3D atlas of human embryology

New insights in human development

de Bakker, B.S.
CHAPTER 5.2 - VARIANTS OF THE HYOID-LARYNX COMPLEX WITH FORENSIC IMPLICATIONS AND CONSEQUENCE FOR EAGLE’S SYNDROME

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Submitted

“Let us only imagine that birds had studied their own development and that it was they in turn who investigated the structure of the adult mammal and of man. Wouldn’t their physiological textbooks teach the following?

Those four and two-legged animals bear many resemblances to embryos, for their cranial bones are separated, and they have no beak, just as we do in the first live or six days of incubation; their extremities are all very much alike, as ours are for about the same period; there is not a single true feather on their body, rather only thin feather-shafts, so that we, as fledglings in the nest, are more advanced than they shall ever be ... And these mammals that cannot find their own food for such a long time after their birth, that can never rise freely from the earth, want to consider themselves more highly organized than we?”

Karl Ernst von Baer, 1828
Abstract

Introduction: The hyoid-larynx complex is one of the most polymorphic regions in the human body. Anomalies of this complex, e.g. Eagle's syndrome and the aberrant hyoid apparatus, are of great importance for radiological examination and surgery of the neck region. The significance of this complex has also been well recognized in forensic sciences, as fractures in this complex are important indicators for strangulation or blunt trauma on the neck. We aimed to provide an overview of the anatomical variations of the hyoid-larynx complex and explain their etiology based on its development.

Material and Methods: To assemble an overview of the deviations from normal anatomy, 284 radiological scans of excised hyoid-larynx were examined and supplemented with rare cases described in literature and from a historical anatomical collection.

Results: Two third of the examined hyoid-larynx complexes deviated from the anatomical standard. This was mostly due to minor variations like age-dependent ankylosis of the hyoid and presence of triticeal cartilages in the lateral thyrohyoid ligament. One fifth of the cases, however, comprised striking anatomical variants, mostly in the trajectory of the second pharyngeal arch cartilage.

Discussion: Our recent observations on the development of the hyoid-larynx complex in human embryos explains the etiology of a variety of anatomical variations. Eagle’s syndrome and the aberrant hyoid apparatus should be considered as two expressions of one entity, preferably referred to as ‘second pharyngeal arch cartilage anomalies’. As some variants mimic a fracture of the hyoid-larynx complex, we emphasize the importance of our work for forensic experts.

Introduction
Anatomical variations of the hyoid-larynx complex occur in 4-30% of the general population (De Paz et al. 2012; de Santana Jr et al. 2006; Petrovic et al. 2008; Naimo et al. 2015). Anomalies of this complex are of great importance for radiological examination and surgery of the neck region (De Paz et al. 2012; Gok, Kafa, and Fedakar 2012). The significance of anomalies has also been well recognized in forensic sciences, as fractures in this complex are important indicators for strangulation and blunt or penetrating trauma on the neck (Gok, Kafa, and Fedakar 2012; Kindschuh, Dupras, and Cowgill 2010; Urbanova et al. 2013; Saukko 2015).

The aim of this study was to provide an overview of variations in the hyoid-larynx complex and explain their etiology based on its development. Our data will be discussed in the light of the currently available literature concerning clinical and forensic relevance, providing an overview of this highly polymorphic complex, from development to death.
Anatomical variants

Normal anatomy
The normal adult hyoid-larynx complex (Fig. 1A) is described as combination of hyoid apparatus (i.e. styloid processes, stylohyoid ligaments and lesser horns of the hyoid), body and greater horns of the hyoid bone, uncalcified thyroid-, cricoid- and arytenoid cartilages and their ligaments proper. The thyroid cartilage encompasses its superior and inferior horns. No additional cartilaginous structures are present in the trajectories of the stylohyoid ligaments, median thyrohyoid ligament and lateral thyrohyoid ligaments and no ankylosis of the joints between hyoid body and greater and/or lesser horns has occurred. Normal length of the styloid process is generally described as 20-30 mm (De Paz et al. 2012; de Santana Jr et al. 2006; Eagle 1949, 1958; Wellinger, Dugast, and Desnos 1966).

Fig. 1. Overview of the normal adult human anatomy of the neck region. A: Lateral view of a schematic representation of the normal anatomy of the adult neck region. B: Ventral view of the hyoid bone, thyroid and cricoid region with emphasis on the anatomical structures mentioned in this paper. Note that the arytenoid cartilages (dashed lines) lie in fact dorsally of the thyroid cartilage.

