Aspiration Techniques

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3.1 Introduction

We live in the era of minimally invasive medicine, often achieved through the use of complex and costly equipment and procedures. Fine needle aspiration cytology (FNAC) runs opposite to this trend, managing to be both accessible and minimally invasive (De Rosa et al. 2020). It requires little equipment and can be performed quickly. It is also safe, causing minimal or no complications for the patient, with little discomfort. Furthermore, when employed correctly, FNAC provides a high sensitivity and specificity in the diagnosis of breast malignancies.

This technique is usually done as an outpatient procedure and enables rapid on-site evaluation (ROSE) for sample adequacy, reducing the need for hospital beds and repeat procedures. This allows for resources to be diverted to other areas, which may be particularly important in low-resource settings and in pandemic contexts, such as that which arose in 2020 due to COVID-19 (Pinto and Schmitt 2020).

It is important to stress, however, that both the aspiration technique and the correct interpretation of cytology slides require practice and skill. Practices unfamiliar with the technique should start with low volumes, progressing safely but surely. Of note, the interpretation of breast cytology was recently made more accessible by the release of The Yokohama System for Reporting Breast Fine Needle Aspiration Biopsy Cytopathology (Field et al. 2020), which defines diagnostic categories with well-defined diagnostic criteria and risks of malignancy.

3.2 Role of Breast FNAC in the Clinical Practice

In the breast, FNAC should be used as a component of the so-called triple test, which is the assessment of suspect lesions integrating clinical,

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imaging, and morphological data. In this context, core needle biopsies (CNB) should only be used in a subset of cases with discordant findings in the triple test.

It is true that CNB have replaced FNACs in most practices in the developed world. This is because they are perceived to enable a more reliable and definitive diagnosis and also provide adequate material for ancillary testing, which is mandatory in the context of breast cancer. This perception is not entirely accurate; however, great strides have been made in improving the diagnostic yield of FNACs of the breast, and in using the material thus obtained for ancillary studies. Therefore, it is no surprise that not only do FNACs remain very important in low-resource settings but are also part of the daily life in certain large, specialized, practices in the developed world (De Rosa et al. 2020). Furthermore, CNB are more expensive, cause more discomfort, and have a higher potential for complications.

The advantages of FNAC are:

- Greater mobility of the needle during aspiration, allowing an increased area of sampling (vs. CNB, which obtain tissue in only one plane).
- Greater sensitivity of the physical nature of the lesion through palpation, and therefore better needle localization.
- Better evaluation of the texture of the lesion during the aspiration, which may hint at the diagnosis and helps to determine if additional needle passes are needed. For example, gritty, rubbery, or "fatty" resistance feelings suggest the possibilities of carcinoma, fibroadenoma, and fat necrosis, respectively.
- Allows for ROSE, that is, to immediately and accurately assess the adequacy of the specimen obtained, avoiding unnecessary repetition of the procedure, and reducing time for diagnosis by the immediate identification of an inadequate aspirate.
- Processing time is significantly less when compared to both paraffin embedding and frozen section processing of needle biopsies, thereby lowering turnaround times and enabling a faster diagnosis.
- Lower cost, discomfort, and complications when compared to CNB.

On the other hand, disadvantages of FNAC include:

- Little experience among pathologists with performing FNAC and in the interpretation of the corresponding slides. In many centers, pathologists are not trained adequately to perform FNAC or in reading cytological slides, particularly in the context of breast lesions and as a result are more comfortable with histology. This is probably one of the main reasons why FNACs have been largely replaced by CNB when cost is no objection.
- Perceived limitations in the available material for ancillary studies and research endeavors. Cytology samples are thought of as inadequate for these ends, particularly for performing immunohistochemistry, which is mandatory for theranostic purposes. However, as demonstrated in other chapters of this book, most ancillary techniques can be performed in cytological material with results comparable to histology, particularly if cell blocks are used.
- Possible low cellular yield in some lesions, namely carcinomas with a highly desmoplastic stroma.
- Difficulty in classifying proliferative lesions that have a degree of atypia but lack unequivocal features of malignancy. Recent research suggests this may be solvable using immunohistochemistry such as 34βE12, however (Hoshikawa et al. 2016).
- Difficulty in distinguishing in situ versus invasive carcinomas. This may be a significant drawback, particularly in the context of neoadjuvant chemotherapy or sentinel lymph node biopsies. However, many of these cases can be resolved by using the triple assessment (Kocjan 2006; Kocjan et al. 2006, 2008). Furthermore, p63 has been successfully used for this purpose in several publications, alone or in combination with a high-molecular weight cytokeratin. These results have not been validated in large, prospective series,

