



Contents lists available at BioMedSciDirect Publications

International Journal of Biological & Medical Research

Journal homepage: www.biomedscidirect.com



Original article

A prospective study to compare the aspiration and non-aspiration techniques in fine-needle cytology of Lymphnode and to evaluate the diagnostic accuracy of aspiration cytology in Lymphnode lumps.

K. Bharathi^{*a}, S. Anuradha^b, Abdul Khaliq^c, Sudha Venkatesh^d

^{a,b} Department of Pathology, Sri Satya Sai Medical College & Research Institute, Nellikuppam, Tamilnadu, India.

^d Department of Pathology, Madras Medical College, Chennai.

ARTICLE INFO

Keywords:

Lymphnode lumps

FNAC

FNNAC

Diagnostic adequacy

Diagnostic accuracy

ABSTRACT

A prospective study to compare the aspiration and non-aspiration techniques in fine-needle Cytology of Lymphnode and to evaluate the diagnostic accuracy of aspiration cytology in Lymphnode lumps. Fine needle cytology is an efficient alternative to the surgical excision in case of lymphnode lumps. Objectives: The aim of this study was to compare the cytological features of fine needle non- aspiration cytology (FNNAC) with that of fine needle aspiration cytology (FNAC) in lymphnode lumps and to evaluate the diagnostic accuracy of fine needle cytology. Methods: Samples were obtained by applying both FNAC and FNNAC techniques, from lymphnode lumps of 100 patients who attended the cytology department. The sampling procedures were done by single operator after getting a written consent. The slides were studied and reported by an expert cytologist, thus bias was avoided in all stages of the study from sampling to reporting. Slides obtained from FNAC and FNNAC techniques were compared using a objective scoring system developed by Mair et al based on the background blood or clot, amount of cellular material, degree of cellular degeneration, degree of cellular trauma and retention of appropriate architecture. In 30 cases aspirated lymphnode lumps were excised surgically and the diagnostic accuracy of fine needle cytology in lymphnode lesions was evaluated by doing histopathological correlation with cytological features. Wilcoxon signed rank test was performed using SPSS14 software for statistical analysis. Differences between FNAC and FNNAC based on the above mentioned five parameters were analysed. The diagnostic adequacy, quality of smears obtained from two techniques were compared. Results & Conclusions: The number of unsuitable smears and failure rate was lower for FNAC technique. The diagnostic adequacy of FNAC (98%) is more than FNNAC technique (80%) and the difference is statistically significant ($p < 0.05$). We conclude in our study that FNAC is the best choice as a diagnostic test for all the lymphnode swellings including cystic lesions. The overall diagnostic accuracy of lymphnode cytology was 90%. The discordance rate was 10%. The sensitivity was 96.42%. The specificity was 66.66%.

©Copyright 2010 BioMedSciDirect Publications IJBMR -ISSN: 0976:6685. All rights reserved.

1.Introduction:

Fine needle cytology (FNC) of lymphnodes is a recently developed popular technique used for rapid diagnosis of all the superficial lumps and even deeply seated lesions with the aid of imaging techniques. The technique is simple, relatively painless OP procedure and economical. It is an essential test employed to rule out metastatic malignancy, to know the tumor type preoperatively,

to decide the line of management and for staging the tumor. The accuracy can approach to that of histopathology, when applied by experienced and well trained practitioners. Fine needle cytology includes two techniques fine-needle aspiration cytology (FNAC) and fine-needle nonaspiration cytology (FNNAC). In this prospective study 100 cases of lymphadenopathy were examined and sampled. An attempt was made to compare the techniques FNAC and FNNAC with reference to the diagnostic adequacy and quality of the material obtained. Then the diagnostic accuracy of fine needle cytology was calculated by histopathological correlation in 30 cases where the enlarged lymphnode was excised surgically as indicated.

* Corresponding Author : Dr.Bharathi.K MD(Path),

Assistant professor of pathology,

Sri Satya Sai Medical College & Research Institute, Nellikuppam.

Kancheepuram district, Pin code - 603108

Tamilnadu, India.

Ph no: 9962531082

E- mail: bharathi.pathos@gmail.com

©Copyright 2010 BioMedSciDirect Publications. All rights reserved.

