Hepatocellular Carcinoma versus Other Carcinomas of Liver: Distinguishing Features

Anwar Ul Haque, Aliya Sani and Nuzhat Sultana

Department of Pathology, Pakistan Institute of Medical Sciences, Islamabad.

Objective: To find out the helpful cytological features distinguishing between hepatocellular carcinoma (HCC) with other carcinomas of liver

Study Design: Fine Needle Aspiration Cytology (FNAC) cases were selected in 35 patients on whom a diagnosis was rendered either of hepatocellular carcinoma or other carcinoma involving liver. The aspirates were examined for slide background, cellularity and specific cytoplasmic and nuclear features in both sets.

Result: There were 20 cases of hepatocellular carcinoma (HCC) & 12 cases of other carcinoma involving liver. Six out of twelve In the current study we examined 12 cytological criteria out of which six were found to be of considerable help in distinguishing hepatocellular carcinoma (HCC) from other carcinoma involving liver. The hepatocellular carcinomas characteristically had central nuclei, discohesive cells, high cellularity and naked nuclei while hyperchromatic nuclei, eccentric nuclei and nuclear cytoplasmic ratio of >75% were the prominent features of other carcinoma involving liver. On the other hand coarse chromatin, nuclear molding and chromatin clearing of nuclei showed no significant difference in distinguishing hepatocellular carcinoma from other primary and metastatic carcinoma of the liver.

Conclusion: Fine needle aspiration cytology (FNAC) of the liver is a very useful diagnostic modality to identify the vast majority of neoplasms of primary or metastatic nature. Meticulous attention to the nuclear features is vital in distinguishing hepatocellular carcinoma (HCC) from other carcinomas of liver.

Keywords: Hepatocellular carcinoma, HCC, FNAC of liver, other carcinomas of liver and metastatic carcinomas of liver

Introduction

Hepatocellular carcinoma (HCC) is the most common liver neoplasm. HCC is a major cause of morbidity & mortality in certain parts of Africa & Asia with the incidence of 100/100,000 people per year.1 On the other hand liver is a common site for metastatic tumors, cholangiocarcinoma and hepatoblastomas. As management of these lesions could profoundly differ, it is imperative to make a clear distinction between them. The Fine Needle Aspiration Cytology (FNAC) is an important useful diagnostic tool in this regard. Its value is further enhanced when it is combined with imaging techniques to determine the exact site for aspiration.2,3 FNAC provides many advantages in the diagnosis of abdominal masses. It permits rapid, accurate & correct diagnosis with a low complication risk. In recurrent or metastatic disease it averts an open biopsy.4 It can be performed in out patient clinic without anesthesia & can be repeated as often as required & results are available within few minutes.5

In many countries including Pakistan HCC constitutes the majority of hepatic neoplasms. At times differentiation between HCC and other neoplasms; primary and metastatic to liver is clinically difficult; FNAC can be used for making diagnosis in most cases. It is usually easy to render a diagnosis of malignancy, however it may be difficult to pinpoint the origin of the tumor.6 In the present study we analyzed 12 specific cytological features in HCC & compared them with other carcinomas involving liver in order to find out the useful distinguishing microscopic features.

Materials & Methods

The 32 cases of liver FNACs performed at Pakistan Institute of Medical Sciences (PIMS) and Islamabad Institute of Pathology were reviewed. The aspirates were either performed blindly or under ultrasound or CT scan guide. These included the cases from July 2003 –December 2005. The slides were stained with Hematoxylin & Eosin stain (H&E stain). All smears containing scant cellularity or significant artifact were rejected. The slides were reviewed by a
consultant pathologist along with a postgraduate student. 12 cytological criteria were evaluated and subjectively quantified on the score of 0-3. These criteria included cellularity, necrosis, discohesive cells, naked nuclei, irregular nuclei, central nuclei, eccentric nuclei, nuclear moulding, prominent nucleoli, coarse chromatin, chromatin clearing and nuclear cytoplasmic (N/C) ratio.

From total of 35 cases, 32 cases fulfilled the above criteria, out of which 20 were diagnosed as HCC and 12 as other carcinomas involving liver, which included cholangiocarcinoma, hepatoblastoma & metastatic adenocarcinomas.

