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DETERMINATION OF HYALURONIC ACID AND N-PEPTIDE OF COLLAGEN TYPE III LEVELS IN SERUM AS PREDICTORS OF THE PERITONEAL ADHESIONS DEVELOPMENT IN CHILDREN

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Summary

Introduction. *Adhesive peritoneal disease (APD) is the most common and formidable complication of abdominal surgery, which led to adhesive processing occurring in 70 to 90 % of operated patients. It is manifested by late adhesion intestinal obstruction (LAIO), chronic abdominal pain, constipation and infertility.*

The aim of the study *was to investigate the levels of hyaluronic acid (HA) and N-peptide collagen type III (N-PC III) in serum to determine the course of APD in children.*

Material and methods. *Serum levels of HA and N-PC type III were determined in 148 children aged 6-17 years (83 boys and 65 girls). I group (main) – 38 patients with APD, LAIO treatment with the use solution of natrii hyaluronas and decamethoxine, II comparison group – 35 children with APD, LAIO treatment by traditional methods, III group – 45 patients operated on for other acute surgical pathologies of GI tract in which after surgery for 5-10 years no signs of APD were noted. The control group consisted of 30 children operated on inguinal hernias.*

All parents were given informed voluntary written consent to conduct the research. Statistical processing of the obtained results was performed using Microsoft Office Excel and Statistica 10.0 (StatSoft Inc.).

Results. *An increase in HA level in the control, main and comparative groups on the 5th day of postoperative period was registered by 31,94, 126,96 and 39,60%, respectively, in comparison with the preoperative values; on the 14th day decrease in HA level was observed in all groups, by 21,24, 32,79 and 25,83%, respectively, in comparison with the 5th day.*

Determination of the level of N-PC type III in all groups showed an increase in its number on the 5th day, compared with pre-surgery by 64.62, 57.40 and 79.32 %, respectively. On the 14th day the level was 2 times higher in the main group and 2.86 times higher in the comparison group, compared with the control indicators. This indicated the stabilization of connective tissue organization and peritoneal regeneration in the main group and the lack of clear peritoneal regeneration in the comparison group.

Conclusions.

1. Serum levels of HA and N-PC type III may be markers of the onset and development of APD in children.

2. An increase in the level of HA by more than 30%, and N-PC type III by 90 %, compared with control indicators, a year or more after undergoing surgery for APD, LAIO, may indicate a high risk of recurrence and be used as an additional prognostic indicator.

3. Intraoperative use of sodium hyaluronate and decamethoxine in abdominal surgery and the use of local adhesiolysis as anti-adhesive measures help reduce the risk of re-adhesion in children.

Key words: *Adhesions; Children; Hyaluronic Acid; N-peptide of Collagen Type III; Treatment.*

Introduction

Adhesive peritoneal disease (APD) is the most common and formidable complication of abdominal surgery, which led to adhesive process occurring in 70 to 90 % of operated patients. It is manifested by late adhesion intestinal obstruction (LAIO), chronic abdominal pain, constipation and infertility [3, 9].

According to studies, operations on LAIO account for up to 2.4 % of the total number of operations on organs of abdominal cavity (OAC) [7], and is 60-70 % of all forms of intestinal obstruction [1]. APD can be suspected based on medical history, symptoms, physical, instrumental examination and the presence of risk factors. Modern examination methods should be used to confirm the diagnosis, identify the location of the obstruction and the development of complications such as ischemia, necrosis and perforation of hollow organs [2]. APD in many patients lead to the development of various complications that occur months or even many years after surgery, such as small bowel obstruction, infertility in women, chronic abdominal pain [5]. The percentage of LAIO development is higher in pediatric surgery than in adult surgery. For the treatment of LAIO open and laparoscopic operations are proposed [7, 8], as well as the use of various anti-adhesive agents [11]. There is no need to peritonize peritoneal defects, because the mesothelium in the abdomen settles on these defects, is implanted and prevents the formation of adhesions [4]. Adhesive intestinal obstruction that develops later than the third week after surgery is associated with the transformation of new connective tissue into fibrous and scar tissue [9].

To date, none of the available anti-adhesive barrier agents has significantly reduced the incidence of adhesions [3]. The effect of different blood markers on the development of APD has not been fully investigated. Studies of serum protein (I-FABP) and D-lactate levels in intestinal ischemia have shown important diagnostic value [10]. The area of this work is to study the levels of hyaluronic acid (HA) and N-peptide collagen type III (N-PC III) in the serum of children with APD, followed by the use of connective tissue metabolism as additional predictors of APD.

