



Detailed gross anatomic and sialographic characteristics of major salivary glands in water buffaloes (*Bubalus bubalis*)

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Abstract

To clarify the detailed general architecture and topography of major salivary glands and demonstrate a fine anatomy of the ductal system of the glands in water buffaloes, we conducted gross anatomic and sialographic investigations of glands in 14 half heads from seven water buffaloes. The position of the mandibular gland, course of mandibular duct relative to monostomatic gland, a rostral extension of polystomatic gland, and site of origin of monostomatic duct in buffaloes essentially differed from those of various ruminants. The shape of the parotid and mandibular glands, and origin of their ducts, lacking filling of retromandibular fossa by parotid gland, the topography of mandibular gland relative to the parotid gland and mandibular lymph node, caudal extension of polystomatic gland, rostral extension of monostomatic gland, and location of polystomatic gland relative to monostomatic gland in buffaloes were very similar to those in ox. However, several considerable differences in morphology of glands in buffaloes and ox were recognized. Major salivary glands in buffaloes almost show ‘grazing ruminants’ morphological and morphometrical characteristics. Within parotid, mandibular, and monostomatic glands in buffaloes, there was a ductal arborization pattern in lateral sialograms. Whereas the main parotid duct was formed by a union of two central branches of the intraglandular duct, main mandibular, and monostomatic ducts were consisted of one central branch. The pattern of peripheral branches from the central branch of intraglandular duct in buffaloes was significantly different among the glands. Our detailed sialography of ductal morphology and morphometry can be helpful in accurate diagnosis of gland diseases in live water buffaloes.

Keywords Anatomy · Salivary glands · Sialography · Water buffalo

Introduction

Major salivary glands of ruminants including parotid, mandibular, and sublingual (monostomatic and polystomatic) salivary glands release the saliva into the mouth (Dyce et al. 2014) and are of remarkable interest because they play a significant role in ruminal digestion (Smith 2014). Many morphological, topographical, and morphometrical studies

on major salivary glands in various domestic and wild ruminants have been conducted over the long course of veterinary history to provide important clues regarding the classification of the species according to the feeding types including browsers, intermediate feeders, and grazers (Hofmann and Stewart 1972; Hofmann 1989; Hofmann et al. 2008). They have also contributed to comparative morphology (Getty 1975; Nickel et al. 1979; Saber and Hofmann 1984; Kay 1987; Smuts and Bezuidenhout 1987; Pérez et al. 2017). According to the findings of these studies, the salivary glands in various domestic (sheep, goat, cattle, and camel) and wild ruminants (red deer, fallow deer, mouflon, pampas deer, blackbuck, Chinese water deer, and muntjac) are quite different in shape, size, position, and structure.

On the other hand, sialography is the only imaging technique and the most detailed way of examining the fine anatomy of the ductal system of the major salivary glands. In fact, sialography is a radiographic technique and an invasive procedure in which radiopaque contrast material is

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injected retrograde into the gland's ductal system via the intraoral opening of either the major salivary glands before imaging with plain films/digital image receptors (Mallya and Lam 2014). Literature review revealed that few conventional sialographic studies of the major salivary glands in animals have been reported including the sialography of parotid and mandibular glands in cattle, camels, sheep, and horses (Dehghani et al. 1994, 1999, 2000, 2005) as well as the sialography of the parotid, mandibular, and monostomatic glands in goats (Tadjalli et al. 2002) and dogs (Harvey 1969; Tadjalli et al. 2004). Recently, computed tomography (CT) sialography of dogs was investigated by Kneissl et al. (2011). Sialography has also been used for the evaluation of salivary gland diseases in large animals (Smith 2014).

Water buffaloes (*Bubalus bubalis*) are placed in the family Bovidae, within the order Artiodactyla. The domestic water buffalo (*Bubalus bubalis*) is an important livestock resource in many countries in Asia, Mediterranean regions and Latin America (Naserian and Saremi 2007). The water buffalo is a species with excellent zootechnical characteristics for both milk and meat production. Because of the steady increase in the consumption of these products, ranches face the need to increase the number of animals raised for these purposes (de la Cruz-Cruz et al. 2014). No description of the major salivary glands of water buffaloes seems to have been published. Therefore, the main objectives of this study are (1) to provide a detailed anatomical description of the parotid, mandibular, and sublingual salivary glands of water buffaloes; (2) to demonstrate fine ductal morphology of the major salivary glands of water buffaloes using conventional sialography; (3) to document the technique used to obtain sialograms of normal major salivary glands; (4) to consider morphological and sialographical similarities and differences between major salivary glands of water buffaloes and those of domestic and wild ruminants; and (5) to determine the feeding type in water buffaloes.

Materials and methods

Fourteen half heads of seven adult male water buffaloes in Khuzestan were used in this study. The specimens were obtained from a local slaughterhouse without any external abnormality or pathology. The age of animals was estimated from the teeth, ranging between 2–3 years (FAO 1977). The greatest length and width of the heads were measured using a measuring tape and a caliper. The heads were also weighed.

Gross dissection

The layer-by-layer dissections of facial muscles, facial vascular and nervous systems, major salivary glands, and

their ducts at both sides were thoroughly performed in fresh heads. Some heads were divided into two halves from the median line by an electric saw (JR3050T, Malcita, China) to dissect and document the anatomy and topography of the medial surface of the glands and ducts. In addition, the mandible was removed to visualize the mandibular duct and the sublingual gland and its duct. In some heads, the latex was injected via a cannula placed in common carotid arteries to distinguish the artery from the surrounding structures. Some dissection was performed under a floating arm fluorescent illuminated magnifier (LTS 120, China, 2.5×). In addition, photographs were documented throughout the dissection with digital images taken with a Canon digital camera (Canon, Tokyo, Japan, G9).

Morphometry of the glands

Liner measurements of the glands and ducts were taken by a caliper (200 mm: Insize, China) to an accuracy of 0.05 mm.

Sialographic procedure

Plain films

Plain radiography in the lateral and ventrodorsal positions were taken by KCD-10M-6AIT, portable X-ray, Toshiba, Japan for the imaging evaluation of the major salivary glands before sialography of each specimen.

Sialography of the parotid gland

The mouth was opened by a mouth gag. The intraoral opening of the parotid duct was easily found in the oral vestibule opposite the upper check teeth. Head-light unit glass was employed during the cannulation of the duct. A blunt cannula was used to avoid perforation of the duct wall of the glands and then the cannula was gently inserted and contrast material was slowly injected. The dose of the agent was dependent on the size of the head and the following sialographic phases.

The sialography was done in two phases: (1) Duct filling which was performed to visualize the ductal branching pattern of the glands (Mallya and Lam 2014). It was accomplished only by injecting 1–1.5 ml of the contrast material (Meglumine Compound Vial 76%, Darou Pakhsh, Iran) by the pressure of the hand. By suboptimal filling of the ductal system in this phase, some areas of the ductal system of the gland may not be seen. (2) Acinar or parenchymatous filling was performed to be able to see the acini (Mallya and Lam 2014). It was accomplished by injecting 2–3 ml of the contrast material. By overfilling the ductal system and a complete acinarization of the gland, the detailed

intraglandular ducts and the arrangement of branches were completely obscured, rendering the examination virtually useless. After each phase, latero-lateral radiographs were subsequently taken with a cassette placed under the side being examined and exposed to the opposite side of the head while the syringe was still connected.

Sialography of the mandibular gland

The orifice of the gland was positioned in the floor of the mouth on the sublingual caruncle. It was smaller than that of the parotid. Thus it was usually more difficult to cannulate the mandibular duct than the parotid duct. To cannulate the duct, the caruncle should be raised and steadied by grasping it with a fine smooth forceps. To better visualize the orifice of the duct, the magnifier was used during mandibular cannulization. Gentle rubbing of the peripheral venous catheter (Gauge: 20; pink color) in a rostrocaudal direction with a slight rotation can help the cannula to enter the orifice. The remaining procedures and the volume of the contrast agent were similar to those in conducting the sialography of the parotid gland.

Sialography of the monostomatic gland

The sialography of the gland was similar to that of the parotid gland except for the following. The peripheral venous catheter (Gauge: 24; yellow color) was used to cannulate the duct. The orifice of the major sublingual duct was the smallest duct of the major salivary ducts. Cannulization of the major sublingual duct was the most difficult and time-consuming among the ducts. The volumes of the contrast agent for filling the duct and the acini were 0.5 ml and 0.5–1 ml, respectively.

Sialographic morphometry of the main parotid and mandibular ducts

The greatest length of the main parotid duct (from the orifice to the site of the union of the central branches of the intraglandular duct) and the greatest length of the mandibular duct (from the orifice to the site of the beginning of the central branch of the intraglandular duct) were measured. The greatest widths of the parotid duct (at the level of its second angle) and mandibular duct (at the level of the beginning of its main or extraglandular duct) were also measured. The measurements of the variables were taken using an electronic caliper (FCR PRIMA V Console software) on the lateral sialograms. All parametric values were analyzed using analytical statistics in Microsoft Excel and were expressed as mean \pm standard deviation.

