Age Characteristics of the larynx in infants during the first year of life

Abstract

Background and Purpose: Investigations of the larynxes in thirty infants during the first year of life of both sexes, randomized trial, were performed by morphological and histologic analysis. Morphometric parameters of the larynx as: length (anterolateral parameter), width (transverse parameter) and thickness (anteroposterior parameter) were determined. These parameters determine the size and shape of the larynx. The reciprocal relation parameters which determine size of the larynx in infants and the body length are in high correlation. Therefore, the equation for calculating the size of the larynx out of the body height was founded. Histologic characteristics of the laryngeal cartilage are constant. They indicate evident changes, that are the basis for approximate determination of a child’s age.

Materials and Methods: The organs of the infants were taken from pathoanatomical autopsies. None had changes in the respiratory system. The major methods of the investigation were anatomical macrodissection, morphological and histologic analysis and statistics.

Results: The average body length in infants was 540 ± 20 mm (54 ± 2 cm) and the average larynx length was 11.9 ± 0.3 mm. There was a correlation between these parameters p < 0.01 (r = 0.75). The average value of the width of the larynx in infants was 17.7 ± 0.5 mm. The width of the larynx was in correlation with the body length p < 0.01 (r = 0.79). The average value of the thickness of the larynx was 12.6 ± 0.4 mm. This parameter was correlated with the body length p < 0.01 (r = 0.82). Histologic analyses results of our investigation, cartilage of the larynx in infants, show that hyalin structure is the result of age changes.

Conclusion: Quantitative anatomical knowledge on the larynx in the pediatric population are necessary for the clinical orientation, particularly for choosing a suitable endotracheal tube. Size of the larynx in premature, neonates and small children, constantly follows the external body dimensions, particularly the body height, that is confirmed by the correlation factor in its highest value. They indicate evident changes, that are the basis for approximate determination of a child’s age. Hyalin structure is in correlation to the children’s age.

INTRODUCTION

First investigations of the larynx were carried out by Bouchut (1). The author used a small, iron-ring shaped device, “virole”, which was inserted between vocal cords with a probe. Data on intubation and artificial ventilation for resuscitation of newborn children were given by Delfraysse (2). The development of laryngeal cartilages were studied by
The authors paid particular attention to the thyroid cartilage and triticate cartilage, particularly to the position of the latter between cornua superiora of the thyroid cartilage and cornua majora of the hyoid bone. The ossification of laryngeal cartilages was studied by Mupparapu and Vuppalapati (4). They concluded that there is a general trend of increased ossification of laryngeal cartilages with advancing. Modern equipment and techniques enable the use of anaesthesia and artificial ventilation particularly in pediatric population, such as premature babies and babies born with smaller gestational age; such cases are present in a large number (Hack et al. and Lee et al., 1980) (5, 6). Frequent intubations in premature babies and neonates are followed by a high risk of laryngeal and tracheal damage, especially by laryngeal stenosis which shows a trend of constant increase (7). The main cause of laryngeal damage and mortality of infants is inadequate application of the endotracheal tube in relation to the anatomical shape of the airway. The shape of the standard endotracheal tube in children is not corresponding to the anatomical shape of the airway, so it represents the most frequent cause of the occurrence of lesions while this procedure is applied for therapeutic and diagnostic purposes. The damages occur on the larynx (cricoid and arytenoid cartilage) and on the trachea. The smallest endotracheal tube available for clinical usage has internal diameter of 1.5 mm, and its external diameter is 2.7 mm. That represents the smallest size of the larynx that can be intubated by this size of tubus described by Schild (8).

Although the incidence of neonatal subglottic stenosis markedly decreased in the 1990s, the current incidence of neonatal subglottic stenosis is about 2 % (9).

Significance of the bifid epiglottis was described by Stevens and Ledbetter as a rare and heterogeneous anomaly (10).

Cricotracheal resection in children of two and less than two years of age was performed by Johnson et al. (11). Navsa et al dissected and measured the neonatal cricothyroid membrane on 27 cadavers. They established that cricoid membrane dimensions are too small for the tracheal tube to pass through. That is why the larynx cartilage fracture may occur. Little is known about the anatomy of the neonatal airways, particularly about the cricoid membrane size (16).

