The role of endoscopic ultrasound-guided fine needle aspiration (eus-fna) for the diagnosis of intra-abdominal lymphadenopathy of unknown origin

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Abbreviations: CT, computer-aided tomography; MRI, magnetic resonance imaging; EUS-FNA, endoscopic ultrasound with fine needle aspiration; PPV, positive predictive value; NPV, negative predictive value

Background and Aims: The diagnosis of intra-abdominal lymphadenopathy is difficult, especially when no primary lesion has been identified. We aimed to evaluate the diagnostic yield of EUS-FNA cytology in patients with enlarged intra-abdominal lymph nodes of unknown etiology.

Patient and Methods: 147 patients with abdominal lymphadenopathy on imaging in whom EUS-FNA was performed with a 22-gauge needle. Performance characteristics of EUS-FNA including the sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and accuracy were compared between the 2 groups.

Results: The location of the enlarged lymph nodes was the celiac axis (8.2%), peri-gastric (34%), peri-pancreatic (25.2%), peri-portal (27.9%), and other intra-abdominal locations (4.8%). The median number of EUS-FNA passes was 5. The final diagnosis were lymphoma (n=27), metastatic adenocarcinoma (n=44) patients, other miscellaneous malignancies (n=22) and benign disease (n=54). The sensitivity, specificity, and accuracy of EUS-FNA were 89.7, 98.3, and 93.5% respectively. A false positive FNA result was present in only 1 case (0.7%); false negative FNA results were present in eight cases (5.8%). Lymph node morphologic features of roundness, echogenicity, and homogeneity on EUS were not a predictor of lymph node malignancy.

Conclusion: In a retrospective cohort trial, EUS-FNA was found to be highly accurate and safe in diagnosing patients with intra-abdominal lymphadenopathy of unknown etiology.

Introduction

The advent of advanced diagnostic imaging modalities such as computer-aided tomography (CT) scanning and magnetic resonance imaging (MRI) has led to the increasing detection of enlarged intra-abdominal lymph nodes of unknown etiology. Without a clear primary lesion it is often times difficult to assess the malignancy potential of these enlarged lymph nodes. While primary lymphoma is a common cause of lymphadenopathy, many benign conditions of infectious or granulomatous may also lead to lymph node enlargement. Diagnostic laparoscopy, mediastinoscopy, and thoracoscopy are invasive and add unnecessary morbidity to a situation where the patient may have benign disease only.

Previous studies have examined endoscopic ultrasound

with fine needle aspiration (EUS-FNA) as a safe and effective method to obtain tissue diagnosis in these cases.^{1,2} Initially it was postulated that endosonographic features alone could predict malignant potential of lymph nodes, but later work has shown that fine needle aspiration adds to diagnostic yield and accuracy, potentially precluding the need for surgical exploration.³⁻⁶ EUS-FNA has repeatedly been proven a safe exam with examination with minimal complication rates and sensitivities and specificities approaching 100%.^{4,7-8} We examine a multi-center experience using EUS and EUS-FNA to predict the malignant potential of purely intra-abdominal lymphadenopathy without clear diagnosis of a primary tumor.

Patients and Methods

Patients

We conducted a retrospective analysis of consecutive patients with intra-abdominal lymphadenopathy of unknown origin found on trans-abdominal ultrasound, CT or MRI scan who presented to our institutions from November 2001 and December 2011.

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Patients were identified using our existing endoscopy database. EUS-FNA reports were then reviewed to obtain demographic information and endosonographic features of lymph nodes. FNA results and available surgical pathology were collected from institution electronic medical records and charts. Our inclusion criteria included the following: (1) The lymph nodes were located at locations that were accessible from the gastro-esophageal junction, stomach, or duodenum on the basis of computed tomography (CT); (2) Abdominal, pelvic and thoracic CTs, and previous endoscopic examinations had not identified a primary lesion. We excluded patients with mediastinal nodes, those with a previous history of malignancies and those with elevated tumor markers. The study was approved by the institutional review boards of the participating centers.