however (Harton et al. 2007; Aiad et al. 2011; Tanaka et al. 2016).

- High rate of insufficiency when aspirating microcalcifications or other non-palpable lesions under imaging guidance due to a paucity of epithelial tissue present. As a result, there is a general agreement that microcalcifications should be assessed by CNB and not by FNAC. This is particularly important in the context of the screening of breast cancer using mammography, which often detects malignancies through microcalcifications.

These perceived drawbacks and the ease of use of CNB have led to a decline in FNAC of the breast, particularly in the evaluation of primary breast lesions. As we have seen, this is not totally warranted. And particularly so in the setting of metastatic disease. Due to their minimally invasive nature, FNACs allow for a quick confirmation or exclusion of disease progression and also enable the gathering of material for repeat testing of biomarkers, as currently recommended by clinical guidelines. They may be used for the confirmation of multicentric disease and for the aspiration of regional lymph nodes, enabling staging of a patient's disease and planning of suitable therapy (Francis et al. 2019). As previously stated, FNAC can be performed quickly and in an outpatient setting. When healthcare resources are otherwise constrained (such as in the COVID-19 pandemic) or limited, the advantages of FNAC may outweigh the disadvantages (Pinto and Schmitt 2020).

FNAC or CNB are complementary, and both techniques should be available. The choice ought to be made based on the particular circumstances and a conjunction of clinical, radiologic, and pathologic findings.

3.3 Breast FNAC Procedure

3.3.1 Equipment

The equipment necessary to perform an FNAC is quite simple. In our practice, breast FNAC is performed as an outpatient service in the clinic. Patients are usually booked in advance, by letter of referral or by telephone. The staff and equipment requirements are minimal and include an assistant (particularly important when performing image-guided FNAC), an examination couch, a writing desk, a work surface, an examination tray (containing instruments), and good lighting. The work surface should ideally have access to running water (i.e., a sink). Consumables should be placed on the work surface, perhaps inside a tray and including: cotton pads/gauze and an antiseptic solution for skin antisepsis before puncture; gloves for hygiene and protection; a syringe, needle, and a syringe holder for the aspiration procedure; glass slides for smear preparation; liquid fixatives, preferentially 95 or 100% alcohol or formaldehyde, for cell block preparation; a small bandage, for post-procedure hemostasis; and a disposal boxes for needles and another for material contaminated with biological fluids (Fig. 3.1).

By placing these materials inside a small box, one can arrange a simple kit enabling the performance of FNACs at any location, be it the radiologist's room, a clinician's office, or an internment ward. If ROSE is to be done, a microscope and quick staining kit should also be available.

Needles are a crucial element of the FNAC procedure and should be chosen with care. In terms of diameter, experienced cytopathologists advocate for the use of needles with 23G or smaller, although others use needles up to

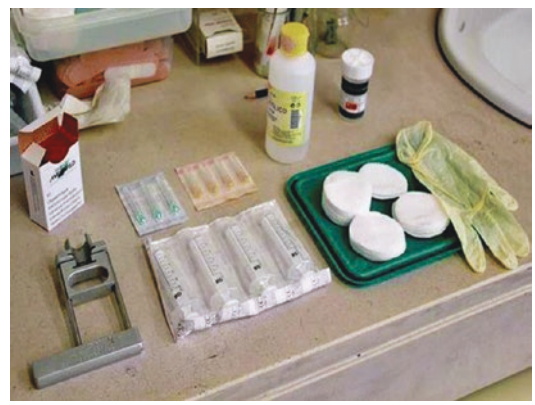

Fig. 3.1 Basic equipment for performing breast FNAC

27G. There are advantages to a smaller diameter: they result in less pain for the patient and in lower rates of complications (such as hemorrhage). As for length, it depends on the target lesion. Most breast lesions can be reached with a 25–30 mm needle.