The anatomy of the larynx in infants

In small infants, the larynx has special anatomical relations. In newborns, larynx has smaller dimensions in comparison to the body than it is the case in adults. At this age larynx is characterized by a high position because it lies on the level of the 4th cervical vertebrae, and descends in the childhood period. The axis of respiratory system is parallel to digestive system axis, which enables contemporary breathing and swallowing in newborns.

Significant anatomical characteristic of the larynx at this age is soft cartilaginous structure of the skeleton and relatively narrow larynx lumen. In small infants the larynx is a narrow gate in the area of vocal cords. It is 7–9 mm wide, and conal subglottis in sucklings is 5.5–6 mm wide (13). If it is narrowed to 4 mm because of edema, breathing is almost impossible. Softness of the cartilaginous skeleton additionally favors deformity of the larynx and trachea. Infants’ laryngeal tissue is in its entirety softer than in adults. The upper end of the larynx is cone-shaped, the cricoid cartilage is inclined backward, vocal cords are shorter, and epiglottis is narrower. In fact, epiglottis hangs above the larynx. Epiglottis often has the shape of the Greek letter omega, but later on it assumes the normal shape of a leaf or a triangle. In a human being, the larynx has a special function as a phonatory organ, not just as a respiratory tube (14).

The Physiology of the larynx in infants

Aristotle is considered a creator of phonetics. He studied anatomy of phonatory organs. He spoke on breath, pneumonia, that produces voice. Galen made a progress performing experiments in this field. He compared the human throat to musical instruments that have a mouthpiece, and the human mouthpiece is the glottis. In the middle ages, phonetics was forgotten.

In the new age, Leonardo da Vinci showed an interest in phonetics. Currently, vocal cords are considered to vibrate mainly in horizontal position, which means that they move during phonation away from the middle and again nearer to the middle. Those excursions are 4 mm long for each vocal cord, which means that distance between vocal cords for some tones is 8 mm. A newborn is capable to produce sounds by the first octave, immediately after birth. It is just a reflex at the beginning, and serves as an unconscious preparation for speech function. In the third or fourth month of life, or during the first half of the first year of life, a child babbles sounds, while at the end of the first year a child pronounces one-syllable words (13, 14, 15).

The objective of this study was to investigate the anatomy of the larynx in infants in the first year of life, including size and shape of the larynx, size of the cartilage and their histologic composition, and to establish characteristics that determine a child’s age.

MATERIAL AND METHODS

The investigations were performed on the larynges of thirty infants, of both sexes, during the first year of life as a randomized trial applying morphological and histological analyses. The organs of the infants were taken from pathoanatomical autopsies. None had changes in the respiratory system. Dissected organs were fixed in 4% buffered formalin, each organ for seven to ten days.

Experimental procedures

The major methods of the investigation were: anatomical dissection, microdissection, morphological and histological analysis and statistics. Morphometric parameters that determine the size of the larynx were measured (30 children). These are: length (anterolateral pa-
rameter – from the middle of the incisurae thyroideae superior to the low margin of the cricoid cartilage), width (transversal parameter – at the level of tubercula superiorna cartilaginis thyroideae), and thickness (anteroposterior parameter – the midst of incisurae thyroideae superior to midst of the upper margin of the laminae cartilaginis cricoideae).

Every single cartilage of the larynges was separated by anatomical preparation and then measurements were performed on the cartilage of thyroidea, cricoid cartilage, vocal cartilages and epiglottis.

On the thyroid cartilage, ventral diameter, dorsal diameter, right and dorsal diameter, left (from the margin of cornu superius to the margin of cornu inferius) were determined. On the cricoid cartilage height of arch (from upper to lower margin), height of laminae (from upper to lower margin), internal sagittal diameter (from the midst of the inner side of arch to the midst of inner side of laminae), external sagittal diameter (from the midst of external side of arch to the midst of external side of laminae), inner transversal diameter (middle point of arch, right and left), and external transversal diameter (between lateral points of laminae). On vocal cartilages, these measurements were performed: height to the left (between apex and basis), height to the right, basal width to the left (between processus muscularis and processus vocalis), and basal width to the right. Length (from front to back position) and width (between the most distant points of the back part) were determined on the epiglottis. Laryngeal cartilages were investigated by microanatomical techniques.

