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Distance Between Endotracheal Tube Cuff and Vocal Cords Measured by Ultrasound in Chinese Adults: A prospective case control study

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Distance Between Endotracheal Tube Cuff and Vocal Cords Measured by Ultrasound in Chinese Adults: A prospective case control study

Abstract

Background and objective Unrecognized malposition of the endotracheal tube can lead to severe complications in patients under general anesthesia. The purpose of this study was to estimate the occurrence of too shallow or too deep intubation using the traditional 23/21 rule and to verify the feasibility of using ultrasound to measure the distance between the upper edge of saline-inflated cuff and the vocal cords.

Design Prospective case control study.

Setting A tertiary hospital in Beijing, China.

Methods In this prospective study, 105 adult patients who required general anesthesia with an endotracheal tube (ETT) were enrolled. Prior to induction, ultrasound was used to identify the position of the vocal cords. After intubation, the ETT was fixed at a depth of 23 cm at the upper incisors in men and 21 cm in women. The depth of intubation was verified by video-assisted laryngoscopy, and the correct ETT position was defined as vocal cords lying between the two insertion depth marks. The distance between the upper edge of the saline-inflated cuff and the vocal cords was measured by ultrasound; the ideal distance was considered to be 1.9 cm to 4.1 cm. The tip-to-carina distance was measured using a fiberoptic bronchoscope. The primary outcome was determination of the sensitivity, specificity, positive predictive value, and negative predictive value of ultrasonography in determining correct ETT insertion depth.

Results Among the 105 cases, two cuffs were too close to the vocal cords and one too far away from the vocal cords. These diagnoses were made by ultrasound and were in agreement with results from direct laryngoscopy. There was no endobronchial intubation, but in 8.6% of the patients the distance between the tip of the ETT and the carina was less than 2 cm. The overall accuracy of ultrasound in identifying malposition of the cuff was 100.0% (95% CI: 96.6%–100%). The sensitivity, specificity, positive predictive value, and negative predictive value of ultrasound were, respectively, 100% (95% CI: 96.5%–100%), 100% (95% CI: 29.2%-100%), 100% (95% CI: 96.5%–100%) and 100% (95% CI: 29.2%-100%).

Conclusion The distance from the cuff to the vocal cords may be too short or too long for some patients when the traditional 23/21 rule is used. Identification of the upper edge of the saline-inflated cuff and the vocal cords by ultrasound to assess the location of the ETT is a reliable method. It can be used to avoid malposition of the ETT cuff and reduce the incidence of vocal cords injury after intubation.

Article summary

Strengths and limitations of this study

- Unrecognized malposition of the endotracheal tube can lead to severe complications in patients under general anesthesia.
- The distance from the cuff to the vocal cords may be too short or too long for some patients when the traditional 23/21 intubation rule is used.
- Identification of the upper edge of the saline-inflated cuff and the vocal cords by ultrasound to assess the location of the ETT is a reliable method.
- It can be used to avoid malposition of the ETT cuff and reduce the incidence of vocal cords injury after intubation.
- Since the number of incorrect tube positions in our study was small, a larger sample size study may be needed to verify our results.

Introduction

Tracheal intubation is a routine procedure of resuscitation and general anesthesia. The appropriate depth of endotracheal intubation should be confirmed after intubation and during surgery because a malposition of endotracheal tube (ETT) can lead to serious complications. Placing the tube too deeply may stimulate the carina, and unrecognized endobronchial intubation may result in single-lung ventilation, hypoxemia, and atelectasis of the nonventilated lung.[1] On the other hand, if the ETT is placed too shallowly, the tube cuff's impingement on the vocal cords may lead to vocal cords injury, compression of the recurrent laryngeal nerve, and even accidental extubation.[2]

An optimal ETT placement should ensure sufficient distance (2–5 cm) between the tip of the ETT and the carina[3] and sufficient distance (1.5–2.5 cm) between the proximal margin of the cuff to the vocal cords.[2 4] Most ETTs for the adults have two black insertion guide marks at 2 and 4 cm above the cuff or one mark at 2 to 3 cm above the cuff. [5] Alignment of the marks with the vocal cords helps to place the ETT at the correct depth. [6] However, this technique relies on visualization of the vocal cords with a laryngoscope (i.e., grade I or II view). A large tongue, prominent teeth, a short neck, or blood and secretions may make visualization of the ETT's position within the glottis difficult. Besides, under some circumstances, such as using blind intubation or a Shikani or Bonfil optical stylet to guide intubation, the depth markers cannot be observed.

Under such circumstances, intubation depth is commonly determined according to experience, usually 23 cm for male and 21 cm for female patients.[7-9] After intubation, auscultation of breath sounds is routinely performed to confirm the location of the tube. If endobronchial intubation is suspected, the tube must be withdrawn until bilateral breath sound can be heard. However, auscultation of breath sounds can be unreliable,[10] and blind withdrawal of the tube can be hazardous for patients with short tracheas. In some patients the distance from the vocal cords to the carina is identical to or even shorter than the distance from the ETT tip to the insertion guide mark. [6] This means that there would not be enough cuff free zone above the vocal cords when breath sounds can be heard from both sides in patients with short tracheas.

Cuff palpation has been suggested as a tactile method to estimate the position of the EET, [11 12] but its accuracy is influenced by the thickness of the soft tissue of the anterior neck and the experience of the operator.[13] Furthermore, a high-volume low-pressure cuff may not be palpable despite correct placement. One study has shown that cuff palpation has only a 26% specificity for indicating incorrect ETT placement. [14] The development of ultrasonography has made it possible to identify the ETT cuff more accurately. In 1987, Raphael et al., for the first time, obtained a clear image of a saline-inflated cuff by ultrasound.[15] Previous studies used ultrasonic images of saline-inflated cuffs at or above the suprasternal notch as indicators of appropriate ETT insertion depth.[16] However, as in the case of auscultation, observing the cuff at or above the suprasternal notch can rule out too deep intubation but not too shallow intubation.

Since both the vocal cords and the ETT cuff can be visualized using ultrasound, a safe distance between the vocal cords and the ETT cuff can potentially be guaranteed by ultrasonography. We hypothesized that ultrasound can be used to estimate the distance between the upper edge of the ETT cuff and the vocal cords in adults so that the depth of the ETT can be adjusted accordingly. The purpose of this study was to review instances of endobronchial intubation and estimate the occurrence of too short a distance between the cuff and vocal cords in adult Chinese using the 23/21 rule and also to determine the feasibility of using ultrasound to measure the distance between the upper edge of saline-inflated cuff and the vocal cords.

Materials and methods

Patient and public involvement

Patients were not involved in the development of the design, recruitment or conduct of the study. The results will not be disseminated to study participants.

Following approval from the institutional review board (Peking University Third

Hospital Ethics Committee, IRB00006761-2016163), patients aged 18 to 70 who were scheduled for elective cervical surgery under general anesthesia were recruited in this prospective case control study. Written informed consent was obtained from all patients. The exclusion criteria included difficult airway (Mallampati classification 3 and 4, mouth opening <3 cm), abnormal airway or chest anatomy, and a history of cervical trauma or cervical surgery.

Equipment and researchers

A reinforced ETT with a 7.0- or 8.0-mm inner diameter (ID) (Covidien Mallinckrodt, USA), with two insertion guide marks was used in the present study, which involved four investigators: an anesthesiologist experienced in airway ultrasonography using a 38-mm 6-13 MHz linear ultrasound probe (Turbo SonoSite HFL, Bothell, WA), an anesthesiologist experienced in fiberoptic bronchoscopy (FOB), a senior resident, and an anesthetic assistant. The two anesthesiologists who performed the ultrasound and FOB examinations were blind to the results of laryngoscopy.

Ultrasound assessment of vocal cords

When the patient reached the operation room, routine monitors (pulse oximeter, noninvasive blood pressure cuff, and electrocardiogram) were placed. The patient was in a neutral position. The ultrasound probe was placed transversely on the neck perpendicular to the skin. The probe was moved cranially or caudally until the true vocal cords could be identified (Figure 1). Along the midpoints of the short axis of the probe, line A was drawn on the patient's skin (Figure 2) to mark the position of true vocal cords. If the vocal cords could not be clearly identified, the patient was excluded.