Syringes are also an integral part of the procedure. Transparent plastic, disposable, sterile syringes are preferred. Most pathologists and clinicians use 10 ml syringes for FNAC, but larger 20 ml ones may also be employed and are particularly useful in the context of large cystic lesions. It is important to remember, however, that in most cases larger syringes will not provide larger or better samples: on the contrary—since the pressure of the vacuum is larger, these may result in more hemorrhage and a higher number of inadequate samples, particularly if the target lesions are highly vascularized.

Finally, the syringe holder. This device allows for suction and release of the syringe to be accomplished with one hand, freeing the other to locate and stabilize palpable lesions. Several types are available, including those made of metal or plastic. The authors prefer those made of metal, which have a higher durability while still weighing below 200 g. As a last piece of advice, the pistol-grip syringe holder allows for the creation of large vacuums, which, as we have discussed, may result in the aspiration of blood and little else. A good quality aspiration sample should fill just the needle hub and not the syringe.

3.3.2 Lesion Localization

Palpable and non-palpable breast lesions can be submitted to the FNAC procedure. Breast aspiration should preferably be guided by imaging, but it is largely accepted that palpable lesions can be sampled using only palpation as a guide.

3.3.2.1 Palpation

The localization of a nodule may be obvious on simple observation, and a thorough palpation of the breast is not necessary for every procedure. However, when the lesion is not easily identified, a good clinical examination of the breast should be performed. In this situation, it is best to examine the patient in the supine position, forearm up and underneath the head, palpating the breast systematically, nipple to axillary tail in a circular fashion. The examining hand should be kept flat with no spacing between fingers. A pillow or towel placed under the shoulder can aid in the appreciation of a subtle nodule. Aspiration may be done in the supine position. However, this can at times prove difficult, due to the fact that the breast will flatten out moving the lesion closer to the chest wall and increasing the risk of pneumothorax. The risk can be mitigated by using a tangential approach, but this is not ideal, since it may lower the diagnostic yield.

Thus, it is usually better to ask the patient to stand upright after the lesion has been located and identified. In the upright position, a mass within the breast will become suspended by gravity and more tethered to the surrounding connective tissue. This added immobilization of the mass enhances material acquisition and moves the nodule away from the chest wall, decreasing the risk of pneumothorax (Staerkel and Sneige 2006).

In contrast to deep organ sites, breast aspirations benefit from the fact that they are not impeded by overlying structures such as muscle. However, given that the breast is largely made of adipose tissue, lesions contained within are usually mobile, which may hinder adequate sampling. Consequently, it is crucial to immobilize the mass so that when a needle is introduced, a coring action is achieved. This can be done by placing the fingers on each side of the mass, stretching the skin, and flattening the breast. When possible the fingers should be placed along the axis of greatest mobility. This procedure also enables almost any mass to be reached with a 30-mm needle. Sometimes, because of the tumor's depth within the breast, the large size of the breast, and/or the infiltrative nature of the mass, a lesion will lack definition. On palpation, only a vague firmness will be appreciated, usually felt over a large portion of the breast. In this context, aspirations may yield little significant tissue, even when multiple passes are performed. This is typically due to an erroneous evaluation

on the part of the operator: since the mass is deep seated, its firmness may be mistaken for the chest wall, leading to superficial aspiration attempts in fear of risking a pneumothorax.

