The Middle Ear of Ruminants: An Anatomical and Bio-Metrical Comparison between the *Lori* Sheep and the *Native* Cow in Shahrekord District

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Abstract: The ossicles consist of three small bones known as the smallest bones in the body that play an important role in transmitting sound to the inner ear. In order to study the details of middle ear ossicles in Lori sheep and native cattle, bio-anatomical parameters of malleus, incus and stapes ossicles were measured. The age factor is not considered and all animals were adult. Investigations showed that there was no significant difference in the levels of the middle ear bone between the two sexes. In this study, the ossicles were carefully dissected by the appropriate devices and after separation, the ossicles were measured using caliper and digital scales. Bio-anatomical changes were evaluated under stereomicroscope magnification. The results showed that all three bones structure is similar to other animals. Biometric aspects indicated that most of the changes belonged to malleus and incus bone. After examining the two species under study, the least significant change was observed between the two animals in the stapes. In conclusion, this study was able to compare middle ear ossicles between sheep and native cattle in Shahrekord district. Anyway, it is conceivable that the biometric sizes of the middle ear bones may be affected by animal species.

Keywords: Sheep, Cow, Ossicles, Middle ear.

INTRODUCTION

The performance of hearing is a specific feeling are observed among the upper classes of the animal kingdom and is one of the most important sensory inputs to maintenance of life. It seems very interesting, such small structure like the ear performs a vital act of hearing. It is even more surprising that there are three small ossicles, called: a malleus, incus and stapes inside the middle ear [1]. In discussion of middle ear evolution, primitive tetrapods such as ichthyostega, have a tympanic membrane at the quadrate position. A high degree of extension was observed for the tensor tympani and stapedius muscles, both of them attached to distinct muscular processes [2]. Wible and Gaudin (2004) reported a lack of processes for muscular fixation in middle ear in Euphractussexcinctus, but they did not indicate the degree of muscular development [3]. Next experiences were developed from the reptilian condition and then extend to reptiles and birds, and the mammalian middle ear [4, 5].

The mammalian middle ear is the most basic morphological features that signalize this class of vertebrates. Middle ear skeletal pattern differs obviously among vertebrates than those of other amniotes and has attracted the attendance of comparative zoologists for years (Kuratani, 1999). The middle ear has three ossicles, the malleus, incus and stapes [6, 7]. Fleischer (2013) demonstrated the more terrestrial mammals can be arranged into two basic middle ear types established upon their ossicular morphologies; with a range of intermediate or transitional morphology between both types. These groups are detected by having a malleus with a long process (processusgracilis) fused to the ectotympanic via a goniale, a wide transversal lamina between the manubrium and the articular surface (for the incus), and a prominent bony mass near the base of the manubrium called the orbicular apophysis [8].

Recent genetic analyses in the mouse have provided information about genes central to middle ear formation [9-11]. However, Goosecoid mutant mice showed defects of the malleus manubrium, processus brevis and the ectotympanic and tympanic membrane in the middle ear region [12]. Low-frequency airborne hearing can be improved by relatively large middle ear ossicles with loose ligament attachments, large tympanic membranes and an increased bullar volume [13]. Therefore, a goal from investigations of auditory genetics and physiology is to express the relation between the middle ear's structure and the function of auditory signal process [14].

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E-ISSN: 2310-0796/21

The purpose of the present study was; i) to perform a morphological description of the middle ear region with special emphasis in the auditory ossicles; ii) to carry out a bio-morphometric comparative study of the middle ear ossicles in the two species *Lori*sheepand *Native* cow.

