Role of Rapid On-site Evaluation in CT-guided Fine Needle Aspiration Cytology of Lung Nodules

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Abstract

Objective: To prospectively investigate the value of rapid on-site evaluation (ROSE) in transthoracic fine needle aspiration cytology (FNAC) of patients with pulmonary nodules. Computed tomography (CT)-guided FNA is commonly employed for the diagnosis of lung lesions and the most common reason for not being able to provide a diagnosis in FNA is inadequacy of samples. Materials and Methods: This was a prospective study conducted in the departments of pathology and radiology of our cancer centre. This study had approval from the institutional review board and ethical committee of our institute. Fifty consecutive cases undergoing CT-guided transthoracic FNAC in our centre were included in the study. The smear submitted for ROSE was stained using toluidine blue stain. The specimen adequacy and diagnosis in ROSE was compared with that of the final cytology report, and the concordance regarding adequacy and diagnosis were noted. Results: Smears were adequate in 34 cases (68%) and inadequate in 16 cases (32%). Out of the 16 inadequate cases, 5 (31%) were converted to adequate due to the application of ROSE, thus increasing the adequate number of cases to 39 (78%). A diagnosis of malignancy was made in all 39 adequate cases. Sensitivity of ROSE in determining adequacy was 92% and specificity was 100%. The most common malignancy was adenocarcinoma in 26 cases. Pneumothorax occurred in 2 cases. No significant complications occurred in other cases. Conclusion: CT-guided FNA with ROSE is a safe and useful tool in the diagnostic work-up of lung cancer patients. A multidisciplinary approach along with onsite evaluation of adequacy will increase the diagnostic utility of cytology in lung lesions.

Keywords: Adequacy, computed tomography, fine needle aspiration cytology, lung mass, rapid onsite evaluation

Introduction

Lung cancer is a leading cause of cancer death. The high mortality associated with lung cancer is mainly because a significant number of lung cancers are detected even today at an advanced stage. This diminishes the role of radical surgery; the current management of these advanced stage patients includes personalized medicine where treatment decisions are taken based on the specific histology and molecular characteristics of the tumor. This requires obtaining samples from the patients by minimally invasive procedures. Thus, the need of the day is use of technology for precise and focused sampling of clinically and radiologically suspicious lesions to provide an accurate cytological/histological diagnosis resulting in an appropriate treatment.

Advanced radiological techniques have improved the detection of and access to smaller lesions of the lung. In the current scenario, the skill of the radiologist is in obtaining representative specimens from the lesion and for the pathologist to establish a diagnosis on limited cytological and histological specimens. Transthoracic fine needle aspiration (FNA) under computed tomography (CT) guidance is widely employed for its high success in morphological diagnosis and low complication rate. The availability of rapid on-site evaluation (ROSE) will further enhance the utility of this procedure by ensuring adequacy of samples so that, if inadequate, the procedure can be repeated immediately to ensure adequacy and avoid delay in report or having to repeat the procedure at a later date.

In this study, we assessed the efficacy and feasibility of ROSE in CT-guided FNA of lung nodules. We also assessed the concordance of ROSE with final cytology regarding adequacy and diagnosis.

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**Materials and Methods**

This single centre, prospective study was conducted in the department of pathology of our cancer centre in association with the department of radiology. The study was started after obtaining approval from the institutional review board and the ethical committee of our centre. Fifty patients having radiologically diagnosed pulmonary mass lesions that can be approached through the transthoracic route were included. These patients had strong clinical suspicion of pulmonary neoplasm. Informed consent was obtained in all the cases prior to the procedure.

Fine needle aspiration cytology (FNAC) was performed as an outpatient procedure in the presence of a pathologist and radiologist. Patients were positioned according to the location of lesion in the preliminary CT-scan. A 20–22-gauge needle with a 20 ml syringe was used for aspiration. When the tip of the needle reached the outer edge of the lesion, a repeat slice of the area of interest was taken to check the exact position of the tip. The patient was advised to hold the breath while the needle was being inserted. The aspirated material was smeared on the slides. Additional material, if any, was submitted for cell block. One smear considered to be visually adequate was submitted for ROSE and was stained rapidly with toluidine blue. The toluidine blue stain was prepared in our laboratory by dissolving 0.5 g of crystalline toluidine blue in 20 ml of 95% ethanol and making the solution up to 100 ml by adding distilled water. The solution was then filtered and refrigerated till use. The smears were reported by an experienced pathologist regarding adequacy, and if adequate, a provisional diagnosis was given. The turnaround time of ROSE was less than 5 min. The result of ROSE was communicated to the radiologist, and based on this the decision was made as to whether a repeat procedure was needed. If the sample was adequate in the first pass itself, as confirmed by ROSE, no further passes were done.

Inadequate cases were subjected to a repeat procedure according to the discretion of the radiologist, i.e., if no complications were expected a repeat procedure was done immediately. If there was any indication of pneumothorax or hemoptysis, then the repeat procedure was deferred to a later date.

The slides not submitted for ROSE were fixed in alcohol and submitted for routine Papanicolaou stain. Patients were kept under observation, and if no subsequent complications such as pneumothorax occurred, they were discharged the same day.

**Results**

Fifty cases included 39 males and 11 females. The maximum numbers of patients were in the age group of 61–70 years. In 34 cases (68%), in the initial procedure itself sampling was adequate. Sensitivity, specificity, and positive and negative predictive values of ROSE in evaluating adequacy were calculated.

Of these 34 cases, ROSE was able to confirm adequacy in 31 cases; however, in 3 cases ROSE gave a false negative result of inadequacy. This was because the slides submitted for ROSE did not have adequate material but slides submitted for routine processing had adequate material.

