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ASSESSMENT OF LIPID PROFILE OF PERITONEAL FLUID FOR DIFFERENTIAL ASCITES DIAGNOSTICS

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The problem of differential ascites diagnostics in different patients remains urgent for modern surgery. Apart from well known methods, authors propose an original algorithm for assessment of ascetic fluid. The main element of such studies is the determination of a lipid spectrum of ascetic fluid and content of lipid components in cell structures. 260 samples were examined which were obtained from patients having examination and treatment in the General Surgery Clinic of the Kirov Military Medical Academy (St. Petersburg). For examination of ascetic fluid, the methods of cytological analysis of ascitic fluid centrifugate were used, its cytochemical examination, as well a complex of biochemical methods: determination of the total protein, amylase, chylomicrons, triglycerides; bacterial inoculation was made. It has been shown that the ratio of chylomicrons, lipoproteins and cytosis permits a more precise differentiation of ascites type which influences the selection of further treatment program especially in chyloperitoneum and lymphatic ascites.

Keywords: *ascites, chylomicrons, lipoproteins, chyle peritonitis.*

Ascites is a pathological fluid accumulation in the abdominal cavity. The most common cause of ascites is liver cirrhosis (LC) (81% of cases) [1-3] and cancer diseases (14%) [4]. Such causes of ascites as cardiac failure, tuberculosis, pancreatic diseases do not exceed 5% rates [2, 5]. An important condition for a favorable prognosis in treatment of a patient with ascetic syndrome is a timely diagnostics.

A modern clinician has not only methods of physical examination at his disposal, but also much more sensitive, reliable, specific instrumental methods of visualization of even insignificant fluid amounts in the abdominal cavity. Thus, it is possible to detect ascetic syndrome even on the earliest (preclinical) stages, sometimes at the first manifestation. A screening, routine method is the

ultrasound examination (USE) [3, 6, 7]. USE is widely available, highly informative, mobile, economically feasible for the wide use, non-invasive, and easily reproducible if a dynamic control is required. Computerized tomography and magnetic resonance imaging also indicate the fluid amount in the abdominal cavity, but do not provide information about its nature. Diagnostic puncture and laparocentesis allow to assess visually the nature of the abdominal content, to make its cytological, laboratory examination.

The differential diagnosis of diseases leading to ascites presents is rather difficult [3]. Modern guidelines on differential diagnostics of ascetic syndrome usually discuss pathogenetic mechanisms, clinical picture and a patient's examination [2, 4], hereby less attention is paid to laboratory and morphological (cytological) diagnostics [8]. The algorithm of examination of patients with ascites is not so far defined and should be supplemented with morphological methods.

Aim of the work: to specify differential and diagnostic criteria for characterization of ascetic fluid.

Materials and Methods

A complex examination of 260 patients with ascites was made. On admission to the hospital each patient had a diagnostic puncture of the abdominal cavity with laboratory examination of ascetic fluid (AF).

AF evacuated during the diagnostic puncture was subject to examinations which may indicate the origin of ascites: determination of cytosis, sediment microscopy with determination of percentage of cell elements. Cytological preparations were made from the ascetic fluid according to the common practice [9]. 10 ml of AF were centrifuged at 1000 rpm. The supernatant fluid was removed from the centrifuged tube within 5 minutes, and the sediment was fixed with 10% neutral formalin within an hour. After that, smears were prepared. Cell composition of smears was examined after Giemsa staining. Cell elements found in microscopy were differentiated to lymphocytes, monocytes, neutrophils, castration cells, considering their morphological peculiarities (size of cells, presence of nuclei).

To assess fat inclusions, Sudan III dye was used which stained the preparations bright orange. The preparations were further examined under binocular microscope "Bimam R-11 LOMO" (Russia). Semi-quantitative assessment of fat inclusions was made.

For a cytological expedient analysis, the simplified method for making preparations was used – without counterstaining with hematoxylin; in such a case, the preparations were studied under low magnifications – x100-200. During the study, it was found that the presence of droplets in the liquid phase is not generally typical for serous ascetic fluid and ascites-peritonitis. Fat inclusions were represented by single, discrete, small-sized and non-fusing droplets.