Results

Morphometry of heads

The mean values of the weights and the greatest length and widths of the heads were 20.01 ± 3.57 kg, 55.71 ± 3.69 cm, and 27.28 ± 1.90 cm, respectively.

Gross dissection

The major salivary glands in water buffaloes were comprised of large paired glands—the parotid, mandibular, monostomatic, and polystomatic sublingual glands.

Parotid gland

The lobulated and club-shaped parotid gland (Fig. 1a,b) was located along the caudal border of the lateral surface of the masseter muscle and extended from the ventral surface of the base of the ear to the angle of the mandible (Figs. 2, 3).

The gland was partly covered laterally by zygomaticoauricularis muscle (Fig. 3), parotid fascia, and parotidoauricularis muscle.

The medial surface of the gland was uneven (Fig. 1b) because of the lateral retromandibular and mandibular lymph nodes, the mandibular salivary gland, the angle of stylohyoid bone, the digastricus muscle, the occipitohyoid muscle, the external carotid artery and its branches, the maxillary vein, the facial nerve, the superficial temporal nerve, the dorsal and ventral buccal nerves, and the transverse facial artery (Figs. 2, 3).

The maxillary vein was pierced at the middle part of the medial surface of the gland. There were no angles at the dorsal and ventral ends of the gland (Fig. 1b).

Parotid duct

The duct was formed by the confluence of the two dorsal radicles (Fig. 12b) within the medial surface of the middle part of the ventral end of the gland (Fig. 1b). Then, the duct together with the facial vessels ran ventral to the ventral buccal nerve over the ventral border of the lateral surface of the masseter muscle. Then it passed medial to the tendon of the insertion of the sternomandibularis muscle. After that, it was inclined dorsally and rostrally to ascend in the vascular groove caudal to the facial vessels along the rostral border of the masseter. After that, the duct passed rostradorsally to open a space between buccinator muscle and the lateral wall of the vestibule by

Fig. 1 Lateral (a) and medial (b) surfaces of the removed left parotid gland in water buffalo. 1. Canal of the maxillary vein, 2. Parotid duct. Scale: 1 cm

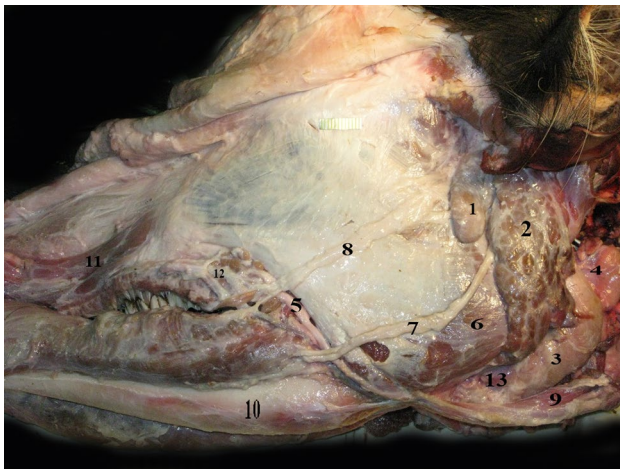
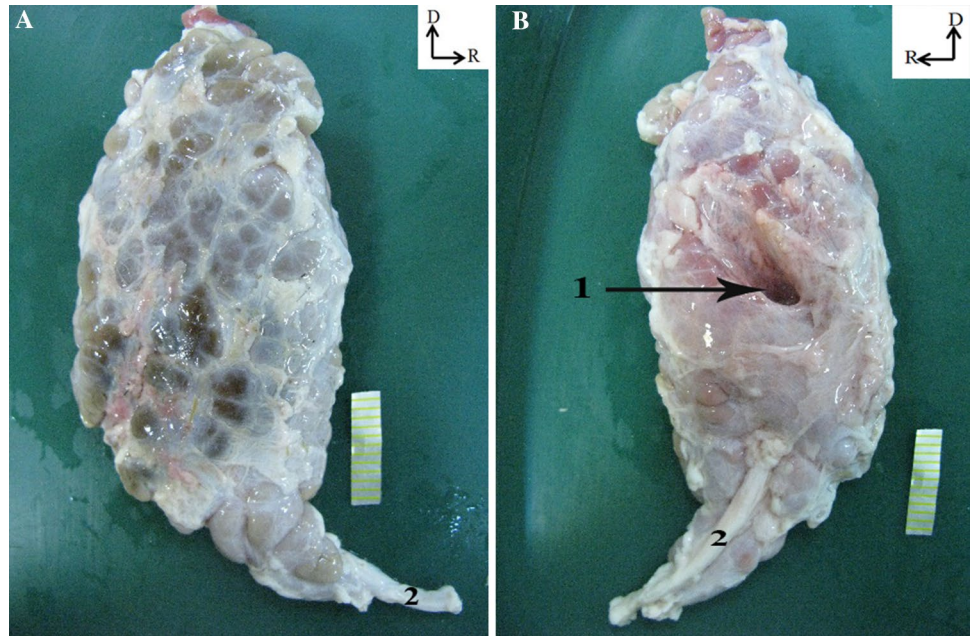


Fig. 2 Lateral view of the left parotid duct in water buffalo. 1. Parotid lymph node, 2. Parotid gland, 3. Mandibular gland, 4. Lateral retropharyngeal lymph node, 5. Parotid duct, 6. Master muscle, 7. Ventral buccal nerve, 8. Dorsal buccal nerve, 9. Sternomandibularis, 10. Mandible, 11. Zygomaticus, 12. Facial vein, 13. Mandibular lymph node. Scale: 1 cm

penetrating the dorsal buccal gland. In this location, the zygomaticus muscle did not pass over the termination part of the parotid duct (Fig. 4).

In six specimens, the intraoral openings of the ducts were opened bilaterally on papillae that lies opposite the longitudinal crest of the first upper molar teeth (M1) (Fig. 5a). In one specimen, the intraoral openings of the ducts were opened bilaterally on large papillae that lies almost between the fourth upper premolar (P4) and the first molar teeth (M1) (not shown).

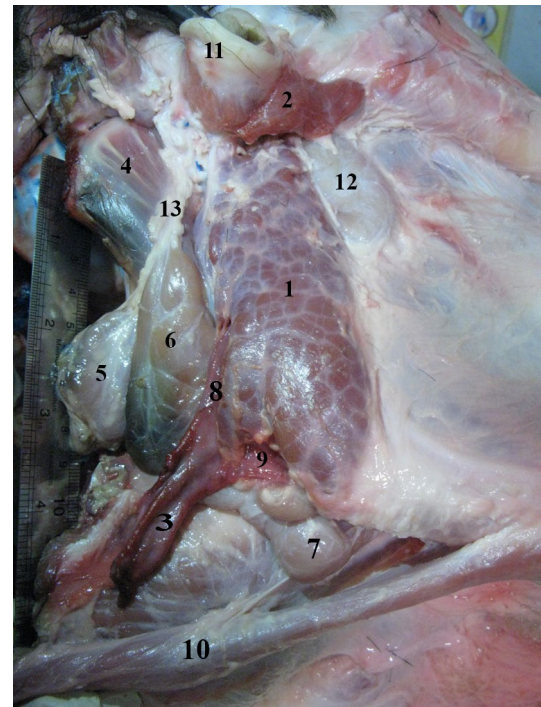


Fig. 3 Lateral view of the left parotid duct in water buffalo. 1. Parotid gland, 2. Zygomaticauricular, 3. Linguofacial vein, 4. Occipitomandibularis, 5. Lateral retropharyngeal lymph node, 6. Mandibular gland, 7. Mandibular lymph node, 8. Maxillary vein, 9. Lingual vein, 10. Sternomandibularis, 11. Auricular cartilage, 12. Parotid lymph node, 13. Supporting connective tissue of the mandibular gland, attaching to the jugular process

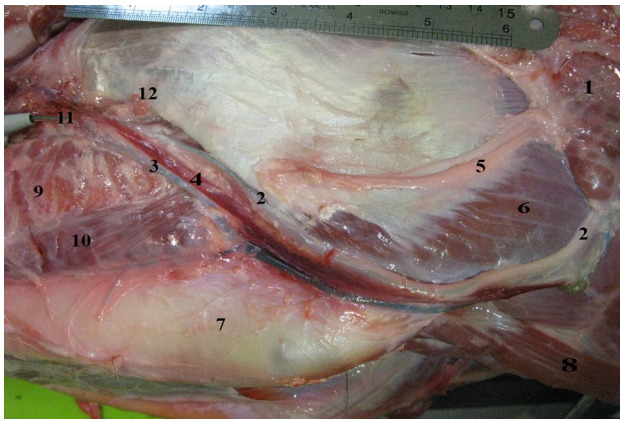


Fig. 4 Lateroventral view of the origin, course, and termination of the left parotid duct in water buffalo. The common carotid artery was injected with blue color latex. The terminal part of the ventral buccal nerve has been cut to expose the structure coursing over the face. 1. Parotid salivary gland, 2. Parotid duct, 3. Facial artery, 4. Facial vein, 5. Ventral buccal nerve, 6. Masseter muscle, 7. Mandible, 8. Sternomandibularis muscle, 9. Buccinator muscle, 10. Depressor labii mandibularis, 11. Orifice of the parotid duct (catheter in place), 12. Dorsal buccal salivary gland

The mean values of the greatest length, the width of the dorsal end, widths of the middle, and ventral end of the gland were 128.92 ± 20.52 , 44.42 ± 5.66 , 50.07 ± 10.03 , 37.42 ± 10.76 mm, respectively. The mean values of the

length and widths of the duct were 233.14 ± 20.5 and 4.57 ± 0.79 mm, respectively.

