THERAPEUTIC AND PREVENTIVE EFFECTIVENESS OF ANTIDISBIOTIC AGENTS

IN RATS WITH LIPID INTOXICATION

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Abstract

Aim: to determine the effect of antidisbiotic agents on the state of the organism of rats under conditions of lipid intoxication and to propose a comprehensive method for assessing therapeutic and prophylactic efficacy.

Methods: lipid intoxication was induced in rats using thermoperoxide sunflower oil. As antidisbiotic agents, drugs containing quercetin, inulin, calcium citrate (Kvertulin), lecithin, quercetin, inulin, calcium citrate (Lekvin), flavolignans, inulin, lecithin, calcium citrate (Lekasil) and lysozyme, quercetin, inulin, gelatin, calcium citrate (Lysozyme-forte). The level of markers of inflammation and dysbiosis (elastase, urease, MDA) and the level of markers of defense systems (lysozyme, catalase) were determined in the tissues of the gums, the mucous membrane of the colon, liver and blood serum. The pathogenic effect of lipid intoxication was assessed by the sum of changes in the level of markers. The therapeutic effect of antidisbiotic drugs was also assessed by the pattern of the change in the level of markers.

Results: lipid intoxication increases the activity of elastase, urease and MDA content in tissues, but decreases the level of lysozyme and catalase. Antidisbiotic drugs have anti-inflammatory effects. Lipid intoxication most of all affects the mucous membrane of the colon and liver. The most effective was Lysozyme-forte.

Conclusions: a comprehensive method for assessing the pathogenic effect of lipid toxins and a comprehensive method for assessing the therapeutic and prophylactic efficacy of antidisbiotic drugs are proposed.

Keywords: *lipid intoxication, markers of inflammation, markers of nonspecific immunity, antidisbiotic agents, methods for assessing therapeutic efficacy.*

Introduction

Lipid intoxication of the body occurs when consuming thermally processed fats [1, 2]. The leading pathogenetic mechanism of the development of pathological complications in lipid intoxication is the development of intestinal dysbiosis and dysbiotic syndrome [3]. The latter is manifested by bacteremia, endotoxinemia, systemic inflammation and multiple organ failure.

For the prevention and treatment of dysbiotic syndrome, antidisbiotic agents (probiotics and prebiotics) are used, and recently polyfunctional antidisbiotic drugs (PFAD), which, along with prebiotics, include antioxidants, antimicrobial agents, and hepatoprotectors [4].

The aim of this study was to develop a comprehensive method for assessing the effectiveness of the therapeutic and prophylactic action of PFAD, based on determining the nature of changes in the level of biochemical markers of dysbiosis, inflammation, antimicrobial and antioxidant defense systems.

The enzymes elastase, urease and the product of lipid peroxidation malondialdehyde (MDA) were chosen as markers of inflammation and dysbiosis [5].

The enzyme lysozyme [6] and the antioxidant enzyme catalase [5] were chosen as markers of defense systems.

Materials and methods

The experiments were carried out on 41 white Wistar rats (males, 7 months old, live weight 238-253 g), distributed into 6 groups: 1st - control (intact animals), 2nd–6th received with food 1 ml of thermally processed sunflower oil (TPSO) daily for 75 days [7]. Starting from the 31st day of the experiment, the rats of the 3rd and 6th groups began to receive the following antidisbiotic agents: Kvertulin (3rd group), Lekvin (4th group), Lekasil (5th group) and Lizozyme-forte (6th group). All drugs were administered daily at a dose of 300 mg/kg for 45 days. The composition of the preparations is presented in table 1.

The animals were euthanized on the 76th day under thiopental anesthesia (20 mg/kg) by total bloodletting from the heart. Serum was obtained and tissues of the gums, colon mucosa and liver were isolated. In these biological objects, the level of elastase [5], urease [8], catalase [5], lysozyme [8] and MDA [5] was determined.

The pathogenic effect (PE) of lipid intoxication was assessed by the degree of increase (in %) of the activity of elastase, urease and MDA content, as well as by the degree of decrease in the activity of lysozyme and catalase.

The therapeutic effect (TE) of antidisbiotic drugs was assessed by the degree of decrease in the level of markers of inflammation and dysbiosis (elastase, urease, MDA) and by the degree of increase in the activity of defense markers (lysozyme and catalase).

Therapeutic and prophylactic efficacy (TPE) was calculated using the formula: $TPE = \frac{TE}{PE} \cdot 100\,\%$, where:

TE — the sum of the absolute values of the change (in%) markers of inflammation and protection (excluding the signs + or -) after the administration of drugs;

PE — similar, but only in animals, that received TPSO without antidisbiotic drugs.

Experimental studies were conducted in accordance with the rules established by the Directive of the European Parliament and the Council (2010/63 / EU), by the order of the Ministry of Education and Science, Youth and Sports of Ukraine No. 249 of March 1, 2012 "On Approval of the Procedure for conducting scientific experiments, experiments on animals by scientific institutions " and methodical recommendations.

Results

Table 2 shows the results of determining the level of inflammation markers and defense systems in the tissues of control rats.

Table 3 shows the results of changes (in %) in the level of markers (an increase in the level of inflammation markers and a decrease in the level of defense markers) in the tissues of rats treated with TPSO. From these data, it can be seen that the most sensitive to this effect of TPSO were the mucous membrane of the large intestine and the liver.

Table 4 shows the results of changes in the markers of inflammation and protection in the tissues of rats treated with Kvertulin against the background of lipid intoxication. To the greatest extent, the therapeutic effect of Kvertulin affected

the blood serum levels, and the lowest - on the indicators of the colon mucosa.

