Unusual histological appearances of barium sulphate—a case report with scanning electron microscopy and energy dispersive x ray analysis

C WOMACK

From the Departments of Pathology, City Hospital and University Hospital, Nottingham

SUMMARY Multiple birefringent rhomboidal crystals seen on histological examination of a resected oesophagus were subsequently found to contain elemental barium and sulphur on energy dispersive x ray (EDX) analysis. Scanning electron microscopy and EDX analysis of readily available commercial barium sulphate suspensions suggested that the two forms of barium sulphate previously described in tissue sections may be a result of the method of preparation of the commercial suspension used.

With increased use of barium sulphate prepared by crushing the natural compound, birefringent rhomboidal crystals should be found with increasing frequency in endoscopic biopsy samples and resected specimens.

In histological sections, barium sulphate classically appears as green, finely granular material which is not birefringent.¹ Larger, birefringent rhomboidal crystals of barium sulphate may also occur, but these are usually in association with the classic green, granular forms.

In the case described in this report, multiple rhomboidal crystals were found on histological examination of a resected oesophagus. No classic green, granular barium sulphate was seen histologically, and attention was focused on other possible sources for the rhomboidal crystals. The nature of these crystals was eventually determined by energy dispersive x ray (EDX) analysis, which showed elemental barium and sulphur, consistent with the patient having undergone several preoperative barium contrast studies.

It has recently been suggested that the birefringent rhomboidal crystals sometimes seen histologically represent additives in commercial preparations of barium contrast and not barium sulphate itself.² It is well known that manufacturers of barium sulphate are reluctant to divulge the precise constituents of their preparations.³ To look for possible additives, samples of three commercial barium sulphate suspensions commonly used in Nottingham hospitals were examined by scanning electron microscopy and EDX analysis.

Case report

HISTORY

A 46 year old man with a past history of attempted suicide swallowed some commercial grade ammonia (strong ammonia solution) in a further suicide attempt. He subsequently developed a lower oesophageal stricture with perforation. Following the failure of conservative management, which included the patient having a sialastic tube in his oesophagus for three weeks to help reduce stricture formation, a partial oesophagectomy was performed about eight weeks after the original injury.

Between the time that he swallowed the ammonia solution and his oesophagectomy, the patient underwent three barium sulphate oesophageal studies. In each study the commercial brand of barium sulphate used was not recorded.

After the operation the anastomosis healed without appreciable stricture formation. The patient is currently in good physical health, on a normal diet, and has been discharged for psychiatric assessment.

MACROSCOPIC APPEARANCES

The resected specimen, received in 10% formol saline, consisted of a 14 cm length of oesophagus with a rim of stomach 1 cm wide at one end. The outer surface was roughened and haemorrhagic. At the mid point of the specimen there was an irregular opening 0.7 cm in diameter which communicated

Fig. 1 (a) Within granulation tissue are many irregular crystals, which vary in size and shape, the larger ones being rhomboidal. (b) When viewed under crossed polarising filters many of the crystals are birefringent. Haematoxylin and eosin. Original magnification $\times 936$.

with the lumen. The oesophageal wall was uniformly thickened to 1 cm and the lumen was narrowed. The mucosal surface was dark red, friable, and showed multiple linear ulcers.

LIGHT MICROSCOPIC APPEARANCES

Light microscopy of routine paraffin embedded, haematoxylin and eosin stained histological sections showed complete replacement of oesophageal mucosa and partial replacement of submucosa and muscularis propria by inflammatory granulation tis-

sue. In addition, there was fibrosis of submucosa and muscularis propria.

Crystals were seen in most of the histological sections of the specimen examined, were present within the superficial granulation tissue, and were seen in sections examined from the whole length of the specimen. The crystals varied in size and were irregular in shape, the larger ones being rhomboidal (Fig. 1a). Many of the crystals appeared birefringent under crossed polarising filters (Fig. 1b). We were unable to stain the crystals using the Rhodizonic acid method for barium⁴ and no classic green granular barium was seen.

Material and methods

Pieces of the resected oesophagus were embedded in epoxy resin. Sections roughly $2\text{ }\mu\text{m}$ thick were cut and mounted on small perspex squares. Samples of three commercial barium sulphate suspensions E-Z-HD (Henley's Medical), Baritop 100 (Nicholas Laboratories), and Micropaque Standard (Concept Pharmaceuticals) were obtained. E-Z-HD barium sulphate was made up in tap water according to the manufacturer's recommendations; the other two (Baritop and Micropaque) are provided ready for use by the manufacturers. Aliquots of each suspension were pipetted on to perspex squares and allowed to dry in air.

All the oesophageal and barium sulphate samples thus prepared were then carbon coated using a Nanotech micropep 250 and examined in a JEOL JSM 35C scanning electron microscope with a Link Systems 860 EDX analysis system.