Mandibular gland

The pale yellow, uniform and lobulated gland was curved (Fig. 6a, b) and located partly caudal to about the middle of the ramus of the mandible and medial to the angle of the mandible, reaching into the caudal intermandibular space (Figs. 2, 3). The lobules of the gland did not have the same dimensions as the parotid lobules and were fitted together much more loosely with fewer connective tissues separating them. The lobules of the mandibular gland (Fig. 6a, b) were larger than those of the parotid gland (Fig. 1a, b).

The mandibular gland was related laterally to the mandibular lymph nodes (rostral and caudal), medial pterygoideus, digastricus, tendinous insertion of the sternomandibularis muscles, linguofacial vein, sternohyoideus muscle, angle of the mandible, and ventral end of the parotid salivary gland (Figs. 2, 3, 7).

The mandibular gland was related medially to the common carotid artery, pharynx, larynx, vagosympathetic trunk, sternocephalicus, and lateral retropharyngeal lymph node (Figs. 3, 7).

The rounded dorsal end of the mandibular gland was related to the lateral retropharyngeal lymph node, linguofacial and maxillary veins, parotid gland, and caudal belly of the

Fig. 5 a Rostrolateral view of the left parotid orifice (catheter in place) and its papilla (arrow) in water buffalo. P2-4: upper second to fourth premolar teeth, M1. First molar tooth. **b** Sublingual floor of the oral cavity in water buffalo. Sublingual caruncle with openings of the mandibular (pink angiocath) and major sublingual (yellow angiocath) ducts are cannulated

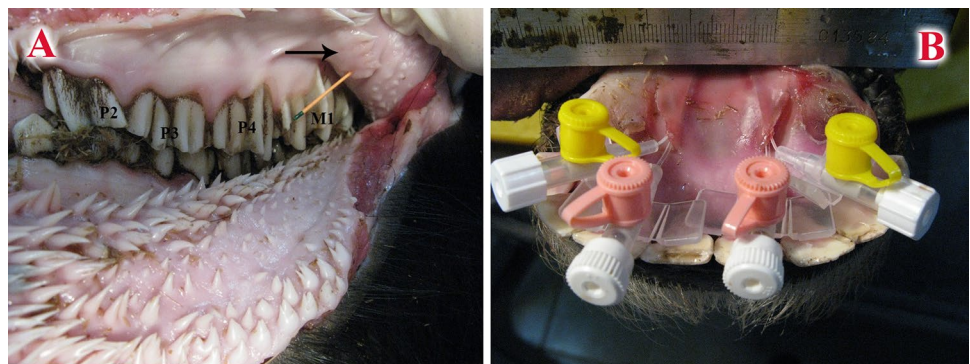
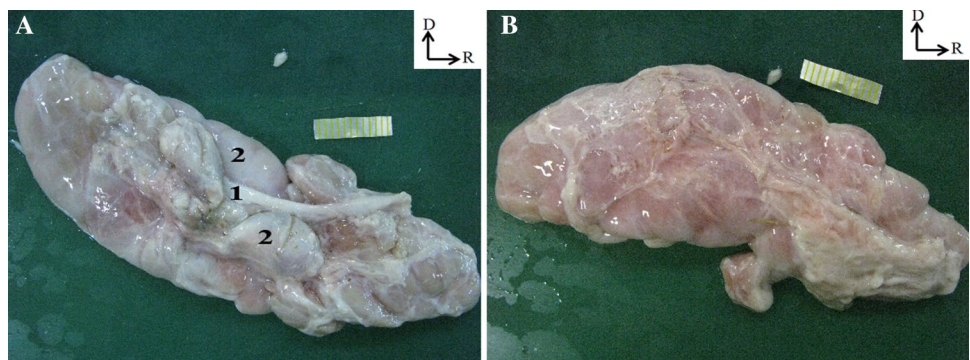


Fig. 6 Lateral (a) and medial (b) surfaces of the removed left mandibular gland in water buffalo. 1. Mandibular duct, 2. Mandibular lymph nodes, R, rostral; D, dorsal. Scale: 1 cm



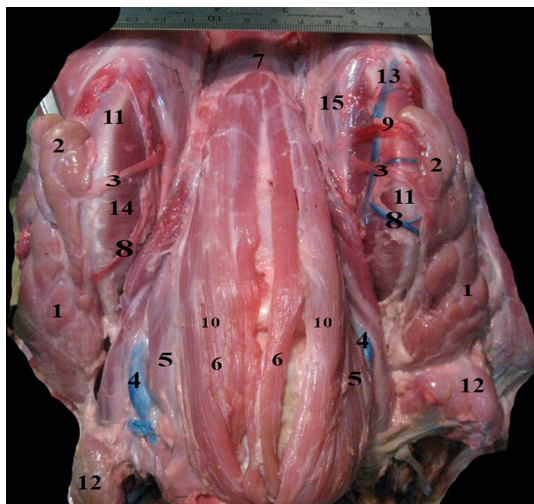


Fig. 7 Ventrocaudal view of the mandibular gland in water buffalo. The gland and sternomandibularis have been displaced laterally to show the surrounding structures of the gland and its duct. The right lingual vein has been cut to show the surrounding structures of the gland. The right and left common carotid arteries have been injected with blue color latex. 1. Mandibular salivary gland, 2. Mandibular lymph nodes, 3. Mandibular duct, 4. Common carotid artery, 5. Mylohyoideus muscle, 6. Sternohyoideus muscle, 7. Larynx, 8. Lingual artery, 9. Lingual vein, 10. Sternothyroideus, 11. Mandible, 12. Lateral retromandibular lymph node, 13. Sternomandibularis, 14. Medial pterygoideus muscle

the digastricus muscle (Figs. 3, 7). The end was connected to the jugular process by a connective tissue (Fig. 3).

The rounded ventral end of the gland was related to the mandibular lymph nodes (caudal one), sternomandibularis, sternohyoideus, and the body of the mandible (Figs. 2, 3, 7). The ventral end was not in contact with the contralateral gland and was not palpable (Fig. 7).

The gland did not have any angles (Fig. 6a, b).

Mandibular duct

The duct was formed by one radicle (Fig. 14), emerging from the middle of the lateral surface of the gland (Fig. 6a), and medial to the angle of the mandible. The duct together with the lingual artery was turned rostrally between the ramus of the mandible and the intermediate tendon of the digastricus muscle over the medial surface of the medial pterygoideus muscle (Figs. 7, 8).

Once reaching the polystomatic sublingual gland, the duct passed rostrally along the ventral border of the gland together with the lingual artery and lingual nerve of the mandibular nerve on the medial surface of the mylohyoideus muscle. Then, for a short distance, the duct together with the lingual nerve was located in the intermuscular septum between the mylohyoideus and stylohyoideus muscles and

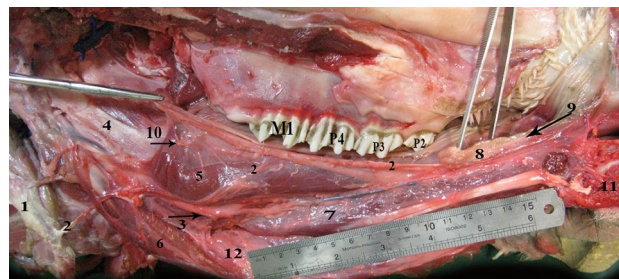


Fig. 8 Lateral view of the origin, course, and relationship of the right mandibular duct in water buffalo. The mandible, genioglossus, and mylohyoideus muscles have been removed. 1. Mandibular gland, 2. Mandibular duct, 3. Mandibular nerve of trigeminal nerve, 4. Stylohyoid bone, 5. Stylohyoideus, 6. Rostral belly of digastricus, 7. Geniohyoideus, 8. Monostomatic gland, 9. Major sublingual duct, 10. Polystomatic gland, 11. Mandibular symphysis, 12. Genioglossus, 13. P2-4: upper second to fourth premolar teeth, M1. Upper First molar tooth

then descended ventrally between mylohyoideus and geniohyoideus muscles (Figs. 8, 9).

Once reaching the monostomatic sublingual gland, the duct passed along the middle of the lateral surface of monostomatic sublingual salivary gland and closely ventral to the major sublingual duct (Fig. 11) and then they passed dorso-rostrally over the lingual surface of the incisive part of the body of the mandible under the mucous membrane to open independently, just behind the central and first intermediate incisors (I1 and I2) into a small orifice on the ventral surface of the large flat sublingual caruncle, close to the midline (Fig. 5b).

