Sputum collection in children has been difficult, as they usually swallow the expectorate coming from the lungs. To obtain respiratory tract secretions, gastric lavage (GL) and bronchoalveolar lavage (BAL) have been used. GL, which collects the respiratory secretions that are swallowed at night, is performed early in the morning after an overnight fast. GL is typically performed in hospitals, which increases diagnosis costs since patients have to stay in hospital for an average of 3 days. In contrast, BAL samples the alveolar epithelial lining fluid directly, and has been found to be useful for the diagnosis of several respiratory infections, including TB. Isolation rates for M. tuberculosis from GL cultures in children have been reported to vary from 20% and 40%, However, the culture positivity rate of BAL in all children reported previously lied between 10% and 16%. Menon et al. suggested that BAL is better than GL for the diagnosis of pulmonary TB in children, whereas Somu et al. reported the opposite.
This retrospective study was undertaken to compare the diagnostic yield of *M. tuberculosis* from self-expectorated sputum to GL and BAL, in children with pulmonary TB.

**Patients and Methods**

This retrospective study included children and adolescents aged 6-18 years, who were referred to our hospital (Kaohsiung Chang Gung Memorial Hospital, Taiwan) for suspected pulmonary TB, and who underwent TB surveys between March 2005 and February 2014. All of the obtained specimens were investigated for *M. tuberculosis* by smear and culture. Only patients who were proven to be positive for *M. tuberculosis*, based on culture, smear or polymerase chain reaction (PCR) results, were enrolled in this study. The collection of data from patients’ medical records was approved by the Institutional Review Board of Kaohsiung Chang Gung Memorial Hospital.

**Gastric Lavage**

TGL was collected from patients on three consecutive mornings, before they ingested food or liquids. Nasogastric tubes were placed on patients with suspected TB, and gastric aspiration was performed with 50 ml of 0.9% saline flush. The lavaged specimens were sent for AFB staining and culture for *Mycobacterium tuberculosis*. Specimens containing a minimum 30-ml volume were sent for analysis. After digestion and decontamination of specimens, a concentration procedure was performed and the sediments were examined by Ziehl-Neelsen staining. Mycobacteria were cultured on Lowenstein-Jensen medium. In some cases, a PCR assay was performed on BAL specimens.

**Bronchoalveolar Lavage**

BAL was obtained by flexible fiberoptic bronchoscopy—the day before gastric lavage—and performed by an experienced bronchoscopist (HR Yu). The procedure was explained, and written informed consents were obtained from parents/guardians. All patients were assessed for complications during the procedure by pulse oximetry and clinical monitoring. Patients were sedated with midazolam (0.1-0.2 mg/kg), and supplementary oxygen was given during the procedure. Lignocaine jelly (2%) was applied to the nasal passage, the flexible fiberoptic bronchoscope (Olympus BF3 C20 or BFXP40 as indicated) was inserted transnasally, and 1 ml of 2% lignocaine was instilled on the larynx. The bronchoscope was advanced into the trachea and wedged into the most affected area (as seen on chest X-rays) or into a segment of the right middle lobe if the lesions were diffuse. After wedging, 1 ml/kg aliquots of sterile non-bacteriostatic 0.9% NaCl solution were instilled through the suction channel of the bronchoscope, and subsequently aspirated into a collecting tube. After the procedure, BAL samples were immediately sent for AFB staining, TB culture or PCR examinations.

**Sputum Expectoration**

Sputum induction was performed by experienced respiratory therapists. All patients with attempted sputum induction were given 3% saline nebulized for up to 15 min, using a Micromist nebulizer (Hudson RCI, Temecula, CA, USA). Sputum samples were collected in sterile containers and transported to the microbiology laboratory for processing.

**Statistical Analysis**

Statistical analysis included the chi-squared test for non-parametric data. All statistical tests were performed using SPSS 19.0 for Windows (SPSS, Inc., Chicago, IL). A p value < 0.05 was considered to have statistical significance.

**Results**

**Demographic and clinical characteristics**

Diagnosis of PCR-positive and/or culture-positive pulmonary TB was confirmed in 12 patients; of these, six were male and six were female. The median age was 14.4 years, (ranging from 9 to 18 years). All of these patients had received Bacillus Calmette-Guérin (BCG) vaccination in the first year of life. Contact histories were identified in four (33.3%) patients. Three patients were symptomatic at admission. The common symptoms included cough, fever, failure to thrive, and difficulty of breathing (16%). One patient had hemoptysis. On
chest radiography, 10 patients had segmental lesions in the form of collapse and/or consolidation; one of these patients had pleural effusion. Two patients, had paratracheal or hilar adenopathy radiographically.