Results

Two distinct patterns were seen on scanning electron microscopy. The preparations of oesophagus (Fig. 2a) and E-Z-HD barium sulphate suspensions (Fig. 2b) showed irregular particles which varied greatly in size and shape, the larger ones being rhomboidal. In contrast, Baritop 100 (Fig. 2c) and Micropaque Standard (Fig. 2d) showed much smaller, smoother, regular particles.

EDX analysis of all the specimens examined with the scanning electron microscope showed only elemental barium and sulphur (Fig. 3). Typical electron energies for the peaks are shown in the Table. No peaks were seen for magnesium or silicon.

Discussion

Initially, several possible sources for the rhomboidal crystals seen in histological sections of the resected oesophagus were considered and these included:

Fig. 2 Scanning electron microscopy. (a) Oesophageal crystals embedded in epoxy resin show particles of varying size and shape, many with jagged edges and similar in morphology to (b) particles of E-Z-HD commercial suspension. Original magnification $\times 4940$. In contrast, the particles of (c) Baritop 100 and (d) Micropaque Standard are rounder, smaller, and more uniform. Original magnification $\times 14\,820$.

1 The ammonia ingested: whether an ammonium salt or a complex containing ammonium ions and formed by direct action on the oesophageal tissues.

2 A contaminant: whether in the ammonia solution ingested, at operation, or in the formalin fixative used.

3 Other ingested material: before operation the patient had taken food orally. He also had a sialastic tube in his oesophagus for three weeks.

4 Contrast medium: although there were three barium contrast studies between the patient's ingestion of ammonia and his oesophagectomy, the histological appearances and negative rhodizonic acid stain were not diagnostic for barium sulphate.

The nature of the rhomboidal crystals was determined rapidly and accurately by EDX analysis. This technique is being used increasingly in many branches of pathology.⁵⁻⁷

Fig. 3 Energy dispersive x ray analysis. Traces of each specimen show four barium (Ba) peaks and one sulphur (S) peak. No other elements were found. (a) oesophagus; (b) E-Z-HD; (c) Baritop; (d) Micropaque.

In the case presented, both the extent of deposition of barium sulphate and its histological appearances are unusual. The extent of deposition of barium sulphate may be explained by the extent of damage to the oesophagus. The ulcerative properties of strong ammonia solution are well recognised. It is also accepted that contrast medium gains access most readily to the wall of the alimentary tract through diseased or traumatised mucosa or, when instilled under pressure, through an intact mucosa.⁸ The highest concentration and deepest penetration

of barium sulphate occurred around the site of perforation, where damage from the ammonia solution was greatest.

The original description of the histological appearances of barium sulphate⁹ has since been confirmed in animal and human experiments, and it is now widely accepted that in histological sections barium sulphate appears as green, finely granular material which is not birefringent. In the first report of a barium granuloma of the rectum¹⁰ attention was drawn to numerous doubly refractile rhomboidal crystals which were scattered among the cellular elements of the granuloma. Chemical and spectroscopic analysis of these rhomboidal crystals showed barium, and on the basis of this and subsequent studies¹¹ it has been accepted that such birefringent rhomboidal crystals are composed of barium sulphate. Rhomboidal crystals are usually associated with green granular barium sulphate, however, and are usually a minor feature.

Typical electron energies for the peaks obtained on energy dispersive x ray analysis

Energy (eV)	Element
2460	S
4480	Ba
4840	Ba
5160	Ba
5520	Ba

Reasons for the difference in the histological appearances of barium sulphate have not generally been stated in previous reports, although changes in physical property, such as birefringence, as a result of phagocytosis have been suggested.²

Commercial barium sulphate suspensions are prepared either by a precipitation process or by crushing the naturally occurring compound.¹² Precipitated barium sulphate suspensions (for example, Baritop 100, Micropaque Standard) contain small, smooth regular particles, whereas crushed preparations (for example, E-Z-HD) contain larger particles which vary in size and have rough edges. Many of the crushed particles of barium sulphate are birefringent whereas the precipitated particles are not. The microscopic appearances of barium sulphate may thus depend on the method of preparation of the commercial suspension used; precipitated suspensions appearing as green granular particles in histological sections and crushed suspensions as rhomboidal crystals.

The two forms of barium sulphate have different properties which can be useful radiologically.^{12,13} Precipitated granular barium sulphate is good for large bowel and single contrast gastrointestinal studies. Since many of the reports referred to above concern large bowel barium granulomas, the finding of green granular barium sulphate histologically may simply reflect the widespread use of precipitated commercial suspensions in barium enemas. The finding of occasional rhomboidal crystals histologically may be explained by the small quantities of crushed barium sulphate which are sometimes added to precipitated commercial suspensions to enhance radiological contrast.¹⁰ High quality double contrast examination of the upper gastrointestinal tract is best obtained using a crushed barium sulphate preparation because the rough edges of the particles facilitate mucosal adherence. Crushed barium sulphate preparations have become readily available commercially only in the last 10 years, which, together with the fact that there are few reports of upper alimentary granulomas, may explain why rhomboidal crystals are not commonly seen histologically.

