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Histological and Microbiological Assessment of the Role of Micro Organisms in Chronic Anal Fistula

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Fistula in ano is one of the commonest ailments pertaining to the ano-rectal area and said to arise from crypto glandular infection of the anal gland, which lies within the intersphincteric space.

Infection would seem to be an integral part of the pathogenesis of fistula in ano. The microbiology of acute anorectal sepsis is well documented, but the studies on chronic anal fistula have continuously raised doubts about the relevance of infection in fistula persistence. The present histological study complements the earlier work, which used microbiological methods alone where important organisms might have been overlooked.

Granulation tissue from twenty patients with non specific fistula in ano was processed within 4h of medicated seton (*Kshara sutra*) therapy. Three samples from the intersphincteric part of the fistula were obtained. Two samples were studied microbiologically. The pus smear was taken from the fistulous opening by sterile cotton swab and sent to the Department of Microbiology, in a sterile container, and pus culture was done. Pus swab was inoculated on nutrient agar medium. The plated media were incubated at 37 C and examined at 48, hours. Smears from colonies that grew on the nutrient agar media were stained with Gram-stain. Gram-positive organisms and Gram-negative organisms were identified by conventional biochemical techniques. The third sample was fixed in 10 per cent formal saline for histological processing. Multiple 4 µm paraffin sections were stained using haematoxylin and eosin, gram, cresyl fast violet, periodic acid – Schiff and Ziehl – Neelsen stains.

The results show the incidence of fistula in ano and the origin of the predominant microorganism present in ano rectal fistula. These were investigated using 100 pus samples obtained from the 20 patients. Isolates of *Staphyloccus aureus*, streptococus Spp and *Corynebactrium* spp. were identified as skin derived organisms. Isolates of *Enterococcus* spp., *Escherichia coli*, *Bacteriodes* spp were considered gastro intestinal tract derived organisms. No mycobacterium species was grown from any of the eight specimens.

Sections of tissue from all twenty specimens showed a similar pattern of intense active chronic inflammatory change characterized by a large number of plasma cells, scattered multinucleate foreign-body giant cells and prominent vascular proliferation. Acute inflammatory change (of variable degree) was superimposed. No granulomas were seen in any specimen. Vegetable matter was demonstrated on the granulation tissue surface in one specimen only and bacteria were seen on histological examination and subsequent Gram staining. Relatively large numbers of organisms were grown from specimen. Stains for Helicobacter species, mycobacterium tuberculosis and fungi were negative in all specimens.

The present study has confirmed the relative paucity of organisms and demonstrates through micro biological study but the histological examination does not reveal many organisms. In fact, i9t is difficult to obtain a detailed picture through microbiological examinations. But details of the inflammatory changes can be witnessed through histological examination. In other circumstances, histological examination has been used to demonstrate important pathogens such as tuberculosis, Mycobacterium leprae and Helicobacter Pylori.