Slides from single pass may sometimes show difference in the distribution of material. Thus, this false negative result cannot be considered a limitation of ROSE. There was one case in our study wherein ROSE was adequate but smears stained with Papanicolaou stain were inadequate. However, we had cell block for this case. In situations where only the toluidine blue stained smear shows material, one can always perform destaining with 95% ethyl alcohol followed by restaining with permanent stains. This is an advantage of toluidine blue staining. Toluidine blue method is not only a reliable method for rapid staining and diagnosis it also permits preservation of cytological material by destaining and restaining with permanent stains.

The sensitivity of ROSE in determining adequacy was 92%; there were 16 inadequate cases. ROSE was able to confirm inadequacy in all 16 cases with a specificity of 100%. Positive predictive value was 100%, and negative predictive value was 84% [Table 1]. Out of the 16 inadequate cases, 5 were converted into adequate with the use of ROSE. A single aspiration was sufficient in 34 cases (87%), two aspirations were required in 2 cases (5%), and three aspirations were required in 3 cases (8%) to attain adequacy. Thus, with onsite evaluation in a single sitting, 5 inadequate cases could be converted to adequate samples and a diagnosis could be given. Thus, with ROSE the adequate samples could be increased from 68% to 78%. We did not come across any true false positive cases in our study group. Though there was one case in our study wherein ROSE was adequate but smears stained with Papanicolaou stain were inadequate, we did not consider this as false positive because the same case showed adequate material in sample submitted for cell block preparation, which was done in the same sitting. Hyperplastic reactive respiratory epithelium is one of the causes for a false positive diagnosis. Pneumothorax occurred in 2 cases. No significant complications occurred in other cases.

In all 39 adequate cases, a diagnosis of malignancy was made. Subtyping of malignancy could also be satisfactorily done on smears of onsite evaluation. Most common malignancy was adenocarcinoma (26 cases) (including primary as well as metastatic disease), 6 cases of squamous cell carcinoma, and 5 cases of small cell carcinoma. In 2 cases, a definite subtyping was not possible and a diagnosis of nonsmall cell carcinoma was given. In most of the cases of adenocarcinoma,
a glandular pattern could be identified [Figure 1a]. Squamous cell carcinomas showed dirty necrotic background and keratinization [Figure 1b]. Small cell carcinoma showed small/medium-sized cells with coarse chromatin and molding of nucleus [Figure 1c]. One of the cases of metastatic carcinoma was from renal cell carcinoma and showed the classical vesicular nucleus with prominent nucleoli [Figure 1d].

A satisfactory agreement was achieved in differentiating the cancer subtypes. There was 100% agreement between morphological diagnosis in ROSE and final cytology. Wherever cell block preparations were available (18 cases), a limited immunohistochemical panel comprising p40 and TTF-1 was used to subcategorize nonsmall cell carcinoma as adenocarcinoma [Figure 2a] or squamous cell carcinoma [Figure 2b]. A panel of CK, synaptophysin, and chromogranin was used in cases of small cell carcinoma.

**Discussion**

At present, different modalities are available to procure samples from suspicious lung lesions. The choice of a particular modality for sampling a lesion is based on the size and location of the lesion.[5] CT-guided FNA is a minimally invasive method for diagnosing lung lesions, especially the peripheral ones. When compared to other modalities it produces only minimum discomfort to the patient. Complications are rare and include hemoptysis and pneumothorax, which are usually self-limiting. However, it is time consuming in terms of patient positioning and localization.

The main limitation of FNAC diagnosis is inadequate samples. Inadequate sampling results in delay in diagnosis and the need to repeat procedure increasing patient anxiety. Onsite evaluation can assess the adequacy of smears at the time of the procedure so that if the sample is inadequate procedure can be repeated in the same sitting and adequacy of sample can be ensured. The presence of a pathologist can even ensure a diagnosis within minutes.

ROSE is a simple technique which is underutilized in the Indian scenario. This is mainly because of shortage of staff and lack of awareness of the potential of this simple technique. Studies have demonstrated that ROSE can allow reduction of the number of needle punctures, reduction in complication rates, lowering procedure time and costs, and most importantly improving the diagnostic yield.[6]

Availability of cytopathologist for onsite evaluation may not be feasible always. An alternative is to train pulmonologist/radiologist/cytotechnicians to assess adequacy. Bonifazi et al. reported an 81% overall agreement between pulmonologist and cytopathologist in the evaluation of ROSE when ROSE was performed by a pulmonologist after a 3-month training in cytopathology.[7]

Furthermore, ROSE can be limited to cases where technical difficulty is anticipated in procuring samples so that in a...
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single procedure itself adequacy of samples can be ensured. Moreover, in centers where due to shortage of staff ROSE could not be routinely employed, it should at least be mandatory in cases where the procedure is being repeated.

There is also the need to educate all concerned personnel regarding the utility of ROSE. Though the application of ROSE may seem time consuming initially in the long run it saves time of clinicians, pathologists, and technicians by avoiding the need to repeat procedure. It also prevents delay in starting the treatment.

This study, similar to previous studies, concludes that CT-guided FNAC is an accurate and safe method with high sensitivity and specificity for the diagnosis of pulmonary nodules. ROSE can subclassify the morphological type of bronchogenic carcinoma in a majority of cases. The use of ROSE helps in ensuring the adequacy of sample, minimize the number of passes required, and thereby minimize complications such as pneumothorax.

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Conflicts of interest
There are no conflicts of interest.

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