The aim of the study was to investigate the levels of hyaluronic acid (HA) and N-peptide collagen type III (N-PC III) in serum to determine the course of APD in children.

Material and methods

Serum levels of HA and N-PC III were determined in 148 children aged 6-17 years (83 boys and 65 girls). The main group is 38 patients with APD, LAIO with sodium hyaluronate and decamethoxine (NGD), which have antimicrobial, antihypoxic, antioxidant effects in addition, group II (comparison) - 35 children with APD, LAIO treated by traditional methods, group II - 45 patients operated on for other acute surgical pathologies of OAC (destructive forms of appendicitis with local and widespread peritonitis, intussusception, Meckel's diverticulitis, Hirschsprung's disease, post-traumatic splenectomy, laparoscopic appendectomy), who did not show signs of APD within 5-10 years after surgery. The control group consisted of 30 children operated on inguinal hernias.

Examinations and treatment of patients were performed on the basis of the surgical department of the City Children's Clinical Hospital in Chernivtsi for the period 2006-2021. Biochemical determinations of serum HA and N-PC III levels were performed in the research laboratory of Bukovinian State Medical University. The study was conducted in accordance with the principles of the Declaration of Helsinki. The study protocol was approved by the Local Ethics Committee of the institution for all participants. All parents/guardians were required to give informed written consent to the study.

Exclusion criteria: patients with early adhesive intestinal obstruction, most often developing against the background of intestinal paresis peritonitis, which is sluggish and does not always lead to the development of APD.

Selection criteria: 73 children from APD, LAIO, who were hospitalized in a surgical hospital more than once and were treated conservatively or surgically. Of the 73 patients, 61 were operated on urgently, 12 underwent surgery for chronic abdominal pain, and were hospitalized 4 or more times for intestinal obstruction. Types of surgical interventions are shown in Table 1.

Table 1

Distribution of children with peritoneal adhesive disease with the development of late adhesion intestinal obstruction, according to the performed surgical interventions, n

Operative interventions performed	Main group I	Comparison group II
Adhesiolysis	12	13
Adhesiolysis and resection of the small intestine	6	4
Adhesiolysis and removal of the ileostomy	1	1
Elimination of the cause without total adhesiolysis	12	13
Elimination of the cause without total adhesiolysis and resection of the small intestine	6	3
Elimination of the cause without total adhesiolysis and ileostomy	1	1
Total	38	35

For biochemical study, blood was collected in an amount of 5 ml. Blood was taken in accordance with the generally accepted reservations for venipuncture. Standard tubes were used for storage. Before centrifugation, we made sure that the coagulation process was complete. Samples were free of fibrin or other solid impurities, cleared of separator, cells and clots, and stored for up to 24 h at 2-8°C. After storage, samples were thoroughly stirred and checked for air bubbles; to ensure optimal results, bubbles were removed before analysis. Kits were used to quantify the content of HA and N-PC type III in vitro in the serum by immunochemiluminescence assay using a fully automated chemiluminescent immunoassay series Maglumi 1000. The analyzer automatically calculated the concentration of HA and N-PC type III in each sample based on the calibration curve, which was built by the method of two-point calibration of the reference curve.

The results of the study were processed using the statistical package Statistica 10.0 (StatSoft Inc.). To assess the prognostic value of levels of HA and N-PC type III in the serum in the development of adhesion used the method of ROC - analysis (Receiving Operating Characteristic), which counted for true - and false positive results by calculating areas under ROC - curves and «goodness-of-fit».

Research results and their discussion. The dynamics of serum GC and N-PC type III in groups of children are shown in the Table 2.

Table 2

The level of hyaluronic acid and N-peptide of collagen type III in groups of children before surgery, ng/ml

Groups of children	Number of children	Hyaluronic acid level, ng/ml	The level of N-peptide collagen type III, ng/ml
Control group	30	87.73±4.13	20.72±3.94
Main group I	38	112.85±7.25 p<0.05	39.51±5.71 p<0.05
Comparison group II	35	117.92±8.14 p<0.05	41.74±6.18 p<0.05
III group of children operated on for other acute surgical pathologies of PPP (blood for the study was collected during a routine examination).	45	91.15±5.75 p<0.001 p1<0.05 p2<0.05	22.91±6.01 p<0.05 p1<0.05 p2<0.05

Note: *p - the degree of probability of the studied indicators in comparison with the control; p1 - the degree of probability of the studied indicators in children of the I main group in comparison with children of the III group, operated on for other pathology of the abdominal cavity without signs of APD development; p2 - the degree of probability of the studied indicators in children of the second comparative group compared with children of the third group, operated on for other pathology of the abdominal cavity without signs of development of APD*

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The initial different concentrations of HA and N-PC type III in the blood serum of patients with signs of APD were considered as independent predictors of the development of the adhesion process, using the methods of ROC analysis. It is established that the area under the curve for the level of HA is 0.93 ± 0.053(97.5 % confidence interval [> 0.9]) (Fig. 1), and for the level of N-PC type III 0.858 ± 0.043 (95% confidence interval [0, 7-0.9]) (Fig.2).