The mean values of the greatest length, width of the rostral end, width of the middle, and caudal end of the gland were 164.73 ± 26.73 , 34.13 ± 4.06 , 40.4 ± 8.05 ,

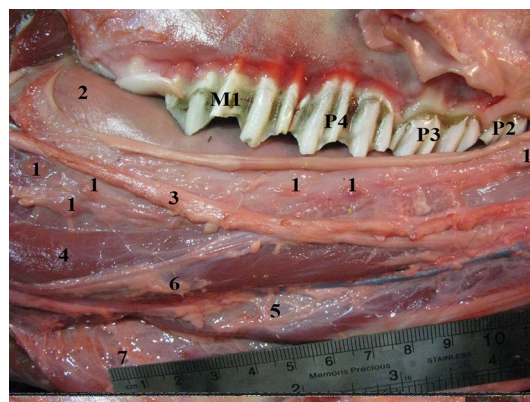


Fig. 9 Lateral view of the right polystomatic gland in water buffalo. Close-up of Fig. 8. 1. Major salivary glands, 2. Glossopalatine arch, 3. Lingual nerve, 4. Styloglossus muscle, 5. Geniohyoideus muscle, 6. Mandibular duct, 7. Genioglossus muscle, P2-4: upper second to fourth premolar teeth, M1. Upper First molar tooth

34.4 ± 4.65 mm, respectively. The mean values of the length and width of the duct were 343.5 ± 12.55 and 1.3 ± 0.07 mm, respectively.

Sublingual salivary glands

These glands consisted of the polystomatic and monostomatic sublingual glands.

Polystomatic gland

The gland was located over the lateral surface of the body and the root of the tongue, laying opposite the rostral border of the second upper premolar tooth to the last upper molar tooth close to the palatoglossal arch (Figs. 8, 9).

The gland was located caudal to the monostomatic sublingual salivary gland (Fig. 8, 9).

The gland was medially in relation with the styloglossus muscle, laterally with the mylohyoideus muscle, dorsally with the sublingual fold, and ventrally with the lingual nerve. The reddish gland consisted of a loose chain of isolated lobules. The lobules which were located more caudally were yellowish and larger (Fig. 9).

Minute and numerous excretory ducts of the gland were opened to visible pores along the sublingual fold (not shown).

Monostomatic gland

The gland was located at the level of diastema and extended from the caudal end of the mandibular symphysis to about the level of the rostral border of the first upper premolar tooth (Fig. 8).

The yellowish gland was the palest gland in color among the studied major salivary glands (Figs. 8, 10) and consisted of more loosely arranged lobules than other major salivary glands (Fig. 10).

The gland was laterally in relation with the mylohyoideus muscle and the mandibular duct, medially with the styloglossus muscle, ventrally with the geniohyoideus muscle

and the lingual nerve, and rostrally with the frenulum of the tongue (Figs. 8, 9).

Major sublingual duct

The duct began from the caudal end of the gland close to the dorsal border of the lateral surface of the gland and passed rostrally throughout the gland closely dorsal to the mandibular duct (Fig. 11) and then passed dorsorostrally over the lingual surface of the incisive part of the body of the mandible under the mucosa membrane to open independently into the ventral surface of the sublingual caruncle just a little rostral and lateral to the orifice of the mandibular duct (Fig. 5b). The orifice of the duct was very tiny and quite difficult to find.

The mean values of the greatest length, width of rostral end, width of the middle, and the caudal end of the monostomatic gland were 84.85 ± 9.87 , 15.71 ± 1.40 , 29.14 ± 1.45 , 17.85 ± 2.33 mm, respectively. The mean values of the

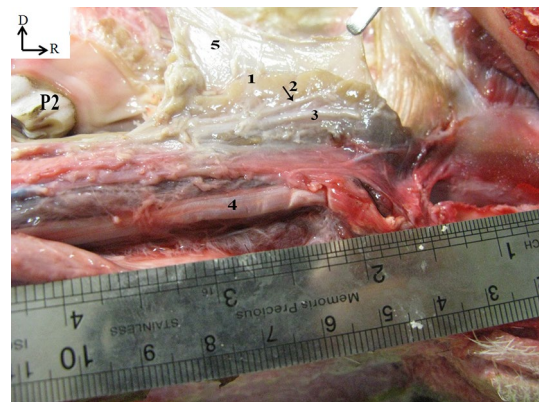


Fig. 11 Lateral view of the monostomatic gland and its duct in water buffalo. The mandible was removed and the capsule of the gland was reflected dorsally to show the monostomatic and mandibular ducts. 1. Monostomatic gland, 2. Major sublingual duct, 3. Mandibular duct, 4. Geniohyoideus, 5. Capsule of monostomatic gland, 6. P2: second premolar tooth, R, rostral; D, dorsal

Fig. 10 Lateral (a) and medial (b) surfaces of the removed left monostomatic gland in water buffalo. R, rostral, D, dorsal. Scale: 1 cm

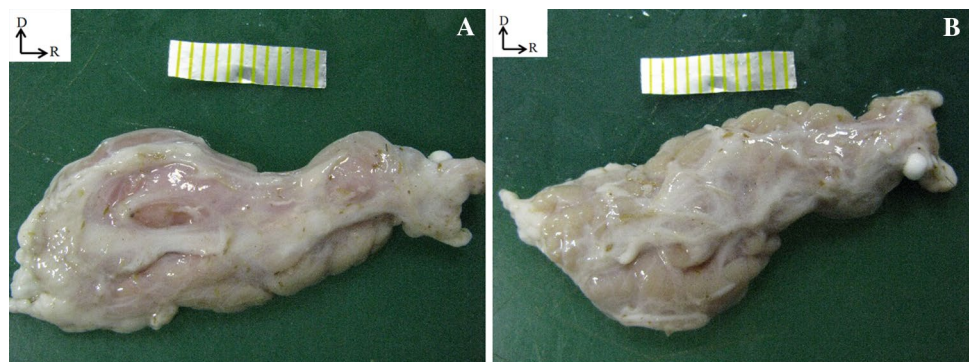


Fig. 12 **a** Lateral view of the plain film of the head in the water buffalo showing none of the major salivary glands is visible. **b** Lateral view of the left parotid sialogram in water buffalo. **a** The orifice of the main parotid duct (catheter in place), **b** Second angle of the main duct, **c** central branch of intraglandular duct

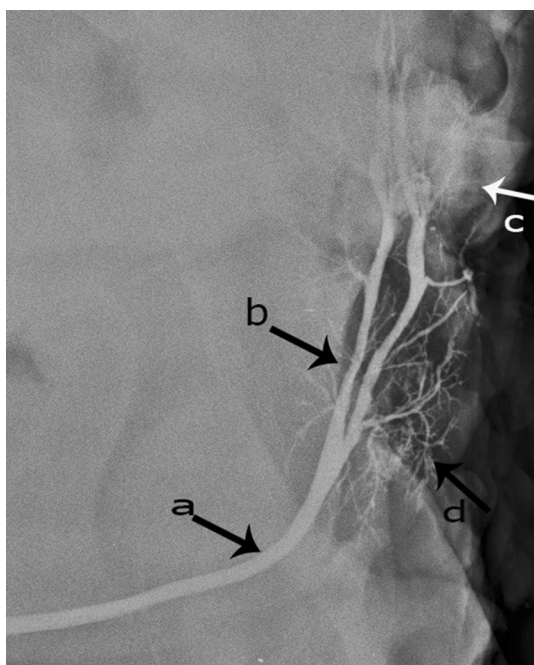
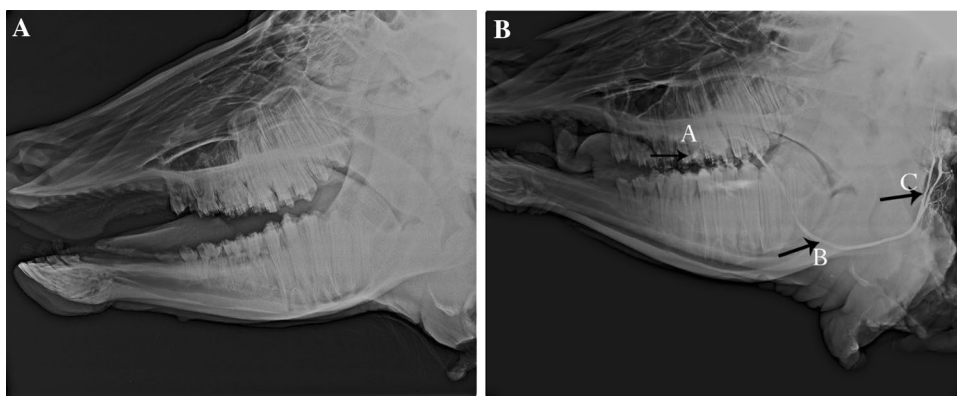


Fig. 13 Cropped lateral view of the left parotid sialogram in water buffalo. **a** Main duct, **b** central branch of the intraglandular duct, **c** the leafless tree is coming into bloom in ventral of the external acoustic meatus, **d** peripheral branches of the central branch from intraglandular duct (leafless tree)

length and width of the duct were 105.28 ± 9.53 and 1.05 ± 0.1 mm, respectively.