Table 5 shows the results of changes in markers in the tissues of rats treated with Lekvin against the background of lipid intoxication. In general, the therapeutic effect of Lekvin differed little from that of Kvertulin, with the exception of the liver, in which this effect was one and a half times greater, possibly due to the lecithin introduced into the composition of Lekvin.

Table 6 shows the results of the effect on markers of the Lekasil preparation containing milk thistle flavolignans, which are widely used in the composition of hepatoprotectors. Indeed, its therapeutic effect was found to be highest in the liver.

Table 7 shows the results of determining the effect of the drug Lysozyme-forte, which differs from Kvertulin by the presence of the antimicrobial enzyme lysozyme. Lysozyme-forte turned out to be the most effective in the action on blood serum markers, almost completely (by 91.7%) normalizing these parameters.

Conclusions

1. A comprehensive method for assessing the pathological effect of toxic substances on the body has been proposed.

2. A comprehensive method for assessing the therapeutic effect of antidisbiotic drugs on the body under intoxication conditions has been proposed.

3. The most sensitive to lipid intoxication were the mucous membrane of the large intestine and the liver.

4. Of the four tested PFAD, Lysozyme-forte was found to be the most effective.

Acknowledgments

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Table 1. The composition of the used PFAD*

№№	Name PFAD	Composition
1	Kvertulin	Quercetin, inulin, calcium citrate
2	Lekvin	Lecithin, quercetin, inulin, calcium citrate
3	Lekasil	Flavolignans milk thistle, inulin, calcium citrate
4	Lysozyme-forte	Lysozyme, quercetin, inulin, gelatin, calcium citrate

*All drugs are produced by Biochimtech LLC (Odessa, Ukraine)

Table 2. The level of markers of inflammation and protection in the tissues of control rats

Markers	Gum	Colon mucosa	Liver	Blood serum
Inflammation				
Elastase, mk-cat/kg (l)	37,3±3,5	44,8±2,8	360±10	120,0±1,4
MDA, mmol/kg (l)	18,4±1,4	4,36±0,21	24,9±2,1	0,96±0,07
Urease, mk-cat/kg (l)	0,73±0,12	0,66±0,17	0,07±0,02	0,70±0,10
Protection				
Lysozyme, unit/kg (l)	141±12	93±5	86±4	79±4
Catalase, mkat/kg (l)	8,59±0,07	1,30±0,09	5,26±0,23	0,41±0,03

Table 3. The degree of change in the level of markers of inflammation and protection in the tissues of rats with lipid intoxication

Markers		Degree of change,%		
		Gum	Colon mucosa	Liver	Blood serum
Inflammation					
Elastase	+	55,2	82,3	23,6	29,2
MDA	+	72,3	52,3	109,3	49,0
Urease	+	126,0	386,9	151,4	120,0
Protection					
Lysozyme	−	17,7	38,7	39,5	29,1
Catalase	−	12,8	4,6	7,4	26,8
Pathogenic action		287,0	564,8	331,2	254,1

Table 4. Effect of Kvertulin on the degree of changes in inflammation and defense markers in the tissues of rats with lipid intoxication

Markers		Degree of change, %		
		Gum	Colon mucosa	Liver	Blood serum
Inflammation					
Elastase	−	15,5	26,3	22,7	22,1
MDA	−	17,9	33,7	27,1	30,8
Urease	−	27,2	17,6	27,8	59,7
Protection					
Lysozyme	+	17,6	21,1	15,4	46,4
Catalase	+	20,1	29,8	13,1	23,3
Therapeutic action		98,3	128,5	106,1	182,3
Therapeutic and prophylactic effectiveness		34,2	22,7	32,0	71,7

Table 5. Effect of Lekvin on the degree of changes in inflammation and defense markers in the tissues of rats with lipid intoxication

Markers		Degree of change, %			
		Gum	Colon mucosa	Liver	Blood serum
Inflammation					
Elastase	−	21,6	36,1	19,2	9,7
MDA	−	33,2	34,3	23,2	37,1
Urease	−	10,9	34,0	81,8	43,5
Protection					
Lysozyme	+	8,8	21,1	19,2	50,0
Catalase	+	19,9	23,1	5,7	53,3
Therapeutic action		94,4	148,6	152,1	193,6
Therapeutic and prophylactic effectiveness		32,9	26,3	45,9	76,2

Table 6. Influence of Lekasil on the degree of changes in markers of inflammation and protection in the tissues of rats with lipid intoxication

Markers		Degree of change, %			
		Gum	Colon mucosa	Liver	Blood serum
Inflammation					
Elastase	−	17,8	38,9	20,4	12,5
MDA	−	29,5	21,1	32,8	40,6
Urease	−	2,2	22,3	26,7	39,0
Protection					
Lysozyme	+	27,5	36,8	5,8	7,1
Catalase	+	7,2	29,0	5,5	20,0
Therapeutic action		84,2	148,1	171,1	119,2
Therapeutic and prophylactic effectiveness		29,3	26,2	51,7	58,7

Table 7. Influence of Lysozyme-forte on the degree of changes in inflammation and defense markers in the tissues of rats with lipid intoxication

Markers		Degree of change, %			
		Gum	Colon mucosa	Liver	Blood serum
Inflammation					
Elastase	−	14,7	29,1	24,6	25,1
MDA	−	19,8	32,1	25,2	42,0
Urease	−	15,2	15,2	43,2	74,0
Protection					
Lysozyme	+	17,6	19,3	38,5	28,6
Catalase	+	14,6	29,0	14,2	63,3
Therapeutic action		81,9	124,7	145,7	233,0
Therapeutic and prophylactic effectiveness		28,5	22,1	44,0	91,7