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ORIGINAL RESEARCH

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Risk factors in the development of acute appendicitis complications

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Abstract

Background: To study the risk factors of the development of acute appendicitis (AA) complications in adults in order to improve the results of surgical treatment.

Material and methods: The research included 449 patients with AA treated surgically during the years 2015-2017 divided into 2 groups: 117 patients who were admitted with complicated appendicitis (intra- and extraabdominal complications) and 332 patients with non-complicated AA were randomly selected from the same period. The rate and characteristic of the complications evolved during the pre- and postoperative period in these two groups were specified and analyzed.

Results: In the acute complicated appendicitis group (CAA), there was a predominance of women with a ratio of 1.60 versus 1.26 in the uncomplicated acute appendicitis group (NAA). The proportion of people aged> 60 years was significantly higher in the case of CAA-23.1% (n=27), while in uncomplicated AA it was only 3.9% (n=13). In the case of AA complications, there was an emphasis on late addressing, the debut-addressing term being higher compared to uncomplicated AA. The low socio-economic status has a significant negative impact on the evolution of AA and its complications, as well as on the results of appendectomy. Thus, uninsured patients (n=59, 49.6%) formed almost half of CAA group. Associated comorbidities were established in 76 or 16.9% of cases, respectively in CAA-21.4% vs 15.4% in NAA group. In summary we note that the presence of associated uncorrected comorbidities has an obvious negative impact on the development of AA.

Conclusions: Our findings suggest that clinical assessment is most important for identifying individuals at risk of developing complications of AA and the above-mentioned risk factors are useful for emergency surgical decisions.

Key words: Complications of acute appendicitis, risk factors.

Introduction

Acute appendicitis (AA) is the most common surgical abdominal emergency and its lifetime risk is 8.6% for men and 6.7% for women, yet the risk of undergoing appendectomy is higher for women (12 vs. 23%) [1-3]. In developed countries the incidence of AA is 400-520 cases per 100000 population, while in poorly developed countries it is 100-320 cases per 100000 population [4, 5]. In the Republic of Moldova the AA frequency denotes 220 cases per 100000 inhabitants [6].

Apparently simple as a pathology, AA does not always find an easy solution, and by its complications it can sometimes generate situations requiring complex therapeutic features. Although surgical treatment is well tolerated by most patients, it is associated with a risk of postoperative complications in 2%- 23% of cases [7, 8]. In addition, notwithstanding the implementation of miniinvasive techniques, it is noted that about 3% of patients who underwent an appendectomy with or without laparoscopy [9] were repeatedly admitted to hospital with a diagnosis of intestinal occlusion, cataloged as a tardive post-operative complication, often a long time after primary surgery [10].

AA complications may evolve either as a natural stage in the pathophysiological process of vermicular appendicitis with plastron formation or depending on wall integrity with its perforation and triggering of generalized or localized appendicular peritonitis [11,12]. This type of complications can be called intraabdominal complications, according to the literature; they have an incidence of about 5-7% cases in the developed countries and up to 30% in the case of countries with poor socio-economic status [13]. Notwithstanding the general decrease in the morbidity rate through AA, an impressive number of studies demonstrate the stability of these rates over the past decades [14-16].

Another group of complications are caused by the purulent processes in the postoperative wound (suppuration, abscess, ligature fistula) at a rate of 18-20%, they do not have a certain tendency to diminish [17,18]. Even despite the frequent and prolonged use of antibacterial drugs for prophylaxis of postoperative wound complications, the frequency of appearance remains at a constant level [19]. In general, post-appendectomy complications rates are usually within 10%-19% range for acute AA without perforation and reaches to 12%-30% for perforated AA [20-24]. Perforation

increases the AA mortality rate from 0.0002% to 3% and causes an increase in morbidity from 3% to 47% [25-27].

Thus, we can see that, despite all the surgical progress achieved, the AA complications remain a problem that still requires increased attention. This determines the need to specify risk and prognostic factors in the development of AA complications for their prophylaxis and improvement of surgical treatment results.

The purpose of the paper: to study the risk factors of the development of acute appendicitis complications in adults in order to improve the results of surgical treatment.

Material and methods

Study Design. The study includes a retrospective analysis of clinical material focused on the estimation of the incidence, character and risk factors of the development of AA complications. We designed a case-control study to compare different perceived risk factors among patients with complicated or uncomplicated AA. The point of reference was the analysis and evaluation of the anamnestic disease, the clinical picture, the laboratory and instrumental data established preoperatively in connection with the morphological changes of the vermicular appendix performed by morphopathological examination of the operative piece. The rate and characteristic of the complications evolved during the pre- and postoperative period in the analyzed patients were specified. For the purpose of assessing the microbial etiological factor, the patients of the study group were subjected to the bacteriological examination which included seeding on aerobic culture media as a source of collected material for samples collected during diagnostic laparoscopy, surgical intervention, pathological leakage from the safety drains or postoperative wound.

Participants and data collection. The research included 449 AA patients treated surgically by classical approach during the years 2015-2017 at Surgical Clinic Nr. 2 and Nr. 3 of Surgery Department Nr. 2 of Nicolae Testemitsanu State University of Medicine and Pharmacy. The analyzed data were extracted from: clinical observation sheets, operative protocols, histopathological examination bulletins and electronic database Sfinta Treime Municipial Hospital (code K35.0-3, K35.9). An individual clinical research file was complemented for each patient. The terms of evolution of the disease were analyzed, the studied cases being classified in 3 time intervals: addressing up to 6 hours after the onset of clinical manifestations, 7-24 hours; more than 24 hours. At the same time, attention was drawn to the socioeconomic status of the patient (insured or uninsured).

The Charlson co-morbidity index (CCI) was used in order to establish the aggressive synergistic action of chronic co-morbidities on AA evolution and its complications (tab. 1).

This score was evaluated in each patient with the consecutive specification in the following groups according to the stage of compensation of the associated condition: √CCI – I (lack of co-morbidity) – 0 points; √CCI- II (compensated co-morbidity) – 1 point, √CCI III (uncompensated co-morbidity) 2-3 points, √CCI-IV (decompensated co-morbidity) >4 points, √CCI-V (decompensated co-morbidity refractory to any treatment).

Table 1

The value and conditions considered in the Charlson co-morbidity index

Value	Conditions
1	Myocardial infarction, congestive heart failure, peripheral vascular pathology, cerebrovascular disease, dementia, chronic pulmonary pathology, connective tissue pathologies, ulcerative disease, non-severe compensated hepatopathy.
2	Diabetes, hemiplegia, moderate or severe kidney pathologies, complicated diabetes with internal organ damage, leukemia, lymphoma.
3	Moderate or severe hepatopathy.
6	Non-metastatic solid tumors, Metastatic tumors, AIDS.

Definitions. In order to standardize the results we divided all patients into 2 groups: Non-complicated acute appendicitis (NAA) group**,** consisting of patients without the progression of complications of cataract and phlegmonous AA without perforation; and complicated acute appendicitis (CAA) group, consisting of patients with complications of acute appendicitis. It included phlegmonous with perforation AA, gangrenous AA, appendicular infiltration or periappendicular abscess, local, diffuse or generalized peritonitis; septic complications of postoperative wound. The same group included 4 patients who were previously operated for AA, who at distant point had acute adherent intestinal occlusion, treated surgically, 1 patient with external intestinal fistula, 6 patients with ligature fistula, and 5 with post-apendectomy hernias.

Statistical analysis. For the purpose of processing quantitative summaries, statistical software was used. Pearson's chi-square and independent sample t tests were used to compare categorical and continuous variables as indicated. A p -value ≤ 0.05 was considered statistically significant.

Results and discussion

AA complications have evolved in 117 out of 449 cases or 26.1%, index compatible with literature data [28-30]. So the control group included 332 patients without complications (NAA), and the CAA group included 117, respectively. Tables 2 and 3 show the structure of AA complications in pre- and postoperative periods.

AA had a higher incidence in women – 257, or 57.2% observations, men representing 41.8% respectively. Specialized literature [14, 31-33] indicates a prevalence of AA in men, the dominance of women in our research was probably determined by a hyperdiagnosis in patients with benign gynecological disorders (tubo-ovarian infections, gynecologic peritonitis). This fact was mirrored in the rate of the so-called negative appendices (catarrhal) which had an incidence of 15.6% in women (n = 40), compared with 3.6% in men (n=7) (P<0.001). In the general group of patients

Table 2

Intra-abdominal pre- and postoperative complications of acute appendicitis in CAA group (n=117)

INTRAABDOMINAL COMPLICATIONS	N=	%
Perforated appendicitis	21	17,9
Appendicular infiltration	11	9,4
Appendicular infiltration with abscessing	7	5,9
Periappendicular abscess	19	16,2
Interintestinal abscess	1	0,9
Typhlitis	9	7,7
Local and diffuse peritonitis	73	62,4
Generalized peritonitis	1	0,9
Acute dynamic intestinal occlusion	31	26,5
Intestinal occlusion by post-appendectomy adhesion	4	3,4
External intestinal fistula	1	0,9

Table 3

Pre-and postoperative extra-abdominal complications of acute appendicitis in the CAA group (n=117)

WOUND COMPLICATIONS	N=	%
Suppuration	15	12,8
Abscess	7	5,9
Infiltration	12	10,2
Seroma	14	11,9
Ligature fistula	6	5,1
Hernia post-appendicectomy	5	4,3

undergoing appendectomy catarrhal rate was 10.5%. We found a higher incidence of postoperative complications of AA in this group compared to phlegmonous appendicitis and a lower incidence of complications compared to gangrenous appendicitis (tab. 4).

Figure 2 shows the stratification of patients with AA according to age groups and sex.

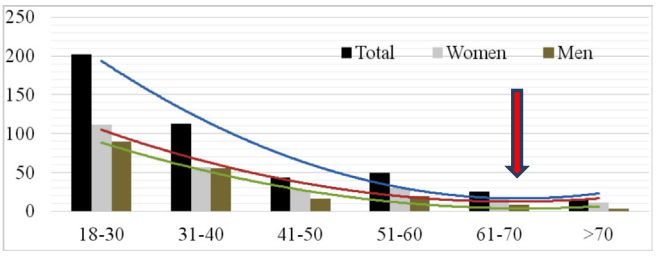

Fig. 2. The incidence of acute appendicitis according to age and sex (n=449).

Data analysis denotes the highest incidence of AA in the age group of 18-30 years, – 202 patients (45%), therefore, practically half of the total number of patients. The decrease in morbidity through AA over the next 3 decades of life (age 31-60 years) is noted, while in patients> 60 years old we have confirmed changes in the epidemiological trend, namely: the trend of AA growth in the general group, especially in women (marked with a red arrow on fig. 3.3). We consider this remark to be important because elderly patients with associated comorbidities are at risk of developing AA complications either on the basis of delayed referral to a physician or because of difficulties or mistakes in establishing the diagnosis and respectively the withdrawal of surgical treatment.

In the complicated acute appendicitis group there was a predominance of women with a ratio of 1.60 (F/B=72/45) comparing to 1.26 (F/B=185/147) in the non-complicated acute appendicitis. The comparative analysis also recorded statistical differences in age, which was 33.5 ± 13.4 years in the NAA group compared to 39.4 ± 16.1 years in the CAA group (P<0.001). Moreover, the share of persons >60 years old was higher in the case of complications of AA – 23.1% (n=27), while in uncomplicated AA it only constituted 3.9% (n=13). Table 5 shows the patients' profile by age and gender.

Table 4

The structure of postoperative complications according to the morpho-pathological form of AA

Complications Catarrhal (n=47, 10.5%)	The morpho-pathological form of acute appendicitis (N=449)		
	Phlegmonous (n=330, 73.5%)	Gangrenous (n=72, 16.0%)	
Complications of the postoperative wound	5 (10.6%)	17 (5.2%)	26 (36.1%)
Intestinal fistula	-	1 (0.3%)	-
Ligature fistula	1 (2.2%)	2 (0.6%)	3 (4.2%)
Postappendectomy hernia		1 (0.3%)	4 (5.6%)
Intestinal occlusion by postappendectomy adhesion	1 (2.2%)	2 (0.6%)	1 (1.4%)
Interintestinal abscess			1 (1.4%)
Total	7 (14.9%)*	23 (7.0%)	35 (48.6%)**

*- siginificant statistic difference between catarrhal and phlegmonous AA ; P<0.05
**- siginificant statistic difference between cataral and gangrenous AA; P<0.001

Table 5

Distribution of complications (CAA) and absence of complications (NAA) of acute appendicitis by age group and sex

INDEX	18-30	31-40	41-50	51-60	>60	Total
AAC M	8	14	7	8	8	**45**
W	19	10	13	11	19	**72**
AAN M	82	42	9	11	3	**147**
W	93	47	15	20	10	**185**

In this context we can conclude that advanced age is a risk factor in the development of complications of acute appendicitis.

The analysis of the onset-address terms in the all patients found a predominance of the term more than 24 hours after the onset of the disease. To standardize clinical manifestations of AA onset, the time when the patient started to feel nausea, vomiting, dyspepsia or any abdominal pain was defined as the time of onset of the symptoms of the disorder. Of the total of 449 patients only 67 (14.9%) addressed before 6 hours have passed, 131 addressed in 7-24 hours (29.2%), while in over 24 hours – 251 (55.9%) patients. In the case of AA complications there was an emphasis on late referral, the debut-addressing term being higher than uncomplicated AA (fig. 3).

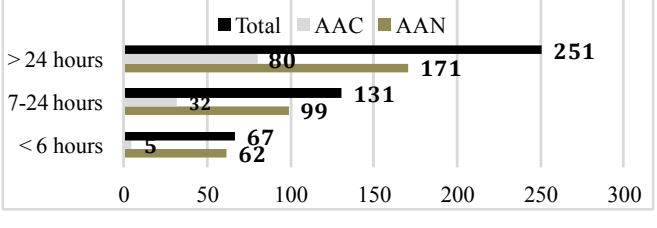

Fig. 3. Distribution of patients according to terms onset-address.

Thus, in the group of patients with complications of AA only 5 (4.3%) patients were addressed in terms of up to 6 hours, 32 (27.3%) – 7-24 hours, and 80 (68.4%) were addressed over 24 hours. In the group of patients without complications these parameters were respectively 18.7%, 29.8% and 51.5% of cases:

Table 6

Division of patients with (CAA) and without complications (NAA) of acute appendicitis depending of the time of address

Variable	< 6 hours	7-24 hours	>24 hours
CAA n (%)	5 (4.3%)	32 (27.3%)	80 (68.4%)
UAA n (%)	62 (18.7%)	99 (29.8%)	171 (51.5%)
P	< 0.001	> 0.05	< 0.05

In this context, the data obtained is consistent with the results of other studies, which have shown that delayed appendectomy is associated with weaker results, so early diagnosis with surgical treatment plays a decisive role in improving outcomes [25,31,34,35]. Busch M. [36] reported that an in-hospital delayed term of more than 12 hours prior to surgery was an independent risk factor for perforation. Kim M. [37] has determined that delayed appendectomy, 24 hours after the onset of clinical manifestations, significantly increases the rate of complications of acute appendicitis. Papandria D. [22] notes that the delay of admission is associated with a higher perforation rate. In contrast, other authors have not recorded a connection between the appendectomy's terms and its perforation rate [20,21,38,39]. For example, Teixeira P. reported that the delay of appendectomy did not increase the risk of perforation [40]. Thus, the ideal or opportune terms for performing appendectomy are currently a controversial problem; the results obtained by various authors are not univocal.

Probably, the different results are determined by what was analyzed – just the terms of the patient in the ward, or the onset – surgical treatment terms. Of course, considering the pathophysiology of AA and its complications, it is rational to study general, complex terms, not just the time when the patient is in custody of the surgeon. On the other hand, it is required to note that the initial symptoms of AA are known to be vague and nonspecific, making it difficult to accurately determine the time of initiation of the pathology. Moreover, the symptoms are subjective, because they depend on the sensations of the patients; the surgeons rely only on their claims. For these reasons, the time at which any known symptom, such as nausea, vomiting, anorexia or abdominal pain are reported by the patient, is to be considered as the first occurrence of AA symptoms.

To conclude on this subject, analyzing our own results, we consider that late referral is a major risk factor for the evolution of AA complications. We believe that the moment symptoms of appendicitis are triggered is important for deciding when to perform emergency surgery. The delayed operation (from the onset of symptoms) is associated with more severe results in the progression of pathology and the risk of developing complications of the disease.

Studying the causes of late referral of patients, we paid attention to their socio-economic status, taking as a criterion the estimation of the presence or absence of medical insurance. The vast majority of patients in our study were insured – 364, or 81.1% of cases. Of the 85 uninsured patients 61 (71.%) went to hospital late, over 24 hours after the first clinical symptoms of acute appendicitis manifested. Figure 4 shows the distribution of patients according to the socio-economic status and the onset-addressing terms.

A direct link was established between the socio-economic state and the terms of addressing for medical care. Probably the absence of medical insurance determines the delay of addressing, the initiation of a self-treatment, which in turn reflects on the evolution of the disease. Thus, namely uninsured patients (n=59.49.6%) formed virtually half of the group of patients with AA complications. Similar results have been obtained by Lin K. [13] who finds a substantially higher incidence of perforated AA in low-income patients compared to normal population, – 37.28% and respectively 26.1%, and the so-called perforation rate constituted 1.34/1.

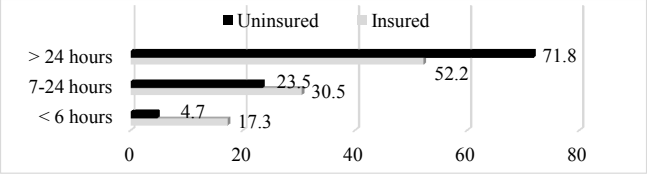

Fig. 4. The distribution of the patients (%) according to the onset-addressing and the socio-economic status.

This study, as well as the results of our research, confirmed that low socio-economic status has a significant negative impact on the evolution of acute appendicitis and its complications, as well as on appendectomy results.

As regards the influence of associated co-morbidities on the natural evolution of the disease, some connections have been noted, namely: with the increase of the co-morbidity score increases the rate of complications as well as the general ones, as well as the abdominal or post-appendectomy wound. This is probably due to the difference in the biological conditions on the background of which the inflammation process occurs in patients with associated co-morbidities, as well as the inflammatory response of the macro-organism itself to the pathogen. Thus, these suggestions are in favor of alternative theories in the field of AA pathophysiology which place the local immunological response made by the vermiform appendix (favored by the richness of lymphoid tissue in the appendix submucosa) and the general release of pro- and antiinflammatory cytokines in response to the pathogenic microbial agent.

Associated co-morbidities were established in 76 or 16.9% of cases, most frequently – cardiovascular (hypertension, ischemic heart disease, angina pectoris, atherosclerotic or postinfarct cardiosclerosis, paroxysmal arrhythmia) – 26 (34.2%) cases. Digestive tract disorders (chronic cholecystitis, chronic gastroduodenitis, chronic viral hepatitis B and C, chronic gastro-duodenal disease in remission) was marked in 12 (15.8%) patients. Type II diabetes was present in 15 (19.7%) patients, and respiratory tract disorders (bronchial asthma, pneumonia, chronic bronchitis) were noted in 6 (7.9%). Urogenital diseases (salpingitis, salpingoophoritis, ovarian cyst, urolithiasis, chronic renal failure) were diagnosed in 7 (9.2%) cases. In 10 (13.2%) patients, co-morbidities were represented by various causes of cerebral infarction, dysmetabolic or atherosclerotic encephalopathy, feripritis anemia, narcotics, varicose veins, chest cancer chemotherapy.

In case of co-morbid score «0» we noticed significant differences, 84.6% of patients being in the NAA group compared to 78.6% of the patients in the CAA group (P <0.001). In other words, the majority of patients who had a favorable affection did not show associated co-morbidities. The association of compensated co-morbidity did not show a severe impact on disease progression, at 13.3% with NAA, compared with 15.4% of patients with CAA (P>0.05). In the case of undercompensated or decompensated associated diseases, or the presence of more co-morbidities, the picture was radically different: 1.5% compared to 3.4% in CCI = 2; and 0.6% vs. 2.6% in CCI ≥ 3 in the respective groups.

Table 7

Characteristics of patients with complications (CAA) and without complications (NAA) of acute appendicitis depending on Charlson Co-morbidity Index (CCI)

Parameter	Total (N= 449)	CAA (n= 117)	NAA (n= 332)	P
CCI "0" (%)	83.1	78.6	84.6	<0.001
CCI "1" (%)	13.8	15.4	13.3	>0.05
CCI "2" (%)	2.0	3.4	1.5	<0.01
CCI "≥3" (%)	1.1	2.6	0.6	<0.001

In brief, we note that the presence of associated co-morbidities has a clear negative impact on AA evolution. The failure to diagnose secondary pathologies or to perform surgical treatment in a patient with insufficiently corrected subcompensated or decompensated co-morbidity and the associations of several co-morbid pathologies are significant risk factors for the development of AA complications.

One of the cardinal problems discussed in the literature is the specification of the pathophysiological mechanisms that underlie the pre- and postoperative complications of AA. It is considered that all septic complications after appendectomy evolved within the surgical wards are due to nosocomial infection. However, the presence of an exogenous source is necessary for its appearance, or otherwise the patient who underwent the operation must contact either a supposed wound of another patient or with contaminated dressing material, instrumentation and so on. The most frequent phenomenon occurs when a septic department is absent in the clinic and the patients with septic complications are not isolated. Another reason could be due to the non-observance of the profile of the hospitalized surgical patients. Such situations may result in outbreaks of in-hospital infection.

In the group of CAA patients, bacteriological cultures were culled intraoperatively in order to analyze whether there is a correlation between the bacterial species normally present in the digestive tract and those isolated from the peritoneal fluid in the appendicular peritonitis. Another objective was the comparative analysis of microbial germs in the case of postoperative wound complications and isolated germs in the peritoneal cavity.

Although peritoneal fluid response was noted in all cases, only 78 patients (66.7%) identified microbes. Constantly, the germs were present in all observations accompanied by diffuse peritonitis and partly in AA with localized peritonitis. In 33.3% observations, the bacteriological laboratory response was negative, probably due to the impossibility of determining the anaerobic flora. This moment is a weak point of the bacteriological research analyzed in our study, because the absence of flora identification does not necessarily signify its absence. The aforementioned increases its importance, because the microbial flora is decisive in the evolution of the disease in the etiopathogenesis of AA, most of the postoperative complications are septic. The aggressiveness of microbial flora in AA is due to the colon, which

in the healthy individual contains anaerobic flora with the predominance of gram negative bacteria Bacterioidis. On the other hand, it is known that the most commonly found in the colon is E.Coli, which is an aerobic germ, and is also an anaerobic microbe (optional) and therefore can also develop under anaerobiosis conditions. These considerations are probably the explanations of a large number of negative bacteriological outcomes in our research.

Analysis of positive crop results (n = 78) revealed the identification of 1 single germ in 6 (7.7%) cases; at least 72 germs were identified in the remaining 72 patients (92.3%). Primary bacteriological research confirmed the prevalence of Gram-negative flora – 57 or 73.1% cases, represented by E. coli – 43 (55.1%), Klebsiella and Enterobacter – 7 (9.0%), Pseudomonas aerugenosae – 4 cases (5,1%), Proteus mirabilis – 3 (3.8%). Of gram-positive microorganisms the following were discovered: (n= 21) Staphylococcus (aureus, epydermidis and heamolyticus) were identified in 9 (11.5%) cases and Enterococcus in 7 (9.0%) observations. Other microbial agents were established in 5 (6.4%) cases.