Biochemical parameters of patient ascetic fluid – total bilirubin, total protein, creatinine, urea nitrogen, amylase – were determined on an automatic analyzer "Technicon"@ SMA 12/60, USA. Percentages of protein fractions were determined by cellulose acetate electrophoresis. In each case, the ascetic fluid was inoculated on enrichment medium for sterility control. Ascetic fluid was placed into a sterile test tube in the amount of 8-15 ml (in this case, the fluid acts as a transport medium), the tube was closed with a sterile rubber cork and sent to the laboratory. A strict adherence to the mentioned conditions for sampling and delivery of materials allowed to preserve aerobic and anaerobic microflora in the viable state.

Differential and diagnostic criteria were determined by a pair-wise comparison of each diagnostic criterion using Mann-Whitney test, and an alternative hypothesis was accepted concerning the existence of differences with the level of statistical significance $p=0.01$. For statistical calculations Excel XP, Access XP and Statistica 5.0 programs for personal computer were used.

Results and Discussion

After examination of 260 samples of the peritoneal fluid 3 groups of patients were with different nature of ascetic syndrome: serous ascites (SA), ascites-peritonitis (AP) and chyloperitoneum (ChP). The complex study of milky-colored peritoneal fluid showed a significant difference in lipid composition in 10 patients which allowed to allocate them to the fourth group of patients with lymphatic

ascites (LA). The puncture in patients of this group resulted in peritoneal fluid of milky color having the composition similar to lymph, but with a lower content of chylomicrons and triglycerides (fig. 1). Direct communication between the peritoneal cavity and the lumen of lymphatic vessels was excluded. It is known that decompensated LC goes with lymphatic hypertension, and the cause for lymph penetration into the peritoneal cavity lies in a high permeability of lymphatic vessels. Lymph enriched by in protein, VLDLP and HDLP penetrates the peritoneal cavity through the thin-walled lymphatic vessels, and also through Glisson's capsule from the dilated lymph collectors below the liver [10-12]. In the settings of dynamic insufficiency of the outflow of lymph due to the generalized blockage of lymphatic vessels, lymph nodes sinuses and the thoracic lymph duct by a tumor mass, microfistulas in lymphatic capillaries are formed opening into the peritoneal cavity [13]. Lipoproteins of the intestinal genesis partly diffuse in the intercellular space throughout the thickness of the intestinal wall and, having reached the serous membrane, enter the peritoneal cavity [14]. Based on our material of 10 observations, hepatic cirrhosis was diagnosed in 2 cases, and in 8 cases – malignant neoplasms. The full-scale lipid profile of lymphatic and serous ascetic fluid from the peritoneal cavity of patients with HC showed the highest concentration of VLDLP. Chylomicrons in lymph amounted to 8.75±1.2% and had the lower proportion in serous ascetic fluid (1.1±0.7%).

The contents of the abdominal cavity in ChP reliably differ from that in SA and LA in lipid composition. A full-scale lipid profile showed higher concentration of chylomicrons (20.7±3.9%), and lower concentrations of VLDLP and HDLP. All patients with ChP showed a direct communication between peritoneal cavity and the lumen of lymphatic vessels.

