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THE EFFECT OF CURCUMIN ON CHANGES IN OXIDATIVE STRESS PARAMETERS IN ANIMAL MODEL OF RHEUMATOID ARTHRITIS: A PILOT STUDY

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Abstract: The aim of this study was to examine the antioxidant capacity of curcumin and whether it modulates RA-induced oxidative stress. Pro-oxidants from plasma and coronary venous effluent were determined spectrophotometrically. The concentration of nitrite was significantly lower in the curcumin treated group compared to the disease group ($p < 0.05$), while the concentrations of superoxide anion radical, hydrogen peroxide and lipid peroxidation index were lower in curcumin group, but without statistical significance between the groups. Curcumin has a beneficial antioxidant effect in the treatment of RA, but considering that this was a pilot study, additional investigations will be performed.

Keywords: curcumin, rheumatoid arthritis, oxidative stress

Introduction

Rheumatoid arthritis (RA) is a chronic autoimmune disease characterized by progressive inflammation of the joints and consequent destruction of cartilage, bone erosion and development of disability (Smolen1 et al., 2016). Oxidative stress plays an important role in the pathogenesis of RA. Reactive oxygen species (ROS) can activate the nuclear factor κB (NFκB) signaling pathway, responsible for the production of proinflammatory cytokines, also can activate enzymes metalloproteases, responsible for damaging extracellular

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matrix components, and increase the level of lipid peroxidation, which causes oxidative damage in RA synovial fluids and tissues (Djordjevic1 et al, 2023; Behl1 et al., 2021).

Curcumin is the active component from turmeric (*Curcuma longa*) and a potent scavenger of a variety of reactive species including superoxide anion radical, hydroxyl radical, and nitrogen monoxide (Pulido-Moran1 et al., 2016). The potential efficacy of curcumin has been recognized in the prevention and treatment of various diseases such as diabetes mellitus, cancers, cardiovascular, inflammatory, autoimmune, neurological and skin diseases (Pourhabibi-Zarandi1 et al., 2021).

Due to the pathogenesis of the disease and the mechanism of action of curcumin, the aim of this study was to examine the antioxidant activity of curcumin and whether it modulates RA-induced oxidative stress in animal model of RA.

Materials and methods

This was experimental study on animal material *in vivo* and *ex vivo*. The study included 12 female *Wistar albino* rats divided into two groups:

1. RA - rats with CFA-induced RA (n=6)
2. QRA - rats with CFA-induced RA treated with curcumin 200 mg/kg three times per week 4 weeks *per os* (n=6)

Animal model of rheumatoid arthritis was induced by subcutaneous injection of 0.1 ml Complete Freund's adjuvant (CFA) into the left hind paw (Zhao1 et al., 2022). Rats were sacrificed 28th day from immunization. After the sacrifice, the blood samples were collected. Coronary venous effluents were collected during the different time moment in the ischemia-reperfusion protocol on *Langendorff* model of isolated heart (Watanabe1 and Okada2, 2018).

Cardiac and systemic oxidation status were investigated by determination of level of prooxidants spectrophotometrically from coronary venous effluent and plasma: superoxide anion radical ($O_2^{\cdot-}$), hydrogen peroxide (H_2O_2), nitric oxide in the form of nitrite (NO_2^-) and lipid peroxidation index - measured as thiobarbituric acid reactive substances (TBARS).

For the statictical analysis IBM SPSS 22.0 statistical package was used. The data are showed tabular and graphical. The Mann-Whitney test and Student's t-test were used to test the variability of certain parameters between the examined groups. The significance level was represented by the values of the obtained results $p < 0.05$.

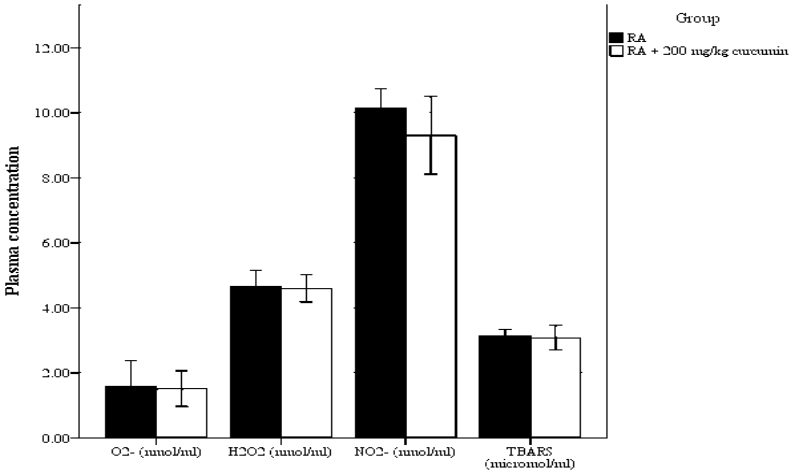
Results and discussion

The concentration of NO_2^- was significantly lower in the QRA group compared to the RA group (9.30 ± 0.60 vs 10.14 ± 0.30, p = 0.017), while the concentrations of $O_2^{\cdot-}$, H_2O_2 and TBARS were slightly lower in the QRA group, without statistical significance between the groups (Table 1, Graph 1).