Intubation

Following the induction of anesthesia, an ETT (7.0-mm ID for females, 8.0-mm ID for males) was inserted by the resident and the insertion depth was determined using the 23/21 cm rule (23 cm at the upper incisor teeth in men and 21 cm in women) using a video-assisted laryngoscope (Zhejiang UE Medical Corporation, Tai Zhou, China). The ETT was held in place by the assistant, then the position of the vocal cords relative to the depth markers was verified under video laryngoscopy. If the vocal cords could not be seen, the patient would be excluded. Once the vocal cords were seen, the ETT was connected to the ventilator for mechanical ventilation and anesthesia was maintained with propofol 6 to 8 mg/kg per hour.

Ultrasound assessment of ETT cuff

The ultrasound probe was placed sagittal on the neck, perpendicular to the skin, and above the suprasternal notch. The ETT cuff was then inflated with 8 mL of saline. A pressure gauge was used to measure the cuff pressure; if the pressure exceeded 30 cmH_2O, the patient would be excluded. After the injection of saline, two parallel hyperechogentic lines would appear on ultrasound screen, as shown in Figure 3. The upper line represented the anterior wall of cuff, and the lower line represented the anterior wall of ETT. The junction of these two lines or the proximal starting point of the upper line represented the proximal margin of the cuff. Then the probe was moved along the midline of the neck until an image of the proximal margin of the cuff appeared at the center of the screen. Line B (Figure 2) was then drawn on the skin along the midpoints of the long axis of the probe. The distance between lines A and B (AB), representing the distance between the vocal cords and the proximal edge of the cuff (VCD), was measured. Then a towel was placed to cover the neck. With the ventilator disconnected, the tip-carina distance and the incisors-carina distances were measured [17] by the an anesthesiologist, who was blind to the marks on patient's neck, using a 2.8-mm FOB (TIC-SD-I, Zhejiang UE Medical Corporation, Tai Zhou China). For patients with suitable VCDs (1.9–4.1 cm), the saline was drawn out of the cuff and the cuff filled with the proper amount of air. Finally, the tube was secured with tape. For patients with unsuitable VCDs (<1.9 cm or >4.1 cm), the saline was drawn out of the cuff and the tube moved cephalad or caudally based on the calculated distance to get the desired VCD. Then the video-assisted laryngoscope was again used to confirm the relative position of the glottis and the two insertion guide marks.

Statistical analysis

The primary outcome was the accuracy of the ultrasound image confirming the proper depth of the ETT. The distance between the proximal margin of the cuff and the first and second insertion guide marks of the ETT was 2 ± 0.1 cm and 4 ± 0.1 cm respectively. The depth by ultrasound was defined as correct (AB distance between 1.9 and 4.1 cm), too shallow (AB distance <1.9cm), or too deep (AB distance >4.1 cm). The depth was also defined by video-assisted laryngoscopy as correct (vocal cords lying between the two insertion guide marks), too shallow (both marks above vocal cords), or too deep (both marks below vocal cords). Both shallow and deep placements were considered to be incorrect.

According to the results of preliminary tests, an accuracy of 90% was considered acceptable; to obtain an α error of 0.05 and a statistical power of 0.8, the calculated sample size was 102 patients using PASS11.0 (NCSS LLC, Utah) software. A total of 120 patients were recruited to provide for potential dropouts. SPSS version 20.0 (SPSS, Inc., Chicago, IL) was used for data management. The normality of data was assessed using the Shapiro-Wilk test. Normally distributed variables were expressed as the mean ± SD and compared between genders using Student's *t*-test. Non-normally distributed variables were expressed as the median (range) and analyzed using the Mann-Whitney U test. Ultrasound was compared with the gold standard of HC video-assisted laryngoscopy as a diagnostic test; accuracy, sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were calculated. The corresponding 95% confidence intervals (CIs) were calculated based on the Clopper-Pearson method in SAS 9.4 (SAS Inc., NC). Agreement between ultrasound and video-assisted laryngoscopy was evaluated by the kappa consistency test. P <.05 was considered to be statistically significant.

Results

A total of 120 patients were initially enrolled in the study and 105 were included in the final analysis. Figure 4 presents the allocation process. The demographic data of the 105 patients who completed the trial are presented in Table 1. The differences in height, weight, and thyromental distance between males and females were statistically significant (*P* <.05). There was no significant difference in VCD detected by ultrasound and the tip-carina distance detected by FOB between males and females (Table 1).

Table 1 Patient Demographics

	Total (n=105)	Female (n=62)	Male (n=43)	P (Male vs Female)
Age（yr）	53（20 , 69）	53 (20, 69)	54 (20, 65)	0.757
Weight（kg）	64.0 ± 9.8	60. 6± 8.9	68.9 ± 8.7	<0.001
Height（cm）	164.8 ± 8.5	160.0 ± 6.6	171.7 ± 5.9	<0.001
BMI	23.0（17.0 , 33.7）	22.8 (18.8, 33.7)	23.1 (17.0, 30.9)	0.669
VCD（cm）	2.9±0.6	2.9±0.6	3.0±0.6	0.607
TCD (cm)	3.6 (1.3, 6.4)	3.4 (1.3, 6.4)	3.6 (1.9, 6.4)	0.124

Variables are presented as mean ± SD (range) or median (range), CVD: vocal cords -cuff –distance, TCD: tip- carina distance, P- comparison between male and female

The diagnoses of too shallow an intubation for one female and one male and too deep an intubation for one female made by ultrasound was in agreement with the diagnoses made by direct laryngoscopy. After inserting the tube forward for about 1.5 cm, the glottis in each of the two shallow intubation patients lay between the two depth makers as confirmed by video-assisted laryngoscopy. After pulling the tube up about 1cm, the glottis position of the deep intubation patient was also corrected. The distances from the ETT tip to the carina measured by FOB were between 1.4 to 6.4 cm with no statistical difference between males and females; this distance was less than 2 cm for 1 male and 8 female patients. The data on these patients are shown in Table 2.

Table 2. Data of the three patients with incorrect insertion depth

Patient	CVD (cm)	Sex	Age (y)	Height（cm）	Weight（kg）	BMI (kg/m²)	TCD（cm）
1	1.1	M	20	188	80	22.6	6.4
2	1.4	F	61	162	60	22.8	3.3
3	4.4	F	61	152	52	22.5	1.3

CVD: Cuff – vocal cords distance, TCD: Tip- carina distance,

Using video-assisted laryngoscopy as the standard criterion (Table 3), the overall accuracy of ultrasound was 100.0% (95% confidence interval [CI]:96.6–100%), with a sensitivity of 100% (95% CI: 96.5%–100%), a specificity of 100% (95% CI: 29.2%–100%), PPV 100% (95% CI: 96.5%–100%), and NPV 100% (95% CI: 29.2%–100%), and 100.0% (95% confidence interval [CI]:96.6–100%) in detecting the correct position of the ETT. Overall, the correct identification of tracheal versus bronchial intubation was 100.0% (95% confidence interval [CI]:96.6–100%). The kappa value was 1.

Table 3. Results of Ultrasound vs. Video assisted laryngoscope

	Video assisted laryngoscope	
Ultrasound	Positive	Negative
Correct	102	0
Incorrect	0	3

Video assisted laryngoscope: identifying the relationship of vocal cords and depth markers.

Positive: vocal cords lied between two depth markers; otherwise it is negative. Ultrasound: identifying the distance between upper edge of cuff and the glottis. Proper: the distance between upper edge of cuff and glottis was in the range of 1.9 to 4.1cm; otherwise it is improper.

Discussion

Among the 105 cases using the 23/21 rule, two cuffs were too close to the vocal cords and one too far away from the vocal cords. The diagnoses of too deep intubation and too shallow intubation made by ultrasound were in agreement with direct laryngoscopy. The distances measured by ultrasound had high accuracy, sensitivity, specificity, PPV, and NPV for identifying the position of ETT at the level of the glottis. It was also found that malposition of the ETT could be corrected based on the results of ultrasound.

Unlike previous studies, which were mainly focused on the avoidance of too deep an intubation, our study was concerned with the safety of the vocal cords as a priority. We believe that for most adults, instances of ETT cuff impingement on the vocal cords or compression of the recurrent laryngeal nerve are more harmful—and more readily ignored—than endobronchial intubation.