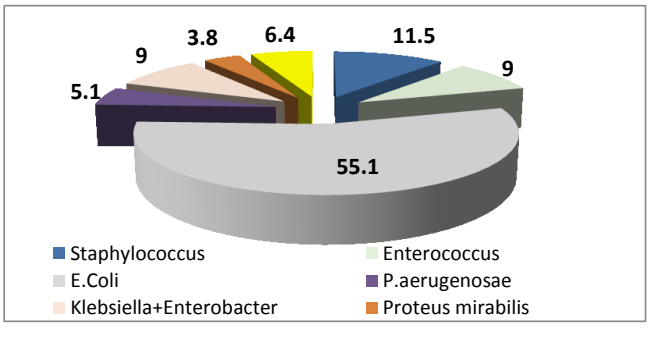

Fig. 5. Microbial germs identified in the study group.

Penicillins possessed a high antimicrobial activity in regards to *E. coli* with a cumulative rate of 29.5% of cases and aminoglucosides in 35% of observations. *Pseudomonas* aeruginosa noted sensitivity to azlocillin – 27.3% and ceftazidime in 31.8% of cases. *Klebsiella and Enterobacter* were sensitive to ciprofloxacin and cyprinol at a cumulative rate of 21.3%. *Proteus mirabilis* was susceptible to cefotaxime and carbapenicillin in 34.6% of cases. Concerning gram-positive flora, cefuroxime (41.7%), ciprofloxacin and oxacillin – 50%; lincomycin – 47.4%, enterococci, vancomycin (26.7%) and netilmicin (20.0%) were effective against Staphylococcus.

Repeated microbiological studies were performed in 48 patients with wound complications. The microflora isolated from wounds usually coincided with seeding taken during abdominal cavity surgery. A total of 53 microbiological tests were performed with positive results in 88.3% of cases. Gram-negative flora observations were found in 67.1%, and gram positive in 32.9% of cases. Microbial associations were noted in 71.2% of cases were represented by 2 microorganisms; in 10.7% – 3 and in 6.4% of cases we confirmed the presence of 4 species of bacteria.

The isolated strains of microbes in repeated bacteriological tests were susceptible to cephalosporins and aminoglycosides. It is known, however, that in-hospital infection is mainly caused by multi-resistant, gram-negative bacteria that produce beta-lactamases and are resistant to cephalosporins, penicillins and aminoglucosides (antibiotic groups commonly used). Specification of microbial flora and its sensitivity to antibiotics allows us to consider flora, which circulates in the abdominal cavity prior to surgery, as the main cause of the suppuration of postoperative wounds. From this point of view, the septic processes of post-appendectomy wounds appear to be essentially a continuation of the purulent-inflammatory process already existing at the point when the patient was hospitalized and treated surgically. In some cases, the lack of identity of the microbial flora may be a consequence of the so-called bacterial translocation in the intestine. These circumstances are important in interpreting the causes of post-operative parietal complications, which by definition are attributed to in-hospital infection, but de facto are a continuation or outcome of the purulent-inflammatory process with which the patient was hospitalized.

Of course, the given deductions do not deny and do not question the importance of nosocomial infection in the evolution of septic-purulent processes in the surgical patient, but we believe that every case of parietal complication of the postoperative wound requires individualized analysis that would allow more accurate determination of the source of the infection (endogenous or intra-hospital). An accurate understanding of this pathophysiological process influences the choice of ways to prevent and treat postoperative complications, including postoperative antiobiotic therapy.

Summarizing the data of the bacteriological research of the patients with AA complications we can observe the following trends: 1) an essential predominance of the gram-negative flora was found in the studied patients – 73.1% of cases part of this study; 2) secondary non-hospital peritonitis with autochthonous flora with a high sensitivity to standard antibiotic therapy usually evolved in patients with CAA; 3) microbial species with high resistance or polyantibiotic resistance are present in case of association of nosocomial infection; 4) when it comes to the general structure of the in-hospital microbial landscape, it is represented by a higher rate of gram-positive flora, represented primarily by staphylococcus aureus and subpopulations of staphylococcus (epydermidis and heamolyticus); 5) the identified gram-positive in-hospital flora possesses a high resistance to the penicillin group antibiotics, hence the productive beta-lactamase-producing species are also resistant to cephalosporins.

Conclusions

1. In our study complications of acute intra-and extra-abdominal appendicitis had an incidence of 26.1%, a value compatible with existing literature data.

2. Risk factors for the evolution of complications were the late referral from the onset of the first clinical manifestations, age> 60 years, and the presence of insufficiently corrected co-morbidity or the association of several co-morbid pathologies. Low socioeconomic status also has a negative impact on the evolution of acute appendicitis and its complications.

3. There was a higher incidence of AA parietal postoperative complications in catarrhal appendicitis compared to phlegmonous appendicitis and a lower incidence of complications compared to gangrenous appendicitis. Thus, more extensive application of diagnostic laparoscopy is necessary in uncertain clinical situations.

4. Specification of microbial flora and its susceptibility to antibiotics allows us to consider the main cause of postoperative wound sedation of native flora susceptible to the usual antibiotic therapy. From this point of view, the septic processes of the post-appendectomy wound appear to be essentially a continuation of the purulent-inflammatory process with which the patient was hospitalized and surgically treated. Any suspicion of postoperative complication through nosocomial infection requires wide-spectrum desiccation antibiotic therapy, including one covering the anaerobic flora.

5. Our findings suggest that clinical assessment is most important for identifying individuals at risk of developing complications of acute appendicitis and the above-mentioned risk factors are useful for emergency surgical decisions.

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The impact of culture media on the endothelial viability of corneas

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Abstract

Background: The complexity of cryopreservation and its potential to damage the endothelium make it so that it is rarely used in routine eye banking, except for occasional, clinically urgent transplants, where the main objective is to save the eye. The culture medium was the method of choice in the Eye Bank.
Material and methods: The study group was divided into 3 subgroups. For penetrating keratoplasty and anterior lamellar keratoplasty, corneas taken up to 24 hours, with a number greater than 2000 endothelial cells per mm^2, are preferably used.
Results: In the first group we found: weakly edematous epithelium; thin, transparent stroma; thin Descemet's membrane; transparent endothelial layer, endothelial cell density greater than 2800 cells/mm^2. The corneas of the second group presented: edematous, but with uncompromised integrity epithelium; slightly edematous, transparent stroma; slightly folded Descemet's membrane; intact endothelial layer and an average of 2600 cells/mm^2. Corneas of the third group: evidently edematous epithelium, with exfoliations in some areas and Bowman's membrane detachment; considerable edema of stroma in all layers; pronounced folds of Descemet's membrane; interrupted endothelial layer along the outline of the folds.
Conclusions: The age of the donor and the preservation time are important factors that influence corneas in culture media and determine the state of endothelial cells. Although the number of endothelial cells usually decreases with age, there are still many corneas from donors over 80 years of age who meet the minimum criteria for transplantation.

Key words: minimum essential medium Eagle, fetal bovine serum.

Introduction

Out of 62 eye banks included in the Association of European eye banks of 2010, 47 – have used culture media, 9 – have used the hypothermic method and 6 – have used both methods: generally, 70% of corneas were stored in culture media [9,13]. The most common organ culture medium is the Eagle Essential Medium (MEM) with 2% fetal bovine serum (FBS), although up to 8% FBS is used by some banks [14]. Most eye banks also include in their medium: penicillin, streptomycin and amphotericin B, but there are alternative antibiotics including biklin, amukin, tazocin and nystatin [12,15]. Another difference in methodology is that the medium is changed during storage; however, 40% of eye banks do not alter the medium during storage, while others refresh the culture medium every 1-2 weeks [11]. The concentration of dextran, used to reverse the stromal edema, which occurs during cultivation in the medium, ranges from 4 to 8%. Regardless of these changes, it would seem that the results of the grafts are similar. Corneas are normally stored for up to 4 weeks [1], but there have been reported successful cornea transplants stored in organ cultures for 7 weeks [2].

The culture medium was the method of choice in the eye bank in the city of Bristol, since it was established in the mid-1980-s, and illustrates the general technical description [3]. Before cutting corneoscleral discs, the eyes are cleaned by rinsing in sterile saline and immersion in povidone-iodine to reduce bacterial and fungal contamination of the surface of the eye. Corneas are suspended in 80 ml of HEPES buffer MEM Eagle, 26 mmol/l $NaHC0_3$, 2% FBS, 2 mmol/l L-glutamine, penicillin, gentamicin and amphotericin B and stored at 34°C [23]. The cornea contains viable cells and cannot be sterilized; however, in comparison with hipothermic storage, culture medium increases the chance of detecting the bacteria and fungi that might cause a postoperative infection, and the addition of antibiotics in the culture medium is more effective, allowing a higher storage temperature [16,17,18]. The first sample probe is taken for bacteriological examination after 7 days of cultivation to examine bacteria and fungi, the second probe – at the end of storage, before the release of the cornea for transplantation [24].

There are three main approaches to preservation and storage of corneas containing living cells: via organ culture, hypothermia, and cryopreservation. Only the latter currently offers the prospect of an unlimited storage time [19]. Techniques for corneal cryopreservation were developed in the 1960s and applied clinically [20, 21]. More recently, retention of endothelial function was reported after ice-free cryopreservation by vitrification of rabbit cornea in a high

concentration of propane – l2-diol [23]. But the complexity of cryopreservation and its potential to damage the endothelium make it so that it is rarely used in routine eye banking, except for occasional, clinically urgent transplants, where the main objective is to save the eye [23].

Material and methods

The study is based on dystrophic changes of endothelial cells in culture medium at different periods of time till sampling and evaluation of cellular conditions during 60 days storage in culture medium "Tissue C", containing Eagle MEM with fetal bovine serum, 2% (FBS), a mix of antibiotic/antimycotic which guarantees effective protection against bacteria and fungi, phenol red indicator which allows a quick view of the changes in pH. During the corneal cultivation, the medium remains unaltered.

250 corneas were collected from 126 donors who died of cardiac arrest. The sampling was performed by the team of the Human Tissue Bank from the Republic of Moldova, Chisinau. All sampling stages have been completed in accordance with the regulations of standard operation protocol (SOP) [4]. The criteria of donor selection [5], the screening of the corneal morphological defects, congenital abnormalities, postoperative scars, post-mortem structural changes were observed. The study group was subdivided into 3 groups, group I – corneas collected up to 20 hours, 160 donors – the stroma is completely transparent, thin, the contours of the boundaries between the iris and pupil are visible; group II – corneas collected from 20 to 24 hours, 60 donors – the surface epithelium is swollen, turbid, its integrity is dubious, the borders of the iris look good; group III – corneas collected after 24 hours, 30 donors – the contour of the iris is poorly distinguished with the presence of an opalescent ring. The thickness of the cornea is proportional to the time the organ is retrieved. Sampling of eye tissue is preferably to be carried out up to 24 hours after cardiac arrest. Corneas were morphologically assessed on transparency. The vast majority of the corneas were excised with surgical instruments specific to ophthalmic interventions (fig 1) [6].

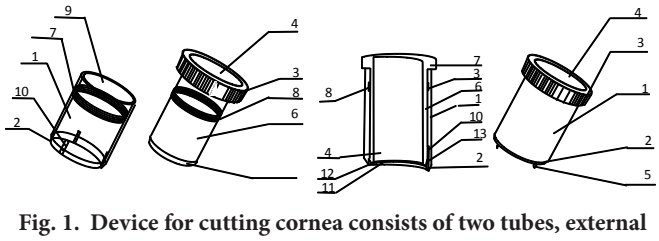

Fig. 1. Device for cutting cornea consists of two tubes, external and internal, made of inoxidable steel, external (1), with the sharp edge (2), the distal end being straight (13), which assembles the blade (5), internal diameter of 18 mm (9), internal tube (6) with distal external thread (8), at the far end of the threaded handle (3), internal diameter of 17 mm (4), proximal external edge (12) and internal (11).

Sampling methods: after aseptic processing of the eye, the eyelids were fixed with blepharostat. The sectional area was set in a ring-shaped manner, afterwards, using a scalpel, a small incision was made in the eye and one of the scissors blade was inserted into the eye section, and the tissue was incised along the line.

Using the ophthalmic tweezers, the edge of the cornea-sclera complex disk was fixed, and the irido-crystalline diaphragm was limited. Using the device for cutting the cornea, the cornea is incised in the center, allowing to make an incision of 2 mm from the edge of the limb, and rotating it clockwise until the conjunctiva and the cornea are completely separated. Subsequently, the device is removed and the cornea is fixed with tweezers, and then the irido-crystalline complex is separated. The corneas obtained were washed with a sterile isotonic solution at a temperature of 20-25 °C in a sterile plastic container with a screw cap. Subsequently, the transport medium, Eussol C", was changed by immersing the tissue in a sterile 50 ml tube with a screw cap containing MEM medium, antibiotics and antifungal agents. Then it is placed in a specialized container for the transportation to the tissue bank.

After receiving the results of the serological test, was assessed the state of the endothelial cells on an inverted contrast microscope with a blunt trepan, in which cells with a uniform redistribution are determined, preferably at the edge of the cornea and in the middle region. In the extracellular space, dead cells are visualized (necrosis/apoptosis) and the state of the Descemet's membrane is observed.

Endothelial cell density should be higher than 2000 cells/mm^2. Moderate or severe symptoms, such as cell pleo-

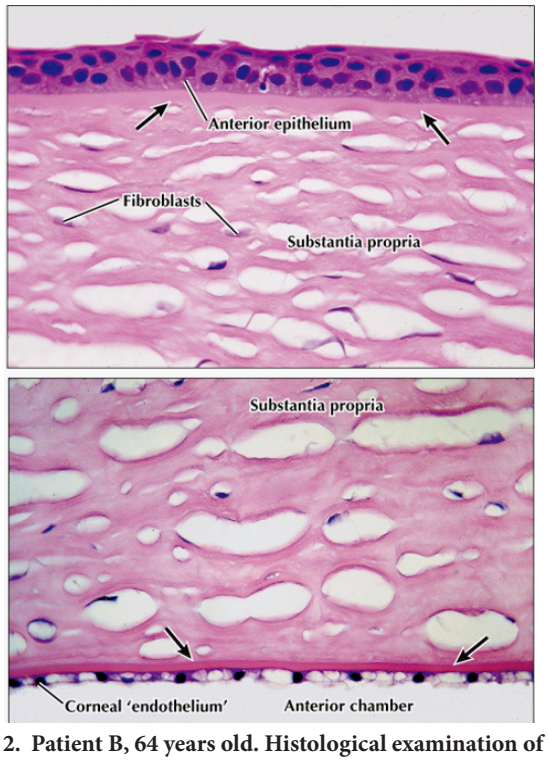

Fig. 2. Patient B, 64 years old. Histological examination of cornea preserved by Tissue C, after 4 days. Anterior epithelium. Layered non-keratinized with planocellular 5-7 cell lines (a), continued with the conjunctival epithelium covering the corneal limbus – injury to stem cells induces fibrosis of the cornea. H-E, x 90.

morphism, significant cell loss during long-term storage, or the presence of dead cells, are considered contraindications to transplantation. The number of cells is calculated in 10 (ten) of 100 squares, diagonally, the average number of endothelial cells is indicated in the calculation.

In the first group, microscopic examination most commonly reveals edematous epithelium, absolutely transparent stroma, rare short membrane folds, thin Descemet's layer; endothelium is completely transparent, intact throughout the area (fig. 2). Areas with a uniform redistribution of cells, mainly on the edge of the cornea and the middle region, the intercellular space and dead cells (necrosis/apoptosis), which occupy small spaces, are identified. The density of endothelial cells is greater than 2800 cells/mm^2 with moderate cell pleomorphism, which is considered as indications for penetrating keratoplasty.

Corneas from group II show: the surface of the epithelium is slightly edematous (Fig. 4), its integrity is not compromised (the exception may be a slight mechanical separation). The stroma is thin, transparent with initial signs of edema in the lower layers. The Descemet's membrane has a single flat shell located centrally and radially; the endothelial layer is intact, and usually, over the folds of the Descemet's membrane, a subtle swelling in the form of an opaque opalescence is allowed. Endothelial layer is arranged evenly, with persistent tesserae (fig. 5), which includes 26 cells per square, giving on average 2600 cells/mm^2.

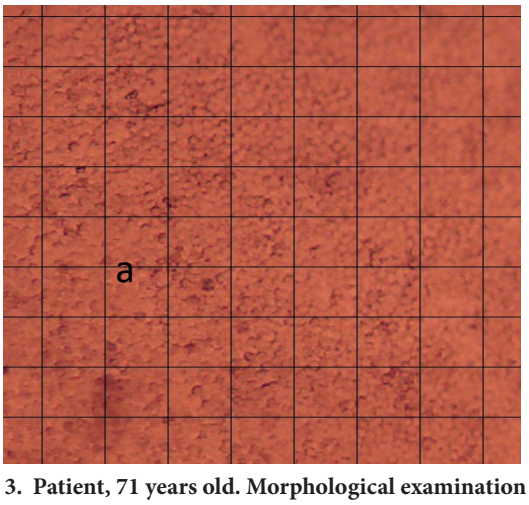

Fig. 3. Patient, 71 years old. Morphological examination of cornea with electron microscopy with inverted phase contrast. Endothelial cells arranged in mosaic form (a), including 26 in a square, constituting 2800 cells/mm^2.

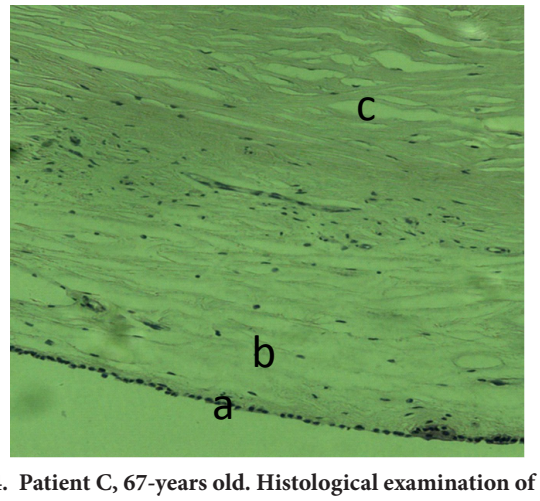

Fig. 4. Patient C, 67-years old. Histological examination of the cornea in the second group, preserved in the culture medium Tissue C, after the 4th day, stroma with initial signs of edema in the lower layers, a thickened, transparent (c) posterior membrane (a) has one central-radial flat shell; endothelial layer is intact (b), usually, along the membrane folds. H-E, x 90.

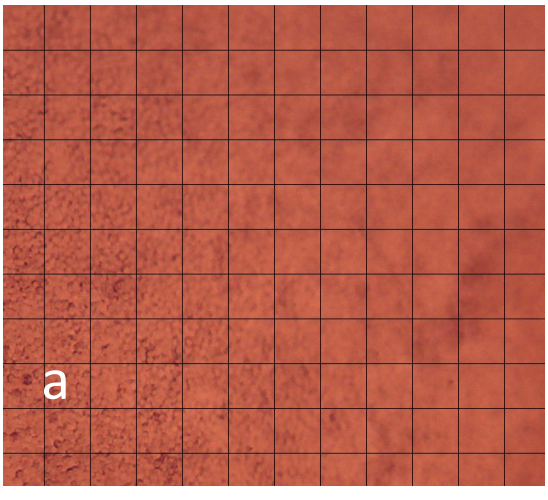

Fig. 5. Patient D, 70 years old. Morphological examination of cornea by electron microscopy with inverted phase contrast. Endothelial cells arranged in mosaic form (a), including 26 cells in a square, totalling 2600 cells/mm^2.

Multi-layered squamous epithelium is isolated from the anterior layer (Fig. 6), highlighting the blast cells at different phases of division along with stem cells, the sources of regeneration of the anterior epithelium.

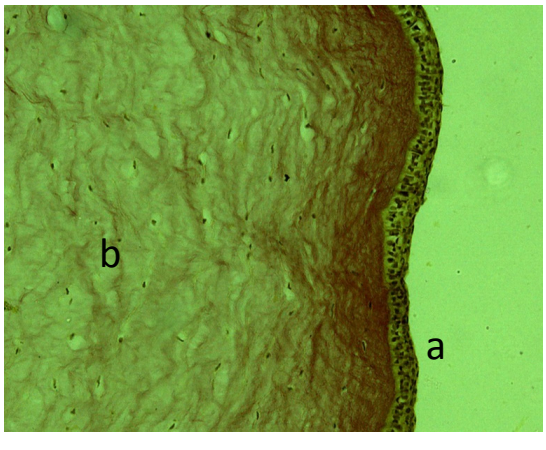

Fig. 6. C. 53-year-old patient. Across the cornea – Tissue "C" preserved on the 5th day. Surface epithelium is edematous (a), its integrity is not compromised (exception can be a minor mechanical exfoliation). Stroma (b) with the initial signs of edema in the lower layers, thickened, transparent. Picrofuxin, x 140.

Corneas of the group III – anterior epithelium edemation in some areas with Bowman's membrane exfoliation (fig. 7). Stroma is edematous in the whole layer and matte colored. Descemet's membrane has pronounced folds,

creases pointing in different directions, giving the aspect of "parquet floor" or "chessboard". Endothelial layer is matte, discontinued along the outline of the folds (fig. 8).

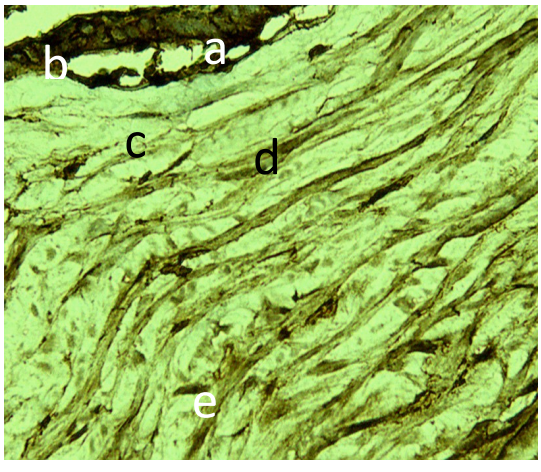

Fig. 7. Patient E, 54 years old. Corneas in the group III. Anterior layer consisting of weakly contoured epithelium (a), enhanced color (+++) with AE1 / AE2 (b), detachment in some areas of the anterior layer (c), the stroma has a frameless thickened fibrillated structure (d), weak fibroblasts (e) AE1/AE2, x 140.

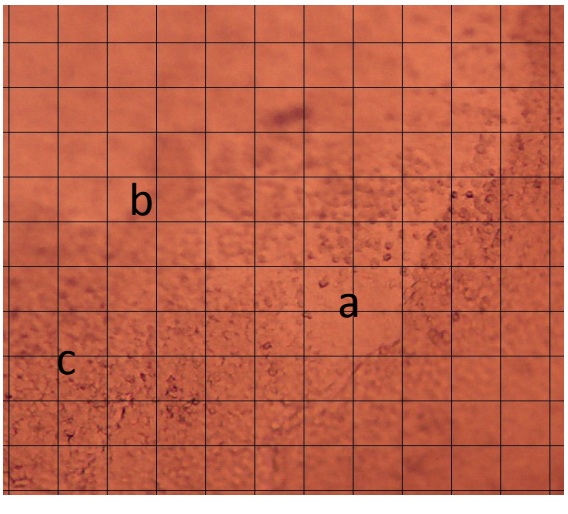

Fig. 8. Patient D, 69 years old. Morphological examination of cornea by electron microscopy with inverted phase contrast. Endothelial cells are sectorally stable with the disappearance of the mosaic (a), which contains 20 cells per square (c), constituting 2000 cells per square millimeter, and detachment of the epidermis (b).

Results and discussion

Corneas are stored for up to 4 weeks, and their suitability for transplantation shall be based on the examination of the corneal endothelium through luminescent microscopy. This allows assessment of endothelial cells (ETC). In Bristol, a minimum of 2200 ETC cells/mm^2 is considered acceptable for transplants, which require a viable endothelium. Other abnormalities and endothelial damage may be also identified and taken into account [25]. Corneas stored in the culture medium for up to 4 weeks have been shown to preserve the integrity of both endothelial and epithelial layers. Although corneal cells may be lost through apoptosis, this seems to affect the epithelial layer more than endothelial cells [7]. Important factors influencing corneas from culture media and determining the condition of the endothelial cells are the age of the donor and, to a lesser extent, the storage duration [3]. Although ETC typically declines with age, there are still many corneas from donors of over 80 years of age who meet the minimum criteria for transplantation.

