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Development of the chondrocranium in the domesticated fowl (*Gallus gallus* f. *domestica*), with a study on the variation of the of hypoglossal foramina

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Abstract

We critically review diverse literature on the cranial development in chicken, aiming at identifying potential areas of research in chondrocranial diversity that may relate to adult variation. Independent of the method used, there is general agreement regarding the onset of most chondrocranial elements. Inconsistencies in the literature concern the chondrification of specific elements as separate cartilage or continuous outgrowth of the preexisting cartilaginous element, the origin of fenestrations, the presence of certain cartilaginous structures (e.g., intertrabecular, cranial ribs), and the number of hypoglossal foramina. Intraspecific variation in the number of hypoglossal foramina in the red junglefowl and chicken is common, and known for other birds. The number of hypoglossal foramina lateral to the exoccipital bone varies between one and four in adult skulls of 75 individuals. To test for differences in chondrocranial shape between the fowl and chicken, timed stages of different breeds need to be investigated.

Key words

Introduction

The domestication of the fowl, *Gallus gallus,* led to a large variation of morphological traits in the vast number of chicken breeds. This variation has been documented for example in the adult bony skull (Stange *et al*., 2018). Less is known about the embryonic chondrocranium, and the variation of its elements among different chicken breeds. The chondrocranium of the red junglefowl (the wild form) has not been described, but there are reports on single specimens (Tonkoff, 1900) and ontogenetic series of some chicken (Parker, 1869; Sonies, 1907; Vorster, 1989; Heyns, 1997). Difficulties in comparing these reports are expected due to different methods used, variation in the stages studied, intraspecific variability, and missing information on the breeds studied. In this paper, we provide a summary and critical assessment of this literature.

A review of previous works

The development of the chondrocranium in chicken

The first anlage of the chondrocranium, the base of the cartilaginous skull, can be divided in a prechordal part (trabecles) and postchordal part (acrochordal and parachardal cartilage): this division is established while the
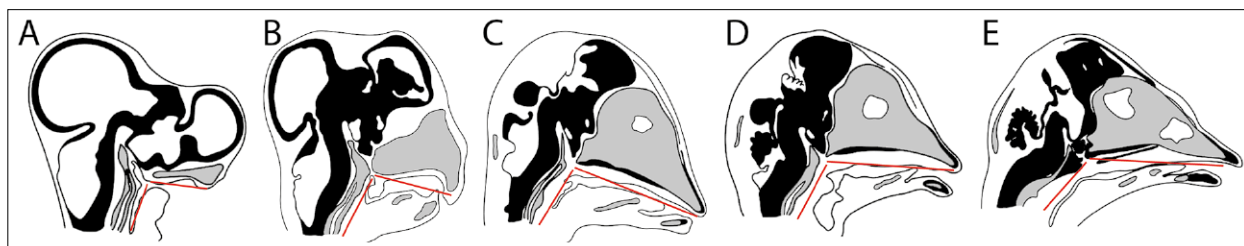
Fig. 1. Reduction of the angle between the pre- and postchordal region in an ontogenetic series of *Gallus gallus*: A) head length (HL) 2.5 mm, B) HL 7 mm, C) HL 13 mm, D) HL 13.5 mm, and E) HL 17 mm. Notice the relation with the developing brain, in black. Drawings modified from Lang (1952). Not to scale.

brain and the mesencephalic flexure form (TAKAMATSU & FUJITA, 1987; PIKALOW *et al*., 1994). The first indications of the cranial flexure are visible in chicken from 33 hours of incubation on (HAMBURGER & HAMILTON, 1951). The prechordal and postchordal cartilages create an angle that is reduced in ontogeny (Fig. 1) (PARKER, 1869; LANG, 1952; LILLIE, 1952; VORSTER, 1989; HEYNS, 1997), as in vertebrates in general. Fate-mapping studies established that the chondrocranium has a mixed origin; neural crest (prechordal) and mesoderm (postchordal) (COULY *et al*., 1993; WADA *et al*., 2011).

The first chondrifications in the chicken head appear around day five of incubation in the region of the chorda, in the base of the future skull (HUXLEY, 1864; PARKER, 1869; TONKOFF, 1900; SONIES, 1907; DE BEER & BARRINGTON, 1934; BELLAIRS, 1958; BELLAIRS & JENKIN, 1960; LILLIE, 1952; VORSTER, 1989; HEYNS, 1997) (Table 1). Between day six and eight (Fig. 2A–C, 3A–C), most features of the chondrocranium are cartilaginous (LILLIE, 1952; VORSTER, 1989). Although first ossifications are present between day seven and day ten (ERDMANN; 1940; JOLLIE, 1957; VORSTER, 1989), and the first signs of resorption of the cartilaginous skull appear around day nine, no significant structural changes take place in the chondrocranium until day 14 of incubation (VORSTER, 1989). The latter is the time when some features of the nasal capsule appear that are the last parts of the chondrocranium developed. Besides the nasal cartilages, little cartilage is left in the adult skull (HEYNS, 1997).

Inconsistencies in the literature on the development of the chondrocranium

Independent versus continuous chondrification of cartilaginous elements

A debated aspect of chondrocranial development in the literature is whether certain structures of *Gallus gallus* f. *domestica* develop in continuity with each other or independently. Compared to *Anas platyrhynchos*, another Galloanserae, in *Gallus gallus* f. *domestica* many elements of the chondrocranium develop in continuity to each other. SONIES (1907) assumed that many skeletal parts of the

