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Anatomical and Computed Tomography Study of the Mandible of the Patagonian Huemul (*Hippocamelus bisulcus*): Ecological and Clinical Insights

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ABSTRACT

Given the high prevalence of skeletal and mandibular diseases in the Patagonian Huemul, comprehensive anatomical studies are essential to understand the impact of nutritional deficiencies and support conservation efforts. The aim of this study is to provide a detailed anatomical characterisation of three exhumed mandibles of Patagonian huemul (*Hippocamelus bisulcus*) through three-dimensional reconstructions obtained from computed tomography data and semi-automatic segmentation, documenting their distinctive features. The findings revealed distinctive features in the mandible, consistent with the browsing habits of herbivores, such as a robust coronoid process, a wide and deep pterygoid fossa, a significantly lower mandibular condyle compared to other deer species and a unique configuration of the mandibular canal with a curved caudal portion and a straight rostral portion. These anatomical adaptations are likely related to the species' feeding habits and behaviour. The study also addressed the challenges of researching an endangered species, given that access to biological material is restricted by strict regulations in Chile and Argentina. These restrictions limited the available sample size and hindered the acquisition of additional specimens, which could affect the generalisation of the results. Despite these limitations, the research provides valuable anatomical insights that are fundamental for the biology, clinical practice and management of specimens. In conclusion, the mandibles of both juvenile and adult Patagonian huemul demonstrate distinctive features characteristic of browsing herbivores. The findings can serve as a basis for future comparative studies on mandibular anatomy and function in this endangered deer species as well as in other herbivorous deer.

1 | Introduction

The Patagonian huemul (*Hippocamelus bisulcus*), an endemic deer species found in Chile and Argentina, is classified as endangered by the IUCN Red List (Black-Decima et al. 2016). This species faces significant threats due to anthropogenic factors such as poaching, habitat loss, invasion by predatory species, excessive grazing and diseases transmitted by domestic livestock, leading to a drastic decline in its population (Pine 1993; Vila et al. 2006; Black-Decima et al. 2016). As of 2020, estimates indicated that the Huemul population had reduced to fewer than 1500 individuals. Despite intense conservation efforts and comprehensive studies aimed at identifying the ecological and pathological factors contributing to their decline, Huemul numbers continue to decrease (Flueck et al. 2022). This situation underscores the urgent need for a deeper understanding of Huemul's biology and habitat to devise more effective management and conservation strategies.

The Huemul employs a *concentrated selection* strategy to consume highly nutritious plant materials, distinguishing it from larger ruminants (Escobar, Smith, and Flueck 2020). Adapted with a slender mouth, it can select small, nutrient-rich foods like leaves and shoots. Its diet varies seasonally and spatially, incorporating up to 145 plant species (Vila et al. 2010). Although grasses are usually scarce in their diet due to low digestibility and rapid molar wear from silica, research in Los Glaciares National Park, Argentina, reveals that grasses constitute 46% of their annual intake, compared to 31% from woody plants (Serret and Borghiani 1997). In captivity, Huemuls adapt by consuming both native and exotic plant species (Escobar, Smith, and Flueck 2020). Contrary to the belief that they are mountain deer residing in high-altitude sanctuaries, recent evidence suggests that Huemul inhabit transitional zones between forests and steppes, with historical and contemporary sightings in steppes (Escobar, Smith, and Flueck 2020). Currently, there is intense debate about the determination of the Huemul's natural habitat, whether it be forested areas or steppes (Corti and Díaz, 2023; Flueck et al. 2022). The proposed hypotheses are based on interpretations of historical accounts and documents that record the presence of this species in certain latitudes. In terms of anatomy, comparisons of their appendicular skeletons with those of other cervids place the Huemul between fast runners and walkers/climbers, challenging the notion that they are specialised climbers (Flueck 2021; Huemul Task Force 2012). Clarifying these points will be vital for establishing the foundations of their conservation based on their real distribution, preferred habitat, migratory patterns and trophic ecology (Flueck 2021). Regarding the latter, studying mandibular anatomy could provide data that confirm the diverse trophic ecology of the Huemul.

In mammals, a remarkable diversity in the shape of the mandible is commonly observed, with convergent adaptations often being primarily related to the trophic ecology of the species (Álvarez and Flores 2019). Numerous developmental and experimental studies have elucidated the relationship between structure and function; namely, the specialisations of herbivorous mammals vary according to the type of vegetation on which they feed, which is considerably diverse (Clauss, Kaiser, and Hummel 2008; Janis and Fortelius 1988). However, this does not discount the influence of phylogenetic factors, as demonstrated in the traits of ungulate mandibles (Zhou et al. 2019; Pérez-Barbería and Gordon 1999), or the impact of hormonal or environmental factors. Nonetheless, evolutionary history has shown that early mandible shapes were optimised for rapid closure and stress resistance but became less optimal for these functions over time, leading to diverse feeding strategies (Deakin et al. 2022). In cervids and other herbivorous mammals, the mandible fulfils various functions; it provides a robust base for the insertion of masticatory muscles necessary for the efficient processing of fibrous and tough plant materials (Cassini and Toledo 2021). It also articulates the teeth, which have adapted to a herbivorous diet, such as the molars used for grinding and chewing vegetation. For the Patagonian huemul, the mandible plays a significant role in social behaviour and reproduction (Escobar, Smith, and Flueck 2020). Males use their antlers and mandibles to compete for mates and establish dominance within their social groups.

In addition to the ecological and conservation aspects, the detailed anatomical characterisation of the Patagonian huemul's mandible has significant clinical implications, particularly in the context of locoregional anaesthesia techniques. A precise understanding of mandibular anatomy is essential for the safe and effective application of anaesthesia in clinical and surgical procedures in this species. However, to date, there is no detailed anatomical or tomographic description of this structure in the scientific literature. This study represents the first comprehensive effort to document the mandibular anatomy of the Patagonian huemul through three-dimensional reconstructions based on computed tomography data and semi-automatic segmentation. The lack of detailed studies limits our understanding of the anatomical adaptations that enable the huemul to feed on a variety of plants and how these adaptations affect its trophic ecology and migratory behaviour.

Given the high prevalence of diseases affecting the skeletal system and mandible of the Patagonian huemul, which hinder population recovery by influencing feeding behaviour and ecology (Flueck and Smith-Flueck 2017; Flueck 2018), comprehensive anatomical studies are urgently needed. This necessity is emphasised by the fact that the mandible in this species has predominantly been studied from a comparative and evolutionary perspective (Heckeberg, 2020) and through clinical case studies (Escobar, Smith, and Flueck 2020; Flueck and Smith-Flueck 2008). These anatomical studies are also essential to understand the impact of nutritional deficiencies on bone thickness and structure described in the skull (Núñez-Cook, Vidal-Mugica, and Salinas 2023, 2022) and on the appendicular skeleton (Salinas, Núñez-Cook et al. 2020; Salinas, Arenas-Caro et al. 2020; Núñez-Cook, Vidal-Mugica, and Salinas 2022) as well as the 63% prevalence of mandibular osteopathies described by Flueck and Smith-Flueck (2008).

In recent years, computed tomography (CT) and 3D reconstruction techniques applied to wildlife have advanced significantly, enabling detailed and non-invasive visualisation of complex anatomical structures, such as the mandible (Ribeiro et al. 2022; İşbilir and Güzel 2023). These technologies have been particularly useful in veterinary studies, facilitating the precise evaluation of pathologies, surgical planning and understanding of mandibular morphology across various species. In conservation contexts, 3D CT has been crucial for studying endangered species like the Patagonian huemul, where access to biological material is limited, offering a valuable tool for detailed anatomical analysis of bone structures and their correlation with ecological adaptations and feeding behaviours (Farha et al. 2021; Keneisenuo et al. 2022).

