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# Effects of magnetic field and solar radiation on body axis alignment in water buffalo

Master's thesis

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# Declaration

I hereby declare that this master's thesis entitled “Effects of magnetic field and solar radiation on body axis alignment in water buffalo” is my own work and all the sources have been quoted and acknowledged by means of complete references. I agree with the work to be stored and available for educational purposes in the library of CULS.

In Prague: 27 April 2017

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Radim Sedláček

*Photo by author*

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# Abstract

**Effects of magnetic field and solar radiation on body axis alignment in water buffalo**

Water buffalo (*Bubalus bubalis)* has a great importance for communities living in remote parts of Central Sulawesi. Water buffalo are highly susceptible to thermal stress and hence should be allowed to perform wallowing behaviour which was not always a practice followed by the farmers in Central Sulawesi, the locality of our research. Therefore, our aim was to analyse the effects influencing behaviour of buffalo and local Donggala cattle (*Bos taurus*) with emphasis on solar radiation and the geomagnetic field of the Earth. Data were collected using scan sampling method for 39 days. Changes in behaviour of each buffalo (n = 6) and cattle (n = 5) were recorded every 10 minutes, 8 hours per day. Parallel, perpendicular or oblique orientation of long body axis relative to incident solar radiation was recorded as well as the direction each individual was facing according to compass orientation that was classified into eight 45° arcs relative to magnetic north. Buffalo showed significant preference for parallel orientation ($P < 0.0001$) whilst cattle oriented significantly more often perpendicular relative to the sun ($P < 0.0001$). Buffalo significantly preferred parallel orientation in morning ($P < 0.0001$) as well as in the afternoon ($P < 0.05$) compared to midday period when the sun was directly over their head and thus prevented them from employment of solar orientation. Parallel orientation was more likely to be employed by both species, as the dry bulb temperatures increased while proportion of perpendicular orientation in cattle decreased with increasing ambient temperature. Cattle further exhibited significant preference for perpendicular orientation in morning ($P < 0.0001$), presumably to gain energy from the solar radiation after cold night. Both species significantly preferred magnetic alignment towards the south (cattle: $P < 0.05$; buffalo: $P < 0.0001$) and the east (cattle: $P < 0.01$; buffalo: $P < 0.001$). Cattle also showed a high frequency of orientation due north and buffalo further shown significant preference for west alignment ($P < 0.001$). Further investigation is required to test the influence of daily activities on solar orientation and magnetic alignment in ruminants.


**Key words**: ungulates, *Bubalus*, cattle, thermoregulatory behaviour, heat adaptations

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**List of the abbreviations used in the thesis**

| | |
|---|---|
| ANOVA | analysis of variance |
| CULS | Czech University of Life Sciences |
| E | east |
| FAO | Food and Agriculture Organization |
| GLM | generalized linear model |
| HSD | Tukey's honest significance tests |
| ITIS | Integrated Taxonomic Information System |
| IUCN | International Union for Conservation of Nature |
| N | north |
| NE | north-east |
| NS | north-south |
| NW | north-west |
| PARB | orientation parallel back to the sun |
| PARF | orientation parallel facing to the sun |
| PERP | orientation perpendicular to the sun |
| S | south |
| SE | south-east |
| SW | south-west |
| W | west |

# 1. Introduction

The Swamp buffalo together with rice harvesting became an icon of human survival for communities living in remote parts of Central Sulawesi. Buffalo are considered animals of great economic importance since their meat, horns, skin and nutritious milk can all be exploited. Water buffalo became a key component in agriculture as beasts of burden and work animals since they are well-adapted to hot and humid climates, although an individual also produces valuable milk of up to 600 kg milk per year (Marai and Haeeb, 2010; Borghese, 2011; de la Cruz-Cruz et al., 2014). As mentioned by villagers during the data collection, the traditional ritual slaughter of water buffalo is the most important component of the renowned elaborate funeral rites of the Toraja ethnic group indigenous to South Sulawesi. Water buffalo are highly susceptible to thermal stress and hence should be allowed to perform wallowing behaviour (De Rosa et al., 2009; Khongdee et al.,2011), search for shade (Gu et al., 2016) or be cooled down by sprinkling water on their body at least 4-5 times per day if automatic spray systems are not used (Marai et al.,1995; Marai and Haeeb, 2010). Since the management of buffalo with access to wallows mentioned above was not always a practice followed by the farmers in the locality of our research, we decided to analyse the environmental conditions in which the buffalo is locally bred and the factors affecting display of their natural behaviour with emphasis on thermoregulatory behaviour in context of solar orientation. Behavioural thermoregulatory strategies referring to deliberate employment or avoidance of thermal heat from solar radiation was already discussed decades ago (Clapperton et al.,1965; Berry et al.,1984; Walsberg, 1983; Stevenson, 1985). Thermoregulatory behaviour in the context of solar radiation allows some mammals to effectively regulate the amount of radiant heat obtained from an environment. Although interest in science and the environment increases worldwide, our knowledge of behavioural thermoregulatory strategies used by large mammals to avoid or utilize directly the energy from solar radiation is based on just a few empirical studies describing, for example, shade-seeking and the principal responses of some species to excess radiant heat load (Maloney et al. 2005; Hetem et al., 2011; Sedláček, 2014) but not much was mentioned about thermoregulatory behaviour under solar adiation in the context of overall behaviour patterns (Mitchell et al. 2002; Maloney et al. 2005; Keren and Olson, 2006 and 2007; Ismail et al., 2011; Sedláček, 2014).

Taking into account the important role of solar radiation on body orientation and consequently thermoregulation of ruminants, there is another factor influencing an animal's orientation and that is the geomagnetic field of the Earth. Animals with the ability to orient their movements with respect to the geomagnetic field are said to have a magnetic compass sense. A well-developed magnetic compass and adaptive orientation have already been detected in various animal taxa such as insects (Hsu et al., 2007; Dommer et al., 2008, Vácha et al., 2008); lobsters (Lohmann *et al*., 1995) fish (Hart et al.,2012); amhibians (Phillips and Borland, 1992; Fischer et al., 2001); turtles (Papi et al., 2000; Lohmann et al., 2008); alligators (Rodda, 1984) and birds (Walcott et al.,1979; Mora et al.,2004; Moritz et al., 2007; Hart et al., 2013). Goal-directed magnetic compass orientation based on geomagnetic field perception was found in mice (Muheim et al.,2006; Phillips et al. (2013) and rats (Frilot et al., 2014). Siberian hamsters (Deutschlander et al.,2003) and mole-rats (Kimchi and Terkel, 2001; Thalau et al.,2006) use information from the magnetic field to remember the position of their nest or to control the direction in which they built the nest. Even some large mammals are capable of magneto-reception and use it to save their metabolic energy (Begall et al.,2008). For instance red foxes (*Vulpes vulpes*) use magnetic alignment when hunting their prey (Červený et al.,2011). Grazing and resting domestic cattle, roe deer and red deer orient their body axes position mostly in north–south (NS) which could be explained as intentional adjustment of their body position in the direction of the magnetic field lines of the Earth (Begall et al., 2008; Slaby et al.,2013). Whereas the bibliographic research revealed a range of striking adaptations of buffalo to survive in hot climates of its natural habitat, the field research pointed out some important findings to be compared with previous scientific studies devoted to other animal species.

## 1.1. Aims of the Thesis

The aim of the thesis was to summarize the current knowledge about anatomical and physiological adaptations of buffalo and cattle as well as their behaviour and ability to overcome long-term high temperatures. Another task was to describe the homeostatic factors affecting thermoregulatory behaviour of observed cattle and bufallo and asses wheather extreme temperatures or differences in weather conditions influence daily activity of animals. A further aim was to investigate whether buffalo and indigenous cattle intentionally orient their body axis alignment in relation with the magnetic field of the Earth or whether the animals change their body position according to the direction of solar radiation during hot summer days. The field research and ethological observation of both species were made in Indonesia to detect the veracity of our following hypothesis.

### 1.1.1. Research questions and hypotheses

**Does the solar radiation affect behaviour of buffalo?**

$H_1$: Frequency of orientation parallel to incident solar radiation increases with ambient temperature and perpendicular and oblique orientation is more frequent in both species at midday when the sun is at zenith.

**Does the magnetic field affect behaviour of water buffalo and cattle?**

$H_2$: Both water buffalo and cattle intend to orient their longitudinal body axis in north and south direction more frequently compared to other cardinal directions.

