Original Article

Indian Journal of Pathology: Research and Practice
Volume 6 Number 1, January - March 2017
DOI: http://dx.doi.org/10.21088/ijprp.2278.148X.6117.9

Correlation of Imprint, Scrape and Crush Cytology with Conventional Histopathology in Diagnosis of Benign and Malignant Lesions

Adlekha Shashikant*, Chadha Tandra**

*Associate Professor, Department of Pathology **Associate Professor, Department of Microbiology, Maharaja Institute of Medical Sciences, Vizianagram, Dist. Vizianagram, Andhra Pradesh, India.

Abstarct

Background: Cytological tools, imprint and crush smear techniques are simple and rapid techniques in diagnosis of various lesions. *Aims and Objectives:* 1. To evaluate the utility of imprint, scrape and crush cytology as diagnostic modality. 2. To study the advantages and limitations of cytological techniques. 3) To correlate, compare and contrast with histopathology examination. *Material and Methods:* A prospective study, comprising of 200 cases of various specimen, submitted in histopathology department. Smears by different cytological techniques were stained by rapid H&E , PAP stain and Giemsa stains. *Results:* Imprint and crush smears showed cent percent diagnostic accuracy in diagnosing CNS lesion, Genitourinary, Bone, Testis and skin and soft tissue lesions. Lesions from Breast, Gastrointestinal lesions, Tumor resection margins , also showed high diagnostic concordance between cytological methods and histopathological examination. *Conclusion:* Imprint, scrape and crush smear methods are rapid cost effective techniques of obtaining accurate diagnosis of benign and malignant lesions.

Keywords: Imprint Cytology; Crush Cytology; Scrape Cytology; Histopathoogy.

Introduction

Rapid diagnosis of surgically removed specimens has created many ripples and controversies and a single completely reliable method has not yet been developed. Histopathology of a paraffin section remains the ultimate gold standard in tissue diagnosis. The utilty of frozen sections is well known and is being routinely used by the surgical pathology laboratories for intraoperative diagnosis. Many studies have been done in the past to evaluate the role of cytology in intraoperative diagnosis of tumor. These studies have concluded that cytology has the advantage of being much less time consuming, easy to adopt, reliable and does not require special instruments[1,2]. Hence, cytology methods can be employed routinely in the intraoperative diagnosis in conjunction with frozen section. It can be used to diagnose small tissue that can be preserved for permanent paraffin block method. The use of either frozen section or cytological examination alone has an acceptable rate (93–97%) of correct diagnosis, with regard to interpretation of benign versus malignant [3-6]. Commonly used methods for obtaining and preparing cells for cytological evaluation are touch preparation, fine needle aspiration cytology (FNAC), scrape and crush smear preparation. Scrape and crush preparations yield higher cellular smears, compared to imprint cytology. In addition, cytological examination of surgical specimens has proved to be a valuable learning tool and has educational value. The skill and expertise developed by routinely practicing intraoperative cytological technique can be applied to the interpretation of FNAC. This significant educational value coupled with its intrinsic simplicity and rapidity and cost effectiveness will likely necessitate the widespread implementation of this diagnostic technique in the near future.

The diagnosis of *H pylori* infection entails invasive and non invasive tests. The invasive tests are

Corresponding Author: Shashikant Adlekha, Department of Pathology, Maharaja Institute of Medical Sciences, Vizianagram, Dist. Vizianagram, Andhra Pradesh – 535217 India, Andhra Pradesh.

E-mail –ruc.isha@gmail.com

(**Received on** 30.11.2016, **Accepted on** 13.12.2016)

endoscopy based tests, which include histipathological examination, cytological examination-Crush and imprint cytology, rapid urease test (RUT) and polymerase chain reaction. Cytological examination, such as imprint and crush smears have been used in detection of malignancy with sensitivity up to 95.2% [7]. These techniques are routinely not used in detection of *H pylori* infection. So we evaluated the usefulness of imprint, squash and crush smears in comparision to conventional histopathological examination .

Material and Methods

200 surgical specimens submitted from various organs and specimens, submitted for histopathological examination in Sree Narayana Institute of Medical sciences and Hospital, Kerala, India were evaluated by imprint and scrape/crush cytology. Gross examination of the specimen of tumor removed from the patient was done by inspection and palpation. The specimen was then cut with a sharp knife into two halves.

