Immunomodulatory influence of early enteral immunonutrition on the dynamics of changes in the cellular immune response after pancreatic resection for cancer

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Abstract

The objective of the study was to investigate the effect of enteral immunonutrition and standard nutrition on the changes of selected cellular immune parameters during the early post-operative period in patients after with pancreatic cancer surgery. Prospective, randomized studies performed in 41 patients after pancreaticoduodenectomy receiving early enteral standard nutrition (group I, n=22) or enteral immunonutrition (group II, n=19). The dynamics of changes for the following cell percentage: CD3+, CD4+, CD4+/CD25+, CD4+/CD38+, CD8+, CD8+/CD38+, CD8+/CD25+, CD19+, CD19+/CD95+, CD19+/CD38+, CD19+/CD34+, CD14+, CD14+/HLADR+, CD14+/CD68+, was evaluated before and on post-operative day 1, 3, 7 and 10 using flow cytometry. A significant improvement in cellular immune response was most frequently noted in the group of patients treated with early enteral immunonutrition. In comparison with pre-operative values for group II, a significant increase in the subpopulation of lymphocytes CD4+ (p=0.02), CD4+/CD25+ (p=0.02), CD4+/CD38+ (p=0.006) was observed on day 7 and 10. In patients receiving immunonutrition between days 3 - 10 after operation, the percentage of CD19+ cells was significantly higher (group I vs. group II, respectively: 5.38±4%; 17.53±14% p=0.001, 5.75±4%; 10.52±5% p=0.01 and 5.12±3%; 17.34±4.8 p=0.001). In patients receiving standard enteral nutrition no significant changes in the subpopulation of CD4+, CD4+/CD25+, CD4+/CD38+ and CD8+ lymphocytes were noted. The percentage of remaining lymphocyte subpopulations (CD3+, CD8+/CD38+, CD19+/CD95+) in both groups did not significantly change. The early enteral immunonutrition as compared with standard enteral nutrition shows an immunomodulative effect on the changes of cellular immune response after pancreatoduodenectomy. Among the evaluated immune parameters only the change of dynamics in percentage of lymphocyte B (CD19+ cells) in patients with no complications after pancreatic cancer resection is an early marker for cellular response to enteral immunonutrition.

Key words: *pancreas surgery, cellular immune response, nutrition.*

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Introduction

Malnutrition, cancer and extensive surgical trauma are the main factors aggravating the post-operative immune disorders, which increase the risk of post-operative complications. It has been proved that malnutrition and improper wound healing correlate with immune disorders [1-4]. This problem refers also to the patients suffering from pancreatic cancer requiring extensive surgery, where the rate of complications is still high [5-7]. One of the ways to improve the immune response and to decrease the number of post-operative complications in this group of patients could be the introduction of immunonutrition.

In properly nourished patients operated on for pancreatic cancer, the application of total parenteral nutrition (TPN) only increased the risk of complications as compared with patients not receiving the above-mentioned nutrition [8]. Previous investigations also suggested that routine applications of post-operative parenteral nutrition to patients undergoing major pancreatic resection for malignancy cannot be recommended because the complications were significantly more serious after TPN compared to the groups not receiving the adjuvant TPN [9]. Baradi H. et al. [10] investigated the best method for delivering enteral feeding in patients following pancreaticoduodenectomy and concluded that early post-operative tube feeding was associated with significantly less frequent use of total parenteral nutrition and the lower rates of readmission and complications. In the last study the enteral feeding administered through a jejunostomy catheter as compared with total parenteral nutrition (TPN) was a safe and beneficial solution for patients who have undergone duodenohemipancreatectomy for a peri-ampullary mass [11]. The established nutritional goal following pancreaticoduodenectomy can be obtained by enteral feeding and the use of immunonutrition, which seems to improve outcome [12]. Early post-operative enteral feeding may safely and effectively replace parenteral nutrition in patients undergoing pancreaticoduodenectomy. Braga et al. [13] proved that early enteral feeding was a suitable alternative to TPN after major abdominal surgery for gastric or pancreatic cancer and the enriched diet appears to be more beneficial in malnourished and transfused patients. In patients with neoplasms of the colorectum, stomach, or pancreas, the perioperative administration of a supplemented enteral formula (enriched with arginine, RNA, and omega-3 fatty acids) reduced post-operative infections and the length of hospital stay. However, both the immune-enhanced and control groups do not differ significantly enough to warrant a change in nutritional dietary supplementation [14]. A study of the same group proved that immunonutrition improves outcome after major pancreatic surgery [15]. Other authors [16] included patients with esophageal, gastric and peripancreatic, or bile duct cancer who underwent resections and were receiving early post-operative enteral feeding with an immune-enhancing formula. The results showed that there were no significant differences in the number of minor, major, or infectious wound complications between the groups. Hospital mortality was 2.5% and the median length of hospital stay was 11 days, which did not differ between the groups. According to this data, the early enteral feeding with an immune-enhancing formula was not beneficial and should not be used in a routine fashion after surgery for upper gastrointestinal malignancies. Another study from the same center indicated that an early standard enteral feeding decreases fat oxidation and the whole body protein catabolism while improving net nitrogen balance. By significantly improving protein metabolism, the standard enteral feeding may decrease post-operative morbidity and mortality after surgery for upper gastrointestinal cancer [17].