# 2. Literature Review

## 2.1. Origin and geographic spread of buffalo

The Asian buffalo or the Water buffalo (*Bubalus bubalis)* was first domesticated in India 5000 years ago and in China a thousand years later compared to the domestication of *Bos Taurus* and *Bos Indicus* that occurred 10,000 years ago. Although some sources suggested that taming of water buffalo occurred in Neolithic China over 7000 years ago (Barker et al., 1997; Borghese, 2011); the DNA analysis of ancient bone samples failed to establish direct links between modern domesticated water buffalo and indigenous, extinct water buffalo (*Bubalus mephistopheles*) from ancient China (Yang et al., 2008). Water buffalo were differentiated based on their morphology in two subspecies, also named types, i.e. the river buffalo and the swamp buffalo. However these types also differ in chromosome number (swamp: 2n = 48 and river: 2n = 50). The two subspecies are inter-fertile and give progeny with 49 chromosomes (Borghese, 2011).Well-defined breeds of the river buffalo are recognized in India and Pakistan, but can be found further west to the Balkans and Italy. Morphologically, the swamp buffalo are more similar to the wild Asian buffalo (*Bubalus arnee*) that inhabit south-east Asia from Assam and Nepal in the west to the Yangtze valley of China in the east (Barker et al., 1997). River buffalo are heavier, the adult male weight ranging between 450 and 1000 kg, while Swamp types weigh between 325 and 450 kg. The buffalo (*Bubalus bubalis*) population in the world is actually about 168 million heads. The Asian population of 168 million heads alone stands for 95.83 % of the buffalo on Earth. 3,717 million are in Africa, practically only in Egypt (2.24 %); 3.3 million in South America (1.96 %); 40,000 in Australia (0.02%) and 500,000 in Europe (0.30 %) of the world's population (FAO, 2005).

## 2.2. Classification of buffalo

The water buffalo (*Bubalus bubalis)* from the genus *Bubalus*, subfamily Bovinae and family Bovidea (Gray, 1821) was first named by Linnaeus in 1758. Kerr further suggested Indian water buffalo (*Bubalus bubalis arnee*) and Anoa (*Bubalus anoa*) as the names for the wild taxon of water buffalo (IT IS, 2017). Nowadays, the IUCN considers the wild forms of water buffalo under the name *Bubalus arnee* as it was not accepted for the name to be based on the domestic form *B. bubalis*. The remaining world population of wild water buffalo is listed as Endangered and totals under 4,000 individuals found in India, Nepal, Assam and Southeast Asia. The estimated population is less than 2,500 mature individuals, a result of a 50% decrease during the last three generations (Hedges et al., 2008). Also Lowland Anoa (*Bubalus depressicornis*) (C. H. Smith, 1827) and Mountain Anoa (*Bubalus quarlesi*) (ITIS, 2017) endemic to the Sulawesi and Buton islands in Indonesia are listed as Endangered species of the genus *Bubalus* with decreasing trends in population (Burton et al. 2016, IUCN). Although the Bovidea family used to belong to the order Artiodactyla also called even-toed ungulates, according to the recent genetic studies the cetaceans (whales, dolphins and porpoises) are considered to be the modern sister group of Hippopotamidae and other artiodactyls and therefore a new order Cetartiodactyla was created. Cetartiodactyla hence became one of the most diversified orders of mammals comprising the suborder Suiformes (pigs and peccaries), the suborder Tylopoda (camels and llamas), the suborder Ruminantia and the suborder Cetancodonta (infraorder Cetacea superfamily Hippopotamoidea). There are currently 332 extant cetartiodactyl species recognized and further classified into 132 genera and 22 families (Boisserie et al., 2005; Vislobokova, 2013).

## 2.3. Thermal stress in domestic ruminants

Heat stress is a worldwide problem for breeders of cattle and it is the main cause of heavy economic losses. It affects about 60 % of the world cattle population (Wolfenson et al., 2000). Exposure to solar radiation, high ambient temperatures and humidity are the main environmental stressing factors affecting domestic ruminants in the context of animal welfare (Das et al.,1999; Silanikove, 2000; De Rosa et al., 2009). Although ruminants developed sophisticated thermoregulatory mechanisms, they do not maintain strict homeothermy during heat stress (Marai *et al*, 2007). Large mammals are able to maintain favourable homeothermy only in well hydrated and nourished condition and when not compromised energetically (Hetem et al.,2016).

### 2.3.1. Management of domestic ruminants in hot climates

Hyperthermia is not only harmful to animal health, it also affects their productivity (Kadzere et al., 2002) and lowers fertility among lactating dairy cows rapidly (Wolfenson et al., 2000; Aarif and Aggarwal, 2015). However thermoregulatory reactions towards increasing ambient temperatures changes depending on breed or stage of adaptation (Mirkena et al., 2010; Pereira et al., 2014). Since the zebu cattle (*Bos taurus*) acquired genes resulting in better maintaining of thermal equilibrium within physiological and cellular levels, their genotype has been used in crossbreeding systems in order to strengthen the thermoresistance of cattle breeds. (Hansen, 2004; Mirkena et al., 2010). An experimental study conducted in a sub-tropical Egyptian desert shows the successful raising of buffalo under dry management housing (Marai *et al*, 2009). A different research analysed the impact of climatic factors of the Brazilian Eastern Amazon on respiratory frequency, rectal temperature and index of tolerance to heat in Murrah buffalo. Surprisingly, the rectal temperature of buffalo remaining in pasture without access to the shade, was lower (38.6 °C) in the afternoon compared to the group of buffalo kept in shade of the silvopastoral system (38.8 °C) (Silva *et al*, 2010). A field study of Gu et al., 2016 found no difference in rectal temperature between buffalo calves kept under non-shaded and shaded management, although the respiratory rate of the non-shaded group was grater at noon and in the evening than for calves in the shaded group. On the other hand Khongdee et al. (2011) pointed out in their study that wallowing-enabled buffalo had a significantly lower mean rectal temperature ($39.1 \pm 0.1$°C) in comparison with buffalo that had no access to wallows ($40.0 \pm 0.1$°C).

### 2.3.2. Biological responses of ruminants to heat stress

Prolonged exposure to thermal stress can trigger extensive biological changes and disturbances in the metabolism of water, protein and energy (Kadzere et al., 2002; Marai *et al*, 2007). Cattle can be defeated by hyperthermia if they fail in maintenance of thermoneutrality. The accurate onset of heat stress in cattle is complicated owing to many factors such as energy balance or metabolism of sodium, potassium and chlorine that represent the constituent elements of sweat (Kadzere et al., 2002). Panting, sweating, greater vasodilation and blood flow are the highlighted physiological consequences of heat stress while increased activity of cortisol and thyroid glands are the substantial hormonal reactions to it (Silanikove, 2000; Aarif and Aggarwal, 2015). Adaptive depression of the metabolic rate is the most significant effect of long-term exposure to heat stress and is related with reduced appetite and imbalance in hormonal secretion, enzymatic reactions, mineral and blood metabolite levels (Chaiyabutr et al.,1997; Kadzere et al., 2002; Aarif and Aggarwal, 2015). Although some breeds employ early increase of respiratory rate and moderate sweating while other breeds respond to thermal stress by moderate tachypnea (abnormally rapid breathing) and lower increase in sweating but increasing rectal temperature (Pereira et al., 2014), respiratory rate is still proved to be the most relevant indicator for monitoring thermal stress in cattle (Brown-Brand et al., 2005). Although Cattles' respiration rate, pulse rate, body temperature and general discomfort increase more slowly in cattle compared to buffalo (Marai and Haeeb, 2010).

#### 2.3.2.1. Anatomical thermoregulatory adaptations in ungulates

Adaptive heterothermy together with selective brain cooling are considered to be thermoregulatory adaptations of great importance for mammals, especially large ungulates living in warm arid environments (Mitchell et al., 2002). Selective brain cooling lowers brain temperature increased during hyperthermia whilst adaptive heterothermy helps animals conserve body water by employing body heat storage leading to a reduction of the necessity to dissipate heat by evaporation, under thermal stress (Mitchell et al. 2002, Strauss et al. 2015). A significant positive correlation was found between selective brain cooling magnitude and osmolality in sheep as they uses selective brain cooling with a greater magnitude when dehydrated as opposed to euhydrated state (Strauss et al. 2015).

##### 2.3.2.1.1 Special morphological adaptations to heat stress in buffalo

The skin of buffalo is rich in blood vessels for more effective heat conduction and dissipation. It is further covered with a sparse hair compared to the good protective coat of the cattle (de la Cruz-Cruz et al., 2014). The evaporative coetaneous cooling mechanism is weaker in buffalo due to the fact that cattle has six times greater density of sweat glands (Marai and Haeeb, 2010). Buffalo hence developed some important anatomical characteristics including a thick epidermis that contains increased amounts of melanin particles which protect the skin from the ultraviolet rays and gives it its typical dark colour (Marai and Haeeb, 2010; de la Cruz-Cruz et al., 2014). The greater activity of well-developed sebaceous glands plays role of further beneficial adaptation of buffalo. These glands produce a fatty substance, called sebum, which lubricates the hornified top layer of the skin and thus prevent water and solutes in mud from being absorbed (Marai and Haeeb, 2010).

