



Fine-Needle Aspiration Cytology and Exfoliative Cytology

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1 Exfoliative Cytology

1.1 General Principles

- Obtaining surface cells for subsequent evaluation by conventional microscopy.
- Easy and quick to perform, low cost, and noninvasive method.
- Technique well accepted by patients.
- No need for local anesthesia.
- Rapid diagnosis.
- It can be repeated several times and used in population screening and clinical studies.

1.2 Indications

Although not commonly used in these groups, they can be useful and indicated (Figs. 1 and 2) in the following:

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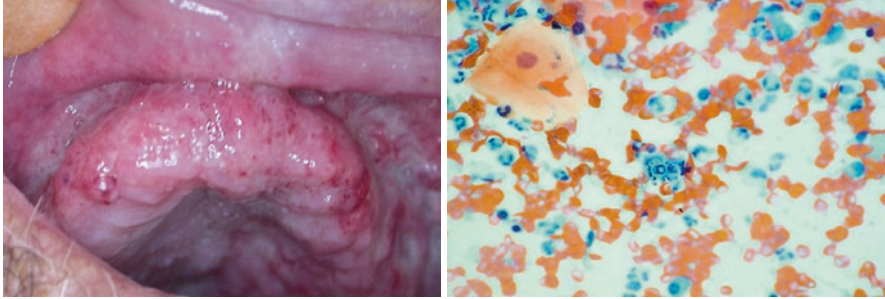


Fig. 1 Paracoccidioidomycosis. Ulcerated lesion of mulberry-like appearance with hemorrhagic spots located in the alveolar ridge that extends to the sulcus and hard palate. Multinucleated giant cells containing birefringent yeast-like structures are characteristic of the fungus *Paracoccidioides brasiliensis* (Papanicolaou, 630×). In detail, rounded cells presented cryptosporulation with a “steering wheel” appearance of the fungus *Paracoccidioides brasiliensis* (Papanicolaou, 630×)

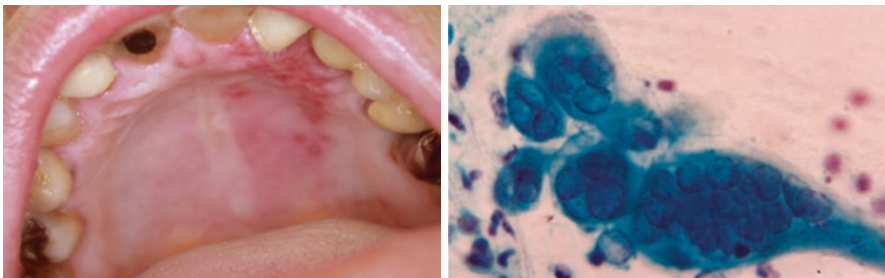


Fig. 2 Herpes simplex. Shallow, punctate ulcers with an erythematous halo, located on the hard palate. In addition, parasitized epithelial cells had enlarged nuclei and scattered chromatin, multinucleation, and giant cell formation (Papanicolaou stain, 630×)

- Diagnostic aid in autoimmune diseases, such as pemphigus vulgaris.
- Identification of bacterial (abscesses, actinomycosis), fungal (candidiasis, paracoccidioidomycosis), and viral herpes simplex and human papillomavirus infectious lesions.

1.3 Materials

Materials included (Fig. 3) are the following:

- Disposable cytobrush (cervical brush) or sterile metal spatula.
- Clean and dry microscopy glass slides with frosted ends.
- Fixed: 95% ethyl alcohol (96°GL) or cytological spray fixative.
- Slide mailer or microscope slide jar to keep the slides.

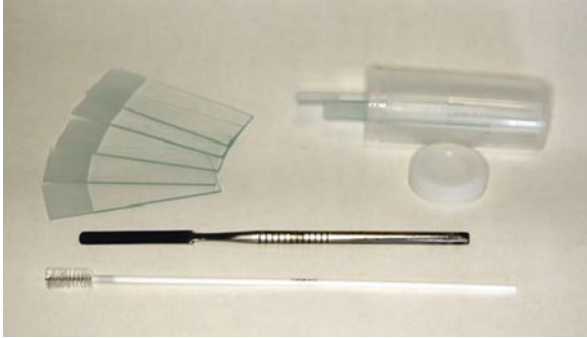


Fig. 3 Material needed for the cytopathological exam. Metallic spatula and cytobrush for sample collection. Glass slides with the frosted end where the sample is distended. The frosted end can be labeled with the patient's name and date or place of sample collection. Microscope slide jar with fixing solution

1.4 Technique

The technique (Fig. 4) consists of the following:

- Rotate the cytobrush in the lesion to be analyzed or scrape the area with a metal spatula. In the case of vesicular lesions, rupture the vesicle for material collection.
- The cells are transferred to a glass slide previously dried and cleaned.
- The material must be spread on the glass slide with rotational movements with the cytobrush or in a thin layer with the metal spatula, usually three slides for each staining.
- Fix immediately with cytological spray fixative or condition the slides in a slide holder containing 95% ethyl alcohol (96°GL).
- Staining: Papanicolaou, Hematoxylin and Eosin (HE), Periodic acid–Schiff (PAS).



