



ORIGINAL RESEARCH PAPER

Respiratory Medicine

DIAGNOSTIC ROLE OF GASTRIC ASPIRATION IN SMEAR NEGATIVE PULMONARY TUBERCULOSIS IN ADULT.

KEY WORDS: Diagnostic role ; Gastric Aspiration ; sputum smear negative pulmonary tuberculosis.

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ABSTRACT

To study the diagnostic utility of gastric aspiration in sputum negative pulmonary tuberculosis by identifying gastric smear for AFB and culture. To compare the outcome among HIV positive and negative individuals. To study how many were false negative due to improper techniques in sputum sample submission. To compare the radiological patterns with yield in gastric aspiration. To investigate the role of bronchoscopy in smears negative pulmonary tuberculosis. From this study, gastric aspirate for smear positivity among sputum smear negative pulmonary tuberculosis was 24% and gastric culture positivity was 22%. Gastric aspirate for AFB smear and culture can be used as a tool in diagnosis of pulmonary tuberculosis in patients who cannot submit sputum and patients who are smear negative as suggested by the study. Gastric aspirate performed on 2 consecutive days may improve the diagnostic yield. Gastric aspirate performed among HIV positive patients was only 10%. Gastric aspirate positivity in culture had a high yield among chest xray with suspected pulmonary tuberculosis with consolidation pattern(38%). There is practice of prescribing fluoroquinolone for community acquired pneumonia in private. The patients seeking treatment in a tertiary care centre are not naive population. Many of them would have had treatment and exposed to fluoroquinolones before presenting to us and hence these patients are at the risk of developing fluoroquinolone resistant pulmonary tuberculosis. DOTS program in India is functioning at the level of primary health centre where sophisticated techniques like bronchoscopy are not available, patient presenting with no sputum under suspicion of tuberculosis, gastric aspirate can be performed as a diagnostic technique which is a simple outpatient procedure.

INTRODUCTION

Tuberculosis is still one of the most deadly infectious diseases among developing countries like India with the mortality of around lives of over 2 million all over the world. Early diagnosis and treatment with effective chemotherapy in adequate dosage will cut down the morbidity and mortality. The annual report of 2011-RNTCP gives the update on the prevalence and mortality rate. There is fall in the prevalence and mortality over 10 yrs and target has been set for better outcome. A pulmonary tuberculosis suspect is a one with Cough for more than 2 weeks associated with night sweats, weight loss, haemoptysis, low grade fever. All these TB suspects have to be subjected for sputum examination. The problem of situation is that there a subset of population who present with symptoms other than cough and pulmonary tuberculosis has to be proved by varied techniques for the presence of acid fast bacilli either in smear or culture. The TB infectiousness among smear negative pulmonary tuberculosis is around 11% as CDC identifies in a study. There are various techniques for obtaining representative of pulmonary specimens for AFB smear and culture. Most common and prime sample to be collected in patients with pulmonary tuberculosis is sputum sample.

MATERIAL AND METHODS

In this study, gastric smear aspirated has been examined under fluorescent microscope and media for culture used is LJ media. Fixed smears are covered with 0.3% auramine and stained for 15 min and washed with 0.5% acid alcohol. Then the slide is stained with 0.1% potassium permanganate. The problem with the technique is highly expensive requires dark rooms and difficult to handle. There are higher chance of false positive where the artefacts were take up the stain. The bulb which is one of the component parts needs to be changed from time to time and may be expensive to procure and repair. Continuous power supply is needed with minimal voltage fluctuations for maintenance. Fluorescent microscope offers shorter time than conventional microscopy after staining by Ziehl-Neelson staining. One of the study quotes that a 100 smears /day examined can be compared with only 30-40 Zn

stained smears. Acid fast bacilli from gastric aspirate fluid appearing as pink shaped rods against blue background in the conventional ZN staining. The conventional ZN staining employs carbol fuchsin as primary staining upon which 25% sulphuric acid is added following which decolourisation with 95% alcohol is done. Counter stain with methylene blue imparts blue colour to the background for identification of slender slightly curved rods under oil immersion field. There are other methods of staining like cold staining techniques like cold staining techniques called KINYON staining with higher concentration of basic fuchsin and phenol. The most common used media is LJ media containing egg yolk which sustains the growth of MTB. Green color to the media is given by malachite green which helps in identification of organism early. The media contains glycerol, amino acids which provide the source of energy for growing mycobacteria. This media supports the growth of both typical and atypical organisms. After obtaining informed consent from the patient, the patient is asked to be on overnight fasting at least for 8 hrs from previous day night. The patient after applying lignocaine gel over nasal passage and over the naso gastric tube of appropriate size is taken. Nasogastric tube is introduced and as soon as tube reached the gaster, position confirmed and at least 15-20 ml of gastric aspirate fluid collected in a sterile container without adding any fluid to it. If patient feels uncomfortable anytime, NG tube is withdrawn. The sample collected is labelled with a requisition slip and transported to the lab immediately. The transit time not more than 10 minutes. After obtaining the sample 4% NaOH is added to it for at least 10 min. The idea of adding NaOH is that it will destroy the bacteria other than mycobacterium, help to buffer the acidity of a gastric aspirate, also help to homogenise the sample. Centrifugation is done at the rate of 3000 rpm/min for at least 5 minutes. After centrifugation supernatant is discarded and equal volume of normal saline is added to the sediment. Again the sample is subjected to centrifugation at the rate of 3000 rpm/min for 5 minutes. Similarly the supernatant is discarded and sediment is taken. The sediment is then taken for preparation of smear at least 2 slides are prepared for each sample and examined under fluorescent