Based on this background, our research question is: how does the anatomy of the Patagonian huemul's mandible, as revealed through three-dimensional reconstructions and semi-automatic segmentation, reflect its diverse trophic ecology and enhance our understanding of its natural habitat? The aim of this study is to provide a detailed anatomical characterisation of the mandible of the Patagonian huemul (*Hippocamelus bisulcus*) using computed tomography and semi-automatic segmentation. By documenting its distinctive features without making causal inferences or testing specific hypotheses, this descriptive approach seeks to establish a basic understanding of the huemul's biology, facilitate more accurate clinical assessments, improve conservation and specimen management strategies and guide future research.

2 | Materials and Methods

2.1 | Location and Study Design

The study was conducted at the Laboratory of Animal and Experimental Morphology at the Institute of Biology of the Pontificia Universidad Católica de Valparaíso. A descriptive and cross-sectional design was used.

2.2 | Specimen Collection

The mandibles of Patagonian huemuls were exhumed and subsequently identified ($n_{\text{total}} = 3$; males = 2; female = 1) (Table 1). All the studied mandibles were legally obtained under the Hunting Law no.19473 framework and resolution no. 1490 (22 December 2003) of the Agricultural and Livestock Service of Chile (SAG, in Spanish). Some specimens were collected from the wild, while others were obtained from captive breeding conservation projects. The causes of death of the studied specimens were parasites, caseous lymphadenitis and intra-specific fights. Given the high heritage value of the specimens, their transportation, handling and storage were performed according to Simmons and Muñoz-Saba (2005). Mandibles with severe post-mortem damage that hindered accurate anatomical assessment were excluded from the present study. The evaluation of the mandible was limited to the right side. This approach was chosen to maintain consistency and ensure accurate comparisons in both the CT and anatomical studies.

2.3 | External Anatomy Description

The external anatomy of the mandible was described based on simple observation and linear measurements (Figure 1), following anatomical studies in deer (Von den Driesch 1976; Onuk, Kabak, and Atalar 2013) and goats (Onuk, Kabak, and Atalar 2013; Wang et al. 2021). Thus, documenting these findings is crucial, following the precedent set by earlier works on the osteology of the Patagonian huemul (Núñez-Cook, Vidal-Mugica, and Salinas 2022, 2023; Salinas, Núñez-Cook et al. 2020; Salinas, Arenas-Caro et al. 2020).

2.4 | CT Scan Protocol for Mandibular Imaging

The mandibles included in this study belonged to both adult and juvenile Patagonian huemuls (Table 1). The age classification was based on information from park ranger records, which provided detailed data on the individual animals. After exhumation, the mandibles were cleaned of any remaining soft tissue using gentle mechanical methods. They were then air-dried in a controlled environment to prevent any structural changes. No fluids were used during the preparation to avoid potential alterations in the bone tissue, ensuring that the anatomical features remained intact for accurate imaging and analysis. The length of the scans was approximately 250 mm, covering the entire mandibular region from the condylar process to the symphysis. The mandibles were positioned prone on the CT table with the occlusal plane parallel to the table surface. CT positioning lasers were aligned along the midline of the mandible to ensure consistent and accurate imaging of the entire mandibular structure.

2.5 | CT Scanning and Image Acquisition

The mandibular canal was studied using a 16-slice helical computed tomography (CT) scanner (Mx8000 IDT 16, Philips Medical Systems DMC GmbH, Hamburg, Germany). Transverse images of the mandible were acquired in axial scanning mode using the following parameters: 120 kV, 250 mAs, 1.0 mm image collimation, 0.75 s tube rotation time, 0.75 pitch, 1 mm section thickness, and a matrix of 512 × 512 pixels. Subsequently, the images were

TABLE 1 | Identification of Patagonian huemul skulls and mandibles along with information regarding sex, skull weight, mandible weight, age and origin (M: male; F: female).

ID mandible	Sex	Skull weight (kg)	Specimen weight (kg)	Age (years)	Origin
1	M	0.302	75	1.5	Fauna Andina (FA), Región de La Araucanía, Villarrica 39°16′00″ S, 72°13′00″ W; 227 m.a.s.l., Chile
2	M	0.489	77	2	Fauna Andina (FA), Región de La Araucanía, Villarrica 39°16′00″ S, 72°13′00″ W; 227 m.a.s.l., Chile
3	F	0.306	73	7	Cerro Castilla (CC), Region de Aysén del General Carlos Ibañez del Campo, 46°03′00″ S, 72°11′00″ W; Chile

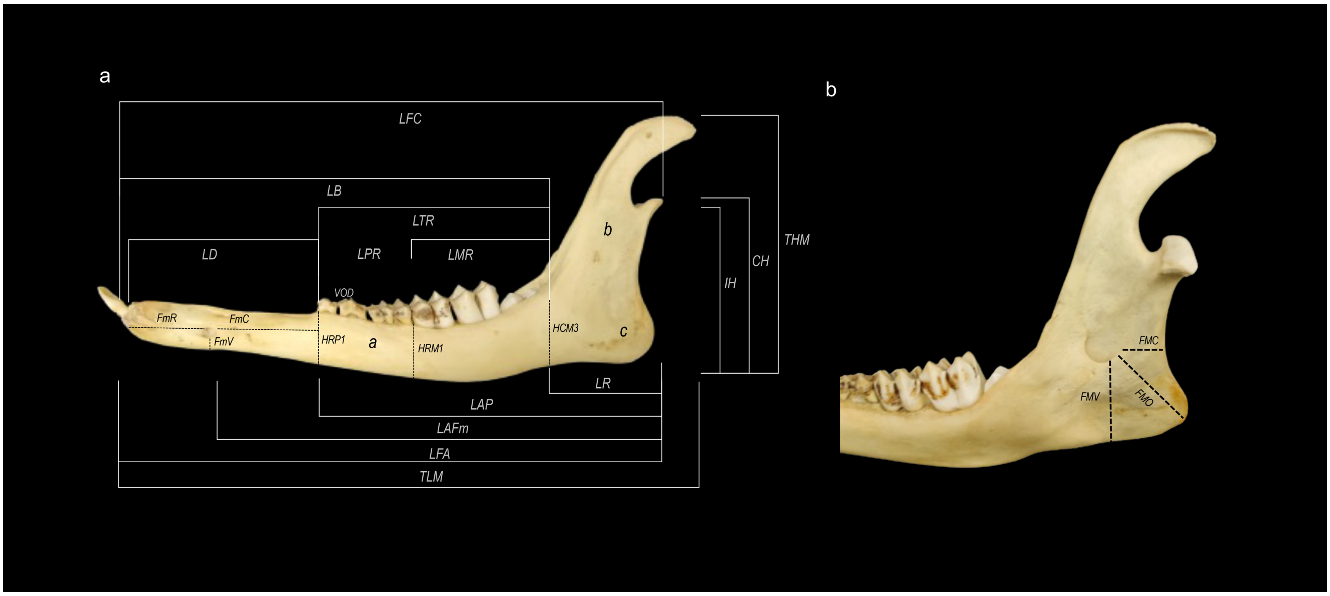
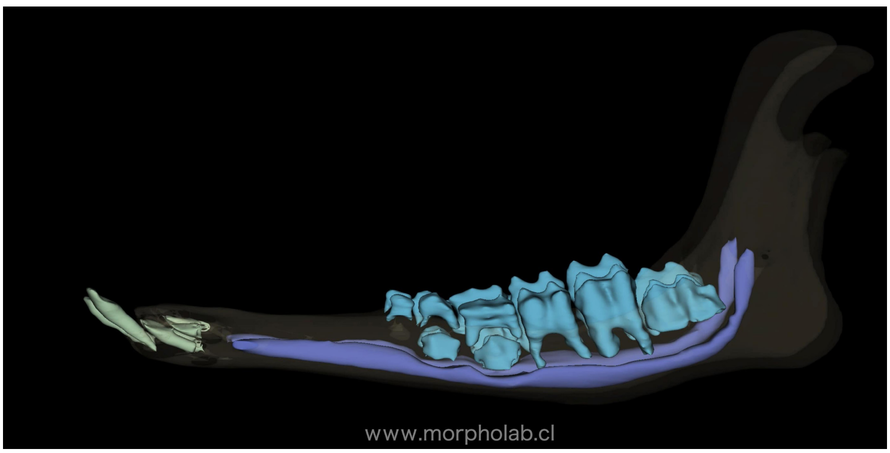