In an early study, Cavo et al.[2] pointed out that the rigid thyroid lamina was lateral to the anterior branch of the recurrent laryngeal nerve; they suspected that the vulnerable point of the recurrent laryngeal nerve lay between 6 and 10 mm below the posterior end of the free edge of the vocal cord. Several studies have shown that extension of the neck and concomitant movement of the ETT was a risk factor for vocal cord injury owing to neck extension during withdrawal of the ETT from the trachea (1.9 cm in Conrardy's study,[3] 1.7 ± 0.8 cm in Kim's study,[18] and 0.9 ± 0.9 cm in Hartrey's study[5]). Under external force, the withdrawal distance of the ETT is increased.[19] Since the cuff-free area of 2 to 4 cm above the vocal cords can be guaranteed by observing the insertion guide marks, this criterion was adopted as the standard for correct ETT positioning in the present study.

Our study showed that in using 23/21 rule, the rate of correct ETT positioning was as high as 97.1%. In only one male patient, by measuring the distance of VCD and observing the intubation guide marks, the tube insertion was considered to be too deep. The endotracheal tube's tip-to-carina distance in this case was 1.3 cm, which was the shortest found in any patient. Apart from this patient, there were 8 patients who had an ETT-to-carina distance of less than 2 cm.

The VCD of one male and one female patient was less than 1.9 cm (1.1 cm and 1.4 cm, respectively). An anesthesiologist has suggested that it would be safer to use a 22/20 rule for Asian people to avoid endobronchial intubation.[9 20] However, if the 22/20 rule had been used in our study, the number of patients with a VCD less than 1.9 cm would have increased to 12, with a greater risk of harmful impingement of the cuff on the vocal cords.

In our study, the VCD was measured with ultrasound. Although some studies have used FOB to make sure that the ETT cuff was 2 to 3 cm below the vocal cords,[21 22] we found it difficult to identify the cuff and the vocal cords via FOB when the ETT was already in place. We used direct video-assisted laryngoscopy to verify the position of the vocal cords with respect to the ETT cuff. Although a quantitative measurement could not be achieved with laryngoscopy, agreement on correct position of ETT using two these two methods can still be justified.

With the increasing popularity of ultrasound use in the field of anesthesia, several studies have used ultrasound to help confirm the depth of intubation. McKay et al.[23] observed an ultrasound image of tracheal ring undulation caused as the ETT was advanced and considered it as an indication of proper ETT depth. Actually, such an image can only be regarded as a sign of endotracheal intubation. Uya et al.[24] defined adequate ETT placement as complete visualization of the saline-injected ETT cuff at the suprasternal notch. Similarly, Tessaroa et al.[16] considered that ultrasonography of the saline-inflated cuff at the suprasternal notch represented correct ETT insertion depth. Some concern has been raised about how far above the suprasternal notch the cuff should be located.[25-27]

Our study was the first to use ultrasound to measure VCD. In awake patients, in the ultrasound transverse view, the hyperechoic vocal ligaments can easily be visualized. During phonation, the true cords move toward the midline. Surface projection of the vocal cords can then be marked on the skin after obtaining the optimal image of the true vocal cords. Surface projection of the proximal edge of the cuff can also be marked on the skin, as in the present study. Our results show that this is a reliable method for identifying incorrect intubation depth. Also, adjustment of insertion depth can be made based on the VCD obtained with this method. One can argue that it is needless to measure VCD in every patient intubated with the 23/21 rule. However, for patients whose necks are in extension during surgery, pre-surgery evaluation of the relative position of the vocal cords and cuff may be necessary. In the present study, although patients with Mallampati classification 3 and 4 and mouth opening less than 3 fingers were excluded, vocal cords could not be completely exposed using video-assisted laryngoscope on 11 of 120 patients. Measuring the VCD as shown in the present study may serve as an alternative under such circumstances.

Limitations of the study

There are several limitations to our study. Firstly, the number of incorrect tube positions in our study was small. A larger sample size study may be needed to verify our results. Secondly, for some male patients with prominent Adam's apples, the probe could not be fully attached to the skin and the vocal cords could not be completely seen. Three male patients were excluded from the study due to unsatisfactory ultrasound images of the glottis. Putting a water-filled pad between the probe and the skin may increase the contact area and enhance ultrasound imaging. Thirdly, although too shallow an intubation can be avoided using the present method, too deep intubation cannot. Using lung ultrasound technique, endobronchial intubation can be determined, and bronchial intubation avoided.[21] For patients with extremely short airways, the proper choice is to select an ETT with a short distance from the catheter tip to the upper edge of the cuff. Lastly, the ultrasound examination was done by experienced anesthesiologists. More training is needed for inexperienced anesthesiologists, although a study has shown that this technique can be easily learned.[24]

Conclusion

Ultrasonography of the upper edge of a saline-inflated ETT cuff appears to be a simple and reliable method for identifying the distance between the vocal cords and the proximal edge of the ETT cuff, and malposition of the ETT can be corrected according to the results of the ultrasound. Confirmation of a safe distance from the upper margin of the cuff to the vocal cords may prevent injury to the vocal cords and recurrent laryngeal nerve.

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Footnotes

Contributors ML designed the study and revised the final manuscript as submitted. XC and WZ contributed equally to this paper, and they were both the first authors. XC, WZ, ZY and JG carried out all data collection. XC and WZ carried out data analyses and drafted the initial manuscript. All authors approved the final manuscript as submitted.

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Data sharing statement: A full anonymized dataset is available from the corresponding author on request.

Figure legend

Figure 1. Transverse scan over the glottis. asterisks, thyroid cartilage; V: vocal cords.

Figure 2. Demonstration of probe placement. Line A: probe placed at the level of vocal cord, Line B: probe placed at the level of proximal edge of tube cuff

Figure 3. Longitudinal view of the intubated trachea, with the cuff of the endotracheal tube inflated with saline. Arrows pointing downwards - anterior wall of the cuff. Arrows pointing upwards - anterior wall of the ETT.

Figure 4. Allocation process

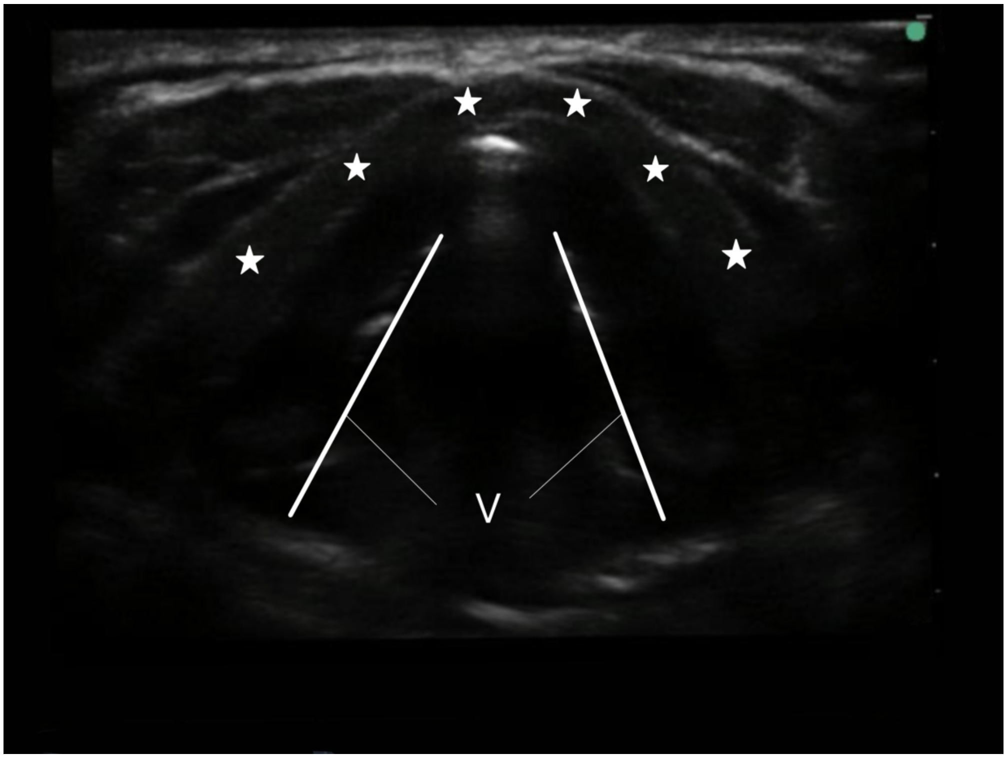

Figure 1. Transverse scan over the glottis. asterisks, thyroid cartilage; V: vocal cords.