The cut surface was wiped off the excess blood, if present, with the help of a filter paper. Again, reinspection and repalpation of the tumor was done. The most appropriate area thought to be representative of lesion was chosen. Imprint/ touch smears were prepared by gently touching the slides, over the cut surface of specimen. Scrape smears were prepared by scrapping with a sharp scalpel or the end of a glass slide, depending upon the type of tissue. Crush/ squash smears were prepared on a CNS tumor by crushing between two glass slides to prepare a smear. On an average, four slides per case were taken from different representative areas. Imprint smears were prepared on all the specimens, crush smears prepared on all endoscopic biopsies and a biopsy from CNS lesion. Scrape smears were prepared on all the biopsies, except for where the crush smears were prepared. The slides were labelled and immediately put into 95% ethyl alcohol and stained with rapid H& E and Papanicolaou stains.

The slides were examined immediately and reported as benign or malignant, with tentative diagnosis. Total time taken for smear preparation, staining and reporting was about 10 minutes. The specimens were then fixed in 10% formal saline. Sections were taken from the same area from where scrapings were taken. Paraffin blocks of the sections were processed in the routine way and 5 µm thick sections were stained with hematoxylin and eosin (H and E).

130 patients undergoing upper gastrointestinal endoscopy in the hospital were enrolled in this study. Three different diagnostic methods were used – histology, imprint cytology and crush cytology. Three antral biopsy fragments were obtained from each patient and two samples sent for pathological examination in unfixed state and one sample being sent for RUT. Imprint smears were prepared from one fragment by keeping one biopsy fragment on a glass slide and gently touching it without pressing. Imprint slides were immediately stained for rapid PAP stains, air dried and stained for Giemsa stain. Subsequently crush smears were prepared on the imprinted smear. The second biopsy specimens were fixed in 10% formalin and processed for three micrometre thick sections and stained with H & E and Giemsa stains. *H. pylori* classically appear as small curved or s-shaped structures (Figure 3). Occasional coccoid forms may be seen and are difficult to be interpreted by routinely used stains.

Diagnostic accuracy was computed for all the methods applied.

Informed consent was taken from each patient and the study was approved by scientific research committee of the institution.

The diagnosis obtained by intraoperative imprint , crush smears and scrape smears were compared with final histopathological diagnosis in terms of diagnostic accuracy.

Results

The diagnostic accuracy of imprint and scrape/ crush cytology was 100% for skin and Soft tissue lesions, Bone, CNS, Testis and Genitourinary tract lesions. 9 out of 10 resection margins, were correctly evaluated for tumor involvement, with 1 false negative for tumor involvement, yielding diagnostic accuracy of 90% for both imprint imprinand scrape cytology methods.

127 and 131 gastrointestinal lesions were correctly diagnosed with imprint and crush cytology, yielding diagnostic accuracy of 92.7% and 95.6% respectively. Evaluation of both benign and malignant breast lesions showed diagnostic accuracy of 85% and 95% for imprint and scrape cytology respectively, with 1 case of DCIS being overdiagnosed as infiltrating carcinoma. Evaluation of thyroid lesions, showed diagnostic accuracy of 75% and 100% for imprint and scrape cytology respectively, with one case of colloid goitre with focal papillae, being misdiagnosed as papillary carcinoma in imprint cytology.