When the culture medium was introduced in the United Kingdom in the mid-1980s, it has had a major impact on supply of corneas to the hospitals across the country [26]. There were two opportunities in the first place, a national distribution service of the cornea similar to an existing one for organs, established within a few years; and, secondly, the culture medium of extended storage time from 2 (whole eyes or corneas in M-K medium) to 4 weeks [8]. This gave time for medical testing and evaluation of donors, as well as for surgical demand. Application of medium culture in the United Kingdom has prompted the storage of corneas in the Bank of eye and they have been made available for elective surgery and for urgent transplants [27]. These benefits of the culture medium have improved the quality, safety and availability of the corneas in the UK and other European countries.

A different technique from corneal organ culture is used for ex-vivo expansion of corneal epithelial stem cells. These cells are extracted from the limbal region between the cornea and the sclera, and they are used in the treatment of ocular surface diseases where the corneal epithelium is defective. This is a painful and sight-threatening condition that is difficult to treat. The most successful application of this technique has been for unilateral ocular diseases where sheets of stem cells can be established from a small limbal biopsy taken from the healthy fellow eye [32]. As with organ culture of corneas, the development of culture media free of bovine serum for limbal stem cell expansion is considered desirable [32, 33]. These tissue constructs echo the trend in corneal transplant surgery for replacing only the defective parts of a cornea rather than relying on full-thickness grafts as the treatment of choice for all corneal deficiencies [34, 35].

The potential for modification of corneas during storage is being investigated. Since allograft rejection is a major cause of corneal graft failure, immunomodulation to reduce the immunogenicity of corneas is being attempted by transfection of endothelial cells to over-express down-regulatory cytokines, such as IL-10 and IL-12 [28]. Another approach is to encourage endothelial cells to divide during corneal storage by transfection with transcription factors such as E2F2, which has been reported to stimulate cell-cycle progression and endothelial replication [30]. There is also the prospect of a tissue-engineered corneal construct [31].

Conclusions

1. The most preferred cornea for penetrating keratoplasty and anterior lamellar keratoplasty is the cornea taken up to 24 hours, with more than 2000 endothelial cells per mm^2.

2. The most effective preservative medium remains the culture medium with the addition of growth factors and antibiotics.

3. Corneas with a low content of endothelial cells of up to 2.000 per mm^2 are used for tectonic anterior keratoplasty.

4. Endothelial cells are thin-walled and require a proper diet with permanent nutrients and oxygen, with more frequent change of media preservation, which gives the possibility to prolong their viability.

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Comparative results in the treatment of clavicle fractures

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Abstract

Background: The negative results of the clavicle fractures treatment represent a high percentage, which implies the search of new methods designed to improve these results. Thus, a new method of stable and functional osteosynthesis of the clavicle fractures were proposed for this improvement, namely that performed with the external fixator. In connection with this, a comparative analysis of the results between the stability and efficacy of osteosynthesis of the the clavicle fractures by using the external fixator proposed by us and the traditional methods was conducted.
Material and methods: For a better deduction and evaluation of the clavicle fractures treatment methods, it was established a task based on a clinical-radiological examination at 124 patients with clavicle fracture, which constitutes 2.7% of the number of patients hospitalized with skeletal fractures. Thus, for better surgical treatment results of clavicle fractures at the patients, was introduced a new stable and functional osteosynthesis method of the clavicle fracture treatment with an external fixator.
Results: The application of the stable-functional osteosynthesis method of the clavicle fractures with an external fixator's help succeeded in a reduction of the medium – term work incapacity, the quantity of mistakes and complications reduction and the considerable increase of the positive results of the treatment, while not neglecting the economic aspect on the background of the shortcomings exclusion of traditional surgical methods.
Conclusions: The stable and functional osteosynthesis method of the clavicle fractures treatment with an external fixator versus the traditional treatment methods ensures the possibility of early functional treatment, also reducing the average of work incapacity term, mistakes and complications appeared as consequences of traditional treatment methods application and offers the possibility to use it at all specialized surgical care levels.

Key words: osteosynthesis, external fixator, clavicular fracture.

Introduction

The main factor in each treatment method of clavicular fracture is not only getting the fractured fragments consolidation but also getting an early, complete functional rehabilitation and the reduction of the work disability term [1, 2, 5, 6, 7, 10, 11]. The fact, that in the surgical treatment of the patients with clavicular fractures are proposed different methods, confirms the existence of multiplicity difficulties faced by trauma surgeons [3, 4, 8, 9,12,13,14], both in identification of the method and its performance, coming out from the inventory of technical resources [15, 16, 17, 18, 20, 25, 26].

Until now, the most spread out method of treatment of a clavicular fracture is the intramedullary osteosyntheses realized with a stainless steel rod made by following the Bogdanov's example (IXI8H9T mark) with the length of 125 mm and 3.5 thickness and on brooches from stainless steel (IXI8H9T mark) made after Kirschner and Elizarov's example with the diameter of 1.8 and 2.2 mm which are not providing a stable fixation of the fragments and require a stable external immobilisation [21, 22, 23, 24, 27, 28]. Therefore, this kind of a method excludes the possibility of an earlier functional treatment. The drawbacks of the traditional methods of described surgical treatment using intraosseous fixators, increase the percentage of complications and poor results.

Objectives

1. Elaboration of a new osteosynthesis method of clavi-wcular fragments fixed with an external fixator, simple in the way of the assembly and manufacturing, which will exclude the shortcomings of traditional methods of surgical treatment and its application to any level of specialized surgical aid.

2. Application of the developed method by the stable-functional surgical treatment of clavicular fracture in the clinic and determining the indications and contraindications on its utilization.

3. Obtaining a reduction of the time limits of incapacity for work, the number of errors and complications and increasing the percentage of the good results, using the proposed new method of treatment.

4. The study and comparison of the results of an early treatment and on time distance, of the period of work incapacity, of the mistakes and complications at the patients treated by applying the new method of stable-functional osteosynthesis, in comparison with the results of treatment through the traditional surgical methods.

Analysis and comparison

The analysis of observations of surgical treatment using traditional methods has been made on 61 patients. Typically, surgical treatment is applied after orthopedic ineffective reduction (59%).

Therefore, as indications for applying the surgical intervention have served the displacements on the longitudinal radius fragments larger than the diameter of the bone.

The term of work disability of the patients treated according to traditional methods accounted 72.7-24.5 days. At the patients, who in the postoperative period never had complications, the term of work incapacity amounted to 58-17.2 days; at the patients, to whom the complications emerged during the postoperative period, it grew up to 81.3-12.5 days. After performing the intraosseous and combined osteosynthesis for the fixator removing, it is required a surgical reintervention. According to our calculations, the period of work incapacity after removing the fixator constituted 6.6-3.0 days, a total of 79.3-27.5 days.

Throughout the surgical treatment, according to traditional methods, surgical errors and complications were detected at the 21 patients (34.7%). A total of 28 were identified for complications. The most common, from 21.4 percent of the total number of complications, have been: migrating rod; skin perforation; slower consolidations; pseud-arthrosis – 14.2%.

Examination of the mistakes and complications confirmed that the main factor that led to their appearance was the multi-traumatic surgical intervention and the instability of the osteosynthesis that have influenced negatively the outcome of the treatment, therefore, the "unsatisfactory" results amounted to 14%.

Results over time of the treatment with traditional surgical methods were examined at 50 (81.8%) patients. According to our calculations, excellent analysis results were obtained from 21 (42.8%) patients, good – 14 (28.0%) patients, satisfactory at – 8 (16.0%) patients and unsatisfactory – 7 (14.0%) patients.

In this context, the unsatisfactory results, obtained by us on the basis of clavicular fractures treatment observations with traditional surgical methods, require new methods of innovation of a less traumatic osteosynthesis that would:

– Ensure a stable fixation of fragments without applying an external immobilization;

– Retain the full function in the joints of upper limb involved in trauma;

– Shorten the period of incapacity for work and the number of errors and quantitative complications;

– Improve the results of treatment;

– Give the option of completing treatment at any level of aid.

The advantages and effectiveness of the external fixator using

In the view of these reasons, the surgical treatment performance at the patients with clavicular fractures is trivial.

The development of a simple and reliable method of treatment of the clavicular fractures, less traumatic, which would provide the opportunity to carry out them at any level of specialized surgical aid, in our view, is timely and actual.

These assumptions have formed the idea of the innovation of an external fixator (innovation certificate No 2784 RM), as well as the elaboration of new methods of osteosynthesis using it (certificate of innovation No 2785 RM) in different variants (innovation certificate No 2809 RM).

In order to provide the effectiveness research of the use of the external fixator and mechanical force of fragments characteristic for the fixator, was evaluated through experimental samples, the comparability of mechanical fixation force of the fragments by using the proposed fixator and the mechanical fixation force of the fragments by traditional methods application of surgical treatment – the rod made by Bogdanov's example and two brooches made by Kirschner and Elizarov's example.

The innovated method of osteosynthesis of clavicular fragments and their fixation with external proposed fixator has been applied in clinical conditions on 63 patients. In the process of applying in clinic, we used 2 variants of the osteosynthesis method. For each variant have been developed specialized indications.

The application of osteosynthesis method of the external fixator assembled in the first version underwent 51 patients, in the second variant – 12 patients. The average time of the fragments fixation with external fixator was 35.3+-3.38 days. The mean time of work incapacity amounted to 46.2+-5.7 days.

In the process of applying the proposed external fixator in clinic were discovered mistakes and complications at 7 (11.4%) patients. It is appropriate to mention that mistakes and complications were common during the method learning in clinic and influenced essentially the outcome of the treatment at 3 (4.74%) patients.

The overtime results were examined by us at 57 (93.6%) patients. Excellent results were obtained at 48 (81.39%), good – 8 (13.59%), satisfactory – 2 (3.2%) patients, unsatisfactory at one sick person (1.6%). The obtained results as a consequence of surgical treatment with the help of the application in clinic of the osteosynthesis method with external fixator mounting, confirm the efficacy of the developed method by obtaining excellent and good results at the 56 (94.98%) patients.

The medium-term work incapacity at the patients treated by surgical implants and osteosynthesis method of the fragments, developed by us and fixed with external proposed fixator, compared to traditional surgical methods (osteosynthesis with Bogdanov's rod, Kirschner's 2 brooches, combined) has been reduced by 1.76 times ($P < 0.01$).

The mistakes and complications detected in the treatment of clavicular fractures according to the method developed by us, in comparison with mistakes and complications detected in the treatment of patients after surgical traditional treatment, have been three times reduced.

The comparison of overtime results, confirms high percentage of excellent and good results of 24.98% versus percentage of excellent and good results in treating patients by traditional surgical methods, which are relative one to another 1.5:1 ($P < 0.01$), to the application of traditional methods in terms of 6.5:1 ($P < 0,05$) when using the method developed by us.

Therefore, the average of the hospital stay of patients treated by the utilization of the developed method was re-

duced by 1.4 times ($P < 0.001$), and the medium-term work disability by 1.8 times ($P < 0.01$).

In this context, taking into account, that the new treatment method ensures reduction of work incapacity term, the reduction of the number of mistakes and complications, substantial improvement of the treatment results, the economical effect increase and the exclusion of the shortcomings of traditional surgical methods, the innovative method can be recommended as an alternative method of treatment of clavicle fractures in trauma practice at any surgical specialized aid level.

Conclusions

The research of literary sources has justified the assumptions of some authors, who used the surgical transosseous method with their external fixation for the osteosynthesis of the fragments.

Therefore, the method is less traumatic, but technically difficult to fulfill, which is possible only in conditions of specialized institutions and clinics in Traumatology and Orthopedics, properly equipped and with highly qualified specialists on the staff.

The developed osteosynthesis stable-functional method of clavicular fractures provides the obtaining of a solid fixation of the fragments with different characters and offers the possibility of early functional therapy.

The application of the osteosyntesis stable-functional method in clinic and its variants, according to developed indications for them at 63 patients allowed obtaining excellent and good results in 94.96% of cases.

Reduction in the number of mistakes and complications by three times, reduction of hospitalization term of patients by 1.4 times and reduction of the period of work incapacity by 1.8 times confirms the essential priority of the proposed osteosynthesis stable-functional method and may be recommended as an alternative method of treatment of clavicle fractures at all levels of specialized surgical aid.

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Hard palate in fetal and early neonatal periods of human ontogenesis

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Abstract

Background: This investigation is important to reveal hard palate ontogenetic transformations in fetal and early neonatal periods, which is important for the facial surgery in fetuses, newborns.
Material and methods: Investigations have been performed in 53 cadavers of fetuses from 4 to 10 months of development, and in 9 cadavers of newborn children of both genders, who died of the reasons not connected with digestive system diseases or anomalies, and were without external signs of anatomic deviations or abnormalities, and were without evident macroscopic deviations of skull structure. Adequate anatomic methods have been used for investigation: macropreparations, topographic anatomical sections, morphometry, and statistical analysis.
Results: Variants of the hard palate shapes during fetal and early neonatal periods of ontogenesis are the following: trapezium (15%), square (10%), and ellipse (7%) shapes. The shapes of the trapezium were detected in most cases in 6-7-month fetuses (20%), and in equal percentage proportion in early and late fetuses (12%). The square form is a characteristic variant form in early fetuses (17%), with fetuses age increase this form was detected in less cases, in 6-7-month fetuses – in 10%, in late (8-10 month) fetuses – in 8%, and within the newborn period the square form was not observed. The form of the ellipse was observed in larger percentage proportion in late fetuses and in newborns (12%), in less proportion (5%) – in 6-7-month fetuses, in early fetuses this form was not detected at all. Forms of the ellipse and of the trapezium were observed in equal number of cases in 8-10-month fetuses and newborns. With fetuses development the hard palate form in sagittal and frontal planes changed from arcuate to flat one.
Conclusions: An ascertainment of typical and variant anatomy of hard palate forms and types would promote implementation of the new methods of major and reconstructive surgical invasions in face and skull.
Key words: hard palate, anatomy, fetus, newborn, human.

Introduction

Cleft lip and hard palate rank first by rate in the structure of dentofacial region development defects, and rank third among all types of congenital anomalies, and belong to the heaviest development defects, leading to significant anatomic (cosmetic) and functional abnormalities. 70% of them are congenital cleft upper lip and palate, and 30% are craniofacial dysostoses. The most various are congenital defects of hard palate development, determined by its size, form, and position variations [1, 11, 12].

The establishment of anatomical variability of organs, structures and their parts at all stages of human development is very relevant. Anomalies of the teeth-jaw system take one of the main places among the pathologies of the maxillofacial area and, according to various authors; their prevalence is from 70 to 80%. One of the common congenital malformations of the maxillofacial area is the cleft lip and cleft hard palate, which are called "hare-lip" and "wolf's jaw". The severity of birth defects is expressed not only by external distortions and functional disorders but also by a negative influence on the child's mental development [10, 17].

The hard palate, which separates oral and nasal cavities, undergoès complex development, and is tightly connected with formation of craniofacial region, dento-mandibular complex, and the skull as a whole. Therefore, knowledge of its individual anatomic variability is important not only for functional anatomy and medical craniology, but also has direct application significance in solving of numerous problems of stomatology, otorhinolaryngology, and mandibulofacial surgery. Wide application of morphometric methods in anatomic sphere investigations would permit to reveal significant signs of certain structure components, objectively reflecting changes, which take place in human ontogenesis [2, 3, 6, 7, 15].

This investigation priority is revealing of hard palate ontogenetic transformations in fetus and neonatal periods, which is important for facial congenital pathology surgery in fetuses, newborns and infants [4, 5, 8, 9, 16, 13, 14].

Investigation objective: to determine forms and types of hard palate in fetuses and newborns.

Material and methods

Investigations have been performed in 53 cadavers of fetuses from 4 to 10 months of development, and in 9 cadavers of newborn children of both genders, who died of the reasons, not connected with digestive system diseases, and were without external signs of anatomic deviations or abnormalities, and were without evident macroscopic deviations of skull normal structure. Adequate anatomic methods have been used for investigation: macropreparations, topographic anatomic sections, morphometry, and statistical analysis. This work has been performed in compliance with general provisions of WMA Declaration of Helsinki – Ethical Principles for Medical Research Involving Human Sub-

jects (1964-2000) and Ministry of Health of Ukraine (Order No 690 of 23.09.2009), and it is a fragment of the complex planned initiative research work of M. Turkevich from Human Anatomy Department, Anatomy, Topographic Anatomy and Operative Surgery Department of Bukovinian State Medical University: "Peculiarities of morphogenesis and topography of organs and systems in pre-natal and post-natal ontogenesis periods" (State Registration No 011U002769).

Macroscopic investigation makes it possible to study peculiarities of the structure and different topographic anatomic relationships of maxilla and hard palate structures, in particular, during fetal and early neonatal periods of ontogenesis. On specimens of maxillae palatine processes and horizontal plates of palatine bones, their anatomic peculiarities were investigated. Linear dimensions were measured with centimeter stripe, trammel, small caliper and slip compass. Further forms and types of hard palate were determined. To determine hard palate types its index was used. Hard palate index was calculated with the formula: maximal hard palate width was divided by its maximal length, and multiplied by 100. Calculated index within 100.0-110.0 indicated dolichouranic type of the hard palate, within 110.1-120.0 – mesouranic type, within 120.1-130.0 – brachyuranic type, within 130,1-140,0 and more – hyperbrachyuranic type [10, 17].

Results and discussion

In fetuses and newborns, the hard palate form syntopically depends on the cellular maxillary process structure. In accordance to geometric figures, five its forms were distinguished: oval form, semicircular form, ellipse form, trapezium form, and square form. It is significant, that typical hard palate forms during fetal and early neonatal periods of ontogenesis are oval (47%) and semicircular form (21%) (tab. 1). Oval form, in percentage proportion, is mostly detected in newborn period (56%), during fetal period this form varies from 40% to 50%. The most seldom was the oval form observed in 7-month fetuses (33%). The highest indices of the hard palate oval form were in 6-7-month fetuses, in early fetuses (4-5 month) – in 24% of cases, in later fetuses (8-10 month) and in newborn period – 16%.

Variant forms of hard palate during fetal and early neonatal periods of ontogenesis are trapezium form (15%), square form (10%), and ellipse form (7%) (tab. 1). Trapezium form was detected mostly in 6-7-month fetuses (20%), in equal percentage proportion– in early and later fetuses (12%). Square form is typical variant form of early fetuses (17%), with age this form was detected in less number of cases, in 6-7-month fetuses – in 10%, later (8-10-month) fetuses – 8%, in newborn period square form was not observed. Ellipse form in larger percentage proportions was observed in later fetuses and in newborns (12%), in lesser ones (5%) – in 6-7-month fetuses, in early fetuses this form was not detected at all. In equal number of cases trapezium and ellipse forms were observed in 8-10-month fetuses and in newborns.

Table 1

Hard palate forms in fetal and early neonatal periods of human ontogenesis

Age, months	Objects number	Oval form	Semi-circular form	Ellipse form	Trape-zium form	Square form
4-5	17	47%	24%	-	12%	17%
6-7	20	40%	25%	5%	20%	10%
8-10	16	50%	13%	13%	12%	12%
Newborns	9	56%	22%	11%	11%	-
		47%	21%	7%	15%	10%

Within fetuses development the hard palate shape in sagittal and frontal planes changes. Whereas in 4-9-month fetuses hard palate in macropreparations of sagittal topographic sections was of the arch form, which convexity was directed cranially to the nasal cavity, in 10-month fetuses and newborns it was flat, convexity was evident in anterior part of the hard palate, near palatine processes of maxilla. During the perinatal period hard palate in frontal plane in equal distance from maxillary palatine processes edge and soft palate looked like an arch, its convexity was directed cranially towards nasal cavity. Dorsally hard palate changes its form from arch-like to flat one, and near transition point of hard palate into the soft one, it becomes flat.

During fetal and early neonatal periods of ontogenesis, based on hard palate index determination, its types were established. The characteristic type of hard palate is dolichouranic one, which in this age period was detected in 64% of cases. Dolichouranic type of hard palate structure in 4-5-month fetuses was detected in 71% of cases, in 6-7-month – in 45%, in 8-10-months – in 63%. In newborns this type of hard palate structure was observed in all cases

Table 2

Hard palate types in fetus and early neonatal periods of human ontogenesis

Age, months	Objects number	Dolichouranic type	Mesouranic type	Brachyuranic type	Hyperbrachy-uranic type
4-5	17	71%	29%	-	-
6-7	20	45%	30%	10%	15%
8-10	16	63%	13%	6%	18%
Newborns	9	100%	-	-	-
		64%	21%	5%	10%

(100%), but in 8-month fetuses the dolichouranic type was not detected.

Mesouranic, hyperbrachyuranic, and brachyuranic types of the structure are variation ones, their proportions in fetuses and newborns are 21%, 10% and 5% of cases correspondingly (tab. 2). Mesouranic type of the hard palate is typical in 5-7-month fetuses, it was observed averagely in 30% of cases, more rarely – in 4-, 8-, 9-month, and this type was never detected in 10-month fetuses. Hyperbrachyuranic and brachyuranic types of the hard palate were observed in 6-8-month fetuses only, these types were detected in early (4-5-month) and in 9-10-month fetuses. The largest indexes of hyperbrachyuranic and brachyuranic types of the hard palate are typical in 6-7-month fetuses, their indexes are 15% and 10% of cases correspondingly.

Conclusions

1. During fetal and early neonatal periods the oval (47%) and semicircular (21%) forms of hard palate are typical, and are characteristic for dolichouranic and mesouranic types.

2. Variant shapes of hard palate are trapezium form (15%), square form (10%), and ellipse form (7%). These shapes are characteristic for hyperbrachyuranic and brachyuranic types. Ellipse form of hard palate may be linked with brachyuranic type, and trapezium and squareforms – with hyperbrachyuranic one.

3. With fetuses development the hard palate form changes its shape in sagittal and frontal planes from arcuate to flat one.

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Techniques of liver decellularization

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Abstract

Background: The growth of the number of people who need the liver transplant and the insufficiency of organ donors, have urged the advancement in bioengineering through the development of new therapeutic strategies which involve generation of functional artificial organs, obtained by the decellularization technology and creation of extracellular matrix and their subsequent recellularization.
Material and methods: Rat livers (n = 9) served as the object of this study which were subjected to decellularization with sodium dodecyl sulfate (SDS) solution 0.1 and 0.5% and the combination of SDS 0.1 to 0.5% and anticoagulant. Subsequently, the extraction of nucleic acids was performed according to the protocol QIAamp Blood Mini Kit (2003). The histological analysis was performed with haematoxylin-eosin (H-E) and the quantification of hydroxyproline via spectrophotometric method.
Results: After the liver tissue decellularization we obtained the matrix of decellularized liver. The genetical, biochemical and histological analysis revealed a better decellularization by the combined method versus the method with SDS solution only.
Conclusions: The quantification of nucleic acids content, hydroxyproline and the histological analysis of decellularized matrix with anticoagulant and detergent SDS method, suggested a more efficient decellularization of liver tissue segments and we achieved a decellularized bioconstruction for recellularization.

Key words: decellularization, recellularization, liver matrix.

Introduction

Liver pathologies with viral or alcohol abuse etiology often results in the need for liver transplantation. The insufficiency of donors and high costs are limiting factors in the ability of the medical system to cure many cases of these diseases [3, 24]. Thus, in order to improve the existing situation, a new field of research is emerging, such as tissue engineering, which presents a promising strategy to reduce the transplanted organ deficiency [5, 23, 25, 29]. Tissue engineering is an interdisciplinary field that allows the production of 3D biological scaffolds with could be used for restoring, maintaining or improving the function of a tissue or organ by combining the principles of tissue engineering and biological sciences. 3D scaffolds obtained by tissue engineering techniques can be oriented in two directions: as matrices for supporting *in vitro* seeded cells to produce a functional tissue which will subsequently be used for transplantation and as a growth factor-releasing device [7]. An important aspect in the strategy based on functional tissue engineering is the technology for obtaining 3D biological matrices by decellularization process with detergents and tissues solubilization. This technology has a specific priority that consists in preservation of the biochemical and structural components of the native extracellular matrix [20]. The native extracellular matrix (ECM) presents the architectural basis of the organs and tissues, which supports the cell types specific to the organs and allows their normal functioning [12]. According to the literature, the extracellular matrices obtained by decellularization are derived from their home tissue, thus serving as a substrate for the cultivation of specific tissue cells, proliferation and efficient differentiation thereof [17].