The results of the above-quoted studies show that the effectiveness of immunonutrition in patients undergoing major surgery for pancreatic cancer depends on the nourishment status of treated patients, the way of administration for nutritional formula (preferred way of administration – enteral nutrition) and its composition (immunonutrition preferred). However, it is also known that despite intensive immunonutrition in the majority of malnourished patients suffering from pancreatic cancer after major surgery, it is impossible to compensate the increasing protein/caloric deficiency within a short period. Another problem makes the immune disorders being very hard to improve (the most serious on post-operative day 1-3) associated with large surgical wound and local tissue infections. The malnutrition increasing in the early post-operative period aggravates immunosuppression and increases the risk of grave complications. The question is: to what extent can we compensate the post-operative drop in immune response by using the early enteral immunonutrition?

The aim of our study was to compare the effects of early post-operative enteral immunonutrition and standard enteral nutrition on the dynamics of changes in the cellular immune response (including the percentage of the following cells: CD3+, CD4+, CD4+/CD25+, CD4+/CD38+, CD8+, CD8+/CD38+, CD8+/CD25+, CD19+, CD19+/CD95+, CD19+/CD38+, CD19+/CD34+, CD14+, CD14+/HLADR+, CD14+/CD68+) in patients after pancreatic cancer resection.

Material and Methods

The prospective and randomized study covered 41 patients (mean age: 58.03±10.2) operated on for pancreatic cancer. All the patients after full clinical diagnostic procedures (image and laboratory tests) were operated on to resect the head of the pancreas (Whipple's pancreatoduodenectomy). The histopathological examination confirmed the diagnosis. Patients were randomized (by using numbered sealed envelopes stratified by the surgeon) to receive either an early standard (group I – 22 patients, Nutrison Nutricia) or immune-enhancing enteral diets (group II – 19 patients, Stresson Nutricia) (tables 1 and 2).

Table 1. Patient characteristics and surgical parameters of the two groups (supplemented versus standard diet)

Characteristics	**Supplemented diet**		**Standard diet**		**P**
age (y)	59.8±6		54.2±4.1		NS
gender (M/F)	14/5		15/7		NS
location of cancer pancreas	19		22		
tumor staging (TNM classification) I II III	 9 8 2		 8 11 3		NS
type of operation pancreatoduodenectomy	19		22		
duration of surgery (min)	330±60		343±45		NS
operative blood loss (mL)	550±300		600±350		NS
transfused patients	6		7		
nutritional status	**before surgery**	**after surgery**	**before surgery**	**after surgery**	
weight loss (%)	6.52±2.1	9.14±2.8	6.34±3.4	9.23±3.2	NS
BMI (kg/m^2)	23.36±4.5	22.43±6.3	22.24±3.2	21.82±3.0	NS
albumin (g/L)	29.8±0.8	24.1±5.4	28.5±3.1	20.3±6.8	NS
TLC (cells/mm^3)	2151±253	1140±262	1900±624	930±145	NS