FIGURE 1 | Linear measurements in the Patagonian huemul mandible. (a) Lateral view: (A) body, (B) ramus and (C) angle. Total mandibular height (THM), total mandibular length (TLM), length from angle to the first incisor alveolus (LFA), length from condyle to the first incisor alveolus (LFC), ramus length (LR), body length (LB), the length between angle and first premolar (LAP), the length between angle and mental foramen (LAFm), tooth row length (LTR), molar row length (LMR), premolar row length (LPR), diastema length (LD), height between angle and mandibular condyle (CH), height between angle and mandibular notch (IH), the distance between mental foramen and first incisor (FmR), the distance between ventral margin of mandible and mental foramen (FmV), distance between rostral margin of first premolar and mental foramen (FmC), height of mandibular body rostral to the first molar (HRP1), rostral to the first molar (HRM1) and caudal to the third molar (HCM3). (b) Medial view: distance between caudal margin of mandible and mandibular foramen (FmC), distance between ventral margin of angle and mandibular foramen (FMV), distance between angle of mandible and mandibular foramen (FMO).



VIDEO 1 | 3D reconstruction of the mandible of the Patagonian huemul. Age: 2 years (male). Right lateral view. In blue: mandibular canal. 'The video shows a 3D reconstruction of the mandible of the Patagonian huemul, focusing on the right lateral view. During playback, the mandibular canal is highlighted in blue, visible along the body of the mandible. The structure is carefully segmented to emphasise the mandibular canal and other components of the bone anatomy'.

reconstructed using a window width of −500 HU (Hounsfield units) and a window level of 2000 HU (smoother kernel).

2.6 | Image Analysis and 3D Reconstruction

For image analysis, the Slicer software version 4.11.2021026 software (Fedorov et al. 2012) with the SlicerMorph extension for DICOM files was used (Rolfe et al. 2021), allowing for the import, visualisation and analysis of 3D models (Video 1). This process included tools for manual and semi-automatic segmentation as well as the creation of high-precision three-dimensional models. Semi-automatic segmentation was performed using the Segment Editor tool in Slicer, which allowed for the delineation and separation of specific anatomical structures within the tomography images. To enhance the precision of the segmentation, additional modules provided by SlicerMorph were utilised, such as Segment Editor Extra Effects, which offered additional segmentation filters, and Lights, which controlled the 3D shading for better visualisation. Linear distances were measured, and shape, volume and surface area analyses were conducted within the mandibular canal using the 3D images obtained from CT scans, following the methods described by Suazo Galdames et al. (2007). Additionally, in transverse section images, the maximum and minimum diameters of the mandibular and mental foramina were measured. The linear distances were categorised by sex to evaluate sexual dimorphism in the mandible (Bucchi, Bucchi, and Fuentes 2016; Morrison and Whitridge 1997; Ohtaishi 1980). Analysis and measurements were conducted exclusively on the right mandibles. This decision was based on preliminary assessments that showed no significant anatomical differences between the right and left mandibles. Focusing on the right mandibles allowed for a standardised approach and ensured consistency in the data collection process.

All quantitative data were organised using Microsoft Excel spreadsheet software and expressed as means and standard deviations. The terminology used throughout this study was based on the (revised) 6th edition of the *Nomina Anatomica Veterinaria* (NAV) (ICVGAN 2017).

3 | Results

3.1 | External Anatomy of the Mandible

The Patagonian huemul presented two mandibles articulated by a symphysis (*Articulatio intermandibularis*), which showed a clearly distinguishable body, ramus and angle. The body (*Corpus mandibulae*) and mandibular ramus (*Ramus mandibulae*) were prominent and formed an obtuse angle. Both mandibles formed a virtual intermandibular space in the shape of an isosceles triangle (Figure 2). The linear measurements of the mandible are shown in Table 2.

The body of the mandible had a labial surface (*Facies labialis*), where the mental foramen (*Foramen mentale*) was observed, either single or with an accessory foramen located rostro-dorsally. The lingual surface (*Facies lingualis*) exhibited a smooth surface and a slightly pronounced mylohyoid line (*Linea mylohyoidea*). The articulation intermandibular was composed of interdigitated septa that fit into depressions on the contralateral mandible. The body of the mandible had 10 alveoli. The incisive portion (*Pars incisiva*) presented three incisor alveoli and one alveolus for a canine tooth. The molar portion (*Pars molaris*) displayed three premolar alveoli and 3 M alveoli. The latter were divided into two compartments by an interradicular septum, except for the third molar, which had two interradicular septa. One mandible presented an incomplete interradicular septum of the third molar. Between the canine and the first premolar, a wide diastema was observed, represented by the interalveolar margin (*margo interalveolaris*), which was straight, smooth and with a fine dorsal margin, giving the mandible a graceful shape.

The mandibular ramus exhibited a slightly pronounced angle (*Angulus mandibulae*), forming an acute ventral margin and a prominent tuberosity for the sternomandibular muscle (*Tuberositas m. sternomandibularis*). Notably, the angle of the female mandible (ID#3) displayed a more concave boundary with the body compared to the other specimens, which, in contrast, showed a straighter continuation between both structures. On the lateral surface, there was a flat, slightly pronounced and rough masseteric fossa (*Fossa masseterica*) in its caudal portion.

