Technical methods

Detection of superficial gastric carcinoma in biopsies and resected stomachs

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Advances in biopsy technique have improved early detection of carcinoma in several organs. The increasing use in this country of upper gastrointestinal endoscopy is likely to promote more frequent search for superficial gastric carcinoma. The histopathological diagnosis of such lesions demands particular care in the handling and examination of the endoscopic biopsies and subsequent gastric resection specimens. However, some of the histopathological methods advocated in early studies of superficial gastric carcinoma seem excessively demanding for routine practice.

In view of these difficulties we wish to describe the simplified methods that we have used to detect nine cases of superficial gastric carcinoma (Machado *et al*, 1975; 1976) in two years.

Methods

GASTRIC ENDOSCOPIC BIOPSIES

The number of tissue fragments is recorded in the macroscopic description of the formalin-fixed biopsy specimen. The tissue is then placed in a plastic cassette (Tissue-Tek) and processed overnight for paraffin embedding. Because of the small size of the endoscopic tissue fragments (2.0 mm is a large biopsy), they are wrapped in gauze. The controlled cooling of the molten paraffin wax attainable with the plastic cassette technique allows tissue fragments to be consistently grouped closely together. Hence this method obviates the need to block each fragment separately, as might be necessary when using metal containers for the molten paraffin.

The paraffin-embedded tissue is sectioned initially at three levels so that about two-thirds of the tissue is cut through. From each level a ribbon of five to eight sections is stained with haemalum and eosin.

A similar set of unstained spare sections is held in reserve for haemalum and eosin, mucin, or reticulin staining as needed. Additional levels from the biopsy blocks are cut only if epithelial dysplasia is found in the absence of overt carcinoma, or if any of the biopsy fragments are not adequately sectioned in the first three levels. The ribboning and the staining procedures for the deeper levels are as described above.

RESECTED STOMACHS

As soon as possible after surgical resection the stomach is taken to the histopathology laboratory. It is opened in the fresh state along the greater curve. After being pinned on a cork board, mucosa uppermost, the specimen may be photographed fresh. Thereafter the stomach is fixed for at least 48 hours by immersion upside down in 10% formal saline. This method of fixation prevents muscular buckling which might otherwise interfere with subsequent systematic examination of the tissue. It is vital during this time to review the specimen with the endoscopist, who can indicate the site of the suspicious lesions. A second set of photographs, including an overall view and appropriate close-ups, may be obtained during fixation. Photography while the stomach is fresh and fixed allows comparison with the endoscopic appearances and yet reveals lesions such as shallow ulcers, which may be inconspicuous in fresh tissue.

Macroscopic description of the stomach is also made in the fresh and fixed states. We find that, even for experienced pathologists, a check-list of descriptive features and a sketch of the stomach aid consistent and comprehensive description. In practice, the check-list is easier to use and is more flexible than a printed protocol with a series of alternative descriptions. Features included in the check-list are: the lengths of the greater and lesser curvatures, the dimensions and macroscopic characteristics of suspicious lesions, their situation and distance from the nearest plane of surgical excision, the presence and location of mucosal ulcers, any distortion of rugae and other visible disturbances of mucosal pattern, and the presence and macroscopic appearances of lymph nodes in the greater and lesser omenta.

To examine the stomach thoroughly many blocks of tissue must be taken in a systematic fashion. Outlining the proposed blocks on a sketch of the

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Figure Sketch of stomach indicating tissue blocked in paraffin. Hatched areas are suspicious lesions. Stippled blocks are those sectioned and stained initially.

stomach helps to plan the incisions. A map of the blocks taken in the cut-up (figure) is invaluable for retrospective review of the distribution of the lesions which are detected histologically. We find that it is preferable to prepare tissue blocks (3 to 4 mm deep and up to 30 mm long) from the entire suspicious area straight away. Such an approach may entail taking 20-30 blocks at this stage. We have found that partial blocking of the suspicious area is unsatisfactory because the trimming in re-blocking precludes complete examination. Care must be taken to orientate the tissue uniformly as it is placed in the plastic processing cassettes, which are labelled, as indicated, in a map of the blocks included in the sketch of the stomach.

For reasons of economy, only a limited number of the blocks need be sectioned at 5 μ m and stained at first, although all are processed and embedded in paraffin. The remainder are held in reserve. Further blocks may then be sectioned as required.

Comment

Very detailed examination of gastric biopsies by means of sections at 100 μ m intervals, as recommended by Hermanek (1973), could entail ribbons from 20 to 30 levels in tissue which is more than 2 or 3 mm deep in the paraffin block. Unless the examination is for exceptional purposes, the work involved for technical and medical staff seems

excessive. The geometrical considerations discussed by Wilkinson and Hause (1974) in the detection of metastasis in lymph nodes lead us to favour the easier method of interval sectioning that we have described. We do not routinely block the endoscopic biopsy fragments separately, and find that the plastic cassette system for processing the biopsies reduces the number of paraffin blocks necessary. Since up to a dozen tissue fragments may be procured at endoscopy, this represents a considerable saving in technical effort. Alternatively, those who wish to study the extent of the lesions in the endoscopic biopsies may block each of the tissue fragments in individual cassettes.

Superficial gastric carcinoma is often inconspicuous and might be missed on cursory inspection of the stomach. For this reason alone the collaboration of the endoscopist is essential. Alternatively, the complete examination of a resected stomach, as described by Mochizuki (1971), would mean blocking over 300 moderate-sized portions of tissue. At first sight the Swiss-roll technique (Magnus, 1937) seems suitable for the complete examination of a stomach. Unfortunately, it is not a method that can be adopted with standard tissue-processing cassettes. The size of the blocks necessitates lengthy hand-processing and the use of large glass slides which are not capable of being accommodated on an automatic staining machine. Further, such large blocks make thin sectioning difficult and increase the problem of histological interpretation in borderline lesions.

We feel that our methods are a reasonable compromise whereby acceptable thoroughness of examination is achieved with an economy of technical and medical effort.

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