Table 2. Composition of diets

Variables	**Composition of the diets/100mL**	
	supplemented diet (n=19)	**standard diet (n=22)**
calories (kcal)	125	100
protein (g)	7.50	4.00
glutamine	1.34	-
arginine	0.89	-
fat (g)	4.2	3.9
LCT	2.0	0.4
MCT	1.5	1.2
EPA	0.079	-
DHA	0.028	-
n6:n3	3.5:1	5:1
L-carnitine (mg)	7.5	-
inositol	63	-
taurine	13	-
choline	46	37
Vit. A (μg RE)	91	82
Vit. E (mg α-TE)	13.0	1.3
Vit. C (mg)	25.0	10.0
osmolarity (mOsm/L)	410	260

The indication for the early post-operative enteral nutrition treatment was the pre-operative loss of body mass (>6% within 2 months) and the extension of surgery (including the advancement of tumor) questioning the possibility of receiving oral diet covering the calorific and protein demand within 7 days after the procedure [18]. The nutritional status was assessed before and on day 7 after the operation based on routine tests revealing a loss in body mass (for initial tests within recent 2 months), body mass index (BMI), albumin concentration and total lymphocyte count (TLC).

The investigations did not include the patients suffering from post-operative infectious complications, unrespectable pancreatic cancer, after transplantations of organs, the patients treated with chemo- or radiotherapy, immunosuppresors, patients with autoimmune diseases, with diabetes type 1 (insulin-dependant), chronic respiratory insufficiency (chronic obstructive pulmonary disease), cardiovascular insufficiency, kidney and lever diseases (biopsy-proven cirrhosis or a serum total bilirubin of >3.0 mg/dL).

The post-operative nutrition in both groups was carried out by using a pump and a tube installed during operation in the distal small bowel loop. The rate of administering the diet was gradually increased from 30 ml/h for the first 24 to 48 hours, and then increased to full feeding depending on the passage of flatus and bowel action. All patients reached their nutritional goal within 72 h. The total feeding time for the entire group of patients amounted to mean 12.3±2 days. Daily supply of the main nutritional substances in standard enteral nutrition amounted on average to: 10.8±1.3 g nitrogen,

208±24.4 g glucose, 66±7.7 g fat (including 101.6±11.9 g of protein and 1693±198 kcal), whereas in enteral immunonutrition: 14.7±2.15 g nitrogen, 177±26 g glucose, 51.4±7.5 g fat, 16.4±2.4 g glutamine, 10.9±1.6 g arginine (including 91.8±13.5 g protein and 1529±224 kcal). The supply of calories and nitrogen did not differ significantly between both groups. Table 2 shows the composition of the diets. The tolerance for both formula diets was excellent.

All patients received antibiotics for prophylaxis (1.2 g augmentin and 2.0 g of cefoperazone), low-particle heparin and were intravenously given crystalline fluids as well as electrolytes depending on actual demand.

The patients signed a written consent after the details of the protocol were fully explained. Protocol of the study was approved by the Medical University Ethics Committee.

Assessing the cellular immune response

Blood samples in all patients were collected from the peripheral vein on the day preceding operation and on days 1, 3, 7 and 10 thereafter. Immunity tests were carried out immediately after collecting blood samples. The phenotype of cellular subpopulations (including the percentage of the following cells: CD3+, CD4+, CD4+/CD25+, CD4+/CD38+, CD8+, CD8+/CD38+, CD8+/CD25+, CD19+, CD19+/CD95+, CD19+/CD38+, CD19+/CD34+, CD14+, CD14+/HLADR+, CD14+/CD68+) was tested by using flow cytometry (cytometer LSR Becton Dickinson, reagents supplied also by Becton Dickinson).

Statistical analysis

Data were presented as a mean standard ± deviation and analyzed by the analysis of variance (ANOVA). To evaluate the statistical significance of difference between pre-operative and post-operative cellular immunity Dunnett's test and Mann-Whitney *U* test for comparisons of lymphocytes and monocytes subsets in standard and supplemented group were used. Chi-squared test and Fisher exact test were used to compare discrete variables. *P* values of less than 0.05 were considered significant.

