA activity based on the DAS28 score, such that disease activity increases as the serum zinc level decreases.

### Limitation

The limitations of this study included the small sample size and single-center design. In this study, samples were selected from a general clinic and may not be representative of the whole population. Additional studies are required with larger sample sizes and wider population sampling.

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## Compliance with ethics guidelines

Elham Rajaee, Karim Mowla, Ali Ghorbani, Mehrdad Dargahi-Malamir, Marzieh Zarei, Faraj Allah Rahimikhah declare that they have no conflict of interest.

## References

Afridi H I, Kazi T G, Brabazon D, Naher S (2011). Association between essential trace and toxic elements in scalp hair samples of smokers rheumatoid arthritis subjects. Sci Total Environ, 412-413: 93–100

Afridi H I, Kazi T G, Kazi N, Shah F (2012). Evaluation of status of zinc, copper, and iron levels in biological samples of normal and arthritis patients in age groups 46-60 and 61-75 years. Clin Lab, 58(7-8): 705–717

Afridi H I, Kazi T G, Kazi N, Talpur F N, Shah F, Naeemullah S S, Arain K D, Brahman (2013). Evaluation of status of arsenic, cadmium, lead and zinc levels in biological samples of normal and arthritis patients of age groups (46- 60) and (61- 75) years. Clin Lab, 59(1-2): 143–153

Ala S, Shokrzadeh M, Pur Shoja A M, Saeedi Saravi S S (2009). Zinc and copper plasma concentrations in rheumatoid arthritis patients from a selected population in Iran. Pak J Biol Sci, 12(14): 1041–1044

Ali H M, Al-Zubaidi M A (2012). Evaluation of trace elements in Iraqi Patients with rheumatoid arthritis by using atomic absorption spectrophotometer (AAS). Iraqi J Pharm Sci, 21(2)

Bonaventura P, Benedetti G, Albarède F, Miossec P (2015). Zinc and its role in immunity and inflammation. Autoimmun Rev, 14(4): 277–285

Chan S, Gerson B, Subramaniam S (1998). The role of copper, molybdenum, selenium, and zinc in nutrition and health. Clin Lab Med, 18(4): 673–685

Farid Y Y, Mohammed A K, Mohammed I S (2000). Serum copper and zinc levels and copper/zinc ratio in patients with rheumatoid arthritis. Iraqi J Med Sci 4(1):49–56

Firestein G S, Budd R, Gabriel S E, O'Dell J R, McInnes I B, Ruddy S, Sergent J S ( 2009). Kelley's Textbook of Rheumatology 8th ed. Philadelphia, PA, Saunders/Elsevier Elsevier

Huizinga T W, Pincus T (2010). Rheumatoid arthritis. Ann Intern Med, 153(1): ITC1–1

Jou J M, Lewis S M, Briggs C, Lee S H, De La Salle B, McFadden S, the International Council for Standardization in Haematology (2011). ICSH review of the measurement of the erythocyte sedimentation rate. Int J Lab Hematol, 33(2): 125–132

Knoell D L, Julian M W, Bao S, Besecker B, Macre J E, Leikauf G D, DiSilvestro R A, Crouser E D (2009). Zinc deficiency increases organ damage and mortality in a murine model of polymicrobial sepsis. Crit Care Med, 37(4): 1380–1388

Leone N, Courbon D, Ducimetiere P, Zureik M (2006). Zinc, copper, and magnesium and risks for all-cause, cancer, and cardiovascular mortality. Epidemiology, 17(3): 308–314

Li J, Liang Y, Mao H, Deng W, Zhang J (2014). Effects of B-lymphocyte dysfunction on the serum copper, selenium and zinc levels of rheumatoid arthritis patients. Pak J Med Sci, 30(5): 1064–1067

Malaviya A N (2003). Outcome measures in rheumatoid arthritis. APLAR J Rheumatol, 6:178–183

Mierzecki A, Strecker D, Radomska K (2011). A pilot study on zinc levels in patients with rheumatoid arthritis. Biol Trace Elem Res, 143 (2): 854–862

Önal S, Nazıroğlu M, Çolak M, Bulut V, Flores-Arce M F (2011). Effects of different medical treatments on serum copper, selenium

and zinc levels in patients with rheumatoid arthritis. Biol Trace Elem Res, 142(3): 447–455

Sahebari M, Ayati R, Mirzaei H, Sahebkar A, Hejazi S, Saghafi M, Saadati N, Ferns G A, Ghayour-Mobarhan M (2016). Serum trace element concentrations in rheumatoid arthritis. Biol Trace Elem Res, 171(2): 237–245

Salgado E, Maneiro J R (2014). [New therapies for rheumatoid arthritis]. Med Clin (Barc), 143(10): 461–466

Strecker D, Mierzecki A, Radomska K (2013). Copper levels in patients with rheumatoid arthritis. Ann Agric Environ Med, 20(2): 312–316

Taneja S K, Mandal R (2009). Assessment of mineral status (Zn, Cu, Mg and Mn) in rheumatoid arthritis patients in Chandigarh, India. Rheumatology Reports, 1(1): e5

Thomas R D, Westengard J C, Hay K L, Bull B S (1993). Calibration and validation for erythrocyte sedimentation tests. Role of the International Committee on Standardization in Hematology reference procedure. Arch Pathol Lab Med, 117(7): 719–723

Tuncer S, Kamanli A, Akçil E, Kavas G O, Seçkin B, Atay M B (1999). Trace element and magnesium levels and superoxide dismutase activity in rheumatoid arthritis. Biol Trace Elem Res, 68(2): 137–142

Uhlig T, Moe RH, Kvien TK (2014). The burden of disease in rheumatoid arthritis. Pharmaco Economics, 32(9):841–851

Ullah Z, Ullah M I, Hussain S, Kaul H, Lone K P (2017). Determination of serum trace elements (Zn, Cu, and Fe) in Pakistani patients with rheumatoid arthritis Biol Trace Elem Res, 175(1): 10–16

Wanchu A, Sud A, Bambery P, Prasad R, Kumar V (2002). Plasma and peripheral blood mononuclear cells levels of Zn and Cu among Indian patients with RA. Ann Rheum Dis, 61(1): 88

Wang C H, Dai J Y, Wang L, Jia J F, Zheng Z H, Ding J, Chen Z N, Zhu P (2011). Expression of CD147 (EMMPRIN) on neutrophils in rheumatoid arthritis enhances chemotaxis, matrix metalloproteinase production and invasiveness of synoviocytes. J Cell Mol Med, 15(4): 850–860

Williamson M A,Snyder L M, Wallach J (2011). Wallachs interpretation of diagnostic tests: pathways to arriving at a clinical diagnosis 10th ed. Wolters Kluwer/Lippincott Williams & Wilkins, 24 (5Part1) :735

Yazar M, Sarban S, Kocyigit A, Isikan U E (2005). Synovial fluid and plasma selenium, copper, zinc, and iron concentrations in patients with rheumatoid arthritis and osteoarthritis. Biol Trace Elem Res, 106 (2): 123–132