MATERIALS AND METHODS

Animals

The study was a biometrical, basically sciences based pragmatic study, performed in the Department of Anatomy in Shahrekord University to monitor the morphological and biometrical differences between ossicles of middle ear for a period of approximately two months, from collecting sample until ossicles separation. Number 5of sheep and cow (in Shahrekord district) heads from both sexes with entire temporal bones and possessing entire ossicular series in the pattern of with no discontinuity and without erosion/deterioration were selected. Bones that seemed to be worn out in consequence of ear disorders and or complicated diseases were excluded from this investigation. The methods used to obtain different bones of the ear was similar to the early stages of "the method of the mastoidectomy" which was carried out by protocol of the researchers [15]. To expose the bones of malleus, incus and stapes, the bulla required to be made accessible by using a very small drill. After that, the middle ear ossicles were separated from the tympanic cavity and examined using an anatomical stereomicroscope (Model Olympus Co., Tokyo, Japan). Following parameters were studied;i) description of the anatomical structure of the middle ear ossicles and ii) the measure the biometric parameters of the middle ear ossicles including weight, thickness and diameter.An X-ray (Ralcos.r.l Comp) set was used to take a radiograph of the middle ear bones, including malleus, incus and stapeson a high-resolution mammography films.

Statistical Analysis

Statistical analyses in data of middle ear ossicles were carried out by using the SPSS statistical software package version 23.0.0 for Windows. Data are expressed as mean ±standard deviation (SD) and statistical variations were tested by Student-T test. The biometric values of variations, including the weight, length, width and thickness in relation to middle ear ossicles were corrected between two the species and the method used was considered with the criteria of a probability of *p<0.05* as statistically significant.

RESULTS

Gross anatomical findings in total available dissected specimens exhibited that the tympanic bulla included three malleus, incus and stapes ossicles (Figures **1** to **3**). Figure **3** demonstrate radiographs of middle ear ossicles in lateral and medial view.

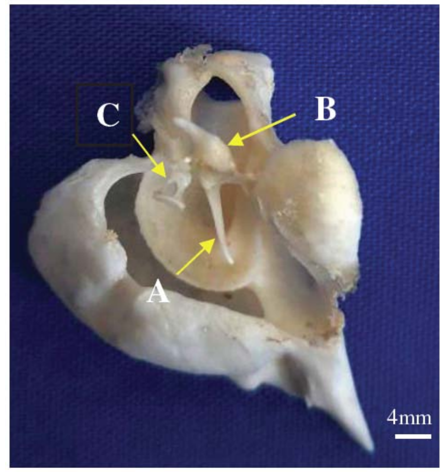

Figure 1: Ossicular chain link in sheep mature (dorsal view); **A**: Handle of malleus; **B**: Incus; **C**: Stapes.

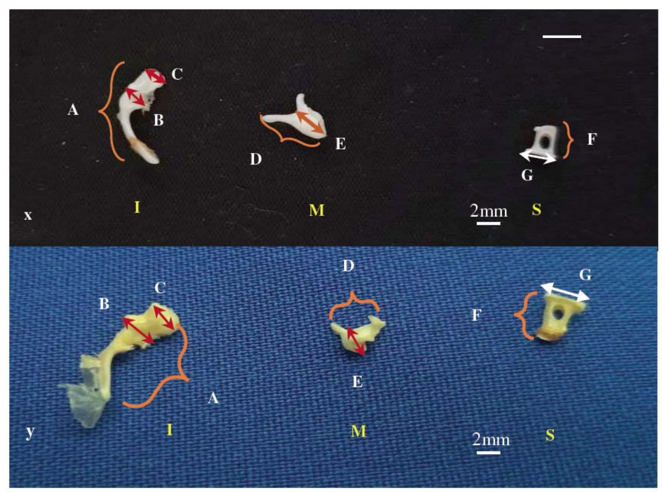

Figure 2: Part **x**, Ossicles in mature cow; I, Incus; M, Malleus; S, Stapes; A, length of malleus; B, width of malleus; C, thickness Of head in malleus; D, length of incus; E, width of incus; F, length of stapes; G, width of stapes; Part **y**,Ossicles in mature sheep; I, Incus; M, Malleus; S, Stapes; A, length of malleus; B, width of malleus; C, thickness of head in malleus; D, length of incus; E, width of incus; F, length of stapes; G, width of stapes.

The malleus was a hammer-molded small bone which joins with next bone (incus) and is attached to the inner surface of the tympanic membrane. The incus ossicle, a bone in the middle ear, was the anvil- molded small bone which connected to the third middle ear bone (Figures **1** and **2**). Stapes, the last bone (Figures

1 and **2**), was the stirrup-molded shaped small bone which was placed on other bones, transports these to the oval opening (an opening that leads from the middle ear to the inner ear cavity [16, 17].