2. Materials and Methods:

This study was conducted during January 2008 - February 2010. Institutional Ethical Committee Clearance was obtained. 100 cytology smears were obtained using the aspiration and nonaspiration techniques. Samples were taken from the patients who attended the cytology department for fine needle cytology with lymphnode lumps as an early presentation. Informed written consent was obtained.

FNAC is the conventional method in which suction pressure was applied to take the cells and tissue sample. Syringe must be attached to the needle (22 gauge or 25 gauge). The basic principle underlying fine needle aspiration cytology is the aspiration of cellular material from the target masses often utilizing fairly high suction pressure. The function of the negative pressure in FNAC is not to tear cells but to hold the tissue against the sharp cutting edge of the needle which scrapes or cuts the tissue as it advances through the tissue [1].

In more recent times a modified technique called fine needle non-aspiration cytology(FNNAC), pioneered in France came into vogue in 1981 [2]. It eliminates active aspiration by syringe, replacing it by the principle of capillary suction of fluid or semi fluid material into a thin channel (fine needle). It is less painful, less traumatic, and smears obtained by FNNAC are of "text book" quality. However no syringe or suction is necessary for this technique. After identifying the swelling with all sterile precautions, the hub of the needle is held in the pencil grip fashion in the right hand and the needle was gently inserted into the swelling and then moved in and out over 5-10 seconds rapidly. Aspirate flows into the needle through capillary action and as soon as the aspirate appears in the hub, the needle is withdrawn and attached to the syringe with air inside. Next the plunger is used to expel the material from the needle onto the glass slides. The slides were fixed in 85% of the Isopropyl alcohol for 20 -30 minutes in Coplin jars. The slides were stained with Harris Hematoxylin & Eosin stain and mounted in D.P.X.

All the needle sampling procedures were made by a single operator, bias was thus avoided in all stages of sampling from patient examination to slide fixation. The slides were studied and a cytological diagnosis was made. In this study we compared the two techniques of fine needle cytology (FNAC and FNNAC) with reference to diagnostic adequacy and diagnostic accuracy. All the slides were objectively analysed using Mair's point scoring system to enable comparison between FNAC and FNNAC techniques as shown in table 1. [Table 1]. On the basis of five criteria tabulated, a cumulative score was obtained for each case which was then categorized accordingly to one of the 3 categories category 1: Unsuitable for cytological diagnosis- (0-2), category 2: Diagnostically Adequate- (3-6), category 3: Diagnostically superior - (7-10). Wilcoxon signed rank test was performed using SPSS14 software for statistical analysis. In 30 cases patients underwent lymphnode excision biopsy. In those cases the cytological features were correlated with the histopathological features and the diagnostic accuracy of FNC in lymphnode lumps was calculated.

3. Results:

In our prospective study 100 cytology smears aspirated by two techniques FNAC and FNNAC were compared. They were categorized according to their age, sex and site. Mean age of the patients with lymphadenopathy in our study was 45 yrs. Age group range involved was 15 – 75 yrs. Cervical group of lymphnodes were commonly involved. Male to female ratio was 1: 1.08. Majority of the cases (58%) belong to reactive lymphadenopathy due to infections. 42% of cases were due to metastatic malignant deposits. 22 cases were reported as Squamous cell carcinomatous deposits and 19 cases as adenocarcinomatous deposits. [Table 2]

We compared the smears obtained through two techniques using Mair's scoring system. The scores obtained by each technique on the basis of five parameters were tabulated. [Table 3] we found that the average score obtained by FNAC(8.14) was more than FNNAC(6.7) [table 4]. We calculated the diagnostic adequacy of each technique by adding the diagnostically adequate as well as diagnostically superior smears obtained. The diagnostic adequacy of FNAC was 98% whereas for FNNAC it was 80%. The difference was statistically significant with p value < 0.05 [Table 5]. Thus we conclude in our study that FNAC is the best choice for aspirating lymphnode lumps.

In 30 cases lymphnode lumps were excised surgically and histopathological diagnosis was made. Most of the excised lymphnodes were due to metastatic deposits since only a small percentage of patients with benign cytological features undergo surgery. Then the cytological features were correlated with the histopathology [table 6]. The diagnostic accuracy of fine needle cytology was thus calculated and found to be 90%. The sensitivity and specificity of fine needle cytology was calculated. The sensitivity was 96.42% and the specificity was 66.66%. We observed in our study that 100% diagnostic accuracy can be achieved through FNC with routine radiological and clinical correlation.