Also, another strong point in favor of using extracellular matrixes obtained by decellularization is that they are non-immunogenic and prevent organ rejection after transplantation [28].

Decellularized liver scaffolds are capable to maintain cellular function, which is a positive indicator in liver tissue regeneration, only a decellularization method should be found which would allow for the preservation of the native extracellular matrix and would not disrupt the conjuctive liver architecture. The methods for decellularization may be of a physical, chemical or enzymatic nature and could influence tissue ultrastructure, biochemical composition and mechanical behavior of the extracellular matrix [8, 10].

The decellularization methods used should maintain the physical properties and integrity of extracellular matrices, but also remove cellular elements as efficiently as possible: DNA, mitochondria, membrane lipids and cytosolic proteins to prevent the occurrence of an adverse inflammatory response and inhibit constructive remodeling [15, 16, 9]. An important aspect in the technology of obtaining of extracellular matrix of the liver is to maintain the integrity and mechanical properties of the microvascular network to ensure the nutrient intake and the exchange of substances in the cells with which the scaffolds will be repopulated [1, 27].

The aim of our study is to obtain by decellularization process the extracellular matrix with integrity of the vascular network.

Material and methods

As object of study were used the livers from Wistar line rats (n=9) weighing 200 - 230g. Prior to isolation of the rat liver, was performed the euthanasia in the carbon dioxide chamber respecting the requirements of the Ethic Committee. After the rats were fixed on the work table, by the median incision the abdominal cavity was opened. Was identified the portal vein, ligated to the proximal end and cannulated with a butterfly catheter with a needle 2 cm length. The excised liver was mounted to the perfusion system and washed with 1-1.5 liters of distilled water and subsequently decellularized by two methods: with decellularization agent – 0.25% or 0.5% sodium dodecyl sulfate (SDS) [20] and the second method, in which we provided perfusion of the liver with anticuagulant solution (citrate phosphate dextrose) before decellularization and then – decellularization with 0.25% or 0.5% SDS. Subsequently, the liver was perfused with 1-1.5 L of 1% PBS solution. The segments of the intact and decelularized liver were fixed in 10% formalin and subsequently was performed histological samples preparation and H-E staining. The extraction of nucleic acids was performed according to the QIAamp Blood Mini Kit extraction protocol (2003). Each sample was quantified by a spectrophotometer (Nano Drop 200 C Thermo Scientific). The hydroxyproline content was determined on a spectrophotometer based on the oxidation of hydroxyproline in pyrrole under the action of chloramine B.

Results

The macroscopic evaluation of the liver at the experiment beginning, showed that after its washing with distilled water (first method) and with distilled water and citrate dextrose phosphate (second method) in the second one the liver became to be more pale in comparision to the first one (fig. 1, 2). However, visual differences in perfusion liquid color were observed: the perfusion liquid obtained after

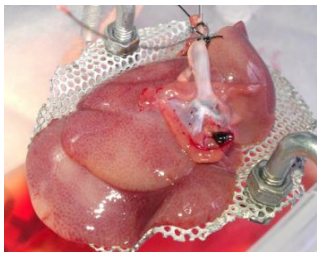

Fig. 1. Liver perfusion with distilled water.

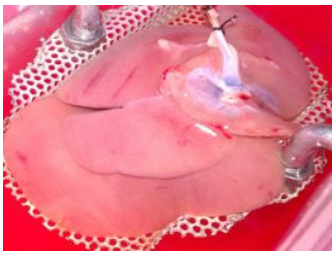

Fig. 2. Perfuzed liver with distilled water and anticoagulant solution.

washing using the distilled and anticoagulant mixture has more intense brown color, which is explained by the more efficient removal of the blood clots.

Also, macroscopic results after decellularization showed a uniform liver decellularization and a whitish color in the second experimental lot (fig. 3 A) compared to the first lot where liver was washed without anticoagulant solution, the yellow color indicating the presence of the cellular residue (fig. 3 B).

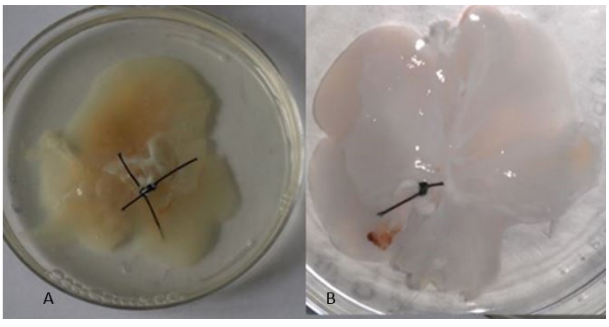

Fig. 3. A – extracellular matrix decellularized by the SDS without anticoagulant washing and B – extracellular matrix obtained by decellularization with SDS and anticoagulant washing.

In the histological analysis of intact liver samples no changes were observed. We have visualized in the inverted with phase contrast microscope KZD the centrolobular and perilobular blood vessels and the hepatocytes which correspond to the native liver (fig. 4).

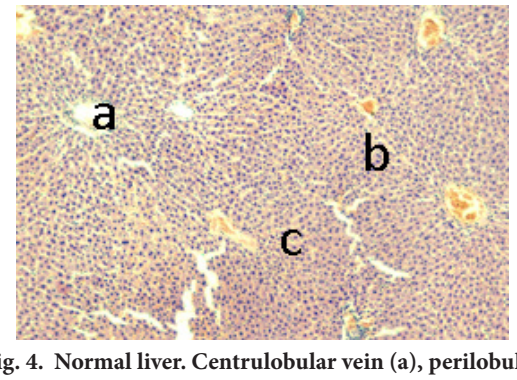

Fig. 4. Normal liver. Centrulobular vein (a), perilobular congested hepatic artery (b), contoured hepatocytes. H-E, x 90.

In the case of decellularized liver scaffolds without using the anticoagulant solution, we observed that there are cellular conglomerates, necrotic detritus and autolysed cells, conjunctival liver architecture is disorganized, the vascular wall is in some portions with the contracted contour and perivascular autolysed cellular elements (fig. 5).

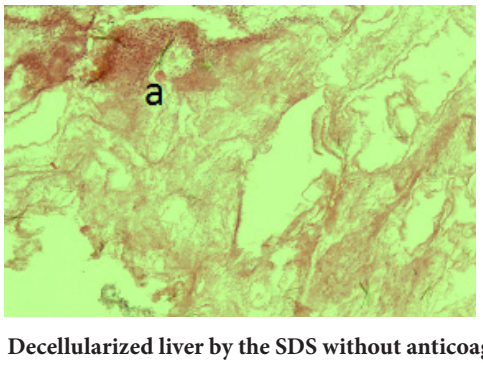

Fig. 5. Decellularized liver by the SDS without anticoagulant washing. Conglomerate of autolysed cells with cariolysis and plasmolysis (a), the disorganization of the connective architecture of the liver (b). H-E, x 90.

The decellularized liver scaffolds with SDS and initially washed with anticoagulant showed histologically: the thickened contour of the centralobular perilobular vascular wall,

the retained conjunctival architecture. The thickened contour of the vascular wall is clarified by the presence of collagen fibers which is a favorable condition for the mattrices used for recellularization (fig. 6).

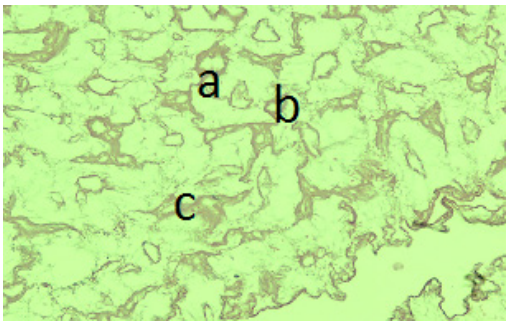

Fig. 6. Decellularized liver by the SDS with anticoagulant washing. Centrolobular vein with thickened contour (a), the thickened liver triad casing (b), preserved liver conjunctive architecture (c). H-E, x 90.

The comparative histological study of decellularized liver scaffolds demonstrated a greater efficacy of decellularization by the previous washing by anticoagulant (phosphate citrate dextrose) with good collagen architecture preserved.

If we compare the DNA concentration obtained from the biologic samples from the liver subjected to decellularization by both methods with versus the DNA concentration obtained from intact liver tissue we notice that it is lower (tab. 1). In samples obtained by decellularization with SDS, which were washed with anticoagulant solution, the DNA concentration was 1.04 ± 0,25 ng/µl, which constituted 5 times less cellular debris than in decellularized SDS samples and was previously washed only with distilled water at which the concentration is 5.2 ± 1,266 ng/µl. This can be explained by the fact that the anticoagulant used allowed more efficient removal of blood clots, thus allowing the ionic decellularization agent to pass through the entire surface of the liver to produce cell lysis and eliminate it.

As the preservation of the extracellular scaffolds after decellularization is a necessary condition for subsequent *in vitro* cellular repopulation, we determined the free hydroxyproline content that is directly proportional to the collagen content in the analyzed biological material. Therefore, we obtained an amount of hydroxyproline of 0.256 ± 0.019 mM/g of dried tissue from intact biological material which is much lower compared to the amounts of hydroxyproline in decellularized samples that are 2.035 ± 1.255 mM per g of dried tissue by the method in which was not used phosphate citrate dextrose and 4.62 ± 0.33 by the method in which was used anticoagulant. The lower amount of hydroxyproline in intact tissue is explained by the presence of cells, that is to say, the same amount of material taken for the research of intact tissue contains cellular components as well as the connective tissue component, but in the extracellular scaffolds tissues predominate the elements of the conjunctive tissues. The fact that the method of using anticoagulant contains higher amount of hydroxyproline results in a more efficient decellularization and the presence of a lower amount of cellular residue.

To see the hepatic vessels integrity the intravascular injection of the blue tripan through the portal vein was done, and it was distributed through all the liver vessels, which revealed that the vessels are intact and that the ionic detergent did not cause their mechanical destruction (fig. 7).

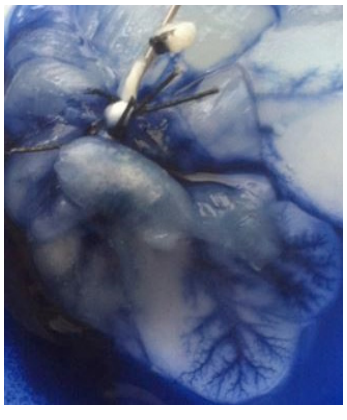

Fig. 7. Contrast with blue trypan of hepatic vessels.

Discussion

A good preservation of the extracellular scaffold would allow future differentiation, proliferation and maintenance of cellular functionality after *in vitro* repopulation, that is a priority in organ bioengineering [6, 18].

In extracellular matrixes obtained from animal organs by decellularisation, which is ideal to completely decode, a mandatory step to estimate the degree of decellularization is needed, which can be done based on the spectrophotometric quantification of total genomic DNA in the residual cell after the step decellularization [26]. The results obtained in our research have shown that the received three-dimensional matrices contain a small amount of residual cellular material, so the matrices are less immunogenic, which would not favor posttransplant rejection. Joery De Kock et

Table 1

The content of DNA and hydroxyproline in intact and decellularized liver

Type of biological material	Hepatic native tissue	Residual liver tissue after decellularization by SDS method without anticoagulant solution washing	Residual liver tissue after decelularization by SDS method and previous anticoagulant solution washing
DNA Concentration (ng/ µl)	93.3 ± 0.41	5.2 ±1.266	1.04 ± 0.25
Hydroxyproline content mM to g of dried tissue	0.256 ± 0.019	2.035 ± 1.255	4.62 ± 0.33

Note: statistically significant difference with the control group * p <0.05.

al. show that extracellular matrices in their research retain their structural components such as collagen and laminin [14], and in our researches the hydroxyproline content revealed the preservation of architecture of the obtained matrix. Also detergents used in the decellularization process cause cell lysis and cell elimination, but their time of action and concentration should be monitorised for the preservation of extracellular matrix proteins [13].

We have obtained extracellular liver matrices using SDS ionic detergent that has cell membrane solubilization potential and complete cell removal, but according to literature it may be used to obtain a matrix with improved chemical, physical, biological properties and combinations of these agents [1]. The concept of organs bioengineering involves not only obtaining a three-dimensional scaffold after removing cells with immunogenic potential, but also preserving vascular network structures for efficient supply of oxygen and nutrients [2, 21, 22]. According to experimental studies of Basak E. et al. the preserving of the microvascular structure of the extracellular matrix after decellularization is essential for its functionality in case of its transplantation [4].

The introduction of the blue tripan solution highlighted the intact structure of the blood vessels of the liver, indicating an eficiency of the used method and favorable conditions for the subsequent repopulation of the obtained extracellular matrices. The extracellular matrices obtained by decellularization can be used not only for transplantation but also as a three-dimensional *in vitro* model for cytotoxicity testing of some drugs because the liver is the main metabolizing organ [11].

Conclusions

1. The comparative evaluating of the DNA tissues content after decellularization by those two methods in relation to intact tissue proves that the method of decellularization of the liver with SDS and washing with phosphate citrate dextrose shows better results.

2. The maintaining of connective architecture and collagen fibers, the higher content of hydroxyproline in ECM obtained by our method demonstrated their high feasibility for the subsequent use as bioengineering structure for recellularization process.

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Evolution peculiarities of the flu in pregnant women

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Abstract

Background: Infection of the pregnant women in advanced period of pregnancy with any of the virus serotypes represents a danger not only for the developing fetus (e.g. fetal intrauterine retardation, fetal distress, oligoamniosis, etc.) but also for the pregnant woman who may develop respiratory and cardiovascular deficiency. We are interested to determine the peculiarities of the clinical evolution, of the flu diagnosis in pregnant women.
Material and methods: The general study group consisted of 189 pregnant women which was divided into 2 clinical subgroups: L1, which included pregnant women with confirmed flu (n_1=42) A(H1N1) and subgroup L2 of pregnant women with seasonal flu (n_2=147).
Results: The results of the examination of pregnant women for the flu virus's DNK confirmed high frequency of pandemic flu in 2009, P<0.001. Since 2010-2017 the epidemiological state is reversed, with a high frequency of seasonal flu 81.6% (110), P<0.001. The development of both forms (seasonal and A (H1N1)) of the flu is presented by two clinical syndromes: toxico-infectious and catarrhal, P>0.05. The study of laboratory investigations of women with the flu demonstrates the presence of similar changes in the both groups: iron deficiency anemia 93 (49.2%); leukopenia 84 (44.4%); leukocytosis with neutrophilia 4 (2.1%); lymphocytosis 42 (22.2%), P>0.05. The most frequent complications of the pregnancy in pregnant women with the flu were premature delivery (early/late periods of pregnancy) – 15 (33.3±7.27) in L_1 and in L_2 – 35 (17.7±3.15), p<0.05, followed by intrauterine fetal infestation syndrome (fetal hypoxia, intrauterine fetal development retardation, oligoamniosis, placental dysfunction) – 33 (17.4%), p<0.05.
Conclusions: Molecular methods of diagnosis to determine the virus serotype underlie the establishment of the definitive diagnosis of the flu. The risk of pregnancy disruption at various terms, premature birth, intrauterine fetus infestation syndrome are the most common complications in pregnant women with the flu in the study.

Key words: evolution, the flu, pregnant women.

Introduction

The flu is a severe infectious contagious disease with a benign evolution in the majority of cases with the exception in children, pregnant women, old people and adults with chronic cardiovascular diseases [1,13,15,19]. It is one of the most frequent viral respiratory infections, very often with severe evolution, being one of the causes of mortality (during epidemics and pandemics) in the general population [2,10,12,16]. Forms of the flu stated as pandemics in the previous years, with new types of viruses, avian A(H1N1), represent a combination between human and animal viruses (swine and birds) [1,2,3,24,25,26] and have been a global health problem. In pregnant women the flu has a tendency towards severe evolution (as a result of immune status suppression), with the disease-related lethality index being doubled as compared to that of the general population [9,10,11,12,23]. It is well known the teratogenicity of flu viruses on the conception product, with a 2,4-fold increase in congenital malformations in pregnant women infected during embryogenesis [17,18,21,22]. Infection of the pregnant women in advanced period of pregnancy with any of the virus serotypes represents a danger not only for the developing fetus (e.g. fetal intrauterine retardation, fetal distress, oligoamniosis, etc.) but also for the pregnant woman who may develop respiratory and cardiovascular deficiency [4.5.6.8].

Paper goal is to determine the peculiarities of the clinical evolution, of the flu diagnosis in pregnant women. Objectives: 1. To find the anamnestic peculiarities (including epidemiological anamnesis), clinical in pregnant women with the flu in the study; 2. Analysis of laboratory parameters in pregnant women with different forms of the flu; 3. State the impact of the flu on maternal and fetal health.

Material and methods

The study is based on the analysis of clinical cases with the flu in pregnant women hospitalized in Municipial Maternity No 2 during 2014-2017. The general study group consisted of 189 pregnant women which was divided into 2 clinical subgroups: L1, which included pregnant women with confirmed flu (n_1=42) A(H1N1) and subgroup L2 of pregnant women with seasonal flu (n_2=147).

The following methods of study have been used to achieve the proposed goal and objectives:

– The accumulation of data from the medical documentation with the elaboration of a questionnaire about the pregnancy history (including the epidemiological anamnesis with the purpose to identify the source of infection), the onset, ambulatory treatment;

– The general clinical (including respiratory) and obstetrical examination;

– Laboratory tests (general analysis of the blood, general urine analysis, biochemical analysis of blood (determining the hepatic, renal functions, general protein, reactive C protein), blood coagulation – platelet number, prothrombin, fibrinogen;

Table 1

Clinical evidence of the flu in the study groups

No	Clinical evidence	Flu A(H1N1) N1=42		Seasonal flu N2=147		t	P
		Abs.	P1±Es1	Abs.	P2±Es2		
1	Fever	38	90.5±4.52	125	85.0±2.95	1.0188	>0.05
2	Myalgia	9	21.4±6.33	43	29.3±3.75	1.0737	>0.05
3	Arthalgya	5	11.9±4.99	16	10.9±2.57	0.1779	>0.05
4	Asthenia	40	95.2±3.29	147	100.0±0.00	1.4552	>0.05
5	Headache	35	83.3±5.76	132	89.8±2.49	1.0362	>0.05
6	Dry cough	33	78.6±6.33	99	67.3±3.87	1.5734	>0.05
7	Wet cough	9	21.4±6.33	48	32.7±3.87	1.5234	>0.05
8	Sore throat	17	40.5±7.57	56	38.1±4.01	0.2801	>0.05
9	Chest pain	3	7.1±3.96	28	19.0±3.24	2.3260	<0.05
10	Running nose	10	23.8±6.57	31	21.1±3.37	0.3657	>0.05
11	Rhinorrhea	32	76.2±6.57	77	52.4±4.12	3.0688	<0.01
12	Dispnoea	3	7.1±3.96	9	6.1±1.97	0.2258	>0.05

– Virusological methods, nasopharyngeal lavage PCR collected in the first 24-48 hours in pregnant women with temperature ≥38 ° C, pulseoximetry (≤95%), clinical symptoms of respiratory failure (tachypnoea, dyspnoea) and intoxication;

– Paraclinical methods: pulseoximetry, chest digital X-ray in pregnant women with temperature ≥38 ° C, pulseoximetry (≤95%), clinical symptoms of respiratory failure (tachypnoea, dyspnoea) and intoxication; electronical fetus recording (CTG), ultrasonography with Doppler;

– Biostatistical methods of processing the primary material using the "Epiinfo 2002" and "Excel" programs from the Microsoft Office package in the personal computer.

To comparatively analyze the indicators, mathematical and statistical techniques that have been applied (proportional indicators, average values, etc.). The veracity of the study was stated by calculating relative value errors ("ESp") and average ("ESm"), the "t- Student" criterion.

Clinical peculiarities of the flu evolution in pregnant women from the study (own results)

The study group consisted of 189 pregnant women, divided into 2 clinical subgroups: with confirmed flu A(H1N1) and with seasonal flu. The pregnant women with all forms of the flu were urgently hospitalized while being in different periods of pregnancy, the general condition at admission being considered satisfactory in 62 pregnant women (33%); average severity in 125 (66%), and 2 cases (1%) of severe condition that resulted in the death of pregnant women in other specialized institutions.

The study of the disease history determined the rapid onset of the flu A (H1N1) with toxic and catarrhal syndrome, which contributed to early admission (in the first 3 days of illness): 32 cases (76%), p<0.05.

The epidemiological anamnesis analysis revealed the appearance of the disease after the contact with sick or in recuperation period people in 14 cases (33.3%) in L1 and 60 patients (41%) in L2. However, in most cases, the source of the infection has remained unidentified and may indicate free circulation of the virus during the epidemic, and the low level of the general population culture in the use of protective measures. Also, a link between the flu illness rate and the seasons of the year in both flu forms 177 (93.6%), P<0.001 has been identified, namely in autumn-winter periods (November, December) and winter-spring (February, March).

The development of both forms of the flu is presented by two known clinical syndromes: toxico-infectious (e.g. general asthenia, fever, headache, myalgia, arthalgya) and catarrhal (dry cough occurring in the first days of the disease, followed by wet cough with expectoration in 3-4 days, sore throat), manifesting themselves identically in both groups (tab. 1).

The clinical examination of the pregnant women in the study reveals objective clinical data of a severe viral infection: increased respiratory frequency (FR) determined in 35 pregnant women (18.5%), P<0.01, light/average tachycardia in 100% (189), P>0.05; decrease of saturation SpO_2 > 95% in 12 pregnant women (6.3%), P>0.05; changes in the auscultation of the lungs – dry breathing with dry rallies (which suggests acute bronchitis), crepitation with wet breathing (diagnosed with pneumonia) in 82 pregnant women (43,3%), P>0.05 (tab. 2).

The clinical examination is an important part in the diagnosing of the flu complications (bronchitis and pneumonia), determined by a similarly rate in both groups of study.

The results of the examination of pregnant women for the flu virus's DNK are presented in the table 3 which confirmed high frequency of pandemic flu in 2009 (3.3 higher than in the case of seasonal flu), P<0,001. During the period of 2010-2017 the epidemiological state is reversed, with a high frequency of seasonal flu 81.6% (110), P<0,001 (tab. 3).

Table 2

Clinical examination of the pregnant women included in the study

Clinical data		Flu A (H1N1) N1=42		Seasonal flu N1=147		P
		aA bs	P1±Es1	aA bs	P2±Es2	
	Lung auscultation					
1	Without changes	11	26.2±6.79	86	58.5±4.06	<0.05
2	Dry rallies+harsh breathing	19	45.2±7.68	56	38.1±4.01	<0.05
3	Crepitation +wet breathing	2	4.8±3.29	5	3.4±1.49	<0.05
1	SpO_2 a) 99-100 % b) 96-98% c) 90-94%	 a) 20 b) 19 c) 3	 47.6±7.71 45.2±7.68 7.1±3.96	 a) 99 b) 39 c) 9	 67.3±3.87 26.5±3.64 6.1±1.97	 <0.05 <0.05 >0.05
2	Ps a) 80-100 b)> 100	a) 39 b) 3	92.9±3.96 7.1±3.96	a)131 b) 16	89.1±2.57 10.9±2.57	>0.05 >0.05
3	FR a) 16-20 b) 20-25	a) 39 b) 3	92.9±3.96 7.1±3.96	a) 115 b) 32	78.2±3.41 21.8±3.41	<0.01 <0.01

Table 3

PCR results of the pregnant women with the flu

Confirmation (PCR)	Flu A (H1N1)N=42		Seasonal fluN=147		t	P
	Abs.	P1±Es1	Abs.	P2±Es2		
2009	26	61.9±7.49	27	18.4±3.19	5.3396	<0.001
2010-2014	16	38.1±7.49	110	81.6±3.19	5.3396	<0.001
2015-2017	0		10	6.8±2.08	3.2749	<0.01

The study of other laboratory investigations (tab. 4) of women with the flu demonstrates the presence of similar changes in the both groups: iron deficiency anemia 93 (49.2%); leukopenia 84 (44.4%); leukocytosis with neutrophilia 4 (2.1%); lymphocytosis 42 (22.2%), P>0.05. Thus, the results of the above parameters determined the presence of leukopenia in the majority of pregnant women due to the immunodepressive and cytotoxic action of the virus on the body affected by the disease. According to literature, severe flu forms complicated by bacterial infection are characterized by leukocytosis with neutrophilia, increased fibrinogen and reactive C protein [23,24,25,26].