Sialography

The major salivary glands and their ducts were not visualized on the lateral plain radiograph (Fig. 12a).

Detailed and complete ductal branching arrangement of the parotid, mandibular, and monostomatic salivary glands were visualized individually (Figs. 12a, 13, 14, 15) and totally (Fig. 16) by sialography.

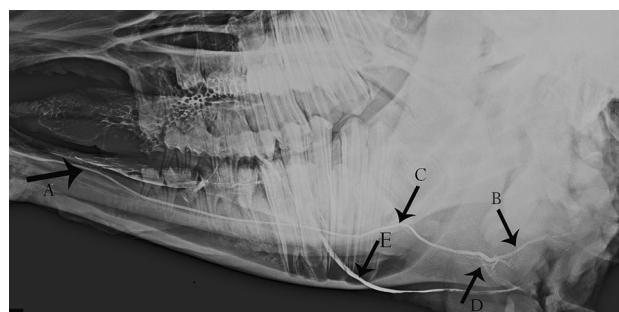


Fig. 14 Lateral view of the left parotid and mandibular sialogram in the water buffalo. **a** Main mandibular duct, **b** central branch of intraglandular duct, **c** angle of the main duct, **d** peripheral branches of the central branch from intraglandular duct (leafless tree or arborizing pattern), **e** main parotid duct

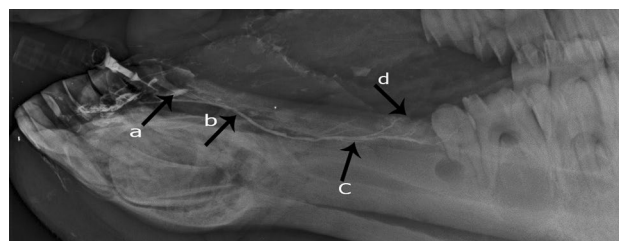


Fig. 15 Lateral view of the left monostomatic sialogram in water buffalo. **a** Orifice of the gland (catheter in place), **b** main sublingual duct, **c** central branch of intraglandular duct, **d** peripheral branches of central branch from intraglandular duct (leafless tree is coming into bloom)

On lateral sialograms, the fine anatomy of the salivary ductal system was clearly identified, while the ductal system and the bony structures of the mandibles, skull, and horns excessively overlapped with ventrodorsal sialograms (not shown).

Sialography of the right and left major salivary glands in one cadaver was virtually useless because of the complete superimposition of the ductal system of the contralateral glands (not shown).

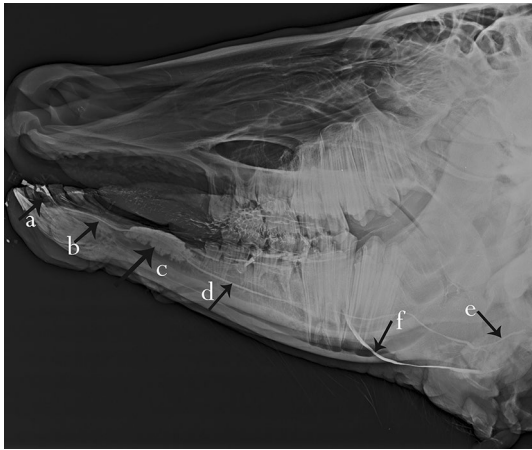


Fig. 16 Lateral view of the left parotid, mandibular, and monostomatic sialogram in water buffalos. **a** Orifices of mandibular and monostomatic ducts (catheters in places), **b** main monostomatic duct, **c** acinar opacification (*filling*) throughout the monostomatic gland and none of the branches of intraglandular ducts is visible, **d** main mandibular duct, **e** central branch of mandibular intraglandular duct, **f** main (extraglandular) parotid duct

Sialography of the parotid gland

About the middle of the caudal border of the ramus of the mandible, the main parotid duct (extraglandular duct) was formed by the union of two long central branches of the intraglandular duct (Figs. 12b, 13). The main duct had a curving pattern from the ventral border of the mandible to its orifice (Fig. 12b).

Peripheral branches originated from different directions of the central branches of the intraglandular duct (Figs. 12b, 13).

As the duct passed at the rostral border of the masseter muscle, the duct was compressed by that muscle so that the shadow of the contrast medium was significantly attenuated at this place (Fig. 12b).

The opacified orifice of the parotid duct was opposite the first upper molar tooth (Fig. 12b).

In the ductal filling phase, the main parotid duct was visible and the central and peripheral branches of the intraglandular parotid duct had a leafless tree or arborizing pattern (Figs. 12b, 13).

In the acinar or parenchymal (opacification or filling) phase, all ductal arrangements were visible but the tree came into bloom, i.e. the terminal ducts and acini (Fig. 13).

The main parotid duct exhibited three angles. The descending angle was almost at the level of the angle of the mandible. The location of the ascending angle was about the level of the vascular groove. The last angle was located at the rostral termination of the duct about the level of the upper molar teeth which was then bent slightly rostroventrally to the end to the orifice (Fig. 12b).

Sialography of the mandibular gland

The main mandibular duct began from the middle of the gland at the level of the angle of the mandible and ran dorsally first and then rostrally parallel to the middle of the molar part of the body of the mandible. Then at the level of the diastema, it continued parallel to the interalveolar border of the incisive part of the body of the mandible to its opening (Figs. 14, 16).

At the level of the distal end of the stylohyoid bone, the main duct on its caudal course suddenly curved ventrally to form an angle (Fig. 14).

The ductal (Fig. 14) and the acinar filling (not shown) of the mandibular gland were noted on the sialograms.

The pattern of the mandibular intraglandular duct was different from that seen in the parotid gland in the following ways: (1) The central branch of the intraglandular mandibular duct consisted of a single branch which extended slightly in caudoventral direction throughout the gland. (2) The peripheral branches of the central branch from the intraglandular duct originated almost ventrally from two different points close to the caudal and rostral end of the central branch.

The peripheral branches of the central branch from the mandibular duct were finer and tapered more suddenly, ending with a poor filling of their terminal ducts (Figs. 14, 16) compared to those in the parotid gland (Fig. 13).

Sialography of the monostomatic gland

Due to the small size and the short duct of the monostomatic gland, it was difficult to achieve an optimal duct filling without complete acinarization of the gland in several specimens. Extensive acinar filling of the gland also obscured the peripheral branches of the intraglandular ductal arrangement in the gland (Fig. 16).

Like the central branch of the intraglandular branch of the mandibular gland, the central branch of the intraglandular duct from the monostomatic gland was a single one but originated from the middle of the caudal end of the gland and passed rostrally along the middle of the gland (Fig. 15).

The central branch of the intraglandular duct and the main duct of the monostomatic sublingual gland followed the dorsal border of the diastema to its opening.

The peripheral branches of the intraglandular duct almost originated dorsally from different parts of the duct which were finer and smaller (Fig. 15).

The main monostomatic duct was the shortest duct (Fig. 15) among the main ducts of the major salivary glands (Figs. 12a, 16) which passed over the interalveolar border of the incisive part of the body of the mandible (Fig. 15).

Once both the complete acinarization of the monostomatic gland and the sialography of the mandibular gland

were achieved, the main mandibular duct passed, for a short distance, ventrally to the monostomatic sublingual gland before reaching the level of diastema. However, from the level of diastema to its opening, the mandibular duct was superimposed over the main monostomatic duct to its opening (Fig. 16).

Sialographic morphometric

The mean values of the greatest length and width of the main parotid and the mandibular ducts were 23.48 ± 0.62 cm and 0.43 ± 0.03 cm as well as 34.69 ± 1.0 cm and 0.28 ± 0.00 cm, respectively.

Discussion

Gross dissection

Parotid gland

Shape and appearance The shape of the gland varies among wild and domestic ruminants. It is club-shaped (Nickel et al. 1979) or elongated (Habel 1970; Budras et al. 2003) in the ox, rectangular in the sheep (Getty 1975), mouflon, red deer, and fallow deer (Saber and Hofmann 1984), and camel (Smuts and Bezuidenhout 1987), or triangular in the pampas deer (Pérez et al. 2017), and ox (Getty 1975) with two rostral and caudal lobes in giraffes (Pérez et al. 2012). In the present study, the parotid gland was club-shaped.

Dorsal and ventral ends of the gland had auricular, cervical and mandibular angles in the mouflon, red deer, and fallow deer (Saber and Hofmann 1984), Chinese water deer and muntjac (Kay 1987), or cervical angles in the sheep and goat (Getty 1975; Nickel et al. 1979) and camel (Smuts and Bezuidenhout 1987). In this study, however, such angles were not found as seen in the ox (Getty 1975; Nickel et al. 1979).