Histologic sections of every single cartilage were made. Sections for histological preparations were made according to the largest diameter of cartilage. Laryngeal cartilages were embedded in paraffin, and sections were cut on Reichter microtome. The sections were coloured by the following methods: hematoxillin-eozin, van Gieson and periodic acid according to Schiff. Histological sections were analyzed by Polyvar-Reichert microscope. All measurement were carried out in duplicate and approximate values considered. For this purpose calliper (Vernier) and nonius were used as measurement instruments.

Standard statistical methods were used such as: mean value x, standard deviation SD, standard error SEM, significance of results was tested by T- and F-test. The coefficient of correlation was used and its statistical significance was taken by a risk under 1% (p < 0.01). The correlation degree was determined among the parameters determining the size of the larynx and body height and parameters of a single larynx cartilages. The coefficients of regressive line y = ax + b were calculated for the measured values that were in correlation.

**Techniques for larynx preparation**

The technique for anatomical preparation of the larynx differs much from the preparational methods of the larynx in persons whose death is determined by forensic expertise. This particularly concerns the persons (adults, infanticide) whose death was caused by strangling (manually or by some ligature). Anatomical preparation is characterized by graduality, precision and diversity in preparational methods. This means that the larynx can be seen in total, isolated with muscles, hyoid bone and thyroid gland; only the cartilaginous skeleton of the larynx can be shown; in total larynx can be isolated together with the pharynx, with no moving away external muscles of larynx. At the end, separation of cartilaginous skeleton can be done by precise dissection.

**RESULTS**

The average body length in infants was 540 ± 20 mm (54 ± 2cm) and the average larynx length was 11.9± 0.3 mm. There was a correlation between these parameters p < 0.01 (r = 0.75). The average width of the larynx in infants was 17.7 ± 0.5 mm. The width of the larynx was in correlation with the body length p < 0.01 (r = 0.79). The average value of the thickness of larynx was 12.6 ± 0.4 mm. This parameter was correlated with body length p < 0.01 (r = 0.82) by means of coefficients (a and b) of the regression line y = ax + b; from the assumed size of body length it was possible to estimate the size of the larynx in infants (Table 1).

**TABLE 1**

<table>
<thead>
<tr>
<th>Estimated size of the larynx (y)</th>
<th>Assumed body length (x)</th>
<th>SEP</th>
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<tbody>
<tr>
<td>LL 0.013 x height + 4.82</td>
<td>0.90</td>
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</tr>
<tr>
<td>WL 0.022 x height + 5.70</td>
<td>1.35</td>
<td></td>
</tr>
<tr>
<td>TL 0.017 x height + 3.56</td>
<td>0.91</td>
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</table>

Legend: LL – length of the larynx, WL – width of the larynx, TL – thickness of the larynx, SEP – Standard error of the predicted value

Functional dependence of the estimated size of the larynx on the assumed body length.

These parameters determine the size and shape of the larynx. The short and wide type of the larynx is dominant in infants during the first year of life (Figures 1, 2). We examined the morphological characteristics of laryngeal cartilages (Figure 5).

The average value of the ventral diameter of thyroid cartilage in infants was 6.79 ± 0.23 mm and the right dorsal diameter was 17.18 ± 0.46 mm and the left dorsal diameter was 17.09 ± 0.43 mm.

The average value of the arch of cricoid cartilage height was 2.54 ± 0.60 mm and the average lamina height was 9.01 ± 0.19 mm. The average value of the inner sagittal parameter of cricoid cartilage was 5.96 ± 0.18 mm and the average value of the external sagittal parameter was 8.85 ± 0.21 mm. The average value of the inner transversal parameter of cricoid cartilage was 6.44 ± 0.15 mm and the average value of the external transversal pa-
The average value of the height of the left vocal cricoid was 5.78 ± 0.15 mm and the average value of the height of the right vocal cricoid was 5.73 ± 0.14 mm. The average value of the basal width of the left vocal cricoid was 5.82 ± 0.13 mm and the average value of the basal width of the right vocal cricoid was 5.82 ± 0.13 mm. The average value of the width of epiglottic cartilage was 8.92 ± 0.30 mm and the average value of the length of epiglottic cartilage was 13.15 ± 0.44 mm.