## 2.4. Behavioural aspects of thermoregulation

### 2.4.1. Behavioural thermoregulation in ectotherms

Behavioural thermoregulatory strategies referring to deliberate employment or avoidance of thermal heat from solar radiation had already been discussed decades ago (Clapperton et al.,1965; Berry et al.,1984; Stevenson, 1985). Further studies devoted to behavioural and metabolic response of ectotherms to solar radiation and thermal stress studied for instance the grasshoppers, locusts (Lactin and Johnson, 1998), iberian rock lizards (*Lacerta monticola*) (Martin et al., 1995), giant tortoises (Coe, 2004) or crocodiles (*Crocodylus porosus*) (Seebacher et al., 1999). Bohorórquez- Alonso et al. (2011) analysed this behavioural pattern in lizards (*Gallotia galloti*) in different localities and structural habitats. They proved that more lizards oriented perpendicular or parallel in relation to the sun rays as opposed to oblique orientations. (Bohorórquez-Alonso et al.,2011). A different experiment revealed that a high proportion of the giant tortoises (*Testudo gigantea*) oriented their long axes parallel facing away from the sun on clear days (Coe, 2004). Perpendicular body orientation to the sun clearly enhances the proportion of body surface exposed to incident solar radiation. On the other hand, orientation parallel in relation to sunlight decreases the animal's surface receiving solar radiation, which allows endotherms to balance the amount of heat received from the environment in dependency on incident solar radiance, especially when the altitude of the sun is low (Berry *al.*,1984).

### 2.4.2. Behavioural thermoregulatory strategies in endotherms

Nowadays, behavioural thermoregulation associated with solar orientation is considered to be an equally crucial survival strategy as those physiological adaptations in endotherms. Studies dealing with behavioural and metabolic response of endotherms to solar radiation are focused for instance on the rock squirrel (*Spermophilus variegatus*) (Walsberg, 1988), Siberian hamsters *(Phodopus sungorus)* (Walsberg et al., 1997), (Thompson et al., 2015), elephant shrews (Elephantulus myurus), Ethiopian hedgehogs *(Paraechinus aethiopicus)* (Baker et al., 2016), yellow baboons (*Papio cynocephalus*) (Stelzner and Hausfater, 1986), springbok (*Antidorcas marsupialis*) (Hetem et al., 2009), cattle (Keren and Olson, 2006 and 2007), sand gazelle (*Gazella subgutturosa marica*) (Hetem et al., 2012a), black wildebeests *(Connochaetes gnou)* (Maloney et al., 2005a), blue wildebeest (*Connochaetes taurinus*) (Berry *et al.*,1984; Hetem et al., 2011), Arabian oryx *(Oryx leucoryx)* (Hetem et al., 2012b), impala (*Aepyceros melampus*) and eland (*Tragelaphus oryx*) (Hetem et al., 2011).

#### 2.4.2.1. Solar orientation in ungulates

The black wildebeests supposedly perceive changes in their body temperature via incident solar radiation affecting their skin temperature. The wildebeest would absorb 30% less radiant heat as a result of parallel body orientation when exposed to the sun for the whole day in comparison with perpendicular orientation of their body axis relative to the sun. (Maloney et al., 2005a). For the different body axis alignments relative to the sun see Figure 1. Solarbehaviour of black wildebeest was found to be a well-developed adaptation to regulate radiant heat in severe environments. The ability of some ungulates to preferentially orient their body orientation in relation to the sun rays was confirmed by Hetem *et al* (2011) after investigation of thermoregulatory behaviour of impala, eland and Blue wildebeest. Preference of perpendicular orientation to the sun in cold winter and parallel orientation during the hot summer was proved in all three species. About 60% of animals analysed in this study preferred perpendicular or parallel body axis alignment to the incident solar radiation dependant on the season, which demonstrates connection with energy saving maintained by the animal itself. For example a small impala managed to save up 16 % of its metabolic energy through this thermoregulatory behaviour (Hetem et al., 2011).

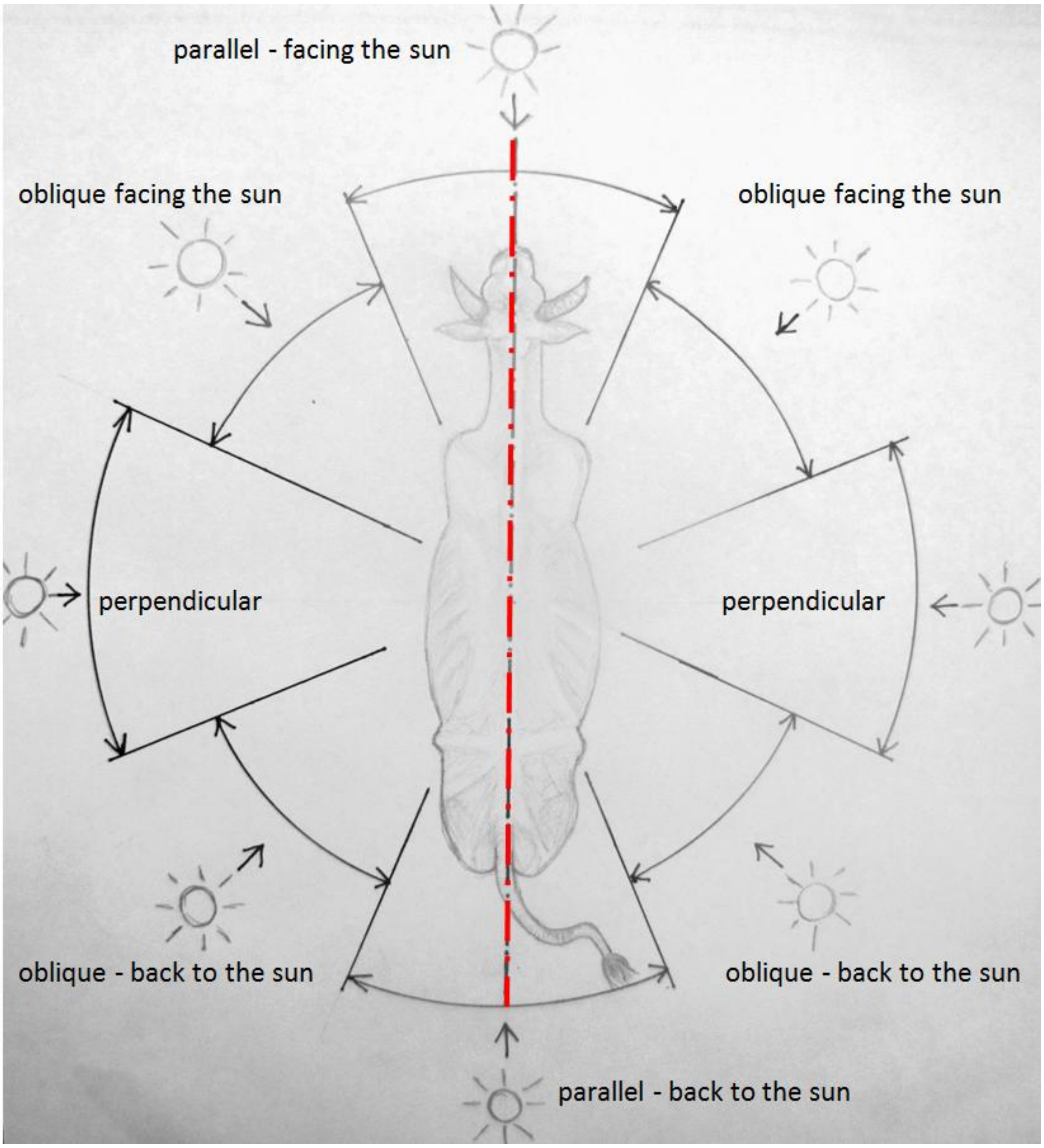


**Figure 1. Body axis alignments relative to sun rays** (*Source: author)*

### 2.4.2.2. Shade seeking and seasonal behaviour in ungulates

Blue wildebeest seek shade more often within the whole day compared to black wildebeest which remains in the sun and employs parallel solar orientation during the hottest part of the day. Such species-specific differences in thermoregulatory behaviour can results in different fitness, body mass and grazing activity of individual species. (Lease et al.,2014). Although black wildebeest seem to use alternative strategy to resist heat stress in shadeless environments, their nycthemeral activity patterns (duration of 24 hours), especially feeding at night increases as days became hotter with the change of seasons. Black wildebeest spend inactive periods mostly by lying during cooler weather conditions but standing as weather becomes hotter (Maloney et al., 2005b).