Table 1: Distribution of cases diagnosed on cytology and histology

Organs/systems	Cytological Diagnosis Imprint Cytology	Scrape/Crush Cytology	Histopathological Diagnosis
Tumor resection Margin(10)	Free of tumor(08) Involved by tumor(02)	Free of tumor (08) Involved by tumor(02)	Free of tumor(07) Involved by tumor(03)
Breast (20)	Fibroadenoma (03) Phyllodes tumor (01) Inconclusive smear (01)	Fibroadenoma(4) Mastitis (1)	Fibroadenoma (04) Granulomatous Mastitis (01)
	Infiltrating ductal carcinoma (14) Mucinous Carcinoma(01)	Infiltrating ductal Carcinoma (14) Mucinous Carcinoma(01)	Infiltrating ductal carcinoma (13) DCIS (01) Mucinous carcinoma (01)
Skin and soft tissue (18)	Benign spindle cell lesion (10)	Benign spindle cell lesion (10	Dermatofibroma (02) Schwannoma (06) Neurofibroma (02)
	Pleomorphic spindle cell lesion (03)	Pleomorphic spindle cell lesion (03)	Malignant fibrous histiocytoma (01) Fibrosarcoma (01) Leiomyosarcoma (01)
	Squamous cell carcinoma (03)	Squamous cell carcinoma (03)	Squamous cell carcinoma (03)
	Granulomatous lymphadenitis (02)	Granulomatous lymphadenitis (02)	Tuberculous lymphadenitis (02)
Genitourinary system (07)	Renal cell carcinoma (02)	Renal cell carcinoma (02)	Renal cell carcinoma (02)
	Carcinoma bladder (01)	Carcinoma bladder (01)	Carcinoma bladder (01)
	Benign hyperplasia of prostate (03)	Benign hyperplasia of prostate (03)	Benign hyperplasia of prostate (03)
	Malignant ovarian tumor(01)	Malignant ovarian tumor(01)	Dysgerminoma (01)
Gastrointestinal system (137)	Tuberculosis of intestine (03)	Tuberculosis of intestine (03)	Tuberculosis of intestine (03)
	Metastic adenocarcinoma-Liver nodule(01)	Metastic adenocarcinoma-Liver nodule(01)	Metastic adenocarcinoma-Liver nodule(01)
	Adenocarcinoma intestine (03)	Adenocarcinoma intestine (03)	Adenocarcinoma intestine (03)
Endoscopic biopsy(130)	H pylori gastritis- Imprint-108 Non specific gastritis-(22)	H Pylori gastritis-Crush -112 Non specific gastritis (18)	H pylori Gastritis(118) Non specific gastritis(12)
Thyroid (04)	Colloid goitre (02) Papillary lesion (02)	Colloid goitre (03) Papillary Carcinoma of thyroid(01)	Colloid goitre (03) Papillary Carcinoma of thyroid(01)
Bone (01)	Osteogenic sarcoma (01)	Osteogenic sarcoma (01)	Osteogenic sarcoma (01)
Central Nervous Ststem (01)	Anaplastic glial tumor	Anaplastic glial tumor with oligodendroglial Component	Glioblastoma with sarcomatous and oligodendroglial components
Testes (02)	Seminoma (02)	Seminoma (02)	Seminoma (02)

Figures in parenthesis indicate the number of cases.

Discussion

Several studies done in the past have discussed the use of imprint and touch preparation, especially as a tool for intraoperative diagnosis.

The frozen section has always been a favoured tool, over cytological methods in rapid intraoperative diagnosis. This is largely attributed to the surgical

Table 2:

Organ/System	Total no. of cases	Imprint cytology-Correct Diagnosis	Scrape Cytology-Correct Diagnosis	Diagnostic Accurcy-Imprint Cytology	Diagnostic Accuracy-Scrape/Crush Cytology
Tumor Resection Margin	10	9	9	90%	90%
Breast	20	17	19	85%	95%
Skin and Soft Tissue	18	18	18	100%	100%
Genitourinary	07	07	07	100%	100%
Gastrointestinal	137	127	131	92.7%	95.6%
Thyroid	04	03	04	75%	100%
Bone	01	01	01	100 %	100%
CNS	01	01	01	100%	100%
Testis	02	02	02	100%	100%

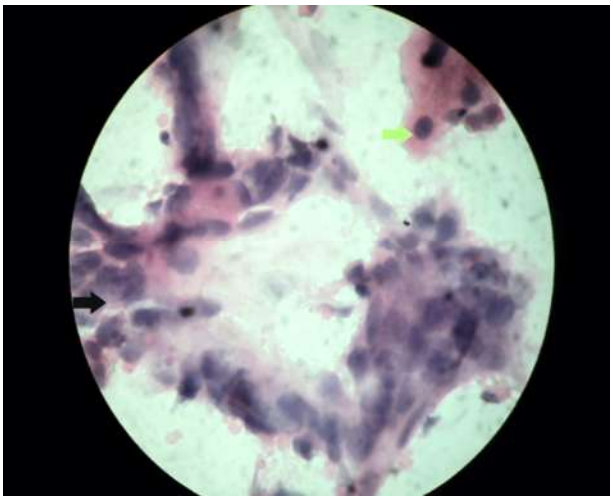

Fig. 1: Image1: Smear showing cluster of atypical columnar cells (Black Arrow) with adjacent benign hepatocytes (Green Arrow)-Haematoxylin and Eosin, X400.