A significant statistical difference in arithmetical mean was found between the ventral diameter of thyroid cartilage and the height of arch of the cricoid cartilage, \( p < 0.05 \). Values of the ventral diameter and the height of arch of cricoid cartilage were in correlation, \( p < 0.01 (r = 0.55) \). It is possible to estimate the height of arch of cricoid cartilage \( y \) out of the assumed value of the ventral diameter of thyroid cartilage \( x \) by means of the coefficient \((a, b)\) of regression line \( y = 0.14X + 1.57 \). Significant difference was found in arithmetic means between the ventral diameter of thyroid cartilage and the length of left epiglottis, \( p < 0.05 \). These parameters are in positive statistical correlation, \( p < 0.01 (r = 0.67) \). It is possible to estimate the length of epiglottis \( y \) out of the assumed value of the ventral diameter of thyroid cartilage \( x \) by means of the coefficients \((a, b)\) of regression line \( y = 1.29X + 4.36 \). (Table 2).

Significant difference in arithmetic means was found between the ventral diameter of thyroid cartilage and and the length of left arytenoid cartilage, \( p < 0.05 \). These parameters are in positive statistical correlation, \( p < 0.01 (r = 0.80) \). It is possible to estimate the height of left arytenoid cartilage \( y \) out of assumed value of the ventral diameter of thyroid cartilage \( x \) by means of coefficients \((a, b)\) of regression line \( y = 0.51 + 2.33 \). Significant difference was found in arithmetic means between the ventral diameter of thyroid cartilage and the height of right arytenoid cartilage \( p < 0.05 \), these parameters were in correlation \( p < 0.01 r = 0.82 \). It is possible to estimate the height of arytenoid cartilage right \( y \) out of the assumed value of the ventral diameter of thyroid cartilage \( x \) by means of coefficients \((a, b)\) of regression line \( y = 0.15X + 2.27 \).
Significant difference was found in arithmetic means between the height of arch of cricoid cartilage and left arytenoid cartilage height, \( p < 0.05 \), and these parameters were in correlation, \( p < 0.01 \), \( r = 0.50 \). It is possible to estimate the height of arytenoid cartilage, left (y), out of the assumed size of the arch of cricoid cartilage (x) by means of coefficients \((a, b)\) of regression line \( y = 1.21 + 2.69 \). Significant difference in arithmetic means was found between the height of arch of cricoid cartilage (x) and the height of right arytenoid cartilage, \( p < 0.05 \). These parameters were in positive statistical correlation \( p < 0.01 \), \( r = 0.58 \). It is possible to estimate the height of right arytenoid cartilage (y) out of assumed height of arch of the cricoid cartilage (x) by means of coefficients \((a, b)\) of regression line \( y = 1.32x + 2.38 \). (Table 3).

Functional dependence of the the estimated height of vocal cartilages on the assumed height of the arch of cricoid cartilage.

Significant difference was found between dorsal diameter of the right thyroid cartilage and the height of the lamina of the cricoid cartilage. The same diameters were in correlation, \( p < 0.01 \), \( r = 0.71 \). It is possible to estimate the height of the arch of cricoid cartilage (y) out of the assumed size of dorsal diameter of the right arytenoid cartilage (x) by means of coefficients \((a, b)\) of regression line \( y = 0.33x + 3.42 \). Significant difference in arithmetic means was found between the dorsal diameter of the left thyroid cartilage and the height of the lamina of cricoid cartilage, \( p < 0.05 \). The same parameters were in correlation, \( p < 0.01 \), \( r = 0.67 \). It is possible to estimate the height of the lamina of cricoid cartilage (y) out of the assumed size of dorsal diameter of the left thyroid cartilage (x) by means of coefficients \((a, b)\) of regression line \( y = 0.32x + 3.49 \), Table 4.

Functional dependence of the estimated height of the lamina of cricoid cartilage on the assumed size of dorsal diameter of the right and left thyroid cartilage.

