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SIRCHAK YE.S.

State Higher Educational Institution «Uzhgorod National University», Medical Faculty,
Uzhgorod, Ukraine

METHODS OF CORRECTING INTESTINE DYSBIOSIS AND THEIR INFLUENCE ON THE DYNAMICS OF CLINICAL MANIFESTATIONS OF HEPATIC ENCEPHALOPATHY AMONG PATIENTS WITH LIVER CIRRHOSIS

Summary. Here are given the results of a complex survey of 169 patients with liver cirrhosis having signs of liver encephalopathy. We found more significant positive dynamics of clinical manifestations of liver encephalopathy in patients ($n = 70$), for whom for correcting microbiocenosis of the colon we prescribed in complex therapy not only prebiotics, but also probiotics.

Key words: liver cirrhosis, liver encephalopathy, intestinal dysbiosis, prebiotic, probiotic.

Actuality of the Problem

Currently, we observe a rapid increase in the distribution of chronic liver diffuse diseases. Risk factors for its development include: food deficit, immune mechanisms infringement, alcohol consumption, genetic and gender factors, viral infections that cause damage to hepatocytes which leads to the formation of the diseases clinical picture and lies within the base of its progressive disease circuit. The main forms of chronic diffuse liver diseases include hepatitis (inflammatory diseases): infectious, autoimmune, toxic (including drugs and alcohol); hepatic disease (dystrophic and exchange character) — hereditary (exchange), toxic (drugs and alcohol), cirrhosis and fibrosis. The most topical nowadays is combination of (viral and alcohol) liver affections — viral and alcohol, which is characterized today as a general medical problem. Infection with hepatitis B virus (HBV) and C (HCV) is diagnosed in 52–54 % of individuals suffering from alcoholism. 25–28 % of patients with alcoholic hepatopathy are infected with HCV, 25–30 % of patients with chronic hepatitis C the major factor that affects the prediction of the disease is chronic alcohol intoxication [1, 4, 6].

It should be remembered that the combined pathology is characterized by a more severe clinical disease circuit, an

early occurrence of complications, higher level of deaths compared to isolated liver damage done by viral or alcohol genesis.

One of the most frequent complications that patients with liver cirrhosis have is hepatic encephalopathy. According to the results of different foreign researches, hepatic encephalopathy occurs with 50–80 % of patients with liver cirrhosis (LC), among them only 40–46 % have clear clinical signs of neurological and psychiatric disorders, and 70–78 % — this occurs in a latent form of hepatic encephalopathy (HE) [5].

HE is the result of endogenous neurotoxins study which normally inactivate in the liver. When damage is done to the liver, the amount of around 20 substances in the liver increases, that can potentially cause brain dysfunction. Among cerebrototoxic substances ammonia takes the main place — the end product of protein metabolism and also ammonia that comes from the intestine into the general blood flow through porto-caval anastomosis, go-

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ing through the liver. Next, ammonia, being in a unionized form, easily penetrates through the blood-brain barrier causing a toxic effect on the central nervous system. The group of endogenous neurotoxins also includes mercaptans, short- and long chain fatty acids, phenol and indole derivatives. Mercaptans are the product of bacteria hydrolysis of sulfur-containing amino acids (methionine, cysteine, cystine) and are mainly situated in the colon. Normally their neutralization occurs in the liver [3]. The dysfunction of the normal synthesis of vitamins, enzymes, amino acids and the replacement of the metabolism of nitrogen containing compounds and urine by the microflora of the colon in case of dysbacteriosis also favors the appearance of clinical manifestations of hepatic encephalopathy in patients with cirrhosis [7].

In the therapy of chronic liver diseases the central place is devoted to topics that concern treatment and preventive measures of their complications, in particular — hepatic encephalopathy, which occurs as a result of severe metabolic disorders. In the treatment of HE, besides the generally accepted traditional methods which include detection and abolition of the provoking factors, prescription of cleansing enemas and antibiotics, use of medications that stimulate the neutralization process of encephalopathy mediators, beneficially prescribe medicine, that normalize the compound of the colon. In this respects, a certain interest is formed by assigning patients with hepatic encephalopathy probiotics into complex, pathogenetically reasoned treatment programs.

Recently a drastic increase is observed in the number of researches on probiotics: in 2001–2005 there were conducted 4 times more researches than in 1996–2000 [2]. Probiotics are being actively studied in terms of nonalcoholic fatty liver disease, but, technically, there is no data on the usage of probiotics in terms of complex treatment of patients with liver cirrhosis.