microscope for acid fast bacilli A part of sediment is taken for culture inoculation also. For each sample 2 tubes are used for inoculation. The conventional media used in our lab is Lowenstein Jensen medium which is egg based enriched medium. The inoculum is incubated in L J slant culture at 37 deg C for a period of at least 4-8 weeks. Reading is taken every week. Most of the time colonies appear during 3-4 week. The contaminated sample with other bacteria most often showed varied growth and spoil the media and those samples are discarded. The sample is considered to have no growth after a wait period of at least 8 weeks after which the samples are discarded. The culture positive specimens are those which grow colonies showing irregular raised dry, wrinkled colonies which are initially white to start with later appear with a buff colour. There are various biochemical tests available to differentiate between typical and atypical mycobacteria but we are not provided with such tests in our lab. Details among patients with history of contact with history of ATT was also noted. Chest skia gram findings are noted: In general, most common presentation of pulmonary tuberculosis is a cavity disease involving upper lobes. Patient may have sputum production if such a cavity is communicating with airway.

RESULTS

Total 165 patients were selected based on eligibility criteria. There were many drop outs as some of them does not want to undergo the procedure. During the procedure there were drop outs. After obtaining the sample some of the specimens were contaminated and discarded. Ultimately we arrived at 125 subjects. The categorisation of smear negative cases with clinical and radiological suspicion of pulmonary tuberculosis were made as follows 1. Sputum smear negative after submitting 2 samples for AFB(82) 2. No complaints of sputum production (26) 3. Unconscious patients (17) In this study about 125 patients participated where 102(81%) males and 23(18%) females. During the selection of patients both male and female were selected. We intended to take both male and female equally. There were more drop outs due to acceptance of procedure and many drop outs at the time of performing the procedure. Many of them were concurrently not willing for bronchoscopy among females. The age group was sliced at range of 10 difference of interval for convenience. It is seen that middle age group contributes to higher proportion of population. Gastric culture positivity was high among younger age group of population for which the P value is significant (p value=0.009). This is evident from the P value because it is statistically significant p value. Chest xray is divided into upper, middle and lower lung zones on each side for descriptive purpose From the above data it is inferred that most of patient had at least 2 Or 3 lung zones involved with the lesion either new cases or treated patients. The idea of collecting the lung zones involvement is that to arrive at whether many zones should be involved or even a small lesion with a cavity can show a positive culture. From the above data it is inferred that lung zones involved has no influence over gastric culture positivity because p value is not statistically significant (p value>0.6). Even a small cavity involving one zone or if many zones are involved does not influence gastric aspirate positivity and bacillary burden. From the above data it is seen that percentage of positivity is marginally more among treated cases. This does not carry much of statistical significance. This may be because many of them are defaulters of antituberculosis therapy as observed from our hospital. This may be reason for marginal high positivity among treated cases. Many of patients in our centre obtained treatment for HIV/TB mostly had an extra pulmonary presentation and hence the percentage may be low among HIV patients. It is inferred that only 22 percent population were HIV infected participated in the study. Among HIV positive patients the yield was low -11 % cases were positivity on culture in L J medium when compared with HIV negative patients. This is because there is varied radiological presentation in HIV disease. Most of presentation to our

hospital is with advanced AIDS presentation. Tuberculosis at this stage presents as a disseminated disease in the form of military tuberculosis or extra pulmonary nodal disease. This may be the reason for low yield among HIV positive individual. From the data, sputum for AFB after subjecting the patients for gastric aspiration, patients were educated as how to give the sputum for AFB. The yield of sputum for AFB was 6 among these set of people, 2 people have been already positivity in gastric aspirate. 4 people had been proved positivity in sputum sample after subjecting them for AFB. Many of the patients who underwent gastric aspiration were not willing for bronchoscopy. Rest 22 of them among 125 were willing for bronchoscopy. Among them 7 cases were proved positive for AFB by bronchoscopic brush. These patients were negative on gastric aspirate both by smear and culture. Whereas gastric aspirate identified 28 patients for AFB by culture. Though bronchoscopy can provide specimen exactly from suspected diseased segment, occupational exposure to AFB and other infectious agent through aerosol is very high while performing a bronchoscopy. Hence bronchoscopy is preferred only as a last resort in our study when there is a high clinical suspicion or doubt in our diagnosis.

