RESEARCH ARTICLE

A STUDY OF MAST CELLS IN SURGICALLY RESECTED APPENDIX.

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Abstract

Appendicitis is one of the common conditions requiring emergency surgery. The exact etiopathogenesis is poorly understood in those appendices which are resected for suspected appendicitis and are classified as normal by histopathology on conventional staining. Acute appendicitis is thought to be initiated by progressive increases in intraluminal pressure that compromise venous outflow. However, faecolith are not found in many cases in which acute appendicitis is confirmed. The present study is a study of mast cells in surgically resected appendix, which includes 150 cases of acute appendicitis, chronic appendicitis, histology negative acute appendicitis, and appendices removed in the course of surgeries done for some other disease/control, who attended D. Y. Patil Hospital and Research centre, Kolhapur. This is a prospective study undertaken from May 2014 to April 2016. The results of this study show that acute appendicitis was the commonest affliction (44.65%) with chronic appendicitis being the second (28%) and histology negative acute appendicitis also sharing the large number of cases at 24%. Mean mast cell count was highest in chronic appendicitis followed by acute appendicitis and by histology negative acute appendicitis and least in appendices removed in the course of surgeries done for some other disease. Therefore, the type I hypersensitivity reaction with release of mediators by mast cells might be a predisposing factor for the sequence of events leading to appendicitis.

Introduction:

In about 20–25% of appendicectomies performed for clinically suspected acute appendicitis, definite morphological changes are lacking on histopathological examination.1

Since appendectomy relieves pain, an unknown pathology is likely to exist.2 The role of inflammatory reactions involving local endocrine cells and neuroproliferation in causing repeated attacks of pain has been described and several staining techniques including immunohistochemistry have been tried in detecting the pro-inflammatory mediators.3 Histopathological examination still remains the gold standard method for the confirmation of the appendicitis.4 Acute appendicitis is thought to be initiated by progressive increases in intraluminal pressure that compromise venous outflow.5 Currently, luminal obstruction due to faecoliths or less commonly submucosal lymphoid hyperplasia especially in children appears to initiate acute inflammation in appendix.6 However, faecolith
are not found in many cases in which acute appendicitis is confirmed. It has also been suggested that obstruction is not an important causative factor in acute appendicitis, but may develop as a result of inflammatory process. Mast cells are constantly present in appendix, the organ most commonly subjected to surgical intervention and removal. Therefore, the type I hypersensitivity reaction with release of mediators by mast cells might be a predisposing factor for the sequence of events leading to appendicitis.

**Methodology:**
The present study is a study of mast cells in surgically resected appendix, which includes 150 cases of acute appendicitis, chronic appendicitis, histology negative acute appendicitis (HNAA), and appendices removed in the course of surgeries done for some other disease/control, who attended D. Y. Patil Hospital and Research centre, Kolhapur. This is a prospective study undertaken in department of Pathology, D. Y. Patil Medical College, Hospital and Research centre, Kolhapur, from May 2014 to April 2016. Haematoxylin and Eosin stained sections examined microscopically and histologic interpretation was done. Toluidine blue stained sections were examined and the mast cell counts were done in 20 non-overlapping high power fields (HPF). The cell counts were done on 20 HPF, the mean cell counts / mm² was calculated to be number of cells in 20 HPF / 3.

On the basis of observations an attempt was made to study distribution pattern of mast cells in relation to various appendicular lesions.

**Results:**
In the present study out of 150 cases, the clinical diagnosis of acute appendicitis was seen in 70.66 % of the total cases, while on histopathological diagnosis only 44.65 % of the cases were those of acute appendicitis including classic acute appendicitis, acute suppurative, perforative and eosinophilic appendicitis. They were followed by 28% of the cases of chronic appendicitis, 24% of Histology negative acute appendicitis (HNAA), and 3.33% of the cases were of appendices removed in the course of surgeries done for some other disease/Control. Patent lumen was seen in 76.66 % of the cases, while obstruction due to fecolith was demonstrated in only 14% of the cases.