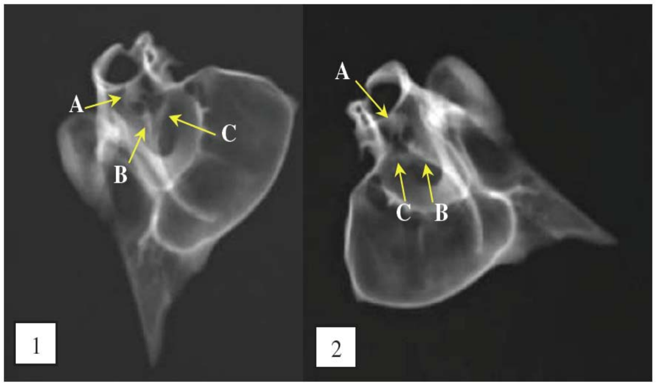

Figure 3: Radiographs of the middle ear ossicles in mature sheep; 1) lateral view; 2) medial view. A, malleus; B, incus; c, stapes.

Data obtained from bio-metric analysis of three malleus bones, incus and stapes in both species (sheep and cow) are shown in Table **1**. The results revealed an obvious increase in length, width, thickness of malleus ossicle in cow than sheep (Table **1**). All of measures except the weight of the Malleus ossicle in the cow showed a significant difference compared with sheep (*p<0.05*). However, only the weight of the Malleus ossicle in the cow no showed a significant difference compared with sheep (*p>0.05*). There was a significant difference in the biometric dimensions of the length, thickness and width (head, middle and tail) of the malleus ossicle between both species (*p<0.05*).

A similar pattern was followed among biometric sizes of the incus ossicle in middle ear (Table **1**), as the biometric dimensions of the length and width of the incus ossicle in the cow were significantly higher than

Table 1: Biometric Parameter of Malleus, Incus and Stapes between Sheep and Cow (mm) (Mean ± *SE*)

row	species	Parameter	Mean± SE	P value
1	sheep	m.sh.l	8.97±.363^{a}	P<0.05
2	cow	m.c.l	10.22±.248^{a}	
1	sheep	m.sh.hwi	2.01±.028^{b}	P<.0.05
2	cow	m.c.hwi	2.20±.051^{b}	
1	sheep	m.sh.twi	.78±.020^{c}	P<0.05
2	cow	m.c.twi	.85±.025^{c}	
1	sheep	m.sh.mwi	3.49±.024^{d}	P<0.05
2	cow	m.c.mwi	3.95±.015^{d}	
1	sheep	m.sh.we	.029±.001	p>0.05
2	cow	m.c.we	.040±.004	
1	sheep	inc.sh.l	4.90±.020e	P<0.05
2	cow	inc.c.l	5.95±.103e	
1	sheep	inc.sh.wi	2.42±.017f	P<0.05
2	cow	inc.c.wi	2.83±.049f	
1	sheep	inc.sh.b2b	3.32±.063	P<0.05
2	cow	inc.c.b2b	4.79±.031	
1	sheep	inc.sh.we	.030±.029	p>0.05
2	cow	inc.c.we	.060±.001	
1	sheep	st.sh.l	3.44±.065	p>0.05
2	cow	st.c.l	3.62±.005	
1	sheep	st.sh.wi	2.49±.043^{g}	P<0.05
2	cow	st.c.wi	2.69±.014^{g}	
1	sheep	st.sh.t	.580±.023	p>0.05
2	cow	st.c.t	.670±.020	
1	sheep	st.sh.we	.024±.011^{h}	p>0.05
2	cow	st.c.we	.025±.010^{h}	

Sh: sheep; C: cow; M: malleus; We: weight;L: length; Wi: width; Hwi: head width; Mwi: middle width; Twi: tail width; Inc: incus; b2b: between two branches; St: stapes; T: thickness. Significant differences between each two values of a single parameter compared between two species are shown with same letters; otherwise there is no letter denotation.

the sheep ($p<0.05$). The differences between two species in weight of incus ossicle do not follow a similar pattern like other biometric values; as there were not significant difference in the parameters mentioned between two animals, in spite of increasing weight of incus ossicle in cow compared sheep ($p>0.05$).