Equally, Arabian oryx search for cooler microclimates with increase of environmental temperatures and aridity (Hetem et al., 2012b), moreover they exhibit seasonal changes in daily activity patterns influenced by ambient temperature (Ismail et al., 2011; Hetem et al., 2012b). Both Arabian oryx and sand gazelle shift from crepuscular or biphasic rhythm during the wet season to a more nocturnal rhythm within the hot dry season. However the sand gazelle has higher minimum, mean but also maximum body temperatures compared to oryx (Hetem et al., 2012a), which could have been explained by the difference between body mass of these two species. Hetem et al. (2016) found out that the mean 24 h body temperatures measured in 17 species of large mammalian herbivores weighting above 10 kg decreased by approximately 1.3 °C with each 10-fold increase in body mass. Although the 24 h amplitude of the body temperature rhythm in mammalian large herbivores is independent of both ambient temperature and body mass in case of sufficient water and energy resources (Hetem et al.,2016).

## 2.5. Factors affecting the production of metabolic heat

The surface of an animal body in the context of solar radiation brings into discussion another topic, that is the influence of solar radiation on metabolic heat production by the animal depending on wind speed and pelt colour (Walsberg, 1983 ; Hetem et al., 2009). Already Walsberg(1983) states the influences of animal's surface coloration affecting exchange of heat with the ambient environment and thus metabolic heat production (Walsberg, 1983).

### 2.5.1. Influence of pelt colour

Although dark skin coloration increases heat gained from solar radiation by 5 %, the levels of ultraviolet transmitted via dark skin represents only one-sixth of those in case of the lighter ventral skin (Walsberg, 1988b). Walsberg and Wolf (1995) reported that no significant differences were found between solar heat gained from a solar simulator by two differently coloured squirrels in the test chamber, conversely Hetem et al. (2009) revealed significant differences among three different colour morphs of springbok in relation to incident solar radiation. The black pelt of springboks conveyed the highest proportion of incident solar radiation compared to the lowest proportion in white morph of springboks. Concurrently the black pelt displayed the lowest solar reflectance, the normal colour morph of springbok intermediate reflectance and the white springboks displayed the highest reflectance in the visible spectral range. The white pelt protects the springboks from the solar heat load in hot temperatures resulting in lower daily maximum body temperatures (40.2 ± 0.2 °C) than in spring (40.7 ± 0.1 °C). Conversely during winter, white springbok had lower daily minimum body temperatures (37.4 ± 0.5 °C) compared to the black pelt that resulted in lower energy cost of homeothermy and daily minimum body temperatures (38.1 ± 0.3 °C) Consequently, diurnal activity of black springbok was lower in winter since they did not need to forage as much due to lower metabolic cost. On the other hand black springbok were disadvantaged in hot periods (Hetem et al., 2009).

### 2.5.2. Influence of wind speed

Concerning warm climates, air movement and consequently wind velocity reduce the heat load gained from the environment assuming skin temperature is higher than ambient temperature (Moran et al., 2004). Stelzner and Hausfater (1986) described thermal effects of wind and sun on body posture of yellow baboons. Wind direction triggered in baboons altering of their trunk orientation, while body posture was primarily influenced by air temperature and solar radiation as to minimize the thermal stress (Stelzner and Hausfater, 1986). Experimental work with Siberian hamsters *(Phodopus sungorus)* showed that production of metabolic heat declined by 27 - 41 % according to the intensity of irradiance and wind speed (Walsberg et al., 1997). Walsberg and Wolf (1995) found out that the change of wind speed from 0.5 m/s to 4 m/s led to a 10-13 % reduction of energy gained from radiant heat in squirrels (*Spermophilus lateralis* and *Spermophilus saturates*) (Walsberg and Wolf, 1995). The heat load measured at skin level below plumage, gained by the emu (*Dromaius novaehollandiae*) from solar radiation was less than 10% of the incident solar radiation at low wind speeds compare to less than 1 % gain of heat load at wind speed above 6 m/s. Remaining 90 % of the heat load from the incident solar radiation was absorbed by the dark plumage (Maloney and Dawson, 1995). Keren and Olson (2007) demonstrated that the ambient temperatures together with the wind velocity had no significant effect on metabolic demands in well-insulated cows; compared to the magnitude of solar radiation incident to body surface that is instrumental for energy conserving (Keren and Olson, 2007). Berry et *al.* (1984) indicated in their study that body position of wildebeest is influenced by sector winds the most in midday when the sun's altitude is high. However cattle are commonly found to turn their backs against strong winds.

### 2.5.3. Influence of thermal insulation structure

Referring to the effective insulation provided by coat of animals. Indeed, differences in hair optics and coat structure can influence the solar heat gain by 40 % between animals of similar colour. However structure of thermal insulation and depths of the inner and outer coats occurring in rock squirrels can minimize solar heat gain. This represents another effective way of adjusting solar heat gain (Walsberg, 1988a).

## 2.6. Magnetoreception

Taking into account the important role of solar radiation on body orientation and consequently thermoregulation of ruminants, there is another factor influencing an animal's orientation and that is the geomagnetic field of the Earth. Begall et al. (2008) stated that some large mammals are capable of magneto-reception and use it as beneficial for saving metabolic energy. Migratory birds use a magnetic compass to keep directions of their target destination during their migratory journeys covering even thousands of kilometres (Wiltschko and Wiltschko, 2003). But how do they perceive the reference of direction provided by the Earth's magnetic field? Mouritsen *et al.* (2004).

### 2.6.1. Magnetic navigation

Animals with the ability to orient their movements with respect to the geomagnetic field are said to have a magnetic compass sense. However, a magnetic compass itself is insufficient to guide migrating animals to specific distant locations. Animals need to further locate where it is according to their final destination in order to be able set an appropriate course. Hence, some animals were hypothesized to possess an additional sense called a map sense (Lohmann and Johnsen, 2000; Gould, 2008). Further evidence supports the ‘‘magnetic map’’ hypothesis which postulates that some amphibians, reptiles and birds may derive information about the geographic position necessary for homing from subtle geographic gradients in the Earth's magnetic field (Philips,1996; Gould, 2008; Freake et al.,2006). This ability would permit animals to reach a target from an entirely unfamiliar site without goal-emanating cues to assist. It is a highly questionable hypothesis since the geomagnetic field vary in time and spatial gradients may be influenced by geological anomalies (Freake et al.,2006). For instance birds (Wiltschko and Wiltschko,1972; Freire *et al.*, 2005), marine turtles (Lohmann et al., 2008) and spiny lobsters *(Panulirus argus)* (Lohmann *et al.*, 1995) or fish (Hart et al.,2012) are indicated to exploit magnetic navigation.

#### 2.6.1.1. Magnetic navigation in aquatic environment

Hart et al. (2012) examined directional bearings of common carps (*Cyprinus carpio*) kept in large circular plastic tubs at traditional Christmas sale at 25 localities in in the Czech Republic. Carps displayed a statistically highly significant preference to orient their body axis spontaneously along the NS axis according to geomagnetic field lines (Hart et al.,2012). Laboratory experiments have revealed the ability of juvenile green turtles (*Chelonia mydas*) to learn the magnetic topography of specific feeding grounds and acquire a magnetic map which is later used for navigation toward particular locations. Adult green turtles had magnets attached to their heads and were relocated from their nesting beaches in this experiment. The fact that those turtles carrying magnets showed diminished homing ability, implies that mature turtles also use the Earth's magnetic field when navigating to specific islands or other destinations (Lohmann et al., 2008).

On the contrary, Papi et al. (2000) implemented similar experiment regarding navigational performance and course straightness of green turtles, they found no differences between 7 magnetically disrupted and 8 untreated turtles. Perhaps turtles exploit multiple cues as a source of navigational information in the marine environment. The magnetic map may be the main cue for navigation into the vicinity of a distant goal, whereas non-magnetic cues such as a sun compass may be employed to guide the turtles to the final target (Papi et al., 2000, Lohmann et al., 2008).

#### 2.6.1.2. Magnetic navigation in semiaquatic environment

For instance, adult eastern red-spotted newts (*Notophthalmus viridescens*) oriented in the direction of their home ponds distant 45 km when exposed either to the ambient magnetic field of the testing site or to an increased magnetic inclination. The changes in magnetic inclination did not affect the shoreward magnetic compass orientation of the newts. This evidence leads to the magnetic map hypothesis and point out that information about an actual animal's geographic position along a single axis (''unicoordinate map'') relative to home may be determined using the earth's magnetic field (Fischer et al., 2001). The ability to orient in the absence of terrestrial landmarks was tested on alligators (*Alligator mississipiensis*) after displacements from their home lakes (11–34 km). Geomagnetic inclination changes of as little as of 0.01–0.02° influenced homing behaviour exhibited by alligators. The age of animals seemed to improve their navigational ability. Alligators younger than a year showed the homing ability despite circuitous displacements (Rodda, 1984).