Methods

All EUS-FNA's were performed by experienced endoscopists who had performed greater than 500 EUS procedures. Target lymph nodes were initially identified using the radial scanning echoendoscope (Olympus GF-UE160, Melville, NY). A curvilinear echoendoscope (Olympus GF UCT 140 or UCT 160) was used then to perform the EUS-FNA with a 22-gauge Echotip ultra needle (Cook Medical, Winston-Salem, NC).

Cytology

Aspirates were placed onto glass slides and preserved with Diff-Quik stain (American Scientific Products, McGraw Park, Illinois, USA). In addition, a smear was also placed in alcohol for Papanicolaou staining. Any additional material was sprayed into Hanks's solution and sent for cell block processing. The cytology technician on-site verified the adequacy of specimens. The cytopathologist evaluated the material collected within 24 hours, after which a final report was submitted. A specimen was considered adequate if there were an adequate number of representative cells from the lesion. If insufficient tissue was present to make a definitive diagnosis, further sampling was obtained via laparoscopy.

The final diagnosis was determined by the results of EUS-FNA cytology, patient follow-up and final surgical pathology diagnosis if available. A final diagnosis of a malignancy was based upon (1) cytologic or histologic evidence of malignancy based upon material obtained by EUS-FNA, ERCP, surgical or percutaneous biopsy or (2) clinical course based upon follow-up in which that patient developed radiographic evidence of local or distant metastasis, or death attributed to a malignant lesion based upon clinical records. A lesion was defined as being benign based upon EUS-FNA results, clinical follow-up of at least 6 months and lack of disease progression. When a final diagnosis could not be established because of the lack of a criterion standard or adequate follow-up, patients were excluded when calculating operating characteristics.

EUS-FNA cytology samples considered to be adequate were interpreted as malignant, benign, or indeterminant. Patients with malignant cytology were classified as "true positive" if final diagnosis results supported initial cytology results and "false positive" if final pathologic results were benign in the face of an initial malignant cytology. Similarly, lesions classified as benign

by EUS-FNA with a final diagnosis of benign were considered "true negative" and those considered malignant on final diagnosis with benign initial cytology were "false negative".

Statistical analysis

Patients were analyzed in regards to their demographics and endosonographic appearance of target lymph nodes. Performance characteristics of EUS-FNA including the sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and accuracy were evaluated. These were determined by comparing EUS-FNA results with the final diagnoses of the lesions, based upon the above mentioned criteria. Accuracy was defined as the ratio of the sum of true positive and true negative values divided by the number of lesions. Categorical variables were reported as frequencies and compared using the two tailed Fisher Exact Test. Statistical significance was determined a priori at $p \leq 0.05$.

Results

Baseline Characteristics

From November 2001 and December 2011, we identified a total of 147 patients with abdominal lymphadenopathy on imaging in which EUS-FNA was performed. The median age of the patients at the time of the exam was 61 years (range, 27-83 years); 56% were female (62% White, 19% African American, 12% Hispanic, 7% other). The clinical presentation in these patients was weight loss (15%), poor appetite (19%), fevers (7%), and abdominal pain (34%); 25% of patients were asymptomatic and lymphadenopathy was found incidentally on imaging studies.

The location of the targeted lymph nodes included celiac axis in 12 cases (8.2%), peri-gastric in 50 cases (34%), peri-pancreatic in 37 cases (25.2%), peri-portal or hilar in 41 cases (27.9%), and in other intra-abdominal locations in 7 cases (4.8%). The median size of the sampled lymph nodes was 29mm (range 9-45mm) in the long axis and 16mm (range 8-25mm). Sampling of the nodes was successful in all cases. The median number of EUS-FNA passes was five (range 1 to 9). The final diagnosis was lymphoma in 27/147 (18.4%) patients, metastatic adenocarcinoma in 44/147 (29.9%) patients, other miscellaneous malignancies in 22/147 (15.0%) patients, and benign disease (reactive lymph nodes, sarcoidosis, or tuberculosis) in 54/147 (36.7%) patients. Patient characteristics and final diagnosis are summarized in **table 1**.