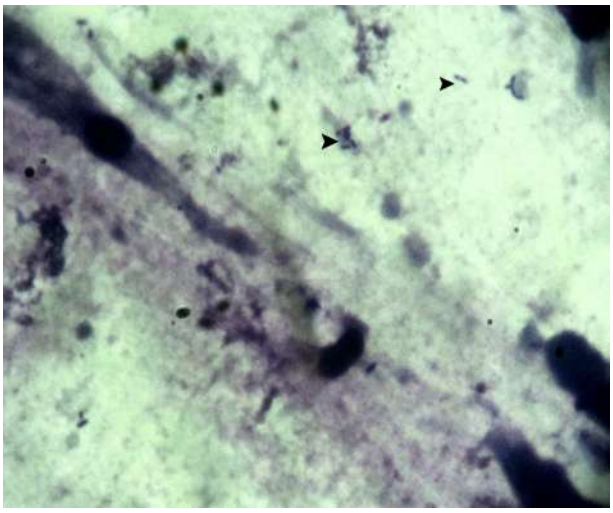

Fig. 3: Imprint Cytological smear showing cluster of tiny curved rod like structures (Arrow head) amidst dirty smear background, Giemsa, 1000x.

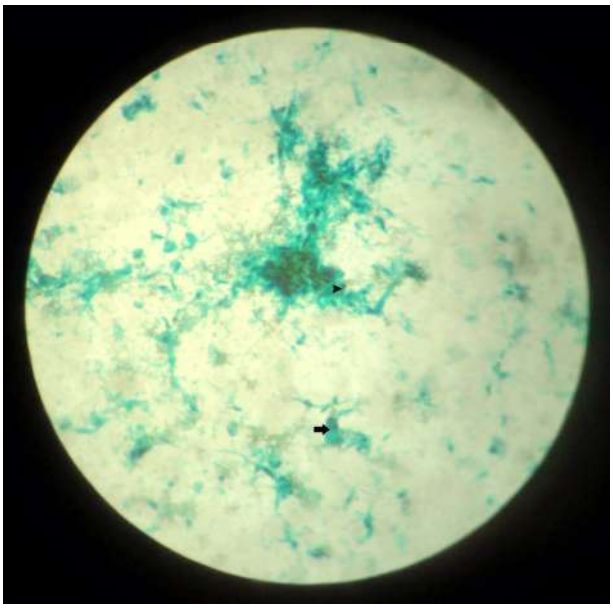

Fig. 2: Imprint PAP stained smear displays a round dark nuclei (Arrow head) and a pleomorphic nuclei (Black Arrow)

pathologist's relatively higher level of confidence in frozen sections, though many studies have demonstrated that the diagnostic efficacy of intraoperative cytology is comparable to that of frozen section. So, this study was undertaken to know the utility of various cytological tools in the intraoperative diagnosis of tumor. We obtained very good results while using scrape and crush cytology.

The preparation of touch or imprint smears entails application of superficial part of the biopsy sample, while crush cytology smears represent the whole biopsy tissue on smear and scrape cytology yields more cellular and uniform smears. Shidham *et al.* [1] and Khunamornpong *et al*. [8] observed that scraping of tumor is the method preferred, because large number of cells can be obtained and cells can be spread well on the slides. We also found that smears prepared after scraping of tumor yielded more cellular and uniform smears.

We also obtained excellent staining results so as to arrive at a diagnosis. This staining completed within 180 seconds and the whole procedure took around 10 minutes, enabling rapid diagnosis with good results as well.

Gross examination is very useful for making a provisional impression, before making any impression by any rapid diagnostic tool. We studied 20 cases of breast lesions, of which 18 could be diagnosed correctly by scrape cytology and 16 by imprint cytology. One case, showed touch smears with clusters of ductal epithelial cells, without any evident lesion, while crush smear showed additional inflammatory component in smear and was diagnosed as mastitis, which was further diagnosed as granulomatous mastitis in histopathology, due to presence of occasional epitheloid cell clusters and area of necrosis. Suen *et al* [9] studied 473 cases of breast lesions with scrape cytology and obtained an accuracy rate of 95.7%. They noted that it was not possible to differentiate between *in situ* and infiltrative carcinoma of breast with scrape cytology, as also noted in our case, with one case of DCIS being overdiagnosed as infiltrating ductal carcinoma.