An additional difficulty may arise in this context, if there is a tendency to approach lesions with the needle positioned tangentially to the skin, rather than with a near vertical or perpendicular approach. This tangential positioning may be more comfortable for the aspirator but it is not optimal for small deep-seated nodules. By the time the needle has reached the level of the lesion, little of the nodule is in the path of the needle. Therefore, a more vertical approach is recommended, even if it feels uncomfortable to some aspirators. The best approach to fix the nodule appropriately is to use the middle and index fingers for immobilization, instead of the commonly used thumb and index finger (Staerkel and Sneige 2006).

3.3.2.2 Ultrasound-Guided FNAC

Nowadays, ultrasound is frequently used to guide breast FNAC, even if lesions are palpable. This technology enables the cytopathologist to visualize the lesion and choose the right area for aspiration (for example, solid areas, instead of cystic or necrotic areas—Liao et al. 2004). There are two methods which can be used for guidance, depending on the site of the lesion: in the first one the transducer probe is used to locate the lesion and place it in the middle of the ultrasound field. The operator then makes a mark using the transducer to pressure the underlying skin, circumscribing the puncture site; this is followed by passing the needle through the skin and advancing it slowly into the lesion, in a way that the transducer probe can be placed perpendicular to the needle, guiding its movements. In the second method, the FNAC procedure is guided step by step so that the ultrasound field encompasses the entire path of the needle, from the moment it pierces the skin and until it is removed. The transducer probe is used to locate the lesion in one of the edges of the ultrasound field; the aspirator then passes the needle through the skin at the edge of the probe and in parallel to it. It is possible, then, to guide the needle into the lesion, avoiding passing through vascular structures that might be in the way of the needle to the lesion. This latter method is preferred to guide the needle in FNAC of the breast (Fig. 3.2).

3.3.3 Aspiration Procedure

FNAC of the breast is a simple procedure. Aspiration of most lesions is painless, and the patient feels only the initial pinprick through the skin. Anesthesia is not required for most breast aspirations. One of the keys to performing an adequate aspiration is the immobilization of the lesion by the aspirator's free hand, which enables better cutting and coring of the mass. Once the lesions are immobilized, as described above, the needle, with a syringe and holder attached is inserted into the mass. Then, the syringe plunger is pulled back, creating negative pressure, as the needle is moved back and forth in a rocking motion. It is not the suction that directly results in obtaining a sample but rather the cutting action of the needle. The suction helps to pull tissue into the cutting path of the needle and to move the resulting fragments up into the needle's shaft (Fig. 3.3). Pumping the syringe plunger does not enhance sampling; in fact, sampling can be reduced as a result of increased bleeding. In general, needle movements that are more frequent, longer in length,

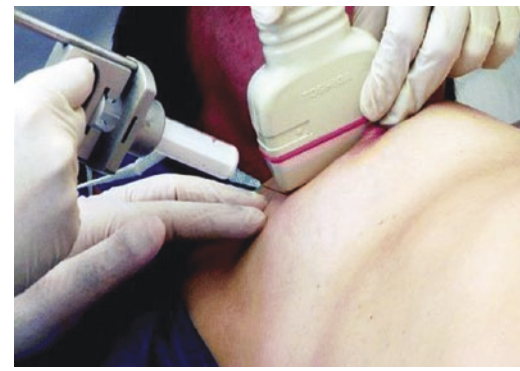

Fig. 3.2 Ultrasound-guided breast FNAC. The transducer probe locates the lesion in one of the edges of the ultrasound field; the aspirator passes the needle through the skin, in parallel with the transducer probe at the edge where the lesion is located in