chicken skull are already associated in the connective tissue or procartilaginous stage. One of the discussed cartilaginous elements in *Gallus gallus* f. *domestica* is the cochlear portion of the otic capsule, which is described in its cartilaginous condition as a constituent part of the basal plate (PARKER, 1869; DE BEER, 1937; ROMANOFF, 1960; VORSTER, 1989; HEYNS, 1997). Likewise, SONIES (1907) mentioned that the cochlear portion of the otic capsule originates in continuation with the parachordal cartilage. His unusual use of the terms "Pars cochlearis (mesotica)" for the cochlear portion of the otic capsule and "Cartilago basiotica (mesotica)" for the anterior part of the basal plate led to a misinterpretation of the development of the cochlear portion of the otic capsule in relation to the basal plate (VORSTER, 1989; ZAHER & ABU-TAIRA, 2013). SONIES (1907) wrote in the paragraph on the Pars cochlearis (mesotica): "Bei 11 m.m. langen *Hühner*embryonen ist der basiotische (mesotische) Abschnitt in knorpeligem Zusammenhang mit der Parachordalplatte entstanden (…).", which means that the cochlear portion develops in cartilaginous connection to the parachordal cartilage. In the summary, he concluded: "Die Cart. Basiotica (…) bildet auch die selbständige Anlage der *Pars cochlearis* der Capsula auditiva", meaning that the anterior part of the basal plate builds the anlage of the cochlear portion independent from the canalicular portion of the auditory capsule. By clarifying this terminological issue, we conclude that there is a general agreement on a continuous development of the cochlear portion with the basal plate in *Gallus gallus* f. *domestica* in the cartilaginous stage. But an independent origin was mentioned by LILLIE (1952) for the mesenchymal condition, and described for the procartilaginous condition by VORSTER (1989). On the contrary, HEYNS (1997) mentioned the procartilaginous cochlear portion fused to the basal plate in the same embryonic stage and breed.

Origin of fenestrations

Contradicting statements in the literature exist on the primary or secondary origin of specific fenestrae. One of these is the fenestra basicranialis posterior. A primary origin of the fenestra basicranialis posterior is reported by SONIES (1907) and BELLAIRS & JENKIN (1960). They stated that the basal plate grows on both sides of the chorda but
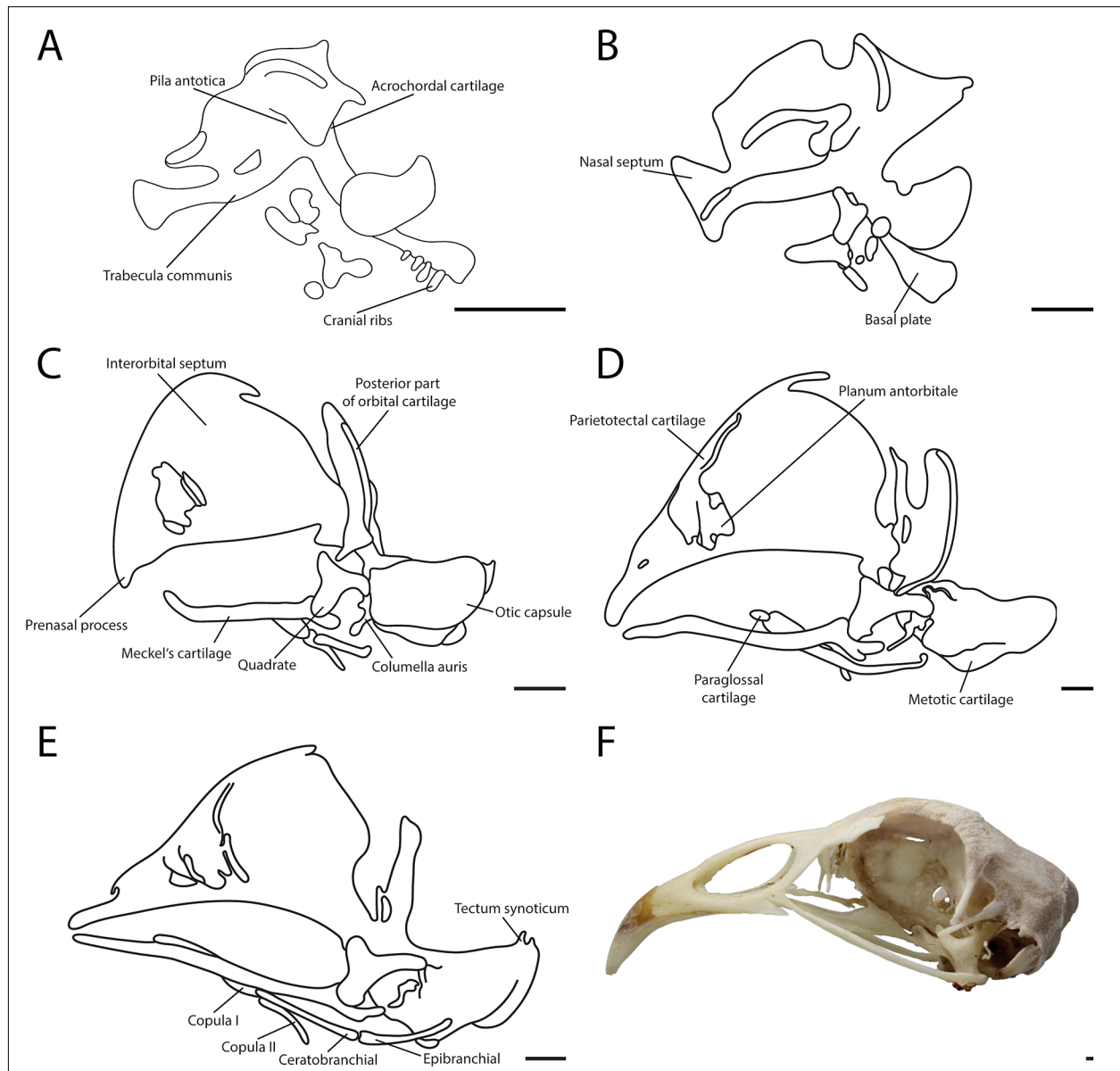
Fig. 2. Lateral view on the developing chondrocranium of the white leghorn chicken (*Gallus gallus* f. *domestica*): A) day 5, B) day 6 – 7, C) day 8, D) day 9, E) day 10 – 11 and F) adult bony skull (A/IV 154). Drawings modified from Vorster (1989). Scale bars equal 1 mm.

not in its anterior region. When the parachordals connect to the acrochordal cartilage, then the fenestra is formed through which the chorda transverses the basal plate. A similar mode of origin is known in other birds (PARKER, 1891; DE BEER & BARRINGTON 1934) and reptiles (YARYHIN & WERNEBURG 2017). Other authors reported a secondary origin through resorption around the chorda in the chicken (PARKER, 1869; TONKOFF, 1900; VORSTER, 1989; HEYNS, 1997), and in other birds (SUSCHKIN 1899; ENGELBRECHT, 1958; ZAHER & RIAD 2009).