Results

In the groups compared (group I vs. group II) the pre-operative values for the percentage of tested cellular subpopulations did not significantly differ. During the post-operative period a significant improvement in cellular immune response was noted in the group of patients treated with early enteral immunonutrition (group II). In comparison with the pre-operative values obtained in group II a significant increase in the number of lymphocyte subpopulation CD4+ (p=0.02), CD4+/CD25+ (p=0.02), CD4+/CD38+ (p=0.006) was noted on day 7 and 10, whereas a significant decrease in CD8+ lymphocyte percentage between days 3-10 (peak on day 7, p=0.0001) was revealed. In patients receiving standard enteral nutrition (group I) no significant changes in CD4+, CD4+/CD25+, CD4+/CD38+ and CD8+ (table 3) lymphocyte subpopulation were noted.

The dynamics of changes in the percentage of CD19+ lymphocyte was similar in both group of patients, whereas only the group receiving immunonutrition showed some significant increase in the number of CD19+/CD38+ lymphocytes between post-operative day 1 and 7 (as compared with pre-operative values, the highest increase was observed on day 3, p=0.007). In patients treated with immunonutrition the percentage of CD19+ cells between days 3-10 after the operation was significantly higher (group I vs. group II, respectively p=0.001, p=0.01 and p=0.001) (figure 1). A significant and longer lasting increase in the number of CD14+ cells was noted in the group receiving immunonutrition up to post-operative day 10 (for all values p=0.01), whereas the percentage of those cells in group I was significantly elevated only on day 1. Very similar changes were observed while assessing the percentage of CD14+/CD68+ cells. A significant increase of those cells was noted only in group II between days 1-10 (the highest values on day 3, p=0.007). A significant response to an extensive surgical trauma was found only while assessing the CD14+/HLA-DR+ cells exclusively in group II (the decrease in percentage of activated monocytes between day 1-7 after the operation, the highest values on day 1, p=0.005). The percentage of remaining lymphocyte subpopulation (CD3+, CD8+/CD38+, CD19+/CD95+ in both groups) did not change significantly or was very low in both groups (below 0.5% for CD8+/CD25+ and CD19+/CD34+ - cells – this data were not included in the table).

Discussion

Pancreaticoduodenectomy is one of the most invasive operations in the upper abdominal surgery and usually involves a high incidence of post-operative complications [19--21]. Malnutrition and loss of gut barrier function as well as immune dysfunction after an extensive surgical trauma are main factors that may increase the patient's susceptibility to post-operative complications. One of the ways to reduce the post-operative suppression and the number of complications can be correcting the post-operative immune disorders using enteral immunonutrition. Several studies suggested that artificial nutritional support through the enteral route improves the outcome of malnourished surgical cancer or critically ill patients, but the impact of early post-operative immunonutrition on the changes of cellular immune response in patients after pancreatic resection with uneventful post-operative course is rarely evaluated.

The current study has investigated whether the early enteral immunonutrition, as compared with standard enteral feeding, affected the immune response defined by dynamics of changes in systemic levels of lymphocytes and monocyte subsets after pancreaticoduodenectomy for cancer. The results of our studies were affected by the pre- and

Table 3. The dynamics of changes in cellular immune response (% of cells) in patients who underwent surgery for pancreatic cancer treated with standard enteral nutrition or enteral immunonutrition