Fig. 4 Collection of material with a cytobrush or metallic spatula, followed by smearing of the collected material on a glass slide with frosted edge. When performed with cytobrush, in rotating movements and with a metallic spatula, in a unidirectional motion. Smear fixation with cytological spray fixative at a distance of approximately 40 cm or place slides in a microscope slide jar with a screw cap containing 95% ethyl alcohol (96°GL) for fixation and subsequent sending to the cytopathology laboratory

2 Fine-Needle Aspiration (FNA)

2.1 General Principles

- It contributes to diagnosis and treatment planning, as it allows the cytological study of a wide group of intra–/extraoral nodular diseases.
- Simple, fast, safe, inexpensive, and accurate technique.
- Well tolerated by patients.
- The patient may present with hematoma, local pain, and infection following FNA.

2.2 Indications

- Diagnosis of nodular diseases, lymph node enlargement or metastases in the head and neck region.
- Cancer staging and monitoring of recurrences.
- Diagnosis of salivary gland tumors.
- Diagnosis of infectious diseases such as paracoccidioidomycosis, tuberculosis, actinomycosis, leprosy, and toxoplasmosis involving lymph nodes among others.
- Diagnosis of lymphomas.
- Diagnosis of adverse reactions to aesthetic fillers, dermoid, epidermoid, and branchial cleft cysts, and other soft tissue diseases.

2.3 Contraindications

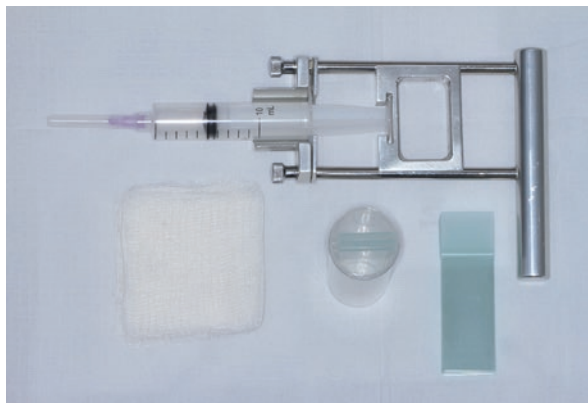
Carefully evaluate the indication of FNA for patients using anticoagulants or presenting coagulation disorders.

2.4 Materials

Materials used (Fig. 5) are the following:

- Sterile gauze, sterile gloves, and antiseptic solution.
- 24 G disposable fine needle (20 × 5.5 mm)
- Disposable syringe of 10 mL or 20 mL supported on syringe mechanical support to facilitate aspiration.
- Clean and dry microscope glass slides with frosted ends.
- Fixative: 95% ethyl alcohol (96°GL), cytological spray fixative, or dry fixation.

Fig. 5 Material needed for the FNA exam. The disposable syringe was supported on an aspiration gun to facilitate aspiration. Glass slides with the frosted end where the sample is distended. The patient's name and date or place of sample collection may be identified on the frosted end with a pencil. Slide holder case with fixing solution



2.5 Technique

The technique (Fig. 6) consists of the following:

- Local antisepsis with 0.1% iodinated alcohol, 10% povidone-iodine (topical PVP-I), or 2% chlorhexidine digluconate solution.
- Choose the best site for needle penetration before starting the procedure. In the case of the intraosseous lesion, introduce the needle in the cavity lumen through a thin area of cortical bone or perforation point, if present.
- Insert the needle almost perpendicularly toward the lesion site.
- Conduct quick and accurate forays in multiple directions for diverse sampling.
- Pull the plunger and observe the presence of material.
- Release the plunger, withdraw the needle, and press the site with gauze.
- Deposit a small amount of material directly on the slide for swab preparation, 2–3 mm in diameter, near the frosted end of the slide.
- With the tip of another slide inclined at 45 degrees, perform the distension of the material in a single movement in a uniform and smooth manner, generally, three slides for each staining.
- Fix by air-drying to be stained by the rapid panoptic technique and in 95% ethyl alcohol (96°GL) for Papanicolaou's technique and HE.
- Staining: Panoptic, Papanicolaou, and HE.
- Immunohistochemistry can be performed on paraffin-embedded material.



Fig. 6 Nodular lesion in the parotid region submitted to FNA. It is important to note that a small amount of aspirated material should be placed close to the frosted end of the slide. With the tip of another slide inclined at 45 degrees, a movement must be done until it touches the material. Slide the glass slide over the other slide in the direction opposite to the frosted end

Acknowledgments The São Paulo State Research Foundation (FAPESP, São Paulo, Brazil, grant number JFS 19/09419-0) and the Coordination of Training of Higher Education Graduate Foundation (CAPES, Brasilia, Brazil, finance code 001).