Minor variations
Minor variations in hyoid-larynx comprise age-related fusion of the body with greater and/or lesser horns by ankyloses of the joints (Fig. 2A2-6), age-related calcification of the thyroid and presence of a triticeal cartilage in the lateral thyrohyoid ligament (Fig. 2D2) (Di et al. 2004; Pollanen and Chiasson 1996; Soerdjbalie-Maikoe and van Rijn 2008; Watanabe, Kurihara, and Murai 1982; Radlanski and Renz 2010). Morphological variations of the hyoid are closely related to sex (Gok, Kafa, and Fedakar 2012; Kindschuh, Dupras, and Cowgill 2012; Miller, Walker, and O’Halloran 1998; Urbanova et al. 2013), race (Gok, Kafa, and Fedakar 2012; Kindschuh, Dupras, and Cowgill 2010, 2012), body proportions (Pollard et al. 2011; Urbanova et al. 2013) and age (D’Souza, Harish, and Kiran 2010; Gok, Kafa, and Fedakar 2012; Gupta et al. 2008; Hansch 1977; Miller, Walker, and O’Halloran 1998; Pollanen and Chiasson 1996; Porrather 1969; Soerdjbalie-Maikoe and van Rijn 2008; Ubelaker 1992; Urbanova
European hyoids are broader and shorter than African ones (Kindschuh, Dupras, and Cowgill 2012). Distal ends of the greater horns are significantly longer in women than in men (Gok, Kafa, and Fedakar 2012; Miller, Walker, and O’Halloran 1998; Pollanen and Ubelaker 1997), whereas male hyoids are generally larger than female ones (Jelisiejewa, Szmurlo, and Kuduk 1968; Miller, Walker, and O’Halloran 1998; Reesink et al. 1999; Shimizu et al. 2005). Inward curving and flattening of the greater horns are typical for the male hyoids (Urbanova et al. 2013). Furthermore, male hyoids are more susceptible to age modifications (Urbanova et al. 2013). Besides that, males show a higher degree of thyroid ossification, ultimately leading to the completely ossified os thyroideum (Jurik 1984; Di et al. 2004). Finally, hyoid muscle attachment sites also show some individual variation. These minor variations occur so often that they cannot be considered as anatomical variants (Soerdjbalie-Maikoe and van Rijn 2008).

**Age-related ankylosis**

Recently, hyoid bone density and ankyloses of the joints between hyoid body and greater and/or lesser horns are getting more attention as possible predictor for age and sex in victim identification (Fisher et al. 2016; Naimo et al. 2015). Age-related ankylosis (Fig. 2A2-6) is a physiological process that increases with age (D’Souza, Harish, and Kiran 2010; Gok, Kafa, and Fedakar 2012; Gupta et al. 2008; Hänsch 1977; Miller, Walker, and O’Halloran 1998; Pollanen and Chiasson 1996; Porrath 1969; Soerdjbalie-Maikoe and van Rijn 2008; Ubelaker 1992). D’Souza reported a mean age of unilateral (Fig. 2A4-5) and bilateral (Fig. 2A2-3) fusion in males of 39.9 and 41.77 years respectively and in females of 37.5 and 45 years (D’Souza, Harish, and Kiran 2010). Body and greater horns usually do not fuse until the 35th to 45th year (Porrath 1969; Pendergrass, Hodes, and Schaeffer 1956) and they might even never fuse (Hänsch 1977; Porrath 1969). Fusion was not reported before the age of 18 (Vanezis 1989) or 20 (D’Souza, Harish, and Kiran 2010; Fisher et al. 2016). Non-fusion (Fig. 2A1) or unilateral fusion (Fig. 2A4-5) has been found in people after the age of 60, which makes this process highly polymorphic (D’Souza, Harish, and Kiran 2010; Miller, Walker, and O’Halloran 1998). Miller et al. suggested that fusion is not a continuous ageing process, but that genetic predisposition is the driving force behind this process (Miller, Walker, and O’Halloran 1998). Furthermore, sex seems to be of no importance to the fusion process (D’Souza, Harish, and Kiran 2010; Miller, Walker, and O’Halloran 1998). Therefore, fusion of the hyoid body with the greater horns can therefore not be used as an indicator for age or sex (D’Souza, Harish, and Kiran 2010).

**Pharyngeal arch cartilage anomalies**

Significant anatomical variants are due to the persistence of embryological cartilage (Dwight 1907). One example is the complete ossification of the lateral thyrohyoid ligament between greater horns of the hyoid bone and superior horns of the thyroid cartilage, called the *congenital hyothyroid bar* (Fig. 2D6-7) (Dwight 1907; Porrath 1969). Other cases comprise anomalies of the second pharyngeal arch cartilage, such as *stylohyoid syndrome* (Eagle’s syndrome) (Fig. 2E2) and the *aberrant hyoid apparatus* (Fig. 2E3-7). The exact incidence of anatomical changes in the stylohyoid chain is difficult to determine, since Eagle’s syndrome and the aberrant hyoid apparatus are often intermingled in literature (De Paz et al. 2012). It seems to vary from 4% or 5% (De Paz et al. 2012; de Santana Jr et al. 2006; Petrovic et al. 2008) to 28% or 30% (de Santana Jr et al. 2006; Shul’ga, Zaitsev, and Zaitseva 2006). Less than 10% of the patients in this group displays clinical symptoms (Chiang, Chang, and Chou 2004; De Paz et al. 2012; de Santana Jr et al. 2006; Petrovic et al. 2008).
Variants of the hyoid-larynx complex with forensic implications and consequence for Eagle’s syndrome

