

Original article

Study of evaluation of role of total leucocyte count (TLC), absolute neutrophil count (ANC), neutrophil lymphocyte ratio (NLR) and ultrasound abdomen in patients with clinical diagnosis of acute appendicitis – A cross sectional study

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ABSTRACT:

Background: The diagnosis of acute appendicitis is essentially a clinical diagnosis where in timely intervention is absolutely important as early diagnosis can result in negative appendicectomies and late diagnosis results in complications. Various parameters were studied from time to time but none of which could unanimously be 100% correct.

Aim and Objectives: To diagnose acute appendicitis so as to reduce negative appendicectomies by including all parameters (TLC, ANC, NLR) in diagnosis and avoid delayed diagnosis to decrease the morbidity and mortality associated with complicated appendicitis.

Material and Methods: The study was conducted at Department of General Surgery at Bhagat Phool Singh Government Medical College for Women, Khanpur Kalan, Sonapat, Haryana. The study was done over a period of 18 months on 100 patients.

Results: Present study evaluated TLC, ANC and NLR parameters in 100 patients belonging to the age group between 15 and 65 years. All patients underwent emergency appendectomy. The blood investigations and ultrasound abdomen was done in the emergency setting. All reports were evaluated with respect to the histopathological results. Patients with co-morbidities were excluded. In the negative appendectomy cases (14%), ultrasound abdomen suggested appendicitis in 12 out of the 14 cases. The total leucocyte count was elevated in 8 out of the 14 cases, Absolute neutrophil count was elevated in 11 out of 14 cases whereas Neutrophil lymphocyte ratio was elevated in 13 out of 14 cases. Total leucocyte count was seen to be in normal range in 54 % of cases. But absolute neutrophil count was seen to be in higher range in 78 % of cases. Neutrophil lymphocyte ratio was elevated in 70% of cases. Ultrasound diagnosed 90 % of cases as acute appendicitis but histopathologically only 78% was found to have acute appendicitis.

Conclusion: All these results helped us to conclude that total leucocyte count is a not specific marker for predicting acute appendicitis, though absolute neutrophil count and neutrophil lymphocyte ratio are better markers for predicting acute appendicitis. Ultrasound of abdomen has 90% accuracy rate in predicting acute appendicitis.

Keywords: Total Leucocyte Count, Absolute Neutrophil Count, Neutrophil Lymphocyte Ratio, Ultrasound Abdomen,

INTRODUCTION

Abdominal pain is one of the commonest complaints encountered by a surgeon in the emergency. Acute appendicitis is the most common surgical cause of abdominal pain¹ It is estimated that as much as 6-7% of the general population will develop appendicitis during their life time, with incidence peaking in the second decade of life². The classical picture of appendicitis like fever, vomiting and right iliac fossa pain, tenderness and rebound tenderness may not be present in all cases.

Despite many diagnostic tools and scoring methods used to aid the clinical diagnosis of Acute Appendicitis such as Alvarado score and RIPASA score, negative appendectomy is somewhat acceptable traditionally even to the tune of 20-30 percent in literature³. However, with the availability of modern gadgets for investigation this rate of negative appendectomy doesn't seem plausible, though the lack of availability of these equipment in rural areas may limit their use.⁴

Leukocytosis has a sensitivity of 87.3% and specificity of 64.3 %, CRP has sensitivity of 70.4% and specificity of 64.3%, IL-6 has a sensitivity of 78% and specificity of 50%, IL -10 has a sensitivity of 40.8% and specificity of 92.9%⁵. USG alone has a sensitivity and specificity of 81% and 88% respectively, CT combined with USG has 96% sensitivity and 89% specificity⁶. None of the laboratory investigations or radiological investigations have been found to be 100% sensitive and specific for the diagnosis of acute appendicitis. A combination of various laboratory investigations such as elevated WBC count and positive CRP, IL-6 IL-10 has been used to diagnose appendicitis with 100 % sensitivity but with only 42.9 % specificity⁵. The total leukocyte count, absolute neutrophil count, neutrophil lymphocyte ratio are simple, quick, inexpensive, easy to interpret without causing any additional financial burden to the patient. The present study has been designed specifically with an aim to diagnose acute appendicitis so as to reduce negative appendicectomies by including all parameters in diagnosis and avoid delayed diagnosis to decrease the morbidity and mortality associated with complicated appendicitis. Ethical consideration definitely come to exist as far as negative appendicectomy is concerned, at the same time the future of negative appendicectomy in relation to medicolegal consideration is not clear.