Sources

- Acha A, Ruesga MT, Rodríguez MJ, Martínez de Pancorbo MA, Aguirre JM. Applications of the oral scraped (exfoliative) cytology in oral cancer and precancer. *Med Oral Patol Oral Cir Bucal*. 2005;10(2):95–102.
- Almeida JD, Lima CF, Brandão AA, Cabral LA. Evaluation of staining methods for cytologic diagnosis of oral lesions. *Acta Cytol*. 2008;52(6):697–701.
- Bertini F, Costa NC, Brandão AA, Cavalcante AS, Almeida JD. Ulceration of the oral mucosa induced by antidepressant medication: a case report. *J Med Case Rep*. 2009;3(3):98. <https://doi.org/10.1186/1752-1947-3-98>.

- Cabral LA, Lima CF, Coutinho de Oliveira ML, Brandão AA, Almeida JD. Oral exfoliative cytology for the diagnosis of paracoccidioidomycosis in a patient with human immunodeficiency virus: a case report. *Acta Cytol.* 2010;54(6):1127–9.
- Cabral LAG, Penteadó AS, Brandão AAH, Almeida JD. Fine needle aspiration cytology in the diagnosis of multiple myeloma. *Health.* 2012;4(8):470–3.
- Díaz KP, Gerhard R, Domingues RB, Martins LL, Prado Ribeiro AC, Lopes MA, Carneiro PC, Vargas PA. High diagnostic accuracy and reproducibility of fine-needle aspiration cytology for diagnosing salivary gland tumors: cytohistologic correlation in 182 cases. *Oral Surg Oral Med Oral Pathol Oral Radiol.* 2014;118(2):226–35.
- Faria KM, Fonseca FP, Silva WG, Silva RN, Vargas PA, de Almeida OP, Lopes MA, Santos-Silva AR. Fine needle aspiration cytology in the diagnosis of perioral adverse reactions to cosmetic dermal fillers. *Oral Surg Oral Med Oral Pathol Oral Radiol.* 2014;117(5):e393–5.
- Leite AA, Vargas PA, Dos Santos Silva AR, Galvis MM, de Sá RS, Lopes Pinto CA, Kowalski LP, Saieg M. Retrospective application of the Milan system for reporting salivary gland cytopathology: a cancer center experience. *Diagn Cytopathol.* 2020;48(9):821–6. <https://doi.org/10.1002/dc.24464>.
- Lingen MW, Abt E, Agrawal N, Chaturvedi AK, Cohen E, D'Souza G, Gurenlian J, Kalmar JR, Kerr AR, Lambert PM, et al. Evidence-based clinical practice guideline for the evaluation of potentially malignant disorders in the oral cavity: a report of the American dental association. *J Am Dent Assoc.* 2017;148(10):712–727.e10.
- Marques SMBM, Pereira JTA. Protocolo de procedimentos do serviço de diagnóstico por imagem. Hospital Municipal e Maternidade Escola Dr. Mário de Moraes Altenfelder Silva. Coleção Protocolos HMEC. São Paulo, 2016. 5ª Edição.
- Pérez-de-Oliveira ME, Petersen Wagner V, do Amaral-Silva GK, Almeida Leite A, Ajudarte Lopes M, Santos-Silva AR, Jorge Júnior J, de Almeida OP, Agustin Vargas P. An audit of cytopathology in the oral and maxillofacial region: 18 years of experience. *Cytopathology.* 2020;31(6):555–63. <https://doi.org/10.1111/cyt.12891>. Epub 2020 Aug 16
- Queiroz JB, Lima CF, Burim RA, Brandao AA, Cabral LA, Almeida JD. Exfoliative cytology of the oral mucosa: comparison of two collection methods. *Gen Dent.* 2010;58(5):e196–9.
- Reys IG, Scarini JF, Fontes KBFC, Barki MCM, Azevedo RS, Tucci R. Análise do diagnóstico citopatológico realizado em pacientes atendidos na clínica de estomatologia do curso de odontologia da Universidade Federal Fluminense, Nova Friburgo /RJ. *Revista Odontológica de Araçatuba.* 2017;38(2):46–50.
- Vargas PA, da Cruz Perez DE, Mata GM, de Almeida OP, Jones AV, Gerhard R. Fine needle aspiration cytology as an additional tool in the diagnosis of odontogenic keratocyst. *Cytopathology.* 2007;18(6):361–6.
- Vargas PA, Prado FO, Fregnani ER, Perez DE, Lopes MA, de Moraes M, Gerhard R. Fine needle aspiration biopsy in central giant cell lesion: a report of 3 cases. *Acta Cytol.* 2006;50(4):449–54.