Table 1: Scoring system According to Mair to classify the quality of cytological aspirate.

Criterion	Quantitative description	points
Amount of cellular material	Minimal to absent; diagnosis not possible	0
	Sufficient for cyto diagnosis	1
	Abundant	2
Retention of appropriate architecture	Minimal to absent ; diagnosis not possible	0
	Moderate ; some preservation of architecture	1
	Excellent ; architecture display closely resembling histology , diagnosis obvious	2
Degree of cellular degeneration	Marked; diagnosis impossible	0
	Moderate; diagnosis possible	1
	Minimal diagnosis obvious	2
Degree of cellular trauma	Marked; diagnosis impossible	0
	Moderate; diagnosis possible	1
	Minimal diagnosis obvious	2
Background blood or clot	Large amount ; great compromise to diagnosis	0
	Moderate amount ; diagnosis possible	1
	Minimal; diagnosis easy; specimen of textbook quality	2

Table 2: Frequency of various lesions of lymphnode:

Cytological diagnosis	Male	Female	Total(%)
Reactive lymphnodes	12	16	28
Tuberculous lymphadenitis	10	20	30
Squamous cell carcinomatous deposits	16	6	22
Adeno carcinoma deposits	9	10	19
Lymphoma	1	0	1
	48	52	100

Table 3: Score obtained by FNAC & FNNAC in individual parameters in our study

Procedure	Adequacy	Archi- tecture	Degene- ration	Trauma	Background blood	Average Score
FNNAC	1.2	1.3	1.4	1.2	1.6	6.7
FNAC	1.87	1.67	1.6	1.4	1.6	8.14

Table 2: Frequency of various lesions of lymphnode:

No of cases	Clinical diagnosis	Cytology Diagnosis	Confirmation by Histopathology diagnosis
6 cases	Carcinoma stomach with secondaries in lymphnode	Adeno carcinoma (ACA) deposits	Adeno carcinoma (ACA) deposits
2 cases	Carcinoma of colon with secondaries in lymphnode	ACA deposits	ACA deposits
8 cases	Carcinoma Breast with axillary lymphadenopathy	ACA deposits	ACA deposits
4 cases	Oral cavity carcinoma with cervical lymphadenopathy	Squamous cell carcinomatous (SCC)deposits	Squamous cell carcinomatous (SCC) deposits
3 cases	Nasopharyngeal carcinoma with cervical lymphadenopathy	SCC deposits	SCC deposits
2 cases	Penile carcinoma with inguinal lymphadenopathy	SCC deposits	SCC deposits
1 case	Tuberculosis	Granulomatous lymphadenitis	Tuberculous lymphadenitis
2 cases	Infective lymphadenopathy	Reactive lymphadenopathy	Reactive lymphoid hyperplasia
1 case	Lymphoma	Undifferentiated carcinoma	Non hodgkins lymphoma
1 case	Lymphoma	Lympho proliferative disease	Non hodgkins lymphoma
30 cases in total	27 out of 30 cases correlated well with histology diagnosis.		

Table 7: Comparison of diagnostic adequacy(%) in lymph node lesions with other studies

Comparison of diagnostic adequacy(%) in lymph node lesions with other studies			
Diagnostic adequacy	FNNAC	FNAC	P value
Dey P and Ray R et al[18] in 1993	95.34%	81.4%	P < 0.05
Braun H et al[19] in 1997	51.2%	51.8%	P > 0.05
CV Raghuvver et al[15] in 2002	85%	87.59%	P > 0.05
Present study	80%	98%	P < 0.05

Table 4: Comparison of the average score and the diagnostic adequacy of FNNAC and FNAC.

