Liquid Based Cytology of Cell Remnants in Needles Used for Breast Fine Needle Aspiration

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Received: July 30th, 2010; revised August 6th, 2010; accepted September 14th, 2010.

ABSTRACT

This study examined cells contained in needles used for the collection of breast fine needle aspirates for the detection of malignant cells trapped in the needles. Remnants of cells contained in 50 needles used for the collection of scanty breast fine needle aspirates were examined by the liquid based cytology technique and compared with the conventional cytological technique of specimens in the corresponding syringes. The breast specimens were collected with clean sterile needles attached to the syringes. Smears were made and stained by the conventional method. The needles were removed from the syringes and a fixative was withdrawn into the syringes and the syringes were recapped with the needles. The fixative containing the specimen was then completely discharged into a centrifuge tube through the needles and treated by the liquid based cytology technique. The study revealed that cells were found trapped in all the needles used for the collection of breast FNA. 6% of them were positive for malignancy, similar to results obtained in the conventional method. Needles used for the collection of breast FNA should be examined before malignancy is completely ruled out particularly in extremely scanty specimens with a clinical suspicion of malignancy.

Keywords: Liquid Based Cytology, Needle Cell Remnants, Fine Needle Aspiration, Breast

1. Introduction

Liquid-based cytology is a technique that enables cells to be suspended in a monolayer and thus making better morphological assessment possible with improved sensitivity and specificity because fixation is better and nuclear details are well preserved in this technique. Abnormal cells are not obscured by other epithelial or inflammatory cells [1]. The method involves collection of specimens directly into a liquid fixative, but in the case cervical specimens, with a brush-like device, Cervex-brush (Rovers medical devices), the brush is used to scrape the cervix according to the manufacturer's instructions, viz. insertion of long bristles into endocervical canal, short bristles against the ectocervix and five full 360º rotations in clockwise direction only. The brush head is then detached and immediately put into a vial containing a special commercial fixative solution such as SurePath preservative fluid [1]. Smears are made from the sediment, stained and evaluated. Two techniques - Thin Prep (Cytyc Corp.) and SurePap (Tripath imaging, Inc.) have been more widely used [1]. The introduction of liquid based cytology has led to improvements in unsatisfactory smear rates, with significant benefits to colposcopic referrals and laboratory turnaround times and colposcopic referrals for repeated unsatisfactory smears has fallen from almost 25% to 0.5%, while the percentage of unsatisfactory smears has fallen from 13.6% to 1.9% [2]. The superiority of the quality of liquid based cytology in comparison with those of conventional smears has been described [3,4]. The sensitivity of a conventional Pap smear is estimated to be 70-80% and about 85-95% for liquid-based cytology tests [5]. Liquid-based cytology is now recommended for cervical cancer screening [6] with a major advantage of allowing ancillary techniques such as those used in immunocytochemistry and molecular biology [7,8]. Liquid-based cytology was introduced as an improvement of the Pap smear technique for cervical specimens but it has also been used for non-gynaecologic cytology [9] and brilliant results have been obtained. The aim of this work was to determine by liquid based cytology the diagnostic value of remnants of cells in needles which are meant to be discarded after the conventional cytological technique for breast specimens.
2. Materials and Methods
Fifty breast aspirates received in needles and syringes were studied. The needles were removed from the syringes after the smears were made by the conventional method. 10ml of liquid based fixative was then aspirated into the syringe and the needle was attached to the syringe. The fixative was then expelled through the needle forcing the cells to be released with the fixative. This was collected in a centrifuge tube and allowed to fix for 2 hours and was centrifuged for about 10 minutes at 1000 rpm and the supernatant removed. 5 ml of a cleaning solution was added to the sediment and spun for 10 minutes. The supernatant was removed and a cellular base added and mixed with vortex for 1 minute. Smears were made from the sediment and stained with haematoxylin and eosin (H&E) by applying Harris haematoxylin for 4 minutes. The smear was then rinsed in water, differentiated in 1% acid-alcohol for 3 seconds, rinsed in water, blued in Scott’s solution for 2 minutes and counterstained in 1% eosin for 30 seconds. Smears were then dehydrated through ascending grades of alcohol, cleared in xylene and coverslipped on DPX. The smears made by the conventional method were also stained with H&E and examined under the X40 microscope objective.

3. Results
The smears from four patients had a clean background with a uniform cell distribution with very few cases of overlapping of cells. The structure of the cells and nuclear appearance were well preserved with a good resolution which enabled a clearer and accurate identification of cells under the microscope. Cells in sheets were also retained without disrupting their diagnostic value.

4. Discussion
Breast carcinomas are the most frequent tumors in women.

Table 1. Appearance and percentage of positive smears in remnants of cells in needles compared with the conventional method.

<table>
<thead>
<tr>
<th>Appearance of smear</th>
<th>Positive</th>
<th>Negative</th>
<th>% of positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>LBC of remnants of cells in needles</td>
<td>Clean background with good morphological assessment, Good fixation and well preserved nuclear details.</td>
<td>3</td>
<td>47</td>
</tr>
<tr>
<td>Conventional method</td>
<td>Similar to liquid based cytology of remnants of cells in needles except with pinkish background with some overlapping cells</td>
<td>3</td>
<td>47</td>
</tr>
</tbody>
</table>

In several countries, mammographic screening allows the early diagnosis of tumors. However, when a tumor is suspected, morphological analysis alone can establish the diagnosis of carcinoma. In this context, guided fine needle aspiration has become increasingly popular for obtaining tissue specimens for the diagnosis of malignant breast diseases [10]. Liquid-based cytology in breast FNA had a good correlation with conventional preparation of breast FNA with the advantages of easier and less time consuming evaluation of cell morphology (clear background, no overlapping, smaller area to screen), reproducibility, a factor of great importance to quality control; and possibility of adjunctive investigations such as immunocytology and flow cytometry on the same material [11]. This has been corroborated with the addition that quantitative analysis of HER-2 mRNA correlated with the results of immunohistochemistry in cancer patients [12]. We have found similar morphological characteristics in our study with the ability to produce multiple clean smears from a single needle containing cells from breast specimen. This study has revealed that cells of diagnostic value are often trapped in the needles used for the collection of breast aspirates and should be examined before malignancy is completely ruled out, particularly in extremely scanty breast specimens where clinical diagnosis suggests malignancy. A major disadvantage of extracting cells from used needles is the risk of accidental puncture of the fingers with such used needles which may contain virulent bacteria and viruses. For this reason, extreme care should be taken when handling used needles [13].

REFERENCES
[1] A. N. Kavatkar, C. A. Nagwanshi and S. M. Dabak,