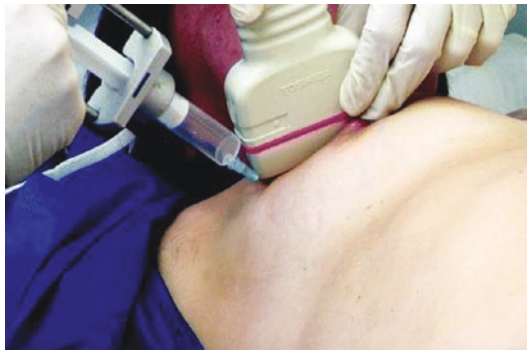

Fig. 3.3 Applying suction while moving the needle helps to pull cells into the needle. In general, needle movements that are more frequent, longer in length, and kept within the tumor during the entire aspiration yield more tissue. A blood-tinged specimen will appear in the hub of the needle. Suction is then released, and the needle is withdrawn

and kept within the tumor during the entire aspiration yield more tissue. Movement and frequency will depend on the size of the lesion and the aspirator's ability to maintain control. Typically, 30–50 excursions with the needle should be made over a 10–20 s period. A blood-tinged specimen will appear in the hub of the needle. Suction should then be released, and the needle is withdrawn. Although little bleeding occurs in fine needle aspirations, it is best to avoid all bleeding, as it can decrease cellular yield and lesion demarcation. Therefore, after withdrawing the needle, a gauze pad should immediately be applied over the puncture site and pressure applied, for at least 1–3 mins (a longer time should be applied in individuals with an easy bruising history or for those currently taking blood-thinning agents).

After removing the needle from the patient, the syringe should be detached from the needle. The plunger should be pulled back, filling the syringe with air. Leaving it in this position, the needle should be reattached. The aspirator then places the needle close to a glass slide, almost touching, and uses the plunger to expel a drop of the sample acquired onto the slide, repeating the procedure in as many slides as necessary until there is no material left. The drops are then smeared, fixed in 95% ethanol or air-dried, and then stained for interpretation.

The procedure outlined above is the most frequently used and is valid for both palpable and non-palpable lesions. There is an alternate technique, however, which is worthy of mentioning, the so-called capillary method. Most steps are similar to what was previously described, but neither a syringe nor a syringe holder is used. Instead, the needle is held directly by the aspirator by gripping the hub. This allows for a heightened appreciation of tissue density and an overall greater sensitivity to movements. Virtually no bleeding occurs; since there is no suction, frequently no material will be seen in the needle hub and the aspiration has to be voluntarily stopped after 15–20 seconds without visual feedback. After the needle is withdrawn, an air-filled syringe is attached, and material is expelled onto slides as previously described. This method may result in low cellular yields in fibrotic and sparsely cellular lesions. If the lesion is a cyst, rapid leakage of fluid from the end of the needle due to pressure may happen. An empty recipient should be kept at hand for this eventuality.

Usually, a single aspiration is sufficient for diagnosis. However, additional aspirations are recommended in certain circumstances that the aspirator should be aware of (Arkoumani and Wells 2006):

- Lesions that are fibrous in nature; can be felt through palpation and appreciated in ultrasound; a fibrous stroma may represent a desmoplastic carcinoma which will tendentially have a low cell yield.
- Small lesions, due to the risk of missing the target.
- A specimen that clots immediately when placed on a glass slide, since this is evidence that significant bleeding has occurred and a diluted/hypocellular sample has been obtained.
- If the sample looks as smooth as a peripheral blood smear, without visible particles.
- If the aspirate is yellow and slightly oily in appearance; this indicates that the specimen is primarily composed of adipose tissue frequently with little or no epithelium; this may be acceptable in some contexts, however,

namely if one is relatively confident that a nonmalignant process is being evaluated—that is, a nodule with low clinical and imaging suspicion, which is soft on palpation and offers little to no resistance on needle penetration.

- Breast cysts: these require complete drainage and the drained area should be reevaluated by palpation or imaging for any residual solid mass, which, if present, will in turn require a separate needle aspiration.

3.3.4 Preparation of Smears

Glass slides should be clean and ready to use. They should be labeled using a pencil at the frosted end. The labeling can include the patient's name (initials), identification number, or site of aspiration. It is safer if at least two identifiers are used.