Technique	Average score	Diagnostic adequacy
FNNAC	6.7	80%
FNACP	8.14	98%
value	P <0.05	P <0.05

Table 5: Grading of smears based on total score in FNAC & FNNAC

Grade	FNAC	FNNAC
Diagnostically unsuitable	2	20
Diagnostically adequate (including the superior quality smears)	98	80
Total	100	100
P value	P<0.05	P<0.05

Table 8: Comparison of diagnostic accuracy(%) of Fine needle cytology in lymph node lesions with other studies:

Comparison of diagnostic accuracy(%) in lymph node lesions with other studies			
Studies	Diagnostic accuracy %	Sensitivity %	Specificity %
Hafez NH, Tahoun NS et al[21] (2011)	82.2	90.9	67.2
Alwan NA, AlHashimi AS et al[22] (1996)	89.6	90.5	98.8
resent study	90	96.42	66.66

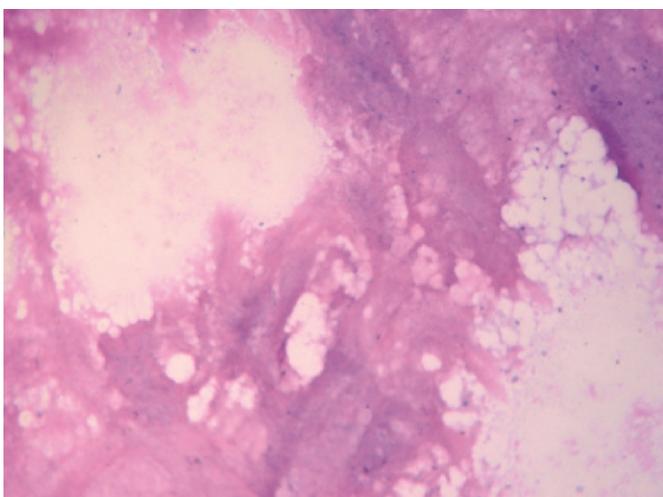
Figure 1: FNNAC smear from Tuberculous lymphadenitis.

Figure 1: Photomicrograph from a FNNAC smear shows caseating necrotic material from Tuberculous lymphadenitis [H and E stain, 100x].

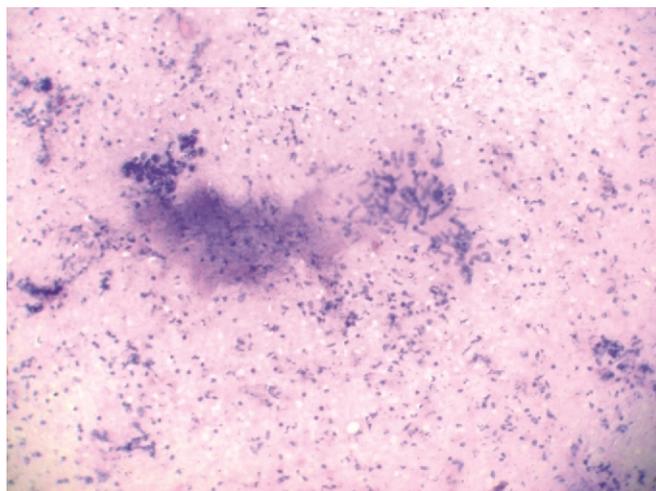
Figure 2: FNAC smear of Tuberculous lymphadenitis

Figure 2: Photomicrograph in FNAC smear shows clusters of epithelioid macrophages forming a granuloma and caseating necrosis [H and E stain, 100x].

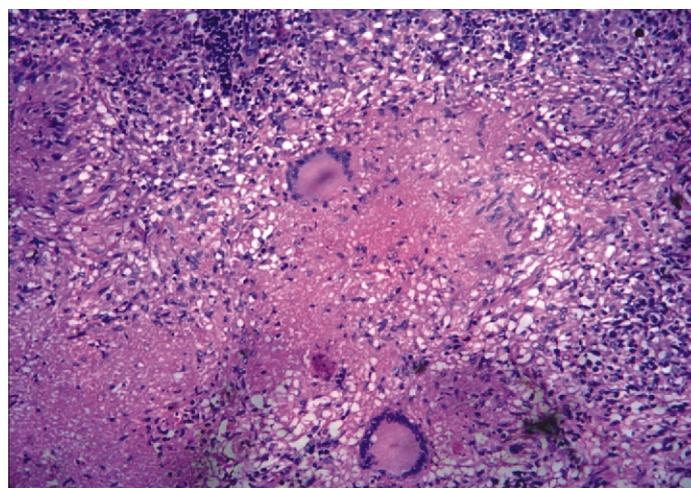
Figure 3: Histopathology picture of tuberculosis.

Figure 3: Histopathology photomicrograph from the excised lymphnode shows caseating granuloma with langhans giant cells [H&E stain, 100x].