Debated existence of particular structures

The intertrabecular is another debated structure. There is general consensus in the literature upon the paired anlage (opposed to single) of the trabeculae in *Gallus gallus* f.

domestica (HUXLEY, 1864; PARKER, 1869; SONIES, 1907; BELLAIRS & JENKIN, 1960; LILLIE, 1952; HEYNS, 1997; WADA *et al.*, 2011), and most authors agree on the origin of the trabecula communis by fusion of the anterior portion of the trabeculae (HUXLEY, 1864; PARKER, 1869; SONIES, 1907; LILLIE, 1952). On the contrary, some authors described the presence of a median bar, the intertrabecular, which fuses subsequently with the trabeculae to form a single structure (BELLAIRS & JENKIN, 1960). These descriptions are in contrast to the observation that there is no sign that the anterior part, the trabecula communis, was paired (VORSTER, 1989; HEYNS, 1997). The observations of an intertrabecular were explained by different degrees of chondrification in the intertrabecular region (BELLAIRS, 1958; VORSTER, 1989; HEYNS, 1997), what is in agreement with the observation that the trabeculae grow together and chondrify in continuity without an independ-
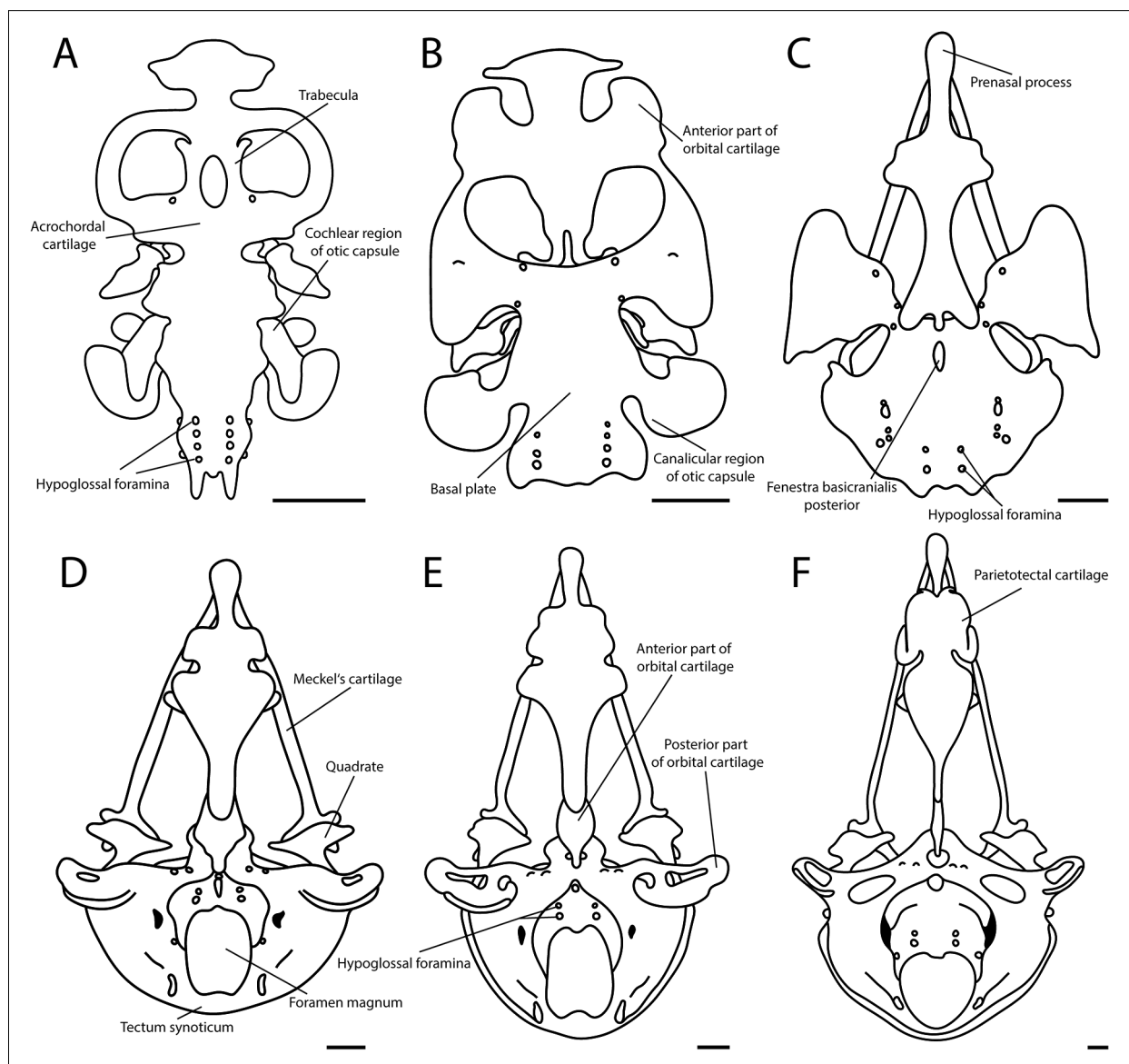
Fig. 3. Dorsal view on the chondrocranium of the white leghorn chicken (*Gallus gallus* f. *domestica*), from left to right: A) day 5, B) day 6–7, C) day 8, D) day 9, E) day 10–11, and F) day 14–16. Drawings modified from Vorster (1989). Scale bars equal 1 mm.

ent anlage of an intertrabecular (SONIES, 1907). A new focus on the developmental origin of the trabecular region revealed that the region is composed of two distinct elements, the trabeculae and the intertrabecular. Both derive from trigeminal neural crest cells that migrate along different pathways (WADA *et al.*, 2011).

One of the few structures of the chondrocranium of *Gallus gallus* f. *domestica*, aside from the intertrabecular, which are questioned to exist by some authors are the cranial ribs. SONIES (1907) did not find any sign of cranial ribs, which he defined as cartilaginous skeletal elements. But transitory mesenchymal cranial ribs (Fig. 2A), present around day five until day eight, are reported by several authors (LANG, 1954; VORSTER, 1989; HEYNS, 1997). Likewise, ROMANOFF (1960) mentioned vestigial ribs lateral and ventral to the basal plate, alternating with the hypoglossal nerve roots.

