Acute appendicitis, acute appendicitis with peritoneal abscess, acute gastroenteritis, lymphadenitis and gastroduodenitis raises serious health problems in children. The purpose of this study consisted in screening of hematological and biochemical markers to establish their significance especially in the case of abdominal diseases with nonspecific or severe symptoms. Data were collected prospectively from 10750 hospitalized children, of which 2538 with abdominal pain, boys 39%, girls 61%, coming from different social backgrounds: 57.8% urban, and 42.2% rural areas. Children with symptoms of acute appendicitis (52.7%) prevailed. Hematological and biochemical parameters of blood were determined by spectrophotometric, potentiometric, immunological and electrical impedance methods. Results. The majority of children with acute abdominal pain investigate (2358) had white blood cells, sedimentation rate of erythrocytes, neutrophils and lymphocytes percentages and C-reactive protein level exceeded the maximum limit of reference. C-reactive protein values were higher in children with clinical suspicion of acute appendicitis and mesenteric lymphadenitis, this marker is the first that react in cases of inflammations and infections. Inflammatory markers analyzed separately taking have no diagnostic value. The levels of white blood cells, sedimentation rate of erythrocytes and C-reactive protein increased together with neutrophil/lymphocyte high ratio, helpful pediatrician to establish an accurate diagnosis in a shorter time for the correct management of acute abdominal diseases.

Keywords: acute abdominal disease, C-reactive protein, blood cells, natremia, potassemia

The aim of this study was to perform the screening of biochemical and hematological markers in children hospitalized with acute abdominal pain and comparing the levels of these markers for different acute abdominal conditions.

Experimental part

The study group was represented by 10750 pediatric patients hospitalized in the Sfântul Ioan Clinical Emergency Hospital for Children in Galati within a period of 2 years, for various diseases, aged between 1-18 years. All patients were included in the study after they, or their legal representatives have signed an informed consent. Subjects who have not signed the informed consent form were excluded from study. For all patients were completed worksheets with respect to demographics, physiologic and pathologic data. For all patients of the study group has been carried out complete clinical examination, have been collected samples to peripheral blood for different laboratory determinations, biochemical (C-reactive protein, alkaline reserve, natremia, potassemia) and hematological (ESR, number of leukocytes, lymphocytes and neutrophils percentages). Biochemical determinations were performed using automatic biochemistry analyzer VITROS 950 and the reagents supplied by Ortho Clinical Diagnostics, Johnson-Johnson Company, UK. The analyzer uses dry technology on slides (analytical elements assembled in support of multi-layer polyester) for performing in vitro quantitative measurements of components in biological fluids (blood, urine). Dry technology allows performing the reactions successively, ensuring precision and accuracy of measurements reflectance spectrophotometric and colorimetric.
comparable with atomic adsorption spectrometry. Determination of concentration of sodium and potassium ions were made through the potentiometric method: a drop of serum for investigation and a drop of reference liquid deposited on each half of the slide; migrate toward the center of the paper deck. Stable liquid junction formed connects between the reference electrodes to the electrode in the sample. Ion-selective electrode for potassium is made from a membrane that contains valinomycin, an ionophore for potassium. Having cavity approximately equal with the diameter of the potassium ion, the ionophore allows the formation of a complex between these ions and valinomycin which causes changes in potential of the electrode. Ion-selective electrode for sodium contains methyl monensin (ionophore for sodium). The difference in potential between reference electrode and electrode in sample is proportional to the concentration of ion in the blood analyzed, expressed in mmol/L. Reference range for Na⁺ is: 137-145 mmol/L and for K⁺: 3.5-5.2 mmol/L. Serum concentration of ECO, (alkaline reserve) was determined by spectrophotometric method using the kit ECO slides. ECO, is the sum of the concentrations of bicarbonate and carbon dioxide dissolved in the serum. Reference range is: 22-30 mmol/L. CRP determination was performed using automatic biochemistry analyzer Rx IMOLA by immunoturbidimetric method in which human protein C antibodies produces agglutination of the latex particles coated with CRP. Agglutination of the particles is proportional to the concentration of CRP that is determined turbidimetrically. Reference range is: 0-0.5 mg/dL. Hemolecugram was performed with automatic hematology analyzer K CELLTAK MEK produced by Nikon Kohden 6400 Firenze SRL, Italy. The analyzer determined cells count (white blood cells, platelets and red blood cells) by electrical impedance method. Leukocytes formula performed was carried out automatically by the analyzer. ESR was determined by manual method Westergreen, the reference range for children is 4-12 mm/h.