Variables	Diet	Pre-operative	POD-1	POD-3	POD-7	POD-10
CD3+	standard	64.24±14.4	60.2±16.9	66.23±13.8	68.92±12.4	69.72±15.0
	supplemented	71.49±9.4	68.91±8.8	71.63±10.8	72.08±10.9	67.89±13.1
CD4+	standard	32.12±7.6	39.72±8.1	40.35±14.6	41.12±9.02	42.06±14.1
	supplemented	36.64±13.0	35.56±8.8	39.9±13.7	43.14±10.9[a]	41.58±14.4[a]
CD4+/CD25+	standard	6.87±4.1	4.47±2.3	4.92±2.9	5.3±3.3	6.34±5.1
	supplemented	3.32±2.1	3.8±2.2	3.6±1.9	4.93±3.1[a]	12.17±8.5[a]
CD4+/CD38+	standard	19.28±8.0	14.65±7.2	18.01±5.9	20.6±14.6	17.12±11.5
	supplemented	18.34±9.0	16.35±4.5	20.11±8.9	24.04±9.8[a]	24.15±10.3[a]
CD8+	standard	30.94±11.4	25.5±11.5	26.33±8.3	29.1±12.3	30.5±13.5
	supplemented	39.65±13.6	37.16±15.23	31.33±16.3[a]	29.9±13.4[a]	33.52±15.8[a]
CD19+	standard	3.63±2.4	7.28±5.41[a]	5.38±4.2[a]	5.75±4.3[a]	5.12±3.1
	supplemented	6.14±3.6	9.91±7.4[a]	17.53±10.2[a, b]	10.52±5.9[a, b]	17.34±4.8[a, b]
CD19+/CD95+	standard	2.83±1.4	1.8±0.6	1.57±0.9	1.86±0.7	1.95±0.8
	supplemented	1.72±1.3	1.6±0.9	1.45±0.5	1.92±1.3	1.77±0.9
CD19+/CD38+	standard	5.34±1.2	6.28±3.6	7.31±6.2	7.28±4.0	5.13±3.2
	supplemented	4.09±1.7	6.51±2.7[a]	9.46±4.9[a]	8.19±3.5[a]	5.68±2.4
CD14+	standard	10.4±8.3	26.92±16.15[a]	23.19±14.5	19.97±17.6	13.06±11.2
	supplemented	10.28±6.8	32.25±14.0[a]	26.95±13.5[a]	24.62±12.2[a]	22.87±14.8[a]
CD14+/HLA--DR+	standard	19.28±15.3	14.86±6.13	13.42±6.7	13.61±8.4	18.31±11.8
	supplemented	31.0±17.7	16.96±7.8[a]	19.82±11.6[a]	21.9±12.3[a]	28.17±13.4
CD14+/CD68+	standard	16.82±14.5	18.21±12.3	20.6±17.4	16.33±10.4	19.1±12.8
	supplemented	15.95±11.4	24.31±14.5[a]	28.4±22.2[a]	19.42±9.6	24.86±15.3[a]

[a] – p<0.05, pre-operative versus post-operative levels and [b] – p<0.05, standard versus supplemented group; POD – post-operative day.

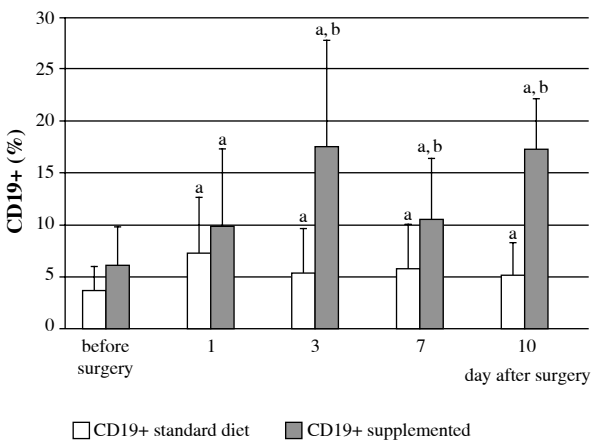

Fig. 1. The dynamics of changes in CD19+ cells (%) after Whipple's pancreatoduodenectomy for pancreatic cancer in patients treated with standard enteral nutrition or enteral immunonutrition. [a] – p<0.05, pre-operative versus post-operative levels and [b] – p<0.05, standard versus supplemented group