Of the 27 patients with a final diagnosis of lymphoma,

Operating Characteristics of EUS-FNA

Overall, EUS-FNA was able to provide an adequate initial sampling of tissue in 95% of patients. False positive FNA results were present in only one case (0.7%); in this patient the initial FNA was reported as malignant cells but final surgical pathology revealed reactive tissue only. False negative FNA results were present in eight cases (5.8%). The sensitivity of EUS-FNA was 89.7% (95% CI 80.3% - 95.1%) while the specificity was 98.3% (95% CI 89.9% - 99.9%). The positive predictive value of the test was 98.5% (95% CI 91.3% - 99.9%) and the negative predictive value was 88.1% (95% CI 77.3% - 94.3%). The accuracy of the

Table 1. Patient characteristics and final diagnosis of intra-abdominal lymphadenopathy

Patient Characteristics		Number of patients	Percentage
Gender	Male	65	44.2%
	Female	82	55.8%
Node location	Peri-Esophageal	12	8.2%
	Peri-gastric	50	34.0%
	Peri-pancreatic	37	25.2%
	Peri-portal	41	27.9%
	Other	7	4.8%
Final Diagnosis	Lymphoma	27	18.4%
	Metastatic Adenocarcinoma	44	29.9%
	- Gastric Cancer	5	
	- Peritoneal Carcinoma	1	
	- Duodenal Cancer	5	
	- Unknown Origin	19	
	- Colon Cancer	8	
	- Lung Cancer	4	
	- Endometrial Cancer	2	
	Cholangiocarcinoma	6	4%
	Pancreatic Cancer	4	2.7%
	Ampullary Cancer	3	2%
	Ovarian Cancer	1	0.7%
	Testicular Cancer	1	0.7%
	Unknown Primary	7	4.8%
	Benign/Reactive	54	36.7%

EUS-FNA exam to obtain the correct diagnosis was 93.5%.

Of the 27 patients with a final diagnosis of lymphoma, EUS-FNA with flow cytometry made an accurate diagnosis in 23/27 (83%) of patients; 3 of the 4 patients who were not diagnosed with EUS-FNA had a large cell lymphoma. Of the 23 patients diagnosed with lymphoma by EUS-FNA, lymphoma subtype was identified by flow cytometry examination to perform immunophenotyping in 16/23 (65%) of patients; the remaining patients underwent excisional lymph node biopsy, bone marrow biopsy and computed tomography (CT)-guided core biopsy to obtain adequate tissue of subtyping.

Echofeatures of Lymph Nodes

The associations between EUS features and lymph node cytology are summarized in **table 2**.

Lymph node shape was reported in 126/147 exams. Shape was defined in two categories: 1) round-appearing or 2) oval/triangular-appearing. Round lymph nodes were appreciated on 81 exams and oval or triangular lymph nodes on 45 exams. Sampling of round lymph nodes resulted in a malignant cytology sample in 44 patients (54.3%), a benign sample in 33 patients (40.7%), and an indeterminant result in 4 patients (4.9%). Sampling of oval or triangular lymph nodes resulted in a malignant cytology sample in 20 patients (44.4%), a benign sample in 22 patients (48.9%), and an indeterminant result in 3 patients (6.7%). No correlation was found between the endosonographic appearance of lymph node shape and prediction of malignancy ($p=0.34$).