The aim of the work: to conduct a comparative assessment of different colon dysbiosis correction schemes using prebiotics and probiotics in patients with LC and to determine their influence on the HE clinical manifestation dynamics.

Materials and Methods

We had 169 patients with LC under observation, which were treated in the gastroenterology, surgical and intensive care departments of Transcarpathian Regional Clinical Hospital of A. Novak. Among the examined patients the number of men prevailed — 102 (60.4 %), aged (56.4 ± 6.3) years, women — 67 (39.6 %), aged (45.9 ± 5.2) years. The control group consisted of 20 practically healthy persons aged 19 to 56 years, whose average age was (38.2 ± 1.8) years. Among them were 11 men (55 %), and women — 9 (45 %).

The diagnosis of LC was made according to their complaints, medical history, laboratory (blood chemistry, identification of markers of hepatitis viruses B, C) and instrumental (ultrasound of the abdomen, fibroesophagogastroduodenoscopy, radioisotope, angiography) methods of research.

Dysbiosis was studied by a quantitative method of iden-

tifying microorganisms that have grown on a nutrient medium of agar, Saburo, Endo and 5% blood agar with the disposition to 1 gram of feces, taking into account the dose of the sawn material and the degree of dilution. Also, on the cup with 5% agar we noted the presence of hemolytic forms such as intestinal and coccal microflora, their parentage comparing to the total number of colonies that have grown, the correlation between the intestinal and coccal microflora. The presence of bifidobacteria was determined by the nature of their growth on the Blaurok medium and microscopy of swabs, colorized by the method of Gram. The number of bifidobacteria and lactobacillus in 1 gram of feces was determined by limiting dilution at which we observed their growth.

Disorders in the central nervous system were determined in patients with LC, who were conscious by the following tests: MMSE scale (Mini-mental State Examination) — a brief scale of mental state assessment, it is used worldwide for the assessment of cognitive functions; self-esteem scale (consisted by Ch.D. Spilberg and Y.L. Khanin) it allows to determine the level of anxiety at the moment through self-esteem (reactive anxiety as a state and personal anxiety as a permanent characteristic of a person); Beck's Depression Inventory — allows to characterize the patients emotional sphere; line copying test (labyrinth) — estimates the constructive apraxia; connection between numbers test — determines the possibility of cognitive movements implementation; symbol-digital test — used for determining the speed and accuracy of movements; numeric square — used to determine the speed of switching attention; Munsterberg's vocational test; number alignment — designed to assess arbitrary attention; memory for numbers methodic — designed to assess the short-term visual memory, its size and precision; image memory methodic — designed to study image memory.

In addition, to identify central nervous system violations among patients with LC we performed an electroencephalographic analysis.

All patients with LC on their diet background received basis treatment with hepatoprotectors, diuretics, desintoxication and vitamin therapy, β -adrenoreceptor blocking agents.

Patients with LC were divided into two groups depending on the intestine dysbiosis correction method. All patients received pebiotic in order to normalize their intestinal microbiocenosis — lactulose (duphalac) 15–20 ml 3 times a day for four weeks and tsyprinol (ciprofloxacin) 500 mg 2 times a day for 5 days. group I consisted of 70 patients, for the complex treatment they additionally received bifiform or bifiform complex probiotics (for patients with constipation) which comprises lactobacilli (*Lactobacillus rhamnosus* and *Lactobacillus acidophilus*), bifidobacterium (*Bifidobacterium*) and inulin (from root *Cichorium intybus*) 1 capsule 2 times a day for two weeks. group II consisted of 99 patients with cirrhosis who didn't receive probiotics, but received only lactulose by 15–20 ml 3 times daily. After 1 month from the beginning of the complex therapy patients with LC were conducted bacteriological stool culture the second time and the determination of HE severity level.

Analysis and processing of the results of patients examination were carried out using a computer system Microsoft Excel 7.0, statistical software package Statistica 6.0.

Results of the Studies and Their Discussion

After clinical and laboratory examinations of all patients with LC we divided them into classes of severity based on Child-Pugh, taking into account the level of bilirubin, albumin, prothombin index and the presence or absence of ascites and hepatic encephalopathy. Class A according to Child-Pugh included 73 (43.2 %) of patients, class B — 20 (11.8 %) of patients, class C — 76 (45.0 %) of patients.

