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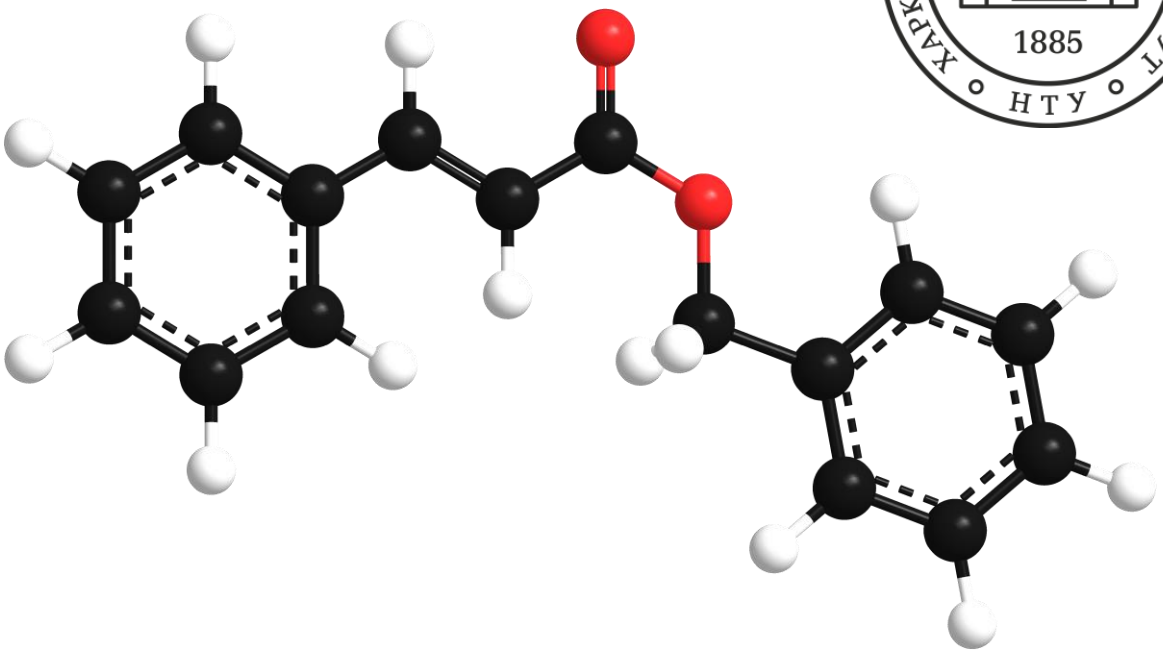

ХІМІЯ, БІО- І НАНОТЕХНОЛОГІЇ, ЕКОЛОГІЯ ТА ЕКОНОМІКА В ХАРЧОВІЙ ТА КОСМЕТИЧНІЙ ПРОМИСЛОВОСТІ

Збірник матеріалів X міжнародної науково-практичної конференції 18-19 листопада 2022

алергенам, які часто спричиняють страждання та смерть.

Об'єкти тестування дуже ресурсомісткі та марнотратні. Вони потребують величезного простору, вентиляції, стабілізації температури та постійного освітлення. Вони утворюють відходи, включаючи хімічні, радіоактивні та біологічно-небезпечні, від очищення, тестування та спалювання трупів.

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FOOD ADDITIVE E407 INFLUENCES LIPOPOLYSACCHARIDE-INDUCED REACTIVE OXYGEN SPECIES PRODUCTION IN PERIPHERAL BLOOD MONONUCLEAR CELLS AND LIPOPOLYSACCHARIDE-MEDIATED CELL MEMBRANE CHANGES IN LEUKOCYTES

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Several studies have suggested that there are links between carrageenan (registered as E407 and E407a) consumption and bacterial intestinal inflammation. However, the mechanisms by which carrageenans may aggravate bacteria-mediated gut inflammation are still poorly understood and required to be scrutinized.