Fig. 2. The anatomical variations of the hyoid-larynx complex. Normal anatomy is shown in the first vertical column. The hyoid bone is shown in purple, second pharyngeal arch cartilage derivatives are shown in blue, third pharyngeal arch cartilage derivatives are shown in yellow and the thyroid complex and the thyrohyoid ligaments are shown in red. Cr: cranial, Ca: caudal, L: Lateral, V: ventral, D: dorsal. Each arrow indicates the location of the variation. The A row shows the normal anatomy of the hyoid bone (A1) and various degrees of ankylosis in ventral view (A2-A6). Ankylosis of the joints of the hyoid bone is considered as normal variation and happens frequently during ageing. A7: Two examples of exostoses of the hyoid bone body; median process and split median process (Di et al. 2004). The B row shows anatomical variations of the greater horn of the hyoid bone in lateral view. B1: normal anatomy, B2: Hypoplastic, B3: Intermittent, note the ankylosis between body and greater horn, B4: Exostosis, B5: Curving upward, B6: Curving downward, B7: Accessory bone. The C row shows the anatomical variations of the lesser horn of the hyoid bone in ventral view. C1: Normal anatomy, C2: Unilateral absence, C3: Bilateral absence, C4: Hypoplastic, C5: Unilateral hyperplastic, C6: Bilateral hyperplastic, C7: Asymmetrical hyperplastic. The D2-6 show the anatomical variations of the thyrohyoid membrane and of the body of the hyoid bone in lateral view. D1: Normal anatomy, D2: The uni- or bilateral physiological triticeal cartilage is so common that it’s not considered as an anatomical variation but merely as a variation. D3: Non fusion of the superior horn of the thyroid to the thyroid cartilage, this could easily be mistaken for a fracture. D4: Unilateral hypoplastic superior horn of the thyroid cartilage. D5: Uni- or bilateral (P van Driessche, personal communication) absence of the superior horn of the thyroid cartilage. D6: An articulating connection between the greater horn of the hyoid bone and the superior horn of the thyroid cartilage. D7: The same as in D6 but with a triticeal cartilage interposed between the two horns. D8: Rare case with a nearly circumferential ankylosed hyoid bone (caudal view) (Klovning and Yursik 2007). The E row shows a lateral view on the anatomical variations of the stylohyoid complex and the stylohyoid ligament, the latter was subdivided by Olivier in 1923 in 3 main types (Lykaki and Papadopoulos 1988). E1: Normal anatomy, E2: Elongation of the styloid process (SP); Eagle’s syndrome. E3: A keratohyal (KH) bone in the stylohyoid ligament. E4: Fundamental type with three bones (stylohyal (SH), keratohyal (KH) and hypohyal (HH)). E5: Major type A (stylohyal, keratohyal, keratohyal and hypohyal). E6: Major type B (stylohyal, keratohyal, keratohyal, hypohyal and hypohyal). E7: Restricted type with a fused keratohyal and hypohyal bone, the so called keratohypohyal (KHH) bone.

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Aberrant hyoid apparatus

Dwight stated in 1907 that Marchetti of Padua’s report from 1652 was the earliest reference to the aberrant hyoid apparatus (Dwight 1907), also known as (incompletely or completely) ossified hyoid apparatus (Lykaki and Papadopoulos 1988; Porrath 1969; Vougiouklakis 2006). Reichert noted the anatomical connection between styloid process and hyoid in 1837, and assigned its origin to the second pharyngeal arch cartilage (Reichert 1837). The hyoid apparatus consists of styloid process, stylohyoid ligament and lesser horn of the hyoid (De Paz et al. 2012; Porrath 1969; Vougiouklakis 2006; Wellinger, Dugast, and Desnos 1966). This chain is completely derived from the second pharyngeal arch cartilage (Reichert’s) and can be subdivided into five, or even seven (Wellinger, Dugast, and Desnos 1966), osteocartilaginous elements from the base of the cranium to the hyoid (Table 1) (von Lanz and Wachsmuth 1955; De Paz et al. 2012; Dwight 1907; Eagle 1958; Harburger 1925; Lesoine 1966a; Marcucci 1959; Petrovic et al. 2008; Vougiouklakis 2006; Wellinger, Dugast, and Desnos 1966). In 1923 Olivier designated the (partly) ossified hyoid apparatus into three main types, depending on the number of bones in the trajectory of the hyoid apparatus (Table 1) (Lykaki and Papadopoulos 1988).

Partial ossification of the hyoid apparatus is not uncommon but the appearance of a complete bony chain is rare in humans (Vougiouklakis 2006). This condition is usually bilateral where both sides can differ in symmetry (Dwight 1907), but it also occurs unilaterally (Dwight 1907; Porrath 1969; Vougiouklakis 2006). This chain passes between internal and external carotid arteries (Dwight 1907). There is usually some movement possible by a joint or a ligamentous connection between different parts of the chain or at least between the ossified chain and hyoid body (Dwight 1907).

