Pseudoglandular formations in clot sections from fine needle aspirates—an artefact caused by bubble formation during aspiration

C E Keen, A Karim

Abstract

Aims—To report the occurrence of an uncommon artefact producing pseudoglandular formations in clot sections from haemorrhagic fine needle aspirations.

Methods—All available histological material from 610 fine needle aspirations by pathologists (23 g needle) over a five year period was reviewed. The frequency and associations of the pseudoglandular artefact was assessed.

Results—Clot sections were prepared in 41 of the 610 cases (7%). Bubbles were present in the clots in 22 of these cases (54%), and in three cases (7%) these were lined by lymphocytes creating pseudoglandular formations. These were two lymph node aspirates and one thyroid aspirate. In four further cases lesser numbers of cells partly lined some of the bubbles; these were lymphocytes, macrophages, or in one case, thyroid epithelial cells.

Conclusions—When clot sections are prepared in cases of haemorrhagic fine needle aspiration, bubbles are often produced during suction; these can on occasion become lined by lymphocytes or other cells, leading to pseudoglandular formations. Recognition of this artefact will prevent unnecessary further investigation of their nature.

(J Clin Pathol 1999;52:532-534)

Keywords: fine needle aspirate; pseudoglandular artefact; clot section; bubbles

Fine needle aspiration (FNA) may be complicated by haemorrhage, which leads to suboptimally stained direct smears. The conventional advice given in manuals of aspiration is to aspirate until the first drop of blood is seen in the transparent needle hub and not to continue after that point, in order to obtain a bloodless aspirate. Some authors advocate aspiration without suction in order to avoid aspirating blood. For example with thyroid aspirates it is probably not necessary to use suction at all in order to obtain sufficient material for direct smears and needle washings. We do use the full suction provided by a 20 ml syringe, but we have modified our practice to turn any haemorrhage to our advantage, by continuing the aspirate and allowing the haemorrhagic component to clot.

We draw attention to an artefact that may be seen in material prepared in this way. In this paper we review all our fine needle clot sections

over a five year period, and assess the mechanism, frequency, and importance of this artefact.

Index case

A 40 year old man presented with a smooth 2 cm oval mass in the submandibular region, of two weeks duration. FNA suggested a reactive lymph node and this was confirmed in clot sections; bcl-2 immunostaining showed negative germinal centres confirming reactive lymphoid follicles. In addition, however, curious pseudoglandular formations were noted (fig 1). These were lined by a single layer of apparently cuboidal cells with a palisaded orientation; they had little cytoplasm and a very straight "luminal" border. Most of the spaces were empty, but some contained thin eosinophilic material. Some appeared partially collapsed. The initial impression was of glandular or neuroepithelial structures. Immunohistochemistry for epithelial and neuroendocrine markers was negative, and no mucin was demonstrated. Some of the cells lining these structures marked as T cells. There were no corresponding structures in the direct smears. Nevertheless, there was continued concern and repeat FNA or excision was advised; this proved clinically unnecessary.

Figure 1 Pseudoglandular structures in a clot section from the index case: reactive lymph node hyperplasia.

Department of
Histopathology and
cytopathology,
University Hospital
Lewisham, London
SE13 6LH, UK
C E Keen
A Karim

Correspondence to:
Dr C Keen, Department of
Histopathology, Royal Devon
and Exeter Hospital
(Wonford), Church Lane,
Exeter EX2 5AD, UK.

Accepted for publication
1 April 1999

Figure 2 Aspirate from mantle cell lymphoma, showing pseudoglandular structures.

When the same appearance was seen in samples from other patients, it became clear that this phenomenon was an artefact.

Methods

ASPIRATION TECHNIQUE

We use a 23 gauge needle (31.75 mm with transparent blue Luer hub), in a Cameco® pistol grip handle, and we sample lesions in a conventional way,¹ with multiple to and fro passes of the needle tip through the lesion. If blood is obtained during aspiration, the procedure is continued regardless until the lesion is well sampled. As usual the aspirating suction is released before withdrawal of the needle. This leaves up to 1–2 ml of blood in the syringe. The needle is immediately detached, and direct smears are prepared in the usual way using a fresh syringe to expel the material in the needle onto slides. Residual needle contents are washed into saline and reserved until the direct smears have been examined. The blood and tissue in the original syringe is allowed to clot and after a few minutes the clot is extracted by removing the piston from the rear of the syringe. The clot is placed directly into a tissue basket in a cassette, fixed in formalin, and processed overnight for routine histology.