A significant difference in arithmetic means was observed between the external transversal diameter of the cricoid cartilage and the basal width of the left arytenoid cartilage, \( p < 0.05 \). The same parameters were in correlation, \( p < 0.01 \), \( r = 0.58 \). It is possible to estimate the basal width of the left arytenoid cartilage (y) out of the assumed size of the cricoid cartilage (x) by means of coefficients \((a, b)\) of regression line \( y = 0.38x + 2.05 \). Significant difference in arithmetic means was found between external transversal diameter of the cricoid cartilage and basal width of the right arytenoid cartilage, \( p < 0.05 \). The same diameters were in correlation, \( p < 0.01 \), \( r = 0.55 \). It is possible to estimate the basal width of the right arytenoid cartilage (y) out of the assumed size of external transversal diameter of cricoid cartilage (x) by means of coefficients \((a, b)\) of the regression line \( y = 0.37x + 2.23 \), Table 5.

Functional dependence of the estimated size of basal width of arytenoid cartilages on the assumed size of external transversal diameter of cricothyroid cartilage.

Significant difference in arithmetic means was found between external transversal diameter of the cricoid cartilage and basal width of the right arytenoid cartilage, \( p < 0.05 \). The same parameters were in correlation, \( p < 0.01 \), \( r = 0.67 \). It is possible to estimate the height of the lamina of cricoid cartilage (y) out of the assumed size of dorsal diameter of the left thyroid cartilage (x) by means of coefficients \((a, b)\) of regression line \( y = 0.32x + 3.49 \), Table 6.

Functional dependence of the estimated height of the lamina of cricoid cartilage on the assumed size of the dorsal diameter of thyroid cartilage, right and left.
tilage and width of epiglottis, p < 0.05. The same parameters were in correlation p < 0.01 r = 0.78. It is possible to estimate the width of the epiglottis (y) out of the assumed size of the external transversal diameter of cricoid cartilage (x) by means of coefficients (a, b) of regression line y = 1.17X – 2.59, Table 6.

Functional dependence of the estimated width of epiglottis on the assumed size of the external transversal diameter of cricoid cartilage

<table>
<thead>
<tr>
<th>Estimated size (y)</th>
<th>Assumed size (x)</th>
<th>SEP</th>
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</thead>
<tbody>
<tr>
<td>WE</td>
<td>1.17 x ETDCC – 2.59</td>
<td>1.08</td>
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</table>

Legend: WE – Width of epiglottis

Histological analysis

**Thyroid cartilage (cartilago thyreodea)**

Thyroid cartilage in children is of hyaline structure with a very thin layer of mucose that lies on the cartilage. The cartilage shows changes in coloring intensity. This is connected to the quantity of chondroid matrix, so some fissures can be seen in the cartilage and they represent artefacts made in the course of preparing the sections as the result of liquid extraction. Mucose is thin and without specific changes (Figure 4).

**Cricoid cartilage (cartilago cricoidea)**

Cricoid cartilage in infants is of hyaline type, with different amount of chondroid substance, which affects the coloring intensity. Mucose is much thicker than the one coating the thyroid cartilage. It is composed out of fibrous fibres with many serosal glands that show different secretion intensities. Perforative ducts with different amount of connective tissue can be frequently found in the cartilage. When cricoid cartilage is observed by contrast-phase microscope, a layer of elastic and collagen fibres can be clearly seen between mucose and cartilage (Figures 5, 6).

**Arytenoid cartilages (cartilagines arytaenoideae)**

Arytenoid cartilages in infants are of hyaline type. They are of unequal thickness. The mucose has a thick layer of fibrous tissue just near the cartilage. This layer is also found in the vocal cartilages of adults, so it probably participates in forming the larynx shape. Mucose is rich due to serosal glands (Figure 7).

**Epiglottis**

Epiglottis in infants is an elastic cartilage characterized by thick mucosa and abundance of serosal glands on both sides (Figure 8). Oral side of this cartilage is coated by cuboidal – stratified epithelium, and partly by respiratory epithelium. Characteristic age change is migration of the cuboidal-stratified epithelium to the laryngeal side. Epiglottis in infants contains more elastic tissue than in adults, in whom certain amount of collagen precipitates, which makes epiglottis firmer.

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TABLE 6

Functional dependence of the estimated width of epiglottis on the assumed size of external transversal diameter of cricoid cartilage.