889x666mm (72 x 72 DPI)

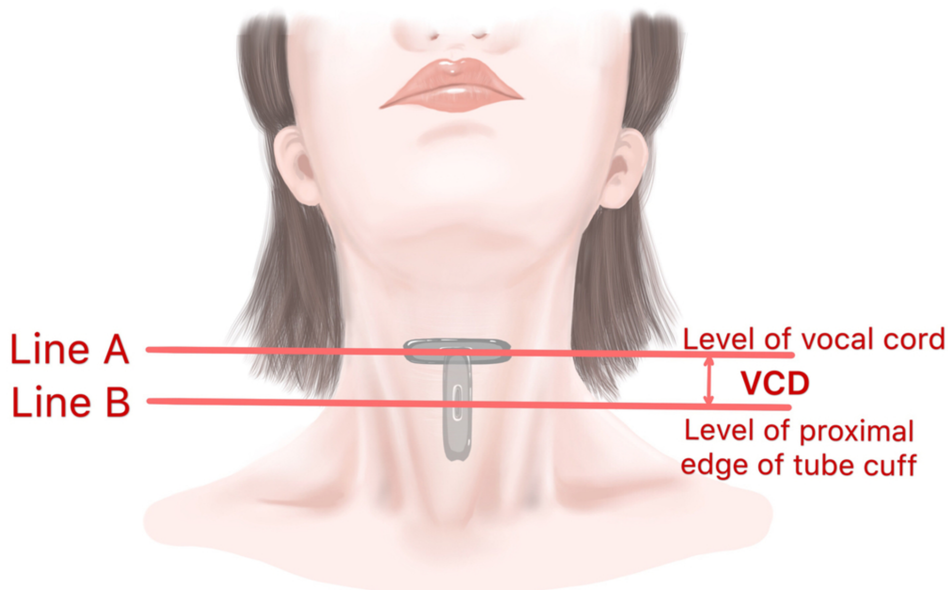

Figure 2. Demonstration of probe placement. Line A: probe placed at the level of vocal cord, Line B: probe placed at the level of proximal edge of tube cuff

90x90mm (300 x 300 DPI)

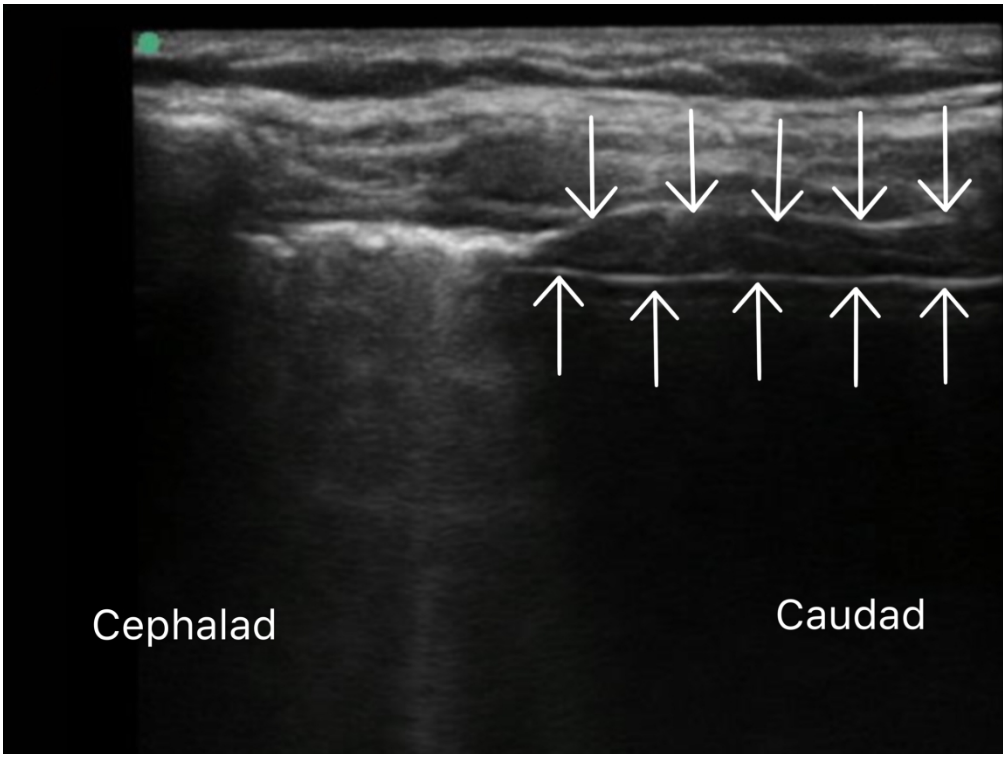

Figure 3. Longitudinal view of the intubated trachea, with the cuff of the endotracheal tube inflated with saline. Arrows pointing downwards - anterior wall of the cuff. Arrows pointing upwards - anterior wall of the ETT.

889x666mm (72 x 72 DPI)

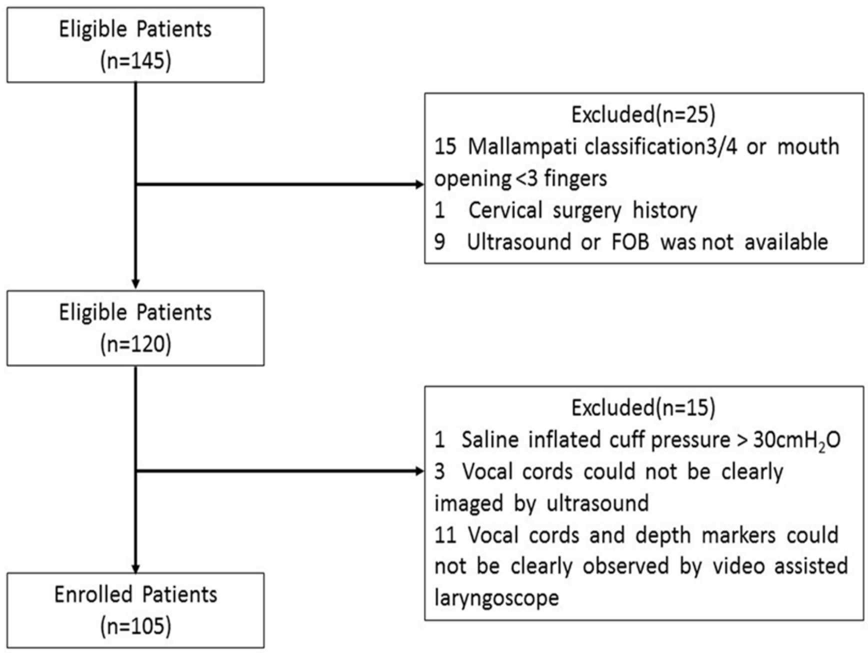

Figure 4. Allocation process

99x99mm (300 x 300 DPI)

Determining correct tracheal tube insertion depth by measuring Distance Between Endotracheal Tube Cuff and Vocal Cords by Ultrasound in Chinese Adults: A prospective case control study

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Determining correct tracheal tube insertion depth by measuring Distance Between Endotracheal Tube Cuff and Vocal Cords by Ultrasound in Chinese Adults: A prospective case control study

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Determining correct tracheal tube insertion depth by measuring Distance Between Endotracheal Tube Cuff and Vocal Cords by Ultrasound in Chinese Adult s: A prospective case control study

Abstract

Objectives Unrecognized malposition of the endotracheal tube can lead to severe complications in patients under general anaesthesia. The purpose of this study was to verify the feasibility of using ultrasound to measure the distance between the upper edge of saline-inflated cuff and the vocal cords.

Design Prospective case control study.

Setting A tertiary hospital in Beijing, China.