The endosonographic pattern of lymph nodes was reported

in 134 of 147 exams. Pattern was defined in two categories: 1) homogeneous-appearing or 2) heterogeneous-appearing. Homogeneous lymph nodes were appreciated on 90 exams and heterogeneous lymph nodes on 44 exams. Sampling of homogeneous lymph nodes resulted in a malignant cytology sample in 47 patients (52.2%), a benign sample in 39 patients (43.3%), and an indeterminant result in 4 patients (4.4%). Sampling of heterogeneous lymph nodes resulted in a malignant cytology sample in 21 patients (47.7%), a benign sample in 20 patients (45.5%), and an indeterminant result in 3 patients (6.8%). No correlation was found between endosonographic appearance of lymph node pattern and prediction of malignancy ($p=0.85$).

The type of lymph node border was reported in 109 of 147 exams. Sharp-bordered lymph nodes were appreciated on 42 exams and indistinct-bordered lymph nodes on 67 exams. Sampling of sharp-bordered lymph nodes resulted in a malignant cytology sample in 26 patients (61.9%), a benign sample in 13 patients (31.0%), and an indeterminant result in 4 patients (7.1%). Sampling of indistinct bordered lymph nodes resulted in a malignant cytology sample in 32 patients (47.8%), a benign sample in 32 patients (47.8%), and an indeterminant result in 3 patients (4.5%). Again, there was no correlation between endosonographic appearance of lymph node border and prediction of malignancy ($p=0.11$).

Lymph node size was reported in 143/147 EUS exams. One hundred and eight patients had lymph nodes <10 mm in short axis and 35 patients had lymph nodes >10 mm in the short axis.

Table 2. Associations between EUS features and lymph node histopathology

Endosonographic criteria of lymph nodes and prediction of malignancy		Malignant	Benign	p
Shape	Round	44	33	0.34
	Oval/Triangular	20	22	
Pattern	Homogeneous	47	39	0.85
	Heterogeneous	21	20	
Border	Sharp	26	13	0.11
	Indistinct	32	32	
Size (Short axis mm)	<10	54	48	0.55
	≥10	15	18	

This cutoff was established from previously published literature.^{1,9,10} Sampling of lymph nodes <10mm in size resulted in a malignant cytology sample in 54 patients (50.0%), a benign sample in 48 patients (44.4%), and an indeterminate result in 6 patients (5.6%). Sampling lymph nodes resulted > 10mm in size resulted in a malignant cytology sample in 15 patients (42.9%), a benign sample in 18 patients (51.4%), and an indeterminate result in 2 patients (5.7%). Size of lymph nodes being > 10mm was not an accurate predictor of malignancy (p=0.55).

Complications

Immediate peri- and post-procedural complications were assessed in all procedures. No bowel perforations or significant intra-procedural bleeding after FNA was encountered. Two patients were admitted for mild abdominal that resolved with one day of hospital admission, a clear liquid diet and intravenous fluid hydration. One patient presented to the emergency room with abdominal pain a day after the procedure and was discharged home after evaluation.

Discussion

The results of this retrospective study demonstrate that EUS-FNA has a high rate of clinical success and accuracy in sampling tissue from enlarged intra-abdominal lymph nodes of unknown etiology. Sixty-three percent of patients with intra-abdominal lymphadenopathy were found to a malignancy while benign disease was diagnosed in 37% of patients. In addition, our findings clearly demonstrate that morphologic features of roundness, echogenicity, and homogeneity on EUS were not a predictor of lymph node malignancy. The current study is one of the largest series in the literature that is specially addressing the role of EUS-FNA for the diagnosis of enlarged intra-abdominal lymph nodes.