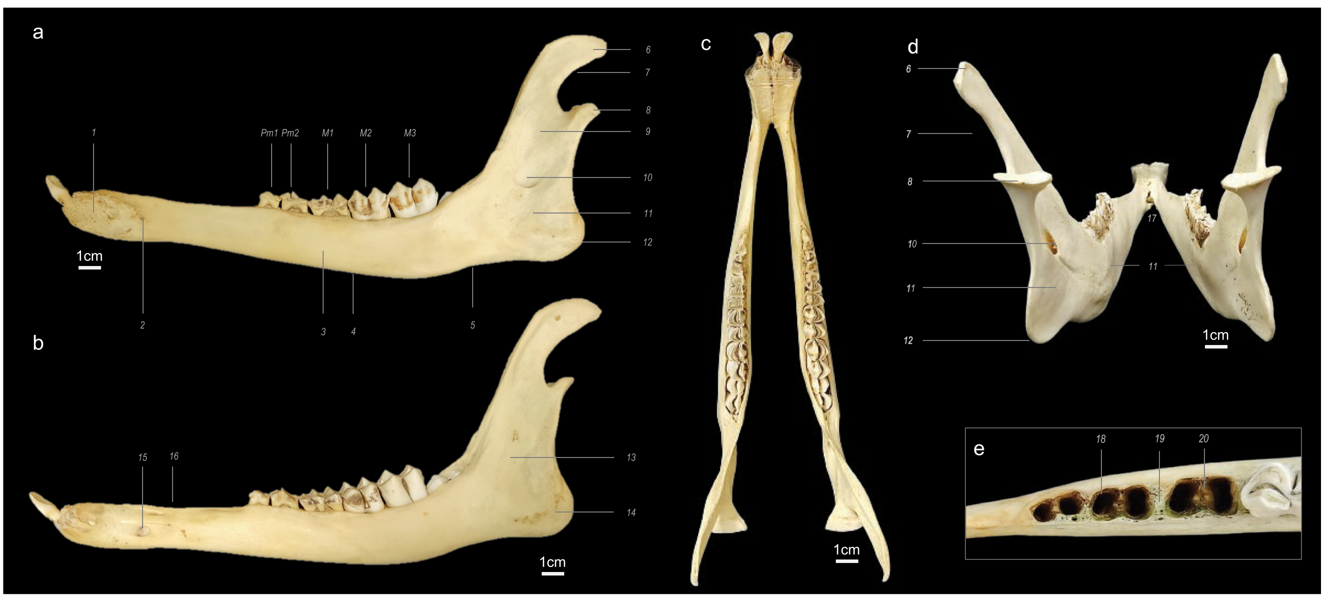


FIGURE 2 | Mandible of the male Patagonian huemul (age: 2 years). (a) Medial view, (b) lateral view, (c) dorsal view, (d) caudal view of articulated mandibles, (e) dorsal view of alveoli (female). (1) articular face; (2) incisive portion; (3) molar portion; (4) ventral margin; (5) incisure for facial vessels; (6) coronoid process; (7) mandibular notch; (8) mandibular condyle; (9) pterygoid fossa; (10) mandibular foramen; (11) mylohyoid groove; (12) mandibular angle; (13) masseteric fossa; (14) sternomandibular muscle tuberosity; (15) mental foramen and accessory foramen; (16) interalveolar margin; (17) intermandibular space; (18) alveolus (for root of the second premolar); (19) interalveolar septum; (20) interradicular septum; M, molar; Pm, premolar.

TABLE 2 | Detailed linear measurements of the mandibles from three Patagonian huemul deer specimens (two males and one female).

Variable	Male ($n=2$)	Female ($n=1$)	Total ($n=3$)
TMH	9.28 ± 0.35	9.77	9.44 ± 0.35
TML	20.07 ± 2.27	22.7	20.94 ± 1.52
LFC	19.7 ± 0.41	22.0	20.46 ± 1.36
LR	4.25 ± 0.45	6.8	5.1 ± 1.51
LB	6.05 ± 0.67	6.87	6.32 ± 0.68
LAP	13.31 ± 0.47	15.01	13.87 ± 1.04
LAFm	16.67 ± 0.0	19.13	17.49 ± 1.42
LTR	8.96 ± 0.42	8.57	8.83 ± 0.38
LMR	5.1 ± 0.09	5.29	5.16 ± 0.13
LPR	3.79 ± 0.11	3.45	3.68 ± 0.21
LD	6.53 ± 0.6	7.45	6.83 ± 0.68
CH	6.23 ± 0.17	6.27	6.24 ± 0.12
IH	5.95 ± 0.04	5.96	5.95 ± 0.03
FMC	1.45 ± 0.12	1.57	1.49 ± 0.11
FMV	2.73 ± 0.09	3.03	2.83 ± 0.19
FMO	3.1 ± 0.37	3.07	3.09 ± 0.23
FmR	2.18 ± 0.35	2.3	2.22 ± 0.26
FmV	0.53 ± 0.14	0.83	0.63 ± 0.2
FmC	3.5 ± 0.61	3.93	3.64 ± 0.5
HRP1	1.84 ± 0.08	2.32	2.0 ± 0.28
HRM1	1.98 ± 0.11	2.37	2.11 ± 0.24
HCM3	2.95 ± 0.01	3.34	3.08 ± 0.23

Abbreviations: CH, condyle height of the mandible; FMC, distance between the caudal margin of the mandible and the mandibular foramen; FmC, distance between the rostral margin of the first premolar and the mental foramen; FMO, distance between the angle of the mandible and the mandibular foramen; FmR, distance between the mental foramen and the first incisor; FMV, distance between the ventral margin of the angle and the mandibular foramen; FmV, distance between the ventral margin of the mandible and the mental foramen; HCM3, body height by caudal to the third molar; HRP1, body height by rostral to the first premolar; IH, incisura height of the mandible; LAFm, length between angle and mental foramen; LAP, length between angle and first premolar; LB, body length; LD, diastema length; LFA, mandibular length from the angle to the first incisor alveolus; LFC, mandibular length from the condyle to the first incisor alveolus; LMR, molar row length; LPR, premolar row length; LR, ramus length; LTR, tooth row length; TLM, total mandibular length; TMH, total mandibular height.

Additionally, a deep pterygoid fossa (*Fossa pterygoidea*) was prominent on the medial surface, housing the oval and elongated mandibular foramen (*Foramen mandibulae*). This corresponded to the beginning of the mandibular canal (*Canalis mandibulae*). Ventral to the mandibular foramen, the start of the mylohyoid groove (*Sulcus mylohyoideus*) was observed, which was more pronounced in the female mandible (ID#3) compared to the males.

Regarding the coronoid process (*Processus coronoideus*), in both males and females, it is flat, medially concave and laterally convex. The observed difference between sexes is that, in males, the coronoid process has a roughened tip, surpassing the height of the mandibular condyle (*Processus condylaris*). The mandibular condyle presented a curved shape, projecting caudally. The mandibular notch (*Incisura mandibulae*) separated the condylar process from the mandibular condyle. The latter had a short neck (*Collum mandibulae*), a prominent head (*Caput mandibulae*) and a convexity dorsally; its articular surface had an oval appearance. Each mandible is articulated with 10 teeth, including 3 incisors, 1 canine, 3 premolars and 3 molars (Figure 2).

3.2 | Mandibular Canal Anatomy

The mandible featured an intraosseous mandibular canal in each bone. It originated at the mandibular foramen, ventral to the temporal muscle's insertion area on the medial (lingual) surface of the mandibular ramus and extended through the mandibular body to the mental foramen on the lateral (vestibular) surface of the mandible.

The mandibular canal comprised three segments: a caudal segment, stretching from the mandibular foramen to the caudal margin of the third molar, oriented rostrally at a 60° angle. Its path started at the medial face of the ramus and continued to the middle of the mandible. An intermediate segment extended from the caudal margin of the third molar to the rostral margin of the first premolar, displaying a slight concave curvature dorsally. Located in the middle of the mandibular body, this segment neared the ventral margin of the mandible along its course, leading to the formation of small alveolar canals for the molar and premolar teeth. A rostral segment, starting at the rostral margin of the first premolar and terminating at the mental foramen, was horizontal and directed laterally in its final portion. Near the mental foramen, a small canal emerged for the accessory mental foramen and rostrally, alveolar canals for the incisor teeth branched off. In transverse sections, each segment exhibited a distinct shape. The caudal segment was wedge-shaped with the apex dorsally oriented. The intermediate segment resembled a water droplet with the apex oriented laterodorsally at the caudal end and was oval-shaped with its major axis horizontal at the rostral end. The rostral segment presented an oval shape with its major axis vertical, concluding in a circular shape in a short segment near the mental foramen.

Throughout its trajectory, the canal was lined by walls of cortical bone. In the rostral and intermediate segments, compact bone was prevalent, while trabecular bone was found in the walls of the caudal segment and the incisive alveolar canals (Figures 3 and 4). Dorsal canals were noted, presumably associated with dental alveoli for incisors, premolars and molars. Morphometric measurements of the mandibular canal are detailed in Table 3.