The data was analyzed using EXCEL 2007 and SPSS version 20 for windows software package. To analyze the data we used statistical procedures included: Pearson’s correlation, ANOVA, T-test. Qualitative variables are presented as percent and quantitative variables as mean ± SD. Between groups, the differences were considered significant when p < 0.05.

**Results and discussions**

2537 (23.6%) of the total investigated children (10750) accused abdominal pain. The characteristic symptoms of acute abdominal pain have been observed at 2358 patients (21.9%), while those of chronic abdominal pain at 179 patients (1.7%). The batch of patients with acute pain was formed in 1439 girls (61%; mean age 11.5 years) and 919 boys (39%; mean age 9.8 years); girls/boys ratio 1.57/1. Distribution by age group showed a higher percentage of children included in the group 10-14 years (938 cases, 39.8%), followed by groups 5-9 years (621 cases, 26.3%) and 15-18 (511 cases, 21.7%). Age group between 0-5 years was represented by 288 children (12.2%). Depending on the environment of residence, children with acute abdominal pain originated mainly from urban areas 57.8% (1364 cases), compared to those in rural areas 42.2% (1173 cases), with an urban/rural ratio 1.37/1. The distribution of cases depending on the cause of the pain showed a higher prevalence of children with acute appendicitis (AA) 44.8% (1056 cases). Significantly lower prevalence was observed for acute appendicitis with peritoneal abscess (AAPA) 7.9% (187), gastroenteritis (GE) 9.3% (218), gastroduodenitis (GD) 8.6% (203), mesenteric lymphadenitis (ML) 5.3% (125) and other diseases 24.1% (569). Cases of AA prevailed in children aged over 5 years (99.4%), as those of AAPA (98.4), ML (87.2%) and GD (81.2%), unlike the cases of GE that had a higher prevalence in children under 5 years old (69.7%).

In tables 1 and 2 are listed the values of blood parameters analyzed in the case of children hospitalized for acute abdominal pain with symptoms of acute appendicitis, acute appendicitis and peritoneal abscess, gastroenteritis, gastroduodenitis and acute mesenteric lymphadenitis.

Figure 1 shows graphically the distribution of values of the percentage of neutrophils and lymphocytes depending on the type of acute abdominal disease.

Analytical data indicate significant statistical differences between the groups of patients with AA and AAPA (p < 0.05, table 1) and insignificant between the GD and GE (p > 0.05) for parameters WBC, ESR and CRP.

In children with acute appendicitis, acute gastroenteritis and gastroduodenitis, age was significantly correlated with the WBC and ESR (p < 0.05, table 1) and insignificant between the GD and GE (p > 0.05). Additionally, in children with acute gastroenteritis age was significantly correlated with the CRP (r = 0.856; p = 0.029). The batch of children with

**Table 1**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Acute appendicitis (AA)</th>
<th>Acute appendicitis with peritoneal abscess (AAPA)</th>
<th>p-value*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No* (%)</td>
<td>Mean</td>
<td>Min.</td>
</tr>
<tr>
<td>Total Leukocytes, (WBC), 10^9/μL</td>
<td>310 (29.4)</td>
<td>12.31</td>
<td>2.70</td>
</tr>
<tr>
<td>C-reactive protein (CRP), mg/dL</td>
<td>485 (46)</td>
<td>3.03</td>
<td>0.33</td>
</tr>
<tr>
<td>Erythrocyte sedimentation rate (ESR), mm/h</td>
<td>952 (90.2)</td>
<td>18.90</td>
<td>5.00</td>
</tr>
</tbody>
</table>