REVIEW OF MATERIAL

We retrieved records of all histological material in our department coded as originating from FNAs in a five year period (1993 to 1997). There were 53 cell pellets prepared from centrifuged needle washings in saline, and these samples were not included in the review. Forty one clot sections prepared as above were reviewed. These were compared with 610 fine needle aspirates coded as taken by the pathologists in the same period.

Figure 3 Pseudoglandular structures in a clot section from a thyroid aspirate.

Results

Of the 41 clot sections, 19 (46%) were prepared from thyroid aspirates, 13 (32%) from lymph node aspirates, and seven (17%) from salivary gland aspirates. Two cases (5%) were from other sites. The corresponding proportions of all 610 aspirates taken by pathologists from those sites in the same period were thyroid, 85 (14%); lymph node, 311 (51%); and salivary gland 128 (21%). The remaining 86 (14%) were from other sites. Pseudoglandular formations were seen in three cases (7%), including the index case. Two were from lymph node aspirates—the index case, reactive lymphadenopathy (fig 1); and another case, mantle cell lymphoma (fig 2): in this case there was no corresponding structure in the direct smears, nor in the subsequent lymph node excision biopsy—and one was from a thyroid aspirate (reported as follicular neoplasm, fig 3). In a further 19 cases, however, groups of round spaces were seen in or at the edge the blood clot. These spaces were of the order of two to five times the diameter of an adipocyte. In most of these cases no cells other than the erythrocytes of the blood clot itself lined the spaces. In two cases a few lymphocytes were noted lining part of the spaces. In one case macrophages were noted lining the spaces and in one case thyroid epithelial cells lined the spaces. In addition to these round spaces noted mainly at the edge of blood clots, there were smaller spaces, of the order of the size of an adipocyte, sparsely scattered throughout the clots.

Discussion

We have shown that bubble formation is common in clotted blood from haemorrhagic aspirates. Aspiration under suction may induce “low temperature boiling” if the vapour

pressure in the fluid aspirate exceeds that in the syringe barrel. If the clotting process begins before the suction is released, these bubbles may persist, either in full or collapsed form. The artefact we describe is dependent on a lymphoid cell rich aspirate that lines the bubbles to produce the pseudoglandular formations. This may explain the uncommon nature of the artefact—lymph node aspirates are not often haemorrhagic. Thyroid aspirates, which are more commonly haemorrhagic, are infrequently lymphocyte-rich, but they will be in thyroiditis and lymphomas.

The physical circumstances in which cells preferentially line the surface of the bubbles remain unclear. We could find no previous description of this artefact searching Medline from 1966 to the present for occurrences of the words artefact or artifact, and bubble, in title or abstract. There is some parallel between the process of bubble formation as described here, and that observed in some other specimen types. Similar bubbles are quite often seen in clotted blood from the endometrial cavity, for example in suction curettage. Floating fragments can sometimes be seen in the gross fixed specimen, containing visible bubbles. The endometrial cavity does not, however, contain the numerous lymphocytes required to complete the pseudoglandular artefact. The smaller spaces seen in our clot sections, about the size of an adipocyte, were diffusely distributed and did not interact with cells in the clots. They are of uncertain origin and significance. It is possible that they represent lipid globules lost in processing.

There is a description of artefactual rosette-like structures in a lymph node aspirate, raising

the possibility of neuroblastoma.² While these were in smears, not clot sections—and are clearly different—they do illustrate the point that one should be aware of pseudostructural artefacts occurring in aspirated material.

We have found our approach to aspiration to be effective and safe, despite the general consensus that haemorrhage means one should stop aspirating. We have had no problems with haematoma, infection, or other complications. We routinely obtain well preserved lesional tissue fragments and cores up to 3 × 0.5 mm, conveniently held in the clotted blood from aspiration. Resultant clot sections are very useful in showing the architecture of the aspirated tissue and allowing recuts for immunohistochemistry or other special stains as required. The artefact, once recognised, is easily disregarded and it should not detract from the use of this technique.

Microbiopsies may also be handled by other techniques such as that described and referred to by Mravunac *et al*,³ under the rubric “where cytology and histology meet.” We endorse the efforts to use the material available in the most efficacious manner, for the benefit of the diagnostic process and hence the patients.

1 Orrell SR, Sterrett GF, Walters MN-I, *et al*. *Manual and atlas of fine needle aspiration cytology*, 2nd ed. Edinburgh: Churchill Livingstone, 1992:10–15.

2 Sethi S, Yu GH. Rosette-like structures in a lymph node aspirate: pitfall in the diagnosis of neuroblastoma [letter]. *Cytopathology* 1997;8:213–15.

3 Mravunac M, Verbeek D, van Heusden C, *et al*. Applications of the microbiopsy technique in non-cervical cytology: where cytology and histology meet. *Histopathology* 1998;33:174–82.