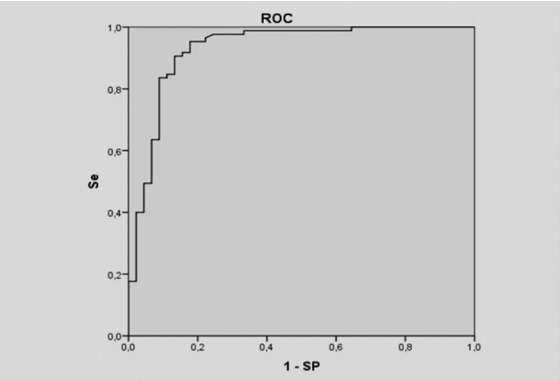

Figure 1. ROC-curve on the level of HA in children, group III, operated on for other acute surgical pathologies of OAC, and group II comparison

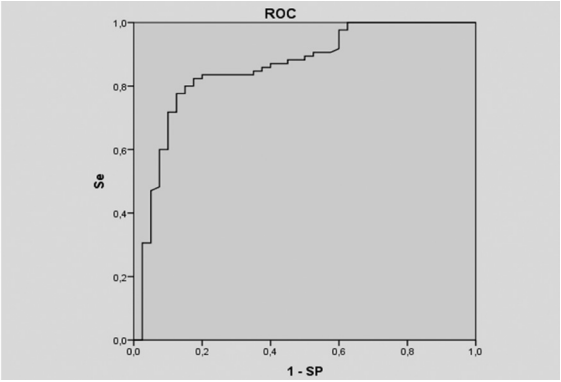

Figure 2. ROC-curve on the level of N-PC III in children, group III, operated on for other acute surgical pathologies of OAC, and II comparative group

When studying the level of HA in the control, main and comparative groups, its growth was noted on the 5th day of the postoperative period, respectively by 31.94, 126.96 and 39.60% compared to preoperative indicators (Tabl. 3).

On the 14th day, there was a decrease in the level of HA in all groups, compared with the 5th day by 21.24, 32.79 and 25.83 %, respectively. However, the level of HA on the 14th day was higher compared to the control in the main group by 88.82%, in the comparison group by 33.94 %. The increase in the level of HA in the control and comparison groups on the 5th day after surgery is associated with increased processes of catabolism of connective tissue proteoglycans and destruction of intercellular substance. The decrease in serum HA in the serum on the 14th day indicated the stabilization of connective tissue organization towards anabolism. The high concentration of HA in the serum of children of the main group compared to others was a consequence of the use of the solution which based on NHD and the ingress of HA from the abdomen into the blood. When determining the levels of N-PC type III in all groups there was an increase in its number on the 5th day compared with pre-surgery by 64.62, 57.40 and 79.32 %, respectively (Tabl. 4).

Table 3

The level of hyaluronic acid in the serum of operated children on the 5th and 14th day of the postoperative period, ng/ml

Groups of children	Number of children	Hyaluronic acid level (ng/ml)	
		5th day after surgery	5th day after surgery 14th day after surgery
Control group	30	115.75±5.53	91.17±6.13
Main group	38	256.12±6.72 p<0.05	172.15±5.18 p<0.05 p2<0.001
Comparison group	35	164.62±7.11 p<0.001 p1<0.05	122.11±9.92 p<0.05 p1<0.001 p2<0.05

Note: *p - the degree of probability of the studied indicators in comparison with the control; p1 - the degree of probability of the studied indicators in children of the main group, compared with the comparison group; p2 - the degree of probability of the studied indicators on the 14th day, compared with the 5th day of the postoperative period*

Table 4

Level of N - serum collagen peptide of operated children on the 5th and 14th day of the postoperative period, ng/ml

Groups of children	Number of children	Level of N - serum collagen peptide (ng/ml)	
		5th day after surgery	5th day after surgery 14th day after surgery
Control group	30	34.11±4.11	22.09±4.56
Main group	38	62.19±5.98 p<0.05	44.15±5.71 p<0.001 p2<0.05
Comparison group	35	74.85±7.34 p<0.05 p1<0.05	63.18±4.91 p<0.05 p_1<0.05 p_2<0.05