post-operative malnutrition of patients, response to extensive surgical trauma and the composition of enterally administered nutritional formulas. The results of our studies unequivocally show that in patients after pancreatic cancer resection (suffering from medium level pre-operative malnutrition), the early enteral immunonutrition increases the cellular immune response in the early post-operative period. The cellular immune response improves independently of malnutrition aggravating in the post-operative period. In comparison with pre-operative values a significant increase in the number of selected subpopulations of lymphocytes ad monocytes was found in patients treated with early enteral immunonutrition. It should be emphasized that the post-operative changes in the assessed immune parameters did not differ significantly (with the exception of CD19+ cells level) in comparable groups of patients (group I vs. group II). It questions the advantage of early post-operative enteral immunonutrition effect as compared with standard nutrition on the level of cellular immune response. One of the most sensitive marker of immune response to early enteral immunonutrition in the investigated group of patients without complications after pancreatic cancer resection was

the post-operative test of lymphocyte B (CD19+ cells) dynamics of percentage changes.

The results obtained can be explained as a result of stimulative effect of immunonutrition to gut immune system (Gut Associated Lymphoid Tissue – GALT). The nutritional formula we used in our studies contained first of all active amino acids (glutamine and arginine), not included in standard nutrition. Glutamine as a nitrogen donor for the synthesis of purines and pyrimidines is the major energy source for the immune system and cells of the small intestine, such as enterocytes. Glutamine maintains the integrity of the gut mucosa. After the enteral administration of glutamine, the number of lymphocytes T increases in Peyer's glands [22]. Some previous studies have shown that glutamine depletion increases spontaneous apoptosis and oxidant-induced cell death in intestinal epithelial cell lines [23]. Intestinal requirements for glutamine appear to increase during catabolic conditions associated with decreased plasma glutamine concentrations and increased cytokine generation by gut mucosal cells [24]. These changes can worsen in malnourished patients after an extensive surgical trauma. The most prominent effect of supplemental arginine is in abrogating trauma-induced immunosuppression and improvement of wound healing. When assessing the mechanism of arginine operation we should remember especially about the key role of nitric oxide and the increased activity of lymphocytes T, whereas the studies of innate immunity should involve the increased activity of NK cells and macrophages [25, 26]. Moreover, the synthesis of nitric oxide from L-arginine may be responsible for the increased splanchnic microperfusion [27].

Studies performed by other researchers found that patients after an extensive surgical trauma for digestive system cancers treated with arginine (25 g per day for 7 days) showed a significant improvement in cellular immune response (increased number of CD4+ lymphocytes), which correlated with better treatment outcome [28]. In our investigations the mean daily dose of arginine was 11g, which could be associated with worse impact of immunonutrition applied to the level of CD4+ lymphocytes. The reduction of impact of immunonutrition used in our studies to cellular immune response could be also associated with immunosuppressive effect of non-saturated fatty acids (EPA and DHA), not contained in the standard nutrition. On the other hand, feeding with moderate amounts of long-chain PUFAs is not clearly immunosuppressive, whereas administering high amounts of these elements might be. Kemen et al. [29] proved that after gastrointestinal surgery for cancer, the supplementation of early post-operative enteral diet with arginine, RNA, and omega-3 fatty acids in the early post-operative period improves post-operative immune responses and helps to more rapidly overcome the immune depression after surgical trauma. The number of T lymphocytes and their subsets, helper T cells (CD4) and activated T cells (CD3, HLA-DR), were significantly higher in the supplemented diet group on post-operative days 10 and 16. B-lymphocyte indices were significantly higher in the supplemented vs. the placebo diet group on post-operative days 7 and 10. Other study showed that in patients with gastric carcinoma after 7 days of post-operative enteral nutritional support, the immunonutrition group (enteral formula enriched with glutamine, arginine and omega-3 fatty acids) had higher levels of immunoglobulin, CD4 cell counts, CD4/CD8 ratio and IL-2 than those in the control group after standard nutrition [30]. The enteral administration of arginine mixed with non-saturated fatty acids as compared with standard non-supplemented diet, increased the lymphocyte response to mitogens, improved nitrogen balance, reduced the number of surgical wound infections and the gravity of those infections, reduced hospital stay and limited the mortality rate in septic patients treated in intensive care units [31, 32]. The glutamine-supplemented enteral nutrition increased the number of CD4+ lymphocytes also in patients who underwent bone marrow transplantations [33, 34]. In our studies the patients receiving immunonutrition showed not only an increased percentage of CD4+ and CD19+ cells, but also the percentage of active T lymphocytes (CD25+, CD38+). On the other hand, the percentage of active monocytes (CD14+/HLA-DR+) in the group of patients treated with immune nutrition was significantly lower, which could be explained as the recovery of normal immune response to an extensive surgical trauma. It was a short-term effect and on day 7 after pancreatic resection in patients without complications the percentage of cells returned to pre-operative values. Similar changes in cellular immune response were found in our earlier studies covering seriously ill patients with massive infections and/or considerable cachexia of parenterally fed patients (TPN) including glutamine [35].