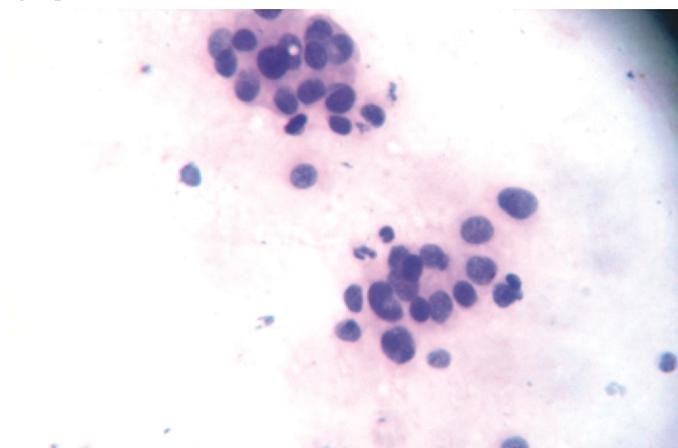
Figure 4: FNC smear of adenocarcinomatous deposits in a lymphnode.

Figure 4: Photo micrograph from the FNC smear shows the glandular adeno carcinomatous deposits in a lymphnode [H&E stain, 100x].

Figure 5: FNC smear of squamous cell carcinoma deposits in lymphnode

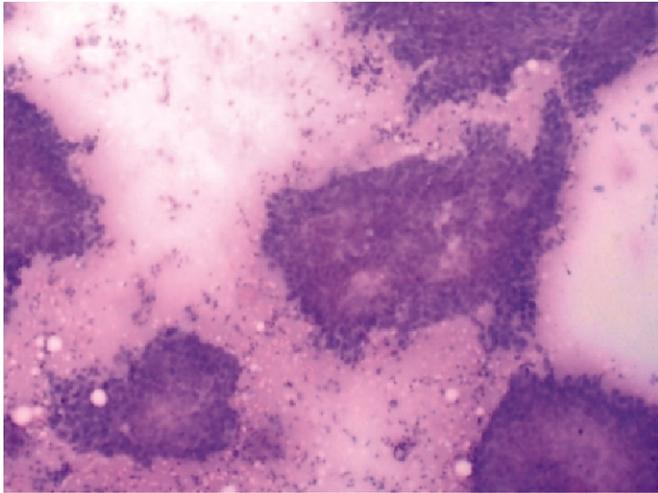


Figure 5: Photomicrograph from FNC smear shows sheets of malignant squamous epithelial cells with atypia and necrotic debris in the background [H and E stain, 100x].

4. Discussion:

Fine needle cytology can be employed on all the superficial palpable lumps that are palpable in skin, subcutaneous tissue, lymphnode, thyroid and breast. The expansion of fine needle cytology in primary diagnosis of lymphoma has been enormous and successful with aids of recent advances like immunohistochemistry and flow cytometry [3,4,5]. The low risk of complications is an additional advantage which allows FNC to be performed in outpatient departments and in radiology rooms. It is suitable for even debilitated patients.

Lymphnode cytology is useful for the clinicians to know whether the lymphadenopathy is due to infections, metastatic malignancy or lymphomas [6,7]. The primary purpose of fine needle cytology is to decide whether surgical excision of lymphnode is indicated or not. In olden days fine needle cytology of lymphnodes has been practiced in central Europe and in Scandinavians for many years particularly by hematologists to diagnose lymphomas in conjunction with bone marrow biopsy.

In 1884, Kronig was the first to diagnose lung cancer by aspirating tissue through a transthoracically inserted cannula. In 1904 Greig and Gray performed aspiration of lymph nodes to isolate causative agents of trypanosomiasis. After that the growth of FNC remained dormant.

Guthrie in 1921 successfully diagnosed cases of syphilis, tuberculosis, lymphomas and metastatic carcinoma by needle aspirations. In 1930, Martin, Stewart and Ellis from United States used a thicker needle (18 gauge) for aspiration [8]. The FNAC technique which was described by Lowhagen et al from Karolinska Institute is generally employed now [9]. They performed the aspiration with a finer needle ie, 22 – 25 gauge [10].