Note: *: p - the degree of probability of the studied indicators in comparison with the control; p1 - the degree of probability of the studied indicators in children of the main group, compared with the comparison group; p_2 - the degree of probability of the studied indicators on the 14th day, compared with the 5th day of the postoperative period.*

On the 14th day, there was a decrease in the level of N-PC type III in all groups: control by 35.24 %, main - 29.01 % and comparative - 15.59 %, compared with the 5th day. However, on the 14th day the level was 2 times higher in the main group and 2.86 times higher in the comparison group, compared with the control indicators. The level of N-PC type III was higher by 43.10 % in the comparison group, relative to the main group. Compared to the preoperative period, in the comparison group it was higher by 51.37 %, in the main group by 11.74 %. This indicated the stabilization of connective tissue organization and peritoneal regeneration in the main group and the lack of clear peritoneal regeneration in the comparison group. Thus, the determination of the levels of HA and N-PC type III in children with APD are statistically significant and have a high reliability of comparative indicators in the study groups.

Conclusions

1. Serum levels of HA and N-PC type III may be markers of the onset and development of APD in children.

2. An increase in the level of HA by more than 30%, and N-PC type III by 90 %, compared with control indicators, a year or more after undergoing surgery for APD, LAIO, may indicate a high risk of recurrence and be used as an additional prognostic indicator.

3. Intraoperative use of natrii hyaluronate and decamethoxine in abdominal surgery and the use of local adhesiolysis as anti-adhesive measures help reduce the risk of re-adhesion in children.

The authors declare no conflict of interest.

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References

1. Bazaev AV, Strel'tsov AA, Galanin IA, Stolyarov EV. Enterostoma v lechenii konglomeratnykh form spaechnoy kishechnoy neprokhodimosti. MediAl' [Enterostomy in the treatment of conglomerate forms of adhesive intestinal obstruction]. MediAl'. 2015;2:28-30 (in Russian).

2. Bodnar O, Konoplitskyi V, Khaschuk V, Proniaiev D, Randiuk R. Surgical treatment's possibilities of late adhesive intestinal obstruction in children. Curr Pediatr Res. 2021;25(S1):609-15.

3. Borysenko VB, Koval'ov AM. Bar'ierna profilaktyka spaikovoi neprokhidnosti kyshechnyka. Art of Medicine [Barrier prophylaxis of adhesive intestinal obstruction]. Art of medicine. 2018;4:12-6 (in Ukrainian).

4. Cartwright SL, Knudson MP. Diagnostic imaging of acute abdominal pain in adults. Am Fam Physician. 2015;91(7):452-9.

5. Catena F, Di Saverio S, Coccolini F, Ansaloni L, De Simone B, Sartelli M, et al. Adhesive small bowel adhesions obstruction: Evolutions in diagnosis, management and prevention. World J Gastrointest Surg. 2016;8(3):222-31. doi: 10.4240/wjgs.v8.i3.222

6. Damodarasamy M, Vernon RB, Chan CK, Plymate SR, Wight TN, Reed MJ. Hyaluronan in aged collagen matrix increases prostate epithelial cell proliferation. In Vitro Cell Dev Biol Anim. 2015;51(1):50-8. doi: 10.1007/s11626-014-9800-z

7. Krielen P, Stommel MWJ, Pargmae P, Bouvy ND, Bakkum EA, Ellis H, et al. Adhesion-related readmissions after open and laparoscopic surgery: a retrospective cohort study (SCAR update). Lancet. 2020;395(10217):33-41. doi: 10.1016/S0140-6736(19)32636-4

8. Pereiaslov A.A., Dvorakevych A.O., Nykyforuk O.M. Laparoskopiia u likuvanni ditei zi spaikovoiu kyshkovoiu neprokhidnistiu. Khirurhiia dytiachoho viku.2018;2(59):46-59. doi: 10.15574/PS.2018.59.46 (in Ukrainian).