Material and Methods

The terminology for the chondrocranial elements follows VORSTER (1989). Five authors described chondrification sequences in *Gallus gallus* f. *domestica* (Table 2). Six developmental stages of VORSTER (1989) and HEYNS (1997) served as reference sequences for comparison across the literature. Both authors, as PARKER (1869), worked on serial sections, whereas SONIES (1907) and BELLAIRS (1958) studied serial sections and whole mounts.

The examined 13 adult skulls of the red junglefowl, 55 skulls of 14 chicken breeds, four skulls of a non-specified breed, and 3 skulls of a feral chicken, are part of the collection of the Paläontologisches Institut und Museum Zürich (PIMUZ) (Table 3). The terminology for the hypoglossal foramina follows MAYR (2018).

Table 1. Chondrification sequence in the chondrocranium of *Gallus gallus* f. *domestica* reported in the literature. Sequences in days of incubation (1), body length (2), or in stages (3) according to the original literature.

Chondrocranial characters	Huxley (1864)[1]	Parker (1869)[3]	Tonkoff (1900)[1]	Sonies (1907)[2]	De Beer & Barrington (1934)[1]	Lang (1955)[3]	Bellairs (1958)[1]	Vorster (1989)[3]	Heyns (1997)[3]
acrochordal cartilage		1		5–5.5	4.5		4–5	1	1
resorption acrochordal cartilage				15.5				4	4
parachordal cartilage	6			6–7			4–5		
basal plate		1	10–11	11				1	1
fenestra basicranialis posterior		3		12		B		3	3
trabeculae	6	1		11			6	1	2
procartilaginous trabecula communis								1	1
trabecula communis		1		11–12					
suprapolar cartilage				15				1	2
polar cartilage				11				1	2
procartilaginous Meckel's cartilage				12					1
Meckel's cartilage		1		12–12.5				2	2
Meckel's cartilage fused								3	3
Meckel's as long as prenasal proocess								5	5
procartilaginous quadrate				10–11					1
quadrate	6	1	10–11					2	2
procartilaginous copula I								2	2
copula I		2		13–13.5				3	3
procartilaginous copula II								2	2
copula II		2		15				3	3
procartilaginous ceratobranchial								2	2
ceratobranchial		1		14				3	3
paraglossal cartilage		2		19.5–20				4	4
fusion of paraglossal cartilage		3						5	5
mesenchymatous/procartilaginous epibranchial		1						2	
epibranchial		2		15				3	3
procartilaginous metotic cartilage								2	2
metotic cartilage				16.5–17				3	
procartilaginous pars canalicularis								2	2
pars canalicularis of otic capsule				14				3	3
pars cochlearis of otic capsule	6	1	10–11	11–12				2	2
procartilaginous columella auris		1						2	2
columella auris		3		16				3	3
fenestra ovalis		2		16				3	3
prenasal process		2	10–11	14–15				3	3
resorption prenasal process								6	
nasal capsule		4						6	6
procartilaginous roof of nasal capsule		1		17			5–6	2	2
parietotectal cartilage		2		18–19			7	3	3
cupola anterior								6	6
procartilaginous maxilloturbinal								4	
maxilloturbinal								4	4
procartilaginious atrioturbinal									5
atrioturbinal								5	
procartilaginous nasal septum								2	2
nasal septum		2	10–11	16			7	3	
resorption nasal septum								6	6
fenestra in nasal septum		4	10–11	20			8–10	4	4
fenestra septi nasi				20				4	4
fenestra craniofacialis		4		20				4	4
planum antorbitale		2		20			7		3
procartilaginous interorbital septum								2	2

Table 1 continued.

Chondrocranial characters	Huxley (1864)[1]	Parker (1869)[3]	Tonkoff (1900)[1]	Sonies (1907)[2]	De Beer & Barrington (1934)[1]	Lang (1955)[3]	Bellairs (1958)[1]	Vorster (1989)[3]	Heyns (1997)[3]
interorbital septum		2	10–11	14			8–10	3	
fenestra septalis of interorbital septum		4	10–11	20				6	6
processus tectalis		2	10–11	21			9–10	5	5
procartilaginous pila antotic								1	1
pila antotic				10–12				2	2
resorption pila antotic								4	4
pila antotica spuria								3	3
anterior part of orbital cartilage		2					5?	1	1
planum supraseptale			10–11	21			8		
procartilaginous supraorbital cartilage								1	1
supraorbital cartilage							6		2
posterior part of orbital cartilage		2	10–11	17–18				3	3
vacuity in posterior part of orbital cartilage			10–11					4	4
mesenchymatous cranial ribs								1	1
cranial ribs disappeared								3	3
hypoglossal foramina		1	10–11	12				1	2
reduction number of hypoglossal foramina								3	3
procartilaginous tectum synoticum								3	3
tectum synoticum		2	10–11	21				4	
dermal ossifications		3	10–11					3	3

Results

Chondrification sequence in *Gallus gallus* f. *domestica*

Although the preparation methods differ among the studies – e.g., serial sections versus whole mounts staining (Table 2) – there is a general agreement on the chondrification sequence in the early skull of *Gallus gallus* f. *domestica* (Table 1) (Huxley, 1864; Parker, 1869; Tonkoff, 1900; Sonies, 1907; De Beer & Barrington, 1934; Bellairs, 1958; Bellairs & Jenkin, 1960; Lillie, 1952; Vorster, 1989; Heyns, 1997). Differences in the reported sequence concern three of the traced characters of the chondrocranium. The three features are the suprapolar cartilage (Sonies, 1907), the fenestra basicranialis posterior (Sonies, 1907), and the tectum synoticum (Parker, 1869).