The values of the weight, the diameter and length observed concerning stapes ossicle in each two animals are presented in Table **1**. It should be noted that there was significant difference in the width value of the stapes ossicle when compared between two species ($p<0.05$). The thickness, length and weight biometrical amounts increased in cow compared with sheep during the anatomical evaluation of this bone; but except the weight there was no significant difference between cow and sheep in biometrical amounts of stapes ossicle ($p>0.05$).

DISCUSSION

Although some researchers evaluated the bony structure of the middle ear from an anatomical viewpoint [1, 18-20], but the study on the comparison of middle ear ossicles (malleus, incus and stapes) between the two species of cattle and sheep from each two sexes has not been elucidated in detail. Therefore, the present article has evaluated these bone structures as far as possible. Findings in the biometric values of the middle ear ossicles displayed that there were significant differences between the two species, but no observed clear difference about the gender impact on biometric amounts in both animals.

The researchers have shown that measuring the middle ear ossicles plays an important role in the output impedance of the ear [14]; On the other hand, it can have other effective effects in artificial hearing aid that replace some of the missing parts of the body. In this way, sodhi *et al*., (2017) in a demographic study at India on the morphometric dimensions of human ear ossicles of males showed that exact measurements of the ossicles could being very helpful in designing the prosthesis in ossicular chain pathology [21].

As previously identified, reports revealed that there was no significant difference between middle ear bones in male and female rabbits. This may lead to the result that diversity in the characteristics of middle ear ossicles, particularly bio-anatomical parameters, does not matter so much in term of the sex [22], as was agree with the results of this study. However, although the findings suggest no gender differences in the structure of the middle ear ossicles [22], but in current study effect of gender factor was taken into account. In a study by Meiring and Oschman (1991) on the morphology of the malleus ossicle of 75 adult human cadaver, in a comparative manner, they was found that there was no significant difference between two sex bones [23].

The anatomical gross structure of the malleus, incus and stapes ossicles in this study was similar to recent reports [14, 24, 25]. The findings in few reports show tangible differences, as kurtul *et al*., (2003) in rabbit showed that some middle ear ossicles, in anatomical appearance, have a clear difference over the rest of the animals. They in own study found that there were variation greatly especially on the processes and handle in this ossicles [22].

Regardless of the significant differences between some biometric quantitative values in the middle ear ossicle in this study, the variations indicated a sensible increase in measured values about ossicles of the cattle relative to the sheep. However, with a brief overview at "*p-value*" of the measured number of ossicles, we were found that malleus ossicle showed the least alteration between the two species of cattle and sheep. While in the other two ossicles, the most alteration was observed regarding of biometrical measurements in incus and stapes ossicles. Researches has shown that between the ossicles, incus is the most fixed ossicle and stapes ossicles have the most variable as far as their morphological changes are of particular importance [26-29]. Previous studies have shown that congenital ossicles anomalies are accompanied by facial nerve abnormalities; furthermore, Padmini and Rao (2013) in evaluating morphological changes of human fetal ear ossicles showed that impoverished human fetuses can be used in form homo-grafts to substitute corroded mature ear ossicles.

The degree and extent of bony chambers could be reliably predicted by the size of its internal structures [30]. It seems that the distinct difference that between the bones of the two species under study can be clearly seen, to be closely related to the dimensions of tympanic bone. According to this point, as is clear, tympanic bone shows a larger dimension in cow than sheep [16, 31]. Therefore, it's easier to understand, the distinct difference between the measured values in the malleus, incus and stapes ossicles between two animal species; however, some bony values did not show significant differences.

ACKNOWLEDGEMENTS

This work was financially supported by the University of Shahrekord, Iran. In the end, the authors of the paper, from experts in the field of anatomy of the department of basic sciences thanks and appreciates.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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Received on 06-12-2020 Accepted on 02-01-2021 Published on 03-03-2021

https://doi.org/10.12970/2310-0796.2021.09.02