4 | Discussion

We have conducted an anatomical and morphometric description of the Patagonian huemul deer's mandible, and we present the first report detailing the mandibular canal using 3D segmentation based on CT data. CT proved to be a highly effective tool and was particularly valuable in describing the

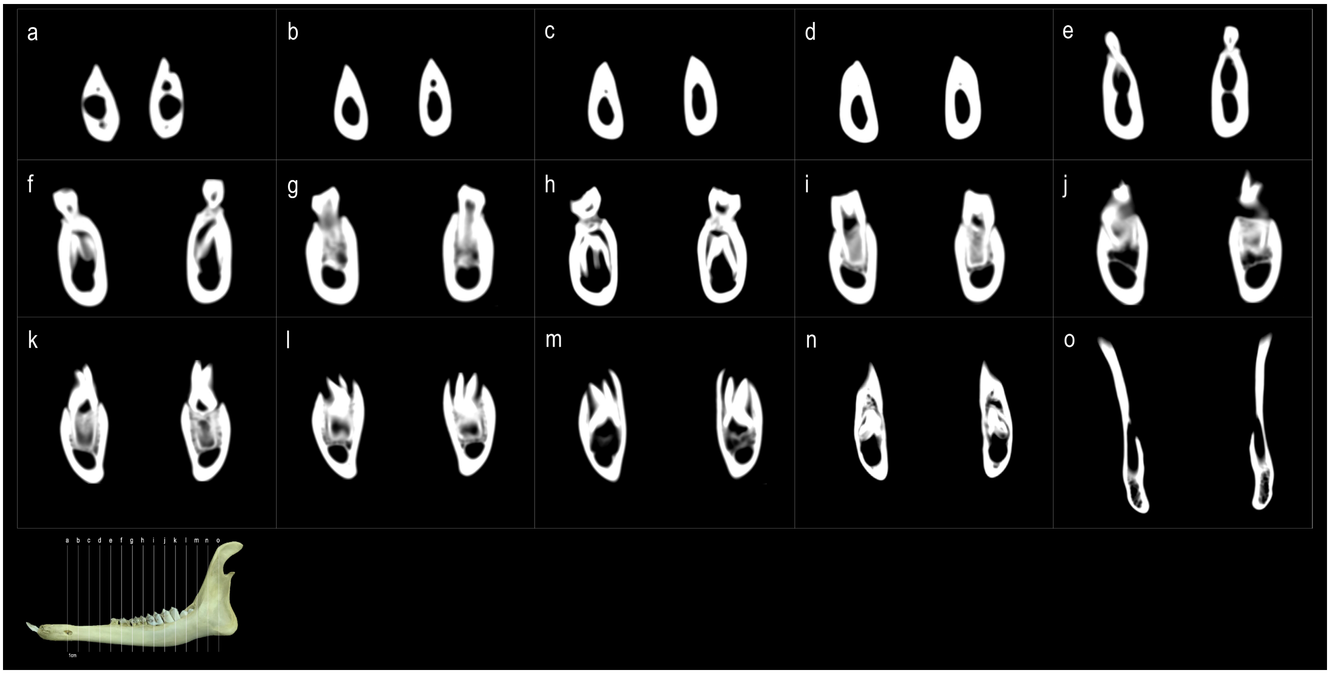


FIGURE 3 | Mandible of the male Patagonian huemul (age: 2 years). Series of transverse sections obtained from computed tomography. From rostral to caudal, separated every 1 cm from the caudal margin of the mental foramen (a) to the mandibular foramen (o).

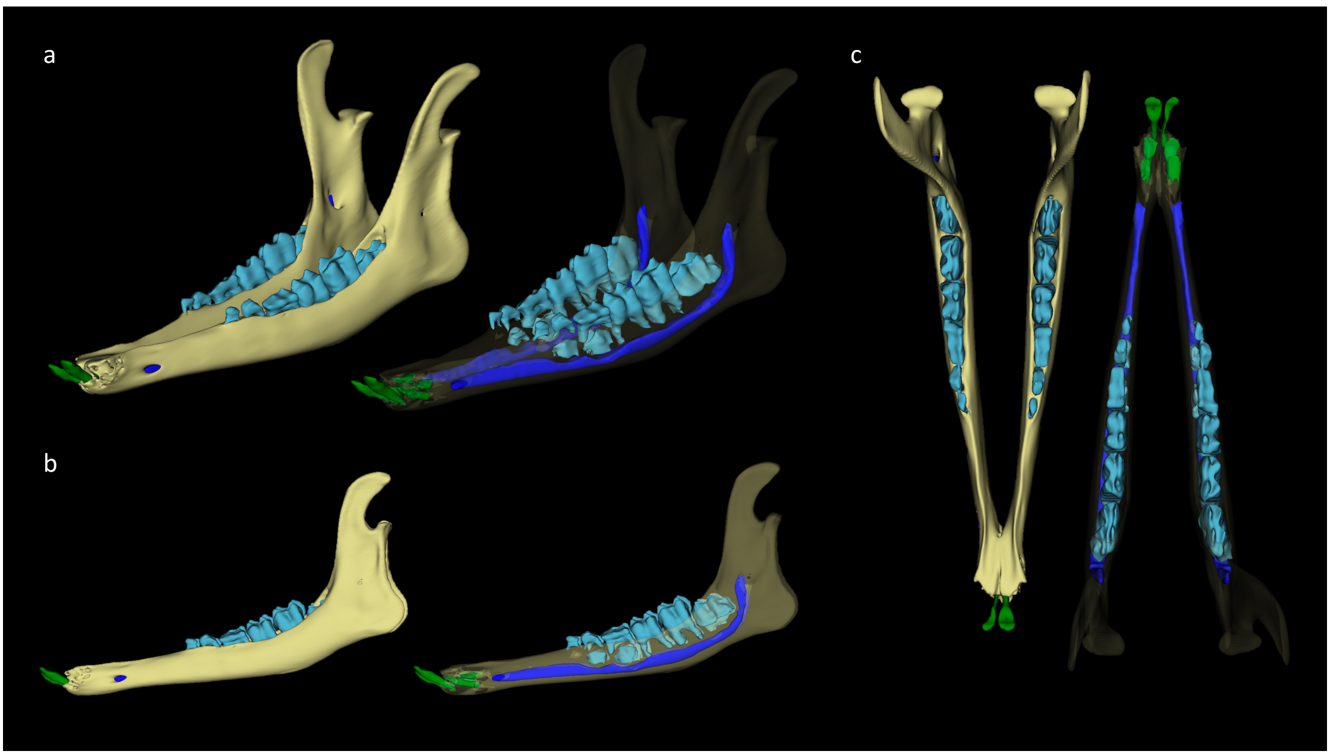


FIGURE 4 | 3D reconstruction of the mandible of the male Patagonian huemul (age: 2 years). (a) Right rostrolateral view, (b) lateral view, (c) dorsal view. In green: incisor teeth; blue: mandibular canal; light blue: premolars and molars.

mandibular canal in the Patagonian huemul deer. This non-invasive technique allowed for a detailed three-dimensional reconstruction of its conformation within the mandible, avoiding interference from adjacent structures that could hinder the study. Although the mandible of the Patagonian huemul deer shares common characteristics with other cervids, such as the fact that it is a paired bone, the presence of an extensive diastema, and a coronoid process projected dorsally (Rees 1969; Pinder and Grosse 1991; Azorit, Analla, and Muñoz-Cobo 2003; Onuk, Kabak, and Atalar 2013; Castaños 2017; Saldivia and Villegas 2019; Coacalla, Curie, and Flores 2021; Cassini and Toledo 2021; Keneisenuo et al. 2022), it also presents particular

TABLE 3 | Detailed linear measurements (mean ± SD; cm) volume (cm^3) and surface (cm^2) of the mandibular canal from three Patagonian huemul deer specimens (two males and one female).