Variation in the number of hypoglossal foramina

In *Gallus gallus* f. *domestica*, the number of reported hypoglossal foramina in embryos is inconsistent (Table 4). Vorster (1989) described variation in ontogeny with four foramina at day five of development and a reduction to two foramina at day eight (Fig. 3A,C). Tonkoff (1900) mentioned two openings on both sides in a chick-en around eleven days of incubation, whereas Parker (1869) depicted two pairs of hypoglossal foramina already in a 4 days old chicken embryo ([Parker (1869)]: plate LXXXI, fig. II) as well as in 10–11 days old embryos ([Parker (1869)]: plate LXXXII, fig. I–III). Likewise, Sonies (1907) described in early stages two pairs of foramina and in later stages a smaller additional foramen occupied by a blood vessel. Vorster (1989) further mentioned intraspecific variation in the number of hypoglossal foramina (Fig. 3B), and Heyns (1997) described in a 6 days old chicken embryo individual variation in the symmetry of the hypoglossal foramina with three foramina on one, and four on the other side (Table 4). Later in ontogeny, two hypoglossal foramina are described in the chondrocranium (Table 4) (Parker (1869); Tonkoff, 1900; Sonies, 1907; Lillie, 1952; Vorster, 1989; Heyns, 1997). Erdmann (1940) mentioned two foramina in two different chicken breeds around day 11 and day 14 of incubation, in which the ossification of the exoccipital bone only started, and Jollie (1957) figured two foramina in the exoccipital bone in a 2–3 day old chick, whereas Parker (1869) depicted two foramina in an individual after hatching ([Parker (1869)]: plate LXXXVII, fig. II, VII; plate LXXXV, fig. IV, VII–VIII, X), or three hypoglossal foramina ([Parker (1869)]: Plate LXXXVII, fig. V) in the bony chicken skull.

In a survey of hypoglossal foramina in the adult skulls of birds, Mayr (2018) reported three foramina (on each side) for Galliformes, including *Gallus gallus*. Bubien-Waluszewska (1968) stated that in *Gallus gallus*

Table 2. Literature on the chondrocranial development in *Gallus gallus f. domestic*a.

Author	Species name (used by author)	Material	Section thickness	Staining method	Number/age range of stages
Huxley (1864)*	chick				
Parker (1869)	common fowl (*Gallus domesticus*)	sections			10/4 days – adult
Tonkoff (1900)	Hühnchen	sections	25 μ		1/10 days 18 hours
Sonies (1907)	Hühnchen (*Gallus*)	sections & whole mounts	15–20μ	Haemalaun, Acid fuchsine & methylene blue & Methylene blue (van Wijhe)	5/4– 10 days
De Beer & Barrington (1934)*	chick	sections & whole mounts		Victoria blue (van Wijhe)	
Lang (1955)	*Gallus gallus*	sections	14μ, 16μ, 31μ		2/-
Bellairs (1958)	chick	sections & whole mounts		Haematoxylin, Eosin, Masson's trichome technique & Methylene blue (van Wijhe)	–/4 – 10 days (sections) & 7–10 days (whole mounts)
Lillie (1952)*	chick				
Vorster (1989)	White leghorn (*Gallus gallus*)	sections	10μ, 12μ	Haemalaun (Mayer's)/ Haematoxylin (Haidenhain's iron) & Bismarck brown & Eosin, Azocarmine & azan	6/4–18 days
Heyns (1997)	White leghorn (*Gallus gallus*)	sections	10μ	Haematoxylin (Ehrlich's), Bismarck brown, Eosin	6/4–18 days

* no information on material

f. *domestica* the two branches of the hypoglossal nerve (faisceau dorsal and f. ventral) leave the skull by two hypoglossal foramina, and the author depicted two foramina in a specimen of a chicken ([Bubien-Waluszewska (1981)]: fig 7.11.). In different chicken breeds observed in our study, the number of hypoglossal foramina in the skull varied between one and four (Fig. 4; Table 2), and there is bilateral asymmetry in the count of hypoglossal foramina in 24 of 75 skulls (Fig. 5).

The hypoglossal foramina differ in size, shape, and arrangement. In all the studied breeds the caudal foramina (h1 and h2) are larger than the rostral ones (h3 and h4) (Fig. 4), which is the case in embryos too (Vorster, 1989; Heyns, 1997). In the case where specimens have only one caudal foramen, the opening is much larger (Fig. 6B) than each of the two separate foramina otherwise present (Fig. 6A). In some skulls, within the large foramen, the incomplete separation of two smaller foramina is visible, what seems an incomplete separation of the two caudal foramina through their development (Rice, 1920; Mayr, 2018). In five of 14 breeds, the nonspecified breed, and in the red junglefowl, the variation with only one caudal foramen on one or both sides can be found. Likewise, the pattern of the foramina differs among the chicken and the fowl. In some, the hypoglossal foramina are arranged in a line (Fig. 4C), whereas in others the pattern is square-like (Fig. 4B) or triangular (Fig. 4A). The shape of the foramina is less diverse than their number or arrangement. Solely the non-specified breed (Fig. 4D) has slit-like foramina, whereas in the other chicken they are round.

Discussion

Although in the literature the description of the developmental sequence of the chondrocranium in chicken is in general consistent (Huxley, 1864; Parker, 1869; Sonies, 1907; Bellairs, 1958; Vorster, 1989; Heyns, 1997), variation (and potentially heterochronic shifts) in the chondrification sequence and variation in the count of hypoglossal foramina do exist.

Variation in the chondrification sequence

The suprapolar cartilage is a small cartilage continuous with the polar cartilage, which chondrifies at the same time as the latter. Due to the continuous chondrification of the polar cartilage with the trabeculae, the first sign of cartilage appears early in the chondrocranium (Vorster, 1989; Heyns, 1997). On the contrary, Sonies (1907) described the appearance of the suprapolar cartilage as a distinct element separate from the polar cartilage. It chondrifies rather late in the chondrification sequence together with the elements of the first branchial arch. This change in the sequence could be an issue of the method used (whole mount by van Wijhe) resulting in cartilages being recorded later than when using histology (De Beer, 1930). Cartilages of the chondrocranium do not develop as homogenous structures, and regions with different cartilaginous stages can be present (Yaryhin & Werneburg 2017). Histology allows the recognition of cartilages

Table 3. Variation in the number of hypoglossal foramina in the skull of the red junglefowl and chicken. Foramina are numbered from medial to lateral and from caudal to rostral.