### 2.6.1.3. Magnetic navigation in birds

Altogether, a magnetic inclination compass has been indicated in all ten bird species tested for it (Wiltschko *et al*., 2011). Landing direction of mallards (*Anas platyrhynchos*) were tested including the effect of sun position, wind, the time of the year, locality, latitude, and magnetic declination. The mallards preferred landing along the NS axis independently of the direction of the bird's arrival. Moreover, the magnetic north was found to be a better predictor for landing direction compared to geographic north, thus pointing out that the Earth's magnetic field provides a landing direction indicator. (Hart et al., 2013). Birds and marine turtles are even described to use magnetic ‘sign posts’ (different magnetic condition in specific location with respect to total intensity and inclination of magnetic field) (Wiltschko and Wiltschko, 2005). In contrary, Benhamou et al.(2003) revealed that white-chinned petrels maintain efficient homing ability even if relying only on non-geomagnetic information. Five displaced white-chinned petrels (*Procellaria aequinoctialis*) were tested to see if they rely on geomagnetic information during homing to isolated breeding islands thousands of kilometres distant in the open ocean. They were prevented from using the geomagnetic field by carrying a mobile magnet on the head, on their way home. All five petrels were able to home successfully (Benhamou et al.,2003).

### 2.6.1.4. Magnetic navigation in terrestrial mammals

Muheim et al. (2006) revealed that mice are capable of goal-directed magnetic compass orientation based on the geomagnetic field perception. Phillips et al. (2013) confirmed that C57BL/6 mice use a well-developed magnetic compass to learn effectively the magnetic direction of a submerged platform inside 4-armed (plus) water maze (Phillips et al.,2013). Siberian hamsters (*Phodopus sungorus*) use information from the magnetic field to remember the position of their nest (Deutschlander et al.,2003), similarly to mole-rats (*Spalax ehrenbergi*) and Ansell’s mole-rats (*Cryptomys anselli*), which both show a directional preference to align their nest due south or the southeast direction (Kimchi and Terkel, 2001; Thalau et al.,2006). In contrast to birds, orientation of Ansell’s mole-rats was not disrupted with change in intensity of broadband field. Mole-rats continued to build nests in the south direction, indicating that their magnetic compass is not based on radical pair processes but a fundamentally different principle, which perhaps involves magnetite (Thalau et al.,2006). Neither artificial shifts of inclination nor light affected the preferred nesting direction, therefore the magnetic compass of mole-rats was specified as a light-independent polarity compass (Moritz et al., 2007). Neurons that respond to magnetic stimuli were identified in the superior colliculus of Zambian mole rats. These neurons are organized within a discrete sublayer and found to be directionally selective. This finding brings evidence for a specific mammalian brain structure being involved in magnetoreception (Němec et al., 2001).

Červený et al. (2011) suggested that Red foxes (*Vulpes vulpes*) use magnetic alignment when hunting rodents and other small animals. Results of the study revealed that most of successful jumps of foxes were clustered to the north-east (74 %) and second biggest cluster of successful jumps roughly to the S direction (15 %), whereas hunting jumps in other compass directions were largely unsuccessful in high vegetation and under snow cover (Červený et al., 2011).

#### 2.6.1.4.1 Magnetic navigation in ruminants

Begall et al. (2008) found out that grazing and resting domestic cattle, roe deer and red deer orient their body axes position mostly in the NS direction which could be explained as intentional adjustment of their body position in the direction of field lines of the magnetic field of the Earth (Begall et al., 2008). After analysis of different data, Begall et al. (2011) confirmed that cattle orient their body axis significantly in the NS direction and further indicated that cattle display magnetic alignment even more often in resting than in standing position. Slaby et al. (2013) observed cattle grazing predominantly in NS direction, however their revealing results stressed out a new phenomenon influencing orientation of cattle and that is herd density. Results of their study pointed out that mutual distances between individual animals influence their NS preference. Social interactions among animals in more dense herds may therefore inhibit the need of NS alignment. Alignment could be consequently explained as an expression of solitary behaviour in situations when animals lack social interactions (Slaby et al., 2013). Begall et al. (2013) investigated body orientation of horses by means of Google Earth aerial images clearly displaying horses in paddocks or pastures in flat areas of

Europe. The study has revealed a preference of horses for the NE/SW axial orientation (Begall et al., 2013).

On the contrary, Hetem et al. (2011) pointed out that none of the three analysed species (impala, eland and Blue wildebeest) preferred northerly alignment of their body axis. More specifically both eland and impala oriented within each compass arc for comparable period of time whilst blue wildebeest revealed preferential use of NW orientation (Hetem et al., 2011).

### 2.6.2. Mechanisms of magnetoreception

For magnetoreception, three major hypotheses are discussed. Electrosensitive marine fish such as rays have been proposed to exploit electromagnetic induction to sense the geomagnetic field. Recent research on magnetoreception of vertebrates focuses on two different mechanisms, one dependent on biogenic magnetite particles and the second mechanism based on chemical reactions that are modulated by magnetic fields (radical pair mechanism) (Lohmann and Johnsen, 2000; Johnsen and Lohmann, 2005). Amphibians are described to use a light dependent compass that is probably based on a radical pair mechanism compared to magnetite based mechanism possibly used in mammals (Wiltschko and Wiltschko, 2005). A study of Mora et al. (2004) confirmed sensitivity of pigeons to at least one component of the geomagnetic field. Further available evidence indicate that birds use both mechanisms. The radical pair mechanism in their right eye provides directional information whereas the magnetite dependant mechanism in the upper beak provides information on actual position as components of the 'navigation map' (Wiltschko and Wiltschko, 2005). The chicks which used their right eye in experiment oriented according to magnetic cues, while chicks using their left eye did not orient towards the magnetic cues. Although Rogers et al. (2008) suggest that left eye might be a matter of hemispheric specialization for specific types of cues, but not meaning that a right eye-left hemisphere is specialized for detecting the magnetic directions (Rogers et al., 2008). Interestingly, the behaviour of birds in darkness differs fundamentally from their normal compass orientation. It originates in the magnetite-containing receptors located in the upper beak instead of relying on the radical-pair mechanism (Stapput et al.,2008).

### 2.6.2.1. Magnetite based mechanism

Well-developed magnetic compass and adaptive orientations were detected already in a larval *Drosophila melanogaster* (Dommer et al., 2008) and the American cockroaches (*Periplaneta americana*) even after loss of their antennae which had been suggested to play a positive role in magnetoreception of insects (Vácha et al., 2008). Honeybees (*Apis mellifera*) were found to undergo iron biomineralization during which superparamagnetic magnetite is formed in iron granules (Hsu et al., 2007). Magnetite is a biogenic material with unique ferromagnetic properties and has been identified also in the magnetotactic bacteria, protoctists, dolphins (Kirschvink and Gould, 1981; Kirschvink, 1989), homing pigeons *(Columba livia)* (Walcott et al.,1979), lobsters amphibians, fish, reptiles, birds and different mammals (Wiltschko and Wiltschko, 2005). Single-domain tiny crystals of magnetite ($Fe_3O_4$) are organized in structures called magnetosomes, biological bar magnets (Kirschvink, 1989; Kirschvink et al., 2001). Magnetite hence may allow the associated cytoskeleton to transduce information received from geomagnetic field to the nervous system and initiate a neural response (Lohmann and Johnsen, 2000).

This mechanism may be applicable to most magnetotactic organisms and provide the basis for their magnetoreception (Hsu et al., 2007). Although the volume fraction of magnetosomes found in higher organisms is rather small, enough of these structures with high energetic potential are present to ensure sensitivity to extremely small fluctuations and gradients in the geomagnetic field (Kirschvink, 1989; Kirschvink et al., 2001). Lobsters exploit for their navigation a polarity compass which has properties incompatible with chemical magnetoreception but it has properties consistent with magnetite (Lohmann *et al*., 1995) Animals developed separate types of magnetic organs specialized for determining location or direction. The magnetite organs occur in innervated tissue of the ethmoid sinus near the nose in most magneto-sensitive vertebrates (Gould, 2008). An experimental study of Mora *et al* (2004) suggests that the detector intended for magnetoreception is localized in the upper beak tissue of birds. Moritz et al. (2007) described association of the magnetite particles found in the upper beak area of birds with afferent trigeminal terminals, especially with the ophthalmic nerve branches (Moritz et al., 2007). The ability to detect electromagnetic fields was tested on Sprague–Dawley rats in two independent experiments using both awake rats and rats under anaesthesia. The field detection took place somewhere in the head, possibly via sensory neurons in facial skin, which further synapse in the trigeminal nucleus and consequently project to the thalamus through glutamate-dependent pathways (Frilot et al., 2014).