**BAL culture had higher sensitivity for TB diagnosis than GL and SP**

Table 1 shows the number of procedures performed in the 12 patients, and the number of smear and/or culture positive results obtained with each method. Due to staff limitations, we were unable to include all techniques in each of the 12 patients. All patients tolerated bronchoscopy and BAL without complications. Eleven of the 12 diagnoses were confirmed (use strong verbs please!) by culture and one by PCR of BALF. Positive results were obtained in 14 samples from 11 patients, including SP (n = 6), BAL (n = 6), and GL (n = 2).

Among all cases confirmed by culture, the sensitivity was 75.0% for SP (six of eight samples), 85.7% for BAL (six of seven samples), and 40.0% for GL (two of five samples) (Table 2). We did not detect a difference in the recovery rates of *M. tuberculosis* by GL or BAL techniques (p = 0.222), (possibly due to the limited number of cases in the present study, this sentence should appear in the section of discussion).

Among the five patients (certainly, you can use cases, but seem to be inhumane) had sputum smear-positive TB, only one patient was positive for each of the three collected samples (Table 2). Three cases had only one smear-positive result, of three collected samples. GL culture for *M. tuberculosis* was positive for two of five patients (40%), gastric lavage culture were negative for five patients. We did not calculate the combined culture rate of BAL with GL or expectorated sputum, due to the limited number of cases.

**Table 1. Diagnostic tests list of the 12 pulmonary *Mycobacterium tuberculosis* infection patient**

<table>
<thead>
<tr>
<th>Case</th>
<th>Age</th>
<th>Sex</th>
<th>SP/S</th>
<th>SP/C</th>
<th>BAL/S</th>
<th>BAL/C</th>
<th>GL/S</th>
<th>GL/C</th>
<th>PCR</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>15</td>
<td>M</td>
<td>-</td>
<td>-</td>
<td>0/2</td>
<td>2/2</td>
<td>-</td>
<td>-</td>
<td>PCR</td>
</tr>
<tr>
<td>2</td>
<td>7</td>
<td>F</td>
<td>-</td>
<td>-</td>
<td>0/1</td>
<td>0/1</td>
<td>0/3</td>
<td>0/3</td>
<td>0/2(BAL)</td>
</tr>
<tr>
<td>3</td>
<td>9</td>
<td>M</td>
<td>0/3</td>
<td>0/3</td>
<td>1/1</td>
<td>1/1</td>
<td>0/3</td>
<td>0/3</td>
<td>1/1(BAL)</td>
</tr>
<tr>
<td>4</td>
<td>15</td>
<td>F</td>
<td>1/3</td>
<td>1/3</td>
<td>0/1</td>
<td>1/1</td>
<td>0/1</td>
<td>0/1</td>
<td>1/1(BAL) 0/1(GL)</td>
</tr>
<tr>
<td>5</td>
<td>15</td>
<td>M</td>
<td>1/3</td>
<td>1/3</td>
<td>0/1</td>
<td>1/1</td>
<td>-</td>
<td>-</td>
<td>0/1(BAL)</td>
</tr>
<tr>
<td>6</td>
<td>16</td>
<td>F</td>
<td>1/3</td>
<td>3/3</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>1/1(BAL)</td>
</tr>
<tr>
<td>7</td>
<td>14</td>
<td>M</td>
<td>0/3</td>
<td>0/3</td>
<td>0/1</td>
<td>1/1</td>
<td>-</td>
<td>-</td>
<td>1/1(SP)</td>
</tr>
<tr>
<td>8</td>
<td>15</td>
<td>F</td>
<td>-</td>
<td>-</td>
<td>0/1</td>
<td>1/1</td>
<td>0/3</td>
<td>3/3</td>
<td>0/1(BAL)</td>
</tr>
<tr>
<td>9</td>
<td>15</td>
<td>M</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0/3</td>
<td>1/3</td>
<td>1/1(BAL)</td>
</tr>
<tr>
<td>10</td>
<td>17</td>
<td>F</td>
<td>0/3</td>
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<td>-</td>
<td>-</td>
<td>-</td>
<td>1/1(GL)</td>
</tr>
<tr>
<td>11</td>
<td>18</td>
<td>F</td>
<td>2/3</td>
<td>3/3</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0/1(SP)</td>
</tr>
<tr>
<td>12</td>
<td>17</td>
<td>M</td>
<td>3/3</td>
<td>3/3</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

