

Research Article**Isolation and Identification of *Candida Species* in Endoscopic Specimens of the Patients with Peptic Ulcer****Rajkumar Narasinghan^{1*}, D. Senthil Pragash², Uma Maheswari Kuppusamy³, Gnanasoory⁴, Kalaiarasan⁴**¹Assistant Professor, Department of Microbiology Dhanalakshmi Srinivasan Medical College Perambalur, Tamilnadu²Assistant Professor, Department of Microbiology, Melmaruvathur Adhiparasakthi institute of medical sciences and Research Melmaruvathur, Tamilnadu³Doctors Diagnostic Centre, Tiruchirapalli, Tamilnadu.⁴Senior Microbiologist, Frontline Hospital. Tiruchirapalli, Tamilnadu.***Corresponding author**

Dr. Rajkumar Narasinghan

Email: drnknmd1972@gmail.com

Abstract: Gastrointestinal tract (GI tract) of humans has inner lining which is known as mucous membrane. These inner lining of GI tract has major role in saving the stomach from acids present in the stomach. It prevents injury to the GI tract by coating the walls. If the mucous layer of GI tract gets damaged then acid can damage the GI tract. The *Candida* is yeast like fungi. The yeasts are spherical, oval or elongated and reproduce by budding. *Candida albicans* is normally present in the mouth, intestine and vagina. *Candida albicans* is a frequent component of the human indigenous microbial flora of the gastrointestinal (GI) tract of apparently healthy individuals. *C. albicans* can increase in numbers when this balance is disturbed to cause candidiasis of the intestinal tract, or yeast infections of other parts of the body. *C. albicans* causes thrush. The aim of the present study is to analyze the various fungi present in endoscopic specimens of acid peptic disorder patients. Eighty samples were collected from the patients who are suffering from signs and symptoms of peptic ulcer disorders. The samples were collected from both male and female patients with age varying from 21 years to 50 years. Samples were processed for fungal etiology. The direct smear Grams stain results showed a positivity of 8.75 % for fungus. All the 7 fungi isolated belong to the *Candida species*. The present study concluded that along with other bacterial isolates, *Candida species* are also present in the endoscopic specimens.**Keywords:** Peptic Ulcer, *Candida albicans*, Pseudo mycelium

INTRODUCTION

Gastrointestinal tract (GI tract) of humans has inner lining which is known as mucous membrane. It prevents injury to the GI tract by coating the walls. It acts as a barrier between the GI tract and acid. Peptic ulcers are defects in the GI mucosa that extend through the muscularis mucosa. Peptic ulcers occur in presence of acid & pepsin. The two main types are Gastric and Duodenal ulcers. They can also occur at gastro-jejunal stoma & lower end of esophagus with ectopic gastric mucosa. According to duration and complexity of symptoms, they can be divided into acute peptic ulcers and chronic peptic ulcers[1]. Acute Peptic Ulcers disruption of gastric mucosal barrier appears as multiple erosions. Classically they present with hemorrhage and are frequently multiple. They can occur in any part of stomach. These acute lesions can progress to chronic ulcers. Chronic peptic ulcers occur in alkaline mucosa & invade muscle coats. Fibrosis from recurrent ulcers causes deformities. It is nearly always situated in first part of duodenum. Anterior ulcer carries risk of perforation while posterior ulcer may cause hemorrhage

due to erosion of gastro duodenal artery[2]. There are several fungal species in nature and they cause many diseases in man, but 90% of the infections are caused by *Candida*. The *Candida* is spherical, oval or elongated yeast like fungi and reproduces by budding. The yeasts which produce pseudo mycelium are usually called Yeast like fungi. *Candida albicans* is a frequent component of the human indigenous microbial flora of the gastrointestinal (GI) tract of apparently healthy individuals[3].

MATERIALS AND METHODS

The study was carried out for a period of six months extending between January 2008 to June 2008. A total number of 80 endoscopic specimens were collected from Endoscopic clinic, Department of General Surgery, Rajah Muthiah Medical College and Hospital. The samples were collected from the patients attending the General Surgery Outpatient department with signs and symptoms of acid peptic disorders. The samples were collected from both male and female patients with age varying from 21 years to 50 years.

Sample Collection

Clinically suspected cases of acid peptic disorder patients were properly instructed for the endoscopic procedure by the clinical co-coordinator on the day before the procedure. With all standard emergency equipments and relevant clinical staff members, the patients underwent endoscopic examination, which was done by an expert Endoscopic Surgeon. Suitable cases with lesions were identified and the endoscopic specimens were collected for microbiological studies in specific containers. For microbiological study, the specimens collected were transferred aseptically to a sterile screw capped bottle with Brain Heart Infusion Broth, labeled properly and transported in ice-box immediately to laboratory without delay. In microbiological laboratory, the samples were immediately processed by standard microbiological methods for isolation and identification of fungal organisms[4].