#### 2.6.2.2. A radical pair mechanism

Birds’ capability to detect the direction of the Earth's geomagnetic field is based directly on a chemical reaction whose product yields are strictly connected with the orientation of the reactants within the field. The avian magnetic compass is thus based on photon absorption leading to the creation of radical pairs (Ritz *et al*., 2000). A radical pair is described as an intermediate product of short-lived reaction and comprises 2 radicals formed in tandem (Rodgers & Hore, 2009). A magnetic compass functions only when the radical pairs are aligned within the host receptor cells which must be spatially distributed within the bird’s body (Ritz *et al*., 2000). The radical pair compass hypothesis has been split into two separate related hypotheses, as mentioned by (Kirschvink, et al., 2010) as follows. The first part of the hypothesis says that “the geomagnetic field modulates the natural, hyperfine-field-driven singlet to triplet interconversion (and vice versa) in a radical pair under physiological conditions and the corresponding chemical signal (magnetically induced change in singlet/triplet yield) is transduced to the nervous system in a specialized sensory cell.“ while the second one states that “ a spatially and temporally coherent alignment of radical-pair host molecules within this sensory cell can exploit the axial anisotropy of the radical-pair reaction to extract information about the axial orientation of the external magnetic field ”(Kirschvink, et al.,2010).

Magnetoreception in birds and effects of geomagnetic fields on radical pairs are poorly understood. We still lack information on various mechanisms including primary biophysical detection events, pathways used for signal transduction, neurophysiology of the process and the mechanism how is the information delivered and evaluated in the brain (Rodgers & Hore, 2009). Thalau et al. (2006) pointed out that even a very weak high-frequency fields affect the orientation of migratory birds, indicating that the avian magnetic compass relies on a radical pair mechanism (Thalau et al.,2006). Radical-pair mechanism has been demonstrated in domestic chickens and two passerines (Wiltschko *et al*., 2011). It has been proposed that the magnetically sensitive photochemical reactions dependent on radical pairs take place in a specialized ocular photoreceptor serving as the primary detector (Rodgers & Hore, 2009). Both theoretical considerations and behavioural evidence have suggested that radical pair processes in light-sensitive molecules of the retina could enable migratory birds to sense the geomagnetic field as visual patterns (Mouritsen *et al*., 2004, Wiltschko *et al*., 2011).

A radical pair might be composed of cryptochrome photoreceptors if they are paired with molecular oxygen as a reaction partner (Ritz *et al*., 2009). Cryptochromes, the photoactive proteins have been suggested as the magnetoreceptor molecules already in 2000 (Ritz *et al*., 2000). Cryptochromes, 50 to 70 kDa blue-light photoreceptor flavoproteins were first identified in plants in 1993 and since detected in bacteria, insects, and animals (Rodgers & Hore, 2009). Cryptochrome has been found in specific cells, particularly in large displaced ganglion cells in the retina of migratory garden warblers (*Sylvia borin)* (Mouritsen *et al*., 2004) and still persist the current candidate protein that may host magnetically sensitive radical-pair reactions (Kirschvink, et al.,2010). Remarkable difference between migrating garden warblers and nonmigrating zebra finches *(Taeniopygia guttata)* was found in cryptochrome expression particularly pronounced in the large displaced ganglion cells that are known to project solely to a brain area where magnetosensitive neurons have been reported (Mouritsen *et al*., 2004).

## 2.6.3. Factors affecting the magnetoreception of animals

### 2.6.3.1. Light-dependency of magnetoreception

A behavioural study on male eastern red-spotted newts (*Notophthalmus viridescens*) revealed that magnetic compass and thus orientation of these semiaquatic salamanders is dependent on the wavelength of light (Phillips and Borland, 1992). Equally Wiltschko *et al*.(2005) reported that the fixed direction behaviour observed in birds represents a specific type of light-dependent orientation based on a different principle in comparison with the normal inclination compass which is used for migratory orientation and based on a radical-pair mechanism (Wiltschko *et al*.,2005). Moreover light-dependency of magnetic inclination compass was proven in five bird species (Wiltschko *et al*., 2011). The birds possess a light-dependent magnetoreception system which is based on magnetically sensitive and antagonistically interacting spectral mechanisms. Perhaps, one of these mechanisms is high-sensitive short-wavelength and the other low-sensitive but long-wavelength mechanism. (Muheim et al., 2002 ).

### 2.6.3.2. Magnetoreception dependent on intensity of the field

Balmori (2015) highlights the effects of anthropogenic electromagnetic radiation in the radiofrequency range on animals and wildlife in general. Exposure to radio frequency fields in the MHz range may alter the receptor organs of insects and birds to orient in the direction of Earth's magnetic field. Such radio frequency fields and radio frequency noises are found near base stations and urban areas that together with increasing use of wireless telecommunication technologies causes growth of electrosmog that may interfere with the process of magnetoreception in some animal species (Balmori, 2015).

For instance magnetoreception was proved to be intensity dependent in European robins (*Erithacus rubecula*) and thus higher intensities can lead to disorientation of migrating birds or other magnetosensitive animals (Muheim et al., 2002). On the other hand an experiment with European robins (Erithacus rubecula) pointed out that a high-frequency magnetic field of 1.315 MHz did not affect their orientation, whereas local anaesthesia of the upper beak resulted in disorientation (Stapput et al.,2008).

#### 2.6.3.2.1 Magnetic sensitivity of domestic ruminants

Burda et al. (2009) demonstrated the sensitivity of magnetoreception in ruminants. Cattle grazing in localities, where high-voltage power lines appeared at a distance of at least 500 m, aligned their body axes significantly in the NS direction. Conversely, cattle grazing within a radius up to 150 m or under the high-voltage power lines exhibited no preference for orientation in a certain direction. Similarly, roe deer shown roughly N-S alignment in localities with no overhead high-voltage power lines compared to those roe deer grazing close to power lines and exhibiting random body orientation (Burda et al., 2009). Although a NE/SW axial preference was detected in horses that were grazing either distantly from power lines and in the vicinity of lines that produced extremely low frequency electromagnetic fields (Begall et al., 2013), behavioural studies analysing orientation of animals should be conducted in the distance of slopes and settlements, but also in certain vicinity of high voltage power-lines (Begall et al., 2011). Neither grazing cattle nor roe deer oriented with power line direction in the vicinity of power lines which indicates that visual perception of the power lines had no significant effect on the orientation of the ruminants (Burda *et al*,. 2009). A retrospective cohort study by Algers and Hennichs (1985) analysed whether 400-kV lines could influence fertility of cows bred on 106 farms in Sweden. Although no significant difference was found between cows grazing beneath 400-kV lines and non-exposed cattle in this study (Algers and Hennichs, 1985), results of different experimental study indicate that exposure to extremely low frequency electromagnetic field may increase the duration of complete oestrous cycle in dairy cows. The mean duration of oestrous cycle was 19.5 ± 0.4 for non-exposed and 21.3 ± 0.4 days for the exposed cows (Rodriguez et al., 2003).

All of the previous studies on bovines were conducted with use of high voltage commercial lines. The amount of current in these lines changes considerably across time therefore it is hard to predict the uniformity of the electric and magnetic fields in the lines. The possible effects of the alternating current in lines on various studies led to construction and experimental use of a bovine exposure chamber within which the animals are exposed to uniform and controlled electric and magnetic fields (Burchard et al.,1996; Burchard et al., 1998; Nguyen et al.,2005).

# 3. Materials and methods

## 3.1. Study area

The behavioural observation was carried out in the Napu valley, North Lore (Poso Regency), Central Sulawesi, the Republic of Indonesia (see Figure 2). Location with a good overview of the whole surface of the enclosure was chosen as a base for observation. Swamp buffalo (*Bubalus bubalis*) (Barker et al., 1997), locally referred to as Napu buffalo and local Donggala cattle (*Bos taurus*) were housed on two separate, nearby located pastures in Sedoa village bordering seasonal tropical forest of Lore Lindu National Park (1°21'02.4"S, 120°20'30.5"E for cattle; 1°20'55.4"S 120°20'09.2"E for buffalo).