Variable	Male (*n* = 2)	Female (*n* = 1)	Total (*n* = 3)
MDMF	14,4 ± 0.28	13.3	14.03 ± 0.66
mDMF	10.15 ± 1.2	8.7	9.67 ± 1.19
MDmF	9.6 ± 0.57	8.6	9.27 ± 0.70
mDmF	4.1 ± 1.7	3.3	3.83 ± 1.28
MDC1	4.8 ± 0.71	5.0	4.8 ± 0.51
mDC1	4.2 ± 0.71	3.0	3.13 ± 0.51
MDC2	5,85 ± 0.21	7.6	6.43 ± 1.02
mDC2	2.8 ± 0.28	2.7	2.77 ± 0.21
MDC3	7.1 ± 0.14	7.4	7.2 ± 0.2
mDC3	2.5 ± 0.28	3.0	2.67 ± 0.35
MDC4	6.6 ± 0.42	8.0	7.07 ± 0.86
mDC4	2.85 ± 0.78	3.0	2.9 ± 0.56
MDC5	5.25 ± 0.78	8.3	6.27 ± 1.84
mDC5	3.3 ± 0.57	3.4	3.33 ± 0.4
MDC6	4.65 ± 0.92	7.7	5.67 ± 1.88
mDC6	3.95 ± 1.06	4.3	4.07 ± 0.78
MDC7	5.15 ± 0.35	7.3	5.87 ± 1.27
mDC7	4.5 ± 0.71	6.0	5.0 ± 1.0
MDC8	5.4 ± 0,14	7.3	6.03 ± 1.1
mDC8	3.7 ± 2.26	7.0	4.8 ± 2.48
MDC9	8.05 ± 3.18	6.6	7.57 ± 2.4
mDC9	5.45 ± 1.2	6.3	5.73 ± 0.98
MDC10	6.05 ± 0.49	6.3	6.13 ± 0.38
mDC10	6.0 ± 0.42	5.3	5.77 ± 0.5
MDC11	6.6 ± 0.42	6.6	6.6 ± 0.3
mDC11	5.45 ± 0.78	6.3	5.73 ± 0.74
MDC12	5.35 ± 0.07	6.7	5.8 ± 0.78
mDC12	4.8 ± 0.28	6.6	5.4 ± 1.06
MDC13	5.6 ± 0.57	6.4	5.87 ± 0.61
mDC13	4.85 ± 0.63	5.3	5–0 ± 0.51
MDC14	7.1 ± 0.28	11.7	8.63 ± 2.66
mDC14	5.95 ± 0.49	5.4	5.77 ± 0.47
MDC15	12.4 ± 1.98	17.0	13.93 ± 3.0
mDC15	2.95 ± 0.35	6.0	3.97 ± 1.78
Volume	5.47 ± 1.69	6.73	5,89 ± 1.4
Surface	42.16 ± 16.34	43.21	42.52 ± 11.57

Note: The measurements were performed according to Figure 3.
Abbreviations: MDC, maximum and mDC, minimum diameter in cross-sectional view according to the height of the mandibular canal; MDMF, maximum diameter of the mandibular foramen; mDMF, minimum diameter of the mandibular foramen; MDmF, maximum diameter of the mental foramen; mDmF, minimum diameter of the mental foramen.

aspects of interest from an eco-morphological and clinical-surgical perspective.

4.1 | Eco-Morphological Aspects

The mandible of the Patagonian huemul deer presented characteristics useful for identifying or differentiating individuals and/or cervid populations. A noteworthy aspect is the presence of a robust coronoid process whose caudal inclination is more pronounced than reported in Red Deer (*Cervus elaphus*, Azorit, Analla, and Muñoz-Cobo 2003), White-tailed Deer (*Odocoileus virginianus*; Rees 1969), Marsh Deer (*Blastocerus dichotomus*, Pinder and Grosse 1991), Reindeer (*Rangifer tarandus*; Castaños 2017), Indian Muntjac (*Muntiacus muntjak*; Keneisenuo et al. 2022) and Sambar Deer (*Rusa unicolor*; Keneisenuo et al. 2022). This characteristic provides a larger anchoring surface for the tendons of the masticatory muscles. The graceful shape of the mandible, a slightly pronounced masseteric fossa, and a straight diastema are consistent with an animal whose diet is primarily composed of dicotyledons (Cassini and Toledo 2021). Additionally, both the temporal fossa in the skull (Núñez-Cook, Vidal-Mugica, and Salinas 2022) and the pterygoid fossa in the mandible are deep and wide, suggesting that the temporal muscle has an agonist function in closing the mandibular angle, with the masseter muscle acting synergistically during chewing, and additionally suggests the presence of strong pterygoid muscles. This relationship enhances mandibular retraction and diduction, which is beneficial for browsing animals (Cassini and Toledo 2021). The height of the mandibular condyle is considerably lower than reported in the Marsh Deer (*Blastocerus dichotomus*, Pinder and Grosse 1991), White-tailed Deer (Rees 1969) and Reindeer (Castaños 2017). This enables the mouth to open at a wider angle, aligning with observations in animals whose diets incorporate fruit consumption (Cassini and Toledo 2021; Escobar, Smith, and Flueck 2020). Regarding dentition, it is specialised and adapted to its herbivorous diet (Escobar, Smith, and Flueck 2020), with molars featuring low crowns and narrow, tall crests for efficient grinding (Clauss, Kaiser, and Hummel 2008; Janis and Fortelius 1988). Concerning the mandibular canal, a curved caudal portion and a straight rostral portion are highlighted, which differs from what has been reported in the sambar and muntjac deer (Keneisenuo et al. 2022). Furthermore, differences were observed in the orientation of the canal compared to domestic carnivores such as canines and felines (Ascaso, Whyte, and Trobo 2021; Martinez et al. 2009) and wild ones like the Common field fox (*Lycalopex vetulus*, Magalhães, Romão et al. 2019), which present a horizontal orientation in the mandibular ramus (Ascaso, Whyte, and Trobo 2021). These differences are likely attributable to the particular shape of the mandible in the Patagonian huemul deer, which has an obtuse angle between the body and the ramus, and a straight interalveolar margin in the diastema, typical of browsing cervids. In contrast, grazing cervids exhibit an acute angle between the ramus and the body and a curved interalveolar margin (Cassini and Toledo 2021). This anatomical feature suggests that the diverse configurations of the mandibular canal result from the inherent mandible shape, the arrangement of teeth, and its biomechanics. It indicates that an animal's trophic ecology can influence both the external morphology and the internal structure of the mandible.