An alternative explanation for these rhomboidal crystals has recently been reported. In a comparative light and EDX analysis study of a barium granuloma of the stomach, rhomboidal crystals seen in association with green granular barium sulphate were found to be rich in magnesium and silicon.² The rhomboidal crystals were interpreted as being talc added to the commercial barium sulphate suspension used and this interpretation seems to have gained acceptance.¹⁴ However, the study of the gastric barium granuloma contained little clinical

information, other possible sources for the magnesium and silicon were not considered, and the authors were unable to explain why pure barium sulphate crystals are birefringent and barium sulphate in the granuloma is not. Furthermore, in the present study samples from the patient and the three commercial barium sulphate suspensions contained only elemental barium and sulphur on EDX analysis. In particular, no magnesium or silicon has been shown. In two other papers which include EDX analysis of barium contrast in human tissue^{15,16} no magnesium or silicon was reported. In one paper¹⁵ barium, sulphur, and aluminium were found in a barium granuloma of transverse colon. The aluminium was thought to represent a contaminant introduced during preparation of the tissue for EDX analysis.

A review of tissue reactions to barium sulphate is beyond the scope of this case report. Further study is needed to determine potential differences between *in vivo* reactions to crushed and precipitated barium sulphate preparations. With the increased use of crushed barium sulphate preparations, particularly in upper gastrointestinal radiology, histopathologists need to be more aware of the histological appearances of these preparations.

I would like to thank Mr WE Morgan, FRCS, for permission to publish the clinical details of his patient. I am indebted to Mr W Brackenbury and Mr S Terras and to Mr E Toulson of JEOL (UK) for expert technical assistance. I should also thank Dr ID Ansell and Professor DR Turner and Professor RE Cotton for helpful advice and Mrs HJ Nice for typing the manuscript.

References

- ¹ Morson BC, Dawson IMP. *Gastrointestinal pathology*, 2nd ed. Oxford: Blackwell Scientific Publications, 1979:578.
- ² Marek J, Jurek K. Comparative light microscopical and X-ray microanalysis of barium granuloma of stomach. *Path Res Pract* 1981;**171**:293-302.
- ³ Miller RE. Barium sulfate suspensions. *Radiology* 1965;**84**:241-51.
- ⁴ Pearse AGE. *Histochemistry theoretical and applied*, 3rd ed. vol 2. London: Churchill Livingstone, 1972:1142.
- ⁵ Underwood JCE. *Introduction to biopsy interpretation and surgical pathology*. Berlin, Heidelberg, New York: Springer Verlag, 1981:118.
- ⁶ Slater DN, Durrant TE, Gray T, Parry EA. Transmission electron microscopy with energy dispersive X ray microanalysis: its application in the medicolegal autopsy. *Med Sci Law* 1983;**23**:5-16.
- ⁷ Harrison JD, Davies KJ. A method for the identification of particulate material in biopsies from the oral tissues. *Br Dent J* 1982;**153**:327-9.
- ⁸ Lewis JR, Kerstein MD, Koss N. Barium granuloma of the rectum; an uncommon complication of barium enema. *Ann Surg* 1975;**181**:418-23.

Unusual histological appearances of barium sulphate

- ⁹ Thomas JC. The disposal of barium sulphate in the abdominal cavity. *J Path Bact* 1936;**43**:285-98.
- ¹⁰ Beddoe H, Kay S, Kaye S. Barium granuloma of the rectum: report of a case. *JAMA* 1954;**154**:747-9.
- ¹¹ Carney JA, Stephens JH. Intramural barium (barium granuloma) of the colon and rectum. *Gastroenterology* 1973;**65**:316-20.
- ¹² James WB. Double contrast radiology in the gastrointestinal tract. *Clin Gastroenterol* 1978;**7**:397-430.
- ¹³ Gelfand DW, Ott DJ. Barium sulfate suspensions: an evaluation of available products. *AJR* 1982;**138**:935-41.
- ¹⁴ Chaplin AJ, Turner ELT. Observations on the histochemistry of

barium. *Histochemistry* 1983;**79**:111-6.

- ¹⁵ McKee PH, Cameron CHS. Barium granuloma of transverse colon. *Postgrad Med J* 1978;**54**:698-702.
- ¹⁶ Crocker PR, Toulson E, Levison DA. Particles in paraffin sections demonstrated in the backscattered electron image (BEI). *Micron* 1982;**13**:437-46.

Requests for reprints to: Dr C Womack, Department of Pathology, University Hospital, Queen's Medical Centre, Nottingham NG7 2UH, England.