Preparation of high-quality smears is one of the most important parts of the aspiration procedure itself (Stanley and Lowhagen 1993). The aim of preparing a smear is to obtain a homogenous layer of well-preserved cells, concentrated in a small area of the slide, which makes the microscopic analysis easier and quicker. Cells should be spread over the slide surface by gentle pressure so that they are not crushed. If the smears are not interpretable, then a reliable diagnosis cannot be made, no matter how adequate the aspirated specimen is or how much experience the cytopathologist has. Keep in mind that an FNAC smear is not a blood cells analysis smear and one does not need the thinnest smear, but one that can maintain some of the lesion architecture, without being thick or crushing the cells.

There are two basic methods of smearing: one-step and two-step methods (Stanley and Lowhagen 1993). The one-step method is preferentially used on small-volume specimens obtained from solid lesions. To perform the smear, the slides should be held as shown in Fig. 3.4. The specimen droplet is placed near the slide label. This specimen slide is then held by the physician's dominant hand in a vertical position. Another slide is used as a spreader, held by the contralateral hand, placed perpendicularly over the other slide, at an angle, so that its superior edge is poised above the specimen droplet. Then, smoothly, the spreader slide is lowered until it touches the specimen slide, homogenizing the droplet, and, while applying a constant and gentle pressure, a downward motion is performed, drawn along the length of the lower slide. The surface of the spreader slide must always be parallel to the surface of the specimen slide, and the smear should finish before the end of the specimen slide, occupying the smallest area of the slide possible.

Fig. 3.4 Preparation of smears using the direct one-step technique. The lower slide holds the material, while the upper slide is used as a spreader slide. The spreader slide is poised over the material droplet, with its lower edge forming a hinge-like contact with the lower slide

The two-step method is used for liquid or hemorrhagic specimens within which cells and tissue particles are suspended. To perform the smear, the slides should be held as shown in Fig. 3.5. The fluid sample is placed from the middle to the labeled edge of the slide. The spreader slide is held at a 45° angle to the specimen-bearing slide, and its end is brought into contact with the fluid. Then, the spreader slide advances toward the specimen slide's label, carrying the fluid and suspended particles. The surface tension causes the fluid to spread out in a line behind the edge of the spreader slide. The spreader slide, thus, returns in the opposite direction, stopping in the middle of the specimen slide, where the tissue particles remain concentrated. The spreader slide is quickly pulled away from the specimen slide,

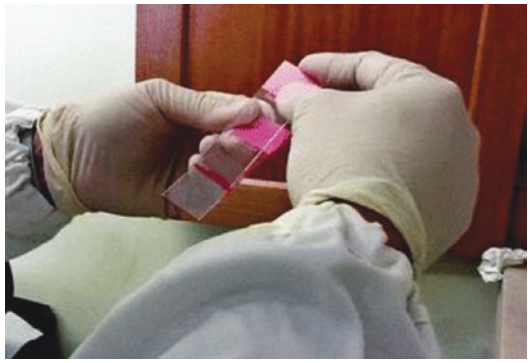

Fig. 3.5 Preparation of smears using the two-step method. Observe the concentration of the material in the middle of the slide that will be smeared according to the one-step technique

which is turned to one side in order to drain the excess fluid. After that, the spreader slide is turned perpendicular to the specimen slide, and the line of sediment tissue particles is smeared as it is in the one-step method. This technique is more complex, and the smears are not as good-looking as in the one-step method, but it allows a better smear quality to fluid or hemorrhagic samples: the tissue particles are concentrated in the middle of the slide, which makes microscopic observation easier, and the excess of fluid or blood is removed, allowing rapid drying and better fixation of the slide.