The most frequent complications of the pregnancy in pregnant women with the flu were premature delivery (early/late periods of pregnancy) – 15 (33.3±7.27) in L_1 and in L_2 – 35 (17.7±3.15), p<0.05, followed by intrauterine fetal infestation syndrome (fetal hypoxia, intrauterine fetal development retardation, oligoamniosis, placental dysfunction) – 33 (17.4%), p<0.05.

Table 4

Laboratory results in pregnant women with the flu in the study

No	Laboratory tests (obtained data)	Flu A(H1N1) N1=42		Seasonal flu N2=147		t	P
		Abs.	P1±Es1	Abs.	P2±Es2		
1AGS:	Anaemia	26	61.9±7.49	67	45.6±4.10	1.9545	>0.05
2	Lymphocytosis	8	19.0±6.05	34	23.1±3.48	0.5874	>0.05
3	Monocytosis	12	28.6±6.97	26	17.7±3.15	1.4248	>0.05
4	Leukopenia	21	50.0±7.72	63	42.9±4.08	0.8134	>0.05
5	Leucocytosis	3	7.1±3.96	1	0.7±0.69	1.5912	>0.05
1. Blood biochemistry							
2	High fibrinogen	3	7.1±3.96	1	0.7±0.69	1.5912	>0.05
3	Reactive protein C	4	9.5±4.52	0			<0.05
4	Hipoproteinemia	6	14.3±5.40	3	2.0±1.15	2.2267	<0.05

Conclusions

1. The risk of getting the flu (60%) is increased in pregnant women who had direct contact with sick people.

2. The clinical symptoms of the flu are similar when infected with different serotypes, manifested by catharal and toxico-infectious syndrome, with a moderate/severe evolution (74%) in pregnant women with the flu A(H1N1).

3. Molecular methods of diagnosis to determine the virus serotype underlie the establishment of the definitive diagnosis of the flu.

4. The risk of pregnancy disruption at various terms, premature birth, and intrauterine fetus infestation syndrome are the most common complications in pregnant women with the flu in the study.

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Relationship between personality disorders and headaches using PID for DSM-5

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Abstract

Background: Studies on the specificity of migraine headache in patients with personality disorders are multiple. Results are often contradictory, which may be explained by psychological, socio-cultural, economic and purely individual differences of subjects.
Material and methods: 128 patients from the Department of Headache and Autonomic Disorders of the Institute of Neurology and Neurosurgery (Chisinau, the Republic of Moldova) were evaluated in this study, in 2 stages: psychometric testing using Personality Inventory Disorders (PID-5) for DSM-5 in the 1st stage and data collection, headache intensity assessment and Headache Questionnaire in the 2nd stage.
Results: The results of psychometric test allowed to separate the examined subjects into 3 groups according to numeric values of facets of PID-5: group I – Normal (0-1), group II – Accentuated Personality (1 – 1.66), group III – Personality Disorder (>1.66), and these results were correlated with intensity and frequency of headache. The analysis of 25 facets of PID-5, which are included in 5 domains of higher order: Negative Affection, Antagonism, Disinhibition, Detachment and Psychoticism, divided the domains into 3 groups: Internalization, Externalization and Psychoticism. These values were correlated again with intensity and frequency of headache.
Conclusions: Female gender has a higher introversion tendency than males, introversion and neurosis is more common among women with migraine; the onset of personality disorders occurs during early youth.
Key words: personality disorder, PID-5, headache.

Introduction

A basic hypothesis of personality theory claims that individual behavior is constant in different contexts. This fundamental assumption was strongly disputed by several authors, with asserting the circumstances as the most important determinant of behaviour. However, there are many statements that the conservation over time of the recurrent migraine attacks features and of the traits of personality are probably linked together.

Over the years, many studies have been focused on the connection between particular personality traits and headache syndromes. Many researchers have used the Minnesota Multiphasic Personality Inventory (MMPI) to investigate the personality profiles of people with headaches. The neurotic triad – hypochondria, hysteria and depression in tensional and migraine headaches has been presented by many studies. It is sensible to assume that certain personality traits may increase the vulnerability of a person suffering headaches. Disputed questions are related to personality traits of patients with tension-type headache and migraine and the differences with healthy subjects [5, 7].

Personality disorders affect about 10% of the general population and are typically refractory to standard pharmacology and behavioral interventions [4, 20]. The rate of migraine and the rate of personality disorder are higher in women, and the gender differences influence the perception of pain as well as the style of coping [9, 13, 19]. The scores of introversion and the incidence of neurosis are significantly higher among women with migraine [8, 23]. A study on female migraine patients has reported a strong correlation between symptoms of neurosis and headache duration (r = 0.51) [13, 15]. Contrariwise another large study, including men and women, has no found relationship between severity of neurosis and duration of headache [6, 20].

Headache is a common phenomenon in everyday life and in the clinic, constituting an important public health problem, with a large impact at individual and society level, confirmed by multiple epidemiological researches, which attests to a high prevalence of headache of the population. Described since ancient times, headache, as well as the problem of pain in general, has become in recent years one of the most advanced chapters of medicine [7, 16].

Tensional headache (TH) is the most prevalent of all headaches, and it is also one of the most expensive clinical conditions for the health system. The knowledge of tensional headache is still quite limited despite the significant

impact of this disorder on patients' lives and the existence of established diagnostic criteria. There is a group of patients with the chronic subtype of this headache, whose quality of life is greatly compromised [18, 14, 24].

The presence of psychological symptoms has been highlighted in patients with migraine. Several studies have investigated personality traits using structured tools: The Personality in Vivo for DSM-5, PID of the Manual of Diagnosis and Statistics of Mental Disorders, 5th (DSM-5) [1, 2, 3]. The International Classification of Headache Disorders (ICHD) was applied for migraine patients. These studies have proved a 2-4-fold greater risk of major depression in migraine patients. This association between migraine and depression is much stronger for migraine with aura. Other findings are: higher risk of panic disorder, phobic disorder, generalized or post-traumatic stress disorder [20].

Onset, development, clinical picture and treatment of headaches depend heavily on the psychological state of the patient. Patients with personality disorders (PD) respond differently to pain than healthy people from this point of view. Several recent studies suggest that people with migraine are more likely to suffer personality disorder than people without migraine. Personality disorders affect 26% of patients with chronic refractory headache, the most common being: borderline, narcissistic, antisocial, avoidant, obsessive-compulsive [11, 20]. The profile of personality seems to be linked with the frequency of headaches, connections to somatic problems, excessive use of medication [20].

A personality disorder or an accentuated personality could explain the resistance to treatment of headache. It is important to note these differences in the therapeutic approach to these patients. The inclusion of psychotherapy, psychiatric drugs, in addition to analgesics, would considerably diminish the suffering of these patients; improve the quality of the therapeutic act, and the quality of life of the suffering person.

Since migraine pain is intense and disabling for patients, it is possible for transforming into chronic form. The association of migraine pain with personality disorders possibly aggravates the development of the disease. Thus, it is important to study the characteristics of migraine pain in patients with personality disorders, to assess the degree of severity, and to choose the appropriate treatment options and prophylaxis.

Recent data on affective and personality disorders in patients with migraine are often contradictory (which can be explained by the socio-cultural, economic and individual psychological differences). Factor analysis identified two factors that are likely to be the two mechanisms that "control" human behavior, as well as his psycho-physiological state. The authors emphasize the methodological difficulties in solving the puzzle of "migraine personality"; especially, the fact that affective disorders are almost an inevitable comorbidity of migraine, which "overshadows" the access to the true personality of the patient, as such. The authors point out the difficulty of future studies that will take into account the insights of recent publications in the study of headache and associated comorbidities, as well as modern approaches related to the new vision of the concept of personality disorder (accordingly to DSM-5) [17]. There are few comparative studies on the patients with two (or more) different headache syndromes. The specificity of migraine pain in patients with personality disorders is still little investigated [5, 6].

The goal is the study of the personality disorder's degree correlation with the clinical profile in patients with migraine headache and tensional headache.

Objectives: 1. Studying the characteristics of migraine pain and tensional headache depending on the degree of personality disorder; 2. Analysis of the specificity of headache according to the type of personality disorders.

Material and methods

128 patients from the Department of Headache and Autonomic Disorders of the Institute of Neurology and Neurosurgery (Chisinau, the Republic of Moldova) were evaluated in this study between March 2016 and February 2017. The diagnosis was already confirmed in all patients. The average age of patients was 32.5 years (from 18 until 59 years of age). The study evaluated the patients in two stages.

First stage – Psychometric testing

The Personality Disorder Inventory (Personality Inventory for DSM-5 (the Manual of Diagnosis and Statistics of Mental Disorders), PID-5, is a tool for assessing personality traits, developed by the American Psychiatric Association (AAP) in 2012. In 2011, the AAP proposed a substantial revision of the methods of diagnosing personality disorders. This proposal included a hybrid model in which a categorical diagnosis of personality disorder is divided on the basis of dimensional and pathological personality features.

The analytical approach is based on modern psychometrics tools [12].

Inventory of Personality for the DSM-5 (PID-5) has been translated and validated by a working group made up of collaborators of the Department of Human Physiology and Biophysics, and the Department of Headache and Autonomic Disorders within the Institute of Neurology and Neurosurgery, in compliance with norms of translation, adaptation and validation of International Test Commission and with the consent of the authors.

The Structure and Contents of the Inventory of Personality for DSM-5

Inventory consists of 220 items of self-report and is used to measure disadaptative traits of personality. The responses are selected from 4 variants, from 0 ("very false or often false") till 3. Thus, PID-5 provides scores evaluated on a scale of 4 points, on the 25 facets.

Each facet includes four to fourteen elements. These characteristics correspond to the features of disability of personality, described in the section III of the DSM -5, included in five domains of higher order: negative affect, separation, antipathy, disinhibition and psychoticism. The score higher than 2 of a certain feature makes quantitative

estimation of one of the six types of personality: Antisocial, Borderline, Schizotypal, Evasive, Obsessive Compulsive or Narcissistic [1, 2].

Generally, based on the results obtained from the PID-5 test, it is possible to allocate the numerical scores for each feature from which the profile of the personality disorder is formed.

For example, for the borderline personality disorder we study the distribution of the numerical results of 7 facets. For a positive result, 4 of the 7 must be above the 2 on the ordinate axis (fig. 1).

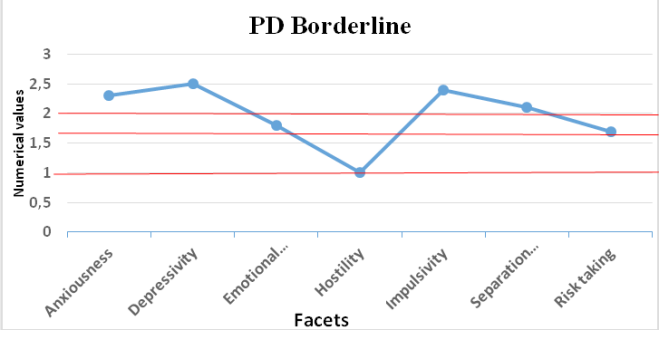


Fig. 1. Profile of personality disorder Borderline (of one patient).

Note: distribution of numerical values of 7 facets is studied for personality disorder borderline. A positive resultat is attested when 4 from 7 values are greater than 2.

For most patients included in the study, PID scores attained high numerical values for facets but did not reach the value of 2 (on the graph of the ordinate axis), these could only be placed "at the limit of disorder", as they did not accumulate the number of points needed to be included in the group of subjects with personality disorders (fig. 2).

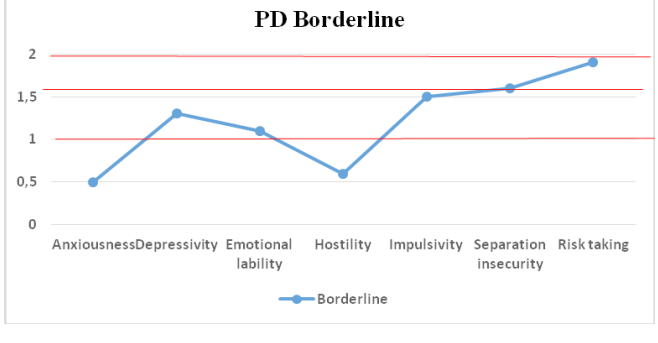


Fig. 2. Profile of the traits of personality in the subject with accentuated personality.

Note: subjects with personality disorder borderline, with numerical values of PID facets greater than 1.66.

Figure 2 shows that the numerical results of the PID that generates the Borderline personality disorder were distributed below the value of two, which indicates that the person does not have a definite disorder, however, up to 1, i.e. within the limits of the norm there are only two facets (anxiety and hostility), the others being between 1 and 2, that is, there is a tendency towards the borderline disorder. The detailed analysis of the degree of accentuation of the facets in 425 patients highlights that facets with numeric value between 1 and 1.66 are included in so called "area of accentuation". Thus, the numeric value of the facets higher than 1.66 exceeds the range of the accentuation and is considered as 2, marking the presence of the characteristic trait of the respective disorder.

Starting from the above, it was decided in this study to modify the numerical values limits in the formation of the groups, so that subjects with numerical results of PID facets less than 1.66 would be considered conventionally subjects with accentuated traits of personality disorder.

The distribution of facets and their numerical values, specifically, a value of 1.66, allows the segregation of the examined subjects into three groups:

– Group I-PID: 0-1 – Normal (hereafter, I-N group);

– Group II-PID: 1-1.66 – Accentuated personality (II-AP);

– Group III-PID: 1.66 < – Personality disorder (III-PD).

The 25 facets resulting from the Inventory correspond to the features described in DSM-5 and comprise five domains of the supreme order, as described in Sequence III: Negative Affect, Separation, Antagonism, Disinhibition and Psychoticism.

The hierarchical structure of the PID-5 traces is derived from the 5-level model [10,19], the first level being the global pathology of personality, at level two the general factor can be divided into problems of externalization and of internalization. At level three, the outsourcing factor maintains its structure, and the internalizing factor is divided into two higher order domains – negative affectivity and detachment. At level four, the negative affectivity of detachment maintains its structure, while the externalization is divided into two areas of superior order – antagonism and disinhibition. At level five, the four top-level domains are preserved, and the fifth is psychoticism.

Drawing from the exposed and analyzing the aspect of the 25 facets (personality traits) of PID-5, which fall into five areas of higher order personality: Negative Affection, Antagonism, Disinhibition, Detachment and Psychoticism, and studying the clinical significance of each field in the Session III DSM-5, it becomes possible to divide the domains into 3 groups according to the tendency the individual manifests when a personality domain or another prevails over the conscious and unconscious. This division allows the creation of the study groups as follows:

I group: Internalization: Negative affection + Detachment;

II group: Externalization: Antagonism + Disinhibition;

III group: Psychoticism.

In this study, it was decided to divide the group of patients according to these three groups, this being the second stage of statistical research.

The second stage of the study included the following research methods

Data collection – anamnesis, objective examination, neurological status of patients' observation file, diagnosis in all patients was already confirmed.

Headache intensity assessment via analogue numerical scale (ANS) – is used to measure pain intensity, maximum score of 10 points (0 points – absence of pain, 10 points – the most intensive pain felt by the patient);

The Headache Questionnaire was adapted by the Center of Headache and Autonomic Disorders of the Institute of Neurology and Neurosurgery in 2011. It consists of 18 compartments, which are analyzed together, generating the characteristics of headache: the age of onset of headache, the number of days with pain per month, duration of access, location of pain, activities that amplify the pain, the type of pain, the signs that precede the pain, the trigger factors, headache accompanying symptoms, behavior during access, pain intensity according to the number of days a month with pain, hereditary anamnesis, use of analgesic drugs, the presence or absence of drug abuse, comorbidities, family status.

Respectively, all patients included in the study based on the already confirmed diagnosis were divided into 3 groups:

Group 1 included 48 patients with migraine: 22 patients with episodic migraine and 26 patients with chronic migraine. Diagnosis in headache patients was established according to the 3rd International Classification of Headache Disorders of the International Headache Society ICD-III.

Group 2 included 14 patients with tension headache (10 patients with episodic CTT and 4 patients with chronic CTT).

Group 3 included 66 patients with affective disorders, of whom 30 are only affective disorders, and 36 have rare episodic headaches that account for an important percentage of their general affliction. The diagnosis was established according to the Diagnostic and Statistical Manual of Mental Disorders (5th Edition) (DSM-V).

Statistical methods. The ANOVA (Analysis of Variance with Bonferroni Correction) and the Independent Proportion Method were used to assess the difference between the scores of the questionnaires in different patient groups. For detecting differences between samples, the x^2 (ch^2 square) index proposed by Helmet and Pearson was used.

Results and discussion

The alternative model of diagnosis of personality disorder states that the scores under the diagnostic criteria (or clinical threshold) do not allow individuals to be qualified as having that disorder. This is useful because it recognizes that there is a potential problem without giving the person the negative label associated with personality disorders. This also prevents a clinician from giving the client complete treatment. A total evaluation by the alternative method measures all the traits associated with personality disorders. Each of the five domains has 3-6 facets that can be examined.

For instance, the antagonisms include six facets: manipulativeness, deceitfulness, grandiosity, attention seeking, callousness and hostility. Any facet that clearly describes the patient, valued by numeric value, is then marked as being present and is then considered clinically important. Facts that have a value in mind are still to be taken into account, thus providing a more comprehensive view of the patient. "Good" facets appreciated numerically are not taken into account for qualifying the personality disorder. There are specific, strict facets describing each personality disorder, and the presence of those facets determines whether the patient has or does not have personality disorder in the event of schizotypal disorder.

Based on the above, the distribution of patients in 3 study groups according to the distribution of facets and numerical values was based on the fact that the majority of patients whose results of PID do not fall within the notion of "pathological personality", but still have a fairly large number facets with a numerical value of 1.5. They can also induce changes in personality, with a tendency towards pathology, changes that we suppose to change painful headache, as well as affective manifestations, which also require corrections in their therapeutic approach. The comparative characteristic of the socio-demographic data of the patients according to the created groups is presented in tab. 1.

Table 1

Socio-demographic data of the patients

	Group I N	Group II AP	Group III PD
Gender Female Male Total	 49 (38.28%) 27 (21.09%) 76 (59.37%)	 31 (24.21%) 12 (9.37%) 43 (33.59%)	 6 (4.68%) 5 (3.90%) 9 (7.03%)
Average age (years)	34.08	30.88	36.66
Family status Married Non-married Divorced Widowed	 48 25 7 2	 26 15 1 1	 0 2 1 0

Note: Group I N - PID : 0-1; Group II AP - PID: 1-1.66; Group III PD - PID: 1.66 <

The table shows the prevalence of women in AP II and III PD lots, which indicates the higher risk for female sex to develop a personality disorder. Family status does not seem to be of any importance, as a risk factor in the occurrence of personality disorders.

The study of the characteristics of migraine pain and tensional headache depending on the degree of personality disorder is shown in fig. 3.

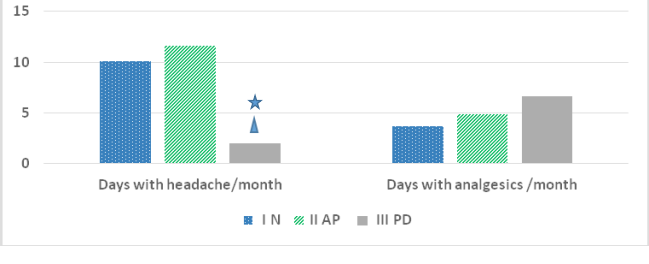
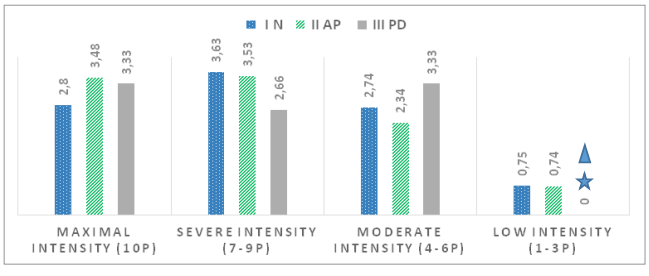


Fig. 3. Numerical values of headache traits according to PID groups (number of days with headache per month, number of days with use of analgesics per month).

Note: P<0.01 ▲ – comparison of groups II and III; ★ – comparison of groups I and III.

Comparison of the headache characteristics between groups made accordingly with scores PID-5 suggests that the use of analgesics is more intense in patients with personality disorders, as well as risk of developing drug abuse is greater (IN group - 3.69 ± 0.63, II AP - 4.88 ± 0.74; III PD - 6.66 ± 1.15). The number of days per month with pain differs significantly between the I-N group, where a lower frequency of headache is recorded than in group II (I-N – 10.12 ± 1.05, II-AP – 11.62 ± 0.95, P <0.01) and a very low frequency is recorded in the 3rd group (I-N – 10.12 ± 1.05, III-PD – 2.00 ± 0.17, P <0.01).



alues of headache traits
(intensity of headache) according to study groups.

Note: P<0.01 ▲ – comparison of groups II and III; ★ – comparison of groups I and III.

The intensity of headache assessed using the headache questionnaire is illustrated in fig. 4, from which the significant changes occur only in low intensity pain, which corresponds to the score 1-3 according to the numerical-analog scale. Thus, between the I-N group (0.75 ± 0.09) and II-AP group (0.74 ± 0.09), P <0.01 and between the I-N group (0.75 ± 0.09) and III-PD (0.00), P <0.01, there is a large statistical difference. Severe pain is more pronounced in patients without personality disorder, and means pain is more felt by patients in the third group [22].

Studying the clinical significance of each domain, according to the DSM -5 (Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition, 2013), we divided the 5 domains into 3 groups, and to the tendency of the individual when a domain prevails on the conscious and the unconscious [9]. These groups are:

1. Internalization: Negative affection + Detachment
2. Externalization: Antagonism + Disinhibition
3. Psychoticism

Based on the above, in the second stage of the statistical analysis in this study, all patients were divided into three study groups. The comparative characteristic of the socio-demographic data of the patients according to the created groups in the 2nd stage of study is presented in tab. 2.

The prevalence of women in the Internalization group suggests the hypothesis that women tend towards greater introversion compared to men. In Group II (Externalization), male gender prevails. The average age of 24.71 years in the third group (Psychoticism) proves an early onset of personality disorders.

The comparison of the numerical data of the headache characteristics in the study groups is represented in fig. 5.