**Results**

There were total 32 hepatic cases of neoplasia, out of which 20 were HCC & 12 were other carcinomas. Their cytomorphological features are listed in table 1. In terms of cellularity HCC did not differ much from other tumors. 60% HCC showed marked, while 40% showed mild to moderate cellularity. The other tumors showed marked cellularity in 33% cases and moderate cellularity in 42%. Central nuclei were seen in 100% hepatocellular carcinoma while they were seen only in 8% of other tumors. Discohesive cells were frequently seen in all case of HCC and they were numerous in 60% cases. On the other hand discohesive cells were not seen in other tumors in 41% cases and they were infrequently seen in another 41% cases. Naked nuclei were seen in overwhelming 75% cases in good numbers in HCC while they were seen only in 16% cases of other tumors in significant numbers. Hyperchromasia was not a distinguishing feature as it was seen frequently in both sets. Eccentric nuclei were frequently seen in (81.8%) cases of other tumors. They were present in all cases of adenocarcinoma, The eccentric nuclei were not seen in any case of HCC (Zero percent). Similarly nuclear cytoplasmic ratio (N/C ratio) of >75% was seen in 83.3% other tumors while this feature was not seen in any HCC. Necrosis was not a discriminatory feature as it was not seen in both sets in significant numbers. Coarse chromatin, moulding of nuclei and clearing of nuclei showed no marked difference in distinguishing HCC & other carcinoma involving liver.

**Discussion**

In the present study we analyzed 12 cytological features in HCC & compared them with other liver tumors. Some authors have used 28

<table>
<thead>
<tr>
<th>Table 1: Comparison Between Hepatocellular ca &amp; Other Neoplasms</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>Cellularity</td>
</tr>
<tr>
<td>Discohesive Cells</td>
</tr>
<tr>
<td>Necrosis</td>
</tr>
<tr>
<td>N/C Ratio</td>
</tr>
<tr>
<td>Moulding of Nuclei</td>
</tr>
</tbody>
</table>
| Hyperchromasia    | 0% | 0% | 35%| 65%| 0% | 0% | 0% | 100%
| Irregular Nuclei  | 0% | 70%| 20%| 10%| 8.3%| 8.3%| 50%| 50% |
| Clearing of Nuclei| 25%| 75%| 0  | 0  | 50%| 8.3%| 25%| 16.7%|
| Naked Nuclei      | 0% | 25%| 35%| 40%| 58.3%| 25%| 16.7%| 0% |
Prominent Nuclei | 35% | 35% | 10% | 20% | 91.7% | 8.3% | 0% | 0%

Figure 1: FNAC HCC showing Cellular & Nuclear Pleomorphism with Granular Cytoplasm, High N/C Ratio, Nuclear Membrane Breaks

Figure 2: FNAC HCC: High N/C Ratio & Coarse Chromatin

Figure 3: FNAC HCC: High N/C Ratio with Coarse Chromatin

Figure 4: FNAC Metastatic Small Blue Cell Carcinoma:

diagnostic cytological features reported in literature using some special techniques.\(^7\) Reported sensitivity and specificity of FNAC on metastatic tumors of liver in a local Pakistani study were 100% and 84.6% respectively.\(^8\) Another study that 87.96% tumors were diagnosed in patients above the age of 40 with male preponderance of 56%. Morphologically, malignant lesions were more common (74.08%) with hepatocellular carcinoma (HCC) as the most common malignancy (42.59%) followed by metastatic malignancies (23.15%), undifferentiated malignancies (5.55%) and Non-Hodgkin’s Lymphoma in 3(2.78%) patients while in two patients (1.85%) only normal hepatocytes were aspirated.\(^9\)

In Indian study reported several useful significant (\(P < 0.001\) to < 0.0001) discriminatory cytomorphological features, which included excessive cellularity, trabecular pattern, nuclear pleomorphism, atypical stripped nuclei, and macronucleoli. The most significant features for differentiating HCC from metastatic adenocarcinoma were trabecular growth pattern, hepatocytoc cells vs. columnar/cuboidal cells, eosinophilic granular cytoplasm, lipid vacuoles, bile pigments, and atypical stripped nuclei (\(P < 0.001\) to < 0.0001). The cytomorphological features which may distinguish poorly differentiated HCC from poorly differentiated adenocarcinoma were polygonal (hepatocytic) cells, eosinophilic granular cytoplasm and lipid vacuoles in HCC, and columnar/cuboidal cells and acinar/glandular formation in adenocarcinoma (\(P < 0.05\) to < 0.001).\(^10\)