Table 1. Terminology concerning the (ossified) hyoid apparatus, as described by Olivier

<table>
<thead>
<tr>
<th>Terminology concerning the hyoid apparatus from cranial to caudal</th>
<th>Fig.</th>
<th>% cases</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Tympanohyal</strong></td>
<td>Intrapetrosic part of the styloid process</td>
<td></td>
</tr>
<tr>
<td><strong>Stylohyal</strong></td>
<td>Styloid process</td>
<td></td>
</tr>
<tr>
<td><strong>Keratohyal</strong></td>
<td>Stylohyoid ligament</td>
<td></td>
</tr>
<tr>
<td><strong>Accessory Keratohyal</strong></td>
<td>Stylohyoid ligament</td>
<td></td>
</tr>
<tr>
<td><strong>Hypohyal</strong></td>
<td>Lesser horn hyoid</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Three types of ossified hyoid apparatus described by Olivier</th>
<th>Fig.</th>
<th>% cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>The fundamental type with 3 bones &gt; stylohyal, keratohyal and hypohyal</td>
<td>2E4</td>
<td>64%</td>
</tr>
<tr>
<td>The mayor type A with 4 bones &gt; stylohyal, keratohyal, accessory keratohyal and hypohyal</td>
<td>2E5</td>
<td></td>
</tr>
<tr>
<td>The mayor type B with 5 bones &gt; stylohyal, keratohyal, accessory keratohyal, accessory hypohyal and hypohyal</td>
<td>2E6</td>
<td>12%</td>
</tr>
<tr>
<td>The restricted type with 2 bones &gt; stylohyal and the keratohypohyal ( = fused keratohyals and hypohyal)</td>
<td>2E7</td>
<td>24%</td>
</tr>
</tbody>
</table>

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The aberrant hyoid apparatus is hypothesized to originate from persisting second pharyngeal arch cartilage that continued to grow and gradually ossified into a bony chain (Dwight 1907; Wellinger, Dugast, and Desnos 1966). The joints in the chain often show some degree of bone clubbing, which implies a continuation of growth (Dwight 1907).

Symptoms include difficulty in swallowing (Dwight 1907) and restriction of neck movement (Lykaki and Papadopoulos 1988), but they rarely occur before the age of 40 because of the age dependent ossification of the cartilaginous bar (Vougiouklakis 2006). However, striking examples have also been seen in children (Porrath 1969). Associated compressive pathologies (De Paz et al. 2012), like glossopharyngeal neuralgia (Graf 1959) or referred pain due to irritation of the sensory nerve branches (Frommer 1974) have been noted. Also, arterial anomalies in the affected region are not uncommon (Vougiouklakis 2006).

Eagle’s syndrome

The stylohyoid syndrome, or Eagle’s syndrome (Fig. 2E2), describes a collection of clinical symptoms related to anomalies in size and location of the styloid process, which disturbs surrounding anatomical structures (Chi and Harkness 1999; De Paz et al. 2012; de Santana Jr et al. 2006; Eagle 1948, 1949, 1958, 1962; Lesoine 1966a; Petrovic et al. 2008; Porrath 1969; Shul’ga, Zaitsev, and Zaitseva 2006). This condition may be uni- or bilateral and varies in severity (Eagle 1949, 1958). The styloid process consists of the tympanohyal and stylohyal part (Table 1) (Dwight 1907; Eagle 1958). This cylindrical, needle shaped bone, with a cartilaginous tip that normally lies between the internal and external carotid artery, projects ventrocaudally from the inferior side of the petrous bone (De Paz et al. 2012; de Santana Jr et al. 2006; Eagle 1949, 1958) and provides an anchorage for the stylopharyngeus, stylohyoid and styloglossus muscles (De Paz et al. 2012). During normal development, the cranial part of the second pharyngeal arch cartilage ossifies and forms the styloid process, which is connected to the lesser horn of the hyoid through the stylohyoid ligament (Reichert 1837). There is no agreement on normal length of the styloid process (Shul’ga, Zaitsev, and Zaitseva 2006). It is described as 20-30 mm (De Paz et al. 2012; de Santana Jr et al. 2006; Eagle 1948, 1949, 1958; Wellinger, Dugast, and Desnos 1966), 30-35 mm (Shul’ga, Zaitsev, and Zaitseva 2006) or even 45 mm (Jung et al. 2004). The length might be age dependent since an elongated process is more often observed in patients of 30 years and older (De Paz et al. 2012; de Santana Jr et al. 2006; Eagle 1949; Jung et al. 2004) though it has recently also been described in a 9-year-old boy (Garriz-Luis et al. 2017).