DISCUSSION

There are many studies and articles on the role of gastric aspirate in children. There are varied techniques available for obtaining a representative sample for proving pulmonary tuberculosis in adults In our study 125 patients accepted to undergo gastric aspiration. Subjecting the patient for gastric aspiration required choosing of right candidate for the study criteria. **DEMOGRAPHIC FACTORS: SEX:** In this study most of patients were male subjects presented with clinical features of pulmonary tuberculosis. This confirms the fact that the disease is more common among men perhaps due to contact from external source, immunocompromised state like chronic alcoholism. From the study, it is inferred that female patients were more reluctant to undergo any invasive procedure whether gastric aspiration or bronchoscopy. Another reason for less female population presented is because of ignorance, inability to produce sputum, family problems, approach to health care centre presenting with diagnosis and treatment delay. It is inferred that the set of people suspected to have pulmonary tuberculosis were falling under middle age group, proving that pulmonary tuberculosis is a disease of middle age working class population. This data addresses the need to suspect the people of middle age group with respiratory complaints for pulmonary tuberculosis. On contrary, were able to prove by L J culture with significant growth for tubercle bacilli among people with age group <25yrs. This is because many of them expectorate the saliva rather sputum for testing or they are very sick to submit the sample for examination. Bahaumann et al in their study concludes that gastric smear for AFB though not sensitive is highly specific to initiate treatment on antituberculosis therapy. He also emphasis here that if gastric aspirate smear is positive for AFB it indicates that the bacillary load is enormous. **GASTRIC ASPIRATE AND ANTITUBERCULOSIS THERAPY:** The gastric aspirate smear for AFB and culture positivity does not seem to be influenced by prior episode of antituberculosis therapy. The yield appears to be almost equal among both treated cases for pulmonary tuberculosis and also among new case suspects. **RADIOLOGY AND GASTRIC ASPIRATE:** Radiological suspicion of pulmonary tuberculosis based on type of lesion adds to the fact from our study that though exudative lesion had more common suspicion, gastric culture positivity was more among people with consolidation like picture. Dooley et al identified that there is delay in the diagnosis of pulmonary tuberculosis by at least 16 days to start on anti tuberculosis therapy in suspected cases of community acquired pneumonia. He also adds in his study that many of the patients with suspected community acquired pneumonia are exposed to respiratory fluoroquinolone before starting them on conventional antituberculosis therapy. This may lead to emergence of fluoroquinolone resistant strains of pulmonary

tuberculosis. This may be one of the reasons for sputum smear negativity due to short course of fluoroquinolone. This also emphasises our need to establish the diagnosis of pulmonary tuberculosis and decide on ATT as soon as possible. **POST GASTRIC ASPIRATE SPUTUM SPECIMEN:** The study population were educated after having undergone a gastric aspiration to submit sputum for AFB. This time they were all hydrated well and asked to give thick tenacious sputum as soon as getting up from the bed and submit the sputum for AFB. It is inferred that there are as small subset of population who among the smear negative category who were not able to give good quality sputum when educated and hydrated well were able to prove positive by conventional sputum smear technique. This emphasis the need to conduct training session for health care professionals regarding good quality adequate sample so that they can help in avoiding the false negative sputum samples. **BRONCHOSCOPY AND GASTRIC ASPIRATE:** Bronchoscopy is a invasive procedure needing technical expertise. We need to prepare the patient, constant monitoring during the procedure and chance of equipment related contamination is possible when performing a bronchoscopy. The patients acceptance and cooperation to undergo bronchoscopy also is less among our study population because all who underwent gastric aspiration were not ready to undergo bronchoscopy. More over gastric aspirate can be performed by staff and paramedics as compared to bronchoscopy which demands technical assistance. The culture studies performed in this study on conventional LJ medium. Rapid culture techniques may help in starting on treatment rapidly. Further studies have to be done on gastric aspirate with rapid culture techniques.

CONCLUSION

Gastric aspirate for smear positivity among sputum smear negative pulmonary tuberculosis was 24% and gastric culture positivity was 22%. Gastric aspirate for AFB smear and culture can be used as a tool in diagnosis of pulmonary tuberculosis in patients who cannot submit sputum and patients who are smear negative as suggested by the study. Gastric aspirate performed on 2 consecutive days may improve the diagnostic yield. Gastric aspirate performed among HIV positive patients was only 10%. This may be because HIV infected in this institution had extra pulmonary forms and disseminated tuberculosis rather than pulmonary presentation. In this study it is also identified that false negative sputum smear samples were also present as some of the cases had post gastric aspirate sputum smear positivity. Gastric aspirate positivity in culture had a high yield among chest x-ray with suspected pulmonary tuberculosis with consolidation pattern (38%). There is practice of prescribing fluoroquinolone for community acquired pneumonia in private. DOTS program in India is functioning at the level of primary health centre where sophisticated techniques like bronchoscopy are not available, patient presenting with no sputum under suspicion of tuberculosis, gastric aspirate can be performed as a diagnostic technique which is a simple outpatient procedure. The sample can be subjected for AFB smear study in the lab immediately. The procedure can be performed by trained health care professional like staff nurses unlike bronchoscopy which needs to be performed by trained bronchoscopist with constant monitoring.

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