Methods In this prospective study, 105 adult patients who required general anaesthesia were enrolled. Prior to induction, ultrasound was used to identify the position of the vocal cords. After intubation, the endotracheal tube (ETT) was fixed at a depth of 23 cm at the upper incisors in men and 21 cm in women. The depth of intubation was verified by video-assisted laryngoscopy. The distance between the upper edge of the saline-inflated cuff and the vocal cords was measured by ultrasound; the ideal distance was considered to be 1.9 cm to 4.1 cm.

Results Among the 105 cases, two cuffs were too close to the vocal cords and one too far away from the vocal cords. These diagnoses were made by ultrasound and were in agreement with results from direct laryngoscopy. The overall accuracy of ultrasound in identifying malposition of the cuff was 100.0% (95% CI: 96.6%–100%). The sensitivity, specificity, positive predictive value, and negative predictive value of ultrasound were, respectively, 100% (95% CI: 96.5%–100%), 100% (95% CI: 29.2%-100%), 100% (95% CI: 96.5%–100%) and 100% (95% CI: 29.2%-100%).

Conclusion Identification of the upper edge of the saline-inflated cuff and the vocal cords by ultrasound to assess the location of the ETT is a reliable method. It can be used to avoid malposition of the ETT cuff and reduce the incidence of vocal cords injury after intubation.

Article summary

Strengths and limitations of this study

- Previous studies on intubation depth were mainly focused on how to avoid too deep intubation. The hazard of too shallow intubation was often neglected.
- We found that the distance between the vocal cord and the upper edge of ETT cuff can be measured with the aid of ultrasonography. This method might be useful to avoid malposition of the ETT cuff and reduce the incidence of vocal cords injury after intubation.
- The technical limitations include the difficulty in fitting the ultrasound prove to the surface of prominent Adam's apple and 5 to 8 min to complete the examination procedure.

Introduction

Tracheal intubation is a routine procedure of resuscitation and general anaesthesia. The appropriate depth of endoracheal intubation should be confirmed after intubation and during surgery because a malposition of endotracheal tube (ETT) can lead to serious complications. Placing the tube too deeply may stimulate the carina, and unrecognized endobronchial intubation may result in single-lung ventilation, hypoxemia, and atelectasis of the nonventilated lung.[1] On the other hand, if the ETT is placed too shallowly, the tube cuff's impingement on the vocal cords may lead to vocal cords injury, compression of the recurrent laryngeal nerve, and even accidental extubation.[2]

An optimal ETT placement should ensure sufficient distance (2–5 cm) between the tip of the ETT and the carina[3] and sufficient distance (1.5–2.5 cm) between the proximal margin of the cuff to the vocal cords.[2, 4] Most ETTs for the adults have two black insertion guide marks at 2 and 4 cm above the cuff or one mark at 2 to 3 cm above the cuff. [5] Alignment of the marks with the vocal cords helps to place the ETT at the correct depth. [6] However, this technique relies on visualization of the vocal cords with a laryngoscope (i.e., grade I or II view). A large tongue, prominent teeth, a short neck, or blood and secretions may make visualization of the ETT's position within the glottis difficult. Besides, under some circumstances, such as using blind intubation or a Shikani or Bonfil optical stylet to guide intubation, the depth markers cannot be observed.

Under such circumstances, intubation depth is commonly determined according to experience, usually 23 cm for male and 21 cm for female patients.[7-9] After intubation, auscultation of breath sounds is routinely performed to confirm the location of the tube. If endobronchial intubation is suspected, the tube must be withdrawn until bilateral breath sound can be heard. However, auscultation of breath sounds can be unreliable,[10] and blind withdrawal of the tube can be hazardous for patients with short tracheas. In some patients the distance from the vocal cords to the carina is identical to or even shorter than the distance from the ETT tip to the insertion guide mark. [6] This means that there would not be enough cuff free zone below the vocal cords when breath sounds can be heard from both sides in patients with short tracheas.

Cuff palpation has been suggested as a tactile method to estimate the position of the EET, [11 12] but its accuracy is influenced by the thickness of the soft tissue of the anterior neck and the experience of the operator.[13] Furthermore, a high-volume low-pressure cuff may not be palpable despite correct placement. One study has shown that cuff palpation has only a 26% specificity for indicating incorrect ETT placement. [14] The development of ultrasonography has made it possible to identify the ETT cuff more accurately. In 1987, Raphael et al., for the first time, obtained a clear image of a saline-inflated cuff by ultrasound.[15] Previous studies used ultrasonic images of saline-inflated cuffs at or above the suprasternal notch as indicators of appropriate ETT insertion depth.[16] However, as in the case of auscultation, observing the cuff at or above the suprasternal notch can rule out too deep intubation but not too shallow intubation.

Since both the vocal cords and the ETT cuff can be visualized using ultrasound, a safe distance between the vocal cords and the ETT cuff can potentially be guaranteed by ultrasonography. We hypothesized that ultrasound can be used to estimate the distance between the upper edge of the ETT cuff and the vocal cords in adults so that the depth of the ETT can be adjusted accordingly. The purpose of this study was to review instances of endobronchial intubation and estimate the occurrence of too short a distance between the cuff and vocal cords in adult Chinese using the 23/21 rule and also to determine the feasibility of using ultrasound to measure the distance between the upper edge of saline-inflated cuff and the vocal cords.

Materials and methods

Study participants

Following approval from the institutional review board (Peking University Third Hospital Ethics Committee, IRB00006761-2016163), patients aged 18 to 70 who were scheduled for elective cervical surgery under general anaesthesia from 2016.10 to 2017.5 were recruited in this prospective case control study. Written informed consent was obtained from all patients. The exclusion criteria included difficult airway (Mallampati classification 3 and 4, mouth opening <3 cm), abnormal airway or chest anatomy, and a history of cervical trauma or cervical surgery.

Equipment and researchers

A reinforced ETT with a 7.0- or 8.0-mm inner diameter (ID) (Covidien Mallinckrodt, USA), with two insertion guide marks was used in the present study, which involved four investigators: an anesthesiologist experienced in airway ultrasonography using a 38-mm 6-13 MHz linear ultrasound probe (Turbo SonoSite HFL, Bothell, WA), an anesthesiologist experienced in fiberoptic bronchoscopy (FOB), a senior resident, and an anesthetic assistant. The two anesthesiologists who performed the ultrasound and FOB examinations were blind to the results of laryngoscopy.

Ultrasound assessment of vocal cords

When the patient reached the operation room, routine monitors (pulse oximeter, noninvasive blood pressure cuff, and electrocardiogram) were placed. The patient was in a neutral position. The ultrasound probe was placed transversely on the neck perpendicular to the skin. The probe was moved cranially or caudally until the true vocal cords could be identified (Figure 1). Along the midpoints of the short axis of the probe, line A was drawn on the patient's skin (Figure 2) to mark the position of true vocal cords. If the vocal cords could not be clearly identified, the patient was excluded.

Intubation

Following the induction of anaesthesia, an ETT (7.0-mm ID for females, 8.0-mm ID for males) was inserted by the resident and the insertion depth was determined using the 23/21 cm rule (23 cm at the upper incisor teeth in men and 21 cm in women) using a video-assisted laryngoscope (Zhejiang UE Medical Corporation, Tai Zhou, China). The ETT was held in place by the assistant, then the position of the vocal cords relative to the depth markers was verified under video laryngoscopy. If the vocal cords could not be seen, the patient would be excluded. Once the vocal cords were seen, the ETT was connected to the ventilator for mechanical ventilation and anaesthesia was maintained with propofol 6 to 8 mg/kg per hour.