**Table no.1:** Age and sex distribution of various appendicular lesions.

| AGE GROUP(yrs) | MALES | FEMALES | TOTAL | PERCENTAGE(%) |
|----------------|-------|---------|-------|---------------|
| 0-10           | 4     | 3       | 7     | 4.66          |
| 11-20          | 25    | 23      | 48    | 32            |
| 21-30          | 38    | 24      | 62    | 41.33         |
| 31-40          | 6     | 9       | 15    | 10            |
| 41-50          | 6     | 3       | 9     | 6             |
| 51-60          | 6     | -       | 6     | 4             |
| >60            | -     | 3       | 3     | 2             |
|                | 85    | 65      | 150   | 100           |

**Table no 2:- Mean mast cell count in various appendicular lesions.**

| HISTOPATHOLOGICAL DIAGNOSIS | NO. OF CASES(n=150) | Mast cell count/mm² | Submucosal mast cell count | Mast cell Count in muscularis propria |
|----------------------------|---------------------|---------------------|-----------------------------|--------------------------------------|
|                            | Range | Mean±SD | Range | Mean±SD | Range | Mean±SD |
| Classic acute appendicitis | 46    | 7-19 | 10±3 | 8-26 | 12.5±3.72 | 7-28 | 14.12±3.8 |
| Acute suppurative appendicitis | 14 | 8-20 | 11.785±3.66 | 9-26 | 14±4.41 | 11-28 | 16.21±4.3 |
| Acute perforative          | 04    | 11-17 | 14.75±3.03 | 14-26 | 18.5±4.7 | 12-23 | 16.75±4.0 |
appendicitis

| Diagnosis                                             | Age (Years) | Mean mast cell count in mucosa | Mean mast cell count in submucosa |
|-------------------------------------------------------|-------------|--------------------------------|----------------------------------|
| Acute eosinophilic appendicitis                       | 03          | 13.66±1.7                      | 16±21                            |
| Chronic appendicitis                                  | 42          | 11.26±3.80                     | 11.78±3.40                       |
| Histology negative acute appendicitis (HNAA)          | 36          | 6.22±3.9                       | 8.6±2                            |
| Appendices removed in the course of surgeries done for some other disease/Control | 05          | 3.8±1.7                        | 5±1.4                            |

Table no.3: Mean mast cell count in various age groups

| Age (in years) | Histopathological diagnosis | Mean mast cell count in mucosa | Mean mast cell count in submucosa |
|----------------|-------------------------------|--------------------------------|----------------------------------|
| <15 Years      | Classic acute appendicitis    | 12.33±2.88                     |                                   |
|                | Acute suppurative appendicitis| 14.3±4.6                       |                                   |
|                | Acute eosinophilic appendicitis| 17±3                           |                                   |
|                | Acute perforative appendicitis| 22±4                           |                                   |
|                | Chronic appendicitis          | 17.24±3.5                      |                                   |
|                | HNAA group                    | 8.8±1.4                        |                                   |
|                | Control group                 | -                              |                                   |

| Age (in years) | Histopathological diagnosis | Mean mast cell count in mucosa | Mean mast cell count in submucosa |
|----------------|-------------------------------|--------------------------------|----------------------------------|
| 15 to 45       | Classic acute appendicitis    | 11.6±3.2                       | 14.5±3.9                         |
|                | Acute suppurative appendicitis| 14.6±1.7                       | 14.66±1.7                        |
|                | Acute eosinophilic appendicitis| 17±0                           | 17±0                             |
|                | Acute perforative appendicitis| 15±2                           | 20.5±5.5                         |
|                | Chronic appendicitis          | 17.7±4.08                      | 16.41±4.1                        |
|                | HNAA group                    | 9.52±2.52                      | 9.19±2.1                         |
|                | Control group                 | 5.25±1.9                       | 4.7±1.4                          |

| Age (in years) | Histopathological diagnosis | Mean mast cell count in mucosa | Mean mast cell count in submucosa |
|----------------|-------------------------------|--------------------------------|----------------------------------|
| >45            | Classic acute appendicitis    | 11±3.2                         | 13.5±4                           |
|                | Acute suppurative appendicitis| 14.5±3.9                       | 17±4                             |
|                | Acute eosinophilic appendicitis| 17±0                           | 15±0                             |
|                | Acute perforative appendicitis| 13.5±2.5                       | 16.5±2.5                         |
|                | Chronic appendicitis          | 19.3±1.32                      | 17.72±3.2                        |
|                | HNAA group                    | 8.8±1.85                       | 8.9±2.7                          |
|                | Control group                 | 5±0                            | 7±0                              |

