

Influences of Probiotic “Biomilk” on Indomethacin-induced Oxidative Injuries of Some Tissues

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The effect of probiotics Biostim LBS (Biomilk) and LBB of pure culture on ulcerogenesis in Indomethacin-induced oxidative stress by means of a model of Indomethacin-induced ulcer in white male rats was studied. Indomethacin was subcutaneously injected in a dose of 20 mg/kg bw 4 hours prior to taking for examination the biopsy material such as blood, liver and brain. Both probiotics Biostim LBS and LBB were introduced per sondam in a dose of 1600 mg/kg bw for 30 consecutive days prior to modelling the Indomethacin-induced ulcers. Malonyldialdehyde (MDA) as a marker of lipid peroxidation was examined in plasma, liver and brain homogenate. The results from the morphometric study of gastric lesions were presented as index of lesions. Indomethacin caused the formation of numerous lesions and haemorrhages and enhanced MDA level in plasma and tissues. Both probiotics Biostim LBS and LBB introduced for 30 days restrict the lipid peroxidation and protect the gastric mucosa from Indomethacin ulcerogenic action.

Key words: Biostim LBS, Indomethacin, gastric ulcer, index of lesions.

Introduction

Indomethacin is a NSAID widely used in the treatment of rheumatoid arthritis, collagenoses etc. and possesses a manifested ulcerogenic effect [9]. It has been proved that it suppresses prostaglandins' synthesis and thus their antiacid and gastroprotective properties. This is accompanied by the occurrence of mucosal lesions, haemorrhagic erosions and delay of reparation processes [3]. There exist data about the involvement of free-radical processes in indomethacin-induced ulcers [10].

Probiotics stimulate the growth and development of bifidum and lactobacilli [2]. There are data about the antioxidant activity of lactic acid bacteria [1, 4, 5]. A protective effect of probiotics containing such bacteria has been established in experimental stress- and alcohol-induced ulcerogenesis as well as in patients with ulcer disease [7].

The objective of the present study is to establish the effect of the probiotic Biostim LBS (Biomilk) containing livecells of *Lactobacillus bulgaricus*, proteins, fats, carbohydrates, minerals and vitamins as well as the effect of LBB-pure culture of *Lactobacillus bulgaricus* on ulcerogenesis in Indomethacin-induced oxidative stress.

Materials and Methods

The study covered white male Wistar rats weighing 180-200 g who were maintained at free access to standard food and water. The animals were divided into 6 groups with 7 animals each: group I – controls, distilled water for 30 days; group II – Indomethacin on the last day of experiment; group III – Biostim LBS for 30 days; group IV – LBB for 30 days; group V – Biostim LBS for 30 days + Indomethacin on the last day of experiment and group VI – LBB for 30 days + Indomethacin on the last day of experiment.

Indomethacin (Fluka Chemie, Switzerland) in the shape of suspension in distilled water and Tween-80 was chosen as an ulcerogenic agent with pro-oxidative action and subcutaneously injected in a dose of 20 mg/kg bw. Stomachs were taken for examination 4 hours after Indomethacin administration under ethereal narcosis. A morphometric investigation of gastric lesions was performed and the results were presented as index of lesions. Malonyldialdehyde (MDA) as a marker of lipid peroxidation after the method of Porter [8] was estimated in blood plasma, liver and brain homogenates.

Results and Discussion

Indomethacin treatment induced the formation of numerous lesions and haemorrhages. The index of lesions in this group was highest (Fig. 1). Independent

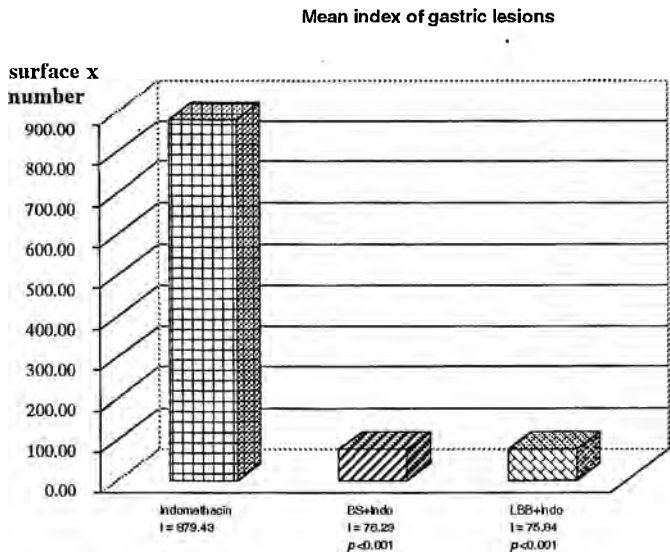


Fig. 1. Mean index of gastric lesions in Indomethacin-treated rats in a model of Indomethacin-induced ulcerogenesis