MATERIALS AND METHODS

The present study was conducted at Department of General Surgery at Bhagat Phool Singh Government Medical College for Women, Khanpur Kalan, Sonapat, Haryana with written and informed consent of the participants. The study was done over a period of 18 months after ethical committee approval. The study was a cross sectional observational study conducted on 100 patients.

Patients with clinical features of acute appendicitis aged between 15 and 65 years were included in the study. Patients having age <15 years and > 65 years, having diabetes, hypertension or other immunocompromised patients, pregnant female and radiologically proven other diagnosis like ureteric colic, Meckels diverticulitis etc. were excluded from the study.

METHODOLOGY

Detailed history taking and clinical examination of abdomen of all patients of clinical diagnosis of acute appendicitis aged between 15 and 65. Venous blood sample was collected from each patient in

purple vacutainer at the time of admission under aseptic precautions. Samples will be send to central hematology laboratory and run on haematological analyser before hand for complete blood count and differential leukocyte count. Slide preparation of Peripheral blood smear with Leishmann staining was done and complete blood count, differential leukocyte count was obtained. In case of disputes, blood counts obtained from Peripheral blood count was considered final. Absolute Neutrophil count was obtained from evaluation of peripheral blood smear. The percentage of neutrophils divided by 100 multiplied to the total leukocyte count gives the Absolute Neutrophil count. The neutrophil Lymphocyte ratio was obtained by dividing the percentage of neutrophils to the percentage of lymphocytes from the data available from differential leukocyte count. All patients was subjected to ultrasound examination of abdomen.

STATISTICAL ANALYSIS

The collected data was analysed statistically by using statistical package SPSS ver. 20. Student 't' test / Kruskal wallis test was used. Percentage and proportion was calculated for qualitative data. Chi Square was used to find out the association for categorical data.

RESULTS

In this cross sectional observational study 100 patients were enrolled after fulfilling the inclusion criteria. Age group between 21-30 years had the maximum number of cases 36 (36%) where as age group age less than 20 years had the minimum number of participants 17(17%). The minimum age of the participants was 15 years and the maximum age was 64 years. The mean age of the study population was 32.65±13.48 years. 32 (32%) patients were of female gender and the remaining 68 (68%) cases were of male gender.

A total of 86 cases (86%) had histopathologically proven features of acute appendicitis. There were 4 cases (4%) of gangrenous appendix. Congestion of appendix (normal appendix) was seen in 14 cases (14%). 90 patients (90%) was diagnosed to have acute appendicitis by ultrasonography. Ultrasound could diagnose appendiceal perforation in 6 cases (6%). Probable appendicitis was given in 11 cases (11%) and normal appendix in 7 (7%) of cases. Appendix could not be visualised in 3 cases (3%). Out of the total 90 cases diagnosed as acute appendicitis by ultrasound only 78 cases were histopathologically proven appendicitis.