Table 2

Socio-demographic data of the patients in study groups

	Group I Internalization	Group II Externalization	Group III Psychoticism
Gender			
Female	73 (57.03%)	11 (8.59%)	5 (3.90%)
Male	21 (16.40%)	16 (12.50%)	2 (1.56%)
Total	94 (73.43%)	27 (21.09%)	7 (5.46%)
Average age (years)	34.28	31.00	24.71
Family status			
Married	59	15	2
Non-marriedi	25	11	4
Divorced	8	0	1
Widowed	2	1	0

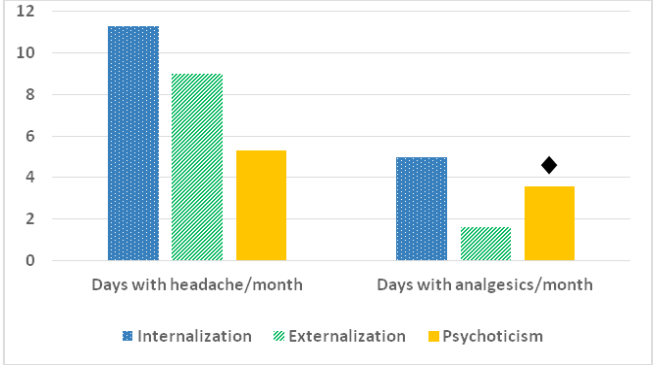


Fig. 5. Numerical values of the headache traits in study groups.
Note: P<0.05 ◆ – comparison between groups I and II.

The use of analgesics is more pronounced in the patients of the Internalization group (4.94 ± 0.75), compared to the Externalization group (1.59 ± 0.41), with a statistically significant difference (P <0.05). Similarly, the number of days with pain per month is higher for patients in the Internalization group. Externalization helps reducing the frequency of pain.

The duration of headache attack is higher in patients in the first group (11.25 ± 0.95), in comparison with the third group (2.35 ± 0.22), with a statistically significant difference between them (P <0.05, fig. 6), which confirms the role of inhibition and introversion in the manifestation of the duration of headache [22].

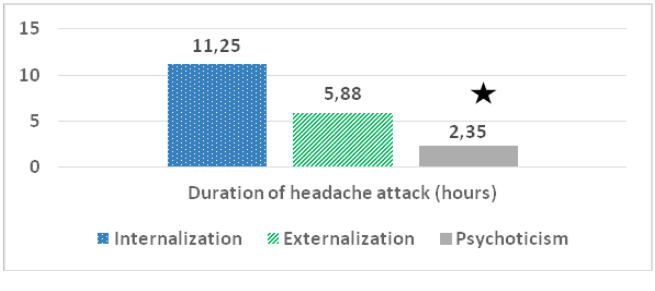


Fig. 6. Numerical values of the duration of the headache attack in study groups.
Note: P<0.05 ★ – comparison between groups I and III

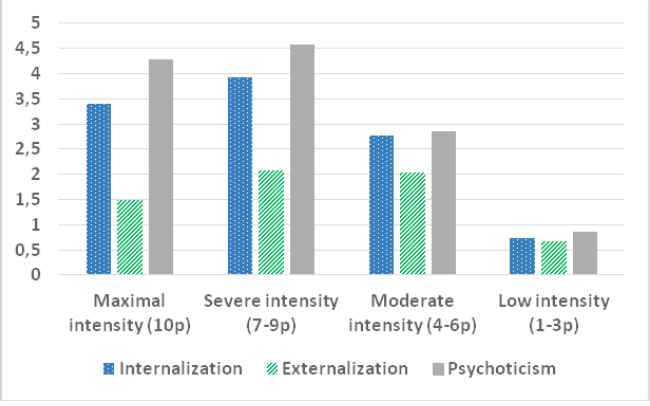


Fig. 7. Numerical values of headache intensity in study groups.

Multiple researchers argue that the personality can be considered as capable of altering the clinical appearance, the evolution of pain, and the response to therapy. The data obtained are shown in fig. 7, namely, that the pain is more intense in patients with psychoticism (group III) and in the Internalization group. Externalization helps reduce pain intensity.

The statistical analysis of the data on the relation between the 6 types of PD according to the latest DSM-5 edition and the higher order personality domains assembled in the three groups – Internalization, Externalization, and Psychoticism are shown in fig. 8. It was noticed that the narcissistic personality disorder is dominating for Psychoticism (1.44 ± 0.53), also high levels in group III are attested to antisocial disorder (1.19 ± 0.5) and obsessive – compulsive disorder (1.39 ± 0.64), with considerable statistical differences (P <0.05) compared to the values in other groups. The borderline disorder is prevalent in Internalization group (1.23 ± 0.94). Externalization manifests itself more in the antisocial and schizotypal disorders [22].

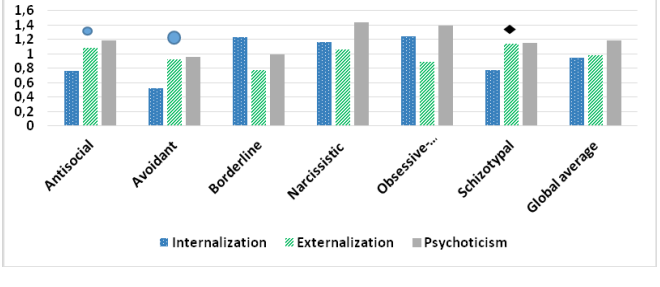


Fig. 8. Numerical values of personality disorders in study groups.

Note: P<0.05 ◆ – comparison between group I and II.

P<0.01 ● – comparison between group I and II.

★ – comparison between group I and III.

⬟ – comparison between group I and III.

Conclusions

The numeric values of the facets of PID-5 test were used to separate the examined subjects in three groups (I – Normal, II – Accentuated Personality, III – Personality disorder), and this division confirms the correlation between personality disorder and migraine, same idea sustained by bibliographic data. The obtained results provide data about a relationship between personality disorder and clinical profile of the pain.

In conclusion, it can be affirmed that the data obtained according to the division based on the numerical bases of the PID-5 facets, the examined subjects in three groups (I – Normal, II – Accentuated Personality, III – Personality disorder) allow to emphasize first the fact that the risk to develop a personality disorder is higher in women, the same is supported by literature data, women show higher rates of migraine and personality disorder than men, and gender differences influence the perception of pain and the style of coping.

The characteristics of headache are influenced by personality disorders, which is confirmed by the use of more potent analgesics, the number of days with pain per month is less influenced, however, it differs in patients without TP from those with a tendency to disorder, the intensity of pain is greatly influenced within the qualifying "Accentuated personality" on the visual-numeric scale.

The distribution of individuals examined in I – Internalization, II – Externalization, III – Psychoticism, based on the clinical significance analysis of the five areas of higher order personalities, allows to state that female gender has a higher introversion tendency than males, introversion and neurosis is more common among women with migraine; the onset of personality disorders occurs during early youth.

The PID test can make a significant contribution to deciphering the definition of "migraine personality".

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REVIEW ARTICLES

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Myocardial revascularization in patients with coronary artery disease and diabetes mellitus

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Abstract

Background: Diabetes mellitus (DM) is one of the most crucial risk factors for morbidity and mortality from coronary heart disease (CAD) in the contemporary world. The prevalence and rapid progression of atherosclerotic lesions leading to worse survival is a defining feature of the course of CAD in patients with DM. Clinical manifestations of CAD often call for revascularization in patients with DM. The contemporary data regarding efficacy of the coronary artery bypass grafting (CABG) and percutaneous coronary intervention (PCI) in patients with CAD and concomitant DM are summarized in the review.

Conclusions: Worse survival prognosis in case of CAD with concomitant DM is associated with systemic atherosclerosis, presence of a number of concomitant risk factors, as well as masked clinical manifestations of ischemia and myocardial infarction common for the patients with DM. The combination of CAD and DM once again proves the benefit of the long-term use of drugs for the secondary prevention of cardiovascular events. From the standpoint of evidence-based medicine the optimal method of revascularization in CAD patients with multivessel coronary artery disease and concomitant DM is CABG surgery. In FREEDOM study surgical myocardial revascularization reduced the number of endpoints compared to PCI. In the case of PCI it is advisable that eluting stents reducing the likelihood of restenosis and the need for repeated revascularization be used.

Key words: diabetes mellitus, coronary artery disease, coronary artery bypass grafting.

Introduction

Diabetes mellitus (DM) is one of the most crucial risk factors for morbidity and mortality from coronary heart disease (CAD) in the modern world. 25-30% of patients with acute coronary syndromes are diagnosed with DM [27]. Clinical manifestations of CAD develop earlier in patients with DM and are associated with a worse prognosis compared with patients without diabetes. CAD associated mortality in patients with DM is two times higher than that in patients without diabetes [31]; CAD is the cause of death of three out of four patients with DM [25]. This allows characterizing DM as an equivalent of CAD with already suffered myocardial infarction (MI) in terms of the risk of coronary events [18]. Clinical manifestations of CAD often call for a specific medical therapy and revascularization in patients with DM. On the other hand, concomitant DM is present in almost 40% of patients with CAD who undergo coronary artery bypass grafting (CABG) [27]. The existing evidence base and existing clinical experience allow us to summarize the current state of the problem of myocardial revascularization in patients with stable coronary artery disease and diabetes.

Diagnosis and the course of CAD in patients with DM

Rapid progression and prevalence of atherosclerotic lesions in the presence of DM is accounted for by a number of adverse effects associated with hyperglycemia, insulin resistance and dyslipidemia. These include, in particular, enhancement of lipid uptake by macrophages, leading to the formation of foam cells, endothelial dysfunction, platelet activation, increased activity of proteolysis processes, and stimulation of smooth muscle cell proliferation, fibrosis and systemic inflammation [29]. Insulin resistance and hyperinsulinemia are associated with other risk factors for CAD, such as impaired glucose tolerance, hypertriglyceridemia, a decrease in high-density lipoprotein cholesterol, and arterial hypertension. At the same time, isolated hyperinsulinemia has recently been considered an independent cardiovascular risk factor [33].

Patients with DM are more likely to have lesions of the left coronary artery and multivessel coronary arteries; they also have more lipid-rich and prone to rupture atherosclerotic plaques [27]. Factors that can determine worse survival prognosis of diabetic patients are diffuse atherosclerotic lesions of the small coronary arteries, the presence of many

associated risk factors, in particular kidney damage, and masking of clinical manifestations of ischemia in patients with DM. Consequently, even with the expansion of choice of medical and interventional treatment, the prognosis in patients with DM and already formed cardiovascular disease is significantly worse than in those without it.

Methods to diagnose CAD in patients with DM do not differ essentially from those used in patients without diabetes. Patients with DM often have symptoms atypical for ischemia, or painless course of CAD. The main non-invasive method of examination in the presence of chest pain is a stress test on a bicycle ergometer or treadmill; out-patient ECG monitoring may have additional significance in certain situations (for example, during nighttime or morning painful attacks). In patients with DM the initial probability of CAD is higher than in those without it. This, in turn, may affect the informative value of instrumental methods of CAD diagnostics. For example, with a positive test result in a patient with DM and atypical pain, the probability of CAD is higher than in a patient without diabetes. However, obtaining a negative result in many cases allows discontinuing the examination [33]. In doubtful situations, it is possible to use the most sensitive and highly specific methods for CAD diagnostics and myocardial viability assessment; in particular, single-photon emission computed tomography (SPECT) with thallium-201 or sestamibi, or stress echocardiography with exercise or dobutamine stress echocardiography.

The potential for reducing CAD associated mortality in patients with type 2 diabetes largely depends on how well risk factors are being corrected, including the use of modern antihypertensive and hypolipidemic agents [16]. The combination of CAD with DM is an additional strong reason for the secondary prevention of cardiovascular events with antiplatelet agents, beta-blockers, statins and renin-angiotensin system blockers [34]. Yet, the greatest hopes for the improvement of the course of CAD in patients with DM are associated with surgical or endovascular myocardial revascularization. Nevertheless, even in the case of percutaneous coronary interventions (PCI) or coronary artery bypass grafting (CABG), the survival prognosis of patients remains worse compared with those without diabetes [29].

Patients with DM who have undergone CABG in our clinic demonstrated hemodynamically significant stenoses of the right and circumflex coronary arteries more often. In these patients a greater number of postoperative complications were recorded (in particular, nephropathy and postoperative atrial fibrillation) compared with those without DM [1]. It is the presence of a diffuse lesion of the coronary bed that explains the high probability of restenosis caused by neointimal hyperplasia following PCI in the presence of DM. To a certain extent the frequency of restenosis was reduced due to the introduction of eluting stents (with a drug coating). However, even with the widespread use of this modern technology of endovascular treatment, DM is associated with poor clinical outcomes. DM appeared one of the most powerful predictors of stent thrombosis in a meta-analysis of 47 studied factors [12].

The information on the effect of DM on the outcomes of interventions is mainly based on the data from observational studies, registers, retrospective analyses of subgroups of patients with DM in large controlled studies of the effectiveness of revascularization, as well as studies comparing the effectiveness of PCI and CABG. Only in some studies, though, interventional treatment of CAD in patients with DM was given special consideration to.

Revascularization or medical treatment?

Generally, the indications for myocardial revascularization in patients with DM do not differ drastically from those in patients without it. In particular, a meta-analysis of nine randomized clinical trials involving 9904 patients with acute coronary syndrome did not reveal any dependence of the benefit of revascularization in the presence of DM [28]. At the same time the absolute risk reduction was larger with concomitant diabetes compared with patients without DM.

BARI 2D study included patients with type 2 diabetes and hemodynamically significant stenosis of at least one coronary artery. Half of the patients had stable angina, 10% had unstable angina, and 18% did not have angina or its equivalents. Patients with indisputable indications for revascularization (for example, multi-vessel lesion with LVD) were not included. In all cases, intensive drug therapy was carried out to correct hyperglycemia, dyslipidemia, hypertension, reduce angina, as well as modify lifestyle. On evaluating the coronary angiography data, the cardiologist chose the potentially most appropriate method of revascularization: PCI or CABG followed by randomization into groups of drug treatment or revascularization. Consequently, PCI was mainly performed in patients with a lesion of one or two vessels (two-thirds of the participants), while patients with a three-vascular lesion underwent CABG [15].

At the same time, among high-risk patients selected for CABG surgery, revascularization reduced the incidence of major cardiovascular events (22% versus 31%, $p = 0.01$) and non-fatal MI (7% versus 15%, $p < 0.01$). Thus, for the first time it was shown that in stable patients with coronary atherosclerosis, CABG operation reduced the risk of the future non-fatal MI, while in the low-risk patients selected for PCI, immediate revascularization did not improve outcomes. The results obtained in BARI 2D study agreed with the data from COURAGE study and other studies in which most patients did not have DM [4, 9].

It is worth noting that BARI 2D study did not include patients with initial coronary atherosclerosis (without convincing indications for revascularization) on the one hand, and, on the other hand, patients with severe symptoms or pronounced stenotic coronary arteriosclerosis (therefore, absolute indications for revascularization). Thus, the results of this study are most relevant to patients with moderate or stable symptoms and /or moderately pronounced atherosclerotic changes in the coronary bed. Moreover, approximately 40% of patients randomized for drug treatment, during 5 years of follow-up, were actually revascularized due

to the progression of angina, acute coronary syndrome or severe myocardial ischemia. Only a third of the patients in the PCI group had eluted stents implanted; in most cases, conventional metal stents were used [31].

Surgical revascularization of percutaneous intervention?

BARI 2D study did not attempt to compare the effectiveness of CABG and PCI, because the groups of patients who used various methods of revascularization differed significantly in angiographic characteristics. The first attempt to answer this question was made in the original BARI study, in which 1.829 patients (mostly with unstable angina and multivessel CAD) were randomized to CABG or PCI groups (at that time – balloon angioplasty) [6]. No differences in death rates or MI were revealed in the compared groups in this study. However, in patients with concomitant DM such differences were found. In particular, the mortality of patients with diabetes and CAD with lesions of one or two vessels of coronary arteries during 5 years of the follow-up was 35% in the PCI group and 19% after CABG [7]. CABG was generally associated with significantly better survival over five (80% vs. 67%) and ten years (respectively, 58% vs. 46%, p = 0.025) compared with balloon angioplasty [7,8]. Undoubtedly, the use of CABG was determined by the use of arterial mammary shunts in 81% of cases. The results of BARI study and some retrospective analyses showed that the presence of DM can affect not only the outcomes of revascularization interventions, but also the choice between CABG and PCI.

The results of retrospective analyses also testified to the possible advantages of CABG in comparison with PCI in terms of the effect on mortality within 5 years in patients with CAD and DM [10]. The use of PCI with stent implantation is often associated in patients with DM with the formation of restenosis. In CARDia study involving 510 patients with DM during a one year observation, the use of stents was accompanied by a greater incidence of restenosis and repeated revascularization compared with CABG [24]. Despite the obvious limitations of retrospective analyses, the existing evidence base testified in favor of performing CABG (and not PCI) as a more reliable method for myocardial revascularization in patients with three-vascular lesion or stenosis of the left coronary artery. In particular, in the subgroup of 452 patients with DM and multivessel lesions of coronary arteries in the SYNTAX study with a 5-year follow-up, no differences between the CABG and PCI groups were found in terms of incidence of "large" cardiovascular complications; however, the need to repeat revascularization was more common in the PCI group [27].

It can be assumed that concomitant DM does not affect the patency of arterial (mammary) shunts, in contrast to venous shunts, where rapid progression of atherosclerotic lesions was observed [32]. Moreover, a lower incidence of non-fatal MI in BARI-2D study [15] revealed that bypass surgery, unlike endovascular interventions, helps to prevent the progression of coronary artery lesions and / or rupture of atherosclerotic plaque in proximal regions [2]. However, despite these arguments, the percentage of PCI in patients with diabetes continued to increase. Obviously, this paradoxical phenomenon was brought about by the advancement of the eluting stents technology, as well as a frequent instant implementation of angiography and PCI, without a proper discussion and giving the patient full information to make a reasonable decision [21].

A FREEDOM study conducted by the United States National Heart, Lung, and Blood Institute randomized patients with DM and multivessel CAD into PCI groups with implantation of eluting stents (such as sirolimus or paclitaxel) or CABG surgery (2.9 shunt per patient, 94.4 % of cases used arterial shunts) [14]. The study included patients with hemodynamically significant stenosis (more than 70%) of two or more coronary arteries, but without stenosis of the left coronary artery trunk. Only about a quarter of patients had previously suffered MI, in the overwhelming majority of cases, the left ventricular (LV) ejection fraction (EF) was preserved (i.e., there were no convincing indications for surgical revascularization). The observation lasted at least two years, on average – 3.8 years. All patients were prescribed modern medical therapy to control low-density lipoprotein cholesterol level (target value less than 70 mg%), blood pressure (less than 130/80 mm Hg) and glycosylated hemoglobin (less than 7%). The primary endpoint was the sum of death outcomes, non-fatal MI, or stroke.

A total of 1900 patients were included (mean age 63.1 ± 9.1 years, 29% – women), with 83% having a three-vessel lesion. The frequency of reaching the primary endpoint after CABG was 30% less: in the CABG group it made up 18.7%, after PCI – 26.6% (P = 0.005). The effect of CABG was achieved by reducing MI frequency (10.9% versus 16.3%, P <0.001) and death from any cause (6.0% versus 13.9%, P = 0.049). Yet, strokes developed more often in the CABG group than after stenting (5.2% vs. 2.4%, P = 0.03), mainly occurring in the early postoperative period.

FREEDOM study was the most significant stage in the discussion on the choice of the optimal method of revascularization in patients with DM. CABG surgery provided better results than PCI due to a decrease in the frequency of end points [14]. A little later, similar results were obtained in VACARDS study [23], where a comparison of CABG and PCI was also conducted in patients with DM. Due to the early termination of the study, a total of only 198 patients were randomized. Deaths, or non-fatal MI cases occurred in 18.4% of patients in the CABG group and 25.9% in the PCI group (P <0.05).

A certain limitation to the use of data from FREEDOM study was a relative "mildness" of patients in terms of the frequency of previous MI, severity of LV dysfunction, and clinical manifestations of heart failure. To a certain extent, this limitation was overcome in a recently published analysis of outcomes in 11.518 patients with LV ischemic dysfunction, conducted as a part of a register in the Canadian province of Alberta [19]. The difference between CABG and PCI in terms of the overall influence on mortality rate was

evident in patients with DM (10.7 and 15.7% respectively, P = 0.0001) and was absent in patients without it (8.4 and 8.7%, unreliable differences). Among 2387 patients with DM, "matches" with similar clinical characteristics were specifically selected who underwent CABG or PCI [26]. The frequency of "major" cardiovascular complications, as well as death from all causes was greater in the cohort of patients with EF less than 35% and 35-49%. The incidence of stroke did not differ in the CABG and PCI groups, regardless of EF indicator. PCI was associated with a greater incidence of MI in the cohort of patients with low EF, whereas repeated revascularization was more often recorded after PCI in cohorts with EF less than 35% and 35-49%. The authors concluded that CABG is more expedient in terms of the effect on the risk of cardiovascular complications and the survival of patients with DM and LVD.

Favorable effects of CABG compared with PCI were also confirmed in a recently published analysis of the results of treatment of patients after acute coronary syndrome in real clinical practice [30]. In a systematic analysis of the results of interventional treatment of 13114 patients with type 2 diabetes cases of major adverse events, repeated revascularization and MI were observed more often in the PCI group than after CABG [11]. It should be noted that groups of patients with and without DM do not differ in the incidence of early stent thrombosis; at the same time, DM is associated with a greater incidence of late thrombosis compared with patients without diabetes [38].

Obviously, the type of stents used can have a significant impact on the results of PCI in patients with DM. In particular, in the New York State register, the use of everolimus-type stents was associated with a similar risk of death, a higher risk of MI (in case of incomplete revascularization) and repeated revascularization, and a lower risk of stroke compared to isolated CABG [3]. In general, the existing evidence base testifies in favor of CABG, rather than PCI, in patients with DM and multivessel CAD [22, 36]. Basing on the available data, patients with DM should be informed about the benefits of CABG for improvement of survival prior to coronary angiography. If there is a concomitant pathology which causes an increased risk of surgery, an individualized decision regarding the type of revascularization should be made by a multidisciplinary team of specialists based on a comparison of the advantages and disadvantages of various methods [20, 27].

Consensus recommendations and contradicting aspects of patient management

Feasibility of sensitive heart stress-visualization methods to detect MI has not yet been proved in asymptomatic patients with DM who have no past history of coronary events [17, 37]. In such cases optimal drug therapy aimed at the primary prevention of cardiovascular events remains the method of choice [34]. Detection of LV dysfunction, high-risk criteria according to the exercise test, as well as cases of insufficient effectiveness of drug therapy form the basis for coronary angiography in patients with anginal attacks (fig. 1). Basing on the results of BARI 2D study, patients with one or two vascular lesions of coronary arteries associated with stable manifestations of MI can continue conservative treatment, while in case of destabilization of ischemia or emergence of disabling symptoms (angina at low loads), surgical

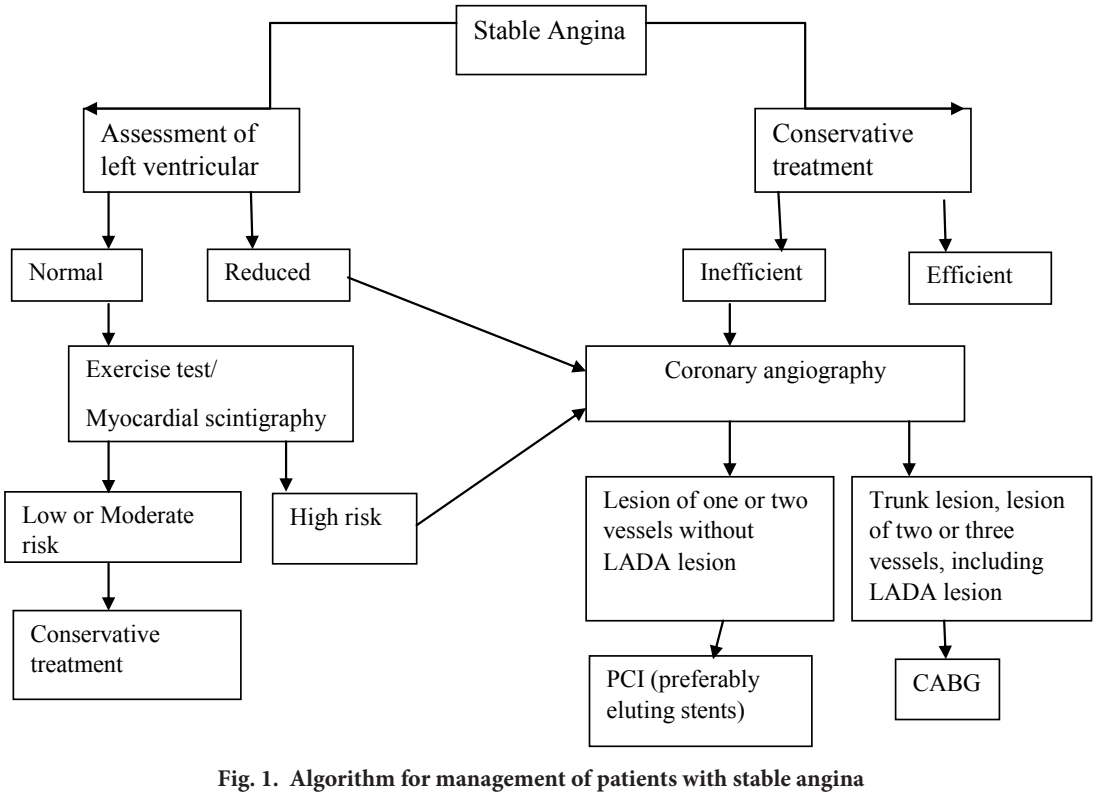

Fig. 1. Algorithm for management of patients with stable angina with / without concomitant DM. LADA – Left Anterior Descending Artery.

myocardial revascularization or PCI should be considered. Finally, in patients with multivessel CAD in the presence of DM, surgical myocardial revascularization has advantages over PCI, which has been primarily, indicated by the results of BARI 2D and FREEDOM studies [14, 15].