Table no.4: Mean mast cell count according to sex.
Figure no.1: Photomicrograph showing mast cells in the mucosa of appendix (40X, Toluidine blue)

Figure no.2: Photomicrograph showing mast cells in submucosa of appendix (Toluidine blue stain 40X)
Discussion:
The results of this study show that acute appendicitis was the commonest affliction (44.65%) with chronic appendicitis being the second (28%) and histology negative acute appendicitis (HNAA) sharing the large number of cases at 24%. It can be concluded that most of the studies including those done by Samsi A.B et al\(^8\), Jess P et al\(^9\), Butler C\(^10\), Crabbe MM et.al\(^11\), Blair NP\(^12\), Andiran F et al\(^13\), Duzgun AP et al\(^14\), Mysorekar VV et al, and Sharma S et al\(^15\) report a slight male predominance, which is found in the present study (1.30:1) as well.

Mean mast cell count was highest in chronic appendicitis followed by acute appendicitis, and histology negative acute appendicitis (HNAA) and least in appendices removed in the course of surgeries done for some other disease which correlated with the studies done by Shah AA et al\(^16\), Kolur A et al\(^6\), Singh U.R et al\(^17\), Coskun N et al\(^18\), Nagaraj G et al\(^19\), Kumaran C et al\(^20\), Yang Z et al\(^21\). Increase in mean mast cell count was also highest in chronic appendicitis followed by acute appendicitis in studies done by Sonti S\(^22\) and Naik R\(^23\) which correlated with our study.

In acute appendicitis, mean mast cell count was higher in submucosa and muscularis propria when compared with mucosa of appendix which correlated with the studies done by Shah AA et al\(^16\), Kolur A et al\(^6\) and, Kumaran C et al\(^20\).

In chronic appendicitis mean mast cell count was highest in mucosa followed by muscularis propria and submucosa which correlated with the studies done by Shah AA et al\(^16\), Kolur A et al\(^6\) and Sonti S\(^22\).

In Histology negative acute appendicitis (HNAA) mean mast cell was highest in mucosa followed by muscularis propria and submucosa which correlated with the studies done by Sonti S\(^22\), Naik R\(^23\) and Yang Z et al\(^21\).

In Appendices removed in the course of surgeries done for some other disease, mast cell counts were almost equal in mucosa, submucosa and muscularis propria.

The mean mast cell count was found to be increased in areas of mild fibrosis especially in mucosa, whereas in areas where degree of fibrosis was more the mean mast cell count was less in when compared with areas of early fibrosis. Mast cells, seen in all lesions of appendix were more easily identified in submucosa.
In submucosa, mast cells were larger, spindle to polygonal in shape and had coarse numerous granules as compared to mast cells in mucosa.

An increase in the mast cell clustering was seen in the vicinity of the blood vessels and nerve fibres.

No correlation was found between mast cells, age and sex of the patient.

When compared with mean mast cell count in all the appendicular lesions in our study there is corresponding increase in mean eosinophilic count which correlates with the studies done by Kolur A et al\(^6\) and Shrestha R et al\(^24\).

Mast cell count was found to be higher in the areas of mild fibrosis indicating the growth interaction between the mast cells and fibroblasts in areas of fibrosis.

**Conclusion:**

Toluidine staining technique is easy, simple, long lasting and gives good result for identification of mast cells in appendix.

Mast cell counts were lowest in appendices removed in the course of surgeries done for some other disease/Control, while they were significantly higher in histology negative acute appendicitis (HNAA) group, acute appendicitis and highest in chronic appendicitis. Also obstruction due to faecolith could not be demonstrated in many cases of appendicitis.

Increased eosinophil count with an increase in mast cell count observed in the present study in appendicitis may be due to the consequence of mediators released by mast cells such as eotaxin.

Therefore, the type I hypersensitivity reaction with release of mediators by mast cells might be a predisposing factor for the sequence of events leading to appendicitis.

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