Is urinary 5-HIAA determination a valuable method in diagnosis of acute appendicitis in children?

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ABSTRACT

The aim of the study is to investigate the significance of spot urine 5-hydroxyindoleacetic acid (5-HIAA) levels in patients admitted with the suspicion of acute appendicitis. Seventy-one patients with the mean age of 9.4±2.9 years, who were admitted to our pediatric surgery clinic between August 2002 and March 2004 with the complaints of abdominal pain were evaluated prospectively. Additionally spot urine samples were collected from 34 healthy children for control. 5-HIAA was detected from the urine samples in all children with high-performance liquid chromatography (HPLC) method. The results were analyzed with one way analysis of variance (ANOVA), post hoc Tukey HSD test and receiver operating characteristic (ROC) curve. After the clinical follow-up appendicitis was detected in 40 patients and abdominal pain was found to be due to causes other than appendicitis in 31 patients. Results were found to be correlated with the final diagnosis in all patients. The mean 5-HIAA levels were 2.5±1.8 µmol/dl in healthy children, whereas 9±5 µmol/dl in nonappendicitis and 18.9±17.8 µmol/dl in appendicitis patients, respectively. The difference between the patients with acute appendicitis and the other two groups were statistically significant (P = 0.001). For a value of 8.9 µmol/dl, this test was found to be 70% sensitive and 67% specific according to the ROC curve. 5-HIAA significantly rises in pediatric acute appendicitis. However, due to high values of SD and relatively low sensitivity and specificity, this test seems to have limited diagnostic power as a single parameter in childhood acute appendicitis.

KEY WORDS: 5-HIAA, acute appendicitis, children, diagnosis, enterochromaffin cells

Appendicitis is one of the most common causes of acute abdomen in children. Early diagnosis is a major concern for the prevention of complications and decreasing general costs of the treatment.[1-4]

Enterochromaffin cells (EC) are found in mucosal layer of entire intestinal system but they are mostly prominent in distal appendix and comprise the major cell type in the intestinal endocrine system.[5] Serotonin is the main secretory product of these EC. In an experimental study done previously in our unit, the increase of serotonin metabolite 5-hydroxyindoleacetic acid (5-HIAA) has been shown in rabbits with acute appendicitis.[6] The aim of this prospective clinical study is to investigate this action clinically in children with acute appendicitis.

MATERIALS AND METHODS

The study was approved by the local ethics committee of our institution. Patients who have been brought to our clinics between August 2002 and March 2004 with the complaint of abdominal pain were evaluated in this prospective randomized clinical study. Totally 50 boys and 21 girls with a median age of 9±2.9 years were included in the study. Spot urine samples were collected in all patients during admission. Pathologies like carcinoid tumors which might increase 5-HIAA as well were differentiated intraoperatively in appendicitis patients and clinically in nonappendicitis patients. The patients were also evaluated for other causes of increased 5-HIAA secretion such as fruit ingestion, medications and clinical conditions like gastroenteritis. Those who had symptoms and physical examination findings suggestive of acute appendicitis were operated. The criteria used for the diagnosis was adapted from the pediatric appendicitis scoring system according to Samuel.[7] Ultrasonography was done in most of the patients for diagnosis but was not evaluated in the present study. The diagnosis was confirmed with pathological examinations postoperatively. Additionally, spot urine samples were collected from 34 healthy children for control. 5-HIAA was determined from the spot urine samples in all children.
with high-performance liquid chromatography (HPLC) method. The mean levels of 5-HIAA of each group were compared.

Five milliliters of spot urine samples were taken into test tubes with 6 N HCl in pH 3. The samples, which were studied shortly after were kept at 4°C and those that were planned to be studied after a while were kept at -20°C. After the urine was stirred, pH was dropped to a level between 4.5 and 6.5 with 2 N NaOH. 5-HIAA determination was done with Hewlett Packard 1100 series HPLC system and ClinRep® (Recipe Chemicals and Instruments GmbH Munich, Germany) urine kit for 5-HIAA. Lyophilized iso-VMA (100 ng/ml) was used for internal standard, which was inside the kit. Hewlett Packard 1049 A Electrochemical Detector (detector no: 3314G01022, Germany) was used in a detector potential of 800 mV and a range of 50 nA. 5-HIAA was calculated with the comparison of peak value at 14-15 min after the injection of 20 µl of sample with the values for internal standard at 3-4 min. The results were given in µmol/dl. Pump flow rate was 0.9 ml/min, column temperature was kept at 30°C and substances that would give interference were removed with sample washings. 5-HIAA was removed from the column chromatographically and detected electrochemically. The levels of 5-HIAA between groups and between perforated and nonperforated appendicitis patients were also compared.

The results were analyzed with one-way analysis of variance (ANOVA), post hoc Tukey HSD test and receiver operating characteristic (ROC) curve with SPSS 11.0 version after the groups were tested for normal distribution. The comparison between the perforated and nonperforated appendicitis patients was done with Wilcoxon signed ranks test. The results were evaluated in mean±SD values. P < 0.05 was considered to be statistically significant.