The results of HLA-DR antigen expression assessment in monocytes obtained by other researchers who used modified (supplemented) nutritional formulas are very close to ours. In a prospective randomized trial in patients undergoing major abdominal surgery, the impact of an enteral formula supplemented with arginine, omega-3 fatty acids and nucleotides on immune parameters (expression of activated surface HLA-DR antigen on CD14+ cells, the phagocyte activity of CD14+ monocytes and granulocytes) was compared with a standard enteral formula [36]. After enteral immunonutrition, the expression of activated surface antigen HLA-DR was diminished on CD14+ cells.

We can presume that the effect of immunonutrition on the cellular immune response in malnourished patients after pancreatic cancer resection would be more intense if the same nutrition was also received in the pre-operative period. The results of the study of Ates et al. [37] in patients after curative gastric or colon cancer surgery suggest that pre-operative nutrition via the enteral route provided better regulation of post-operative immune system restoration (CD4+, CD8+, NK cells) than parenteral nutrition and the enteral immunonutrition should be started rather in the

pre-operative period than post-operatively. The results of most recent studies performed in patients operated on for colorectal cancer suggest that post-operative cellular immune response disorders (Th1/Th2 imbalance) should be corrected by introducing short-term (5-day-long) pre-operative enteral immunonutrition [38]. Similar studies [39] covering patients operated on for colorectal cancer treated both pre- and post-operatively with nutrition including arginine and n-3 fatty acids showed that an improvement in cellular response (phagocytosis ability of polymorphonuclear cells and delayed hypersensitivity response to skin tests) correlates with the reduction of frequency of intestinal anastomosis leakage. In addition, this study showed that the post-operative prolongation of such nutrition provides no additional benefit. However, it is known that in the group of patients suffering from colorectal cancer malnutrition is much less frequent than in patients with pancreatic cancer. On the contrary, data reported by McCarter et al. [40] in patients with upper gastrointestinal cancer and pre-operative oral supplementation with immunonutrients (arginine and n-3 fatty acids) failed to show any significant modification of several immunometabolic parameters and the rate of post-operative complications was similar among the groups. Another randomized trial [41] performed in severely malnourished patients with head and neck cancer, showed no effect of perioperative immunonutrition (with arginine alone) on nutritional, immune and outcome variables. In patients with pre-operative weight loss <10% and the cancer of the gastrointestinal tract, pre-operative supplementation with arginine, omega-3 fatty acids, and RNA is as effective in perioperative administration in improving outcome. Both pre- and perioperative immunonutritional strategies were superior to the conventional approach, but in this study immune response was not measured [42]. The basic factor obstructing the comparison and interpretation of results obtained is the considerable differentiation in the groups of patients under investigation and differences in selecting the compositions of nutritional formulas as well as in choosing certain immune parameters.

Concluding, our study clearly shows that the early enteral immunonutrition in comparison with standard nutrition has an immunomodulatory effect on the changes in cellular immune response after an extensive surgical trauma. However, the immunomodulative influence of enteral immunonutrition seems to be selectively marked and refers mainly to lymphocytes B, which may call into question the advantage of effect of this nutrition on the cellular immune response, as compared with standard enteral nutrition. In addition, it should be emphasized that among all assessed immune parameters only the dynamics of changes in percentage of CD19+ cells in patients without complications after pancreatic cancer resection can be used as an early marker of cellular immune response to enteral immunonutrition.

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