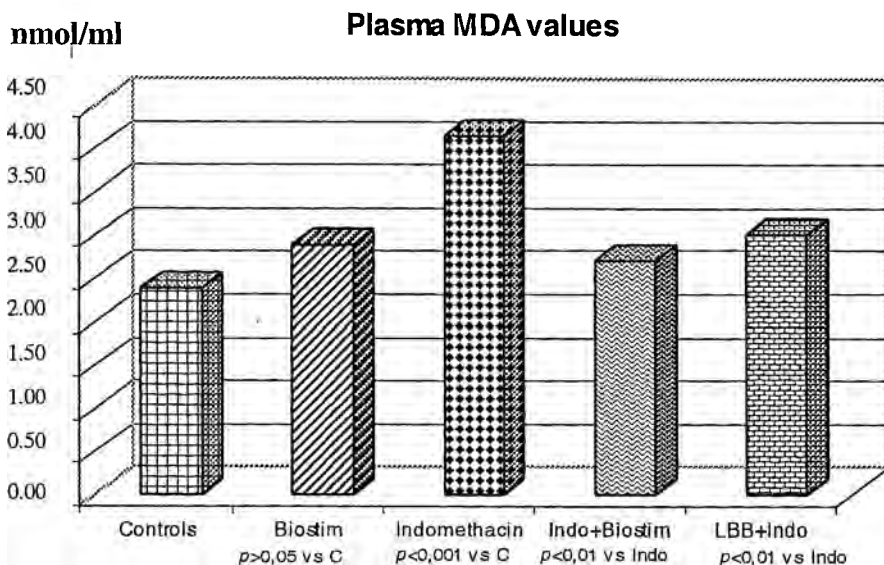


Fig. 2. Plasma MDA values

chronic treatment with Biostim LBS and LBB did not cause any ulcer formation at all. The probiotics Biostim LBS and LBB introduced for 30 days strongly protect the gastric mucosa against the ulcerogenic action of Indomethacin.

MDA in plasma

In the animals treated Biostim LBS or LBB only MDA values did not differ significantly from these of the controls ($p > 0,05$). Indomethacin treatment caused a significant ($p < 0,001$) MDA increase in plasma as compared with that of the controls. MDA in plasma reduced in the groups with Biostim LBS+Indomethacin and LBB+Indomethacin in comparison with the group with Indomethacin only ($p < 0,01$) (Fig. 2).

MDA in liver homogenate

In the animals treated with Biostim LBS or LBB only MDA values in liver tissue did not differ significantly from these of the controls ($p > 0,05$). Indomethacin treatment caused a significant ($p < 0,01$) MDA increase in liver homogenate as compared with that of the controls. In the liver homogenate, MDA values reduced significantly ($p < 0,01$) in the animals of the groups with Biostim LBS+Indomethacin and LBB+Indomethacin when compared with these with Indomethacin only (Fig. 3).

MDA brain homogenate

In the animals treated with Biostim LBS or LBB only MDA values in a homogenate from brain tissue did not differ significantly from these of the controls ($p > 0,05$). Indomethacin treatment caused a significant ($p < 0,001$) MDA increase in brain homogenate as compared with that of the controls. In the brain homogenate, MDA values reduced significantly ($p < 0,01$) in the animals of the groups with Biostim LBS+Indomethacin and LBB+Indomethacin when compared with these with Indomethacin only (Fig. 4).