*Number of children (Percentage) with biochemical and hematological parameters altered in blood; ** p-value is significant if < 0.05, ***DS - standard deviation.
Acute appendicitis is characterized by inflammation of the cecal appendix and is the most common cause of acute abdomen both the child and the adult. Since there is no drug treatment of AA, diagnosis is necessary to be established quickly for the patient to be operated closer to the onset of disease. The evolution of AA, from the shape debut (catarrhal), to phlegmonous and gangrenous forms, the peritoneal wall is compromised and can occur perforation, postoperative recovery complicated and increases the risk of death. Positive diagnosis is mainly based on symptoms and physical examination. For patients with suspected appendicitis without classic manifestations the doctor recommended laboratory tests that can support the diagnosis in advanced stages of AA and abdominal ultrasound. For children with abdominal pain and suspicion of AA, we performed laboratory tests of blood for the number of neutrophils, the total number of leukocytes, and reactive C protein and sedimentation rate of erythrocytes. The results of these tests indicate values that does not fit into the ranges of variation recommended for all analyzed parameters (GR, WBC, CRP and ESR; Table 1). Thus, the number of neutrophils has ranged from 25.6% to 97% in the case of acute appendicitis and from 30.3% to 92.1% in case of AAPA; most of the values were outside the range of normal (42-85%). It is known that neutrophils help the body in the fight against infection and the healing of the lesions; increase in neutrophils appears as a necessary reaction to the body, while it tries to heal or to remove an invading microorganism or a foreign substance. The increase of the number of neutrophils and their activity can be determined by bacterial, viral, fungal and parasitic infections, as well as inflammatory disorders, including autoimmune diseases and certain medications. For this reason it is believed that neutrophils alone cannot be considered a reliable parameter in the diagnostic for appendicitis in children [9]. However, some authors recommend neutrophil/lymphocyte ratio (GR/L) as a sensitive marker in the diagnosis of appendicitis in children, values of the ratio GR/L higher than 3.5 and can be used in prediction of acute appendicitis [9, 12-14]. Values of GR/L greater than 8 are positive predictive values for gangrenous appendicitis [15, 16] having a higher sensitivity than leukocytosis. WBC count greater than the mean value ± standard deviation or a value greater than 9500 cells per µL in children is defined as Leukocytosis. In both forms of appendicitis total numbers of leukocytes observed by us

<table>
<thead>
<tr>
<th>Diagnostic</th>
<th>Parameters</th>
<th>No. (%)</th>
<th>Mean</th>
<th>Min.</th>
<th>Max.</th>
<th>Std. Dev.**</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gastroenteritis (GE)</td>
<td>Total Leukocytes (WBC), x 10^3/µL</td>
<td>200 (96)</td>
<td>11.46</td>
<td>7.90</td>
<td>35.6</td>
<td>5.04</td>
</tr>
<tr>
<td></td>
<td>C-reactive protein (CRP), mg/dL</td>
<td>218 (100)</td>
<td>4.82</td>
<td>1.34</td>
<td>9.00</td>
<td>2.93</td>
</tr>
<tr>
<td></td>
<td>Erythrocyte sedimentation rate (ESR), mm/h</td>
<td>216 (99.3)</td>
<td>20.77</td>
<td>11.0</td>
<td>82.00</td>
<td>10.32</td>
</tr>
<tr>
<td></td>
<td>Alkaline reserve (RA), mmol/L</td>
<td>216 (99.3)</td>
<td>17.16</td>
<td>8.00</td>
<td>21.00</td>
<td>2.89</td>
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<tr>
<td></td>
<td>Natremia (Na⁺), mmol/L</td>
<td>121 (55.6)</td>
<td>142.62</td>
<td>128.00</td>
<td>166.00</td>
<td>8.42</td>
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<tr>
<td></td>
<td>Potassium (K⁺), mmol/L</td>
<td>117 (53.7)</td>
<td>5.17</td>
<td>2.10</td>
<td>7.80</td>
<td>1.07</td>
</tr>
<tr>
<td>Mesenteric lymphadenitis (ML)</td>
<td>Total Leukocytes (WBC), x 10^3/µL</td>
<td>103 (82.8)</td>
<td>12.05</td>
<td>3.60</td>
<td>25.90</td>
<td>5.61</td>
</tr>
<tr>
<td></td>
<td>C-reactive protein (CRP), mg/dL</td>
<td>125 (100)</td>
<td>2.83</td>
<td>1.52</td>
<td>6.21</td>
<td>2.26</td>
</tr>
<tr>
<td></td>
<td>Erythrocyte sedimentation rate (ESR), mm/h</td>
<td>122 (98.0)</td>
<td>17.84</td>
<td>10.00</td>
<td>48.00</td>
<td>7.56</td>
</tr>
</tbody>
</table>

*Number of children (Percentage) with biochemical and hematological parameters altered in blood; **DS - standard deviation

Figure 1. Distribution of neutrophil and lymphocyte percentage values depending on the diagnosis of mesenteric lymphadenitis age was significantly correlated only with the WBC (r = -0.576; p = 0.001).
have overtaken 2.8 and 2.7 times the upper limit of the normal range (4000-9500 cells per μL) in the case of acute appendicitis and acute appendicitis with peritoneal abscess respectively. The total number of leukocytes has diagnostic value only in combination with high levels of CRP, with population of neutrophils and increased GR/L ratio [12, 17-22].