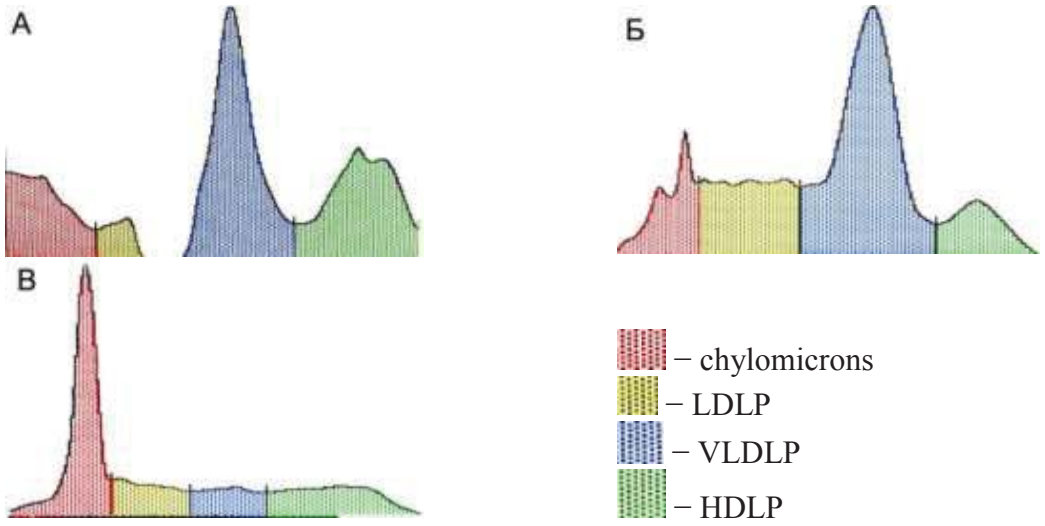

Fig. 1. Lipid profiles of peritoneal fluids:
A − serous ascites; B − lymphatic ascites; B − chyloperitoneum

Milky color of ascetic fluid with a high protein concentration protein was an indicator for examination of its lipid spectrum. The full-scale lipid profile showed that in ChP triglycerides amounted to 4.17±0.9 mmol/l, and chylomicrons – 23.5±6.2%. The comparison with SA: triglycerides 0.72±0.29 mmol/l, chylomicrons 1.2±0.76%. The differences between the mentioned pairs of values are reliable (p=0.001).

The content of chylomicrons in the peritoneal fluid in the range from 3.5% to 13.5% indicated LA. Concentration of chylomicrons above 13.5% confirmed the presence of ChP.

Cytological and cytochemical (Sudan III) examination of smears (n=101) of AF centrifugate showed different cell composition and lipid content of AF in different types of ascites. It was found that the presence of fat droplets in liquid phase in serous ascetic fluid

and in ascites-peritonitis was not, in general, typical. They were seen as single discrete small-sized non-confluent droplets (fig. 2a). The smears obtained from patients with lymphatic ascites contained a larger number of fat components were. Fat droplets were also scattered in all fields of vision, were small and did not fuse with each other. However, the conglomerates of desquamated mesotheliocytes has a positive Sudan III staining (fig. 2b).

A principally different picture was seen in ChP – high number of lipid droplets of different diameter in all fields of vision – from 0.3-0.5 μm to 0 μm (fig. 2c).

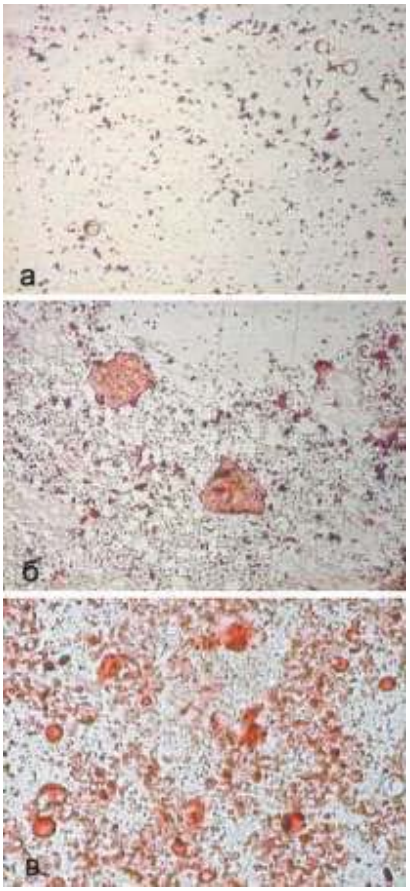

Fig. 2. Smears of peritoneal fluid centrifugate:
a – SA; c. – LA; c – ChP. Staining: Sudan III. Magnification: a – ×200, b, c. – ×100

To make the study objective, authors proposed the semi-quantitative scale for assessment of the content of lipid components in the cytological smears of the peritoneal fluid:

1 score: single small (0.1-0.3 μm) fat inclusions identified in some fields of vision scattered beyond cells; intracellular droplets (up to 0.3 μm) were seen only under high magnification in macrophages;

2 scores: isolated small fat droplets (up to 0.3 μm) beyond cells in all fields of vision; intracellular – small single cells in macrophages;

3 scores: numerous small and moderate fat droplets (0.3-0.5 μm) beyond cells in all fields of vision; intracellular – numerous small droplets in macrophages and in desquamated mesotheliocytes;

4 scores: all fields of vision were covered with confluent fat droplets of different size (0.5-1.0 μm), intracellular – diffused confluent fat droplets of different size (up to 1.0 μm).

