The American Journal of Surgery (2015) 209, 1043-1047

The American Journal of Surgery®

Clinical Science

Diagnostic markers in acute appendicitis

Yahya A. Al-Abed, M.D., Ch.M., M.R.C.S. (Eng)*, Nasser Alobaid, M.B.B.S., F.R.C.S., Fiona Myint, F.R.C.S.

Department of General Surgery, North Middlesex University Hospital NHS Trust, Sterling Way, London N18 1QX, UK

KEYWORDS:
Appendicitis;
Diagnosis;
Diagnostic markers;
Bilirubin;
Hyperbilirubinemia

Abstract

BACKGROUND: An adequate medical history combined with clinical examination is usually enough to make the diagnosis of acute appendicitis. The aim of this study was to determine the value of elevated white cell count (WCC), C-reactive protein (CRP), and bilirubin as diagnostic markers of acute appendicitis.

METHODS: A retrospective analysis was carried out on consecutive patients who underwent appendicectomy over a 3-year period. All data including patients' age, sex, blood test results, and appendix histology were summarized. Sensitivities, specificities, positive, and negative predictive values of WCC, CRP, and bilirubin were calculated separately or in combination for all patients.

RESULTS: A total of 447 patients were included. There is a significant difference in the results between patients with negative and positive appendicitis with regards to CRP (32 vs 73; $P < .001$), mean total WCC (10.9 vs 14.0; $P < .001$), and the mean levels of bilirubin (10.9 vs 17.2; $P < .001$).

CONCLUSIONS: The diagnosis of appendicitis remains multifactorial and blood tests may help to guide the surgeon in the decision making.

Acute appendicitis is one of the most common surgical emergencies and appendicectomy remains among the most frequently performed emergency operations worldwide.[1–3]

An adequate medical history combined with clinical examination to elicit common physical signs associated with localized peritonitis is usually enough to make the diagnosis of acute appendicitis. However, the diagnosis of appendicitis is not always straight forward especially in female patients as a gynecological pathology may mimic acute appendicitis. Moreover, the variability in appendicular locations such as in hidden or retrocecal appendicitis may not allow patients to exhibit enough peritoneal signs to support the diagnosis of acute appendicitis.[4] To date, reliable specific marker of acute appendicitis has not yet been identified. Despite advances in technology and investigation modalities, the rate of negative appendicectomies remains between 15% and 50%.[5]

Several Scoring systems have been developed to aid in the diagnosis of acute appendicitis (Alvaredo,[6] Lintula[7] and RIPASA[8]). However, these systems have their own limitations and are mainly used in children and have not shown to be accurate in the adult female population.[6]

In practice, the diagnosis of acute appendicitis is supported by the presence of elevated inflammatory markers, that is, white cell count (WCC) and C-reactive protein (CRP). However, some studies have shown that neither of these markers is diagnostic nor specific for acute appendicitis.[9]

The authors declare no conflicts of interest.

* Corresponding author. Tel.: +44-1-206-742447; fax: +44-1-206-742030.

E-mail address: yalabed@yahoo.co.uk

Manuscript received March 16, 2014; revised manuscript May 20, 2014

0002-9610/$ - see front matter © 2015 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.amjsurg.2014.05.024

Table 1 Patients according to sex and appendix histology result

	Positive for appendicitis	Negative for appendicitis	Total
Men	248	19	267 (59.7%)
Women	138	42	180 (40.3%)
Total	386	61	447

Recently, serum bilirubin has been found to play a useful role in the diagnosis of perforated appendicitis with a sensitivity of 70% and specificity of 86%.[10] The diagnostic accuracy of appendicitis based on hyperbilirubinemia remains uncertain.

The aim of our study is to determine the value of elevated WCC, CRP, and bilirubin as diagnostic markers of acute appendicitis.

Patients and Methods

A retrospective analysis of collected data was carried out on all consecutive patients who underwent appendicectomy at our institution over a 3-year period (from January 2007 to April 2010). Patients were identified from our research and audit department using the search terms "appendicitis," "appendicectomy," and "laparoscopic appendicectomy." All data including patients' age, sex, blood test results (WCC, differential leukocytes count, CRP, and liver function tests including bilirubin), and appendix histology results were obtained from our audit department and hospital computerized record system (NMPath). All collected data were fed into and summarized on an EXCEL spreadsheet (Microsoft Excel 2007, Redmond, WA). All patients' identifying data were stored on a secure password protected computer, access of which was provided by the hospital login.

