Efficacy of Ultrasound in the Diagnosis of Pleurodesis in Rabbits*

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Introduction: **The treatment of recurrent pleural effusion or recurrent pneumothorax frequently involves the creation of a pleurodesis. Ultrasound is being used more frequently to assess the presence of pleural fluid or pneumothorax. With ultrasound, the gliding sign displays the gliding of the visceral pleura over the parietal pleura during respiration. The absence of a gliding sign is indicative of a pneumothorax.**

Hypothesis: **We hypothesized that the presence of pleurodesis would be indicated by the absence of a gliding sign on ultrasound.**

Methods: **To create a pleurodesis, a single intrapleural injection of transforming growth factor-β_2 at a dosage of 1.70 μg/kg or doxycycline at a dosage of 10 mg/kg in a volume of 2.0 mL was administered randomly to one side of a New Zealand White rabbit. Prior to death on day 14 after intrapleural injection, all rabbits underwent an ultrasonic examination at three marked sites on each side of the chest. At each site, three ultrasonic features (gliding sign, pleural thickening, and pleural effusion) were evaluated and graded. The gliding sign was graded as follows: 0 = gliding sign definitely present, 1 = gliding sign questionable, 2 = gliding sign absent.**

Results: **In a preliminary study for developing skill in assessing the gliding sign, the correlation between the gliding sign and the pleurodesis score in 16 rabbits was highly significant ($r = 0.568$, $p = 0.02$). In the subsequent main study with 18 additional rabbits, the correlation between the gliding sign score and the pleurodesis score was even better ($r = 0.806$, $p = 0.00009$). The gliding sign was definitely present on the noninjected side in all rabbits**

Conclusions: **The presence of a pleurodesis is indicated by the absence of a pleural guiding sign on ultrasound.** ***(CHEST 2005; 128:934–939)***

Key words: gliding sign; pleural effusion; pleurodesis; pneumothorax; ultrasound

Abbreviation: TGF = transforming growth factor

Pleurodesis is the fusion of the visceral and parietal pleura. Pleurodesis usually results from pleural inflammation produced by an injury to the pleura. The treatment of recurrent pleural effusion or recurrent pneumothorax frequently involves an attempt to create a pleurodesis.[1] In clinical practice, a pleurodesis is most commonly created by injecting a sclerosing agent into the pleural space through a chest tube.[2]

The assessment of the presence of a pleurodesis is difficult. In most clinical articles on pleurodesis for pleural effusion, its presence is assessed by the presence or absence of pleural fluid. However, this assessment only indicates whether fluid has reaccumulated but does not indicate if a pleurodesis has occurred. Neither standard chest radiographs nor CT scans demonstrate whether pleurodesis has occurred.

Advances in technology have greatly improved the imaging capabilities of ultrasound. Ultrasound has been proved to be a reliable, efficient, and informative imaging modality for the evaluation of a wide variety of chest diseases.[3–7] Using ultrasound, the parietal pleura and the visceral pleura are normally seen as two thin, bright echogenic lines just beneath the chest wall. The two pleural lines normally glide

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over one another during respiratory movements in real-time ultrasonography. This movement is termed the *gliding sign* of the pleura. The absence of a gliding sign with ultrasonography has been used to diagnose pneumothorax.[7–12]

We hypothesized that the assessment of the pleural gliding sign with ultrasound would be an efficient imaging modality for the evaluation of pleurodesis, and the presence of a pleurodesis would be indicated by the absence of a gliding sign. The objective of this study was to investigate whether ultrasound can be used for the diagnosis of pleurodesis.

Materials and Methods

Pleurodesis Model

The protocol was approved by the animal care committee of Vanderbilt University, Nashville, TN. A chest tube was inserted into a pleural space of New Zealand White rabbit (1.5 to 2.0 kg) using a method previously described.[13,14] The right or left pleural spaces were selected randomly to receive the chest tube. The other pleural cavity was used as the control. There was a sham incision (< 2 cm) on the control side so that the ultrasonographer would not know which side received the pleurodesis agent. To create different grades of pleurodesis, a single intrapleural injection of either transforming growth factor (TGF)-β_2, 1.70 μg/kg (Genzyme; Framingham, MA) or doxycycline, 10 mg/kg in a volume of 2.0 mL (Fujisawa; Deerfield, IL) was administered in a random fashion to each rabbit via its chest tube.