Breed	Collection number	Left side				Right side				Total no. of fora-mina
		h1	h2	h3	h4	h1	h2	h3	h4	
Red junglefowl	A/IV 158	1	0	1	0	1	0	1	0	4
	A/IV 159	1	1	1	1	1	1	1	0	7
	A/IV 160	1	0	0	0	1	0	0	0	2
	A/IV 194	1	1	1	0	1	1	1	1	7
	A/IV 195	1	0	1	0	1	0	1	0	4
	A/IV 196	1	0	1	1	1	0	1	0	5
	A/IV 197	1	0	1	0	1	0	1	0	4
	A/IV 198	1	0	1	0	1	0	1	0	4
	A/IV 199	1	1	1	0	1	1	1	0	6
	A/IV 200	1	1	1	0	1	0	1	0	5
	A/IV 201	1	0	1	0	1	0	0	0	3
	A/IV 202	1	1	1	0	1	1	1	0	6
	A/IV 203	1	1	1	0	1	0	1	0	5
Appenzeller Barthuhn	A/IV 211	1	0	1	0	1	0	1	0	4
	A/IV 212	1	1	1	0	1	1	1	0	6
	A/IV 213	1	1	1	1	1	1	1	1	8
	A/IV 214	1	0	1	1	1	1	1	1	7
	A/IV 175	1	1	1	1	1	1	1	1	8
	A/IV 176	1	0	1	0	1	1	1	0	5
	A/IV 177	1	1	1	0	1	1	1	0	6
	A/IV 188	1	1	1	1	1	1	1	1	8
Appenzeller Spitzhaubenhuhn	A/IV 204	1	0	1	1	1	0	1	0	5
	A/IV 205	1	1	1	0	1	0	1	1	6
	A/IV 206	1	1	1	0	1	1	1	0	6
	A/IV 207	1	1	1	0	1	1	1	0	6
	A/IV 208	1	0	1	0	1	1	1	1	6
	A/IV 209	1	1	0	0	1	1	1	0	5
	A/IV 210	1	1	0	0	1	1	0	0	4
	A/IV 151	1	1	1	0	1	0	1	0	5
	A/IV 152	1	1	1	0	1	1	1	1	7
	A/IV 153	1	0	1	0	1	0	1	1	5
	A/IV 178	1	1	0	0	1	1	0	0	4
	A/IV 179	1	0	1	0	1	0	1	0	4
	A/IV 180	1	1	0	0	1	1	1	0	5
	A/IV 181	1	1	1	0	1	0	1	0	5
Araucana	A/IV 161	1	1	1	0	1	1	1	0	6
	A/IV 162	1	1	1	1	1	1	1	0	7
	A/IV 163	1	0	1	0	1	1	1	0	5
	A/IV 164	1	0	1	0	1	0	1	0	4
	A/IV 165	1	1	1	0	1	1	1	0	6
	A/IV 166	1	1	1	0	1	1	1	0	6
Hybrid Ross 308	A/IV 142	1	0	1	0	1	0	1	0	4
	A/IV 143	1	0	1	0	1	0	1	0	4
	A/IV 144	1	0	1	0	1	0	1	0	4
JA57+New Hampshire (mother+father, hybrid)	A/IV 182	1	0	0	0	1	0	1	0	3
	A/IV 183	1	0	1	0	1	0	1	0	4
	A/IV 184	1	0	1	1	1	0	1	0	5
Nagoya Koshin	A/IV 186	1	1	1	1	1	1	1	1	8
	A/IV 187	1	1	1	0	1	1	0	0	5
Puduaner	A/IV 185	1	1	1	0	1	1	1	0	6
Schweizerhuhn	A/IV 215	1	1	1	0	1	1	1	0	6
	A/IV 216	1	1	1	1	1	1	1	1	8

Table 3 continued.

Breed	Collection number	Left side				Right side				Total no. of foramina
Schweizerhuhn	A/IV 217	1	1	1	1	1	1	1	1	8
	A/IV 218	1	1	1	1	1	0	1	1	7
Shamo	A/IV 149	1	0	1	1	1	1	1	1	7
	A/IV 150	1	1	1	1	1	1	1	1	8
Ukokkei	A/IV 167	1	0	1	1	1	0	1	0	5
	A/IV 168	1	0	1	0	1	1	1	0	4
	A/IV 169	1	1	1	0	1	1	1	0	6
Welsumer	A/IV 170	1	1	1	0	1	1	1	0	6
	A/IV 171	1	1	1	0	1	1	1	0	6
	A/IV 172	1	1	1	0	1	0	1	0	5
Pennsylvania Naked Neck	A/IV 155	1	1	1	0	1	1	1	0	6
	A/IV 156	1	1	1	0	1	1	1	0	6
White Leghorn	A/IV 157	1	1	1	1	1	1	1	0	7
	A/IV 154	1	1	1	1	1	1	1	0	7
Zwergwelsumer	A/IV 173	1	1	1	0	1	1	1	0	6
	A/IV 174	1	1	0	0	1	1	0	0	4
Non-specified breed	A/IV 219	1	0	1	0	1	0	1	0	4
	A/IV 220	1	0	1	0	1	1	1	1	6
	A/IV 221	1	0	1	0	1	0	1	0	4
	A/IV 222	1	0	1	0	1	0	1	0	4
Feral chicken	A/IV 191	1	1	0	0	1	1	0	0	4
	A/IV 192	1	1	1	0	1	1	1	0	6
	A/IV 193	1	1	0	0	1	1	0	0	4

before they are fully mature, whereas the technique of van Wijhe only stains mature cartilage (Sonies, 1907; De Beer, 1930). Since we registered chondrocranial elements as soon as first chondrifications were mentioned, this could have led to a relative early appearance in the chondrification sequence compared to the cartilages prepared by the van Wijhe method and described by Sonies (1907).

The shift in the timing of the appearance of the fenestra basicranialis posterior (Parker, 1869) does reflect the mode of observed formation of the fenestra. Where a primary origin is mentioned (Sonies, 1907) the fenestra basicranialis posterior appears as early as the basal plate is formed (Lang, 1954), whereas in the cases of reports in which the fenestra appears later in ontogeny, together with first dermal ossifications (Heyns, 1997), its formation through resorption was observed (Parker, 1869; Tonkoff, 1900; Vorster, 1989).