It is noteworthy that, for most of the mandibular canal's course, it is surrounded by compact bone rather than spongy bone, as has been reported in humans (Homo sapiens, Suazo Galdames et al. 2007) or domestic canines (Martinez et al. 2009). The distribution of bone tissue types observed in the Patagonian huemul deer has also been reported in Göttingen Minipig pigs (*Sus domesticus*, Corte et al. 2017), rabbits (Evcim 2021), domestic ruminants (Evcim 2021), and other cervids such as the sambar and muntjac deer (Keneisenuo et al. 2022), which can be explained by the herbivorous feeding behaviour of these animals, as it is known that compact bone in the mandible can bear greater loads than spongy bone (Bozkaya, Muftu, and Muftu 2004). As for the volume and diameter of the mandibular canal in other cervids, no previous literature on the subject was found. However, the volumetric data of the mandibular canal reported in Göttingen Minipig pigs are of interest, with a difference in volume and diameter between individuals of 12 and 21 months of age, with these values being larger than those obtained in our study, despite the pig's mandible being smaller compared to that of the Patagonian huemul deer (Corte et al. 2017). Regarding the volume and diameter of the mandibular canal in other cervids, no previous literature on the subject was found. However, volumetric data from the mandibular canal reported in Göttingen Minipig pigs indicate a difference in volume and diameter between individuals aged 12 and 21 months, with these values being larger than those obtained in the present study, despite the pig's mandible being smaller compared to that of the Patagonian huemul deer (Corte et al. 2017). As the individual grows, both the volume and diameter of the mandibular canal increase due to the gradual loss of spongy bone adjacent to the tooth roots. Although the Patagonian huemul deer does not show drastic changes in the shape and volume of the mandibular canal like those reported in the Göttingen Minipig, there may be a similar cause that determines the type of tissue surrounding this canal, as documented in this study. This inference is based on the differentiation process and changes in the spongy bone as the animal matures, transitioning from a nursing fawn to an adult that primarily browses (Escobar, Smith, and Flueck 2020). This change would imply a greater biomechanical demand on the mandible, evident in the mylohyoid groove and mandibular angle, in adult specimens compared to the juvenile mandibles. The reported linear distances are useful as references for zooarchaeological and biomedical studies. The present study categorised them by sex based on their significance in detecting sexual dimorphism in mandibular anatomy (Hohl et al. 2014). Accordingly, differences in linear distances were noted between mandibles, which could be attributed to sex, including differences in the total length of the mandible and the diastema in females. Mandibular sexual dimorphism has been reported in the Red Deer (Azorit, Analla, and Muñoz-Cobo 2003), so it is reasonable to infer that the Patagonian huemul also exhibits this trait. However, due to the limited number of samples recorded by sex and age, it cannot be confirmed that these differences are significant. Nonetheless, we deem this initial record important for proposing hypotheses that can be explored in future studies. After evaluating the available literature, no morphometric data are available for the Taruca (*Hippocamelus antisensis*) mandible. Given the current geographic distribution of the Taruca and its phylogenetic proximity to the Patagonian huemul, it would be reasonable to expect that the linear measurements reported in this study are, on average, larger than those of the Taruca mandible, following Bergmann's biogeographic rule (Freckleton, Harvey, and Pagel 2003). This can be observed when comparing our data with other cervids inhabiting warmer environments or being generalists, such as the Roe Deer (And and Reig 1993; Pėtelis and Brazaitis 2003; Avdić et al. 2013), or with deer adapted to colder environments, such as the Reindeer (Castaños 2017). This relationship is based on the direct correlation between mandible size and animal size, as observed in previous studies of cervids (Azorit, Analla, and Muñoz-Cobo 2003). However, it is important to consider that biogeographic rules are not absolute laws but general patterns observed in nature. This means that there are cases that do not comply with these rules due to the specific characteristics of the ecosystem and the evolutionary history of each species (Oishi et al. 2010). An example of this can be observed when comparing the morphometric data recorded in the Marsh Deer (which inhabits a more northern region than the Patagonian huemul) (Pinder and Grosse 1991) with those obtained in our study, probably due to the ecological implications of the wet habitat in which this deer is distributed.

It was observed that the mandibular anatomy of the Patagonian huemul is consistent with a diet predominantly comprised of browsing, yet it also suggests the capability of grazing. This adaptability allows the animal to handle a variety of food resources, including those that are hard, fibrous, and of varying shapes. These findings further suggest broad flexibility in the choice of trophic resources, which aligns with migratory behaviours, as resource availability tends to decrease with altitude (Dezzotti et al. 2019; Sérsic et al. 2011). The primary diet of the Patagonian huemul, mainly composed of dicotyledons, further demonstrates its adaptive capacity to various habitats, including valleys, steppes and mountains (Escobar, Smith, and Flueck 2020). That said, there is an intense debate about determining the natural habitat of the Patagonian huemul, whether in forested areas or steppes (Corti and Díaz, 2023; Flueck et al. 2022). Clarifying this point is vital for establishing the foundations of its conservation. Although this discussion cannot be resolved solely with our findings, the anatomical record presented in this study suggests that the Patagonian huemul has the ability to adapt to a variety of environments based on the availability of plant resources, which would influence its mandibular morphology. After reviewing the available literature, there are no studies on the mandibular anatomy of *Odocoileus lucasi*, an extinct cervid with primarily mountainous habits (Morejohn and Dailey 2004). However, photographic records of this animal's fossil skeleton, taken and published by James St. John (with Attribution CC BY 2.0), reveal a robust mandible. Conducting a detailed comparative study of mandibular anatomy between the Patagonian Huemul and the *Odocoileus lucasi* is considered pertinent. Such a study would help determine whether the mandibular morphology of the Patagonian Huemul supports its migratory behaviour, using the mountainous habits of *Odocoileus lucasi* as a reference.

It is important to emphasise that all qualitative findings presented in this study were observed in both juvenile and adult individuals. While juvenile Huemul deer largely remain dependent on their mothers, it is during this phase that they begin to develop a degree of independence and experiment with various types of food, extensively exploring their territory (Escobar, Smith, and Flueck 2020). This may explain the qualitative similarities across the juvenile and adult mandible, as well as the quantitative differences. However, it is crucial to highlight that the mandibular shapes of the juvenile specimens studied could undergo changes during further development until reaching adulthood. Additionally, the inferred relationships between ecology and morphology in this study may be influenced by phylogeny (Zhou et al. 2019; Pérez-Barbería and Gordon 1999). Comparing the data obtained from Patagonian huemul mandibles with those of phylogenetically close cervid species decreases the likelihood that the phylogenetic effect is the sole or primary factor influencing these traits. When comparing closely related species, it can be inferred that similarities or differences in mandibular morphology are more closely related to specific adaptations to the environment and ecology of each species than to a shared evolutionary history (Zhou et al. 2019). Therefore, we regard the study's conclusions on the relationship between ecology and mandibular morphology of the Patagonian huemul as relevant, even in the presence of phylogenetic influence.

Based on the study findings and the previous discussion, further studies are crucial to deepen the understanding of mandibular morphology in the Patagonian huemul and its ecological implications. It is recommended to conduct studies on dental morphology to reaffirm previous findings and provide a more comprehensive understanding of dental adaptability to the available food types. Exploring the ecological and biomedical implications of these dental characteristics may reveal important aspects of the species' ecology and health. Furthermore, conducting multivariate and biomechanical studies is essential to assess the relationship between mastication and the external and internal morphology of the mandible. These approaches will allow us to understand how differences in external form translate into variations in the internal structure of the mandible and other relevant anatomical aspects. It is also important to understand the relationship between mandibular skeletal morphological variability, sexual dimorphism and the effect of age in the Patagonian huemul. Specific studies on this issue will provide valuable information about the evolution, reproductive behaviour and population dynamics of this threatened species. Finally, demonstrating Bergmann's rule between the Patagonian huemul and the Taruca regarding body size and morphological characteristics will provide insights into their adaptation to different environments and the factors influencing their geographic distribution.