Another technique should be kept in mind. In certain circumstances, it may be necessary to prepare more than one smear from a single droplet of material. This may happen for different reasons, for example, to divide material across multiple slides for ancillary tests or when too much liquid is placed on a single slide accidentally. In the first case, the entire sample should be expelled onto a single glass slide. The slides are then held as described at the beginning of the one-step method. One of the corners furthest away from the frosted end of the spreader slide is quickly brought in contact with the sample, carrying a part of it. The spreader slide can then be turned around, and the opposite corner touched to the slide as well. The corners closest to the frosted end may also be used in a similar fashion. This can be done at least four times with the same specimen slide. Each portion of the original sample in the spreader slide is then smeared onto a new slide, and in the end this slide is used to smear the original sample, following the one-step method.

Other than smearing, samples may be directly expelled to a vial for several purposes. This may be particularly useful in liquid or bloody specimens. The vial may be filled with saline or a fixative, such as formaldehyde or ethanol. The former, enables a sample to be sent for flow cytometry, for example, while the latter two enable the preparation of cell blocks and liquid-based cytology slides. Cell blocks are particularly useful for immunohistochemistry.

In the authors' experience, processing of aspirated materials by liquid-based technique should only be used in specific situations, such as those mentioned above. For routine morphological observation, it is preferable to prepare smears, given their simplicity and better preservation of architectural features and extracellular matrix.

3.3.5 Fixation and Staining

Although Papanicolaou stain is the most widely used staining method in cytology, most cytopathologists prefer to use a combination of two stains in FNAC, which are complementary: Papanicolaou and Romanowsky. Papanicolaou staining includes the hematoxylin nuclear stain and two cytoplasmic counterstains, orange G and EA. There are at least three types of stains under the designation of the Romanowsky method—Giemsa, May-Grünwald-Giemsa, and Diff-Quik—but all three present the same staining pattern that characterizes this method. Some cytopathologists prefer to stain FNAC slides using hematoxylin and eosin (H&E) as in histology, mainly due to better familiarity with this stain.

The smears prepared after the aspiration procedure can be either intentionally air-dried for Romanowsky staining or immediately fixated in 97 or 70% ethanol for Papanicolaou or H&E stains. The air-dried smears are submitted to post-fixation with methanol.

The best staining is obviously the one with which the cytopathologist is most familiar with, but each method has its advantages, and they work better together. The air-drying required for the Romanowsky method results in cell swelling, and the cells tend to look larger than in Papanicolaou or H&E methods. Romanowsky stains have the ability to react with several tissue components in a metachromatic way, giving them a reddish-purple color. This can be observed in nucleic acids, mucins, and extracellular matrix components, such as in fibroepithelial tumors and metaplastic carcinomas.

On the other hand, nuclear details are not the strength of the Romanowsky method. Nuclei not only look larger using these stains, but they also lack chromatin and membrane contour details. For this reason, a Papanicolaou stain should be performed whenever the nuclear detail is important for the diagnosis or the subclassification of tumors.

While in a Papanicolaou-stained sample nuclear detail provides important diagnostic clues, one has to bear in mind that FNAC usually provides highly cellular smears in which the tissue architecture is almost always maintained. The tumor cells are closely related to the stromal and extracellular matrix elements, and sometimes, evaluation of other additional parameters such as smear pattern, cellularity, and nuclear size may be sufficient to allow a specific diagnosis. The assessment of an FNAC may thus be supplemented by Romanowsky stains. H&E stain has the same characteristics in FNAC as it has in histologic sections, and for this reason, many pathologists prefer to use this method. Nevertheless, as Romanowsky and Papanicolaou stains are complementary stains and provide additional information, they are recommended to be used preferentially in cytological specimens.

3.3.6 Reporting of the Results

Reporting the results is one of the most important parts of the FNAC procedure. The cytopathologist should have in mind the possible consequences of the report and be cautious when making a suspicious or inconclusive diagnosis. Comments and explanatory notes should be used judiciously and only if they are thought to be helpful to the clinicians in making a therapeutic decision. Negative results should always be correlated with clinical and imaging findings in the context of the triple test. If the aspirated lesion is suspicious for malignancy in the clinical or imaging setting, a negative result in the FNAC specimen may not rule out malignancy, and an alternative method, such as CNB or surgical excision should be performed in order to obtain a reliable diagnosis.