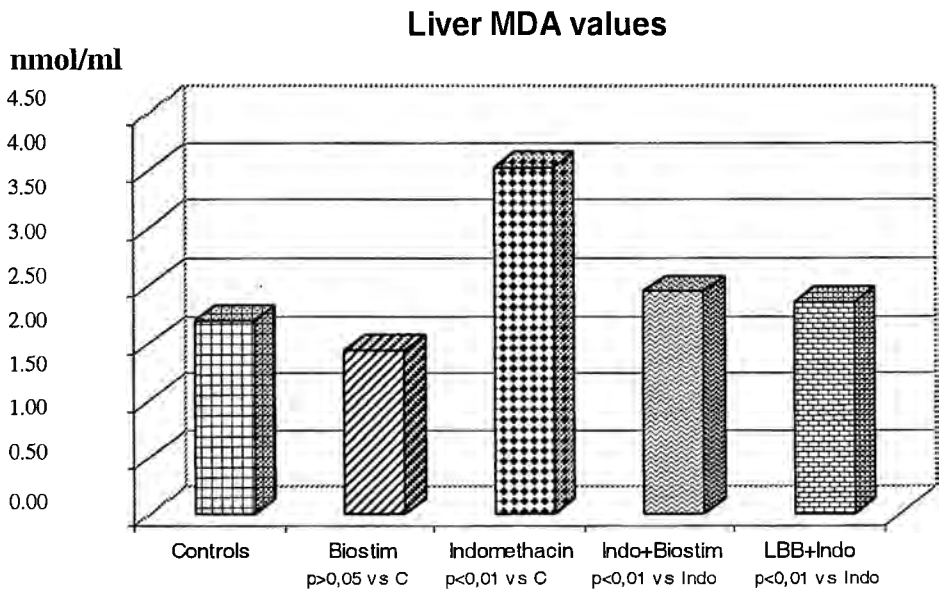


Fig. 3. Liver MDA values

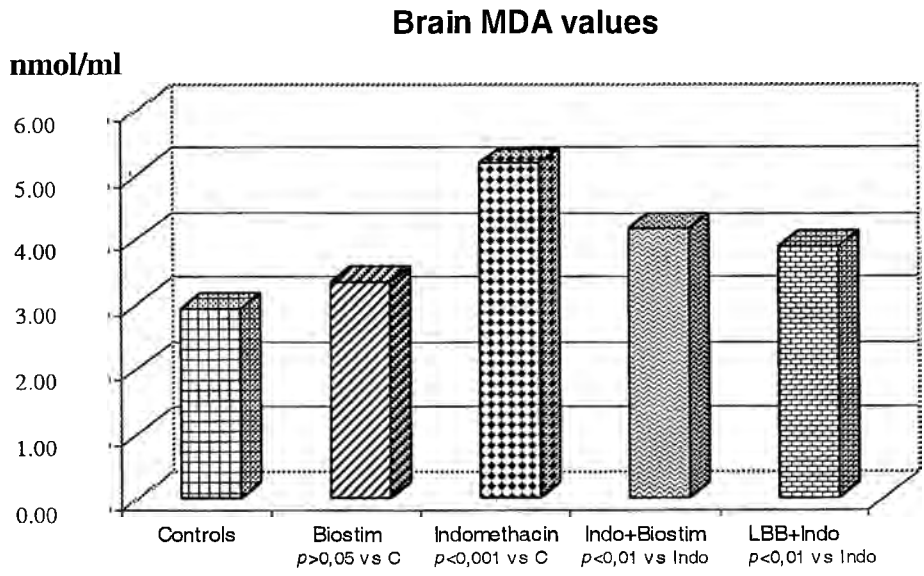


Fig. 4. Brain MDA values

Indomethacin causes MDA elevation in plasma along with that in liver and brain homogenate that allows the assumption of oxidative stress involvement. In the groups with Biostim LBS (Biomilk)+Indomethacin and LBB+Indomethacin these

parameters were favourably influenced upon. It is known that lactic acid bacteria possess an antioxidant activity [5, 6]. It is possible that the protective action of the probiotics Biostim LBS and LBB on the gastric mucosa is due to the restriction of lipid peroxidation. It stresses that MDA as a marker of this peroxidation in liver and brain decreases to a greater extent in the group of LBB+Indomethacin than in that of Biostim LBS + Indomethacin. LBB representing a pure culture with a greater amount of live cells of *Lactobacillus bulgaricus* proved to exert a stronger effect in the examined organs than Biostim LBS. Probably, the antioxidant activity is determined by the number of live cells of *Lactobacillus bulgaricus*. The present results have shown that there exists a parallelism between the degree of the antioxidative effect and the number of live cells of *Lactobacillus bulgaricus*.

In stress-induced gastric ulcers a protective effect of alpha-lactalbumin has been established. The authors assume that this antiulcerogenic activity is due to the stimulated prostaglandin synthesis [9, 10, 11].

Based on the aforementioned data we could draw the conclusion about the clinical significance of the chronic application of both Biostim LBS (Biomilk) and LBB in Indomethacin-provoked pathology of the gastric mucosa that corresponds with the data of other authors about the protective properties of these probiotics against stress- and ethanol-induced ulcer.

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