Position The gland filled the retromandibular fossa in the mouflon, red deer, fallow deer (Saber and Hofmann 1984), and pampas deer (Pérez et al. 2017), water deer and Muntjac (1987), camel (Smuts and Bezuidenhout 1987), and sheep (May 1964). However, in the current study, the gland did not fill the fossa as seen in the ox (Getty 1975; Nickel et al. 1979).

Unlike the ox (Habel 1970; Getty 1975; Nickel et al. 1979; Budras et al. 2003), the dorsal end of the gland in water buffaloes did not reach the zygomatic arch or temporomandibular joint.

Relationship of the parotid gland and its lymph node The dorsal end of the rostral border of the parotid gland partially

covers the parotid lymph node in the ox (Nickel et al. 1979; Getty 1975; Dyce et al. 2014), mouflon, red deer, and fallow deer (Saber and Hofmann 1984), as well as red deer, muntjac, and blackbuck (Kay 1987) or completely in the sheep and goat (Getty 1975; Nickel et al. 1979). The gland also covers two-thirds of the lymph node in the red deer. However, in the present study, the gland did not cover the parotid lymph node as is seen in camels (Smuts and Bezuidenhout 1987) and giraffes (Pérez et al. 2012). The node is also routinely incised with masseter in meat inspection of ruminants (Habel 1970).

The origin, course, and termination of the parotid duct Like the ox (Getty 1975; Nickel et al. 1979), pampas deer (Pérez et al. 2017), red deer, and fallow deer (Saber and Hofmann 1984), the parotid duct in water buffaloes runs rostrally on the ventral part of the lateral surface of the masseter (Habel 1970; Nickel et al. 1979). However, the duct passes on the medial side of the ventral border of the mandible in the ox (Getty 1975; Budras et al. 2003).

Like the mouflon, red deer, fallow deer (Saber and Hofmann 1984) and pampas deer (Pérez et al. 2017), the parotid duct was formed by the union of the two radicles or branches. However, it was formed by four converging radicles in the camel (Rezk and Shaker 2017), or numerous radicles in the ox (Nickel et al. 1979; Budras et al. 2003).

The relationship between the parotid duct and the buccinator muscle was reported only in few publications of medical history. Like the sheep (May 1964) and pampas deer (Pérez et al. 2017), the parotid in water buffaloes pass superficially to the buccinator muscle. However, the parotid duct passed through the muscle in the sheep (May 1964), mouflon, red deer, and fallow deer (Saber and Hofmann 1984), camel (Curasson 1947), and human (Kang et al. 2006). In humans, the buccinator muscle plays a functional role in saliva secretion from the parotid gland (Kang et al. 2006).

The orifice of the parotid duct on the oral vestibule is very variable among the domestic and wild ruminants. Like the camel (Smuts and Bezuidenhout 1987), Chinese water deer, blackbuck deer, and muntjac (Kay 1987), the orifice in the 6 heads of water buffaloes under study was located bilaterally opposite the first upper molar tooth (M1). In addition, the duct in one water buffalo was positioned bilaterally opposite and between P4 and M1, similar to those reported in the sheep (May 1964; Getty 1975; Nickel et al. 1979).

On the other hand, the orifice was situated opposite P4 in the goat (Habel 1970), mouflon, red deer, fallow deer (Saber and Hofmann 1984), and pampas deer (Pérez et al. 2017), and opposite M2 in the ox (Getty 1975; Nickel et al. 1979; Barone 1997) and giraffe (Pérez et al. 2012).

Unlike the pampas deer (Pérez et al. 2017) and ox (Barone 1997), the parotid papilla was present in water buffaloes and all previous reports in the wild and domestic ruminants.

Mandibular gland

Shape and appearance The gland consisted of two separable lobes in the mouflon (Saber and Hofmann 1984), pampas deer (Pérez et al. 2017), and blackbuck deer (Kay 1987), or four lobes in the camel (Rezk and Shaker 2017). In addition, the gland is composed of two lobes with the rostral lobe having four smaller lobes in the giraffe (Pérez et al. 2012). However, the gland was totally and distinctively lobulated in water buffaloes.

Unlike the mouflon, red deer, and fallow deer (Saber and Hofmann 1984), the mandibular gland of water buffaloes did not exhibit any angles as seen in the ox, sheep, and goat (Nickel et al. 1979).

Like the ox (Getty 1975; Nickel et al. 1979), and blackbuck deer (Kay 1987), the mandibular gland was curved in shape. However, the shape of the gland is triangular in the camel (Smuts and Bezuidenhout 1987), mouflon, red deer, fallow deer (Saber and Hofmann 1984), muntjac and Chinese water deer (Kay 1987), and sheep and goat (Getty 1975) or is irregularly quadrilateral in the sheep (May 1964).

Position The gland in water buffaloes was situated caudal to the ramus of the mandible and extended rostrally into intermandibular space as seen in the mouflon, red deer, and fallow deer (Saber and Hofmann 1984) and ox (Habel 1970; Getty 1975; Nickel et al. 1979).

The gland is extended caudally from the wing of the atlas (atlantal fossa) in the mouflon, red deer, and fallow deer (Saber and Hofmann 1984), camel (Smuts and Bezuidenhout 1987), and ox (Habel 1970; Getty 1975; Nickel et al. 1979) or from the paracondylar process in the ox (Budras et al. 2003). In the sheep, the gland is located partly caudal and medial to the angle of the mandible (May 1964). Also, the gland is placed medial to the ventral part of the parotid in the Chinese water deer, and red deer (Kay 1987). In the present study, however, the gland was not extended as far caudally as it is in the above-mentioned animals.

Unlike the ox (Getty 1970; Budras et al. 2003; Dyce et al. 2014), the ventral end of the gland in water buffaloes was not palpable and did not meet its fellow in the midline of the intermandibular space.

Relationship of the mandibular gland relative to the parotid gland and mandibular lymph node The coverage of the mandibular gland by the parotid gland is quite different among wild and domestic ruminants. Like the ruminants (Nickel et al. 1979), and blackbuck (Kay 1987), the middle of the lateral surface of the mandibular gland in water buffaloes was covered by the parotid gland. However, the greater part of the gland is also covered by the parotid gland in the mouflon, red deer, fallow deer (Saber and Hofmann 1984), Chinese water deer, and

muntjac (Kay 1987). The mandibular gland of camels is completely covered by the parotid gland except for its caudal and ventral border (Smuts and Bezuidenhout 1987). Caudal lobe of the gland is covered by the parotid gland in the pampas deer (Pérez et al. 2017), and giraffe (Pérez et al. 2012).

Like the ox (Getty 1970; Nickel et al. 1979), red deer, and fallow deer (Saber and Hofmann 1984), and sheep (Getty 1975), the gland of water buffaloes was related laterally to the mandibular lymph node. However, it was not laterally connected to the lymph node in the camel (Smuts and Bezuidenhout 1987), mouflon (Saber and Hofmann 1984), and goat (Getty 1975).

The origin, course, and termination of the duct Like the fallow deer (Saber and Hofmann 1984), the duct in water buffaloes consisted of one radicle. However, the duct is formed by the union of the dorsal and ventral radicles in the ox (Getty 1975), and three or four radicles in the sheep (May 1964), or several radicles in the mouflon, and red deer (Saber and Hofmann 1984).

Like the sheep (May 1964) mouflon, red deer, and fallow deer (Saber and Hofmann 1984), the duct in water buffaloes emerges from the lateral surface of the gland. However, the duct emerges from the medial surface of the gland in the ox (Getty 1975), or from the rostral border of the gland in the camel (Smuts and Bezuidenhout 1987; Rezk and Shaker 2017), and ox (Nickel et al. 1979).

The mandibular duct is opened alongside the orifice of the monostomatic gland in the ruminants (Getty 1970; Nickel et al. 1979; Budras et al. 2003; Dyce et al. 2014) and in the sheep (May 1964) or joins the monostomatic gland in the ruminants (Getty 1975; König and Liebich 2004), sheep (May 1964), mouflon, red deer, and fallow deer (Saber and Hofmann 1984), and pampas deer (Pérez et al. 2017). In camels, with regard to the absence of the monostomatic gland, the mandibular duct opens individually to the sublingual caruncle (Smuts and Bezuidenhout 1987). However, no researcher has reported the exact location of the orifice of the mandibular duct relative to the orifice of the monostomatic gland on the sublingual caruncle. In this study, the mandibular duct in water buffaloes was found opening independently medial and slightly rostral to the major sublingual duct on the ventral surface of the sublingual caruncle.

The course of the mandibular duct relative to the monostomatic gland is variable among the ruminants or even among the authors in the same species. The duct passes on the dorsal surface of the gland in the ox (Habel 1970; Budras et al. 2003), on the medial surface of the gland in the ox (Getty 1975; Nickel et al. 1979; König and Liebich 2004), mouflon, red deer, and fallow deer (Saber and Hofmann 1984) or on the ventral surface of the gland in the goat (Nickel et al. 1979). However, in this study, the mandibular

duct passed over the middle of the lateral surface of the monostomatic gland.