Changes of the colon microbiocenosis in patients with LC and in the control group are presented in Table 1. Changes in anaerobic flora were characterized by a decrease of the level of bifidobacteria and lactobacilli among all patients with LC in both groups. The changes in aerobic flora among all patients with LC in both groups before the treatment were characterized by the decrease of the general number of *E.coli* — among 73 (73.7 %) patients in group II and among 42 (60.0 %) patients in group I. Low fermentative *E.coli* were determined among 80 (80.8 %) patients in group II and among 47 (67.1 %) patients in group I. hemolytic *E.coli* was determined among 17.2–21.4 % of patients, respectively. *Opportunistic enterobacteria* (mainly *Enterobacter aerogenes*, *Enterobacter cloacae*, *Citrobacter freundii*) were determined among 51 (51.5 %) patient in group II, and among 43 (61.4 %) patients in group I. Among 8.1–10.0 % of patients we found *Staphylococcus aureus* in titer of greater than 10³.

When comparing the indicators of microbiological examination of stool in two groups we can see the changes in microflora of the colon were diagnosed in patients of both groups with an approximately equal frequency of therapy (Table 1).

The conducted complex therapy was more effective in patients from group I who received in complex treatment both prebiotic and probiotic. After the second bacteriological examination of stool in patients with cirrhosis from group I in 57.1 and 61.4 % of cases the normalization of bifido-

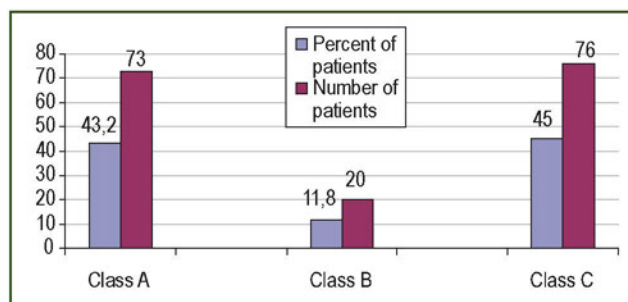


Figure 1 — Distribution of patients with LC depending on the severity level according to Child-Pugh

and lactobacteria was established ($p < 0.05$). In patients from group II, who received lactulose on the background of basic therapy of LC and did not receive probiotic, only in 25.3 and 27.3 % of patients the normalization of bifido- and lactobacteria was revealed. Those changes were accompanied by positive dynamics of aerobic flora indexes which was also more pronounced in patients from group I. Herewith, in patients from group I a decrease in total *E.coli* was noted in 22.9 % of cases, while in patients from group II — in 31.3% of cases. Low enzymatic *E.coli* decreased in patients from group I after treatment by 4.3 times, while in patients from group II — only by 3.1 times; hemolytic *E.coli* decreased in patients from group I by 3.0 times, and in patients from group II — only by 2.1 times; conditionally pathogenic enterobacteria in patients from group I by 3.9 times, and in patients from group II — by 2.9 times.

While assessing the effectiveness of the conducted treatment on the of intestinal dysbiosis severity in patients with cirrhosis it was found, that the most effective therapy was that among the patients group I, which received probiotic in combination with prebiotic. This therapy was better than lactulose monotherapy (patients with LC from group II). However, after the conducted treatment level IV dysbiosis hasn't been discovered in patients from group I, while in patients from group II it was observed in 2.0 % of the cases. Level III dysbiosis decreased by 11.7 and 12.1% in patients from group I and II. In patients with LC from group I level

Table 1 — Changes in colon microflora among patients with LC before the complex treatment, n (%)

Indicators	Control group (n = 20)	Group I (n = 70)		Group II (n = 99)	
		Before treatment	After treatment	Before treatment	After treatment
Bifidobacteria < 10 ⁷	2 (10.0)	70 (100.0)*	40 (57.1)**	99 (100.0)*	74 (74.7)
Lactobacilli < 10 ⁶	2 (10.0)	70 (100.0)*	45 (61.4)**	99 (100.0)*	72 (72.7)
Decrease in the general number of <i>E.coli</i>	2 (10.0)	42 (60.0)	16 (22.9)	73 (73.7)*	31 (31.3)**
Low fermentative <i>E.coli</i>	1 (5.0)	47 (67.1)*	11 (15.7)**	80 (80.8)*	26 (26.3)**
Hemolytic <i>E.coli</i>	0	15 (21.4)*	5 (7.1)	17 (17.2)*	8 (8.1)
Opportunistic enterobacteria	0	43 (61.4)*	11 (15.7)**	51 (51.5)*	17 (17.2)**
Fungi of Candida family	0	0	0	0	0
Number of pathogenic staphylococci	0	7 (10.0)	1 (1.4)	8 (8.1)	2 (2.0)

Notes: differences between indicants of the control group and patients with LC: * — $p < 0.05$; differences between indicants before and after treatment are accurate: ** — $p < 0.05$.