The authors recommend the use of the Yokohama System for Reporting Breast Fine Needle Aspiration Biopsy Cytopathology, which divides breast FNAC diagnoses into five categories, with well-defined risks of malignancy: insufficient for diagnosis, benign, atypical, suspicious for malignancy and malignant. The risks of malignancy and reproducibility of criteria for each of these categories have been validated in the literature (Montezuma et al. 2019).

Using these categories, communication with clinicians is improved, enabling better patient management. Additionally, the Yokohama system provides a framework for standardized reporting, while at the same time being adaptable to different realities. Namely, the system states that all reports should include not only the relevant category (in full, never as a code), but also a clear cytological description, including cellularity and presence or absence of key diagnostic features. A concise comment or conclusion should also be included, providing a diagnosis that is as specific as possible, or, in alternative, a list of most likely differential diagnoses.

When appropriate, synoptic reports may be used, providing a checklist for the cytopathologist, and streamlining communication with clinicians.

Based on the recommendations of this classification system, a report for breast FNAC should be structured as a report in surgical pathology, providing all the clinical information, including specimen type and localization technique, followed by a microscopic description and the diagnostic conclusions, including the final diagnosis

and category in full. An additional comment may be added, including recommendations to correlate the cytology with clinical and imaging findings and/or the need for clinical follow-up or biopsy of the lesion for histologic assessment.

3.4 Clues to Enhance Diagnostic Accuracy

Some additional information is essential to enhance breast FNAC diagnostic accuracy. Cytopathologists should pay particular attention to these:

- Patient age. Different pathologies have different prevalences across the age spectrum. For example, advancing age increases suspicion for carcinoma and decreases the likelihood of fibroadenoma.
- Lesion location. Different lesions arise in different areas of the breast. For example, if the location is subareolar, one should consider the possibility of a papillary neoplasm, nipple adenoma, or subareolar abscess.
- A cystic lesion suggests fibrocystic disease. Exception occurs when the aspirate is markedly cellular with single columnar cells; then one should consider a papillary neoplasm. In addition, the acquisition of thin, watery green-gray fluid typical of benign cyst fluid of fibrocystic changes should caution against an overdiagnosis of carcinoma even when some of the cells present show degenerative nuclear atypia.
- Previous trauma or surgery at the aspiration site requires careful exclusion of a reactive/reparative process, such as fat necrosis, before a malignant diagnosis is made.
- Past history of another malignancy. The breast can be involved by other neoplasms such as melanomas, lymphomas, or metastatic carcinomas from other sites.
- Needle penetration findings are also helpful. Soft, low-resistance aspiration suggests benign disease, fat necrosis, or mucinous carcinoma, whereas firm, rubbery texture favors fibrocystic changes or fibroadenoma. A firm, gritty sensation suggests carcinoma.

The diagnosis derived from breast FNAC should always be correlated with both clinical and radiological findings to determine patient management (the triple test—Kocjan et al. 2008; Arkoumani and Wells 2006). Benign triple test results in the patient being followed clinically with a return visit in 6 months or 1 year. A patient with malignant triple results is referred for definitive therapy. A mixed (inconclusive) triplet requires incisional or excisional biopsy of the lesion in question.

3.5 Complications of Breast FNAC

Complications occurring in breast aspirations are rare and, when they occur, are usually bleeding, infection, and/or pneumothorax. The most likely complication is soft tissue bleeding with a resulting hematoma; however, this can be avoided if, after aspiration, firm pressure is applied directly over the puncture site. Infection is uncommon but when present is of little consequence and may be treated with antibiotics. Pneumothorax is extremely rare but, when present, may require chest tube insertion. In individuals with small breasts where an underlying rib can be palpated, one can position a mobile nodule over a rib for protection. Also, for deep-seated lesions adjacent to the chest wall, the use of a 25-gauge needle instead of a larger bore needle minimizes injury and therefore the risk of pneumothorax.

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