Sublingual salivary glands

Like all ruminants except for camels, the gland in water buffaloes consists of monostomatic and polystomatic sublingual glands. In camels, the monostomatic gland is absent (Smuts and Bezuidenhout 1987).

Position The polystomatic gland extends rostrally beyond the level of the first lower premolar tooth (P2) in the red deer (Saber and Hofmann 1984), or to the level of the third lower premolar tooth (P4) in the fallow deer and mouflon (Saber and Hofmann 1984). The exact position of the rostral extension of the gland has not been reported in ruminants (Getty 1975; Nickel et al. 1979; Budras et al. 2003; König and Liebich 2004). However, in this study, the gland was at the level of P2.

The gland extends caudally up to the rostral border of the pterygoideus muscle in the red deer (Saber and Hofmann 1984), or beyond the palatoglossal arch in the fallow deer and mouflon (Saber and Hofmann 1984), and to the level of the palatoglossal arch in the ox (Getty 1975; Nickel et al. 1979) or to the last lower molar tooth (M3) in the sheep (May 1964). Our findings were very similar to those of the ox and sheep.

Like the ox (Nickel et al. 1979; Budras et al. 2003; König and Liebich 2004), the location of the polystomatic gland in water buffaloes was caudal relative to the monostomatic gland. However, the gland lies ventral to the half of the polystomatic gland in the ox (Getty 1975).

Topographic relations of the polystomatic gland relative to surrounding structures in water buffaloes was similar to those of the fallow deer, mouflon, red deer (Saber and Hofmann 1984), and sheep (May 1964).

Monostomatic gland

The gland extends rostrally to the mental foremen in the fallow deer, mouflon, and red deer (Saber and Hofmann 1984), or to the incisive part of the mandible in the ruminants (Nickel et al. 1979). Our findings were similar to those of the animals reported above.

The gland extends caudally to the level of the first premolar tooth (P2) in the red deer (Saber and Hofmann 1984), and to the first molar tooth (M1) in the fallow deer (Saber and Hofmann 1984), or to the level of the incisive part of the mandible in the ruminants (Nickel et al. 1979), or even to the level of the third premolar tooth (P4) in the fallow deer (Saber and Hofmann 1984). The exact location of the caudal extension of the gland has not been documented but it is said that the gland is ventral to the rostral half (Getty 1975) or about the middle of the polystomatic gland (Nickel et al. 1979). Thus, the gland in water buffaloes did not extend as far as caudally as the above-mentioned animals.

The origin, course, and termination of the duct While the duct originates from the medial surface of the gland and is continued after the mandibular duct to the sublingual caruncle in the ox, sheep, goat (Getty 1970; Nickel et al. 1979), the fallow deer, mouflon, and red deer (Saber and Hofmann 1984), the duct in water buffaloes began from the lateral surface of the gland and continued the mandibular duct to the sublingual caruncle.

Morphometry of the major salivary glands and their ducts

Based on morphometric results, the dimensions of the parotid, mandibular, and monostomatic glands of water buffaloes were smaller than those of the ox but were larger than those of other animals (Tables 1, 2, 3). On the other hand, Rezk and Shaker (2017) documented that the parotid gland of the camel was longer than that of the ox, goat, and sheep.

Table 1 Dimensions of the parotid gland and its duct in the various ruminants (cm)

| Animals | Length | Width | Duct length | Duct width |
|--------------------------------------|-----------------------------------|-----------------|-------------|-------------|
| Water buffalo (present study) | 12.89 ± 2.05 | 5 ± 1.0 | 23.3 ± 2.0 | 0.45 ± 0.07 |
| Ox (Dehghani et al. 1994) | 21.1 ± 0.75 | 6.1 ± 0.5 | – | 0.42 ± 0.03 |
| Sheep (Dehghani et al. 2000) | 6.0 ± 0.9 | 3.6 ± 0.5 | – | 0.31 ± 0.1 |
| Goat (Tadjalli et al. 2002) | 8.7 ± 0.9 | 3.9 ± 0.5 | – | 0.34 ± 0.1 |
| Camel (Rezk and Shaker 2017) | 11 ± 0.54 | 7 ± 0.62 | 20 ± 0.41 | 0.5 ± 0.2 |
| Red deer (Saber and Hofmann 1984) | 9.5–10.5 | 3 | – | – |
| Mouflon (Saber and Hofmann 1984) | 6.5 | 3.5 | – | – |
| Fallow deer (Saber and Hofmann 1984) | 7 | Rostral lobe: 3 | 11.5 | – |
| Giraffe (Pérez et al. 2012) | Rostral lobe: 6 Caudal lobe: 8 | Caudal lobe: 4 | – | – |

Note. The author did not record the values which are marked with dash (–). Saber and Hofmann (1984) and Pérez et al. (2012) only reported the mean values

Table 2 Dimensions of the mandibular gland and its duct in the various ruminants (cm)

| Animals | Length | Width | Duct length | Duct width |
|--------------------------------------|------------|----------|-------------|------------|
| Water buffalo (present study) | 16.4±2.6 | 4±0.8 | 34.35±0.12 | 0.1±0.00 |
| Ox (Dehghani et al. 1994) | 31.65±0.65 | 5.2±0.34 | – | 0.28±0.04 |
| Ox (Nickel et al. 1979) | 18–20 | 8–10 | – | – |
| Sheep (Dehghani et al. 2000) | 9.4±1.7 | 3.2±0.4 | – | 0.1±0.02 |
| Goat (Tadjalli et al. 2002) | 9.7±1.7 | 3.3±0.6 | – | 0.17±0.04 |
| Camel (Rezk and Shaker 2017) | 9±0.5 | 3±1 | 28±2 | 0.4±0.08 |
| Mouflon (Saber and Hofmann 1984) | 6 | 2.5 | – | – |
| Fallow deer (Saber and Hofmann 1984) | 7.5 | 3.5 | 18.5 | – |
| Giraffe (Pérez et al. 2012) | – | – | – | – |

Note. The author did not record the values which are marked with dash (–). Nickel et al. (1979), Saber and Hofmann (1984) and Pérez et al. (2012) only reported the mean values

Table 3 Dimensions of monostomatic gland and its duct in the various ruminants (cm)

| Animals | Length | Width | Duct length | Duct width |
|--------------------------------------|----------|-----------|-------------|------------|
| Water buffalo (present study) | 8.48±0.9 | 2.91±0.14 | 10.5±0.95 | 0.1±0.01 |
| Ox (Nickel et al. 1979) | 10–12 | 2–3 | – | – |
| Ox (Habel 1970) | 10 | 1–2 | – | – |
| Red deer (Saber and Hofmann 1984) | 6 | 2 | – | – |
| Fallow deer (Saber and Hofmann 1984) | 6 | 1–1.3 | – | – |
| Mouflon (Saber and Hofmann 1984) | 3.5 | – | – | – |

Note. The author did not record the values which are marked with dash (–). Only the mean values were reported in the ox, red fallow, red deer, and mouflon

The mandibular gland is larger than the parotid gland among various wild and domestic ruminants regardless of camels (Tables 1, 2). The dimensions of the parotid, mandibular, and monostomatic glands and their ducts in the domestic ruminants were greater than those in the wild ruminants (Tables 1, 2, 3). Rezk and Shaker (2017) reported that the length and diameter of the parotid duct of the camel were smaller than those of the ox, but were larger than those of the goat and sheep. However, the dimensions of the parotid and mandibular ducts in water buffaloes were greater than those of the camel (Tables 1, 2).

Although the mandibular gland of the ox is considerably larger than the parotid gland (Getty 1975; Nickel et al. 1979; Dyce et al. 2014), the mandibular gland in water buffaloes was only 3.7 cm larger than the parotid gland. Therefore, the difference between the dimensions of the parotid and mandibular glands in water buffaloes was not as much as those in the ox. The main difference was also related to the shorter length of the mandibular gland in water buffaloes in comparison with the ox (Tables 1, 2, 3). The smaller major salivary glands in water buffaloes as compared with the ox is supported by the following findings: (1) ruminant species ingest more grass (grazers) have smaller salivary glands (Kay 1987; Hofmann 1973, 1988, 1989; Hofmann et al. 2008) and (2) water buffaloes have a low-quality, and high-roughage diet (Czerniawska-Piatkowska et al. 2010).

In summary, the basic arrangement of the glands was constant in all the examined buffaloes except for the orifice of the parotid duct. Comparative anatomy of the major salivary glands among domestic and wild ruminants has clarified that although the presence of the glands is consistent within the reported ruminants, regardless of the absence of the monostomatic gland in camels, there are significant intra- and interspecific variations in detailed morphology and topography of the major salivary glands among the wild and domestic ruminants.