The otolaryngologist Watt Eagle described two clinical presentations of stylohyoid syndrome (Eagle 1948, 1949, 1958, 1962). First the more common classic type, which is characterized by foreign body sensation in the throat (de Santana Jr et al. 2006; Eagle 1948, 1949; Lesoine 1966a; Shul’ga, Zaitsev, and Zaitseva 2006) and dysphagia (de Santana Jr et al. 2006; Lesoine 1966a; Shul’ga, Zaitsev, and Zaitseva 2006). The recurrent dull and not sharply localized facial and cervical pain (Chi and Harkness 1999; de Santana Jr et al. 2006; Eagle 1948, 1949; Petrovic et al. 2008; Porrath 1969; Shul’ga, Zaitsev, and Zaitseva 2006), radiates towards temporomandibular joint (Eagle 1962; Shul’ga, Zaitsev, and Zaitseva 2006), mandible (Shul’ga, Zaitsev, and Zaitseva 2006), maxillar or mandibular teeth (Shul’ga, Zaitsev, and Zaitseva 2006), ear (De Paz et al. 2012; de Santana Jr et al. 2006; Eagle 1948; Porrath 1969; Shul’ga, Zaitsev, and Zaitseva 2006), mastoid region (de Santana Jr et al. 2006), neck (De Paz et al. 2012; Shul’ga, Zaitsev, and Zaitseva 2006), tongue (De Paz et al. 2012), and throat. Pain usually increases toward the end of the day, with turning of the head and after long speaking or singing (Shimada and Gasser 1988; Shul’ga, Zaitsev, and Zaitseva 2006). Eagle also included all cases with distortion of
nerve function by the elongated styloid process, involving sensory and motor fibers of the 5\textsuperscript{th}, 7\textsuperscript{th}, 9\textsuperscript{th} and 10\textsuperscript{th} cranial nerves (Eagle 1948; Lesoine 1966a). Patients can suffer from increased salivation (de Santana Jr et al. 2006; Eagle 1948), a distorted sensation of taste (Eagle 1948), esophageal and pharyngeal spasms and recurrent coughing (Eagle 1948; Lavallee and Turcotte 1984). The above described symptoms generally occur immediately after tonsillectomy and Eagle believed that the cause of the symptoms was the scar tissue formation, stretching the nerve endings (Eagle 1948, 1949; Petrovic et al. 2008).

In the second clinical presentation of stylohyoid syndrome, the \textit{stylo-carotid syndrome}, symptoms are found along the distribution of the internal or external carotid artery, due to impingement on the vessel. The styloid process affects the circulation of the carotid arteries and induces irritation of sympathetic nerves in their arterial sheaths (Eagle 1949; Lesoine 1966a). With an affected internal carotid artery, patients will complain of parietal headaches and pains in the distribution area of the ophthalmic artery (Eagle 1948, 1949; Lesoine 1966a; Petrovic et al. 2008). The elongated styloid process can push the internal carotid artery laterally, which may be painful on palpation (Eagle 1948). When the external carotid artery is affected, pain will be referred to the temple and infraorbital region (Eagle 1949; Lesoine 1966a). There is even a hypothesis that tinnitus can be caused by stylo-carotid syndrome. This could be explained because pulsating waves from the artery are conducted through the elongated styloid process towards skull and cochlea (Eagle 1948; Lesoine 1966a).

A vegetative syndrome, including pallor, sweating, hypotension and even brief loss-of-consciousness episodes, due to irritation of the carotid perivascular plexus or carotid body by the elongated styloid process, has also been reported (De Paz et al. 2012; de Santana Jr et al. 2006; Lesoine 1966a; Warot 1976). Wilmoth and Leger described this phenomenon as ‘Syncope styloidea’, which occurred in patients with a combination of a high bifurcation of the common carotid artery and an elongated styloid process (Lesoine 1966a).

The elongated styloid process can be palpated in the tonsillar fossa during clinical examination (de Santana Jr et al. 2006; Eagle 1948). For radiological imaging, the computed tomography (CT) preferably with 3D reconstruction is the modality of choice (de Santana Jr et al. 2006; Petrovic et al. 2008). Sagittal CT-angiography can be useful in diagnosing stylo-carotid syndrome.

Differential diagnostic considerations for Eagle’s syndrome are numerous cranio-facio-cervical pain syndromes (De Paz et al. 2012; Petrovic et al. 2008), e.g. neuralgias of the glossopharyngeal nerve, trigeminal nerve (De Paz et al. 2012) and pterygopalatine ganglion (Eagle 1949), temporomandibular disorders (Eagle 1949; Zaki et al. 1996), dental problems (De Paz et al. 2012; Eagle 1949), cervical arthropathies or pharyngeal infections and tumors (De Paz et al. 2012). Therapy consists of conservative management with anti-inflammatory drugs and analgesics, or transoral surgical resection of the styloid apophysis (de Santana Jr et al. 2006; Eagle 1948; Lesoine 1966a; Petrovic et al. 2008), an operation that has been performed since 1872 (Eagle 1958).
Materials and Methods

**Radiology**
Two-hundred eighty-four excised hyoid-larynx complexes were radiologically examined and collected in a forensic-radiological database between 2002 and 2013 (de Bakker et al. 2016). Age ranged from 0 to 98 years (mean: 44 years), male-female ratio was 1:1. Radiological examination of the excised complexes in 8 directions started in the early 2000s with the use of a mammograph and was then continued by radiography in 8 directions with a digital bucky and CT of the excised complex. This combination became the gold standard. In later phases of the study a whole-body CT was often performed before autopsy, in addition to radiological examination of the excised complex with bucky and CT. The complex could then be virtually extracted from the whole-body CT dataset: see de Bakker et al (2016) for protocols (de Bakker et al. 2016). An independent researcher together with an experienced radiologist scored all radiological cases for deviations from standard anatomy as shown in figure 1.