After the intrapleural injection, the chest tube was aspirated at 24-h intervals for any pleural fluid. The chest tube was removed under light sedation when the pleural fluid drainage was < 5 mL over the preceding 24 h. Gentamicin (2 mg/kg; American Pharmaceutical Partners; Schaumburg, IL) was administered IM during the surgery and at 24-h intervals as long as the chest tubes were in place.

Thoracic Ultrasound

Prior to surgery, all rabbits underwent an ultrasonic examination at three marked sites (one anteriorly in the midclavicular line, one laterally in the mid axillary line, and one posteriorly in a line below the scapula) on each side of their chest in the seated position. At each site, three ultrasound features (gliding sign, pleural thickening, and pleural fluid) were evaluated and graded as shown in Table 1. The total score for each side is the sum of the three scores for each of the three sites. Other ultrasonic findings were noted. Each rabbit was again examined using ultrasound 14 days after the intrapleural injection. Thoracic ultrasound was performed by an experienced radiologist (E.D.) [ATL Ultramark 9; Philips; Bothell, WA] with a compact, linear 10–5 MHz probe. The ultrasonographer was blinded as to which side received the pleurodesis agent. Prior to ultrasonic examination, the fur over the entire thoracic area of each rabbit was removed.

Pleurodesis Scoring

Immediately after the final ultrasound examination, the animals were killed with carbon dioxide after sedation. After death, the thorax was removed *en bloc* and the lungs were expanded by the injection of 50 mL of 10% neutral-buffered formalin into the exposed trachea via a plastic catheter (6 mm in diameter) as previously described.[13,14] The trachea was then ligated and the entire thorax submerged into 10% neutral-buffered formalin solution for at least 48 h before the pleurodesis was graded. A consensus pleurodesis score was reached by two investigators (K.B.L. and R.W.L.) blinded to the treatment group and the scores on ultrasound. The degree of pleurodesis was semiquantified by the scheme outlined in Table 2, a modified version of the scoring system we have used in previous studies.[14] The only modification of this scoring system from the previously used eight-point system is that categories 1 through 4 in the previous system were all combined with the same score (1) since there is no pleural symphysis in any of the first four categories in the system used previously. Any evidence of hemothorax, pleural fluid, or pleural thickening was recorded if present.

Statistical Analysis

All data are presented as mean ± SD unless otherwise stated. Linear regression was used to analyze the correlation between the pleurodesis score (dependent variable) and the ultrasound scores (independent variable). All data were analyzed with statistical software (SigmaStat V3.0; SPSS; San Rafael, CA); *t* tests were used to compare the pleurodesis scores in the two different study groups, while the χ^2 test was used to compare the frequency the two different sides and the two different pleurodesing agents were used; $p < 0.05$ was considered significant.

Results

In the preliminary study, 16 rabbits were studied in order to develop skill in assessing the gliding sign. For these 16 rabbits, there was a significant correlation ($r = 0.568$, $p = 0.02$) between the pleurodesis score and the total gliding sign score (Table 3, Fig 1). At the time of the ultrasonic examination before the

Table 1—*The Grade of Ultrasonic Features*

Grade	Absence of Gliding Sign	Pleural Thickening	Pleural Effusion
0	Gliding sign definitely present	Normal pleura	No fluid
1	Gliding sign questionable	Pleural thickening questionable	Mininal; the echo-free space is seen within the costophrenic angle
2	Gliding sign absent	Pleural thickening definitely present	Small; the echo-free space is greater than the costophrenic angle but < 25% of hemithorax
3			Moderate; the echo-free space is 25 to 50% of hemithorax
4			Massive; the echo-free space is > 50% of hemithorax

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