In 1955, Franzen et al introduced a special aspiration syringe holder [11]. However FNAC came into wide acceptance in North America and India in 1980s. A new modified technique of FNC termed as FNNAC pioneered in France by Zajdela et al came into vogue in 1981 [2]. The same procedure was called as "cyto puncture" by Brifford et al in 1982 [12]. FNNAC technique has gained popularity nowadays for its various advantages and has been used widely. FNNAC gives the operator a more direct and better feeling of consistency of the tissue through the tip of the needle and very little admixture of blood in smears.

Santos and Leiman et al in 1988 were the first to compare the FNAC and FNNAC smear in thyroid nodules. They observed that the number of unsuitable specimens in their study was not different with both the techniques [13].

Sharon Mair and Fiona Dunbar et al in 1989 did their comparative study of FNAC and FNNAC and the smears were scored on the basis of following five objective parameters diagnostic adequacy, retention of architecture, degree of trauma, degree of cellular degeneration and background blood or clot obscuring the background as shown in table 1 [14].

Mair et al concluded that there was no statistical difference between FNAC and FNNAC, but they observed that FNNAC smears were diagnostically superior and of text book quality and it allows for greater ease of sampling and a more sensitive probing of the mass to be sampled [14]. FNAC was diagnostic for fibrous lesions and cystic lesions and suggested that the technique of fine needle sampling employed for cytodagnosis could be left to the personal preference of the operator.

In our prospective study, we observed that infections and reactive lymphadenopathy was common in younger patients. Majority of the cases (58/100) belong to reactive lymphadenopathy due to infections. 30% of cases belong to tuberculous lymphadenitis [Figure 1, 2 and 3]. Lymphadenopathy in older patients was mainly because of metastatic malignancy. 42 out of 100 cases were due to metastatic malignant deposits [Figure 4, 5]. Incidence of adenocarcinoma deposits and squamous cell carcinoma deposits were almost equal.

On analyzing the smears obtained by FNAC & FNNAC, we observed that the average score obtained by FNAC was more than FNNAC. The diagnostic adequacy of FNAC was 98% whereas for FNNAC it was 80%. The difference was statistically significant with p value < 0.05. Thus we conclude in our study that FNAC is the best choice for aspirating lymphnode lumps. FNAC technique yielded more cellular smears with more diagnostic adequacy.

Similar studies in literature report as follows: CV Raghuvver I Leeka et al in 2002 reported in their comparative study that FNNAC was superior in quality and diagnostic accuracy than FNAC in thyroid and lymph node lesions. FNNAC seemed to be better for diagnosing malignant lesions while FNAC appeared better for diagnosing benign ones. Both the techniques therefore would be

complementary to each other [15]. Kumarasingh M et al in 1995 observed in their comparative study lesions both the techniques was comparable in all lymphnode [16]. Amrita Ghosh et al in 2000 did a comparative study and found that contamination with blood was more in lymphnode lesions in FNAC smears than FNNAC smears and values were statistically significant [17]. Dey P et al and Braun H Walch et al also did a similar study and obtained contradicting results [18,19] [Table 7]

In our study we have also calculated the diagnostic accuracy of fine needle cytology in lymphnode lumps by doing histopathological correlation. Cytological features were correlated with the histopathology. The overall diagnostic accuracy of fine needle cytology was thus calculated and found to be 90%. The sensitivity and specificity of fine needle cytology was found to be 96.42% and 66.66% respectively.

On comparing our results with other studies we observed that it was similar. Nasuti JF et al demonstrated that lymphnode cytology can be an accurate, economical and rapid diagnostic procedure [20] Hafez NH et al reported a diagnostic accuracy of 82.2%, sensitivity of 90.9% and specificity of 67.2% [21]. AlAlwan et al reported a diagnostic accuracy of 89.6% [22].

5. Conclusion:

Fine needle cytology though not an substitute for histopathological examination but an extremely valued complement to it. It allows the distinction between non neoplastic and neoplastic conditions and helps the clinician to decide the line of management. The clinical value is not only limited to neoplastic conditions but is also valuable in infections where the sample collected by FNC can be used for microbiological and biochemical analysis. Fine needle cytology is readily repeatable and as a preliminary investigation can reduce the hospitalization period and avoid overcrowding in hospitals.

Acknowledgements: Our grateful thanks to the department of Surgery, Madras Medical College, Chennai.