A study by Bottles K, Cohen MB et al in 1988 revealed polygonal cells with centrally placed nuclei, malignant cells separated by sinusoidal capillaries & bile were identified as key cytologic features in diagnosing HCC.\(^11\)

Cohen et al identified 3 cytologic criteria: increased N/C ratio, trabecular pattern & atypical naked nuclei as most predictive of HCC with sensitivity of 100% & specificity of 87%.\(^12\) Sole et al while reviewing 102 HCC, 46 cases of metastasis and 28 cases of non neoplastic lesions. HCC showed irregular arrangement of cells in 82.4%, large nucleoli in 70.6%, naked nuclei in 67.6%, ill defined cytoplasmic membranes in 66%, irregular nucleoli in 59.4% and high cellularity in 55% of cases.\(^13\)

Tekenaka et al described certain cytological criteria for well differentiated, moderately differentiated & poorly differentiated HCC. The criteria set for well-differentiated HCC, included monotonous and scant cytoplasm, well-defined cytoplasmic borders, thick cytoplasm, eccentric nuclei, increased N/C ratio, thick nuclear membranes and increased chromatin density. He also established the criteria for moderately and poorly differentiated HCC as including three cytologic parameters: increased N/C ratio, irregular nuclear contours and increased chromatin density. Both sensitivity and specificity were 100% by
concurrently employing both criteria. These findings are not consistent with our observations. However their comparison was only among various types of HCC and not between HCC and other tumors. Their criteria would certainly lose value if the differential included non HCC tumors.

Andrew et al reviewed the cytological features of 16 cases of metastatic adenocarcinomas that were misclassified as HCC and compared them with classical 17 cases of metastatic adenocarcinoma. On review, cases that were frequently misclassified most often had moderate amounts of granular cytoplasm (16/16 cases) and round nuclei with even chromatin (13/16 cases). Trabeculae (3/16 cases), bare nuclei (2/16 cases), and endothelial wrapping (1/16 cases). In contrast, cases that were rarely misclassified were more likely to have areas with cells showing scant cytoplasm that were crowded and overlapped or molded (13/17 cases) and contained dark hyperchromatic chromatin (13/17 cases) compared to cases that were frequently misclassified (P < .001 and P = .002, respectively). Trabeculae (2/17) and bare nuclei (2/17 cases) were also rarely present. Another study by Andrew et al reviewed cases that were frequently misclassified as adenocarcinoma were misclassified 39% of the time (range, 18%–70%), while cases that were rarely misclassified were classified as adenocarcinoma 2% of the time (range, 0%–8%). The difference was statistically significant (P < .001). On review, 4 cytotologic patterns were found. The most common pattern for cases that were rarely misclassified was prominent trabeculae of cells and endothelial cells wrapping the trabeculae (6/10 cases vs 2/9 cases that were frequently misclassified). The most common pattern among cases that were frequently misclassified was clusters of cells with granular cytoplasm and associated stripped nuclei (5/9 cases vs 2/10 cases that were rarely misclassified). However, the distribution of neither pattern was significantly different (P = .16 for both). One case with large atypical granular cells, as seen in the fibrolamellar variant, was rarely misclassified. The remaining 3 cases (2 frequently misclassified, 1 rarely misclassified) had a nonspecific pattern of cells with granular cytoplasm without obvious trabeculae or stripped nuclei.

Conclusion: We evaluate 12 cytological criteria for distinguishing between HCC and other tumors including metastatic one. We found discohesive cells, naked nuclei, central nuclei, were the prominent features of HCC. While eccentric nuclei and N/C ratio >75 % were the prominent features of other carcinoma involving liver. On the other hand coarse chromatin, nuclear molding and chromatin clearing of nuclei showed no significant difference in distinguishing hepatocellular carcinoma from other primary and metastatic carcinoma of the liver. Further larger studies are required to crystallize the discriminating cytomorphological features.

References