INTRODUCTION

Advent of modern imaging techniques has given more impetus to the art of fine-needle aspiration (FNAC) in the field of diagnosis.\(^1\) Popularised by Bottle and Cohen since their published work in 1991, FNAC of the liver has been increasingly utilised as a minimally invasive procedure for diagnosis of focal liver lesions.\(^2\) Only a few contra-indications have been reported in South Asian patients which include bleeding diathesis, suspected haemangioma and hydatid disease of the liver.\(^3\)

The technique of fine-needle aspiration cytology is effectively being practiced as a diagnostic modality in the entire world. Since Hassan and Ahmad's review of liver FNA more than a decade ago, the technique is being put to greater diagnostic use in Pakistan.\(^4\) Yousuf and co-workers reported use of the procedure as a routine pre-operative investigation for superficially accessible lesions in many institutions.\(^5,6\) A few centres are carrying out ultrasound-directed FNAC from deep-seated organs as well.\(^4,7\)

Liver remains one of the most common organs for the lodgment of metastasis.\(^8\) Differentiation between benign and malignant primary or secondary tumours is extremely important from management point of view. Presence of metastases usually rules out surgery whereas; if HCC is diagnosed at an early stage, surgical resection is possible and may assure cure.\(^9\) Primary cancer of the liver is also not a rare entity in our society as concluded by Parvez and Anwar.\(^10\) The clinical and radiological presentations of both primary and metastatic tumours can be similar – as a space-occupying focal mass. Here, FNAC can play a major decisive diagnostic role.

The present study was conducted to evaluate the diagnostic efficacy of FNAC in making a morphological diagnosis of these lesions by differentiating between benign and malignant lesions and from primary and metastatic neoplasms of the liver verified subsequently.

ABSTRACT

Objective: To determine the diagnostic accuracy of fine-needle aspiration cytology (FNAC) in liver masses to isolate malignant from benign tumours and hepatocellular carcinoma (HCC) from metastatic tumours.

Study Design: Cross-sectional, observational.

Place and Duration of Study: Department of Histopathology, Combined Military Hospital, Peshawar, from June 2004 to June 2005.

Methodology: All the patients with liver masses confirmed by ultrasonography, irrespective of age and gender, were included. Patients with inflammatory lesions were excluded from the study. Selected patients underwent fine-needle aspiration under ultrasound guidance followed by needle biopsy. The cytological slides were stained by haematoxylin and eosin (H&E) stain, while Papanicolaou's stain was employed in selective cases. Needle biopsy fragments were fixed in formalin followed by paraffin embedding and staining with H&E stain. Sensitivity and specificity of FNAC in the diagnosis of liver masses was determined using histological diagnosis on liver biopsy as gold standard.

Results: There were one hundred subjects. The mean age at presentation was 55±12 years with male to female ratio of 1.7:1. Cytological diagnosis in 19 cases was benign/non-neoplastic and 81 was malignant. Out of the latter, 49 (60.49%) were HCC and 32 (39.51%) were metastatic tumours on cytology. The overall sensitivity, specificity and accuracy of FNAC in the diagnosis of malignant lesions was 95.2%, 100% and 96% respectively using histological diagnosis on liver biopsy as gold standard. Sensitivity of FNAC to differentiate HCC from metastatic tumours in liver was 96% while specificity was 100% having a diagnostic accuracy of 97.5%. The discrepancy in cyto-histological comparison was mainly seen in well-differentiated and poorly-differentiated HCCs.

Conclusion: FNAC of the liver masses is a simple, safe, accurate, economical screening test without significant morbidity that can be used to identify the vast majority of hepatic neoplasms of primary or metastatic nature with high sensitivity, specificity and diagnostic accuracy.

Key words: Fine-needle aspiration. Metastatic tumour. Hepatocellular carcinoma.
through histological evaluation of the biopsy material obtained simultaneously.

METHODOLOGY

Patients of any age and gender presenting to Combined Military Hospital, Peshawar, from June 2004 to June 2005 with liver masses and confirmed by ultrasonography were included in the study after getting informed consent. Patients with inflammatory conditions, abscess of liver, bleeding diathesis, suspected hydatid cyst, haemangioma or already diagnosed cases were excluded from the study.

FNAC was performed under ultrasound guidance with the help of a radiologist. A disposable spinal needle (22/23 gauge), connected to 10 ml plastic syringe was used. In case of multifocal lesions, the largest or the most easily accessible lesion was selected. The slides were prepared on bedside and immediately fixed in absolute alcohol, for cytological evaluation. In the second step, biopsy was attempted from the same track using a 16 to 18 gauge spinal needle or a core needle biopsy using disposable Menghini’s needle in a few cases. The larger fragments were separated and immediately fixed in 10% formal saline. The fine fragments and haemorrhagic material were treated as “cell-block preparation” for histological studies. The slides were stained by haematoxylin and eosin. Papanicolaou’s method was employed in some of the cases.