Depending on the concentration of lipid components, patients were allocated to different groups per the score scale in the following way (tab. 1).

During the cytological examination of SA smears (n=60) stained with Sudan III, 76% of samples were assessed as 1 score. In some fields of vision, single small fat inclusions scattered beyond cells were present. The

Table 1

Allocation of patients per the score scale of assessment of lipid component content in peritoneal fluid

Ascites (n=101)	Score scale for assessment of lipid components content			
	1 score	2 scores	3 scores	4 scores
Serous ascites (n=60)	50	12	4	0
Ascites-peritonitis (n=19)	13	4	2	0
Lymphatic ascites (n=10)	0	2	8	0
Chyloperitoneum (n=12)	0	1	3	8

different picture was typical of ChP. The microscopy of 67% of smears revealed that confluent fat droplets of different size were scattered in all fields of vision (4 scores). Based on allocation of various groups of patients per score scale it followed that if peritoneal fluid by the content of lipid components was assessed as 4 scores, it meant ChP. In LA patients, 75% of peritoneal fluid smears were assessed as 3 scores. However, the small number of observations did not allow permit to differentiate between LA and ChP. 3 scores points suggested the presence of lymphatic component in the peritoneal fluid. And if smears were assessed as 1 score allows to exclude LA and ChP. If scarce small fat droplets (less than 0.3 μm) were detected beyond cells in all fields of vision and small single droplets in macrophages (point 2), it was not possibly to differentiate evidently the ascites type, and the examination of lipid composition was required.

Thus, cytological and cytochemical analysis made it possible to differentiate the ascites type as soon as possible (expedient analysis). LA patients were allocated to a separate group, since LA was significantly different from ChP not only by lipid composition, but had a quite different origin.

The average cytosis index in ChP was 604±101 cells in 1 μl. For comparison: the average cell count was 320±140 in 1 μl in serous ascetic fluid of patients with hepatic cirrhosis. The significant reliable difference was shown between ChP and SA and AP (p= 0.001), however, it was impossible to make differential diagnostics of LA only by cytosis index, since the difference was not significant (p=0.2).

As well, the relative lymphocyte count in AF plays a big role. As part of calculation of cell element percentage in serous ascetic fluid, lymphocytes accounted for 56.8±15.2%, and neutrophils were a significantly lesser proportion – 9.5±6.6%. Percentages of cell elements of the fluid in ChP were: segmented neutrophils 20.2±4.7%, lymphocytes 80.6±6.7%. The comparison of all groups of patients for the lymphocyte count revealed significant differences (p=0.001).

Table 2 presents the main laboratory parameters of the peritoneal fluid in various groups of patients.

Table 2

Main parameters of peritoneal fluid in various groups of patients

Ascites (N=260)	Cell and Biochemical Composition of Ascetic Fluid				
	Cytosis (cells per 1 μl)	Lymphocytes (%)	Triglycerides (mmol/l)	Chylomicrons (%)	Total protein (g/l)
SA (n=204)	320±140	56.8±15.2	0.64±0.33	1.1±0.7	17.6±5.1
AP (n=27)	1312±623	20.9±7.1	0,59±0.25	1.2±0.6	19.5±4.2
LA (n=10)	420±106**	71.7±6.1	3.32±0.96	8.8±1.2	28.4±6.4
ChP (n=19)	604±101*	80.6±6.7*	4.9±1.12*	20. 7±3.9*	35.3±7.8

Note: * – p=0.001 (relative to SA), ** – p=0.2 (relative to ChP)

To develop differential and diagnostic criteria, a linear model was made with centroids reflecting the mean values of parameters for different groups of patients. The values of confidence intervals were plotted over the distance of 2 sigmas and encircled with the vertical dimension (on Y axis) reflecting the number of observations. With the absence of intersection for more than one third of the confidence intervals (spheres), a separation line was drawn in the middle of the distance between centroids showing the diagnostic criterion for the respective parameter. The location of laboratory parameters within the range of values limited by the line allowed to allocate a patient to this or another group with 95% reliability.