The third element that differs from the consensus sequence is the tectum synoticum. Parker (1869) described the tectum synoticum (exoccipital lamina) in an earlier stage, around the chondrification of the first branchial arch, than documented in other specimens (Sonies, 1907; Vorster, 1989; Heyns, 1997). Vorster (1989) and Heyns (1997) mentioned the procartilaginous anlage of the tectum early in development, similar to the stage mentioned by Parker (1869), and described the cartilaginous stage of the tectum synoticum in a later stage, around the formation of the fenestrae in the nasal and interorbital septa.

Variation of the hypoglossal foramina in chondrocrania and skulls of chicken breeds

The number of hypoglossal foramina in the chondrocranium of *Gallus gallus* f. *domestica* is variable. An inconstant number of foramina in ontogeny and intraspecific variation are reported, and asymmetry in individuals was illustrated (Vorster, 1989; Heyns, 1997). These variations are also known for other birds (Sonies, 1907; De Beer & Barrington, 1934; Crompton, 1953; Engelbrecht, 1958) and reptiles (Rice, 1920). Sonies (1907) reported an increase from two hypoglossal foramina to three foramina in the ontogeny of chicken, but mentioned that the third hypoglossal foramen is related to an atrophied nerve root and occupied by a blood vessel. Likewise, two hypoglossal foramina in later ontogeny are documented (Parker, 1869; Tonkoff, 1900; Sonies, 1907; Erdmann, 1940; Lillie, 1952; Jollie, 1957; Vorster, 1989; Heyns, 1997). Our observation of 75 skulls revealed variation in the number of hypoglossal foramina among different breeds, and bilateral asymmetry, both common in the red junglefowl and the chicken breeds.

Conclusion

The variation in the morphology of the adult skull among chicken (Stange *et al.*, 2018) is not reflected in the chon-

Table 4. Number of hypoglossal foramina (left and right side) in ontogeny of *Gallus gallus* f. *domestica* as reported in the literature.

Literature	Age [day]							
	4	5	6	7−8	8	9	10-11	adult
Parker (1869)	2|2	2|2					2|2	2−3|2−3
Tonkoff (1900)							2|2	
Erdmann (1940)							2|2	
Sonies (1907)			2|2?	2|2*				
Lang (1955)		2|2						
Jollie (1957)							2|2	
Vorster (1989)		4|4	4|3		2|2	2|2	2|2	
Heyns (1997)			4|3		2|2	2−3|2−3	2|2	

*reports on an additional foramen occupied by a blood vessel later in ontogeny, and refers to it as 'hypoglossal foramen'.
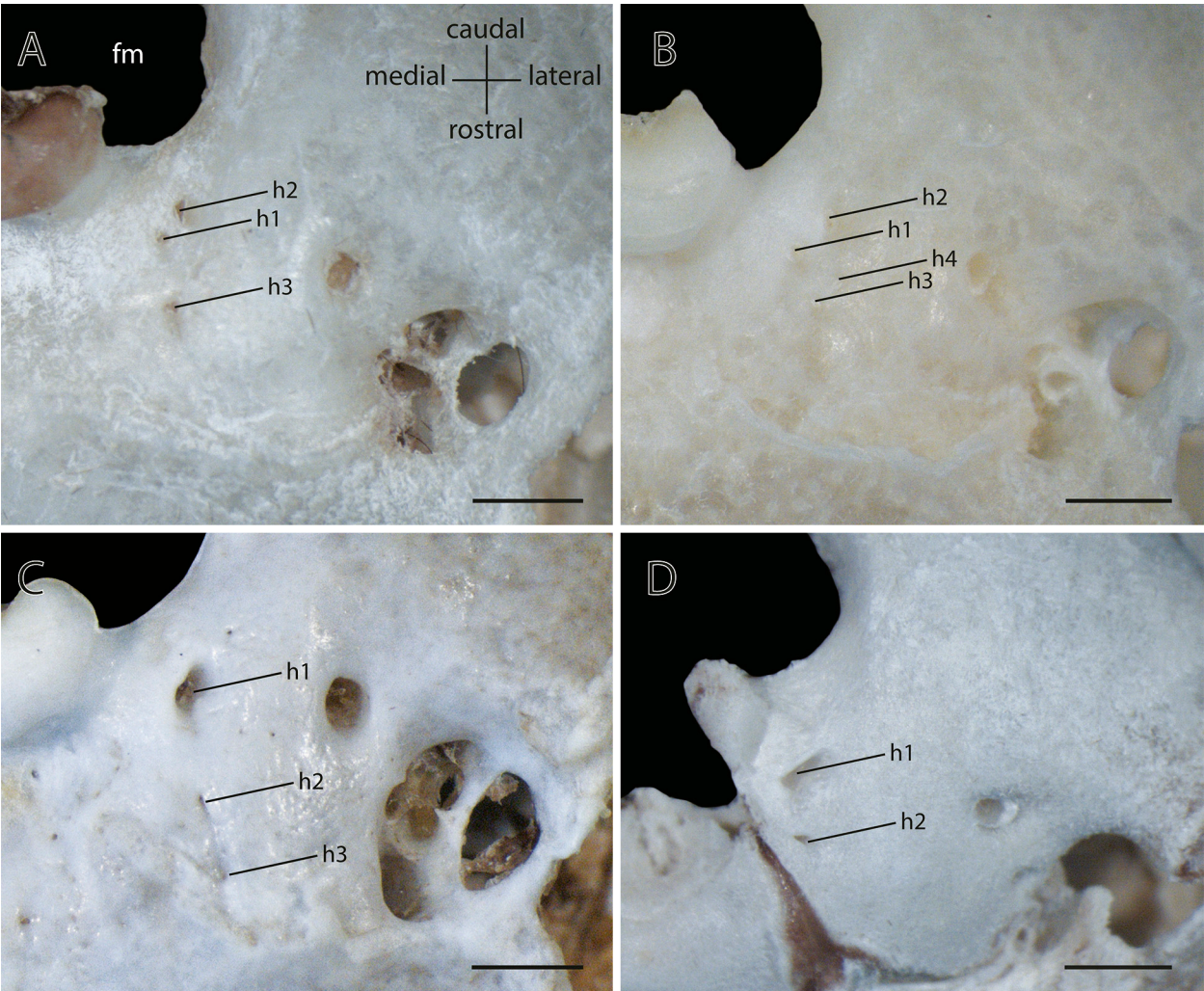
Fig. 4. Variation in the count of hypoglossal foramina among different chicken breeds (only right exoccipital bone photographed). A) Red junglefowl (A/IV 199), B) Schweizerhuhn (A/IV 216), C) Ukokkei, Japanese silkie (A/IV 168), and D) a non-specified breed (A/IV 221). Scale bars equal 2 mm.