Bacterial lipopolysaccharides (LPS) are a well described component of cell wall in Gram-negative bacteria, which acts as a pathogen-associated molecular pattern (PAMP) and ligand for receptors of the innate immune system, especially TLR4. Downstream effects of TLR-4-LPS complex formation in immune cells include the activation of NADPH oxidase with the corresponding overproduction of reactive oxygen species (ROS) and overexpression of NF-kB-dependent pro-inflammatory cytokines. It is important to note that LPS is capable of inducing the inflammatory ROS-mediated response in peripheral blood mononuclear cells (PBMCs).

The aim of the study was to assess the effects of semi-refined carrageenan (E407a) on lipopolysaccharide (LPS)-induced reactive oxygen species (ROS) generation in peripheral blood mononuclear cells (PBMCs) and LPS-mediated cell membrane alterations in leukocytes.

Methods. Blood samples collected from 8 intact rats were incubated with LPS (2 μg/ml), E407a (10 mg/mL) + LPS (2 μg/ml), E407a (50 mg/mL) + LPS (2 μg/ml) and without those compounds (controls) for 24 h in RPMI 1640 medium enriched with 5% fetal bovine serum. ROS generation in PBMCs obtained from the incubated samples was estimated by flow cytometry using 2',7'-dichlorodihydrofluorescein diacetate (H2DCFDA) staining. The impact of E407a+LPS mixture on leukocyte cell membranes was evaluated spectrofluorimetrically using the fluorescent probe 2-(2′-hydroxy-phenyl)-5-phenyl-1,3-oxazole.

Results. Expectedly, in this study, direct incubation of blood with LPS was found to promote ROS generation in PBMCs and diminish the fluidity of cell membranes in leukocytes, supported by the observed changes in the fluorescence values of fluorescent probe, since the fluidity of phospholipid bilayer decreases in response to free radical-induced peroxidation of polyunsaturated fatty acids, which are essential components of cell membranes responsible for the maintenance of bilayer fluidity. Thus, LPS-mediated changes in the fluidity of cell membranes develop at least partially due to ROS-triggered lipid peroxidation, which causes the reduction of the relative amount of polyunsaturated fatty acids (PUFAs) in

phospholipids. This reduces the lipid order in cell membranes making them more rigid.

Surprisingly, in this study, semi-refined carrageenan was found to partially inhibit LPS-induced ROS generation and LPS-mediated changes in phospholipid bilayers of cell membranes *in vitro*. This is inconsistent with reports stating that carrageenans stimulate LPS-induced generation of TNF-α in leukocytes without affecting expression of this cytokine alone in the absence of LPS. However, *in vivo* studies demonstrate that carrageenan stimulates macrophages even in a more pronounced way compared with LPS. Moreover, experiments on murine models show that kappa-carrageenan magnifies LPS-induced inflammation via the bcl-10-NF-kB pathway. These inconsistencies between our findings and literature data may be explained by the direct interactions of LPS and carrageenans, since carrageenan has been shown to directly alter the morphology and, thus, functional activity of LPS suggesting, on the contrary, the protective and inhibitory role of carrageenans in LPS-induced inflammation.

Thus, our results don't exclude the synergistic pro-inflammatory effects of carrageenans and LPS *in vivo*. Therefore, future researches should aim at closing the gap in elucidating the crosstalks between the ingested carrageenan, gut microbiome and host immune system. This is especially important for individuals with the compromised intestinal barrier integrity and already existing intestinal inflammation.

Conclusion. E407a attenuates LPS-induced alterations of redox homeostasis in rat PBMCs and LPS-mediated modifications of cell membrane lipid order in leukocytes. Thus, semi-refined carrageenan (E 407a) reduces the LPS-mediated pro-inflammatory effects in leukocytes as a whole and PBMCs in particular.

THE STUDY OF THE SEMI-REFINED CARRAGEENAN (E407a) INFLUENCE ON REACTIVE OXYGEN SPECIES PRODUCTION IN PERIPHERAL BLOOD MONONUCLEAR CELLS AND ON LEUKOCYTE CELL MEMBRANE

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