Measurements

The ultrasound probe was placed sagittal on the neck, perpendicular to the skin, and above the suprasternal notch. The ETT cuff was then inflated with 8 mL of saline. A pressure gauge was used to measure the cuff pressure; if the pressure exceeded 30 cmH_2O, the patient would be excluded. After the injection of saline, two parallel hyperechogentic lines would appear on ultrasound screen, as shown in Figure 3. The upper line represented the anterior wall of cuff, and the lower line represented the anterior wall of ETT. The junction of these two lines or the proximal starting point of the upper line represented the proximal margin of the cuff. Then the probe was moved along the midline of the neck until an image of the proximal margin of the cuff appeared at the center of the screen. Line B (Figure 2) was then drawn on the skin along the midpoints of the long axis of the probe. The distance between lines A and B (AB), representing the distance between the vocal cords and the proximal edge of the cuff (VCD), was measured. Then a towel was placed to cover the neck. With the ventilator disconnected, the tip-carina distance and the incisors-carina distances were measured [17] by the an anesthesiologist, who was blind to the marks on patient's neck, using a 2.8-mm FOB (TIC-SD-I, Zhejiang UE Medical Corporation, Tai Zhou China). For patients with suitable VCDs (1.9–4.1 cm), the saline was drawn out of the cuff and the cuff filled with the proper amount of air. Finally, the tube was secured with tape. For patients with unsuitable VCDs (<1.9 cm or >4.1 cm), the saline was drawn out of the cuff and the tube moved cephalad or caudally based on the calculated distance to get the desired VCD. Then the video-assisted laryngoscope was again used to confirm the relative position of the glottis and the two insertion guide marks.

Patient and public involvement

No patients were involved in setting the research question or the outcome measures, nor were they involved in developing plans for design or implementation of the study. No patients were asked to advise on interpretation or writing up of results. Study reports will be disseminated to investigators and patients through this open-access publication.

Statistical analysis

The primary outcome was the accuracy of the ultrasound image confirming the proper depth of the ETT. The distance between the proximal margin of the cuff and the first and second insertion guide marks of the ETT was 2 ± 0.1 cm and 4 ± 0.1 cm respectively. The depth by ultrasound was defined as correct (AB distance between 1.9 and 4.1 cm), too shallow (AB distance <1.9cm), or too deep (AB distance >4.1 cm). The depth was also defined by video-assisted laryngoscopy as correct (vocal cords lying between the two insertion guide marks), too shallow (both marks above vocal cords), or too deep (both marks below vocal cords). Both too shallow and too deep placements were considered to be incorrect.

According to the results of preliminary tests, an accuracy of 90% was considered acceptable; to obtain an α error of 0.05 and a statistical power of 0.8, the calculated sample size was 102 patients using PASS11.0 (NCSS LLC, Utah) software. A total of 120 patients were recruited to provide for potential dropouts. SPSS version 20.0 (SPSS, Inc., Chicago, IL) was used for data management. The normality of data was assessed using the Shapiro-Wilk test. Normally distributed variables were expressed as the mean ± SD and compared between genders using Student's *t*-test. Non-normally distributed variables were expressed as the median (range) and analyzed using the Mann-Whitney U test. Ultrasound was compared with the gold standard of HC video-assisted laryngoscopy as a diagnostic test; accuracy, sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were calculated. The corresponding 95% confidence intervals (CIs) were calculated based on the Clopper-Pearson method in SAS 9.4 (SAS Inc., NC). Agreement between ultrasound and video-assisted laryngoscopy was evaluated by the kappa consistency test. P <.05 was considered to be statistically significant.

Results

A total of 120 patients were initially enrolled in the study and 105 were included in the final analysis. Figure 4 presents the allocation process. The demographic data of the 105 patients who completed the trial are presented in Table 1. The differences in height, weight, and thyromental distance between males and females were statistically significant (P <.05). There was no significant difference in VCD detected by ultrasound and the tip-carina distance detected by FOB between males and females (Table 1).

Table 1 Patient Demographics

	Total (n=105)	Female (n=62)	Male (n=43)	P (Male vs Female)
Age（yr）	53（20 , 69）	53 (20, 69)	54 (20, 65)	0.757
Weight（kg）	64.0 ± 9.8	60. 6± 8.9	68.9 ± 8.7	<0.001
Height（cm）	164.8 ± 8.5	160.0 ± 6.6	171.7 ± 5.9	<0.001
BMI	23.0（17.0 , 33.7）	22.8 (18.8, 33.7)	23.1 (17.0, 30.9)	0.669
VCD（cm）	2.9±0.6	2.9±0.6	3.0±0.6	0.607
TCD (cm)	3.6 (1.3, 6.4)	3.4 (1.3, 6.4)	3.6 (1.9, 6.4)	0.124

Variables are presented as mean ± SD (range) or median (range), VCD: vocal cords -cuff –distance, TCD: tip- carina distance, P- comparison between male and female

It took about 30 sec to 1 min to find the vocal cords and mark on the skin, and it took another 3 to 5 min to finish the rest of the procedure, from filling the cuff with saline to withdrawing FOB. The ventilator was stopped for no more the 20 seconds during FOB examination, and no change in SpO_2 was found. The diagnoses of too shallow an intubation for one female and one male and too deep an intubation for one female made by ultrasound was in agreement with the diagnoses made by direct laryngoscopy. The data on these patients are shown in Table 2. After inserting the tube forward for about 1.5 cm, the glottis in each of the two shallow intubation patients lay between the two depth makers as confirmed by video-assisted laryngoscopy. After pulling the tube up about 1cm, the glottis position of the deep intubation patient was also corrected. The distances from the ETT tip to the carina measured by FOB were between 1.3 to 6.4 cm with no statistical difference between males and females. This distance was less than 2 cm for a total of 1 male and 8 female patients. Except for the No.3 patient in

Table 2, the tubes were not withdrawn. For these 8 patients, fibreoptic bronchoscopy was performed during surgery to ensure no endobronchial intubation occurred.

Table 2. Data of the three patients with incorrect insertion depth

Patient	VCD (cm)	Sex	Age (y)	Height（cm）	Weight（kg）	BMI (kg/m^2)	TCD（cm）
1	1.1	M	20	188	80	22.6	6.4
2	1.4	F	61	162	60	22.8	3.3
3	4.4	F	61	152	52	22.5	1.3

VCD: vocal cords-cuff distance, TCD: tip- carina distance,

Using video-assisted laryngoscopy as the standard criterion (Table 3), the overall accuracy of ultrasound was 100.0% (95% confidence interval [CI]:96.6–100%), with a sensitivity of 100% (95% CI: 96.5%–100%), a specificity of 100% (95% CI: 29.2%–100%), PPV 100% (95% CI: 96.5%–100%), and NPV 100% (95% CI: 29.2%–100%), and 100.0% (95% confidence interval [CI]:96.6–100%) in detecting the correct position of the ETT. Overall, the correct identification of tracheal versus bronchial intubation was 100.0% (95% confidence interval [CI]:96.6–100%). The kappa value was 1.

Table 3. Results of Ultrasound vs. Video assisted laryngoscope

	Video assisted laryngoscope	
Ultrasound	Positive	Negative
Correct	102	0
Incorrect	0	3

Video assisted laryngoscope: identifying the relationship of vocal cords and depth markers. Positive: vocal cords lied between two depth markers; otherwise it is negative. Ultrasound: identifying the distance between upper edge of cuff and the glottis. Correct:

the distance between upper edge of cuff and glottis was in the range of 1.9 to 4.1cm; otherwise it is incorrect.

Discussion

Among the 105 cases using the 23/21 rule, two cuffs were too close to the vocal cords and one too far away from the vocal cords. The diagnoses of too deep intubation and too shallow intubation made by ultrasound were in agreement with direct laryngoscopy. The distances measured by ultrasound had high accuracy, sensitivity, specificity, PPV, and NPV for identifying the position of ETT at the level of the glottis. It was also found that malposition of the ETT could be corrected based on the results of ultrasound.

Unlike previous studies, which were mainly focused on the avoidance of too deep an intubation, the safety of the vocal cords was concerned as a priority in our study. We believe that for most adults, ETT cuff impingement on the vocal cords or compression of the recurrent laryngeal nerve are more harmful—and more readily ignored—than endobronchial intubation.

In an early study, Cavo et al.[2] pointed out that the rigid thyroid lamina was lateral to the anterior branch of the recurrent laryngeal nerve; they suspected that the vulnerable point of the recurrent laryngeal nerve lay between 6 and 10 mm below the posterior end of the free edge of the vocal cord. Several studies have shown that extension of the neck and concomitant movement of the ETT was a risk factor for vocal cord injury owing to neck extension during withdrawal of the ETT from the trachea (1.9 cm in Conrardy's study,[3] 1.7 ± 0.8 cm in Kim's study,[18] and 0.9 ± 0.9 cm in Hartrey's study[5]). Under external force, the withdrawal distance of the ETT is increased.[19] Since the cuff-free area of 2 to 4 cm below the vocal cords can be guaranteed by observing the insertion guide marks, this criterion was adopted as the standard for correct ETT positioning in the present study.