SP/S: sputum smear, SP/C: sputum culture, BAL/S: bronchoalveolar lavage smear, BAL/C: bronchoalveolar lavage culture, GL/S: gastric lavage smear, GL/C: gastric lavage culture

* the numerator means how many positive results in the total number (denominator)
DISCUSSION

Pulmonary TB remains a major public health problem, especially in developing countries. In Taiwan, incidence of TB was estimated to be 62 cases/100,000 population/y in 2008, and the control of TB remains a priority for public health welfare.\(^\text{13}\) The diagnosis of pulmonary TB are usually based on sputum AFB smear and TB culture. Several methods are available for obtaining samples in patients with suspected pulmonary TB. Flexible bronchoscopy with BAL and GL can be used for patients who cannot expectorate, or for patients whose sputum is negative for acid-fast bacilli (AFB). In the present study, we compared the diagnostic yields from sputum, BAL and GL. We found that BAL had the highest diagnostic yield. Whether culture results for GL or BAL are superior in children with pulmonary TB remains controversial. Menon et al. reported that, for AFB smears, GL alone have a positive result in three samples among patients (5.67%), while BAL alone was positive in eight samples (15.38%).\(^\text{12}\) These authors concluded that the diagnostic yield for AFB from BAL was better than from GL in children with probable pulmonary TB. However, the combination of GL and bronchoscopy may further improve AFB yield in children with pulmonary TB. Thus, while Singh et al. reported equal culture positive rates for GL and BAL in pulmonary TB in children,\(^\text{14}\) they also found that GL and BAL complemented each other when combined, doubling the diagnostic yield.\(^\text{14}\) Other reports, in contrast, showed that bronchoscopy does not add to the number of positive results obtained from mycobacterial cultures of GL specimens in children with pulmonary TB.\(^\text{10,11}\) These reports contradict our finding that bronchoscopy is superior to GL for culture confirmation of TB diagnosis. This difference probably results from different disease processes or sample number. In addition, the lack of standardization of GL protocols can produce inconsistent results.

Due to the low yield of GL, the procedures are generally performed on three consecutive mornings.\(^\text{10}\) Indeed, our results also concurrent that sputum collections on three consecutive mornings are necessary for accurate diagnosis. Among five cases of positive sputum smears in the present study, three had only one positive smear result from three collected samples. Our results also showed that the rate of positive GL smear was very low for \textit{M. tuberculosis}, indicating that GL smears are suboptimal for TB diagnosis. Although one previous study found the diagnostic yield of a single induced sputum sample was equivalent to a single GL sample,\(^\text{15}\) we were not able to compare the methods in this study because some of our patients could not expectorate sputum. Malekmohammad et al. reported that evaluation of post-bronchoscopy sputum smears is helpful for earlier diagnosis of pulmonary TB,\(^\text{16}\) because mucosal injury during bronchoscopy or irritation of the tracheobronchial tree resulting more efficient expectoration of sputum.\(^\text{16}\) Nonetheless, whether post-bronchoscopy sputum smears are useful for TB diagnosis in children needs further study.

There were several limitations of the present study. Firstly, the patients of this study were highly biased population in a referral center that treats probable TB patients. Secondly, the number of patients in this study is small, generalizability is greatly limited. Thirdly, no patient in this study received BAL, GL and SP at the same time.

In conclusion, flexible bronchoscopy with BAL is a useful tool for the isolation of \textit{M. tuberculosis} in children with pulmonary TB. In this study with only 12 patients, we found that the diagnostic yield

| Table 2. Sensitivity comparison of different diagnostic tests of the 12 pulmonary \textit{Mycobacterium tuberculosis} infection patients |
|-----------------|-------|-------|-------|-------|-------|-------|-------|
| SP/S | SP/C | BAL/S | BAL/C | GA/S | GA/C | PCR  |
| 5/8  | 6/8  | 1/7   | 6/7   | 0/5  | 2/5  | 6/10 |
| Sensitivity | 62.5% | 75.0% | 14.3% | 85.7% | 0%   | 40%  | 60%  |

*PCR positive: 1 in 2 of sputum. 4 in 7 of BAL, 1 in 2 of gastric lavage*
of *M. tuberculosis* from BAL is higher than GL in children with pulmonary TB, and when combined with expectorated sputum and bronchoscopic BAL, the diagnostic yield of *M. tuberculosis* in children can be increased. However, infection control in bronchoscopy suites should not be neglected. Moreover, BAL requires the availability of expensive bronchoscopy units and experienced bronchoscopists. For patients highly suspected of pulmonary TB, when the GL for AFB were negative, flexible bronchoscopy with BAL may increase the diagnostic yield in such instances.

REFERENCES