A linear statistical model was plotted for parameter of lymphocyte count on which the mean values (centroids) were established for different groups of patients. Double sigmas in all groups did not cross and did not have common values. Variations of this parameter among the patients' groups were reliable. Separating points are positioned in intersection point 2δ between groups of SA and LA, LA and ChA patients – 72% and 78% (fig. 3).

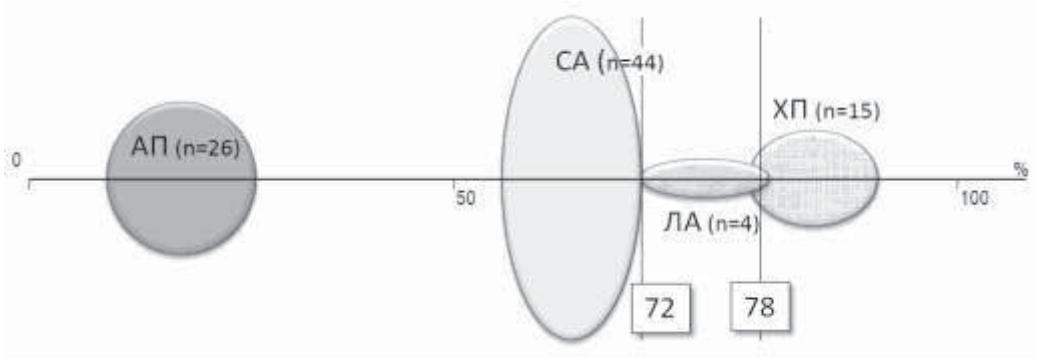

Fig. 3. Lymphocyte count in ascetic fluid in different groups of patients
(linear statistical model with centroids)
АП – AP; СА – SA; ЛА – LA; ХП – ChP

Thus, relative lymphocyte count below 72% indicates serous ascites (excluding AP), in the range from 72% to 78% inclusive – LA, above 78% – ChP.

The necessary and sufficient condition to diagnose AP was the increase of the cytosis in AF above 800 cells per 1 μl and neutrophilia above 250 cells per 1 μl. To exclude contamination of peritoneal fluid, its bacterial control was made in each case by its inoculation on the enrichment medium. Despite the cloudy character of the fluid in LA and ChP, no microbial growth was noted.

Further differential diagnostics of ascites included determination of: the presence of atypical cells (present in 55% of cases in hepatocellular carcinoma, in 22% in metastatic liver disease); aerobic and anaerobic microflora; serum-ascetic gradient (examination of serum albumin/ascitic albumin; parameter ≥ 1.1 confirmed the relationship of ascites with portal hypertension, the method sensitivity was 80%); amylase (ascetic/serum gradient > 0.4 indicated pancreatic genesis of ascites, perforation of the hollow organ); bilirubin (differential diagnostics with perforation of billiary tract); lactate dehydrogenase (differential diagnostics with inflammatory process in the abdominal cavity, oncopathology). Based on the laboratory examination of AF, the ascites type was determined in all patients (n=260) as soon as possible.

Table 3 shows the criteria of ascetic fluid in various diseases.