**Vrolik specimens**
In addition to the radiological cases, we studied some profound cases with anatomical variants from the anatomical museum of the Academic Medical Center of the university of Amsterdam, *Museum Vrolik* (de Rooy and van den Bogaard 2010). Images of these cases served as illustration of rare variants.

**Results**
Radiological examination of the 284 excised hyoid-larynx complexes showed that only 37% met the anatomical standard (Fig. 1). A remarkable 63% of the 284 cases showed various degrees of anatomical variants. Two variations were most observed: the age-dependent uni- or bilateral ankylosis of the hyoid body with the greater horns (n=33 and 70 respectively) and uni- or bilateral presence of triticeal cartilages in the lateral thyrohyoid ligament (n=11 and 12 respectively) (Table 2). These minor variations do not have clinical implications. Nineteen percent of this sample of 284 excised complexes, however, portrayed relevant anatomical variants (Table 2, last column).

All variations found in this study (Table 2) supplemented with rare cases described in literature and from *Museum Vrolik* were summarized in figure 2, in an attempt to provide an overview of all currently known deviations from normal anatomy of the hyoid-larynx complex. Moreover, 27 out of 33 cases with unilateral fusion between hyoid body and greater horn from which laterality and sex were known, were tabulated in table 3. Right sided fusion was observed more frequently (n=17) than left sided fusion (n=10) in both sexes.
Table 2. Overview of hyoidal and stylohyoidal variations found in 284 forensic radiological hyoid-larynx scans

<table>
<thead>
<tr>
<th>Variation</th>
<th>Panel figure 2</th>
<th>Cases</th>
<th>Percentage of 284</th>
<th>Corrected %*</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Hyoid body</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bilateral ankylosis body with greater horns</td>
<td>A2/A3</td>
<td>70</td>
<td>24.6</td>
<td></td>
</tr>
<tr>
<td>Unilateral ankylosis body with greater horns</td>
<td>A4/A5</td>
<td>33</td>
<td>11.6</td>
<td></td>
</tr>
<tr>
<td>Unilateral ankylosis greater and lesser horn</td>
<td>A6</td>
<td>2</td>
<td>0.7</td>
<td>0.7</td>
</tr>
<tr>
<td>Exostosis median process</td>
<td>A7</td>
<td>7</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td><strong>Subtotal</strong></td>
<td></td>
<td>112</td>
<td>39.4</td>
<td>3.2</td>
</tr>
<tr>
<td><strong>Hyoid greater horn</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypoplastic on one side</td>
<td>B2</td>
<td>1</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Intermittent, ankylosis body and greater horn</td>
<td>B3</td>
<td>1</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Exostosis</td>
<td>B4</td>
<td>1</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Curved upwards</td>
<td>B5</td>
<td>2</td>
<td>0.7</td>
<td>0.7</td>
</tr>
<tr>
<td>Curved downward</td>
<td>B6</td>
<td>1</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Accessory bone</td>
<td>B7</td>
<td>3</td>
<td>1.1</td>
<td>1.1</td>
</tr>
<tr>
<td>Articulates with superior horn</td>
<td>D6</td>
<td>1</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Articulates with triticeal and superior horn</td>
<td>D7</td>
<td>1</td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td><strong>Subtotal</strong></td>
<td></td>
<td>11</td>
<td>3.9</td>
<td>3.9</td>
</tr>
<tr>
<td><strong>Hyoid lesser horn</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unilateral absence</td>
<td>C2</td>
<td>9</td>
<td>3.2</td>
<td>3.2</td>
</tr>
<tr>
<td>Bilateral absence</td>
<td>C3</td>
<td>7</td>
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<td>2.5</td>
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<tr>
<td>Hypoplastic on both sides</td>
<td>C4</td>
<td>1</td>
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<tr>
<td>Unilateral hyperplastic</td>
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</tr>
<tr>
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<td>C6</td>
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<td>C7</td>
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<td></td>
<td>27</td>
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<td>9.5</td>
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<tr>
<td><strong>Thyroid superior horn</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unilateral triticeal cartilage</td>
<td>D2</td>
<td>11</td>
<td>3.9</td>
<td></td>
</tr>
<tr>
<td>Bilateral triticeal cartilage</td>
<td>D2</td>
<td>12</td>
<td>4.2</td>
<td></td>
</tr>
<tr>
<td>Non fusion between superior horn and thyroid</td>
<td>D3</td>
<td>2</td>
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</tr>
<tr>
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<td>D5</td>
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<tr>
<td><strong>Subtotal</strong></td>
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<td>1.4</td>
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<tr>
<td><strong>Stylohyoid complex</strong></td>
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<td>Fundamental type</td>
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<tr>
<td><strong>Subtotal</strong></td>
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<td>2</td>
<td>0.7</td>
<td>0.7</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td></td>
<td>63</td>
<td>18.7</td>
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* The corrected percentage of cases comprises only the relevant anatomical variants, without the uni- or bilateral ankyloses of the greater horns and the uni- or bilateral presence of triticeal cartilages that do not have clinical implications.
Discussion