**Figure 2. Location of data collection**

*(Source: Google Maps. 2017. Available at https://www.google.cz/maps /: Accessed 2017-03-26.)*

Despite the tropical climate, the temperatures were quite constant due to the altitude of the pastures approximately 1,200 m above sea level. Mean precipitation in the Napu valley is 4.77mm, mean air temperature 20.98°C, mean solar radiation 17.44 MJ/m$^2$, wind speed 0.98 m/s and air humidity 81.56 % reffering to the time of the year when the observation took place and that was in October and November. The vegetation in the area consists of grassland, several shrubs fe. *Coffea* sp. and *Acacia* species. The vegetation of native grasses is dominated by *Themeda triandra, Sporobolus* sp. *Paspalum scrobiculatum L* and *Hyperrhenia* sp. The elephant grass is the most planted introduced grass species to be found everywhere on the road side or unused land.

## 3.2. Animals

Thorough identification of individual animals preceded the initiation of my own observation and it was assisted by their breeders (the single breeder of the buffalo herd and 3 different breeders of the cattle from whom we directly learned the age of the animals. Although the individual animals could be distinguished by differences between their physical structure (body condition), shapes and sizes of their horns, position of ears or colour pattern; we decided to differentiate the animals by tying up ribbons of various colours around their neck to assure their easy recognition (see Figure 3)

**Figure 3. Example of marking by ribbons for easier recognition.** *(Source: author)*

The swamp buffalo herd consisted of two bulls (average body mass 366 kg) and 4 four cows (average body mass 290 kg) compared to the herd of cattle that was composed of two bullocks (average body mass 161 kg) and four heifers (average body mass 137 kg), as mentioned in Table 1. However, one of the females had to be omitted from the data analysis in case of cattle observation, as the individual was not of the same breed (Balinese cattle) and therefore could influence the results. Average age of buffalo was 7 years and ranged from 4 to 7 years whereas the average age of observed cattle was only a year with no significant variance (P = 0.672) as presented in Table 1.

Both buffalo and cattle were brought on the pasture (place of the observation) after sunrise and left the pasture at dusk. Animals grazed on natural vegetation plus additional Elephant grass and *Acacia* sp. were supplemented daily. Local conditions could not provide a permanent water source, thus drinking water was provided to animals periodically several times a day. Nor shaded areas, nor bathing reservoirs were accessible at either site. Animal weighing was carried out at the end of the data collection with the assistance of their breeders using a portable digital scale for weighing livestock.

**Table 1. General information on individual animals participating in the observation**

| Buffalo | age [years] | weight [kg] | mean weight [kg] |
|---|---|---|---|
| Male | 5 | 396 | 365.5 |
| | 4 | 335 | |
| Female | 11 | 310 | 290.3 |
| | 11 | 339 | |
| | 7 | 290 | |
| | 4 | 222 | |

| Catttle | age [months] | weight [kg] | mean weight [kg] |
|---|---|---|---|
| Male | 8 | 143.5 | 161.3 |
| | 18 | 179 | |
| Female | 17 | 156 | 137.2 |
| | 8 | 125 | |
| | 9 | 130.5 | |

Behavioural observations had no impact on display of natural behaviour in participating animals, thus documentation of ethical approval was not needed.

## 3.3. Data collection

The observations were conducted from September to November 2015 (out of a main monsoon season). Data collection was held for 39 days. Water buffalo were observed for 20 consecutive days “ local Donggala cattle were subsequently observed for 19 days; water buffalo from 30 September to 20 October and Donggala cattle 21 October to 9 November. Scan sampling method (Altman, 1974) was used to record changes in behaviour of each individual from the buffalo and cattle herds every 10 minutes 8 hours per day from 8 am until 4 pm.

Parallel (PAR), perpendicular (PER) or oblique (OBL) orientation of long body axis relative to incident solar radiation was recorded as well as the direction each individual was facing according to compass orientation that was classified into eight 45° arcs relative to magnetic north (Maloney et al. 2006). The orientation of the long body axis according to the eight created compass groups is further referred as the magnetic alignment. For instance north body alignment is always mentioned as body axis orientation of individual heading north according to compass group.

Behavioural changes were classified into eight specific categories (lying = L, standing = S, movement = M, feeding = F, drinking = D, excretion = E, social interaction = SI, other conduct = OC). Scan samples were recorded into previously prepared tables at 10 min intervals (Altman, 1974).

The dry-bulb temperature (°C) was measured in an open area by the portable thermometer and wind speed ($m \cdot s^{-1}$) was detected by use of portable digital anemometer. Both physical quantities were measured at the height of 1m above ground and each of the sampling sites were protected from solar radiation by a tent which was ventilated from the sides.

## 3.4. Data analysis

Statistical evaluations as well as all graphs presented in the thesis were conducted with use of statistical software StatisticaCz 12 (StatSoft, Inc., 2013). The body axis orientation of both species was analysed using various methods to verify our hypotheses. Data were statistically evaluated using filters for specific categories of obtained scans and different statistic tests. The data used for the statistical analysis were filtered only by different categories “lying”, “feeding” and “standing” to prevent the results from unintended effects on body axis alignment in either buffalo or cattle. After all, those other behavioural categories like movement, drinking, excretion and social interaction, that were omitted from analysis account for only 5% of collected data in buffalo and 6% in cattle.

### 3.4.1. Solar orientation

Only data from times when the sun was visible, were used in this part of analysis. Mean ± SE air temperature and wind speed from the whole period of observation were analysed by ANOVA taking individual scans as fixed factors, see Tab.2 in Results chapter. Univariate Tests of Significance detected significant difference between the proportion of parallel, oblique and perpendicular alignment of body axis towards the solar radiation in both species. Analysis of variance (ANOVA) was used to determine the species-specific preference of body axis alignment relative to the sun depending on the dry-bulb temperature.

Linear regression of data without normal distribution was tested using the generalized linear model (GLM). Proportion of individual solar orientations was used as a fixed dependant variable throughout the GLM analysis whilst the independent variables changed depending on the specific influences that were tested (i.e. orientation, species, day period, day time, weather, temperature, individual, sex or age).

Between-Subject Effects were set up during GLM to examine differences between individual animals when the independent variable was continuous or between groups when the independent variable was. Between-Subject Effects were always based on combination of variable “orientation” plus another variable such as orientation, species, day period, day time, weather, temperature, individual, sex, or age.

Tukey's honest significance tests (HSD) were used to identify the sources of significant differences between analysed categories. The pos-hoc tests supported us with more specific details of the results.

### 3.4.2. Magnetic alignment

For the assessment of body alignment of cattle and buffalo depending on magnetoreception we used the same statistic methods as for the analysis of Solar orientation of buffalo and cattle, mentioned above. Proportion of magnetic alignments relative to eight previously designed compass groups was used as a fixed dependant variable throughout the GLM analysis compared to independent variables which changed depending on the specific influences tested (i.e. orientation, species, day period, day time, weather, temperature, individual, sex or age). Between-Subject Effects used in this analysis were always based on “magnetic alignment” plus other variable such as species, day period, day time, weather, temperature, individual, sex, or age.

Direction in which each individual aligned according to compass was recorded and divided into eight 45° arcs relative to magnetic north (Maloney et al. 2006). Compass groups for different orientations contained following arcs: N= 338 - 22°; NE= 23 - 67°; E= 68 - 112°; SE= 113 - 157°; S= 158 - 202°; SW= 203 - 247°; W= 248 - 292° and NW= 293 - 337°. Univariate Tests of Significance pointed out statistically highly significant difference between magnetic alignments of both species. Thus both species were further analysed separately.

### 3.4.3. Other influences

Specific relations between solar orientation, sex, age, day period, day time and magnetic alignment, were analysed after setting up different filters for the whole data set, according to the targeted outcomes.

#### 3.4.3.1. Influence of day time and day period

For instance, the influence of day time on orientation of animals was assessed after categorizing the data by hourly groups. Lease et al.,2014 states that the black wildebeest employed parallel orientation towards the sun most likely during the hottest daily times when the sun was not in overhead direction. Solar noon (+/- 1 hour) could affect our results (Lease et al.,2014), that is why we decided to analyse to assess the impacts of certain parts of the day. Category day period was created dividing the period of daily observation into three parts based on times of sunrise, solar noon and sunset. The category “morning” was set up for day times between 8:00 AM and 11:00 AM, category “afternoon” accounts for data collected between 1:00 PM and 4:00 PM while the data collected between 11:00 AM and 1:00 PM were used as a control for the times when the sun was directly above the animals based on Lease et al (2014) and the fact that in period of data collection (October and November) was the precise solar noon at our location between 11:44 AM and 11:51 AM. Sunrise ranged between 5:40 and 5:47 AM and sunset ranged between 5:49 PM and 5:54 PM in period of field research *(Source: timeanddate.com. 2016. Available at http://staging.timeanddate.com/ worldclock/astronomy.html?n=2647&month=10&year=2015&obj=sun&afl=-12&day=1/:Accessed 2016-10-24.)*

#### *3.4.3.2.* Influence of weather on magnetic alignment

Combination of two independent variables “weather” and “temperature” was used for multiple regression to assess the impact of dry bulb temperature on proportion of magnetic alignment relative to all compass groups within the three individual categories for weather. The selected categories include cloudy, sunny and changeable weather.