Source of Support: Nil

Conflict of Interest: None Declared

6. References

- [1] THOMSON. Thin needle aspiration Biopsy. Acta CyTOL.1982;26:262-263.
- [2] Zajdela A, Zillhardt P, Voillemot N. Cytological diagnosis by fine needle sampling without aspiration. Cancer 1987;59:1201-1205.
- [3] Lopes cardazo P: The cytologic diagnosis of lymphnode punctures. Acta cytol.1964;8: 194-198.
- [4] Keith VE, Harsharan SK, Jerald GZ. Fine needle aspiration biopsy of lymph nodes in the modern era: reactive lymphadenopathies. Pathol Case Rev. 2007; 12 (1) :27-35.
- [5] Howlett DC, Harper B, Quante M, Berresford A, Morley M, Grant J. Diagnostic adequacy and accuracy of fine needle aspiration cytology in neck lump assessment: results from a regional cancer network over a ONE YEAR PERIOD. J LARYNGOL OTOL. 2007;121(6):571-579.
- [6] Chhotray GP, Acharya GS. Fine needle aspiration cytology in diagnosis of metastatic lymphadenopathies. Indian journal of medical research. 1987;85: 685-8.
- [7] Hirachand M. Lakhey J. Akhter B. Thapa. Evaluation of fine needle aspiration cytology of lymph nodes in Kathmandu Medical College, Teaching hospital. Kathmandu Univ Med J. 2009;26:139-142.
- [8] Martin H E and Ellis E. Biopsy Needle puncture and aspiration . Ann Surg 1930 ; 92 :169-181.
- [9] Lowhagen T, Willems JS, Lundell G, Sundblad R, Granberg P. Aspiration Biopsy Cytology in Diagnosis of Thyroid Cancer. World J Surg. 1981;5: 61-73.
- [10] Frable WJ : Thin Needle Aspiration Biopsy. American journal of clinical pathology. 1976; 65:168-80.
- [11] Franzen S, Giertz G, Zajicek J. Cytological diagnosis of prostatic tumours by Trans rectal aspiration biopsy, a preliminary report. Br. J. Urol 1960; 32:193 – 196.
- [12] Briffod M, Gentile A, Hebert H. Cytopuncture in the follow-up of breast carcinoma. Acta Cytol. 1982; 26:195-200.
- [13] Santos JEC, Leiman G. Non aspiration Fine Needle Cytology, Application of a new technique to nodular thyroid disease. Acta Cytol. 1988; 32:353-356.
- [14] Mair S, Dumbar F, Becker P, Plessis WD. Fine Needle Cytology - Is Aspiration Suction Necessary? A study of 100 masses in various sites. Acta Cytol. 1989;33 :809-813.
- [15] Raghuvver CV, Leekha IL, Pai MR, Adhikari P. Fine needle aspiration cytology versus fine needle sampling without aspiration. A prospective study of 200 cases. Indian J Med Sci. 2002;56: 431-9.
- [16] Kumarasinghe MP, Sheriffedeen AH. Fine Needle Sampling without aspiration. Pathology 1995;27 :330-332.
- [17] Ghosh A, Misra RK, Sharma SP, Singh HN, Chaturvedi AK. Aspiration Vs Nonaspiration Technique of Cytodiagnosis - a critical evaluation in 160 cases. Indian J, Pathol Microbiol. 2000; 43:107-112.
- [18] Dey P, Ray R. comparison of fine needle sampling by capillary action and fine needle aspiration. Cytopathology. 1993; 4:299-303.
- [19] Braun H, Walch C, Beham A. FNAC in head and neck region swellings. Laryngo otologie, 1997; 76(6):358-63.
- [20] Nasuti JF, Yu G, Boudousquie A, Gupta P. Diagnostic value of lymphnode fine needle aspiration cytology : an institutional experience of 387 cases observed over a 5 year period. Cytopathology. 2000; 11(1):18-31.
- [21] Hafez NH, Tahoun NS. Reliability of fine needle aspiration cytology (FNAC) as a diagnostic tool in cases of cervical lymphadenopathy. Journal of the Egyptian national cancer institute. 2011; 23(3):105-114.
- [22] AlAlwan NA, AlHashimi AS, Salman MM, AlAttar EA. Fine needle aspiration cytology versus histopathology in diagnosing lymphnode lesions. Eastern Mediterranean Health Journal. 1996; 2(2):320-25.