<table>
<thead>
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Legend: WE – Width of epiglottis

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**Figure 4.** Thyroid cartilage (van Gieson 10X). Ratio of the connective tissue, mucose and cartilage.

**Figure 5.** Cricoid cartilage (van Gieson 10 x) Ratio of the cartilage and mucose rich by glands.

**Figure 6.** Cricoid cartilage (phase contrast 10 x). View of the cartilage and corresponding mucose.

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**Figure 7.** Arytenoid cartilages (cartilagines arytaenoideae). View of the mucose rich by glands and serosal glands.
DISCUSSION

Anatomical investigations in the field of laryngometry show that parameters of the larynx follow the growth of the body (Noback; Klock; Eckel and Sittel; Ward and Tigklia) (16–19). Schild (1984) investigated mutual relations of larynx dimensions and body size and gestational age in prematures, neonates and infants (8). Larynges of 27 infants, weighing 250 gr – 9160 g were investigated. Twenty-two infants were in 24–40 weeks of gestational age. Five infants died before reaching six weeks of life, post partem, but they were involved in the investigation because of their small size. Mutual relation between larynx size and standard clinical measures (body height – head – foot and height, height – head – buttock) was linear.

Results of this investigation show that larynx size is in correlation with body size, i.e. with body height. Our results are partially in accordance with other authors’ results. The main reason for possible difference is in research methodology. Our results show that the larynx size can be estimated out of the size of the body height, by arithmetic and statistical methods (correlation factor and coefficient of the regression line, with standard error in prediction). Some authors think that larynx dimensions are more closely connected to the body weight than to the infants’ age (17).

Results of histological analyses of cartilage of the larynx in infants our investigation show that hyalin structure is the result of age changes. Changes in the cartilage composition are in strict correlation to the age. It is known that larynx cartilages are of the hyalin type at the beginning of life, later on they differentiate into different forms, so that huge cartilages remain and the small ones turn elastic. The laryngeal cartilages composed of elastic connective tissue are prone to calcification, and the cartilages composed of the hyalin tissue are prone to ossification. In the available literature there are no data on histological investigations of laryngeal cartilages in infants.

Eckel and Sittel (1995) performed laryngeal morphometry on adults in horizontal position (18). They investigated the entire organ in the series of larynx sections which were histologically analyzed.

Clinical importance of the investigation

Laryngotomy in little children is connected to a high risk and followed by many complications. Therefore, findings of anatomical investigations on the larynx size (laryngometry) that are the basis for cannula size determination, as well as the cannulation timing and complications, are of great importance (20, 21).

Fracture of the laryngeal cartilage has a special forensic-medical importance, as it is associated to the neck trauma (hanging, strangling – infanticide), because it happens most frequently on the cornua superiora of the thyroid cartilage (3). The data on laryngeal angles, air –lumen and thickness of the laryngeal skeletal parts can significantly help in planning endolaryngeal surgeries, or during transcutaneous insertion of electrodes for laryngeal electromiography.

Garel et al. (1992) examined larynges in children by means of ultrasonography, which represented a new method for researching normal and pathologic states, particularly functional disorders of the larynx (22). Phonosurgical laryngoplastics is a relatively new branch in surgical laryngology (23). Kaufman (2001) described the problems and complications that occur when performing laryngoplastics, because of the anatomical variations in larynx and unexpected events in surgery connected to this procedure, such as: variable size of the lower thyroid incisure, or its lack, which can cause thyroid cartilage fracture; ossification of thyroid cartilage in the early childhood; tubercules occurring in the region of cricoarytenoid joint which, if large, represents a good orientation in searching this joint; occurring ankylosis and pseudoankylosis. If a surgeon has not learned about existence of this type of variations, he/she may cause ankylosis of the larynx joint (24).
CONCLUSION

Quantitative anatomical knowledge on the larynx in pediatric population is necessary for clinical orientation, particularly for choosing a suitable endotracheal tube. The size of the larynx in prematures, neonates and small children constantly follows the external body dimensions, particularly the body height, which is confirmed by the highest value of correlation factor. Therefore, equation was established for calculating the size of the larynx based on body height.

Histologic characteristics of the laryngeal cartilage are constant. They indicate evident changes that are the basis for approximate determination of a child’s age. Hyalin structure is in correlation to the children’s age.

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