Data was entered in SPSS version 11.0 and statistical analysis was done to determine frequency of descriptive variables. Predictive value model of Galen and Gambino was utilized to calculate sensitivity, specificity, positive and negative predictive values and efficacy of FNAC in the diagnosis of liver masses taking false positive, false negative, true positive and true negative cases while using histological diagnosis on the tissue as gold standard.

RESULTS

During the study period, 109 patients reported for aspiration of liver masses, out of whom 9 cases did not fulfil the inclusion criteria and therefore, 100 patients were studied. The mean age at presentation was 55±12 years ranging from 29-80 years. The maximum number of cases was seen between 55-65 years of age showing a male predominance with a male to female ratio of 1.7:1.

Histopathology revealed 85 malignant cases with 55 hepatocellular carcinomas and 30 metastatic tumours, while the remaining 15 were benign lesions. However, FNAC evaluation of the liver masses showed 81 malignant and 19 benign/non-neoplastic lesions confirmed by histopathological diagnosis on the biopsy of the same patient. There were 4 false negative results for malignancy while sensitivity for the diagnosis of malignancy was 95.3% with a negative predictive value of 78.9% and diagnostic accuracy of 96% respectively (Table I).

Out of the 81 malignant lesions on cytology, 49 (60.5%) were cases of HCC and 32 (39.5%) were considered to be metastatic deposits. Analysing the cyto-histopathological comparison of 81 neoplastic cases, biopsy examination confirmed 51 cases to be of primary hepatocellular carcinoma whereas 49 cases were labelled as HCC on cytological smears. Thus sensitivity of FNAC for HCC and to differentiate from metastatic tumours was 96% with a specificity of 100% and having an overall diagnostic accuracy of 97.5% (Table I).

On the other hand, 32 cases were diagnosed as metastatic tumours on FNAC whereas only 30 cases could be confirmed on histopathological examination, giving a false positive rate of 6.67% for metastatic lesions on FNAC (Table I).

Out of the 55 cases of hepatocellular carcinoma, 49 cases were correctly labelled, whereas 2 were diagnosed metastases and 4 as benign/non-neoplastic regenerative nodule at FNAC. Grading of hepatocellular carcinoma into well, moderate and poorly differentiated categories was attempted on aspiration smears and its cyto-histopathological correlation is depicted in Table II. An attempt to adjudge the primary type of tumour in cases of metastasis was also made. Neoplastic cells clumped as clusters with no specific architectural arrangements were present in 10 cases, while acinar

### Table I: Cyto-histological correlation of focal liver masses (n=100).

<table>
<thead>
<tr>
<th>FNAC diagnosis</th>
<th>Histopathology assessment</th>
<th>Metastatic tumour</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>PPV</th>
<th>NPV</th>
<th>Diagnostic accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Malignant</td>
<td>81</td>
<td>10</td>
<td>49</td>
<td>0</td>
<td>30</td>
<td>100%</td>
<td>96%</td>
</tr>
<tr>
<td>Benign/non-neoplastic</td>
<td>4</td>
<td>15</td>
<td>0</td>
<td>96%</td>
<td>100%</td>
<td>96%</td>
<td>93.7%</td>
</tr>
<tr>
<td>Hepatocellular carcinoma</td>
<td>-</td>
<td>-</td>
<td>0</td>
<td>96%</td>
<td>100%</td>
<td>93.7%</td>
<td>97.5%</td>
</tr>
<tr>
<td>Metastatic tumour</td>
<td>-</td>
<td>2</td>
<td>30</td>
<td>100%</td>
<td>96%</td>
<td>93.7%</td>
<td>100%</td>
</tr>
</tbody>
</table>

NPV: Negative Predictive Value; PPV: Positive Predictive Value.

### Table II: Cyto-histological correlation of primary HCC (n=55).

<table>
<thead>
<tr>
<th>Histological diagnosis</th>
<th>No. of cases</th>
<th>Cytological diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Well differentiated HCC</td>
<td>26</td>
<td>Regenerative nodule</td>
</tr>
<tr>
<td></td>
<td></td>
<td>with atypia</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Well differentiated HCC</td>
</tr>
<tr>
<td>Moderately differentiated HCC</td>
<td>13</td>
<td>Moderately differentiated HCC</td>
</tr>
<tr>
<td>Poorly differentiated HCC</td>
<td>16</td>
<td>Poorly differentiated HCC</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Metastases</td>
</tr>
</tbody>
</table>

HCC= Hepatocellular Carcinoma
formation was seen in 14 and papillary formation in one case. Collectively those 25 cases were labelled as metastases of adenocarcinomas (Figure 1). Two cases were labelled malignant melanoma on the basis of melanin pigmentation (Figure 2) and 3 cases as carcinoid tumours.