Appendicectomy was performed conventionally or laparoscopically. There was no age cut off. Patients with incomplete blood test results (WCC, CRP, or liver function tests) and patients with known liver disease, Gilbert's syndrome, or with persistently elevated liver function tests were excluded from the study.

Blood test results were deemed positive if they were above the upper limits of the hospital's laboratory values. Hyperbilirubinemia is thus defined as bilirubin levels of greater than 15 μmol/L. Leukocytosis is defined as WCC of greater than 11 × 109/L, and CRP was considered elevated if levels were more than 10 mg/L.

Patients were classified into 3 groups according to appendix histology result. The first group (control group) contains patients with negative appendicectomy, that is, normal appendix on histology. The second group has patients with appendicitis proven on histology without evidence of perforation, while patients with evidence of appendicular perforation form the third group.

Data were analyzed using SPSS version 19.0 (SPSS, Inc, Chicago, IL). Continuous variables were expressed as mean and standard deviation or range and median. Sensitivities, specificities, positive predictive value (PPV), and negative predictive value (NPV) of WCC, CRP, and bilirubin were calculated separately or in combination for all patients. One-way analysis of variance test was used to analyze difference between means of variables among patients' groups. Results were considered statistically significant when P value was of less than or equal to .05.

Results

Between January 2007 and April 2010, appendicectomy was performed in 567 patients. By applying exclusion criteria, 120 patients were excluded (73 patients had positive appendicitis). A total of 447 patients were included in the study, of which 267 were men and 180 were women. Mean age was 27.1 years (range: 5 to 83 years). The total number of children (age < 16 years) was 76 (17%). The number of patients with negative appendicectomy (control group) was 61 (13.6%), of which 42 were women (Table 1).

The total number of patients who had appendicitis was 386, of which 18.1% (n = 70) had histology confirming perforated appendicitis.

WCC was elevated in 52.6% of men, while it was elevated in 40.5% in women in the negative appendicectomy group. For patients with appendicitis, WCC was elevated in 79.4% of men and 63% of women. In the perforated appendicitis group, WCC was elevated in 82.4% of men and 61% of women (Table 2).

Table 2 Summary of percentages of positives laboratory results including WCC, CRP and bilirubin according to gender and appendix histology results

Criterion	Sex	Negative %	Positive %	Perforated %
WCC	Male	52.6	79.4	82.4
	Female	40.5	63	61.1
CRP	Male	47.4	78.2	92.2
	Female	40.5	73.2	94.4
Bilirubin	Male	26.3	54.4	60.8
	Female	16.7	29.7	50

CRP = C-reactive protein; WCC = white cell count.

Table 3 Results of blood markers according to appendix histology

	WCC	Neutrophils	Lymphocytes	Monocytes	AST	ALP	Bilirubin	CRP
Negative	10.9	8.1	1.9	.69	23.9	96.8	10.9	32.4
Positive	14.0	11.5	1.5	.86	26.2	104.0	17.2	73.3
ANOVA	$P < .001$	$P < .001$	$P < .001$	$P = .002$	$P = .161$	$P = .454$	$P < .001$	$P < .001$

ALP = alkaline phosphatase; ANOVA = analysis of variance; AST = aspartate transaminase; CRP = C-reactive protein; WCC = white cell count.

In the appendicitis group, CRP was similarly elevated to 78.2% and 73.2% in men and women, respectively. With regards to the perforated appendicitis group, CRP was elevated in the majority of patients approaching 95% (Table 2).

Bilirubin was elevated in 26.3% of men and 16.7% on women in the negative appendicectomy group. This figure doubles in patients with appendicitis (54.4% in men and 29.7% in women), while in patients with perforated appendicitis group, this figure reaches more than 60% (Table 2).

Twenty-five patients (6.4%) in the appendicitis group noted to have normal WCC and CRP. Of those, 23 patients had normal WCC, CRP, and bilirubin.