C-reactive protein is a protein synthesized by the liver whose blood levels are increased in inflammation. In the case of both forms of appendicitis C-reactive protein levels (CRP) exceeded the recommended maximum level (0.5 mg/dL). CRP test shows that levels of this protein were consistent with the severity of appendicitis, the average values are approximately 3 times higher in the case of AAPA, compared with acute appendicitis. Our results confirm those reported by Yokoyama [23] and Grönroos [6] which recommends increased levels of CRP as a marker of surgical indication for AA. C-reactive protein is not a diagnostic test but may help confirm the diagnosis in patients with atypical presentation. CRP is often used as an inflammatory marker not only in case of acute appendicitis [10, 23, 24, 25], but in other inflammatory diseases [26, 27]. Very high levels of CRP in patients with appendicitis indicate to form gangrenous disease progression, particularly if associated with leukocytosis and appendicitis indicate to form gangrenous disease [26, 27].

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In the case of acute GE, the most sensitive acute phase reagents were: CRP (100% cases), ESR (99.3%) and WBC (96%), 59.6% of children with acute GE showed increased levels of neutrophil count and 53.7% in the number of lymphocytes (table 2). CRP levels found in new results confirm the findings of other authors [38-40], indicating higher levels for this parameter in the case bacterial infection as compared with those viral. Determination of the concentration of serum C-reactive protein permits early differentiation between bacterial and viral GE, the positive predictive value for this parameter alone is ≥ 2 mg/dL [40]. In addition, the measurement of inflammatory markers we have investigated and levels of serum electrolytes (sodium and potassium), because the diarrhea and vomiting are symptoms in acute GE. GE in children tested caused both hyponatremia (sodium levels in the blood stream less than 137 mmol/L) and hypokalemia (potassium levels < 3.5 mmol/L) and hypernatremia (sodium levels in the blood stream > 145 mmol/L) and hyperkalemia (potassium levels > 5.2 mmol/L). Sodium levels in the serum of patients with acute GE have varied between (128-166) mmol/L and potassium between (2.1-7.8) mmol/L; the prevalence of children with acute GE with altered levels of electrolytes was 55.6% in case of sodium and 53.7% in case potassium. Serum sodium levels are enhanced when the body loses more water than electrolytes [41] and decrease when the body loses more water than sodium, especially in cases of severe diarrhea and vomiting. Signs of hyponatremia include headache, confusion and lethargy. High sodium level causes thirst, confusion and seizures, and hyperkalemia causes the dangerous arrhythmias or abnormal heart rhythms. Hypokalemia usually cause milder symptoms, such as muscle cramps, fatigue and constipation. Analysis of alkaline reserve to 99.3% in patients with acute gastroenteritis highlight values (8.0-21 mmol/L) below the minimum limit of the range of normal variation (22.0 mmol/L). These low values ECO, suggests metabolic acidosis associated with dehydration of children due to diarrhea and vomiting children. In suspected cases of dehydration > 10% is required intravenous administration of fluids. The prevalence of children with gastroodudinitis (GD) who presented changes in biochemical and hematological parameters ranged from 1 to 100% depending on the parameter investigated (table 2). Leukocytosis and lymphocytosis observed indicate the inflammation process or a possible infection with Helicobacter pylori [42]. For all diseases analyzed reports neutrophil / lymphocyte were relatively small as 1.76 in the case of AA, 2.76 AAPA, GE 1.37, 2.5 ML and 3.0 for GD (fig. 1), they are below the level reported by other authors [10, 12-16].

Conclusions

WBC, CRP, ESR, neutrophil and lymphocyte percentages have shown significant changes at more than 50% of the children hospitalized for abdominal pain with specific symptoms of AA, AAPA, GD, GE, or ML, which confirm the nature of the inflammatory and infectious diseases.

Hematological and biochemical parameters analyzed (WBC, CRP, ESR, GR, and L) are not diagnostic criteria for abdominal diseases when considering their individual values, due to low specificity.

A combination of CRP and WBC, with Neutrophil/Lymphocyte ratio determine increased positive predictive value of diagnostic and can help in making decisions regarding the diagnosis of these diseases especially in cases of severe and those severe requiring surgery.
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References