DISCUSSION

The FNAC technique has recently been introduced in Pakistan and since then it has been used very effectively for pre-operative diagnosis in liver tumours.6 In our series the break-up of FNAC results revealed predominantly malignant lesions (81%). This is in accordance with the previous reports of similar ratio in neoplastic/non-neoplastic pathologies presenting as focal mass lesions in the liver. Hassan et al. reported a figure of nearly 50%, in their series of 236 FNA cases relating to focal liver masses in 1991.4 Considering the high prevalence of Hepatitis B and C in Pakistan, buttressed by Rehman, a higher figure for malignant lesions is understandable.11 Lower rate of benign lesions in this series could be due to specific inclusion criteria.

A comparative evaluation of cytology results with the biopsy diagnosis revealed 4 false negative results (4.7%) for the malignant lesions. The present results tally with the previously reported figures of false negative value of 6.7% in some studies.12,13 There were no false positive results for malignancy. The sensitivity (95.3%) and specificity figures (78.95%) for the diagnosis of malignant masses in this series, are very close to the results of local studies. Considering the possibility of false negative results for malignant masses, it is emphasised that a negative cytology report must be evaluated in the light of clinical, radiological and serological findings.

Regarding the cyto-differentiation between HCC and the metastatic deposits, our results indicate an over-diagnosis of metastasis with 6.67% false positive and an under-estimation of HCC with a 10.91% false negative rate. All the over-diagnosed cases of metastasis proved to be cases of HCC histologically. Similarly the greater majority of misdiagnosed cases of HCC were cytologically inferred as metastatic deposits indicating a considerable morphological overlap between the two conditions. Das, in a comprehensive monograph, has stated that the gold standard for the cytological diagnosis of metastatic deposits remains the identification of malignant cells of non-hepatocytic origin.14 However, a strong morphological similarity occurs between the cell morphology of poorly differentiated HCC and poorly differentiated adenocarcinomas. Difficulty of recognising the hepatocytic morphology of neoplastic cells in this grade has been highlighted in several previous studies. Former workers report a figure of 69% in their series for resemblance of cells to hepatocytes, which is in close accordance with our findings. At the other end of the spectrum, WDHCC also remains a diagnostic pitfall cytologically. Granados et al. have also reported unsatisfactory results with FNA for WDHCC.8 Five cases of WDHCC revealed a cytological overlap with the non-neoplastic condition, regenerative nodule. Inherent atypia of cells associated with cirrhosis has been acknowledged as the biggest diagnostic pitfall on FNA. On the contrary, the possibility of false negative results for HCC has also been highlighted, where the cells of WDHCC can be interpreted mistakenly as reactive hepatocytes. Concurrent with the observation of later workers, 5 cases of WDHCC were misdiagnosed on FNA as being re-generative nodule with atypia.

By using the standard criteria, metastatic deposits can usually be diagnosed correctly on the basis of FNAC.
Localising the primary site is one of the most challenging problems in the field of aspiration cytology. Judicious use of supporting techniques such as immunocytochemistry and electron microscopic examination has been advocated strongly to help in ascertaining the primary site or origin of tumour in FNAC. In this series, an attempt was made in this direction solely on the basis of cell morphology and architectural pattern of aspirate. However, considering a wide ranging differential diagnosis amongst conditions like; metastatic carcinoid, oat cell carcinoma and Islet cell tumours with overlapping cytological features, the diagnoses remain debatable without supportive techniques. Here, a battery of immunocytochemical markers is required for confirmation. Precision of final diagnosis for neuroendocrine tumours can be enhanced by ultrastructural demonstration of neurosecretory granules or biochemical demonstration of elevated urinary 5-HIAA.

Most of the metastases in this series were from adenocarcinoma, presumably abdominal in origin. This diagnosis was solely based on cytology as metastasis with 6.67% false positive rate and an under estimation of HCC with a 10.91% false negative rate. All the over-diagnosed cases of metastasis proved to be cases of HCC histologically. Similarly, the majority of mis-diagnosed cases of HCC were cytologically inferred as reactive nodules indicating a considerable morphological overlap between the two conditions. The standard criterion for the cytological diagnosis of metastatic deposits remains the identification of malignant cells of non-hepatic origin on aspiration cytology that can prove to be an effective tool for primary diagnostic work-up of various focal hepatic masses. Metastatic deposits being quite common, 100% sensitivity for its cytological diagnosis supports the effectiveness of this procedure. Since hepatocellular carcinoma is commoner in this study, better expertise is needed in its cytological diagnosis. For this purpose cytopathologist must be properly trained and experienced in evaluating aspirates from hepatic lesions.

CONCLUSION

FNAC of the liver is a simple, safe, accurate, economical screening test that can be used to identify the vast majority of neoplasms of a primary or metastatic nature. The diagnostic accuracy of FNAC is high enough that treatment and prognosis can be predicted with certainty. Accurately sampled and well prepared FNA smears, in conjunction with cell-block preparation, close collaboration of the pathologist/radiologist and correlation with the clinical history, yield the best results. Team-work and expertise in cytopathology remain the key to accuracy.

REFERENCES