EUS-FNA has proven to be useful in the minimally invasive diagnosis of lymphadenopathy without a clear primary lesion. Original reports focused on mediastinal lymph nodes and reported a diagnosis rate of up to 90%, with a sensitivity and specificity of 85% and 100% respectively.^{2,10,11} Later work by Yasuuda et al demonstrated sensitivity of 96%, specificity of 100%, and PPV and NPV of 100% and 96.4% with a complication rate of only 1%, cementing the utility of EUS-FNA in the diagnosis of mediastinal and intra-abdominal lymphomas.¹² Furthermore,

the combination of EUS-FNA with flow cytometry and immunohistochemistry boosted the specificity of lymphoma diagnosis and allowed for the sub-classification of lymphoma types based on cytologic sampling alone.^{13,14} Future technologies such as quantitative spectral analysis of EUS images may provide additional discriminatory power between malignant and benign tissue, but such techniques are still in their infancy.¹⁵

The unique anatomic considerations of the lymphatic drainage of the intra-abdominal cavity provide a challenge to minimally invasive diagnosis not seen in the mediastinum. Metastatic disease from other sites accounted 30% of the cases and benign or reactive disease, including tuberculosis and sarcoidosis was seen in nearly 37%. This reinforces the utility of a minimally invasive approach such as EUS/FNA as these are the patients who benefit the most from the potential avoidance of surgical intervention.

Our overall operating characteristics of EUS-FNA are in keeping with previously published literature.^{5-8,10} Specificity and positive predictive value were over 98%, echoing previously reported low false-positive rates and reiterating the utility of EUS-FNA in the diagnosis of malignant lesions. Accuracy of combined diagnosis was 93.5%.

Relaying on endosonographic features alone was not supported by our findings. Older studies have shown some promise in identifying potentially malignant lymph nodes based on ultrasound features alone.^{1,6,9,10,16} More recent work has demonstrated that there is no one reliable endosonographic feature to suggest malignant potential.^{17,18} Size has been pointed to as a potential concerning feature but our rather large series showed no correlation between size and malignancy.^{1,3, 9,10,18} Likewise the pattern of lymph node echo-scattering, the sharpness of its border, and lymph node shape were not predictive of malignant potential in our series, suggesting that with current, readily available technology, FNA and cytology will remain the mainstay of discriminating a diagnosis.

We were in favor of using the radial echoendoscope prior to linear EUS and FNA since it allowed us to obtain a more “global” view of the gastrointestinal tract, aided in visualizing the target lymph node(s), in determining the depth of scope insertion needed to visualize the target, and identifying adjacent structures to evaluate for overt mass lesions.

Our study had some limitations. First, it was retrospective. In addition, surgical pathology was not available in all our cases since the majority of patients at the time of diagnosis. However,

we had good clinical follow-up and therefore were able to follow the natural history of the patient over at least six months.

In summary, EUS-FNA is a reliable and minimally invasive way to establish a diagnosis in the setting of isolated intra-abdominal lymphadenopathy. Its operating characteristics are reliable to make it the clear first line test, potentially precluding the use of diagnostic surgical techniques. Endosonographic features alone are not sufficient to establish a diagnosis, although emerging technologies may change this in the future.

Conflict of interests

The authors attest that they have no commercial associations (e.g., equity ownership or interest, consultancy, patent and licensing agreement, or institutional and corporate associations) that might be a conflict of interest in relation to the submitted manuscript.