Method of Analysis

Gram Straining was done on the specimen collected. Grams stained film was examined under oil immersion (100 X objective). Presence of Gram-positive budding yeasts with or without pseudohyphae, and number of yeast cells were noted and recorded. Fragments of pseudo and true mycelium which are characteristically pinched off at septations are also seen. Fungal culture was done with a small piece of the endoscopic specimen collected was inoculated in two sets of Sabouraud's Dextrose Agar (one set of plate containing antibacterial antibiotic and another set of plate without antibiotic). One set of plates were incubated at 25°C on BOD incubation and another set of plates were incubated at 37°C for three weeks. Every day the plates were examined for the fungal growth. The emerged fungal growth were examined further and recorded. The fungal colonies grow in 3-4 days and appear as cream colored, smooth and pasty growth. The plates failed to give any fungal growth even after 21 days of incubation, they were discarded. The isolated *Candida* species was treated with serum and further incubated at 37°C for 2 to 4 hrs. A drop of suspension was examined under microscope. Long tube like projections, with no constriction at the point of attachment to the yeast cell, were seen (Germ tube test).

RESULTS

A total of 80 Endoscopic samples were collected from patients with various acid peptic disorder conditions, who attended the Department of General Surgery, Raja Muthiah Medical College and Hospital. The specimens were collected from various age groups of both male and female patients given in Table 1.

Table 1 shows the age wise distribution of male and female patients

Age group (in years)	Number of male patients	Number of female patients	Total
21-30	11	7	18
31-40	21	14	35
41-50	16	11	27
TOTAL	48	32	80

Legend: 1 the endoscopic specimens were collected from patients suffering from gastritis, gastric ulcer, duodenal ulcer, the highest prevalence of symptomatic infection was found in the age group of 31-40 yrs (43.75%). It was least among the age group of 21-30 yrs (22.5%). All the 80 Endoscopic specimens were processed for the presence of bacterial and fungal agents using standard diagnostic methods.

Table-2: Grams stain result direct microscopic examination

S. No	Microbes present	No of positive specimens (n = 80)	Positivity percentage
1.	Bacteria	48	60%
2.	Fungus	07	8.75%

Legend : 2 Among the 80 endoscopic specimens screened for bacterial and fungal etiology, the direct smear - grams stain result showed 48 samples were positive for bacteria and 7 samples were positive for fungus as shown in TABLE II.

DISCUSSION

Peptic ulcer is a group of disorders which happen in upper gastro-intestinal tract in which mucus layer is damaged by the action of acid and other secretions of the stomach. It is characterized by inflammation of the lining of the stomach that leads to stomach ulcers. Fungi found to be associated with the peptic ulcer infection is often found to live in the gastrointestinal tract. The thrush in the mouth spreads down to the esophagus and causes candidial oesophagitis, which leads to dysphagia and sometimes fever, may occur if there is further spread[4, 5]. Present study validates this point by the isolation of 8 *Candida* species out of 80 specimens collected from the acid peptic disorder patients. Hence in this perspective, this study has been taken out to find out the presence of fungal species in the endoscopic specimens from acid peptic disorders, and if present to isolate them and to find out the incidence of the same. One condition which favours growth of fungal species in the gastrointestinal tract is the lower range of acid pH between 5 to 6. This is produced by irrational use of antacids and antibiotics which leads to loss of normal flora which includes the acid producing

enterobacteriaceae group of organisms. Cohen R *et al* in 1994 stated that the presence of chronic infection of various etiologies, or frequent recurrent gastro-intestinal infections of various etiologies which leads to frequent use of antibiotics. This leads to gastrointestinal mucosal damage by both various organisms and adverse effects of medications. This also leads to mild suppression or alteration of immune mechanism and altered normal gastrointestinal flora[7]. DuPont B *et al* in 1992 found that these patients, those who have found to have signs and the symptoms of acid peptic disorders that were discussed above, were included. This chronic nature of disease combined with medications has led to neutropenia, which is associated with an increased rate of *Candida* infections[8]. In the present study, out of 80 endoscopic specimens collected and processed 7 specimens showed positive for fungal culture growth by the presence of creamy white colony on SDA. The direct smear and gram stain result showed either budding yeast cells or with large septate hyphae in 7 out of 80 specimens collected in this study. This indicates the association of *Candida* with acid peptic disorders and the presence of pseudohyphae indicates the diseased status of the patients and the involvement of *Candida* in ulcer patients. Of the 80 specimens subjected to direct microscopic examination by Grams stain method, 7 specimens showed the presence of gram positive budding yeast cells adherent to the gastro-intestinal epithelial cells with or without septate hyphae. The yeast cell adherence may be due to either active infection status of the patients or the heavy colonization of the *Candida* in the epithelial cells. According to Rosenberg S W *et al* in 1997. *Candida albicans* is the frequent fungal component of the human gastrointestinal tract. This point has been substantiated by our study, where 7 specimens (8.75%) yielded positive significant fungal growth on SDA. Since *Candida* exists as one of the common flora, the interpretation of Candidal culture growth gains importance because the interpretation should correlate with confirmation of the diseased condition[9]. Our Study results showed that the *Candida albicans* was the predominant fungal species isolated from the patients who attended the endoscopic clinic. This study conducted by Varrtivarian *et al* in 1993 also reveals that the fungal infection of *Candida* species is prevalent in the patients with severe stomach and abdominal pain and also seems to be more prevalent in the patients with chronic acid peptic disorders[10].

CONCLUSION

The present study concluded that along with other bacterial isolates, *Candida* species are also present in the endoscopic specimens. Further the results revealed that the presence of *Candida* species in acid peptic disorders is an alarming level. Therefore, to study the patho-physiological role of *Candida* species in acid peptic disorders, it should be studied elaborately to rule out their pathogenic involvement.

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