



The interferon production and 2',5'-oligoadenylate-synthetase activity in rat spleen lymphocytes at hypoacidity evoked by omeprazole injection and at administration of multiprobiotic «SYMBITER®»

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ABSTRACT

The IFN production by rat spleen lymphocytes was shown to increase by 2.5 times in comparison to control against a background of hypoacidity evoked by 28-days of OM treatment (14 mg/kg). The activity of IFN-induced enzyme 2',5'-OAS declined by 16 % in these cells. The simultaneous administration of multiprobiotic SYM and OM resulted in IFN titer increase in comparison to hypoacidic animals. The 2',5'-OAS activity in spleenocytes didn't change. SYM displayed interferonogenic properties via gastric microflora normalization and suppressing inflammation.

Keywords: interferon, 2',5'-oligoadenylate-synthetase, omeprazole, multiprobiotic

INTRODUCTION

Gastric hypoacidity is the effect of long-term treatment with H⁺-K⁺-ATPase inhibitors [1]. The increase of gastric juice pH leads to augmentation of gastrin production by antrum G-cells and disbacteriosis development [5]. An inflammation developing in gastric mucosa is accompanied by proinflammatory cytokines secretion [9]. The probiotics are used for microbiocenosis normalization and prevention of structurally functional changes in stomach caused by hypoacidity [6]. The role of cytokines in processes that occur in immune system during hypoacidity has not been clarified yet. Interferons (IFN) are cytokines that bring into broad spectrum of effects on cell metabolism: the defense of viruses and microorganisms, antiproliferative and immunomodulatory action [4]. Type I IFN induces in cells 2',5'-oligoadenylate signaling pathway. The 2',5'-oligoadenylate-synthetase (2',5'-OAS) is it's key enzyme [11]. As far as the augmentation of

IFN secretion by stomach mucous lymphocytes was established at hypoacidity [8] the changes of its production by lymphoid organs are expected.

The aim of the work was to study the interferon production by rat spleen lymphocytes and 2',5'-OAS activity in these cells at hypoacidity caused by 28-daily omeprazole (OM) injection as well as simultaneous omeprazole and multiprobiotic "Symbiter® acidophilic concentrated" (SYM) administration.

MATERIAL AND METHODS

The research was conducted on white nonlinear rats (males) weighing 170–200 grams. Animals were divided into 4 groups. Animals of the 1st group (control) were intraperitoneally injected with 0.2 ml of H₂O (i.p.). Animals of the 2nd group were orally (p/o) treated with SYM (O.D. "Prolisok", Ukraine) (0.14 ml/kg, diluted in 0.5 ml of H₂O) during of 28 days. Animals of the 3rd group were treated with OM ("Omez®" "SIGMA", India) (14 mg/kg, diluted in 0.2 ml of H₂O). Animals of the third group were simultaneously treated with OM and SYM.

The rats were sacrificed by cervical dislocation 1 day after last drug treatment, spleen lymphocytes were isolated

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[2]. Spleenocytes (5·10⁶ cells / ml) were incubated *in vitro* during 24 h at 37°C for IFN induction. Cells of each group of animals were separately incubated with IFN inducers: PHA (“SIGMA-Aldrich”, USA) (20 mkg/ml) and cycloferon (“Polisan”, Russia) (50 mkg/ml). Cell suspensions were centrifuged at 200 g for 5 min. The IFN titer was evaluated by micromethod [10]. The 2',5'-OAS activity was estimated by method [7]. The statistical evaluation was calculated by Student’s t-test.

RESULTS

It was established that spontaneous IFN titer was increased by 2.5 times in comparison to control in supernatants of cultivated rat spleen lymphocytes after 28 days of OM treatment (Table 1). The SYM administration to animals caused the growth of this parameter by 42%. The spontaneous IFN titer turned up to the control level at combined OM and SYM treatment. The enhancement of IFN production induced by PHA and cycloferon was more expressed at common OM and SYM action than at OM action only.

Table 1. The interferon (IFN) titer in supernatants of cultivated rat spleenocytes and its correlation with 2,5-oligoadenylate-synthetase (2,5-OAS) activity in rat spleenocytes after 28 days omeprazole-evoked hypoacidity and multiprobiotic “Symbiter® acidophilic concentrated” (SYM) treatment (M±m, n = 5)

	Control	SYM (0.14 ml/kg)	Omeprazole (14 mg/kg)	Omeprazole + SYM
IFN titer (log₂ titer)				
Without inducer	1.90.07	2.70.09*/#	2.50.85*	2.70.08*/#
PHA	2.70.09*	3.30.11*/#	3.80.11*	50.2*/#
Cycloferon	3.70.14*	4.30.17*/#	5.10.14*	5.70.21*/#
2',5'-OAS (nmol P_i / (min·mg of protein))				
Without inducer	39.021.59	38.11.11*/#	32.671.14*	31.90.86*/#
PHA	53.851.88*	46.11.29*/#	47.371.75*	47.531.89*/#
Cycloferon	70.632.54*	57.532.18*/#	61.421.78*	64.762.26*/#

* – p<0.05 in comparison to control; # – p<0.05 in comparison to animals that were not treated with SYM

The 2',5'-OAS activity in 3rd group rat spleenocytes decreased by 16% after 28 daily OM injection and approximated to 3rd group values at combined OM and SYM administration (Table 1). The SYM administration to control group animals had not influence on enzyme activity. The 2',5'-OAS activity augmentation in cells incubated with PHA and cycloferon was maximal at simultaneous OM and SYM administration in comparison with the third group at OM administration only.

DISCUSSION

The secretion of spontaneous and induced IFN by rat spleen lymphocytes was shown to increase at hypoacidity evoked by 28-days of OM treatment. Probably the IFN synthesis is stimulated in these cells that may be their response to stomach inflammation evoked by bacterial

colonization. It correlates with the facts that IFN- induction occurs owing to bacterial lipopolysaccharide interaction with lymphocyte Toll-like receptors [12]. It was also established that IFN- synthesis is amplified in gastric mucosa lymphocytes against a background of inflammation in consequence of reduced hydrochloric acid secretion [8]. Spleen is extremely sensitive to organism immunological status changes. Therefore, the functional state of this lymphoid organ can be changed under the influence of inflammation that is developed at gastric disbacteriosis. Gastrin can also influence IFN production by spleen lymphocytes, because its blood level rises by gastric hypoacidity [9]. We have demonstrated that SYM leads to IFN level increase in healthy animals and intensification of this effect in hypoacidic ones. Therefore, we suppose that this multiprobiotic leads to inhibition of inflammation via gastric microflora recovery, it has an effect on lymphoid organs functioning.

2',5'-activity in rat spleenocytes falls along with IFN production increase in conditions of hypoacidity. It may occur owing to IFN signaling pathway dysfunction at the stages of IFN-receptor interaction or stimulation of 2',5'-OAS gene expression. The study of IFN system functioning in conditions of gastric hypoacidity is of a great importance for clarifying the mechanism of the immune system reaction on developing pathological process as well as for design of therapeutic and prophylactic preparations.

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