Two thirds of the examined hyoid-larynx complexes deviated from the anatomical standard (Fig. 1). This was mostly due to minor variations like age-dependent ankylosis of the hyoid and presence of triticeal cartilages. Almost one fifth of the cases comprised more striking anatomical variants. Some of these variants have not been described in literature before. Note, however, that this sample of 284 cases may be a biased sample, because it is based on the suspicion of the pathologist of a fracture in the hyoid-larynx complex in a forensic context. Also, we examined explanted complexes and therefore elongation of the styloid process could not be determined.

Embryonic etiology of anatomical variants

When comparing the most profound deviant cases with 3D reconstructions of embryonic development (de Bakker et al. submitted to The Laryngoscope), we noted how their etiology could be explained by embryonic development (Fig. 3).

The median process

A median process of the hyoid body (n=7) can be explained by the body’s embryological origin (Fig. 3A). The hyoid bone anlage (Dwight 1907; Rodriguez-Vazquez et al. 2011), a cylindrical shaped growth center ventrally positioned along the cranio-caudal axis in-between the left and right-sided bar of the second pharyngeal arch cartilage, marks the first appearance of the hyoid body (de Bakker et al. submitted to The Laryngoscope). We hypothesize that the median process is a remnant of this cylindrical shaped anlage.

Second pharyngeal arch cartilage anomalies

Elongation of lesser horns (Fig. 3B), styloid process (Fig. 3E2) and ossification of the hyoid apparatus (Fig. 3C) can all be explained by a degree of stylohyoid ligament ossification. The second pharyngeal arch cartilage persisted as cartilaginous bar in this trajectory after which it ossified partially (Fig. 3B1 and E2) or completely (Fig. 3C1). Up until now, Eagle’s syndrome (Fig. 2E2) and the aberrant hyoid apparatus (Fig. 2E3-7) are traditionally discussed in literature as two separate entities, leading to much confusion concerning definitions and clinical presentation. Considering the embryonic etiology of these syndromes, we propose that they are merely two expressions of a broad spectrum, all due to the partial or complete persistence of the second pharyngeal arch cartilage. These anomalies are all found in the trajectory between the lesser horn of the hyoid and the styloid process. Therefore, they should be considered as one entity, preferably referred to as ‘second pharyngeal arch cartilage anomalies’.

Third pharyngeal arch cartilage anomalies (congenital hyothyroid bar)

The 3D reconstructions of the embryological development of the hyoid-larynx complex show a clear connection between the dorsal part of the bars of the third pharyngeal arch cartilages and the superior horns of the thyroid, derived from condensed mesenchymal tissue. This profound connection will become the lateral thyrohyoid ligament. Figures 3D1, E1 and F1 all show various degrees of a persisting connection between greater and superior horns. The most common variation in this trajectory is presence of a triticeal cartilage (n=11 unilateral and n=12 bilateral). In figure 3D1 the left greater horn describes a 90-degree angle pointing caudal towards superior horn. In figure 3E1 an accessory bone articulates between the elongated superior and greater horns. In figure 3F1 the greater horn articulates directly with the
ossified superior horn. These cases can be explained by ossification of persisting embryological cartilaginous components in the trajectory of the lateral thyrohyoid ligament and is therefore referred to as congénital hyothyroid bar (Porrath 1969).

One case of duplication of the greater horns has been reported (Gok, Kafa, and Fedakar 2012). The hyoid would miss the lesser horns and shows a duplication of the greater horns. We suggest that these duplicated horns, in fact, are more likely to be elongated lesser horns (Fig. 2C6-7).

Clinical relevance

Symptoms of variants in the hyoid-larynx complex are often not recognized by clinicians (Shul’ga, Zaitsev, and Zaitseva 2006). Great care should be taken in situations of tracheal intubation in these patients, because of the risk of regurgitation and aspiration (Ames and McNiellis 1998; Aris, Elegbe, and Krishna 1992; Brimacombe et al. 2004). Anomalies of this complex are of great importance for radiological examination and surgery of the neck region, but they should also be known by forensic experts, anthropologists, anatomists and dentists (De Paz et al. 2012; Gok, Kafa, and Fedakar 2012) to appreciate the clinical impact of these variants and to avoid judicial errors in cases of assumed strangulation or blunt neck trauma.