Our study showed that in using 23/21 rule, the rate of correct ETT positioning was as high as 97.1%. In only one male patient and one female, the tube insertion was considered to be too deep. Some anesthesiologists suggested that it would be safer to use a 22/20 rule for Asian people to avoid endobronchial intubation.[9, 20] However, if the 22/20 rule had been used in our study, the number of patients with a VCD less than 1.9 cm would have increased to 12, with a greater risk of harmful impingement of the cuff on the vocal cords.

In our study, the VCD was measured with ultrasound. Although some studies have used FOB to make sure that the ETT cuff was 2 to 3 cm below the vocal cords,[21, 22] we found it difficult to identify the cuff and the vocal cords via FOB when the ETT was already in place. We used direct video-assisted laryngoscopy to verify the position of the vocal cords with respect to the ETT cuff. Although a quantitative measurement could not be achieved with laryngoscopy, agreement on correct position of ETT using two these two methods can still be justified.

With the increasing popularity of ultrasound use in the field of anaesthesia, several studies have used ultrasound to help confirm the depth of intubation. McKay et al.[23] observed an ultrasound image of tracheal ring undulation caused as the ETT was advanced and considered it as an indication of proper ETT depth. Actually, such an image can only be regarded as a sign of endotracheal intubation. Uya et al.[24] defined adequate ETT placement as complete visualization of the saline-injected ETT cuff at the suprasternal notch. Similarly, Tessaroa et al.[16] considered that ultrasonography of the saline-inflated cuff at the suprasternal notch represented correct ETT insertion depth. Some concern has been raised about how far above the suprasternal notch the cuff should be located.[25-27]

Our study was the first to use ultrasound to measure VCD. In awake patients, in the ultrasound transverse view, the hyperechoic vocal ligaments can easily be visualized.

During phonation, the true cords move toward the midline. Surface projection of the vocal cords can then be marked on the skin after obtaining the optimal image of the true vocal cords. Surface projection of the proximal edge of the cuff can also be marked on the skin, as in the present study. Our results show that this is a reliable method for identifying incorrect intubation depth. Also, adjustment of insertion depth can be made based on the VCD obtained with this method. One can argue that it is needless to measure VCD in every patient intubated with the 23/21 rule. However, for patients whose necks are in extension during surgery, pre-surgery evaluation of the relative position of the vocal cords and cuff may be necessary. In the present study, although patients with Mallampati classification 3 and 4 and mouth opening less than 3 fingers were excluded, vocal cords could not be completely exposed using video-assisted laryngoscope on 11 of 120 patients. Measuring the VCD as shown in the present study may serve as an alternative under such circumstances.

Limitations of the study

There are several limitations to our study. Firstly, the number of incorrect tube positions in our study was small. A larger sample size study may be needed to verify our results. Secondly, for some male patients with prominent Adam's apples, the probe could not be fully attached to the skin and the vocal cords could not be completely seen. Three male patients were excluded from the study due to unsatisfactory ultrasound images of the glottis. Putting a specially designed water-filled pad or gel-like pad which fits the surface of the probe between the probe and the skin may increase the contact area and enhance ultrasound imaging. Thirdly, although too shallow an intubation can be avoided using the present method, too deep intubation cannot. Using lung ultrasound technique, endobronchial intubation can be determined, and bronchial intubation avoided.[21] For patients with extremely short airways, the proper choice is to select an ETT with a short distance from the catheter tip to the upper edge of the cuff. Lastly, although the ultrasound examination was done by experienced anesthesiologists, it still took 5 to 8 min to complete the whole examination procedure.

Training is needed for inexperienced anesthesiologists but a study has shown that this technique can be easily learned.[24]

Conclusion

Ultrasonography of the upper edge of a saline-inflated ETT cuff appears to be a simple and reliable method for identifying the distance between the vocal cords and the proximal edge of the ETT cuff, and malposition of the ETT can be corrected according to the results of the ultrasound. Confirmation of a safe distance from the upper margin of the cuff to the vocal cords may prevent injury to the vocal cords and recurrent laryngeal nerve.

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Footnotes

Contributors ML designed the study and revised the final manuscript as submitted. XC and WZ contributed equally to this paper, and they were both the first authors. XC, WZ, ZY and JG carried out all data collection. XC and WZ carried out data analyses and drafted the initial manuscript. All authors approved the final manuscript as submitted.

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Competing interests: None.

Patient consent: Patient consent obtained.

Ethics approval: The study was approved by Peking University Third Hospital Ethics Committee (IRB00006761-2016163).

Provenance and peer review：Not commissioned; externally peer reviewed.

Data sharing statement: A full anonymized dataset is available from the corresponding author on request.

Figure legend

Figure 1. Transverse scan over the glottis. asterisks, thyroid cartilage; V: vocal cords.

Figure 2. Demonstration of probe placement. Line A: probe placed at the level of vocal cord, Line B: probe placed at the level of proximal edge of tube cuff

Figure 3. Longitudinal view of the intubated trachea, with the cuff of the endotracheal tube inflated with saline. Arrows pointing downwards - anterior wall of the cuff. Arrows pointing upwards - anterior wall of the ETT.

Figure 4. Allocation process

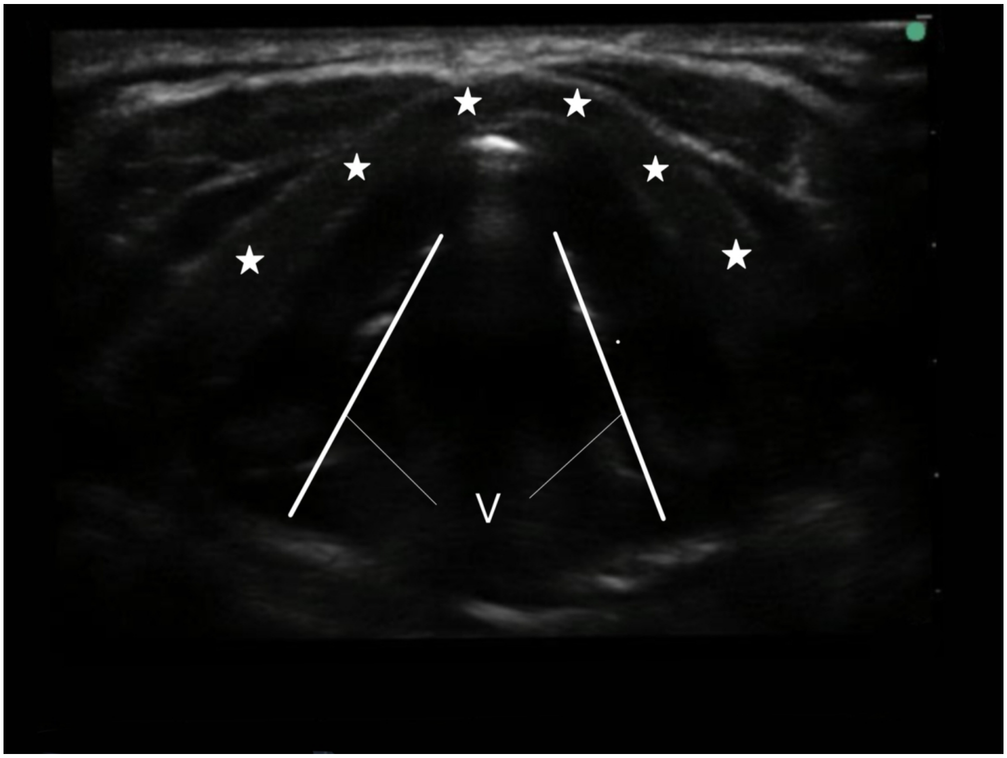


Figure 1. Transverse scan over the glottis. asterisks, thyroid cartilage; V: vocal cords.