9. Ten Broek RPG, Krielen P, Di Saverio S, Coccolini F, Biffl WL, Ansaloni L, et al. Bologna guidelines for diagnosis and management of adhesive small bowel obstruction (ASBO): 2017 update of the evidence-based guidelines from the world society of emergency surgery ASBO working group. World J Emerg Surg [Internet]. 2018[cited 2022 Jun 8];13:24. Available from: https://wjes.biomedcentral.com/articles/10.1186/s13017-018-0185-2 doi: 10.1186/s13017-018-0185-2

10. Shi H, Wu B, Wan J, Liu W, Su B. The role of serum intestinal fatty acid binding protein levels and D-lactate levels in the diagnosis of acute intestinal ischemia. Clin Res Hepatol Gastroenterol. 2015;39(3):373-8. doi: 10.1016/j.clinre.2014.12.005

11. Strik C, Wever KE, Stommel MWJ, Goor HV, Ten Broek RPG. Adhesion reformation and the limited translational value of experiments with adhesion barriers: A systematic review and meta-analysis of animal models. Sci Rep [Internet]. 2019[cited 2022 Jun 10];9(1):18254. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6890766/ doi: 10.1038/s41598-019-52457-2

ВИЗНАЧЕННЯ РІВНІВ ГІАЛУРОНОВОЇ КИСЛОТИ ТА N-ПЕПТИД КОЛАГЕНУ ІІІ-ГО ТИПУ В СИРОВАТЦІ КРОВІ ЯК ПРЕДИКТОРІВ РОЗВИТКУ СПАЙКОВОЇ ХВОРОБИ ОЧЕРЕВИНИ У ДІТЕЙ

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Резюме

Вступ. Спайкова хвороба очеревини (СХО) є найчастішим та грізним ускладненням після операцій на органах черевної порожнини (ОЧП), що супроводжується спайкоутворенням, що спостерігається у 70 – 90 % прооперованих хворих. Проявляється пізньою спайковою кишковою непрохідністю (ПСКН), хронічним абдомінальним болем, хронічним тазовим болем, закрепами та безпліддям тощо.

Мета дослідження – дослідити рівні гіалуронової кислоти (ГК) та N-пептид колагену ІІІ-го типу (N-ПК ІІІ) в сироватці крові для визначення перебігу СХО у дітей.

Матеріал та методи дослідження. Визначення рівнів ГК та N-ПК ІІІ типу в сироватці крові проведено у 148 дітей, віком 6-17 років, з них 83 – хлопчики та 65 – дівчатка. І група (основна) – 38 хворих зі СХО, лікування ПСКН із застосування розчину натрію гіалуронату та декаметоксину, ІІ група (порівняння) – 35 дітей зі СХО, лікування ПСКН традиційними методами, ІІІ група – 45 пацієнтів, оперовані з приводу інших гострих хірургічних патологій ОЧП, у яких після операції протягом 5-10 років не були відмічені ознаки СХО. Контрольну групу склали 30 дітей, оперованих з приводу пахових гриж.

У всіх батьків/опікунів було взято інформовану добровільну згоду у письмовому вигляді на проведення досліджень. Статистична обробка отриманих результатів виконувалась у програмах Microsoft Office Excel та Statistica 10.0 (StatSoft Inc.).

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Результати дослідження. При вивченні рівня ГК у контрольній, основній та порівняльній групах відмічено його зростання на 5-ту добу післяопераційного періоду, відповідно на 31,94, 126,96 та 39,60 %, порівняно із показниками до операції, на 14-ту добу спостерігали зниження рівня ГК в усіх групах, порівняно з 5-тою добою на 21,24, 32,79 та 25,83 % відповідно.

Визначення рівня N-ПК III у всіх групах супроводжувалося збільшенням його кількості на 5-ту добу, порівняно із показниками до операції на 64,62, 57,40 та 79,32 % відповідно. На 14-ту добу рівень був в 2 рази вищим в основній групі та в 2,86 рази у групі порівняння відносно з показниками контролю. Це свідчило про стабілізацію організації сполучної тканини та регенерацію очеревини в основній групі та відсутність чіткої регенерації очеревини у групі порівняння.

Висновки.

1. Рівні ГК та N-ПК III типу у сироватці крові дітей можуть бути маркерами виникнення та розвитку СХО у дітей.
2. Збільшення рівня ГК більше ніж на 30 %, а N-ПК III типу на 90 %, порівняно з показниками контролю, через рік та більше після перенесеної операції з приводу СХО та ПСКН, може свідчити про високий ризик розвитку рецидиву та використовуватись як додатковий прогностичний показник.
3. Інтраопераційне застосування натрію гіалуронату та декаметоксину при операціях на черевній порожнині та застосування локального адгезіолізису, як антиадгезивні заходи, сприяють зменшенню ризику повторного спайкоутворення у дітей.

Ключові слова: спайкова хвороба очеревини; гіалуронова кислота; N-пептид колагену III-го типу; діти; лікування.

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