In any case, the decision on the treatment strategy and the choice of revascularization method in particular should be made taking into account the patient's opinion, not only the information about the anatomy of coronary bed and the point of view of the attending physicians. For many patients, objective, balanced information about the expected length of stay in the clinic, recovery time, quality of life, risk of stroke, as well as the possible need for re-revascularization is crucial to make a final decision (tab. 1).

Table 1

The advantages and disadvantages of various treatment strategies for patients with moderate CAD with concomitant DM (adapted from [31])

Compared criteria	Drug therapy	PCI	CABG
Symptom control	Moderate	Good	Good
Primary or repeated revascularization over 5 years	42% (BARI 2D study)	30%	9%
Repeated revascularization over 1 year	----------	12-20%	2-6%
Mortality risk	Reference	Similar	Similar
MI risk	Reference	Similar	Lower
Non-fatal stroke	Not known	0.4-0.9%	2.5-2.8%
Hospital stay and rehabilitation	Reference	Days	Weeks and months
Quality of life in a year	Reference	Similar	Better
Extent of revascularization	----------	Moderate	Good
Cost-effect ratio: Over 4 years Over life span	 Reference Reference	 Worse Worse	 Worse Better

In the European recommendations for myocardial revascularization in 2018, a special section is devoted to the features of treatment of patients with DM [27]. First of all, this document clearly states that anatomical indications for revascularization in patients with DM are the same as in patients without it. In stable patients with CAD multivessel coronary artery disease and / or lesion of the left coronary artery is an argument in favor of CABG, rather than PCI.

Management problems and worse outcomes in patients with DM who previously underwent surgical myocardial revascularization or endovascular interventions are due to the progressive nature of atherosclerotic vascular lesions, severe endothelial dysfunction, platelet activation and blood clotting disorders [34]. These pathophysiological features determine the priorities of additional drug therapy after revascularization, with particular attention to the treatment of associated diseases and correction of risk factors [16]. However, there has been no convincing data on the effect of glycemia on the frequency of restenosis after PCI or the patency of shunts after CABG surgery so far. Special attention is given to the assessment of renal function before angiography in patients receiving metformin, with the suspension of the drug for 48 hours before the study in the presence of renal failure, and in other patients – in case of deterioration in renal function after angiography [27].

Undoubtedly, the use of renin-angiotensin system blockers, beta-blockers, antiplatelet agents after revascularization in patients with DM seems expedient. Aggressive control of the lipid profile by drugs from statin group, whose dose in patients with a very high level of risk (including patients with CAD and DM) is determined depending on the achievement of the target level of low-density lipoprotein less than 1.8 mmol/l also seems reasonable [13, 35].

European Survey Study on Coronary Revascularization assessed the potential impact of the presence of DM on the physician's choice of drug treatment or revascularization. It has been found that DM was not included in the list of the main factors determining the physician's decision in case of a stable course of CAD [5]. It should be noted that when choosing the revascularization method for patients with DM high probability of restenosis after PCI should be taken into account [34].

Conclusions

The prevalence and rapid progression of atherosclerotic lesions is a defining feature of the course of CAD in patients with DM. Worse survival prognosis in case of concomitant DM is associated with systemic atherosclerosis, presence of a number of concomitant risk factors, as well as masked clinical manifestations of ischemia and MI common for patients with DM. The combination of CAD and DM once again proves the benefit of the long-term use of drugs for the secondary prevention of cardiovascular events. From the standpoint of evidence-based medicine the optimal method of revascularization in CAD patients with multivessel coronary artery disease and concomitant DM is CABG surgery. In FREEDOM study surgical myocardial revascularization reduced the number of endpoints compared to PCI. In the case of PCI it is advisable that eluting stents reducing the likelihood of restenosis and the need for repeated revascularization be used.

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Fundamental aspects of cardiovascular regulation in predisposition to atrial fibrillation

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Abstract

Background: Atrial fibrillation is the most common sustained arrhythmia in cardiology. The structural factors leading to atrial fibrillation are well known, but there should be also regarded the functional factors. In 2014, the Task Force published guidelines for atrial fibrillation describing the importance of the vegetative nervous system in creating predisposition to atrial fibrillation although it describes that the mechanism is not completely clear. Furthermore, it is important to understand this mechanism, regarding the increasing number of patients affected by atrial fibrillation without any structural heart diseases. The aim of this work is to understand the physiological background of the predisposition to the appearance and recurrence of atrial fibrillation regarding the role of neural regulatory systems of the heart, especially when no structural heart diseases are present. Therefore, the following is a fundamental analysis of the neural regulation of heart rhythm, including the vegetative nervous system at its medullar and central levels and also the cerebral cortex input in heart regulation.

Conclusions: The predisposition to atrial fibrillation regarding the neural regulatory systems of the heart can be pinpointed to three key factors: 1. Central over-activity; 2. Sympathetic efferent overflow towards the heart in rest state; 3. Parasympathetic exhaustion and break-down of the parasymphatetic protective function.

Key words: atrial fibrillation, neural heart rhythm regulation, central over-activity.

Introduction

Atrial fibrillation is the most common sustained arrhythmia in cardiology, which affects about six million people in the European Union [1]. The "Worldwide Epidemiology of Atrial Fibrillation: Global Burden of Disease" study in 2013 revealed, that 33.5 million people around the world have atrial fibrillation, which results in around 0.5 percent of the world's population. Though the influence of structural factors of the heart on the arrhythmogenesis of atrial fibrillation is well known [2], it is important to understand the conditions for occurrence of atrial fibrillation in patients without any eminent structural changes in the heart [3]. Thus, the state of the regulatory mechanisms of the heart should also be taken into consideration [4]. The vegetative nervous system plays a key role in the neural regulation of the heart [5]. In 2014, the Task Force published guidelines for atrial fibrillation describing the importance of the vegetative nervous system in creating predisposition to atrial fibrillation [6].

The aim is to understand the physiological background of this predisposition and in addition to analyze the role of the cerebral cortex input on the heart regulation in creating the predisposition to atrial fibrillation. Therefore, the following is a fundamental analysis of the neural regulation of heart rhythm.

The neural regulation of the heart

Regarding the neural regulation of the heart it is predominantly regulated by the vegetative nervous system at the level of the medulla oblongata [7]. There is a pressor center (cardio-accelerator center) and a depressor center (cardio-inhibitory center). The first is driven by nervi sympatici cordis and the second is driven by nervus vagus [7]. At this level, the vegetative regulation of the heart is non-stop and also it is modulated by breathing, by the central part of the vegetative nervous system and by the cerebral cortex impulses [7, 8, 25]. Regarding the central efferent impulses, these are impulses that reach the heart via passing through the pressor center of the hemodynamic center in the medulla oblongata [9]. These impulses fulfill their action on the heart by utilizing the sympathetic fibers [9]. It is the first key moment in understanding the mechanism of neural predisposition to atrial fibrillation [10, 11]. This means that the primary reaction of the heart on all central efferent impulses is sympathicotonic [9, 12]. The secondary reaction is the vagal counteractivation at medullar level and the parasympathetic counteractivation at the central level [12, 5]. This reaction is important and will be explained further in detail below.

The protective function of parasympathetic couneractivation at medullar level and the central input

Usually the parasympathetic part of the vegetative nervous system is regarded as the one which is responsible for vasodilation and for the known effects on the heart (negative bathmotropic, dromotropic, inotropic, tonotropic, and chronotropic) [5, 7], however, there is another very important function. It functions also to protect the heart from an increase in central efferent impulsatory activity [12]. It is imperative to analyze in detail what occurs during an increase in impulsatory activity and what kind of protective mechanisms the parasympathetic vegetative nervous system has. The pressor center always has an inhibitory influence on the depressor center [5, 7]. This is also important in order to protect the heart when the central regulation of the heart increases during rest state [12]. It is a necessary protective mechanism because when the central regulation increases, the intensity of the sympathetic efferent impulses to the heart in rest state also increases [9, 12]. As it was already mentioned above it occurs due to the fact that the central regulatory input exerts its influence via the sympathetic fibers passing through the pressor center [9]. When the regulation of the heart works physiologically, the sympathetic activation driven by the central regulatory impulses is compensated or counterbalanced by the parasympathetic part of the vegetative nervous system. At the medullary level, this means that the depressor center acts as a filter membrane [12]. By its permanent inhibitory action on the pressor center, the depressor center filters out a part of the central efferent regulatory impulses reaching the pressor center (fig. 1). In other words, not all efferent impulses of the central heart regulation reach the heart. This is the protective function of the parasympathetic part of the vegetative nervous system [13].

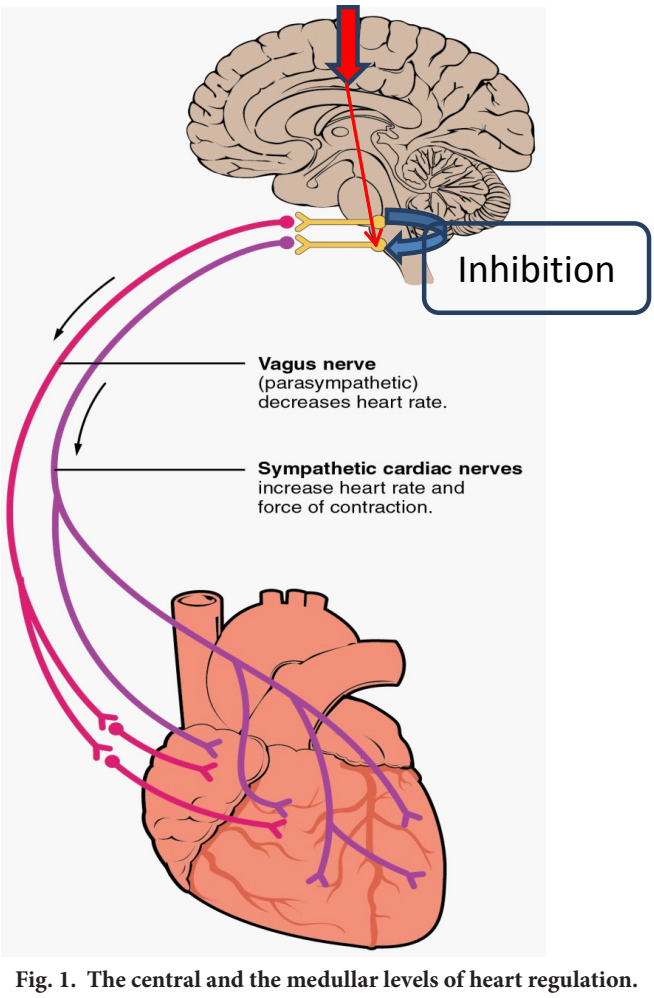

Fig. 1. The central and the medullar levels of heart regulation.

The central level includes the influence of cerebral cortex, lymbic system, hypothalamus, reticular formation. It fulfills its action on the heart via the pressor center. So it acts always at the heart sympathicotonic. In order to avoid a sympathetic overflow of the heart driven by the central efferent impulses, there is a protective mechanism at the medullary level: the depressor center of the hemodynamic center always has an inhibitory influence on the pressor center. This means that the depressor center acts as a filter membrane, so it filters out a part of the central efferent regulatory impulses reaching the pressor center before they further stream to the heart. This is the protective function of the parasympathetic part of the vegetative nervous system.

Pathological counterbalance at the medullar level

Described above is an overview of how the protective parasympathetic counterbalance works at the medullary level. When the parasympathetic vegetative nervous system does not function properly, the following occurs: the central modulation increases, which in turn stimulates its sympathetic action on the heart, but the parasympathetic inhibitory counteractivation on the pressor center is insufficient [14]. This means that the parasympathetic protective filtering of the central efferent impulses is not capable of inhibiting enough of these impulses [13]. Under such conditions, more central efferent impulses stream towards the heart than what is physiologically normal during the rest state [9, 15, 16]. This leads to an increase in sympathetic activity on the heart during this time. As the parasympathetic barrier becomes less active and effective, a large amount of efferent central impulses reach the heart [8]. As a consequence, this results in higher sympathicotony of the heart at rest [8, 17]. The conditions which provoke a permanent chronic decrease in functional activity of the parasympathetic part of the vegetative nervous system can lead to a permanent overflow of the heart in its rest state [12]. This is because under such conditions the efferent modulative impulses are hardly inhibited in the hemodynamic center by its depressive center. As a result, efferent modulatory impulses of high intensity pass through the pressor center to the heart. This is the sympathetic overflow of the heart during the rest state [12]. Such a state itself leads to an ongoing exhaustion of the parasympathetic part of the nervous system. The exhaustion leads to an ongoing decrease in the ability to inhibit the modulative impulses [14, 17, 11]. Thus, the amount that is streaming to the heart continues to in-

crease. This is *circulus vitiosus.* Under these conditions, the modulative regulation of the heart becomes dominant versus the medullary one [18]. This is a pathological situation which creates predisposition for the appearance or recurrence of atrial fibrillation [12, 18].

The central regulation of the heart

Under normal conditions, the heart is regulated by the medullary level and modulated by the central level [5, 7, 18]. This explains why heart regulation occurs autonomously and why the central level has a modulative function. Autonomic heart regulation is ensured by the baroreflex. Normally under central regulation is understood the hypothalamus which represents the highest vegetative center [7]. In this article the central regulation is regarded in a large sense. It includes cerebral cortex, motor cortex, lymbic system, hypothalamus, reticular formation [7, 10, 13]. It should be noted that both groups of pathways were taken into account: the group of pathways from the reticular formation to the hemodynamic center and the group of pathways via hypothalamus to the hemodynamic center [19, 20]. So, the cerebral cortex impulses pass through cardiovascular control center in medulla oblongata. The central modulative influence of the heart also occurs in a calm state because the heart always has to supply effectively with blood every action and state of an organism. Even in a calm state, the heart receives many central impulses necessary to maintain the muscle tonus, the position of the body in space, as well the basic vital functions [7, 20]. It is essential that all this information is received by the heart in order to properly respond by regulating frequency and blood pressure.

Of course the question arises whether there are conditions that can increase the central regulation of the heart in calm state. There are both physiological and pathological conditions that can increase the central regulation of the heart. An example of physiological condition is observed during increased mental activity or during psychoemotional stress [17, 26]. This increase of central regulation is transitory, occurring only during stress or mental activity [17, 18]. After its action stops, the physiological counter-balance switches. As a consequence, the central regulation decreases until its physiological level, so no sympathetic overactivity occurs [17]. A pathological increase of the central heart regulation is characterized by a permanent increase, even in a calm state when the person is relaxed [12, 17, 18, 22]. This leads to a permanent sympathetic overactivity of the heart and can result in permanent positive bathmotropy [26]. This is a dangerous state because it creates favorable conditions to atrial fibrillation [11, 12, 23]. Such a mechanism could explain the appearance of atrial fibrillation and the recurrence of atrial fibrillation in patients without any structural heart diseases identified by an echocardiogram [6, 11, 21, 24].

By advance analysis of heart rate variability, the physiological and pathological central heart modulation can be detected [12, 24, 25, 27]. This can also be used to identify the level of sympaticotony in a calm state as well as the level of activity of the parasympathetic part of the vegetative nervous system [16, 25]. Our future aim is to research possible factors which can lead to an increase in central heart modulation during a rest state, when the person is relaxed.

Conclusions

1. The predisposition towards atrial fibrillation appearance and recurrence can be pinpointed to three key factors:

a) Central overactivity.

b) Sympathetic efferent overflow towards the heart in rest state.

c) Parasympathetic exhaustion and break-down of the parasymphatetic protective function.

2. The parasympathetic part of the vegetative nervous system has not only the well-known functions of vasodilation and the known effects on the heart (negative bathmotropic, dromotropic, inotropic, tonotropic, and chronotropic), but also the very important function of protecting the heart from an increase in central efferent impulsatory activity.

3. An increase of central modulative influence on the heart in rest state could be regarded as a possible mechanism in creation of the neural predisposition for atrial fibrillation.

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Supportive principles in the pharmacological management of the patients with epilepsy

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Abstract

Background: Pharmacological management of patients with epilepsy is still a very challenging approach for the best outcome of these patients. When considering the appropriate treatment choice for patients it is necessary to take into account several factors that can influence the effectiveness and quality of life. Cancelling or changing treatment suddenly can lead to uncontrolled seizures. After a short period without seizures, many patients are tempted to abandon treatment. Cessation of treatment can be discussed after a seizure-free period for at least two years. Treatment should be discontinued gradually by reducing the dosage and constant supervision of the physician. This paper analyses briefly the general pharmacological and treatment methods in several forms of adult epilepsy.

Conclusions: Management of epilepsy means more than observing the medication prescribed by the specialist. It is also important for the patient to maintain his general health status, monitor the symptoms of epilepsy and response to treatment and take care of his safety. Involvement in the management of one's own affection can help the patient to control his condition and to continue his routine in usual manner. The objective of antiepileptic treatment is to reduce epileptic seizures to zero without intolerable side effects. New treatments should focus not only on reducing the frequency and intensity of seizures but also improving the quality of life of patients.

Key words: patient, epilepsy, therapy and dynamics.

Introduction

The analysis of the specialized literature reveals that many issues regarding differential treatment of epilepsy require subsequent clarification. As far as we are concerned, we have designed and developed therapeutic recommendations, in our opinion, effective, supporting the results of treating epilepsy in its various stages, from premonition to status variants. In this context, the main element in the choice of preparations, besides the trivial clinical signs, was the use of sub-curative monitoring data, including repeated EEG examinations, which fixed the subjective response of patients. Choosing the best possible medicine or an optimal combination of medicines is sometimes difficult. The perfect antiepileptic should be long, nonsedative, well tolerated, very active in various types of convulsive and with non-harmful effects on vital organs and functions. In addition, it must be effective in various forms of active epilepsy and in treating underlying epileptic seizures and capable of restoring the electroencephalogram between seizures to its normal form [5; 9; 10; 18; 23; 24; 27; 31; 38; 40; 41; 43].

It is still debatable whether such a drug will ever be discovered, and especially one that will control all types of epilepsy. The thorough study of pharmacological properties allows us to appreciate which of the existing antiepileptics will meet the current requirements of our patients under study. Due to the fact that patients differ considerably after clinical response to known anticonvulsants and the possibilities of treatment with associated drugs are insufficiently and superficially researched, testing of more efficient substances including new combinations continues. Due to the modern medication, which benefits from a wide and sufficiently efficient range of specific drugs, a large proportion of the recurrent and the disabling sequelae of the disease can be prevented. The adverse effects of drugs are low, so many of the past patients who have been labelled for life by this suffering can now live a productive life. The actual ability to control this disease effectively prevents more of its severe consequences [12; 13; 15; 22; 29; 46; 50].

General principles of pharmacotherapy of epilepsies

In the treatment of psychiatric disorders of our patients with epilepsy we have taken into account the following principles:

Appropriate selection of the remedy, its dosing, routes of administration and possible side effects. And we took into account the following:

1. The syndrome of psychic state – the gradual expression of the disorders, the relationship between productive and negative alterations and the type of impairment of psychic processes.

2. The dynamic characteristics of the psychic state – the duration of the disturbances, the changes in the presence of paroxysmal manifestations.

3. The somatic and neurological condition of the patient with epilepsy. This parameter is important in the context

of the evidence of side effects of favorable and unfavorable preparations. Somatic mood dictates and the route of administration of drugs: parenteral in gastrointestinal disorders, endonasal or transorbital (by electrophoresis) when parenteral administration is not preferred.

Individual features of the patient with epilepsy (age, weight, response to anticonvulsant therapy and others) are also considered. It is often forgotten that lower doses are indicated for children and older people as the exchange of substances in them is slow and standard dose treatment leads to accumulation of preparations and adverse effects [6; 7; 14; 19].

We recommend the gradual increase of the doses, with the preference of the minimal effective doses of the drugs. All the above-described drugs are initially indicated at minimal doses, then the dose gradually increases until the first positive effects are displayed, the subsequent increase of the doses is made after a certain period of time to stabilize the positive effect.

Complex treatment – it is necessary to prescribe unimoment of anticonvulsant remedies from different classes and groups in combination with non-medication methods. Polipharmacologic treatment has certain priorities in comparison with monotherapy because it addresses different links of the pathological process. It is important to avoid the multidimensional effects of many drugs, the doubling of the mechanisms of action and the predilection of some and the same psychological processes.

Continuous therapy. The treatment of productive disorders is done until their complete jugulation (sometimes with the purpose of preventing relapse and longer), of the deficient ones by alternating the cures, with gradual modifications [28; 30; 34; 39; 42].

Principles of medication of psychosomatic syndromes in epilepsy

Criteria for the effectiveness of psychotropic remedies administered in epilepsy are those of improving the knowledge and behavioral processes. More differentiated treatment is based on syndrome of mental disorders.

1. Deficient disorder (transient dementia, mental-mental diminution, etc.) The treatment is continuously practiced, alternating the belts. It is rational to indicate the preparations of different subgroups. The following criteria are taken into consideration when drawing up the treatment scheme:

a) Main mechanism of action: nootrop, general metabolism, cerebrovascular or actoprotector;

b) Predominant action on mediating processes: GABA (piracetam, fenibut, gamma-aminobutyric acid); cholinergic (gliatiline); dopaminergic (nakom); and combined (meclofenoxate, glycine, glutamic acid);

c) With predominant action on the function of the encephalic structures: the cerebral and subcortical (nakom), on the left hemisphere (gliatilline); on the right hemisphere (cortexil);

d) With action on psychomotor activity: major stimulation (piracetam, nakom vinpocetine), mean enhancement (aminalone, gamma-aminobutyric acid, cerebrolizine, nicergoline, tanakan), diminishment (fenibut, glycine, cinarizine);

e) Route of administration: parenteral, internal, endonasal, transorbital (by electrophoresis), mixed. Duration of treatment: from 7 days to 4 months (nakom, fenibut). On the basis of this therapy it is also possible to indicate prophylactic doses of anticonvulsants.

2. For different types of excitation (chaotic, twilight, delusional, manic, psychopathic, etc.) the support treatment are the sedative neuroleptics. Major tranquilizers, barbiturates and other anticonvulsants, may also be indicated sedative antidepressants.

3. Hallucinatory delusions. More rational are antipsychotic neuroleptics. In the case of neuroleptic syndrome with caution are added corrective remedies. That adjuvant preparations use daytime tranquilizers, in depressive or anxious states are used antidepressants.