137x102mm (300 x 300 DPI)

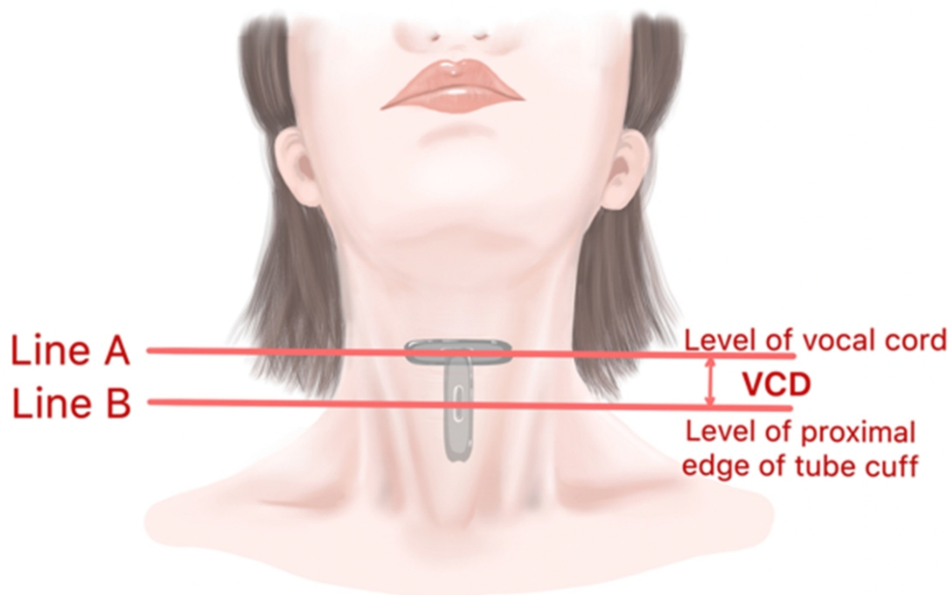
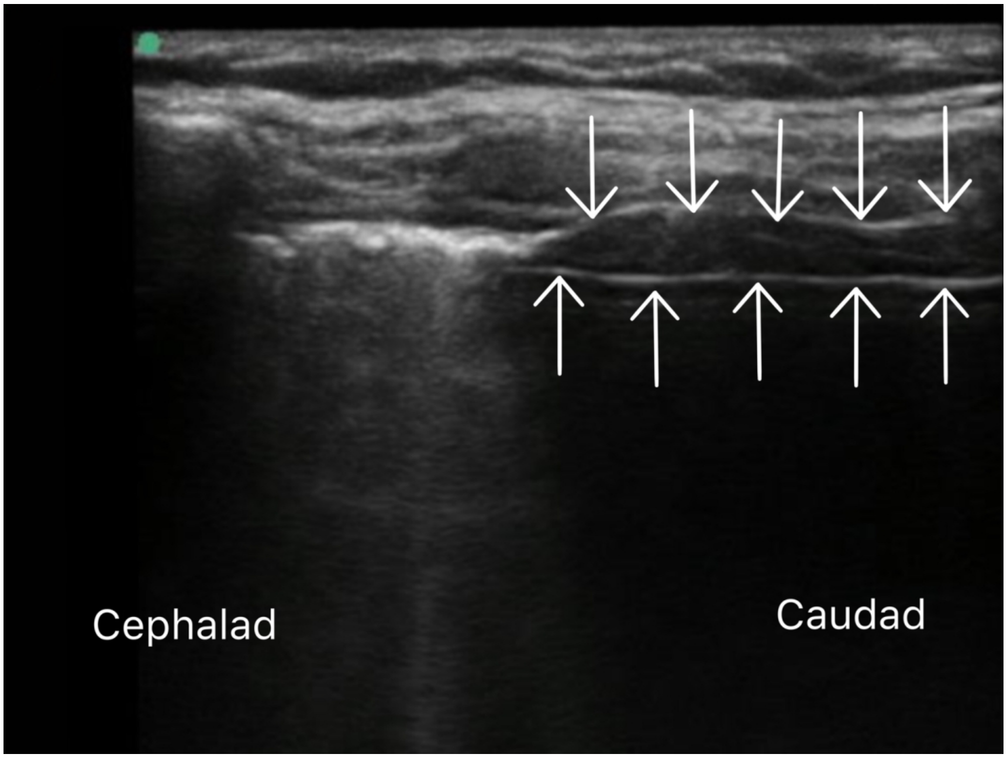


Figure 2. Demonstration of probe placement. Line A: probe placed at the level of vocal cord, Line B: probe placed at the level of proximal edge of tube cuff

99x74mm (300 x 300 DPI)



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137x102mm (300 x 300 DPI)

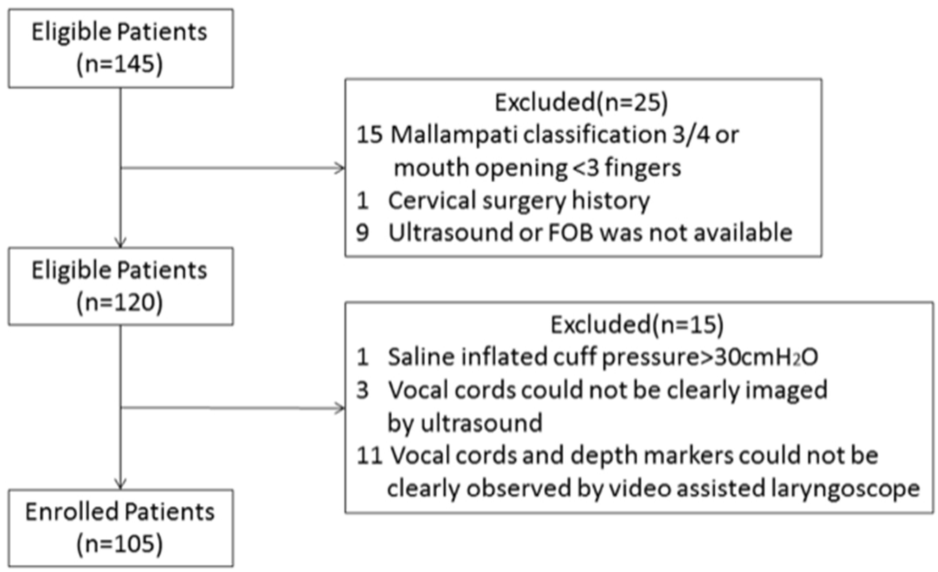

Figure 4. Allocation process

99x63mm (300 x 300 DPI)

STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No.	Recommendation	Page No.	Relevant text from manuscript
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	2	
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	2	
Introduction				
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	3-4	
Objectives	3	State specific objectives, including any prespecified hypotheses	5	
Methods				
Study design	4	Present key elements of study design early in the paper		
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	5	
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants	5	
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case		
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	7	
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	7	
Bias	9	Describe any efforts to address potential sources of bias		
Study size	10	Explain how the study size was arrived at	8	

Continued on next page

Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	8
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	8
		(*b*) Describe any methods used to examine subgroups and interactions	
		(*c*) Explain how missing data were addressed	
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed *Case-control study*—If applicable, explain how matching of cases and controls was addressed *Cross-sectional study*—If applicable, describe analytical methods taking account of sampling strategy	
		(*e*) Describe any sensitivity analyses	
Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	
		(b) Give reasons for non-participation at each stage	
		(c) Consider use of a flow diagram	Figure 4
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	9
		(b) Indicate number of participants with missing data for each variable of interest	
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount)	
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time	
		Case-control study—Report numbers in each exposure category, or summary measures of exposure	*9*
		Cross-sectional study—Report numbers of outcome events or summary measures	
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	10
		(*b*) Report category boundaries when continuous variables were categorized	
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	

Continued on next page

Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	
Discussion			
Key results	18	Summarise key results with reference to study objectives	11
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	13
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	11-13
Generalisability	21	Discuss the generalisability (external validity) of the study results	13
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	16

*Give information separately for cases and controls in case-control studies and, if applicable, for exposed and unexposed groups in cohort and cross-sectional studies.

Note: An Explanation and Elaboration article discusses each checklist item and gives methodological background and published examples of transparent reporting. The STROBE checklist is best used in conjunction with this article (freely available on the Web sites of PLoS Medicine at http://www.plosmedicine.org/, Annals of Internal Medicine at http://www.annals.org/, and Epidemiology at http://www.epidem.com/). Information on the STROBE Initiative is available at www.strobe-statement.org.