Many considerable morphological differences in the glands of water buffalo and ox were recognized, in spite of morphological similarities (See Abstract). These differences include the lack of the coverage of the parotid lymph node by the parotid gland, the lack of parotid gland reaching the temporomandibular joint, the orifice of the parotid duct, the position of caudal extension of the mandibular gland, the formation of the parotid and mandibular ducts, the site of origin of mandibular and monostomatic ducts, the rostral extension of polystomatic gland, and the exact position of the orifices of the mandibular and monostomatic ducts. From a comparative morphometric viewpoint, the mandibular gland is larger than the parotid gland among various domestic and wild ruminants, regardless of the camel in which the parotid gland is larger than the mandibular. Accordingly, the major salivary glands in water buffaloes almost show large

'grazing ruminants' gross morphological and morphometrical characteristics, following the terminology of Hofmann (1989) and Hofmann et al. (2008). These characteristics consist of lacking filling of retromandibular fossa by parotid gland, mandibular gland reaching more rostral towards the mandible, caudal extension of the polystomatic gland, location of polystomatic gland relative to monostomatic gland, and smaller major salivary glands. However, we should also keep in mind that the classification of ruminants into three feeding types (browser, intermediate feeders, and grazers) was based on morphological investigations of mainly African, European and North American species (Hofmann 1973, 1988) and that most of the ruminant species in Asia have been not examined from this perspective.

Sialography

From the following perspectives, we compare and discuss our findings with the published data on the conventional sialography of animals.

1. *Contrast agents*: Similar to all previously published data (Dehghani et al. 1994, 2000; Tadjalli et al. 2002; Kealy et al. 2010), water-soluble agents were used for sialography in this study. Water-soluble agents are watery to remain within the ductal system for enough time. In addition, they are rapidly diffused and diluted by saliva to make ductal anatomy as clear and sharp as those made by lipid-soluble agents although oily contrast media have inflammatory reactions (Mallya and Lam 2014).

2. *Volume of contrast agents*: The dosage of the contrast media used for the sialography of the parotid gland was 15 ml in the cattle (Dehghani et al. 1994), 3–4 ml in the sheep (Dehghani et al. 2000), and goat (Tadjalli et al. 2002), and 5–10 ml in the camel (Dehghani et al. 1999). In this study, however, the dose was totally 5 ml for the two phases.

While the dosage of the contrast media used for the sialography of the left monostomatic gland was 1.5 ml in the goat (Tadjalli et al. 2002), the dose used for the glands in the current study was totally 2 ml for the two phases.

With regard to the presented comparative morphometric data in Tables 1, 2, 3, it seems that Dehghani et al. (1994, 1999, 2000) and Tadjalli et al. (2002) administrated too much of the contrast agent, which resulted in the extensive acinarization of the glands and consequently obscured almost all the details of the intraglandular system. This has been regarded as one of the causes of sialogram failure according to Mallya and Lam (2014).

3. *Radiographic examination or view*: In all publications regarding sialography of the large animals including the cattle, sheep, goat, horse, and camel, the lateral radiograph of the head was taken like what was done for the water buffalo. However, the ventrodorsal radiograph of the head has been

prepared only in the sialography of the dog (Harvey 1969; Kealy et al. 2010; Evans and de Lahunta 2012).

Dehghani et al. (1994) in cattle reported that in the cadaver studies only a single sialogram could be done because of the partial superimposition of the two glands. The parotid sialograms together with the mandibular and monostomatic sialograms were not present in the cattle, sheep, (Dehghani et al. 1994, 2000) and goat (Tadjalli et al. 2002). However, in the present study, the sialograms of the parotid gland together with the mandibular, and monostomatic glands were demonstrated.

4. *Normal ductal anatomy*: Although sialography is the only modality to visualize the fine branches of the intraglandular ductal system, such information was neglected greatly in sialography of the major salivary glands in the cattle (Dehghani et al. 1994), goat (Tadjalli et al. 2002), sheep (Dehghani et al. 2000), and camel Dehghani et al. 1999). They mainly focused on demonstrating the shape and location of the glands and the extraglandular duct of the parotid and mandibular gland. However, the present study is the first report on sialography of the animals to provide detailed and complete information on the system.

Parotid duct

Like the cattle (Dehghani et al. 1994), and goat (Tadjalli et al. 2002), the course of the main parotid duct or the extraglandular duct in water buffaloes exhibited three angles whereas the course of the main parotid duct had two angles in the sheep (Dehghani et al. 2000).

In this study, the main parotid duct was formed by the union of the two central branches. Although Tadjalli et al. (2002) described that the main parotid duct of goats within the gland was formed by two branches, based on their parotid sialograms it seems that the duct was formed by the union of more than two branches and opacification of the gland was also clearly visible on the sialograms. Moreover, based on the sialograms of the parotid duct in the cattle, camel, and sheep (Dehghani et al. 1994, 1999, 2000), the intraglandular system was obscured because of too much injection of the contrast medium.

Mandibular duct

The main mandibular duct was formed only by one central branch in the water buffalo. Dehghani et al. (1994) explained that the duct of cattle was composed of a rostral branch and a caudal branch, but their mandibular sialograms did not support such a description. Moreover, based on the sialograms of the mandibular duct in the goat (Tadjalli et al. 2002), camel, and sheep (Dehghani et al. 1999, 2000), the intraglandular system was obscured due to the excessive injection of the contrast medium.

According to the sialograms of the mandibular duct in the camel, sheep (Dehghani et al. 1999, 2000), and goat (Tadjalli et al. 2002), the course of the main mandibular duct follows a straight line with very short folds. However, such folds were not noted in the water buffalo, similar to the sialograms in the cattle (Dehghani et al. 1994).

Like the sheep (Dehghani et al. 2000), at the level of the distal end of the stylohyoid bone, the main duct (extraglandular duct) of water buffaloes on its caudal course suddenly curved ventrally to form an angle. In the cattle, however, the caudal branch had a semicircular turn prior to joining the rostral branch (Dehghani et al. 1994). In other words, it had an angle in the central branch of the intraglandular duct. The main mandibular duct in the mandibular sialograms of the goat shows an angle, but the authors have not described the location of the angle (Tadjalli et al. 2002).

Major sublingual duct

Although the sialography of the monostomatic gland was reported only for the goat (Tadjalli et al. 2002), the ductal organization of the gland was obscured due to the administration of too much contrast agent and extensive acinarization of the gland. In addition, the sialography of the gland was not performed in the cattle due to the complex anatomy of the gland (Dehghani et al. 1994). In this study, however, the fine anatomy of the monostomatic ductal system and its course relative to the extraglandular duct of the mandibular duct were defined in details for the first time.

From the clinical perspective, the major salivary glands of water buffaloes can be affected by a variety of diseases which, in the order of frequency, include ectasia (dilatation) and fistula of the parotid duct (Misk et al. 1991, 2014), sialoceles of the mandibular gland (Misk et al. 2014), and ranula in the sublingual gland (Sagar et al. 2010; Krishna et al. 2010; Kamalakar 2016). Although Misk et al. (1991) and (2014) indicated some anatomical reasons for lesions of the ducts, they ignored to mention such reasons for the mandibular sialoceles in water buffaloes. However, based on our findings, a longer main mandibular duct and an angle in the duct can likely play a role in the involvement of the mandibular gland.

Sialographic morphometry

There are few morphometric studies documenting the ducts of ruminants. Dehghani et al. (1994, 1999, 2000) and Tadjalli et al. (2002) recorded the mean diameter of the main parotid and mandibular ducts of the cattle, camel, and sheep on the lateral sialograms. However, they did not measure the length of the parotid and mandibular ducts. According to their findings, the mean widths of the main parotid and

mandibular ducts were 4.2 and 2.8 mm in the cattle, 3.5 and 3 mm in the camel, 3.1 and 1.4 mm in the sheep, and 2.9 and 1.8 mm in the goat, respectively. The mean widths of the main parotid and mandibular ducts were 4.3 and 2.8 mm in water buffaloes, respectively. Thus, the width values in water buffaloes were very similar to those in the cattle although in the present study the mean lengths of the parotid and mandibular ducts were documented for the first time. Moreover, the knowledge of morphometric characterization of the normal major salivary ducts may contribute to understanding their potential etiological roles in salivary diseases of water buffaloes.

In summary, the detailed and comprehensive visualization of extra- and intra-glandular ductal systems of the parotid, mandibular, and monostomatic glands in water buffaloes were done individually and totally in lateral sialograms. The fine and delicate anatomy of the ductal system of the glands was mostly consistent among the specimens. Based on our findings and the reported sialograms in ruminants, it seems that no consistent pattern of intraglandular ductal arrangement and course of the main duct (extraglandular duct) exist. The mean widths of the parotid and mandibular ducts in sialograms of water buffaloes were very similar to those in the ox. In short, the present comprehensive and detailed sialography of ductal morphology and morphometry provide very useful information which leads to an accurate diagnosis of the major salivary gland diseases in live water buffaloes. In this regard, the presence of the sialoceles at the middle of intermandibular space does not reveal the affected side of the mandibular gland and sialography can be helpful in the differential diagnosis of the healthy side from the affected side (Misk 2008; Misk et al. 2014).

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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