4. Emotional productive disruptions. In the states of excitation are indicated predominantly sedative neuroleptics and tranquilizers, antidepressants – in depression, tranquilizers and antiepileptics – in dysphoria, in anxiety states – neuroleptics and tranquilizers.

5. Productive districts nearby. Psychoparticular depressions are typically treated with "inor" euroleptics, preferably "behavioral correctors" or low doses of risperidone and tranquilizers; in neurotic manifestations (asthenia, obsessions, hysteria, hypocondria) are used tranquilizers and low doses of antidepressants [1; 2; 3; 4; 8; 11; 25; 26].

Complex epilepsy medication programs

Over the past decades has been made a remarkable progress in assisting epilepsies. In most cases, however, treatment of epilepsy is symptomatic since it is known that only the treatment can prevent recurrences that often result from the action of noxes chronic epileptogenes when it becomes necessary to correct some metabolic disorders in the brain lesions. Etiological is also the specific anti-infectious treatment in the case of epileptic-generating neuroinfections. The antiepileptic treatment performed in newly examined patients aims to control crises and ensure a social life that is close to normal. This treatment was based on the use of last-minute anticonvulsants (carbamazepine, sodium valproate, phenobarbital, diazepam, apaurine, phenytoin), which in most cases have more or less pronounced side effects. They were adapted by us to the form of epilepsy, evolutionary gravity and individual tolerance. The general principles of treatment of epilepsy were in selecting the optimal preparation depending on the type of epileptic seizures, selecting the optimal dose, usually minimal, which allows plenary control of epileptic seizures. The treatment prescribed by us must be lengthy and uninterrupted. However, the optimal duration of specific treatment is purely individual for each patient with epilepsy, as opposed to the complete cessation of crises, which ranged from 6 months

to 5 years. Sometimes, depending on the type of epileptic seizures, the medication was indicated lifelong. Interruption of treatment was performed gradually for 3-8 months with clinical and electroencephalographic monitoring. We have also been very careful in avoiding the factors that trigger epileptic and cautious seizures in observing the optimal life regime [17; 20; 21; 32; 33; 44].

Antiepileptic treatment divides into:

- Emergency (juggling of crises);
- Long-term treatment (crisis prevention);
- Etiological treatment.

The medical treatment of epilepsies should represent as far as possible a monotherapy. The drug selection is based on the type of epilepsy. In generalized seizures and partial seizures that have been reported in our patients, we examined the use of new carbamazepine, sodium valproate, phenobarbital. At patients with petit mal absence, the elective drug we administered was sodium valproate. In the category of patients with epileptic encephalopathy in the treatment we have prescribed sodium valproate and corticoids that have given good results. Some treatment failures, marked by the repeating crises, originally focused on the idea of whether it was correctly applied. In this regard, it is useful to determine the serum concentration of the drug. If the serum level of the drug is satisfactory, but the crises are repeated, the change in treatment by administering or associating the second antiepileptic should be called into question. Continuing treatment is essential, and the sudden abolition of specific medication increases the risk of the crisis. The basic principle that we recommend following our examinations of patients with epilepsy over several years for the treatment of epileptic seizures is one of the highest urgency, which has two essential components, namely:

- Anticonvulsants for stopping epileptic seizure as soon as possible.
- Secondary measures, but of vital importance: Maintenance in resuscitation and intensive care units. In the case of the 259 patients with epilepsy, we found that (58.7%) received stable care, (41.3%) were outpatients. They were hospitalized repeatedly – (19%) of patients. When using an anticonvulsant in maximum therapeutic doses with the occurrence of adverse events, it is reasonable to reduce the dose slightly and to associate other anticonvulsants with the summary of the effects of the preparations. Thus, their potential became known in various combinations, therefore in their prescription requires a thorough dynamic control of the functional status of the kidneys, liver and peripheral blood.

The basic principle of the therapy is complex treatment, based on a strict evidence of the structural features of paroxysms and the clinical evolution of the disorder in both in onset and in different stages. The analysis of the therapeutic measures has demonstrated that the most effective in benign sleep epilepsy are those with minimal hypnotic effect and with tropism predilection for subcortical formations (finlepsin, diphenin, chloracon, hexamedine, sodium tetraborate, elenium).

In patients with epilepsy, whose seizures throughout the full illness are produced in sleep with the hours of peak during the first half of the night; the drug is usually given half an hour before bedtime.

Thus, it has fallen definitively the need to administer in three divided doses of anti-epileptic drugs in patients with epilepsy, even more so that the effect was obtained with a single dose. Therefore, the medication possesses the etiopathogenetic character and is composed according to indications:

- Anticonvulsant;
- Antiphlogistic;
- Dehydration treatment;
- Desensitizing product;
- Fortifying;
- Neuroleptic;
- Nootropic;
- Resorptive;
- Roborant.

The application of treatment of the patient with epilepsy is carried out in three steps:

1) Preferential drug choice;

2) Obtaining therapeutic remission, strengthening, prevention of worsening and patient behavior without seizures for a period of 3-5 years;

3) The suspension of treatment according to the lack of clinical manifestations and EEG of epilepsy [18; 26; 49].

Treatment in ambulatory conditions

Controlling seizures caused by epilepsy requires the daily schedule of treatment recommended by the psychiatrist.

Antiepileptic drugs should be administered according to the prescription without any deviation from it. Disregarding the treatment scheme is one of the main reasons for the inefficiency of control over the seizures.

Antiepileptic drugs are effective only when the level of concentration of drugs in serum is kept constant. The doctor is in view of this fact, recommending a specific scheme for each drug (a specific dosage, a certain scheme of administration, etc.). Not administering a dose compromises the whole treatment. The same rule applies and in the case of patients with epilepsy who follow a cetogenic diet (the complete exclusion of fats and protein) recommended by the physician to improve the control of the drug-resistant epilepsy. Cetogenic diet can be difficult to follow it, but it is mandatory to respect it.

In addition to medical treatment, the physician recommends identifying and avoiding situations, conditions which determine the occurrence of seizures, such as [16; 17; 25; 28; 47]:

- Drinking alcoholic beverages;
- Drinking beverages or foods that contain caffeine;
- Drug use;
- Emotional stress;
- Excessive sun exposure;
- Lack of adequate sleep;

- Prolonged use of computers;
- Reducing the number of daily meals;
- Watching TV for several hours.

Epilepsy medication

The drugs used to prevent seizures are called *antiepileptics*. The goal consists in finding effective anti-epileptic medication which may lead to the least adverse effects. The efficiency of antiepileptics in the prevention of seizures is 60–70%. Although many people suffer the side effects of these drugs, this is the best way to prevent convulsions. The benefits usually outweigh the shortcomings of these medicines.

There are many types of anti-epileptic drugs (also called anticonvulsants), but not all of them treat the same type of seizures. The first step in the choice of anticonvulsants is the correct diagnosis of the type of epilepsy and seizures that the patient with epilepsy produces. It may take more time to find the best combination of drugs, by adjusting the doses, by testing multiple drugs. The goal is to prevent seizures with adverse effects as low as possible.

Once it has been found the most effective treatment, it is very important for the patient to follow the physician's recommendations. Monotherapy (single drug use) is strongly recommended as the treatment of first intention.

Monotherapy causes fewer side effects and does not have the risk of interacting with other drugs. Also, the chances of forgetting or mistaking the time of drug administration are much lower in the monotherapy compared to a multi-drug treatment.

When monotherapy is ineffective, adding a second medication can improve seizure control. Also, in the case of patients with multiple types of seizures, it needs more than a single drug [2; 3; 4; 17; 28; 45].

Interaction with other drugs. Most antiepileptic remedies do not induce side effects, but there are exceptions. Therefore, always carefully read the information in the leaflet. Some medications, for example those based on medicinal plants are not accompanied by adequate information. In such situations, consult with your doctor.

Timetable schedule. Since the effect of most antiepileptic drugs is of short duration, they must be administered daily at the same time, according to doctor's recommendations. Administration of medicines before or after meals does not affect the efficiency.

It is very important that the antiepileptic drug to be taken regularly to maintain its blood concentration at a relatively constant level. If for any reasons you have not taken the drug according to the prescriptions, consult with your doctor. However, there is no standard solution for such situations because other factors are involved, such as the type of anti-epileptic drug, the dosage and any other drug that you take that may affect the disease or the treatment that it is followed.

Pregnancy and breast-feeding. If you are planning to have a baby, consult with your doctor to ensure that your health allows this, that the disease is under control and that your future baby will be born healthy. Ideally, you should consult with a medical specialist and with a gynecologist to discuss antiepileptic treatment during pregnancy.

Epilepsy is not a contraindication to breast-feeding. Although the drug delivery mechanisms are not fully developed in the newborn, and anti-epileptics received through milk may be stored in the body, it may be breast-fed if closely monitored and treated in the long term.

Cessation of antiepileptic medication. Since antiepileptic drugs control only the excessive electrical activity of the brain and do not cure it, the treatment should be followed for an indefinite term. The sudden cessation of the treatment or changing it can result in uncontrolled seizures. After a short period without seizures, many patients are tempted to abandon the treatment. Practice has shown that the cessation of treatment can be discussed after a seizure-free period of at least two years. Treatment will cease gradually, by constant dose reduction and mandatory under the supervision of the attending physician [19; 24; 25; 27; 31; 37; 49].

Tips for minimizing the occurrence of seizures among epileptics [4; 9; 13; 15; 36]:

- Know your triggers and try to avoid them
- Avoid drugs and alcohol
- Have a regular sleep schedule
- Eat a healthy diet
- Minimize factors in your life that cause stress
- Find a competent physician and follow his instructions
- Do not ever feel weird if you call your doctor to ask questions about your health.
- Take your anti-seizure medication as prescribed.
- Do not skip your anti-seizure medication and do not stop taking it abruptly.
- If the doctor is too busy, the nurse will help you.
- Ask your doctor before taking any other medicines.
- If you have an allergy, do not start taking antihistamines before getting your doctor's approval, especially if you have not taken antihistamines before.
- Educate yourself. Read everything you can about seizures and treatment.

Tips for increasing safety around the house, and how to feel more confident about going out:

- If you suffer from seizures, wear a seizure-alert bracelet (doctors will see it and know what type of treatment to administer)
- Carry your medical information in your wallet.
- Use plastic glasses and plates instead of glass and porcelain.
- Put a "usy" sign on the bathroom door, because if you have a crisis the door is not locked and the people can get to you quickly.
- If your child is suffering from seizures, place an interphone in his / her room.
- If child suffers from epilepsy and sleeps overnight at a friend's house, ask your child to always sleep in a low bed, and never use a top bunk.
- Use tightly fitted sheets
- Sleep without a pillow, or use firm pillows.

• Put emergency telephone numbers and first aid instructions on the refrigerator.
• Beware of tobacco, avoiding the risk of a fire.
• Install smoke alarms.
• Minimize sharp edges and corners
• Carpet the floors with thick under-padding
• Put barriers in front of hot stoves or fireplaces
• Swim with a friend.
• Never swim alone.
• Keep the water level in the tub as low as possible, to avoid drowning in the event of a seizure.
• Do not drive until permitted by law and your physician
• Knowledge is power.
• The more you learn about epilepsy, the more you can deal with your illness.

Responding to seizures. First aid during seizures

An epileptic seizure or seizure attack can be scary. A convulsive crisis temporarily interferes with muscle control, movement, speech, vision, or consciousness. It can cause violent shaking of a person's entire body for a few seconds or minutes, accompanied by loss of consciousness [7; 12].

Convulsions can be mild or severe and affect patients differently. Even if people feel helpless when they are around a patient who has convulsions and is difficult to look at, there are many things they can do to help [8; 14; 35]

If you see someone having a seizure, you can do the following:
• Keep yourself and others calm;
• Protect the person from injuries;
• Protect from falls if possible;
• Move furniture or other objects that could harm the person during the crisis;
• Do not insert anything, including fingers, into the patient's mouth

The person responding and performing first aid can be bitten, to minimize the risk, you can do the following [9; 11; 19]:
• Turn the patient to one side, with the mouth down (if it can be moved).
• Do not try to keep the patient lying down.

After a seizure:
• Examine whether the person has suffered injuries.
• Turn the patient to one side during convulsions, do this after the crisis has ended and the individual is more relaxed.
• Clean the mouth and remove vomit, if the patient has dyspnea (difficulty breathing).
• Choose a safe area where the patient can be seated to rest.
• Do not let the person eat or drink until completely aware.
• Survey the patient until he/she is aware and familiar with the surroundings.

Precious information can be provided to the treating physician about the person experiencing a seizure disorder. Remember and inform the doctor about these features:
• Way of movement of the patient's body
• Duration of seizures
• Behavior of the patient before the attack
• Patient's behavior immediately after convulsions
• If the individual has suffered a lesion during the seizure

Convulsions do not always require urgent medical attention. However, call emergency services immediately if one or more of these are true:
– The seizure happens in water;
– The person has never experienced a seizure before;
– The person who has experienced a seizure has breathlessness (apnea) for more than 30 seconds;
– The seizure is longer than 5 minutes (the patient may have a life-threatening condition);
– In case of prolonged convulsions, called epileptic status (more than one seizure occurs within one hour);
– The person who has had a seizure disorder does not normally react within one hour after seizures (abolished reflexes, diminished attention or the patient is not completely awake; confusion, nausea or vomiting, dizziness; inability to walk or sit down; fever).
– Seizures occur after the patient complains of sudden headache; following signs of vascular accident such as speech disorder (dyslexia) or speech impairment; visual disturbances; inability to move different body segments to one side (apraxes) [22; 29; 30; 34; 45; 48].
– The person has other health conditions like diabetes or heart disease.
– The person is pregnant.

Conclusions

Management of epilepsy means more than observing the medication prescribed by the specialist. It is also important for the patient to maintain his general health status, monitor the symptoms of epilepsy and response to treatment and take care of his safety. Involvement in the management of one's own affection can help the patient to control his condition and to continue his routine in usual manner. The objective of antiepileptic treatment is to reduce epileptic seizures to zero without intolerable side effects. New treatments should focus not only on reducing the frequency and intensity of seizures but also improving the quality of life of patients.

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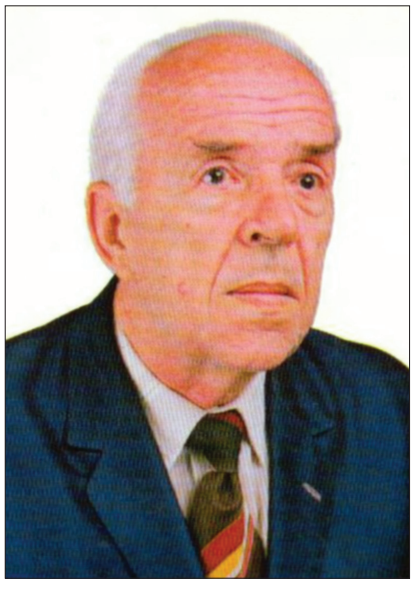
Victor Gheteul – to the 90th anniversary
22.12.1928 – 24.02.2018



Professor Victor Ghețeul was one of the representatives of the second graduation of the State Medical Institute in Chisinau (today *Nicolae Testemitanu* State University of Medicine and Pharmacy of the Republic of Moldova). He was one of the distinguished personalities of the native medicine who, through diligent work, contributed to the founding and consolidation of the Institute, to the creation of the pedagogical staff for the Faculty of Pediatrics and the local pediatric service, to the establishment and development of the national health system.

Professor Victor Gheteul was born on December 22, 1928 in Cluj Napoca, Romania. During 1946-1952 he studied at the Faculty of General Medicine, State Medical Institute in Chisinau, attending the courses of the most prominent teachers of that time. Subsequently, he successfully graduated from the Institute of Pediatrics of the Academy of Medical Sciences in Moscow. His valuable scientific activity was reflected in two theses: Doctor of Medical Sciences (1963) and Philosophy Doctor of Medical Sciences (1981), estimated with an "Excellent" qualification, both of them involving important topics in pediatric pneumology.

In 1986, Professor Victor Gheteul founded the Department of Pediatry No 2, which he led with competency and authority until retirement (1997), continuing though acting as a professor of the same faculty until February 2018.

The great human Victor Gheteul was a remarkable personality, dignified, honest, and wise. The pediatrician Victor Gheteul inspired in his patients trust and love, finding for each one a suitable word, which would give them optimism. A highly skilled diagnostician, a specialist with a unique clinical intuition, an excellent thinker of pediatric science and practice, with a special sense in the development of personalized therapeutic tactics and prevention of complications, severe morbid evolutions, Professor Victor Ghetul succeeded through his prodigious activity to make a renowned name in the pediatricians society of the country.

An integral personality, endowed with a deep civic sentiment, aware of the future of medicine in the republic, Professor Gheteul has prepared a plenty of disciples who work with the same dedication. His special tact imposed respect, excluded negative attitudes, creating around him a climate of understanding, without conflicting situations, a benevolent atmosphere that allowed every member of the team to be professionally accomplished as a doctor, teacher and researcher. The staff of the Department, under the leadership of Professor Victor Gheteul, formed the foundation and the core of a major local scientific school – the Pediatric Pneumology School.

Professor Victor Ghetul was a talented teacher – students of many generations listened to his lectures with satisfaction; he educated through his bright nobility, many disciples, greatly contributed to the training of young pediatricians, who, teaching nowadays in various departments in the country and abroad, keep a vivid and beautiful memory about him.

He was a scientist with a reputation in the field, who left a lot of scientific papers of genuine value, published in the country and abroad. Under the leadership of Professor Victor Gheteul, four doctoral theses and one philosophy doctoral thesis in medical sciences were confirmed.

The illustrious pediatrician, Victor Ghetul, was an expert in the profession, he held the position of main pediatrician and the main pediatric pneumologist of the Ministry of Health in the country, but he also had an international experience as a counselor and expert of World Health Organization in service of mother's and child protection in Laos and Vietnam.

Throughout his activity, Professor Victor Gheteul has manifested his developed sense of duty, his love to work and enthusiasm; he followed his professional path, which has become the essence and meaning of his life. He was an innate intelligent, with the real sense of being good and carefull to the others' pains and needs. These qualities have earned him the honorary title of Homo Emeritus, made him very esteemed in all the posizions which he held.

Professor Victor Gheteul – a bright personality, an experienced clinician, dedicated pediatrician and scientist, as well as Human of Humanity, with much dignity and education, personality with an original intelligence – thereby he became appreciated by us, those who met him, so it remains in our memory as a flame, which throbs and generates the kindness of soul, a fire of desire to know, which still burns.

Ion Ababii, MD, PhD, Professor, Academician
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Chisinau, the Republic of Moldova

Moldovan Medical Journal. December 2018; 61(4)

Efim Alexeevici Muhin – to the 100th anniversary 28.12.1918 – 26.11.1999

This year marks the 100th anniversary from the birth of a remarkable scholar – Efim Alexeevici Muhin, professor, talented organizer of science, Emeritus Scientist, Moldovan State Prize Laureate, personality of great charm. Efim Muhin's name is associated with the implementation of important pharmacological fields, the creation of the scientific school, the foundation of the Scientific Society of Pharmacologists and many other achievements.

Efim Alexeevici Muhin was born on December 28, 1918 in the village of Ciulkovo, Belozersk district, Vologda region, in a family of officials, with old traditions and a great culture. At the guidance and counsel of parents, Efim Muhin successfully passes the admission examinations (1937) to higher medical studies at S. M. Kirov Medical and Military Academy in Leningrad. He studies in this institution for only four years because of the difficult years of the World War II. In September 1941, he was released from the Academy in advance and was enrolled in the army as a military doctor. During the war he worked in various medical positions in the Rifle Division No 177, on the fronts of Leningrad and Volhov. For participation and for his work in the years of the war, Efim Alexeevici Muhin was decorated with: 2 Orders of the Red Star, the Order of the Patriotic War of the 2-nd degree; 9 medals.

In 1947 Efim Muhin graduated with honors from S. M. Kirov Medical and Military Academy, being employed in this institution as a researcher, and subsequently passed through all stages of a scientific-didactic activities at the Pharmacology Department (1948-1951), lecturer (1951-1953), senior lecturer (1953 -1956) and associate professor of Pharmacology Department (1956-1968). The last period of associate professor Efim Alecseevici Muhin includes the two years (1959-1960) as Head of the Pharmacology Department of the Medical and Military Academy. In 1952 E. Muhin defended the thesis of doctor in Medical Sciences on the theme "Pharmacological Characteristics of Dihydrosferofizin" and in 1967 the PhD thesis on the theme "Materials in the the pharmacology of isothioureic derivatives".

In 1968, Efim Alexeevici Muhin is invited by the State Institute of Medicine of Chisinau (SMIC) to be the Head of the Department of Pharmacology. Efim Muhin manifested himself as a highly qualified specialist with a great experience and a high level of organization of the educational-training and scientific activity. In 1970, he was awarded the scientific and didactic title of university professor, and in 1975 he was a scientist emeritus. The remarkable pedagogue Efim Muhin developed 3 manuals for students and trainees, 6 monographs, 4 guides for medicinal preparations and 7 methodological guidelines, published about 180 scientific papers in international and republican magazines, and presented materials in editions in the country and abroad, summaries to international symposiums and congresses. Among the most valuable are: "Pharmacology" – manual (1st and 2nd edition) for students of the Medical and Military Academy and the students of the Chisinau Institute of Medicine, "Pesticides", "Antibiotics", "Pharmacology and Recipe", "Practicum of pharmacology", "Pharmacology of drugs with predominant action on metabolism and antimicrobial preparations", "Pharmacotherapeutic guide for the sector therapist", etc. Based on a set of didactic works, edited with his disciple, corresponding member of the Academy of Sciences of the Republic of Moldova, Professor Victor Ghicavii, and other colleagues from Chisinau and St. Petersburg (associate professor Tamara Pariiskaia), Efim Muhin became, in 1996, the laureate of State Prize of the Republic of Moldova.

Efim Alexeevici Muhin sets up a new scientific direction – **experimental hyperbaric pharmacology**, which studied problems of drug interaction with hyperbaric oxygen, prophylaxis problems in oxygen poisoning. The research in this field is finalized with the defense of 8 doctoral theses in medical sciences and 2 theses of PhD in medical sciences and the editing of the monographs: "Notes of the hyperbaric pharmacology", "Hyperbaric pharmacology", "High-pressure oxygen and hormones". Another direction of scientific research is dedicated to the pharmacology of the cardiovascular system and, in particular, to the antihypotensive remedies, which has resulted in 3 theses of Doctor of Medical Sciences and 2 PhD theses, with the editing of the monographs "Antihypotensive", "Pharmacology of antihypotensive remedies", "Pharmacology of amidin compaunds" and with the development of a new antihypotensive drug – Izoturon.

Efim Muhin is the founder and first president of the Scientific Society of Pharmacologists of the Republic of Moldova; he was a member of the Scientific Society of Pharmacologists of the USSR.

Professor Efim Alexeevici Muhin worked with dedication in education and studies, being the dean of the Faculty of Pharmacy (1970) and General Medicine (1970-1986) of the State Institute of Medicine from Chisinau, enjoying an impressive authority among teachers and among students.

Scientific and professional ties and relationships, founded by Professor Efim Muhin, his work style, intellectual spirit, devotion and professionalism remain in the work of his disciples and friends who continue his ideas in the scientific and pedagogical domain. Numerous disciples, colleagues and friends, those who have known Professor Efim Alexeevici Muhin express their sincere conviction that he was the representative of the Old Russian intelligence, who devoted his intellectual knowledge and skills to the prosperity of medical science. The name of Professor Efim Alexeevici Muhin will remain in the memory of all who have known and worked with him – a person of high intelligence, great dignity and kindness, a model of devotion and responsibility.

Veaceslav Gonciar, MD, PhD, Professor
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1. Submitted articles are first put under consideration to decide whether a given article fits into the area of the journal thematic, then articles are sent to reviewers for further approval, usually to leading experts in the field. The names of the authors and reviewers and their affiliation are not shown to each other.

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