Comparison was made between patients with negative appendicectomy group and patients with confirmed appendicitis (simple and perforated appendicitis groups) with regards to total and differential WCCs, CRP, bilirubin, aspartate transaminase, and alkaline phosphatase. Table 3 summarizes the results.

Analysis of variance test is used to test significance between groups. There appears to be a significant difference in the results between patients with negative and positive appendicitis with regards to CRP (32 vs 73; $P < .001$), mean total WCC (10.9 vs 14.0; $P < .001$), and differential WCC including neutrophils (8.1 vs 11.5; $P < .001$) and lymphocytes (1.9 vs 1.5; $P < .001$).

There is a significant difference in the mean levels of bilirubin between patients with positive appendicitis and those with negative appendicectomy (17.2 vs 10.9; $P < .001$). This difference is not significant with regards to mean levels of the other liver function tests including aspartate transaminase and alkaline phosphatase, indicating significant isolated hyperbilirubinemia (Table 3).

Sensitivity, specificity, PPV, and NPV were calculated in isolation for WCC, CRP, and bilirubin. These parameters were further evaluated by combining diagnostic markers such as WCC and CRP, CRP and bilirubin, and finally combining all markers (WCC, CRP, and bilirubin) (Tables 4 and 5).

In Table 4, WCC and CRP appear to have similar sensitivity, specificity, PPV, and NPV. Here, they both have low sensitivity and low specificity with a high PPV of 91%.

Furthermore, more than 93% of patients with elevated levels of bilirubin have positive appendicitis as indicated by PPV.

Specificity and PPV have significantly improved after combining markers such as WCC and CRP and CRP and bilirubin. Furthermore, specificity and PPV have dramatically increased after combining WCC, CRP, and bilirubin approaching 95% and 98%, respectively (Table 5).

Comments

Diagnosis of acute appendicitis can be difficult, especially in women. A delay in the diagnosis and management can lead to appendix rupture and subsequent peritonitis. Despite advances in technology and imaging modalities, there is no blood marker for acute appendicitis and therefore we cannot reliably make the diagnosis of acute appendicitis based on one test or sign but rather by a combination of clinical, laboratory, and radiologic examinations were indicated.

Over the last decade, some attention has been drawn to the association between hyperbilirubinemia and appendicitis. Perhaps this could be explained by the over ordering of "routine" blood tests in the emergency department. As a result, more studies are performed to test this hypothesis.

Jaundice in the context of appendicitis has been well described in the literature over 60 years ago.[11] It is postulated that elevated serum bilirubin occurs as a result of portal sepsis or empyema resulting in liver hepatocytes dysfunction or damage.[5,12,13] This is thought to be caused by bacterial endotoxins or cytokines. The result is either a direct damage to hepatocytes, cholestasis, or both leading to hyperbilirubinemia. Studies on sepsis have shown the negative impact of *Eschierichia coli* and *Bacteroides fragilis* endotoxins on physiological bile flow in vivo.[10,14] These

Table 4 Sensitivities, specificities, PPV, and NPV of WCC, CRP, and bilirubin

	Sensitivity %	Specificity %	PPV %	NPV %
WCC	73.8	55.7	91.3	25.2
CRP	76.4	55.7	91.6	27.2
Bilirubin	45.9	80.3	93.7	19

CRP = C-reactive protein; NPV = negative predictive value; PPV = positive predictive value; WCC = white cell count.

Table 5 Sensitivity, specificity, PPV, and NPV after combining results of WCC and CRP, CRP and bilirubin, and WCC, CRP, and bilirubin

	WCC + CRP	CRP + bilirubin	WCC + CRP + bilirubin
Sensitivity %	57.3	38.3	31.1
Specificity %	73.8	91.8	95.1
PPV %	93.2	96.7	97.6
NPV %	21.4	19	17.6

CRP = C-reactive protein; NPV = negative predictive value; PPV = positive predictive value; WCC = white cell count.

studies have demonstrated bacterial-induced impairment of hepatocytes microcirculation and hepatocytes damage as seen on a rat liver model. Furthermore, endotoxins are shown to result in hemolysis, which then adds further increase in bilirubin levels.