Table 1: Ultrasound abdomen and Histopathology reports

STATISTIC	VALUE	95% CI
Sensitivity	90.70%	82.49 % to 95.90%
Specificity	14.29%	1.78 % to 42.81%
Positive likelihood Ratio	1.06	0.85 to 1.32
Negative Likelihood Ratio	0.65	0.15 to 2.76
Positive Predictive Value	86.67%	83.86 to 89.05%
Negative Predictive Value	20.00%	5.58% to 51.41%
Accuracy	80.00%	70.82% to 87.33%

The chi-square statistic is 0.3322. The *p*-value is .564351

TLC was in the range of 4000 -11000 cells/mm³ there 54 cases (54%) and number of cases with total leucocyte count more than 11000cells /mm³ was 46 (46%). 48 patients out of the 86 histopathologically proven cases had leucocyte

count within a normal range of 4000-11000cells /mm³. 38 cases out of the 86 cases had a elevated leucocyte count. 6 out of the 14 cases of congestion of appendix had leucocyte in normal range.

Table 2: Total leucocyte count and histopathology report

STATISTIC	VALUE	95% CI
Sensitivity	44.19%	33.48% to 55.3%
Specificity	42.86%	17.66 % to 71.14%
Positive likelihood Ratio	0.77	0.46 to 1.29
Negative Likelihood Ratio	1.3	0.69to 2.45
Positive Predictive Value	82.61%	74% to 88.00%
Negative Predictive Value	11.11%	6.22% to 19.06%
Accuracy	44.00%	34.08% to 54.28%

The chi-square statistic is 0.8137. The *p*-value is .367025

Absolute neutrophil count (ANC) was divided into categories. There was 22 cases (22%) with ANC in normal range (between 2000-6000cells /mm³), and 78cases (78%) with ANC more than 6000 cells/mm³. Elevated ANC (>6000cells/mm³) was

seen in 63 cases of acute appendicitis and remaining 19 patients had normal ANC. All 4 patients with gangrenous appendix had elevated ANC. 11 out of 14 patients with congestion of appendix had elevated ANC.

Table 3: Absolute neutrophil count and histopathology report

STATISTIC	VALUE	95% CI
Sensitivity	77.91%	67.67 % to 86.14%
Specificity	76.60%	61.97% to 87.7%
Positive likelihood Ratio	3.33	1.96 to 5.65
Negative Likelihood Ratio	0.29	0.19to 0.44
Positive Predictive Value	85.90%	78.20 to 91.18%
Negative Predictive Value	65.45%	55.28% to 74.39%
Accuracy	77.44%	69.39% to 84.23%

The chi-square statistic is 0.0031. The *p*-value is .955615

Neutrophil lymphocyte ratio (NLR) categorised as number of cases with NLR between 0 -2.5, 2.5 to 5, 5 – 7.5 and 7.5 and above. There were 17 (17%), 63 (63%), 12 (12%), 8(8%) cases in each of these cases respectively. Out of the 86cases of acute

appendicitis 16 patients had NLR between 0-2.5 and remaining 70 patients had NLR > 2.5. 49 patients had NLR between 2.5 to 5, 9 patients with NLR between 5 to 7.5and 8 patients with NLR > 7.5.

Table 4: Neutrophil lymphocyte ratio and histopathology report

STATISTIC	VALUE	95% CI
Sensitivity	81.40%	71.55 % to 88.98%
Specificity	7.14%	0.18 % to 33.87%
Positive likelihood Ratio	0.88	0.73 to 1.05
Negative Likelihood Ratio	02.6	0.37 to 18.12
Positive Predictive Value	84.34%	81.86%to 86.54%
Negative Predictive Value	5.88%	0.89 % to 30.30%
Accuracy	71.00%	61.07 % to 79.64%

The chi-square statistic is 1.121. The *p*-value is .289704

The minimum value of TLC in this study was 4500cells/mm³ and maximum TLC was 18,000 cells/mm³ with a mean of 10,650 cells /mm³ and standard deviation of 2823.22. The minimum value of ANC was 2679 and maximum value was 15,120 cells /mm³ with a mean of 8033 cells /mm³ and standard deviation of 2624. The minimum value of NLR was found to be 1.25 and maximum value was 12.71 with a mean of 4.04 and standard deviation of 1.94.