Table 3

Differential and diagnostic criteria for ascites in various diseases

Cause of ascites	Laboratory parameters of ascetic fluid						
	Color of fluid	Total protein	Lympho-cytes	Chylomicrons	Triglycerides	Sudan III stain-ing of smears	Serum-ascetic albumin gradi-ent, 10 g/l
Hepatic cirrhosis	straw-yellow, clear	<23 g/l	<72%	<3.5%	<1.8 mmol/l	1 point	>
	milky yellow	≥ 23 g/l	72-78%	3.5-13.5%	≥ 1.8 mmol/l	3-4 points	>
Lymphatic fistula	milky white	≥ 23 g/l	>78%	≥ 13.5%	≥ 1.8 mmol/l	3-4 points	<
Ascites-peritonitis	cloudy yellow	<23 g/l	<72%	<3.5%	<1.8 mmol/g	1 point	<
Pancreatogenic	cloudy, hemorrhagic, brown	>23 g/l	>78%	3.5-13.5%	≥ 1.8 mmol/l	1 point	<
Nephrogenic	straw-yellow, clear	<23 g/l	<72%	<3.5%	<1.8 mmol/l	1 point	<
Heart failure	straw-yellow, clear	≥ 23 g/l	<72%	<3.5%	<1.8 mmol/l	1 point	>
Tuberculosis, tumor of the abdominal cavity	straw-yellow, clear	>23 g/l	<72%	<3.5%	<1.8 mmol/l	1 point	<
	milky, hemorrhagic, cloudy	≥ 23 g/l	72-90%	3.5-13.5%	≥ 1.8 mmol/l	3-4 points	<

Such a screening examination of peritoneal fluid allowed the authors to determine the cause of ascetic syndrome in all patients. The examination of ascetic fluid for chylomicrons, triglycerides and content of lipid components confirmed the hypothesis on lymphogenic origin of ascites and allowed the differential diagnostics of ChP with lymphatic ascites (rationalization proposal N10030/5 dated 25.10.2016). A surgical approach to treatment for resistant ascites in patients with hepatic cirrhosis should be selective with regards to the leading pathogenetic factors of ascites and the degree of decompensation of the underlying disease. Early diagnostics of ascetic syndrome allowed to start pathogenetic treatment in all patients and to determine indications for surgery as soon as possible.

The identification of chyloperitoneum by clinical signs, as a rule, was difficult in 100% of cases [15, 16]. It can be stated that the results of laboratory and cytological examinations of the peritoneal fluid were of priority in diagnostics of chyloperitoneum

Thus, according to the author, the most important element in examination of patients with ascites is to determine a lipid spectrum of the ascetic fluid and content of lipid components in cell structures. The ratio of chylomicrons, lipoproteins, cytosis allowed to differentiate more precisely ascites type and to determine further treatment program, especially in the event of ChP and LA.

Such a screening examination of AF allowed to differentiate ascites associated with other diseases. Thus, tuberculosis-associated peritonitis was revealed in five patients, ascites in ten patients was associated with cardiac diseases and cardiac failure. 25 patients showed a malignant neoplasm of IV stage with carcinomatosis of the peritoneum.

The range of obligatory examinations of ascetic fluid was specified for identification of the disease nature, timely diagnostics of ascites-peritonitis. The percentage of cell composition of AF, in particular, neutrophilia, played the key role in diagnostics of CP. The presence of infectious complications as ascites-peritonitis required effective therapeutic approaches that included a full-scale sanitation of the infectious focus (complete evacuation of ascites), endolymphatic and intravenous antibacterial therapy, and lymphosorption as an effective detoxication method. Early diagnostics allowed to achieve positive treatment results in more than the half patients (13 of 19) with ascites-peritonitis.

Conclusion

The biochemical and cytochemical examination of ascetic fluid is found to be an important method of differential diagnostics of ascites type. The full-scale lipid profile demonstrating significant differences in the proportion of different lipid components of transudate in various ascites is of a particular importance. Thus, if ascites is detected in a patient, together with conventional instrumental examination the methods of diagnostic puncture of the peritoneal cavity should be used with subsequent laboratory, cytological and cytochemical examination of the ascetic fluid.

The complete examination of ascetic fluid allows to identify the cause of ascites in a short time, to make the diagnosis and to start timely pathogenetic treatment.

Authors have no conflict of interest to declare.

References

1. Podyimova SD. *Bolezni pecheni: rukovodstvo 4-e izd., pererab. i dop* [*Diseases of the liver: guide. 4th ed., pererab. and additional*]. Moscow: Medcine; 2005. 768 p. (in Russian)

2. Arroyo V, Gines P, Rodes J, Schrier RW. *Ascites and renal dysfunction in liver disease: pathogenesis, diagnosis, and treatment*. Malden, Mass.: Blackwell Science. 2005.

