Usefulness of Cytokeratin CK7 and CK20 in the Diagnosis of Barrett’s Esophagus (BE)


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Introduction:

Diagnosis of BE is hinged on the presence of intestinal metaplasia (IM) and inflammation in the lower esophagus. However IM can occur in the cardia and lower esophagus following reflux and helicobacter infection. The special stain for demonstrating intestinal metaplasia cannot distinguish IM in BE and that in cardia or lower esophagus due to other causes(1). Quite often biopsies labeled lower esophagus by the endoscopist may in fact be from the cardia especially in cases where the gastro-esophageal junction is indistinct.

Distinguishing BE from mimics in the lower esophagus and cardia is very essential. BE is a recognized major risk factor for adenocarcinoma of the esophagus(2,3), therefore accurate diagnosis is imperative. Management of such a lesion is entirely different as it is regarded as a pre-malignant lesion.

Ormsby et al(4) seemed to have solved this diagnostic dilemma by using antibodies to cytokeratin CK7 and CK 20 staining pattern in the esophagus and stomach. They found that the staining pattern of gastric intestinal metaplasia is totally different from that of Barrett’s epithelium. However El Zimaity and Graham(5) in their study could not reproduce such high specificity and sensitivity of Barrett’s Esophagus CK7/20 pattern in the esophagus and stomach. We therefore decided to conduct similar study with the aim of verifying if in fact there is a specific Barrett’s CK7/20 pattern in the esophagus.

Material and Method:

30 esophageal and 20 gastric biopsies obtained at endoscopy were fixed in 10% formalin. 20 esophageal biopsies were from lower esophagus, 10 from gastro-esophageal junction while the gastric biopsies were as follows, cardia 10, body 5 and antrum 5. These were processed and paraffin embedded tissue slides were cut at 5um and stained with Hematoxylin and Eosin, (H&E), Alcian Blue/Periodic Acid Schiff (ALB/PAS), and Alcian Yellow.

For immunohistochemical stains, 5um thick sections cut on commercially coated slides were stained with antibodies for CK7 and CK20 using Avidin-Biotin complex method in automatic immunohistochemical stainer (Dako mate).

Two pathologists examined all the H&E and immunohistochemically stained slides independently and the presence of inflammation, intestinal metaplasia and presence of Helicobacter organisms were noted. For the immunohistochemically stained cases positive and negative stains of the superficial, deep epithelium and glands of the esophagus and the stomach were noted and graded only as positive or negative. There was no attempt to grade the intensity of the positive stains.

Results:

25 cases from the esophagus and gastro-esophageal junction and a total of 15 cases from the stomach, (7 from cardia, 3 from the body and 5 from the antrum) demonstrated intestinal metaplasia on the ALB/PAS stains. 20 cases from esophageal and gastro-esophageal junction, which demonstrated intestinal metaplasia, showed a pattern of CK7/20 found in the studies of Ormsby et al. CK 20 stained the superficial lining epithelium, Figure 1 while CK7 stained both the superficial, deep epithelium and deep glands Figure 2. Sections from the cardia, body and antrum of the stomach showing intestinal metaplasia stained negative with CK7 Figure 3, while only the superficial lining epithelium was stained with CK20 Figure 4. Normal mucosa from gastric cardia, body and antrum stained negative with CK7 while CK 20 antibodies reacted positively with superficial epithelium from gastric cardia, body and antrum. The staining was on the surface epithelium and foveolar but the gastric pits and glands were negative.

Discussion:

Barrett’s esophagus represents a specialised columnar epithelial metaplasia in the lower esophagus and it has been
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Recognized as a major risk factor for increasing incidence of adenocarcinoma of esophagus, gastric cardia intestinal metaplasia has not been shown to increase the risk of cardia adenocarcinoma. It is then obvious that intestinal metaplasia in the esophagus and cardia are two different entities posing different health risks; therefore must be managed differently. This means that accurate diagnosis of Barrett’s epithelium and its separation from any other form of metaplasia in lower esophagus, gastro-esophageal junction or cardia is imperative.

Identification of Barrett’s epithelium in patients with long segment intestinal metaplasia poses little if any diagnostic problem. However, biopsies from gastro-esophageal junction with short segments of columnar mucosa, which in fact, is the finding in the majority of the patients become problematic. This is so because the end of the esophagus and the beginning of gastric mucosa at the cardia are often difficult to determine on endoscopy.

Histochemically, acid mucin in both cardia and esophageal Barrett’s epithelium are indistinguishable on routine special stains such as ALB/PAS.

Given the above difficulty in anatomizing the end of the esophagus and the beginning of the gastric cardia, and the similarity of the acid mucin in both locations, discovery of a specific marker for Barrett’s esophagus would be clinically useful in the accurate diagnosis and management of this condition.

Over 20 cytokeratins have been described. These are cytoplasmic proteins and show variable expression. Some are ubiquitous but CK7 and CK20 show some restriction in their distribution in the tissues.

Our study showed specificity of the CK7/20 pattern for the Barrett’s epithelium. This pattern was defined as staining of

Figure 1: Lower esophagus, Barrett’s esophagus; Immunohistochemical Stain. Note staining of only the superficial epithelium; Antibodies to CK 20.

Figure 2: Same section as in Figure 1. Barrett’s esophagus; superficial epithelium and deep glands and epithelium; Antibodies to CK 7.

Figure 3: Antrum of stomach. Note complete negative stain with antibodies to CK.

Figure 4: Same section as in Figure 3; Note superficial epithelial staining with antibodies to CK 20.
superficial epithelium with CK20, negative deep metaplastic glands and staining of both the superficial and deep normal and intestinal metaplastic epithelium with CK7/20 pattern was observed in 97% of specimen with long segment Barrett’s and was not seen in gastric intestinal metaplasia by Ormsby et al(4). Consequently they asserted that this observed CK7/20 pattern is specific for Barrett’s esophagus.

El-Zimaity and Graham(5) in their study could not reproduce such CK7/20 pattern specificity and sensitivity, showing only 39% in the long-segment Barrett’s and 35%, 4%, and 24% respectively for intestinal metaplasia in the gastric cardia, body and antrum.

Our findings support the studies of Ormsby et al(4). 20 out of 25 cases with intestinal metaplasia in the lower esophagus and gastro-esophageal junction demonstrated this specific pattern. All the cases of intestinal metaplasia from all the parts of the stomach only demonstrated superficial stains with CK20 while CK7 was completely negative.

The five cases that did not demonstrate this Barrett’s pattern were from specimen labeled gastro-esophageal junction and in fact their staining pattern was that of the cardia. These biopsies must have been from the anatomic site of gastric cardia, and were therefore not false negative CK7/20 Barrett’s esophagus. This pattern is most useful in short segments columnar epithelium in the lower esophagus. Incidentally the short segment is the majority of the Barrett’s therefore the CK7/20 pattern will eliminate difficulty in diagnosis and distinguishes intestinal metaplasia unassociated with cancer risk. Though antrum and the body were added in our study these sites are good distances from the gastro-esophageal junction and lower esophagus, therefore, intestinal metaplasia in these sites indeed pose no diagnostic difficulty irrespective of their staining pattern. Routine use of CK7/20 immunohistochemical stains on lower esophageal and gastro-esophageal biopsies will improve the accuracy of diagnosis of Barrett’s esophagus, which will in turn enhance surveillance, and treatment of the premalignant lesion of Barrett’s esophagus.

Acknowledgement:

We wish to thank the secretaries and technologists of the Division of Anatomic Pathology, Department of Laboratory Medicine and Pathology for their immense assistance and contribution to the successful completion of our study.

References:

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