NEGATIVE APPENDICECTOMY ANALYSIS

Out of the 100 cases 86 cases were histopathologically proven to be acute appendicitis, remaining 14 cases showed congestion of Appendix suggesting negative appendicectomy. Ultrasound diagnosed acute appendicitis in 12 cases in which histopathology showed congestion of appendix. TLC was elevated in 8 out of the 14 cases of negative appendicectomy. ANC was elevated in 11 out of 14 cases of negative appendicectomy. NLR was elevated in 13 out of 14 cases of negative appendicectomy. There was one case in which all the parameters were normal in the study and histopathology was positive for appendicitis.

DISCUSSION

Appendicectomy is one of the most common emergency surgeries performed by young Surgeons. Acute Appendicitis is the most common surgical cause of abdominal pain¹. Even with the availability of modern techniques Acute Appendicitis is essentially a clinical diagnosis, which can result in negative appendicectomies. The negative Appendicectomy rate is around 20-30% in literature³. The incidence of negative appendicectomy is higher in females than males and highest in women belonging to the child bearing age.

The current study is a cross sectional observational study to evaluate the role of total leucocyte count, absolute neutrophil count, neutrophil lymphocyte ratio and ultrasound

abdomen in patients with clinical diagnosis of acute appendicitis. Evaluating the above parameters in acute appendicitis is economically feasible, easily available in the emergency settings. In a developing nation like India where rural healthcare is of utmost importance, we conducted this study in our institute located in a rural area of state Haryana to evaluate the role in diagnosis of acute appendicitis.

This study was conducted in 100 patients belonging to the age group between 15-65 years who underwent appendicectomy. The total leucocyte count, absolute neutrophil count, neutrophil lymphocyte ratio was evaluated and ultrasound abdomen was done preoperatively. The histopathological report was followed up. No patients details was lost during the study. In this study there was 32 females (32%) and 68 males (68%). The minimum and maximum age was 15, 65 years respectively with a mean of 32.65 and a standard deviation of 13.48. The minimum total leucocyte count was 4500cells/mm³ and maximum value was 18,000cells/mm³ with a mean of 10,650 cells/mm³ and a standard deviation of 2823. The absolute neutrophil count varied from 2679 cells/mm³ to 15,120 cells/mm³ with a mean of 8033 and standard deviation of 2624. The neutrophil lymphocyte ratio varied from 1.25 to 12.71 with a mean of 4.04 and a standard deviation of 1.94. Ultrasound abdomen showed acute appendicitis in 90 cases (90%).

In a study by Hajibanded, a systematic review and meta analysis of neutrophil lymphocyte ratio to predict acute appendicitis and its severity, the NLR at 4.7 had sensitivity and specificity of 88.8 % and 90.9% respectively⁷. Another retrospective study by Sahin et al on 1067 patients showed that a NLR of 4.68 was associated with acute appendicitis, with a sensitivity and specificity of 65.3% and 54.7% respectively. A NLR of 5.74 was associated with complicated appendicitis⁸. In a

study by Salman et al in 2019, the sensitivity and specificity neutrophil lymphocyte ratio in diagnosing acute appendicitis was done as retrospective analysis. 372 patients were included in the study. The median age was 27 years and the cut off values for diagnosing acute appendicitis was 4.2. The sensitivity and specificity was 79.5 and 67% respectively.⁹

A retrospective cross sectional study of neutrophil to lymphocyte ratio in diagnosing acute appendicitis was done by Ahmad et al The NLR for acute Appendicitis was 3.11 and perforated appendix was 6.17. The sensitivity was 75.23%, 76.32% for NLR in acute appendicitis and perforated appendix respectively and specificity was 68.70%, 58.72% respectively. The study results were concluded that NLR can be used as reliable adjunct in diagnosing acute appendicitis⁴.