RESULTS

A total of 40 patients with appendicitis and 31 patients with abdominal pain due to causes other than appendicitis were evaluated. The diagnosis of appendicitis was confirmed with intraoperative findings and pathological examinations and the diagnosis in nonappendicitis groups was confirmed with clinical follow-up. In all cases, the spot urine samples were collected during initial admission. Thirteen of the appendicitis cases were found to be perforated in the operation (32.5%). There were two cases with negative appendectomies (4.8%) and their final diagnoses were gastroenteritis and salmonellosis. The causes of abdominal pain in nonappendicitis group were mesenteric lymphadenopathy (n: 9), nonspecific abdominal pain (n: 8), chronic constipation (n: 4), gastroenteritis (n: 3), urolithiasis (n: 2), urinary tract infection (n: 2), salmonellosis (n: 1), cholelithiasis (n: 1) and intussusception (n: 1).

The mean 5-HIAA levels were 2.5±1.8 µmol/dl in healthy children, whereas 9±5 µmol/dl in nonappendicitis and 18.9±17.8 µmol/dl in appendicitis patients [Figure 1]. The comparison of 5-HIAA levels between nonappendicitis and appendicitis patients was found to be statistically significant (P < 0.01) and the comparison between nonappendicitis and control groups was not significant. On the other hand, the comparison of 5-HIAA between patients with perforated appendicitis and nonperforated appendicitis was not significant either (5-HIAA in nonperforated cases 18.8±17 vs 19.2±20 µmol/dl in perforated cases, respectively, P > 0.05).

When the 5-HIAA levels in appendicitis and nonappendicitis patients were evaluated in ROC curve, for a value of 8.9 µmol/dl, this test was found to be 70% sensitive and 67% specific [Figure 2].

DISCUSSION

Early and correct diagnoses are the main goals in the treatment of acute appendicitis. Many diagnostic modalities, both biochemical and radiological, have been suggested in the literature for this purpose. Ultrasonography (US) has been told to be a useful tool in diagnosis and additional use of computed tomography...
(CT) has been observed to increase diagnostic accuracy. However, in other studies CT examinations were found to be ineffective in reducing the negative appendectomy rates when compared with US. Still in a North American study, 61% of surgeons participating in a survey, preferred CT examination over US in their daily practice. Laboratory studies such as leukocyte and neutrophil counts, C-reactive protein (CRP), interleukin-6, phospholipase A2 and other inflammatory determinants have also been proposed in the differential diagnosis of acute appendicitis with relatively low sensitivity and specificity. Physical examination has been emphasized as the most effective diagnostic tool when used with other modalities to form scoring systems in acute appendicitis.

Recent interest has been focused on the level of spot urine 5-HIAA in appendicitis in adults. Rørdam et al has previously showed the increase of serotonin from the enterochromaffin cells in the appendix in cases of acute appendicitis. We have also demonstrated this increase in an experimental model in rabbits. Various other studies have confirmed this observation but others have found that this test is not useful in appendicitis.

We have observed from this study that 5-HIAA significantly increases in spot urine samples of children with acute appendicitis. For a cut-off value of 8.9 µmol/dl, this test is found to be 70% sensitive and 67% specific. Bolandparvaz et al. have found a decrease in 5-HIAA in perforated appendicitis in adults. Rørdam et al. have demonstrated the increase of plasma serotonin to be 45% sensitive and 95% specific in appendicitis. According to Ilkhanizadeh et al, the increase in spot urine of adults was 98% sensitive and 100% specific, whereas the sensitivity was 84% and specificity was 88% in the study of Bolandparvaz et al. On the other hand, Mihmanli et al. have given a sensitivity of 22% and a specificity of 95% in their study and considered this test to be unreliable in the diagnosis of acute appendicitis. However, as the distribution of EC which locate mostly in the extraepithelial layer of appendix is age-dependent and has been known to be maximal in young adults, these results can hardly be applied for children. This study is the first to evaluate the possible value of urinary spot 5-HIAA determination in childhood appendicitis as far as we have searched the English literature.

When we consider our findings, due to a high standard deviation and relatively low sensitivity and specificity, this test seems to have a questionable superiority over the conventional methods in the diagnosis of acute appendicitis in children in spite of the findings of Bolandparvaz and Ilkhanizadeh in adults. Urinary 5-HIAA has been determined with HPLC method in this study. This method is fast, easy and practical. It is cheaper than US and CT and equivalent to other blood tests. However, it may not be feasible to do this test in centers with low number of referrals or in developing countries as its routine determination may be limited. 5-HIAA levels may be changed after ingestion of certain foods and in certain clinical conditions like carcinoid tumors, acute gastroenteritis, inflammatory bowel disease, apudomas, coeliac disease and patients treated with aminosalicylates. 5-HIAA in spot urine has been known to have no diurnal change and no variation between males and females. Even though these factors have been considered for differential diagnosis in this study, high values of SD shows that it may not be easy to standardize this test for all patients which decreases its reliability.

In conclusion, 5-HIAA significantly increases in acute appendicitis of children. However, due to the difficulties in standardization and relatively low sensitivity and specificity, its clinical diagnostic power is limited as a single parameter. Future sensitivity and specificity studies in large patient populations should be done to evaluate its exact efficacy as a major determinant of appendicitis.

REFERENCES