Meat Science 113 (2016) 35–40

Contents lists available at ScienceDirect

Meat Science

journal homepage: www.elsevier.com/locate/meatsci

Magnetic resonance imaging and computer tomography of brain lesions in water buffaloes and cattle stunned with handguns or captive bolts

Barbara K. Schwenk [a,1], Isabel Lechner [b,1], Steffen G. Ross [c], Dominic Gascho [c], Beat P. Kneubuehl [d], Matthieu Glardon [d], Michael H. Stoffel [a,*]

[a] Division of Veterinary Anatomy, University of Berne, Berne, Switzerland
[b] Veterinary Public Health Institute, University of Berne, Berne, Switzerland
[c] Department of Forensic Medicine and Radiology, Institute of Forensic Medicine, University of Zurich, Switzerland
[d] Institute of Forensic Physics/Ballistics, University of Berne, Berne, Switzerland

ARTICLE INFO

Article history:
Received 6 October 2014
Received in revised form 3 November 2015
Accepted 10 November 2015
Available online 10 November 2015

Keywords:
Bubalus bubalis
Stunning
Skull
Anatomy
Diagnostic imaging

ABSTRACT

Owing to the demand for genuine mozzarella, some 330 water buffaloes are being slaughtered every year in Switzerland albeit a stunning procedure meeting animal welfare and occupational safety requirements remains to be established. To provide a basis for improvements, we sized anatomical specifics in water buffaloes and cattle and we assessed brain lesions after stunning with captive bolts or handguns by diagnostic imaging. In water buffaloes and cattle, the median distance from the frontal skin surface to the inner bone table was 74.0 mm (56.0–100.0 mm) *vs* 36.6 mm (29.3–44.3 mm) and from skin to the thalamus 144.8 mm (117.1–172.0 mm) *vs* 102.0 (101.0–121.0 mm), respectively. Consequently, customary captive bolt stunners may be inadequate. Free bullets are potentially suitable for stunning buffaloes but involve occupational safety hazards. The results of the present study shall be used to develop a device allowing effective and safe stunning of water buffaloes.

1. Introduction

Over the last decade, water buffalo husbandry has become increasingly popular in Switzerland. According to the Swiss Animal Tracing Database, water buffalo livestock amounted to approximately 1750 animals in 2013 (Federal Food Safety and Veterinary Office, Dr. Alexandra Briner, personal communication). The interest in rearing this species increased as a consequence of an ongoing price decline for cow's milk (Zemp, 2012) paralleled by an increasing demand for genuine mozzarella. This has made the production of buffalo milk an economically interesting niche product. Since 1996, when the first animals were imported from Romania, the number of buffaloes has increased continuously. In response to stock management requirement to eliminate culled animals and market demand for water buffalo meat, approximately 330 water buffaloes are slaughtered per year in Switzerland.

To meet the Swiss legal requirements to ensure animal welfare at the time of slaughter, adequate stunning must produce deep unconsciousness (Tierschutzgesetz, 2014). As consciousness arises from thalamocortical projections, stunning methods aim to disrupt the information transfer to and within the cerebral cortex. In domestic cattle, this is usually achieved by producing mechanical damage to the forebrain and possibly the brain stem using percussive devices (Fries, Schrohe, Lotz, & Arndt, 2012). Provided that energy transfer is adequate, inertia interactions from a blunt blow to the skull will produce concussion by means of shock waves, coup and contrecoup as well as other impacts on the brain. Penetrating captive bolt devices potentiate this action by producing an additional deleterious shock wave within, and direct damage to the brain tissue (Kneubuehl, 2011; Lambooy, 1981; Perdekamp et al., 2010). As the infringement on the cranial cavity is an essential requirement when using captive bolts, the effectiveness of a penetrating stunning device is highly dependent on the given anatomical conditions and the device being used. Although *Bubalus bubalis* belongs to the same family of *Bovidae* as domestic cattle (*Bos primigenius taurus*), the anatomical characteristics of the head differ considerably between the two species. To date, several studies dealing with the anatomy of the head of water buffaloes have been published (Kamel & Moustafa, 1966; Lakshminarasimhan, 1974; Meyer & Fiedler, 2005; Moustafa & Kamel, 1971; Saigal & Khatra, 1977; Singh, Soni, & Manchanda, 1972). They revealed the skull bones to be substantially thicker and the frontal and paranasal sinuses to be noticeably wider in buffaloes compared to bovines (Saigal & Khatra, 1977). Moreover, these anatomical features vary markedly with the sex and age of the animals (Dyce, Sack, & Wensing, 2010). Furthermore, hide thickness may play a role as well.

