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# Short Communication Comparative evaluation of gastric biopsies for detection of Helicobacter pylori

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ARTICLE INFO	A B S T R A C T
Article history: Received 13-11-2019 Accepted 18-11-2019 Available online 07-01-2020	<ul> <li>Helicobacter pylori earlier known as Campylobacter pylori is a gram negative bacterias that colonizes the gastric epithelium of Humans.</li> <li>Helicobacter pylori is considered as the most successful pathogen as more than 50% population have Helicobacter pylori in their upper Gastrointestinal tract.</li> <li>It is usually observed that 80% of the infected population is asymptomatic to the bacterium. The infection of Helicobacter pylori is more common in developing countries like India than developed western countries. Helicobacter pylori is a major cause of gastro-duodenal injury and is also related to gastric cancer, thus detection of the bacterium is must for proper treatment.</li> <li>This paper is a comparative evaluation of traditional stains used for the diagnosis of H. pylori. Main focus is laid on special stains such as Giemsa stain, toluidine blue and modified H&amp;E staining method.</li> <li>The most common way of detecting Helicobacter pylori is by histopathological examination in which biopsies from stomach region (Body, fundus and antrum) are examined.</li> </ul>
<i>Keywords:</i> Helicobacter pylori diagnosis histopathology GI Biopsies tissue staining	
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## 1. Introduction

*Helicobacter pylor*i is a gram negative spiral shaped bacteria which is usually found in the mucosa layer of the stomach. At places, the mucosa of the stomach extends down to the sub-mucosa to form gastric glands. These gastric glands contain a variety of cells responsible for acidity in the stomach. *Helicobacter pylor*i is usually found in the lumen of the gastric pits.<sup>1–3</sup>

*Helicobacter pylori* is reported to have strong relationship with chronic gastritis, peptic ulcers and in some cases of gastric extrazonal marginal zone lymphoma.<sup>4</sup>

A number of tests can be performed for detection of *Helicobacter pylori* such as haematoxylin and eosin stain coupled with special stains such as toluidine blue and giemsa stain.<sup>2</sup> Often these methods are used in government hospital.

The detection of *Helicobacter pylori* is difficult due to many possible factors such as density and distribution of bacteria in the mucosa layer.

Recent studies suggest that action of antibiotics can lead to decrease in the density of the bacteria. While Consumption of proton pump inhibitor leads to the migration of bacteria to body (of the stomach) when the biopsies are taken only from antrum region.<sup>2</sup>

These factors can give rise to false reading. Usually these problems are resolved by taking multiple gastric biopsies i.e. from fundus, antrum and body regions (after careful clinical correlation).

These biopsies were fixed using 10% buffered formalin, carefully grossed and then processed. Tissue block were formed using paraffin wax. The blocks were then subjected to microtome and slides were formed.

Slides are the deparaffinised and stained with different stains such as H&E stain, giemsa stain etc.

India is a developing country, the medical facilities in rural areas of India are limited. The main purpose of this article is to enlighten different practical ways of diagnosing *Helicobacter pylori* using the resources available.

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#### 2. Materials and Methods

A number of patients underwent upper gastrointestinal endoscopy out of which a major portion of patients were suffering from gastritis. Gastric biopsies were collected (from patients suffering from gastritis). The biopsies were from body, fundus and antrum region of stomach. In some cases, gastric ulcers were also taken.



Fig. 1:

All biopsies were formalin fixed, processed and were embedded in paraffin wax. The sections of biopsies were taken on a slide.

These sections were subjected to different stains such as Toluidine blue stain, Giemsa stain, Modified Haematoxylin and eosin stain and likewise a comparative study was done on human gastric biopsies.

In cases of Modified Haematoxylin and eosin stain, the eosin was modified. 1% (w/v) solution of Eosin Y was made using distilled water. A small amount of concentrated Acetic acid (200 microliter per 100 ml) is added to the solution. The solution is used for staining the slides (same as the normal solution) but the time of staining is doubled.

In case of Giemsa stain, a 2% (w/v) solution of giemsa powder is prepared in distilled water. Once the solution is prepared, it is used to stain the slides.

In case of Toulidine blue stain, a stock solution is prepared by mixing 2 gram of toluidine blue powder in 100 mL of distilled water. The working solution of toluidine blue is prepared by mixing 4ml of stock solution with 96 ml of water.

Standard protocols of staining were followed.

### 3. Conclusion and Result

Upon evaluation, the results were same for all the stains. Giemsa was taken as a gold standard.

The modified haematoxylin and eosin method showed co mparable result (as compared to Giemsa stain and toluidine blue) owing to the small amount of acetic acid. The bacteria was stained reddish-pink by eosin stain.

The intensity of the eosin stain was increased by acetic acid helping in getting a good contrast.

From the data, we can conclude that, all the three stains (Modified Haematoxylin and eosin, toluidine blue and giemsa stain) can be used for the diagnosis of *Helicobacter pylori*.

One of the three stains can be used depending upon the availability, hence promoting an idea of Sustainable development; development using best resources available.

#### 4. Illustrations

In the picture we can see *Helicobacter pylori* in the lumen of gastric glands, top right and left shows the Modified Haematoxylin and eosin stain, while bottom left depicts Toluidine blue and bottom right depicts *Giemsa stain*.

### 5. Conflict of interest

None

#### 6. Source of funding

None

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