Table 2 – Time course of intestinal dysbiosis severity in patients with LC under the influence of complex treatment, n (%)

Degree of dysbiosis	Group I (n = 70)		Group II (n = 99)	
	Before treatment	After treatment	Before treatment	After treatment
Absent	–	19 (27.1)*	–	16 (16.2)*
I degree	3 (4.3)	31 (44.3)*	35 (35.3)	44 (44.4)
II degree	51 (72.9)	16 (22.9)*	30 (30.3)	22 (22.2)
III degree	12 (17.1)	4 (5.7)	27 (27.3)	15 (15.2)
IV degree	4 (5.7)	–	7 (7.1)	2 (2.0)

Note. Differences between indicants before and after treatment are accurate: * – $p < 0.05$.

I–II dysbiosis increased by 40.0–50.0 % ($p < 0.05$), while in patients from group II – only by 9.1–8.1 %. The absence of dysbiotic changes after the treatment was established 10.9 % more often in patients from group I compared with patients from group II. Thus, a combined usage of prebiotics in combination with probiotics on the background of basic therapy in patients with LC with the aim of correcting intestinal dysbiosis severity has proven to be more effective than monotherapy using prebiotics.

At the moment of admission to the hospital and after the course of complex treatment we assessed the neurological condition, mental and cognitive functions condition among patients with LC using psychometric testing, and also conducted an electroencephalographic study. While admitting to the day and night clinic we observed complaints of headache, dizziness, impaired memory and attention difficulties in performing practical skills, irritability, periodic apathy, anxiety almost among all patients in two groups.

After summing up the results of the conducted tests among patients with LC we found subclinical and clinical signs of HE before treatment. The change of the frequency of determining patients with complaints on cognitive and emotional disturbances before and after the treatment and their distribution according to the severity of HE are presented in Table 3.

The following fact draws our attention to the patients in group I, that in addition to standard treatment of LC also received prebiotic and probiotic, more evident positive dynamics of clinical manifestations of HE is observed. After a course of treatment there was no stage III encephalopathy observed in group I, while among the patients in group II it was diagnosed in 5.10 % of cases. Also, after the treatment of patients in group I clinical symptoms of HE were often absent compared to the patients of group II (37.1 and 24.2 %, respectively).

The data received demonstrates significant changes in the cognitive sphere, neurological and emotional status of patients with LC with signs of HE. In patients with HE evident changes in neurological symptomatology are caused by disorders of the brain blood supply in case of liver cirrhosis in conditions of dysmetabolic disorders and toxic influence of ammonia, and also in case of liver cirrhosis these pathological changes develop on the background of a defective microbiocenosis of the colon that was revealed in 100 % of examined patients with liver abnormality.

Thus, medical complex prebiotics (lactulose) in combination with probiotic bifiform and bifiform complex have proven to be effective in treatment of patients with LC. Along with improved indexes of microbiological examination of stool among the examined patients we also observed a significant positive dynamic of clinical manifestations of HE (decrease in cognitive disorders, improvement of mental and emotional condition, movement disorders) among patients with LC that were confirmed by positive dynamic of the conducted neuropsychometrical testing and electroencephalographic study, compared to patients who didn't receive probiotic in terms of complex treatment, but were only receiving lactulose.

Conclusions

1. We can observe evident disorders of the colon microbiocenosis among patients with liver cirrhosis with signs of hepatic encephalopathy.
2. A combined therapy with the use of prebiotics in combination with probiotics is a more effective method for intestinal dysbiosis treatment in patients with cirrhosis than monotherapy using prebiotics.
3. Colon microflora correction using medical complex prebiotics (dufalac) in combination with probiotics (bifiform or bifiform complex) in patients with cirrhosis promotes marked reduction of clinical manifestations of hepatic encephalopathy.