* Corresponding author at: Division of Veterinary Anatomy, University of Berne, Länggassstrasse 120, POB 8466, CH-3001 Bern, Switzerland.
E-mail addresses: isabel.lechner@vetsuisse.unibe.ch (I. Lechner), s.ross@spitalmaennedorf.ch (S.G. Ross), Dominic.Gascho@irm.uzh.ch (D. Gascho), bpkneubuehl@bpk-thun.ch (B.P. Kneubuehl), matthieu.glardon@irm.unibe.ch (M. Glardon), michael.stoffel@vetsuisse.unibe.ch (M.H. Stoffel).
[1] Equally contributing first authors.

http://dx.doi.org/10.1016/j.meatsci.2015.11.010
0309-1740/

To ensure adequate stunning in compliance with animal welfare requirements (Atkinson, Velarde, & Algers, 2013; Gouveia, Ferreira, Roque da Costa, Vaz-Pires, & Martins da Costa, 2009), these anatomical characteristics need to be taken into account. This is well illustrated by the fact that conventional captive bolt devices usually fail to produce deep unconsciousness in adult water buffaloes when used as in cattle (Camisasca & Calzolari, 1995).

The goal of the present study was to revisit the anatomical specifics of the head of water buffaloes (*B. bubalis*) compared to domestic cattle (*B. primigenius taurus*). The heads from buffaloes and various cattle breeds were investigated after stunning, taking into account both sex and age. The anatomical specifics and brain lesions were assessed by gross anatomical dissection and diagnostic imaging using magnetic resonance imaging (MRI) and computer tomography (CT). The use of these imaging techniques allowed gaining accurate information on the topography of the skull and brain with respect to the landmarks that are relevant to stunning. In addition, brain lesions were assessed to judge the potential of various stunning devices in producing deep unconsciousness.

2. Materials and methods

2.1. Head collection and stunning techniques

The heads of the water buffaloes were collected from eight different abattoirs that were selected based on data from the Federal Food Safety and Veterinary Office FSVO. One single person was in charge of stunning for any given slaughterhouse. Three butchers used a different handgun each, one butcher used a bullet casing gun and four butchers relied on three different types of captive bolt stunners (Table 1). The heads were collected from animals that had been stunned in the context of regular slaughterings. Immediate collapse was considered as evidence of effective stunning, that was the case for all the animals included in this study. Heads were assigned to three different groups according to sex and age, *i.e.* males from 15 to 30 months of age (m1, $n = 13$) and older than 30 months (m2, $n = 9$) as well as females older than 30 months (f2, $n = 13$). As young female water buffaloes hardly ever get slaughtered and as this category poses the least challenge with respect to stunning, females under 30 months of age were not included in the present study. On the other hand, maximizing the number of male animals older than 30 months was deliberately pursued, as any method providing adequate stunning in this category may be expected to be effective in all the other groups as well.

Two buffalo brains were removed immediately after slaughter and fixed in 10% formalin to macroscopically assess the severity of the lesions. Another two heads were sectioned in a mid-sagittal plane after diagnostic imaging to monitor the dimension of the sinuses, the extent of the cavum cranii and their topographical relationships.

Thirty-five heads were used for diagnostic imaging. Four MRI scans were discarded for technical reasons, thus yielding 35 CT datasets and 31 MRI datasets (Table 1).

The stunning of water buffaloes was performed with one of the following methods: conventional penetrating captive bolt devices (Cash Magnum 9000S, EFA Schmid & Wezel GmbH & Co. KG Maulbronn, Germany or Schermer KL, Karl Schermer GmbH & Co KG, Ettlingen, Germany; penetrating depth: 121 and 125 mm, respectively) in either the frontal or occipital position; pneumatic captive bolt gun (EFA VB 215, EFA Schmid & Wezel GmbH & Co. KG Maulbronn, Germany; penetrating depth: 135 mm) in the occipital position; bullet-casing gun in the frontal position (“humane killer”, no manufacturer's data available) (Anonymous, 2008); with a revolver or pistol in the frontal position (44 S&W Magnum, Smith and Wesson, Springfield, USA; Ruger GP 100 Double Action Revolver, Sturm, Ruger & Co., Inc., Mayodan, USA; Swiss Army Pistol SIG P220, SIG Sauer GmbH & Co. KG, Eckernförde, Germany). Specifications of the stunning devices and ammunition as well as information regarding their application are provided in Table 1. The heads of the water buffaloes were documented photographically with respect to the bullet holes and all but two heads were dehorned prior to further examination.