drification sequence. Comparisons among breeds and the wild fowl reveal developmental penetrance (Richardson, 1999) of the adult (personal observation), although variation in the latter concern in great part the dermatocranium and not the chondrocranium (Stange *et al*., 2018). Subtle or larger differences in the shape of the chondrocranium may exist between the red junglefowl and diverse breeds (personal observation), but we cannot know based on the published literature given the artifacts of histological and whole mount preparation and visualization. Future stud-
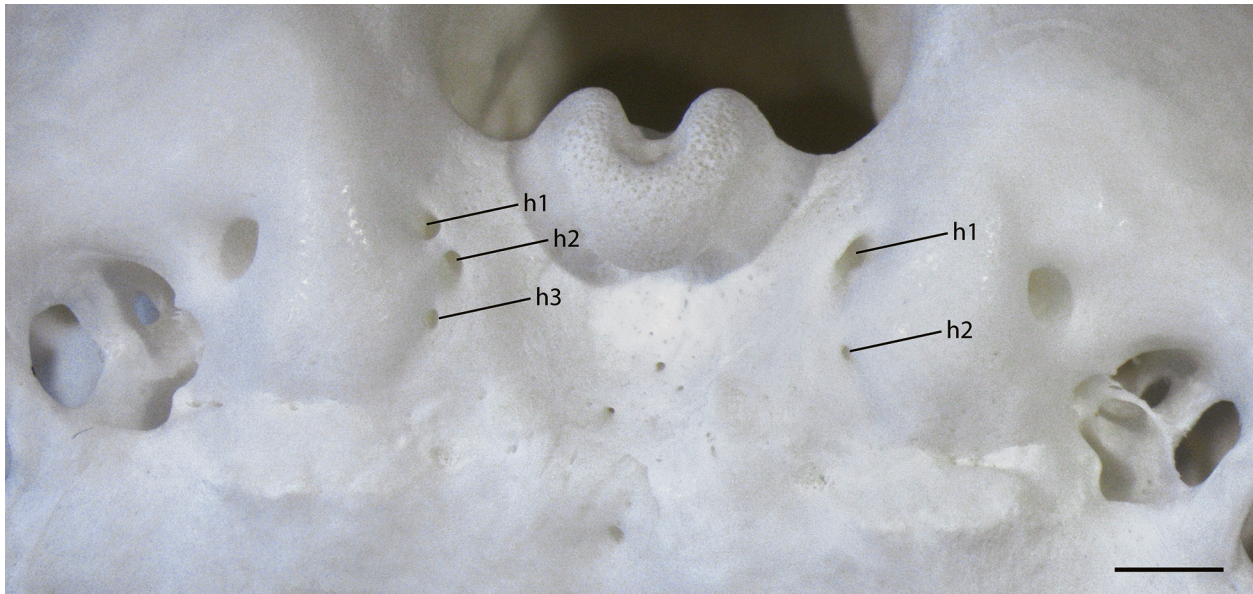
Fig. 5. Bilateral asymmetry in the number of foramina in a Welsumer hen (A/IV 172). Scale bar equal 2 mm.
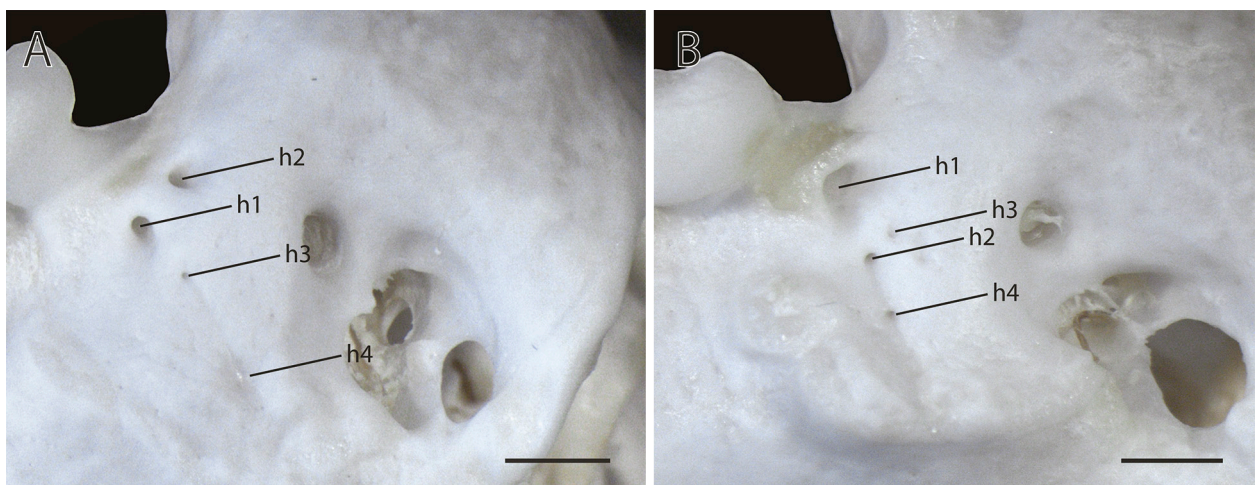

Fig. 6. Variable arrangement and size of hypoglossal foramina within the Shamo breed (only right exoccipital bone photographed). A) Shamo (A/IV 150) and B) Shamo (A/IV 149). Scale bars equal 2 mm.

ies on detailed shape changes in aged series of embryos of diverse breeds would be need to test for intraspecific variation in prenatal skulls following domestication.

We found that variation in the number of hypoglossal foramina in the bony skull among and within breeds, as bilateral asymmetry within individuals, is common in *Gallus gallus* f. *domestica*, though in literature mostly two hypoglossal foramina are reported or figured (PARKER, 1869; BELLAIRS & JENKIN, 1960; BUBIEN-WALUSZEWSKA, 1981). The bony adult skull exhibits the variation already visible in the chondrocrania.

Acknowledgements

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