ULTRASONOCALLY INDUCED NO˙ FORMATION

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Ultrasound is routinely used in physiotherapy and in a wide range of applied research fields.

Our studies were undertaken to describe the influence of ultrasound on whole erythrocytes, their membranes and haemoglobin at the doses used in medical therapy.

Biophysical ultrasound interactions lead to thermal but also to nonthermal effects, which include the cavitation process. We avoided thermal influence thus the obtained results evidence the inercial cavitation effect initiating free radical reactions.

In our experiments 1 MHz continuous wave at the intensities of 0.61 to 2.44 W/cm^2 for 5 min was applied. From the pool of radicals we studied the level of generated NO˙ as a parameter representing ultrasound influence.

The obtained results demonstrate a considerable increase of NO˙ generation in air-saturated water.

Sonication of full erythrocytes (5% Ht) results in considerable increase in NO˙ generation depending on growing ultrasound wave intensity.

The same effect was observed in erythrocyte membranes exposed to ultrasound. It was also found that a clear dependency between biological ultrasound effect and protein concentration exists. The cavitation process more intense in biological samples of lower protein concentration so the rise of NO˙ is poor in the membranes with higher protein content.

Ultrasound interaction with haemoglobin causes slight generation of NO˙ in the sonicated samples. NO˙ life-time is really very short because of the fast reaction with the haem group. As a result of ultrasound action a very small part of haemoglobin is oxygenated and the initial haemoglobin concentration does not influence the level of methaemoglobin forming.

Application of fluorescence probe DCFH-DA demonstrated an increase in the level of reactive oxygen species forms in the whole erythrocytes exposed to ultrasound. We conclude that ultrasound stimulated NO˙ generation and that cavitation is a process which evidently causes free-radical generation.