The current study also shows a sensitivity and specificity of 81% and specificity 07% at a cut off value of 2.5. The results obtained are similar to that available in literature. NLR alone cannot be used in the accurate diagnosis of acute appendicitis, by along with other parameters like total leucocyte count, absolute neutrophil count and ultrasound abdomen, it can be useful adjunct in diagnosing acute appendicitis and thereby preventing negative appendectomy. In a prospective study by Fatima et al, a combined diagnostic accuracy of total leucocyte count, neutrophil percentage and ultrasound abdomen was done in 2019-20. TLC > 11,000 cells/mm³ and neutrophil >75% were considered positive for appendicitis and in ultrasound non-compressible, blind-ending, non-peristaltic bowel loop originating from the cecum (appendix), loculated para-cecal collection, and/or the finding of an appendicolith was considered positive. The diagnostic accuracy of TLC >11,000/mm³ was 82.94%, with sensitivity 83.10% and specificity 82.14%. The accuracy of the neutrophil test was 88.82%, with sensitivity 88.03% and specificity 92.86%. The accuracy of ultrasound was 88.24% with sensitivity 89.44% and specificity 82.14%. cases having TLC >11,000/mm³, neutrophil count >75%, and ultrasonographic features showing non-compressible, blind ending loop, and diameter of the appendix > 7 mm were combined into one group. The combined accuracy was 94.71% with sensitivity and specificity was 94.71%, 97.18%, 82.14% respectively¹⁰. The results of ultrasound were similar in term of Sensitivity and diagnostic

accuracy with our current study. The results with respective to neutrophil count were comparable but the results of total leucocyte count were not similar.

A cross sectional observational study of total leucocyte count on predicting the degree of acute appendicitis was done by Bilal et al in 2019. The study included 238 patients. 198 patients (82.3%) had normal TLC range¹¹. The study concluded that contrary to the impression that a normal TLC rules out acute appendicitis. The results of the study was similar to the results of the current study. The age groups included in the study were comparable. Males were more in both the age groups. The median age was 27 years and in current study was 32 years. 54% of patients had normal TLC range who were histopathologically diagnosed as acute appendicitis.

The predictive value of leucocytosis in diagnosing acute appendicitis was done by Saleem et al in 2017. A total of 180 patients were included in the study. This study was similar to current study in gender distribution only. The total count was elevated in 129 patients (71.6%), which was higher than a current study which had elevated total counts only in 46 cases (46%). The difference in results could be due the inclusion of cases as leucocytosis as total count >10000cells/mm³ when compared to the current study in which >11000cells/mm³ is considered as leucocytosis. The study also included patients aged >10years also in inclusion criteria²³.

A systematic review and meta analysis on the role of ultrasound in appendicitis was done by Giljaca et al. 17 studies were combined which included 2871 participants from a time duration from 1994 to 2014 by two authors independently. The Sensitivity and Specificity of ultrasound was 69 % and 81 % respectively corresponding to 95% confidence intervals¹³. In the Current Study ultrasound had high sensitivity of 90 % but a lower Specificity of 14% . The value of Ultrasound for diagnosis of acute appendicitis was done as a descriptive prospective study by Benedetto et al from April 2015 to April 2016. The sensitivity and specificity was 90% and specificity was 100 %¹⁴. The Sensitivity of Ultrasound was similar to that of our current study.

CONCLUSION

All these results helped us to conclude that total leucocyte count is a not specific marker for predicting acute appendicitis, though absolute neutrophil count and neutrophil lymphocyte ratio

are better markers for predicting acute appendicitis. Ultrasound of abdomen has 90% accuracy rate in predicting acute appendicitis. None of the investigations could single handedly predict the presence of early appendicitis, a combination of all parameters together can reduce negative appendectomy rate. Acute Appendicitis is

essentially a clinical diagnosis and the parameters studied can aid the clinical diagnosis. A combination of the above parameters and its careful interpretation would enable the surgeon to accurately diagnose acute appendicitis thereby preventing negative appendectomy and wastage of national resources.

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