We found that it is not difficult to diagnose malignancy of breast on cytology, but highly cellular smears of benign lesion should be carefully screened as scrape smears would yield more cellular smears. The most significant factor affecting the diagnostic accuracy of intraoperative cytology may be the number of cases in the low-grade or well-differentiated category in a particular study. We obtained an accuracy rate of 95% with scrape cytology. Our study included 130 cases of endoscopic biopsies of stomach, out of which in 108 cases, *H pylori* (Figure 3) was correctly identified in imprint cytology and 112 cases in crush smears, The section study, showed *H pylori* positivity in 118 cases. The high diagnostic accuracy of crush cytology in identifying *H pylori*, was also evident in study of Ahsan K et al [10] with diagnostic accuracy of 90%. One case of metastatic adenocarcinoma in liver nodule was correctly identified by both imprint (Figure 1) and scrape smears. Two cases of testes, one case of CNS tumor (Figure 2) and one case of bone tumor were diagnosed correctly on cytology. The rates of accuracy achieved were comparable with that observed by many other authors. Intraoperative cytology has high accuracy rates, excellent preservation of cellular details, and the possibility of identifying focal, macroscopically undetectable neoplastic lesion in large tissue fragments. Martinez et al [11] studied 100 CNS lesions by touch preparation and frozen section and compared both these techniques with paraffin sections. They observed 76% accuracy in imprint smear and 88% in frozen section as compared to paraffin section. They also observed that touch preparations were superior to frozen section particularly for evaluating soft or highly cellular tumour and for preliminary diagnosis from a minute surgical specimen (eg. stereotactic biopsy).

The disadvantages of intraoperative cytology are very few and high accuracy rates can be achieved with experience. It is though not possible to accurately distinguish *in situ* from infiltrating carcinoma and to satisfactorily evaluate the depth of invasion and/or margins of resection. In our study two and one cases of tumor involving the resection margin were missed by imprint and scrape cytology respectively. This may be attributed to multifocality of tumor involvement, which is unable to be detected by cytological tools.

Apart from its diagnostic role, intraoperative cytology can become a very useful learning tool. It can promote interpretation of cytology smears and its histological correlation, as the material obtained can be interpreted as FNAC smears.

Conclusion

Apart from fine needle aspiration cytology, scrape smears, crush smears and touch smears are important adjunct and complementary tool in diagnosis of various benign and malignant lesions. Cytological tools are cost effective diagnostic tools and an additional boon to the centres where frozen section is not available.

References

1. Shidham VB, Dravid NV, Grover S, Kher AV. Role of scrape cytology in rapid intraoperative diagnosis: Value and limitations. Acta Cytol. 1984; 28:477–82.
2. Gal R. Scrape cytology assessment of margins of lumpectomy specimens in breast cancer. Acta Cytol. 1988; 32:838–9.
3. Mair S, Lash RH, Suskin D, Mendelsohn G. Intraoperative surgical specimen evaluation: Frozen section analysis, Cytologic Examination, or Both? Am J Clin Pathol. 1991; 96:8–14.
4. Scucchi LF, Di Stefano D, Cosentino L, Vecchione A. Value of cytology as an adjuvant intraoperative diagnostic method. Acta Cytol. 1997; 41:1489–96.
5. Shidham VB, Gupta D, Galindo M, Haber M, Grotkowski C, Edmonds P, et al. Intraoperative scrape cytology: Comparison with frozen sections, using receiver operating characteristic (ROC) curve. Diagn Cytopathol. 2000; 23:134–9.

6. Tribe CR. A comparison of rapid methods including imprint cytodiagnosis for the diagnosis of breast tumors. J Clin Path. 1973; 26:273–7.
7. Kocchar R, Bhasin DK, Gupta SK, Malik AK, Mehta SK. Crush preparations of gastro-oesophageal biopsy specimens in the diagnosis of malignancy. Actacytol 1990; 34:214-6.
8. Khunamornpong S, Siriaunkgul S. Scrape cytology of the ovaries: Potential role in intraoperative consultation of ovarian lesions. Diagn Cytopathol. 2003; 28:250–7.
9. Suen KC, Wood WS, Syed AA, Quenville NF, Clement PB. Role of imprint cytology in intraoperative diagnosis: Value and limitation. J Clin Pathol. 1978; 31:328–37.
10. Ahsan K, Hossain MZ, Uddin MR. Role of Crush cytology for the detection of Helicobacter Pylori in gastroduodenal diseases. J Dhaka Med Coll. 2008; 17:88-93.
11. Martinez AJ, Pollack I, Hall WA, Lunsford LD. Touch preparations in the rapid intraoperative diagnosis of central nervous system lesions. A comparison with frozen sections and paraffin-embedded sections. Mod Pathol. 1988; 1:378-84.