Forensic relevance

Fractures in the hyoid-larynx complex are one of the best indicators of strangulation (Gok, Kafa, and Fedakar 2012; Kindschuh, Dupras, and Cowgill 2010; Urbanova et al. 2013; Saukko 2015), but they can also be caused by for instance hanging, traffic accidents, osteoporosis senilis, sporting accidents and after intubation (D’Souza, Harish, and Kiran 2010; Dunsby and Davison 2011; Kleinberg 1934; Leseine 1966b; Marcucci 1959; Porr, Laframboise, and Kazemi 2012; Soerdjbalie-Maikoe and van Rijn 2008; Ubelaker 1992; Saukko 2015). Due to the considerable frequency of anatomical variations of the hyoid-larynx complex, great care should be taken when diagnosing traumatic lesions of this complex (Di et al. 2004; Gok, Kafa, and Fedakar 2012; Saukko 2015).

In the normal process of ageing, the triticeal cartilage (Fig. 2D2) in the lateral thyrohyoid ligament often calcifies, which enables radiographic detection. However, when this cartilage is intensely and inhomogeneous calcified, it should not be mistaken for an avulsed fracture of the upper horn of the thyroid cartilage (Khokhlov 1997; Porrath 1969). Therefore, oblique radiographs should be used for further radiological examination, to distinguish a fracture from an inhomogeneously calcified cartilage (Di et al. 2004).

Most fractures are found in the upper thyroid horns (Saukko 2015). Fracturing of the hyoid occurs mainly between greater horns and body, when ankylosis is incomplete, or in the posterior part of the greater horn (Leseine 1966b; Pollanen, Bulger, and Chiasson 1995; Soerdjbalie-Maikoe and van Rijn 2008; Saukko 2015). Since ankylosis of the hyoid joints is age dependent, fractures occur more frequent in persons aged above 30 (D’Souza, Harish, and Kiran 2010; Gok, Kafa, and Fedakar 2012; Gupta et al. 2008; Hänsch 1977; Leksan et al. 2005; Miller, Walker, and O’Halloran 1998; Pollanen and Chiasson 1996; Porrath 1969; Soerdjbalie-Maikoe and van Rijn 2008; Ubelaker 1992; Saukko 2015). D’Souza even stated that when a victim is over 38 years, clinicians and forensic experts can expect a fractured hyoid, after pressure on the neck (D’Souza, Harish, and Kiran 2010). Joint luxation between hyoid body and greater horn has also been reported in cases of strangulation (Lesoin 1966b).
Table 3. Number of cases with left or right sided fusion of the hyoid body with the greater horn

<table>
<thead>
<tr>
<th></th>
<th>Male</th>
<th>Female</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Left sided fusion</td>
<td>8</td>
<td>2</td>
<td>10</td>
</tr>
<tr>
<td>Right sided fusion</td>
<td>12</td>
<td>5</td>
<td>17</td>
</tr>
<tr>
<td>Total</td>
<td>20</td>
<td>7</td>
<td>27</td>
</tr>
</tbody>
</table>

Fig. 3. Anatomical variants of hyoid explained by embryological development. Overview of six of the most profound anatomical variants on the left sides, compared with the embryonic development of that part of the hyoid-larynx complex on the right sides (de Bakker et al. submitted to The Laryngoscope). The hyoid bone (anlage) is shown in purple, second pharyngeal arch cartilage derivatives are shown in blue, third pharyngeal arch cartilage derivatives are shown in yellow and the thyroid cartilages are shown in red. The ‘L’ indicates the left side of the patient. Each arrow indicates the location of the variant. The shown variants are: conventional radiograph of an exostosis of the hyoid body (A1)(Fig. 2A7), conventional radiograph of an elongation of both lesser horns (B1)(Fig. 2C7), dried specimen of an ossified hyoid apparatus; the fundamental type (C1)* (Fig. 2E4), conventional radiograph of the left greater horn curved downward (D1)(Fig. 2B6), conventional radiograph of a bony connection between the greater and the superior horn, i.e. the congenital hypothyroid bar (E1 & F1)(Fig. 2D7 & D6).

*On display in Museum Vrolik. Collection Louis Bolk, 1912. Photo by Sanne Mos & Marco de Marco; courtesy of Museum Vrolik, Academic Medical Center, Amsterdam. With permission.
Some anatomical variants resemble fractures. All examples in figure 2 should be kept in mind when the hyoid-larynx complex is examined during medico-legal examination but especially examples B3, B7, D2, D3, D5 and D6 should always be considered when a suspected fracture is found in those locations. When the difference between variant and fracture remains ambiguous after autopsy and radiological examination, histological examination of the affected part should be performed.

Conclusion

We provided an overview of the known anatomical variants of the hyoid-larynx complex, with relevance for clinicians and forensic experts. Etiology of the variants has been declared by their development. Since the aberrant hyoid apparatus and Eagle’s syndrome are often intermingled in literature as they are both explained by persistence of second pharyngeal arch cartilage, we propose to refer to them as ‘second pharyngeal arch cartilage anomalies’.

Acknowledgments

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Variants of the hyoid-larynx complex with forensic implications and consequence for Eagle’s syndrome

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