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References

1. Catalano MF, Sivak MV Jr, Rice T, Gragg LA, Van Dam J. Endosonographic features predictive of lymph node metastasis. *Gastrointest Endosc* 1994; 40: 442-6.
2. Williams DB, Sahai AV, Aabakken L, Penman ID, van Velse A, Webb J, et al. Endoscopic ultrasound guided fine needle aspiration biopsy: a large single centre experience. *Gut* 1999; 44: 720-6.
3. Eloubeidi MA, Wallace MB, Reed CE, Hadzijahic N, Lewin DN, Van Velse A, et al. The Utility of EUS and EUS-guided fine needle aspiration in detecting celiac lymph node metastasis in patients with esophageal cancer: A single center experience. *Gastrointest Endosc* 2001; 54:714-9.
4. Chen VK, Eloubeidi MA. Endoscopic ultrasound-guided fine needle aspiration is superior to lymph node echo features: A prospective evaluation of mediastinal and peri-intestinal lymphadenopathy. *Am J Gastro* 2004; 99: 628-33.
5. Nakahara O, Yamao K, Bhatia V, Sawaki A, Mizuno N, Takagi T, et al. Usefulness of endoscopic ultrasound-guided fine needle aspiration (EUS-FNA) for undiagnosed intra-abdominal lymphadenopathy. *J Gastroenterol*. 2009; 44: 562-7.
6. Bhutani MS, Hawes RH, Hoffman BJ. A comparison of the accuracy of echo features during endoscopic ultrasound (EUS) and EUS-guided fine needle aspiration for diagnosis of malignant lymph node invasion. *Gastrointest Endosc* 1997; 45: 474-9.
7. Williams DB, Sahai AV, Aabakken L, Penman ID, van Velse A, Webb J, et al. Endoscopic ultrasound guided fine needle aspiration biopsy: A large single center experience. *Gut* 1999; 44: 720-6.
8. Chhieng DC, Jhala D, Jhala N, Eltoum I, Chen VK, Vickers S, et al. Endoscopic ultrasound guided fine needle aspiration biopsy: A study of 103 cases. *Cancer* 2002; 96: 232-9.
9. Wiersema MJ, Hassig WM, Hawes RH, Wonn MJ. Mediastinal lymph node detection with endosonography. *Gastrointest Endosc* 1993; 39:788-93.
10. Catalano MF, Alcocer E, Chak A, Nguyen CC, Rajjman I, Geenen JE, et al. Evaluation of metastatic celiac axis lymph nodes in patients with esophageal carcinoma: accuracy of EUS. *Gastrointest Endosc* 1999; 50:352-6.
11. Catalano MF, Nayar R, Gress F, Scheiman J, Wassef W, Rosenblatt ML, et al. EUS guided fine needle aspiration in mediastinal lymphadenopathy of unknown etiology. *Gastrointest Endosc* 2002; 55:863-9.
12. Yasuda I, Tsurumi H, Omar S, Iwashita T, Kojima Y, Yamada T, et al. Endoscopic ultrasound guided fine needle aspiration biopsy for lymphadenopathy of unknown origin. *Endoscopy* 2006; 38:919-24.
13. Ribeiro A, Vazquez-Sequeiros E, Wiersema LM, Wang KK, Clain JE, Wiersema MJ et al. EUS guided fine needle aspiration combined with flow cytometry and immunochemistry in the diagnosis of lymphoma. *Gastrointest Endosc* 2001; 53:485-91.
14. Al-Haddad M, Savabi MS, Sherman S, McHenry L, Leblanc J, Cramer H, et al. Role of endoscopic ultrasound-guided fine-needle aspiration with flow cytometry to diagnose lymphoma: A single center experience. *J Gastro Hepatol* 2009; 24(12): 1826-33.
15. Kumon RE, Pollack MJ, Faulx AL, Olowe K, Farooq FT, Chen VK, et al. In vivo characterization of pancreatic and lymph node tissue by using EUS spectrum analysis: a validation study. *Gastrointest Endosc* 2010; 71: 53-63.
16. Faige DO. EUS in patients with benign and malignant lymphadenopathy. *Gastrointest Endosc* 2001; 53: 593-8.
17. Kanamori A, Hirooka Y, Itoh A, Hashimoto S, Kawashima H, Hara K, et al. Usefulness of contrast-enhanced endoscopic ultrasonography in the differentiation between malignant and benign lymphadenopathy. *Am J Gastroenterol*. 2006; 101: 45-51.
18. Song HJ, Kim JO, Eun SH, Cho YD, Jung IS, Cheon YK, et al. Endoscopic ultrasonographic findings of benign mediastinal and abdominal lymphadenopathy confirmed by EUS-guided fine needle aspiration. *Gut Liver* 2007; 1: 68-73.