Table 3 – Distribution of patients with LC according to severity of HE before and after treatment, n (%)

Stages of hepatic encephalopathy	Group I (n = 70)		Group II (n = 99)	
	Before treatment	After treatment	Before treatment	After treatment
Absent	–	26 (37.1)*	18 (18.2)	24 (24.2)
Latent	37 (52.9)	20 (28.6)*	21 (21.2)	18 (18.2)
Stage I	19 (27.1)	18 (25.7)	10 (10.1)	19 (19.2)
Stage II	12 (17.1)	6 (8.6)	38 (38.4)	33 (33.3)
Stage III	2 (2.9)	–	12 (12.1)	5 (5.1)

Note. Differences between indicants before and after treatment are accurate: * – $p < 0.05$.

References

1. Звягинцева Т.Д. Эффективное лечение хронических диффузных заболеваний печени // *Здоров'я України*. — 2007. — № 11–12 (168–169). — С. 50–51.
2. Маевская М.В. Возможности применения пробиотиков в гастроэнтерологии // *Российский журнал гастроэнтерологии, гепатологии и колопроктологии*. — 2009. — № 5. — С. 65–72.
3. Печінкова енцефалопатія: особливості медикаментозного та дієтичного лікування / Н.В. Харченко, Г.А. Анохіна, Н.Д. Опанасюк та ін. // *Сучасна гастроентерологія*. — 2010. — № 6 (56). — С. 68–72.
4. Степанов Ю.М. Лікування алкогольної хвороби печінки // *Здоров'я України*. — 2007. — № 20/1. — С. 90.
5. Целесообразность и эффективность включения эссенциальных фосфолипидов в комплексное лечение больных циррозом печени с начальными проявлениями печеночной энцефалопатии / С.В. Данилюк, Г.И. Лысенко, Е.Ф. Вакуленко и др. // *Новости медицины и фармации*. — 2009. — № 294. — С. 14–17.
6. Iwakiri Y. The molecules. Mechanisms of arterial vasodilatation observed in the splanchnic and systemic circulation in portal hypertension // *J. Clin. Gastroenterol.* — 2007. — Vol. 41 (suppl. 3). — P. 288–294.
7. Pande C. Small intestinal bacterial overgrowth in cirrhosis is related to severity of liver disease / C. Pande, A. Kumar, S.K. Sarin // *Aliment. Pharmacol. Ther.* — 2009. — Vol. 29, № 12. — P. 1273–1281.

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Сирчак Е.С.

ГВУЗ «Ужгородский национальный университет», медицинский факультет

МЕТОДЫ КОРРЕКЦИИ ДИСБИОЗА КИШЕЧНИКА И ИХ ВЛИЯНИЕ НА ДИНАМИКУ КЛИНИЧЕСКИХ ПРОЯВЛЕНИЙ ПЕЧЕНОЧНОЙ ЭНЦЕФАЛОПАТИИ У БОЛЬНЫХ ЦИРРОЗОМ ПЕЧЕНИ

Резюме. Представлены результаты комплексного обследования 169 больных циррозом печени с проявлениями печеночной энцефалопатии. Обнаружена более выраженная положительная динамика клинических проявлений печеночной энцефалопатии у больных (n = 70), которым

для коррекции микробиоценоза толстого кишечника кроме пребиотика в составе комплексной терапии назначали и пробиотики.

Ключевые слова: цирроз печени, печеночная энцефалопатия, дисбиоз кишечника, пребиотик, пробиотик.

Сірчак Е.С.

ДВНЗ «Ужгородський національний університет», медичний факультет

МЕТОДИ КОРЕКЦІЇ ДИСБІОЗУ КИШЕЧНИКА ТА ЇХ ВПЛИВ НА ДИНАМІКУ КЛІНІЧНИХ ПРОЯВІВ ПЕЧІНКОВОЇ ЕНЦЕФАЛОПАТІЇ У ХВОРИХ НА ЦИРОЗ ПЕЧІНКИ

Резюме. Наведені результати комплексного обстеження 169 хворих на цироз печінки із проявами печінкової енцефалопатії. Виявлено більш виражену позитивну динаміку клінічних проявів печінкової енцефалопатії у хворих (n = 70), яким для ко-

рекції микробиоценозу товстого кишечника крім пребіотика в комплексній терапії призначали і пробіотики.

Ключові слова: цироз печінки, печінкова енцефалопатія, дисбіоз кишечника, пребіотик, пробіотик.