The cattle heads were collected from three slaughterhouses (Table 1) in which the stunning was performed with captive bolt devices in the frontal position (Kuchen, NATURaktiv AG, Winterthur, Switzerland or Cash Magnum 9000S, EFA Schmid & Wezel GmbH & Co. KG Maulbronn, Germany; penetrating depth: 90 and 121 mm, respectively; Table 1) or with an air-operated captive bolt gun (EFA VB 215, EFA Schmid & Wezel GmbH & Co. KG Maulbronn, Germany; penetrating depth: 135 mm) according to standard procedures (Fries et al., 2012; Des Verordnung, 2014). Diagnostic imaging was also used to assess the 12 heads from domestic cattle as controls, yielding 12 CT datasets and eight MRI datasets as 4 MRI datasets had to be discarded for technical reasons. MRI data sets were obtained from two females over 30 months (f2), four males between 15 and 30 months (m1) and two males older than 30 months (m2). Cattle heads were not photographed because stunning was performed according to established standard procedures.

2.2. Head/skull examination

Based on topographical relationships as determined on sagittally sectioned heads of water buffaloes, the frontal point of entry for captive bolt guns was chosen at the intersection of two imaginary lines connecting the lower edge to the upper edge of the contralateral horn. The occipital point of entry was located at the level of the lower edge of the horns, *i.e.*, above the insertion of the nuchal ligament. For cattle, the point of entry for captive bolt stunning was selected according to the standard procedure, *i.e.* at the intersection of the two imaginary lines connecting the nasal ocular angle to the lower edge of the contralateral horn. Handguns used by three butchers were fired at a distance of approximately 5 cm from the head. All the corresponding points of entry of the free projectiles were between a line connecting the medial ocular angles and a line at the midlevel of the horn bases. When scanning was carried out within 2 days of slaughter, the heads from both water buffaloes and cattle were stored at 4 °C. Otherwise, the heads were collected and stored at −20 °C in a deep freezer for 2 to 7 weeks and thawed for 72 h before scanning. For the scanning procedure, all the heads were wrapped in heavy-duty plastic bags. The heads were examined with a 3-Tesla MR scanner (Achieva 3.0 TX, Philips Medical System, Best, The Netherlands) equipped with a Q-Body coil. A 16-channel SENSE-XL-torso coil was used unless heads were too big. The sequences included an axial, coronal and sagittal T2-weighted (T2 W) sequence with fat saturation. Slice thickness was 3.0 mm for all the sequences. Computer tomograms were obtained with a dual-source CT scanner (SOMATOM Definition Flash, Siemens, Forchheim, Germany) with 2×128 slices. Data reconstruction was performed with 0.6 mm slice thickness in a soft (B30) and a hard (B70) reconstruction algorithm. Multiplanar and 3-dimensional reconstructions were performed at a multimodality workstation (LEONARDO, SynGo, Siemens Medical Solutions, Forchheim, Germany). Data were analyzed with the Osirix® software (Pixmeo, Bernex, Switzerland).

The following landmarks were identified prior to proceeding with data assessment: optic canal including its lateral edge, nasal, frontal, occipital, basisphenoid and presphenoid bones, hypophyseal fossa, cribriform plate and crista galli of the ethmoid bone, hard palate and nasal septum.

Several anatomical measurements were taken. Hide thickness and sinus width were assessed in CT datasets whereas MRI datasets were used to determine brain damage and the fate of free projectiles. Hide thickness (HT) without coat was measured at a right angle to the frontal bone at the level of the dorsal end of the crista galli (HT1) and at the level of the rostral end of the hypophyseal fossa (HT2). Similarly, the distance between inner and outer tables of compact bone delimiting the frontal sinus was measured twice at the same locations as for the