3. Fuger K. Aszites. In: *Differenzial diagnose internistischer Erkrankunger nach Leitsymptomes.* Hrsg. Aufl. Munchen-Jena: Urban & Fischer; 2003. P. 63-80.

4. Glickman R. M. Abdominal swelling and ascites. In: *Harrison's Principles of Internal Medicine*. New York: McGraw-Hill Med. Publ. Div; 2005. P. 243-6.

5. Gerbes AL, Gülberg V, Wiest R, Sauerbruch T, Gerbes A. German Society for

Digestive and Metabolic Diseases (DGVS) *Diagnosis and treatment of ascites, spontaneous bacterial peritonitis, and hepatorenal syndrome in cirrhosis: S3-guideline*. 2010. P. 43-56.

6. Moore K. Guidelines on the management of ascites in cirrhosis. *Gut.* 2006; 55: 1-12.

7. Gulov MK, Kalmyikov EL, Zardakov SM, Muhabbatov DK, Sadriev ON. Ehinokokkoz pecheni: rol kompyuternoy tomografii i morfologicheskoy diagnostiki sostoyaniya tkani pecheni [Echinococcosis of the liver: the role of computed tomography and morphological diagnosis of liver tissue]. *Rossijskij mediko-biologicheskij vestnik imeni akademika I.P. Pavlova* [*I.P. Pavlov Russian Medical Biological Herald*]. 2016; 4: 104-11. (in Russian)

8. Andreeva IV, Vinogradov AA. Perspektivyi ispolzovaniya sovremennyih metodov vizualizatsii v morfologicheskih i eksperimentalnyih issledovaniyah [Prospects of using modern imaging methods in morphological and experimental studies]. *Nauka molodykh (Eruditio juvenium)* [*Science of young (Eruditio Juvenium)*]. 2015; 4: 56-69. (in Russian)

9. Sarkisov DS, Perov YuL., red. *Mikroskopicheskaya texnika: rukovodstvo dlya vrachej i laborantov* [*Microscopic technology: a guide for doctors and laboratory technicians*]. Moscow: Medcine; 1996. 544 p. (in Russian)

10. Nazyirov FG, Horoshaev VA, Devyatov AV. Osobennosti portalno-limfaticheskoy gipertenzii i hirurgicheskoe lechenie bolnyih tsirrozom pecheni s rezisientnyim astsitom [Features of portal-lymphatic hypertension and surgical treatment of patients with cirrhosis of the liver with resistant ascites]. *Vestn. hirurgii im. I.I. Grekova* [*Herald surgery them I.I. Grekova*]. 1999; 142 (2): 104-6. (in Russian)

11. Sedova TN. *Hiloreya kak sindrom zabolevaniy i povrezhdeniy limfaticheskih protokov: (Diagnostika, klinika i lechenie)* [*Chilorea as a syndrome of diseases and damages of lymphatic ducts: (diagnosis, clinic and treatment)*]: dis. doct. (Med. Sci.). Moscow; 1997. (in Russian)

12. Rector W. Spontaneous chylous ascites of cirrhosis. *J. Clin. Gastroenterol*. 1984; 6: 369-72.

13. Kaas R, Rustman LD, Zoetmulder FA. Chylous ascites after oncological abdominal surgery: incidence and treatment. *Eur. J. Surg. Oncol.* 2001; 27: 187-9.

14. Aalami OO, Allen DB, Organ CH. Chylous ascites: a collective review. *Surgery*. 2000; 128: 761-78.

15. Chichetka AA, Biryukova LN. Hileznyiy peritonit, simuli-rovavshiy ostryiy appenditsit [Chileous peritonitis, simulating acute appendicitis]. *Vestn. hirurgii im. I.I. Grekova* [*Herald surgery them I.I. Grekova*]. 1991; 2: 44. (in Russian)

16. Markov IA. Hileznyiy peritonit [Chylious peritonitis]. *Med. zhurn. Chuvashii* [*Medical Journal of Chuvashia*]. 1995; 3-4: 98-9. (in Russian)

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