Currently, there is a wealth of literature to support association of hyperbilirubinemia and appendicitis.[5,9,10,13,15–24] However, the majority of these studies have examined the value of elevated serum bilirubin in "perforated" appendicitis. Clinical signs in patients with perforated appendicitis are not subtle and although hyperbilirubinemia can be helpful it should not change or accelerate our management. In our retrospective case–controlled design, we found that this effect is significantly noted in patients with appendicitis and not only observed in the perforated appendicitis group.

We accept that our study population would have included some patients with Gilbert's syndrome. We believe, however, that this would only be a small number as Gilbert's syndrome is a rare disorder and affects between 3% and 10% of western European population.[25,26] Therefore, elevated serum bilirubin noted in over than 45% of our patients with appendicitis would be significant enough to indicate that this hyperbilirubinemia is rather secondary to appendicitis.

A recent systematic review published in the Scandinavian journal of surgery[27] concluded the need for further studies to evaluate predictive values of WCC, CRP, and bilirubin combined as diagnostic markers for acute appendicitis. In our study, we evaluated these makers as diagnostic tools in isolation as well as combined to ascertain the diagnostic accuracy of blood markers. Sensitivities of these blood markers after combining results remain poor with a sensitivity of 31.1% after combining WCC, CRP, and bilirubin. This would indicate that such tests are not very sensitive and are rather helpful to aid with the diagnosis, hence the very high specificities. Here, we established a high specificity of 95% and a high PPV approaching 98% of appendicitis after combining WCC, CRP, and bilirubin. Therefore, patients presenting with right iliac fossa pain with elevated levels of WCC, CRP, and bilirubin, their likelihood of having appendicitis is 98%.

A significant number of studies in the literature have examined the value of hyperbilirubinemia in the prediction of appendicular perforation. In a recent diagnostic meta-analysis study done by Giordano et al,[24] the authors who studied 5,000 patients concluded that hyperbilirubinemia alone is not a strong enough predictor of perforation. A study by Khan[5] showed that 86% of patient with appendicitis and its complications have developed hyperbilirubinemia. Another study published by Emmanuel et al in 2011[9] showed that hyperbilirubinemia is a significant marker for simple acute appendicitis and not only for appendiceal perforation. The authors have also found that elevated serum bilirubin had a high specificity of 88% and a PPV of 91% for simple acute appendicitis. These results are mirrored by the findings of our study, which showed almost identical results (specificity 80.3% and PPV 93.7%).

Our study has its limitations, allowing the retrospective nature of our study; it was not possible to identify the main pathology or diagnosis made in those patients who have normal appendix histology with raised inflammatory markers. It is, however, only acceptable to assume that a possible urologic or gynecologic pathology could have been the cause.

Conclusion

In conclusion, this study highlights the value of blood markers in the diagnosis of acute appendicitis. This is particularly important in patients with equivocal symptoms or in female patients with a differential diagnosis of a gynecologic pathology. Furthermore, it highlights the importance of hyperbilirubinemia in the diagnosis of appendicitis. The diagnosis of acute appendicitis, however, remains multifactorial and such tests simply help to guide the surgeon in the decision-making process. Patients presenting with elevated levels of serum bilirubin in the context of right iliac fossa pain warrant early surgical intervention.

Acknowledgment

The authors would like to thank Dr Amy Mitchell, MD, for her kind assistance in the statistical analysis of this study.

References

1. Cusheiri A, Grace PA, Darzi A, et al. Disorders of small intestine and vermiform appendix. In: Cuschieri A, Grace PA, Darzi A, et al.