4.2 | Clinical-Surgical Aspects

In anaesthetic procedures, such as regional nerve block, precise anatomical knowledge and the ability to identify bony prominences through palpation are crucial, as they are often a practical means of perioperative and postoperative analgesia in painful procedures, allowing for less aggressive treatment during recovery (Beckman and Legendre 2002). In the case of the mandible, the exact location of the mental and mandibular foramina in wild animals are osteometric data particularly evaluated in dental studies that involve regional nerve block procedures (Kierdorf et al. 2016; Paulo et al. 2020; Magalhães, Ferreira Júnior et al. 2019; Magalhães, Romão et al. 2019; Choudhary et al. 2018, 2017, 2016, 2015; Glatt, Francl, and Scheels 2008). In the Patagonian huemul, we have identified clinically relevant structures that must be considered in this context. Firstly, the mandibular foramen is of clinical-surgical importance as a reference for identifying the mandibular nerve (Hall, Clarke, and Trim 2000; Clarke and Trim 2013; Audisio et al. 2011). It is important to note that the exact position of the mandibular foramen can vary among species (Wang et al. 2021). Therefore, to precisely locate the mandibular foramen in the Patagonian huemul, it is suggested to use the mandibular angle as a reference and trace a 3 cm axis in the rostrodorsal direction, considering an approximate height of 2.8 cm from the ventral margin of the mandible and a distance of 1.5 cm from the caudal margin of the mandible. Secondly, the mental foramen is of clinical importance. For its correct palpation, two reference points are suggested: either the alveolus of the first incisor, tracing a 2.2 cm axis rostrally, or the rostral margin of the first premolar, tracing a 3.6 cm axis rostrally. Precise knowledge and identification of these anatomical structures in the Patagonian huemul are fundamental for safely and effectively performing anaesthetic procedures. These guidelines provide a practical guide for conservation medicine professionals performing regional nerve blocks in this species. It is important to highlight that we have observed the presence of an accessory mental foramen in the mandible of the Patagonian huemul, which constitutes a relevant finding of clinical importance for orofacial interventions due to the neurovascular bundles present in this region (Iwanaga et al. 2016). If an intervention involving the region around the mental foramen, especially in its rostral and dorsal part, is necessary, explorations using cone beam computed tomography (CBCT) are suggested as part of the preoperative diagnosis due to the advantages of geometrically more precise image resolution it provides (Barría-Pérez et al. 2021; Muinelo-Lorenzo et al. 2016), as currently demonstrated in its efficacy with animals such as horses (Stewart et al. 2021). A precise understanding of these anatomical structures and the use of advanced imaging techniques like CBCT will enable safer and more effective planning and execution of orofacial interventions in the Patagonian huemul.

Another relevant aspect is the prevalence of osteopathologies in the mandibular symphysis of the Patagonian huemul, with up to 63% of cases reported in some populations (Flueck and Smith-Flueck 2008). These pathologies include osteomyelitis, periodontitis, osteoarthritis and incisor exfoliation (Flueck 2015, 2018, 2020; Flueck and Smith-Flueck 2017, 2018, 2020; Smith-Flueck, Flueck, and Escobar 2018; Escobar, Smith, and Flueck 2020). Given that road accidents have been recorded (Escobar, Smith, and Flueck 2020), the presence of a weakened mandibular symphysis associated with osteomyelitis is concerning, as it could facilitate traumatic separation as observed in alpacas and canines (Giraldo 2009; Mellado 2012; Pezo et al. 2017). The higher proportion of spongy tissue observed in the mandibular symphysis has important biomechanical implications, as it is more porous and elastic, allowing it to better absorb forces than cortical tissue, which is denser and more resistant. It provides greater deformation capacity and cushioning forces during mastication, adapting to the diverse biomechanical demands associated with its type of food. Additionally, it can influence bone healing and regeneration capacity in case of fractures or injuries, as spongy tissue is more vascularized, facilitating blood supply and nutrient delivery for tissue repair (Lee, Jain, and Alimperti 2021). On the other hand, the relationship between compact bone and spongy bone is one of the factors to consider for locating fracture lines in bone structures and in dental implant placement techniques (Paz Roca et al. 2001). The mandibular symphysis exhibited the largest area of spongy bone, while the mandibular body showed the largest area of compact bone. These findings suggest that the mandibular body is the most suitable and resistant region for anchoring dental implants for overdentures.

From the perspective of applied anatomy, the 3D visualisation of the mandibular canal is a valuable contribution to advancing the knowledge of anaesthetic protocols in endangered wildlife species. Detailed knowledge of the location and shape of the mandibular canal is essential for preventing injuries to neurovascular components during dental procedures, such as extractions, endodontic treatments, orthopaedic surgery and oncological procedures (Cotrim et al. 2015; Martinez et al. 2010). In the case of the Patagonian huemul, the evidence suggests that the mandibular canal cannot be considered a medullary canal, and it is important to note that treating body fractures with an intramedullary nail through this canal could damage the associated neurovascular bundles.

4.3 | Study Limitations

It is important to highlight that research on the Patagonian huemul faces significant challenges due to the laws of Chile and Argentina, which restrict access to biological and cadaveric material of animals in danger of extinction for analysis and study, limiting the advancement of anatomical knowledge. It is recognised that the sample used in our study is limited, which prevents establishing significant differences between variables and affects the discussion. However, this does not prevent us from formulating hypotheses based on the observed trends. The descriptive findings reported are useful as a reference for the quantitative variability in the mandibles of these deer. Therefore, they constitute a valuable starting point, and we suggest that future studies be conducted with larger and more varied samples to validate and extend our results. Furthermore, the absence of teeth in some of the mandibles studied presented a challenge for establishing precise anatomical associations between the dorsal canaliculi and the ascending alveolar branches and vascular structures. However, no variation was observed in the count of alveolar canals, which contrasts with findings in domestic animals (Nickel et al. 1984), where the number and shape of the alveolar branches vary both intraspecifically and bilaterally. Despite the clinical relevance of the mandibular canal in deer, it has been underestimated in previous anatomical descriptions of mandibles in species such as the Taruca (Coacalla, Curie, and Flores 2021), the Pudu (Saldivia and Villegas 2019), the Reindeer (Castaños 2017) and the Roe deer (Onuk, Kabak, and Atalar 2013). The lack of anatomical and morphometric descriptions of the mandibular canal in deer has limited our discussion and understanding of the potential clinical implications in these animals, as well as the branching of the trigeminal nerve and the evolutionary adaptations aimed at enhancing cranial sensation in this taxonomic group with different ecologies and lifestyles, as described in other herbivores of research interest (Lessner et al. 2023; Evcim 2021; Kierdorf et al. 2016). Despite these limitations, the results obtained in this study provide a solid foundation for understanding the anatomy and specific characteristics of the juvenile and adult Patagonian huemul mandible. These findings contribute to the overall knowledge of the biology and management of the species and can serve as a starting point for more extensive and detailed future research.

In conclusion, the mandibles of both juvenile and adult Patagonian huemul exhibit distinctive features typical of browsing herbivores. These features include a robust coronoid process, a wide and deep pterygoid fossa, a mandibular condyle that is significantly lower than that of other deer, a mandibular canal with a curved caudal portion, and a straight rostral portion that varies in shape in transverse images depending on the segment, and an obtuse angle between the body and ramus. These attributes reflect specific adaptations to the species' dietary and behavioural needs. Such results provide a solid foundation for future comparative studies on the mandibular anatomy and functionality of the Patagonian huemul and other herbivorous deer species. Understanding these anatomical adaptations deepens our knowledge of their biology and has significant implications for improving deer population management practices. The outcomes of this study furnish essential information for managing specimens and highlight the mandible's specialised features in relation to the Patagonian huemul's ecological niche.

Author Contributions

Study conception and anatomical description were performed by Samuel Núñez-Cook and Paulo Salinas. Data collection and the first draft of the manuscript was written by Samuel Núñez-Cook. Samuel Núñez-Cook, Fernando Vidal and Paulo Salinas contributed to data analysis and discussion and approved the final manuscript. Paulo Salinas was responsible for supervision of the study.

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Ethics Statement

Approval by research ethics committees was not required to achieve the objectives. Skeleton care and handling were carried out in accordance with Chilean Law 20.380.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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