Table 1. Mean values of pro-oxidants levels in plasma in examined groups

Plasma pro-oxidant	RA (mean ± SD)	QRA (mean ± SD)	p
$O_2^{\cdot-}$	1.56 ± 0.39	1.51 ± 0.28	0.677
H_2O_2	4.66 ± 0.25	4.60 ± 0.21	0.672
NO_2^-	10.14 ± 0.30	9.30 ± 0.60	0.017*
TBARS	3.14 ± 0.10	3.08 ± 0.20	0.529

*Statistical significance p < 0.05



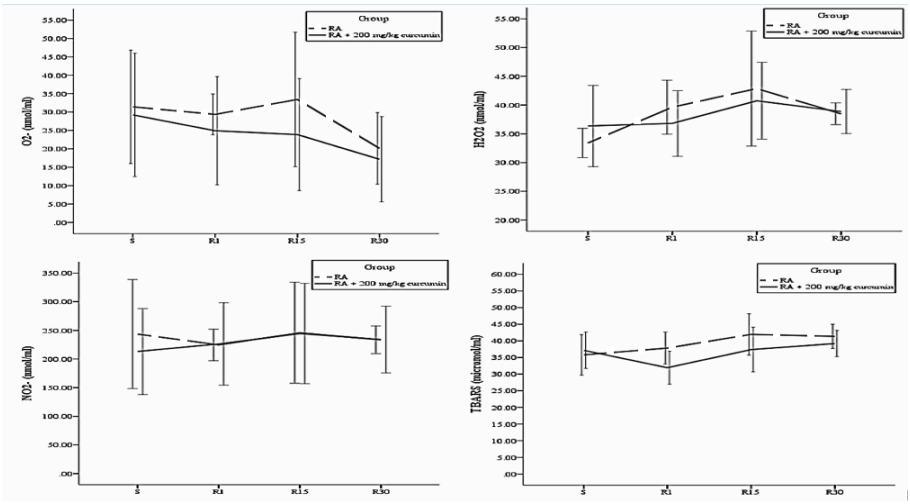
Graph 1. The comparison of the pro-oxidants levels in plasma between the examined groups ($O_2^{\cdot-}$, H_2O_2, NO_2^- and TBARS)

The $O_2^{\cdot-}$ values were decreased at all time intervals in the curcumin group compared to the RA group, but no statistically significant difference was noted (p > 0.05). The H_2O_2 and TBARS values of the QRA group were decreased in the R1-R30 time interval compared to the RA group, but without statistical significance (p > 0.05). Lower NO_2^- values were recorded in the QRA group at

the time of stabilization, but were not statistically significant compared to the RA group ($p > 0.05$) (Table 2, Graph 2).

Table 2. Mean values of pro-oxidants levels in coronary venous effluent in examined groups

Pro-oxidant	Moment in time	RA (mean ± SD)	QRA (mean ± SD)	p
O_2^{-}	S	31.43 ± 10.90	29.23 ± 16.76	0.858
	R1	29.37 ± 5.53	24.93 ± 14.69	0.603
	R15	33.44 ± 18.26	23.88 ± 15.22	0.453
	R30	20.13 ± 9.74	17.17 ± 11.58	0.709
H_2O_2	S	33.40 ± 2.54	36.36 ± 7.05	0.476
	R1	39.62 ± 4.68	36.81 ± 5.72	0.476
	R15	42.86 ± 9.99	40.74 ± 6.69	0.738
	R30	38.47 ± 1.89	38.87 ± 3.82	0.861
NO_2^{-}	S	243.46 ± 47.53	213.16 ± 37.49	0.358
	R1	224.60 ± 13.70	226.27 ± 35.96	0.935
	R15	245.77 ± 44.09	244.37 ± 43.77	0.966
	R30	233.41 ± 12.11	233.99 ± 29.08	0.972
TBARS	S	35.79 ± 6.10	37.15 ± 5.45	0.751
	R1	37.82 ± 4.80	31.93 ± 4.98	0.139
	R15	41.93 ± 6.21	37.37 ± 6.69	0.356
	R30	41.34 ±3.65	39.19 ± 3.95	0.455



Graph 2. The comparison of the pro-oxidants levels in coronary venous effluent between the tested groups (O_2^{-}, H_2O_2, NO_2^{-} and TBARS)

The results of this study indicate a significant decrease in NO_2^- level in the curcumin treated group compared to the group with RA. These finding suggests that curcumin has role in modulating reactive nitrogen species levels, which is consistent with previous research on experimental diabetic rats (Machado1 et al., 2022). Two studies concluded that curcumin attenuates parameters of oxidative stress on different experimental models (Lin1 et al., 2019; Samarghandian1 et al., 2017).

Although the concentrations of $O_2^{\cdot-}$, H_2O_2 and TBARS were lower in QRA group compared to the RA group, these differences were not statistically significant. These results may indicate that the antioxidant effect of curcumin may depend on a plenty factors. There is evidence that therapeutic use of curcumin has many limitations: unfavorable pharmacokinetic and pharmacodynamic properties, chemical instability, low efficacy in various *in vitro* and *in vivo* disease models. Changes in the route of administration play an important role in overcoming pharmaceutical problems related to the use of curcumin in order to improve its bioavailability and therapeutic efficacy (Gera1 et al., 2017).

Conclusion

Treatment with curcumin significantly reduced concentration of NO_2^- in plasma of rats with RA. Also, curcumin reduced other biomarkers of oxidative stress, but without significance. Due to the results and the fact that this was pilot study, next step should be the experiment with larger sample and with different way of curcumin application.

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