editors. Clinical Surgery. 2nd ed. UK: Blackwell Publishing Ltd; 2003. p. 405.
2. Paterson – Brown Simon. Acute appendicitis. In: Paterson-Grown Simon, Ellis Brian W, Paterson-Brown Simon, editors. Hamilton Bailey's Emergency Surgery. 13th ed. London: Arnold; 2003. p. 399.
3. Smith PH. The diagnosis of appendicitis. Postgrad Med J 1965;41:2–5.
4. Guidry SP, Poole GV. The anatomy of appendicitis. Am Surg 1994;60: 68–71.
5. Khan S. Evaluation of hyperbilirubinemia in acute inflammation of appendix: a prospective study of 45 cases. Kathmandu Univ Med J (KUMJ) 2006;4:281–9.
6. Ohle R, O'Reilly F, O'Brien KK, et al. The Alvarado score for predicting acute appendicitis: a systematic review. BMC Med 2011;9:139.
7. Lintula H, Kokki H, Pulkkinen J, et al. Diagnostic score in acute appendicitis. Validation of a diagnostic score (Lintula score) for adults with suspected appendicitis. Langenbecks Arch Surg 2010;395:495–500.
8. Chong CF, Adi MI, Thien A, et al. Development of the RIPASA score: a new appendicitis scoring system for the diagnosis of acute appendicitis. Singapore Med J 2010;51:220–5.
9. Emmanuel A, Murchan P, Wilson I, et al. The value of hyperbilirubinaemia in the diagnosis of acute appendicitis. Ann R Coll Surg Engl 2011;93:213–7.
10. Sand M, Bechara FG, Holland-Letz T, et al. Diagnostic value of hyperbilirubinemia as a predictive factor for appendiceal perforation in acute appendicitis. Am J Surg 2009;198:193–8.
11. Ogilvie TA. Acholuric jaundice and appendicitis. Br J Surg 1951;38: 529–30.
12. Käser SA, Fankhauser G, Willi N, et al. C-reactive protein is superior to bilirubin for anticipation of perforation in acute appendicitis. Scand J Gastroenterol 2010;45:885–92.
13. Estrada JJ, Petrosyan M, Barnhart J, et al. Hyperbilirubinemia in appendicitis: a new predictor of perforation. J Gastrointest Surg 2007;11:714–8.
14. Utili R, Abernathy CO, Zimmerman HJ. Cholestatic effects of *Escherichia coli* endotoxin endotoxin on the isolated perfused rat liver. Gastroenterology 1976;70:248–53.
15. Dawes T, Burrows C. Abdominal pain and jaundice: appendiceal perforation an important differential. Emerg Med Australas 2007;19:276–8.
16. McGowan DR, Sims HM, Shaikh I, et al. The value of hyperbilirubinaemia in the diagnosis of acute appendicitis. Ann R Coll Surg Engl 2011;93:498.
17. Atahan K, Üreyen O, Aslan E, et al. Preoperative diagnostic role of hyperbilirubinaemia as a marker of appendix perforation. J Int Med Res 2011;39:609–18.
18. Ermich S, Kantor Z. [Jaundice in the course of acute appendicitis]. Pol Tyg Lek 1970;25:1583–4.
19. Seller RA. Jaundice in acute appendicitis. Lancet 1969;1:838.
20. Miller DF, Irvine RW. Jaundice in acute appendicitis. Lancet 1969;1: 321–3.
21. Orellana JM, Muñoz S. [Jaundice and appendiceal peritonitis (endotoxic bilirubinostasis)]. Rev Méd Chil 1982;110:233–5.
22. Beltran MA, Mendez PE, Barrera RE, et al. Is hyperbilirubinaemia in appendicitis a better predictor of perforation than C-reactive protein? - a prospective study. Indian J Surg 2009;71:265–72.
23. Hong YR, Chung CW, Kim JW, et al. Hyperbilirubinemia is a significant indicator for the severity of acute appendicitis. J Korean Soc Coloproctol 2012;28:247–52.
24. Giordano S, Pääkkönen M, Salminen P, et al. Elevated serum bilirubin in assessing the likelihood of perforation in acute appendicitis: a diagnostic meta-analysis. Int J Surg 2013;11:795–800.
25. Watson KJR, Gollan JL. Gilbert's syndrome. Baillières Clin Gastroenterol 1989;3:337–55.
26. Claridge LC, Armstrong MJ, Booth C, et al. Gilbert's syndrome. BMJ 2011;342:d2293.
27. Burcharth J, Pommergaard HC, Rosenberg J, et al. Hyperbilirubinemia as a predictor for appendiceal perforation: a systematic review. Scand J Surg 2013;102:55–60.