# 4. Results

The data set contains precisely 320 hours of data from observation that were further analysed. Six buffalo and five heads of cattle indigenous to Sulawesi provided us with 11,400 scan samples

## 4.1. Solar orientation and thermoregulatory behaviour

Relative frequency of PAR and PER alignment of body axis due the solar radiation differed significantly between species ($F_{2,840}$ = 37.25, P < 0.0001).Whilst buffalo exhibit a significant preference for PAR orientation, compared to OBL and PER (Tukey HSD, P <0.0001), cattle were found significantly more often oriented PER towards sun rays compared to other orientations (Tukey HSD, P <0.0001) as presented in Figure 4.

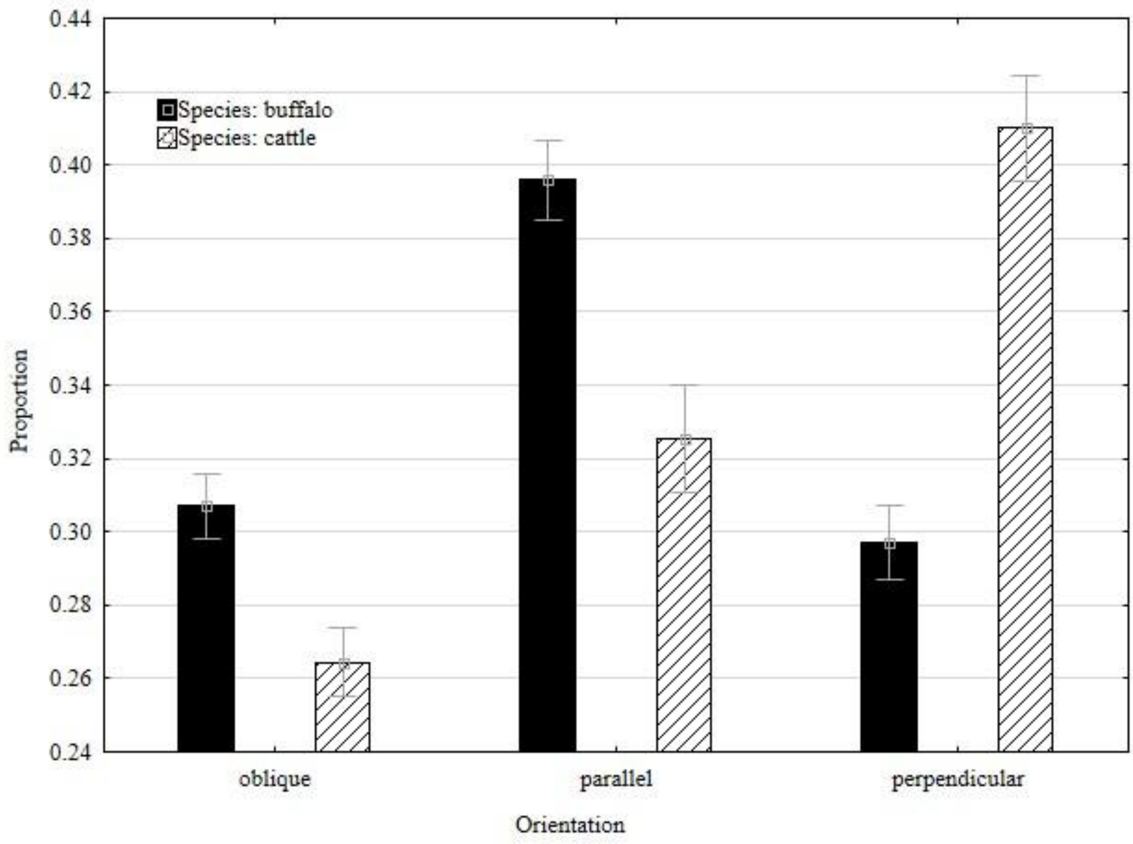


**Figure 4. Species specific differences between mean ± SE proportions of body axis orientation relative to solar radiation**

### 4.1.1. Impact of day period and day time on solar orientation

Day time ( individual hours) significantly influenced solar orientation of both species (buffalo : $F_{14,444} = 9.273$, $P < 0.0001$; cattle : $F_{14,354} = 5.615$, $P < 0.0001$). Day period was not proved to influence orientation of buffalo any significantly by GLM ( $F_{4,459} = 2.373$, $P = 0.052$), however significant differences were detected by Post hoc tests and are obvious from Figure 4 and 5. Buffalo significantly preferred PAR orientation in morning (Tukey HSD, $P < 0.0001$) as well as in the afternoon (Tukey HSD, $P < 0.05$) in comparison with other directions. Day period significantly influenced solar orientation of cattle ($F_{4,369}= 8.779$, $P < 0.0001$). Cattle significantly preferred PER orientation in morning (Tukey HSD, $P < 0.0001$) compare to other directions. No significant preference for orientation relative to sun in midday was detected in cattle (Tukey HSD, $P > 0.05$). Cattle exhibited significant preference for PAR and PER orientation compared to OBL in the afternoon (Tukey HSD, $P < 0.05$).

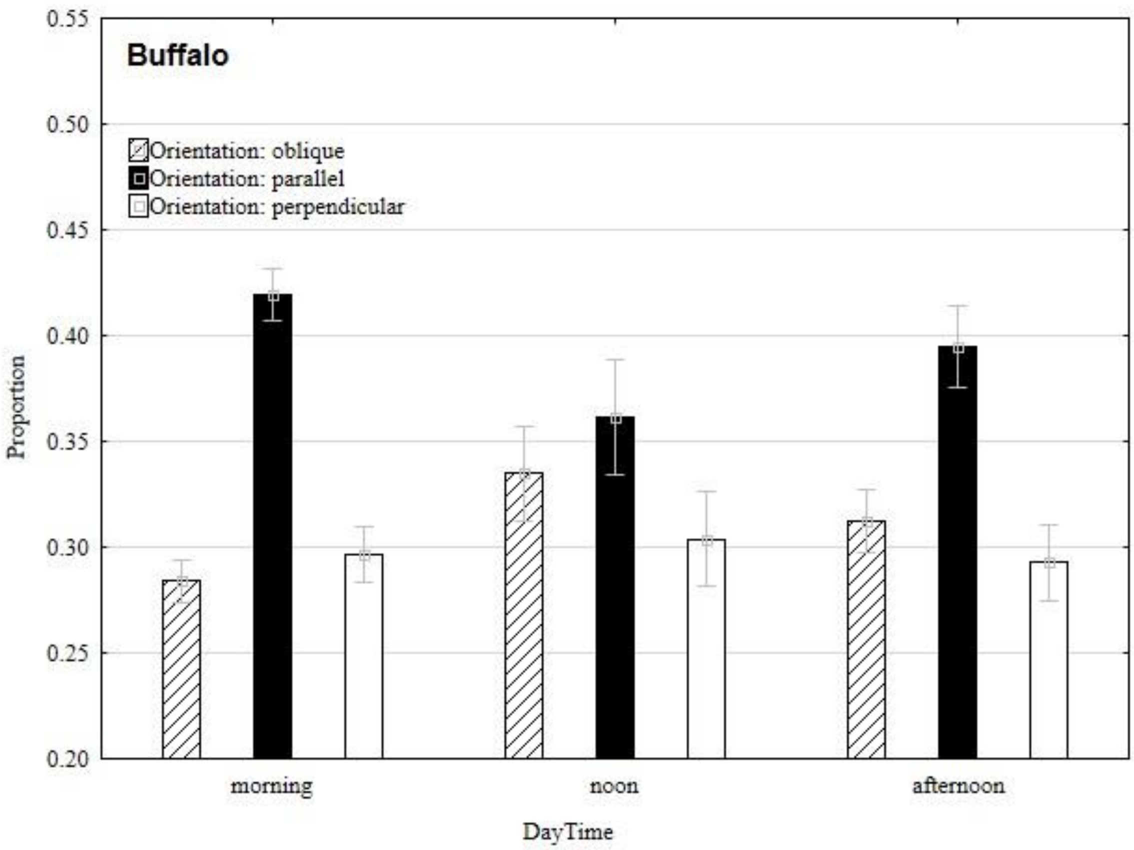


**Figure 5. Distribution of buffalo orientation mean ± SE relative to solar orientation in different day periods**

When comparing preference of individual solar directions in terms of whole day, PER orientation was preferred significantly more in morning compared to preference of PAR orientation in midday and afternoon (Tukey HSD, P < 0.05) (Figure 6).

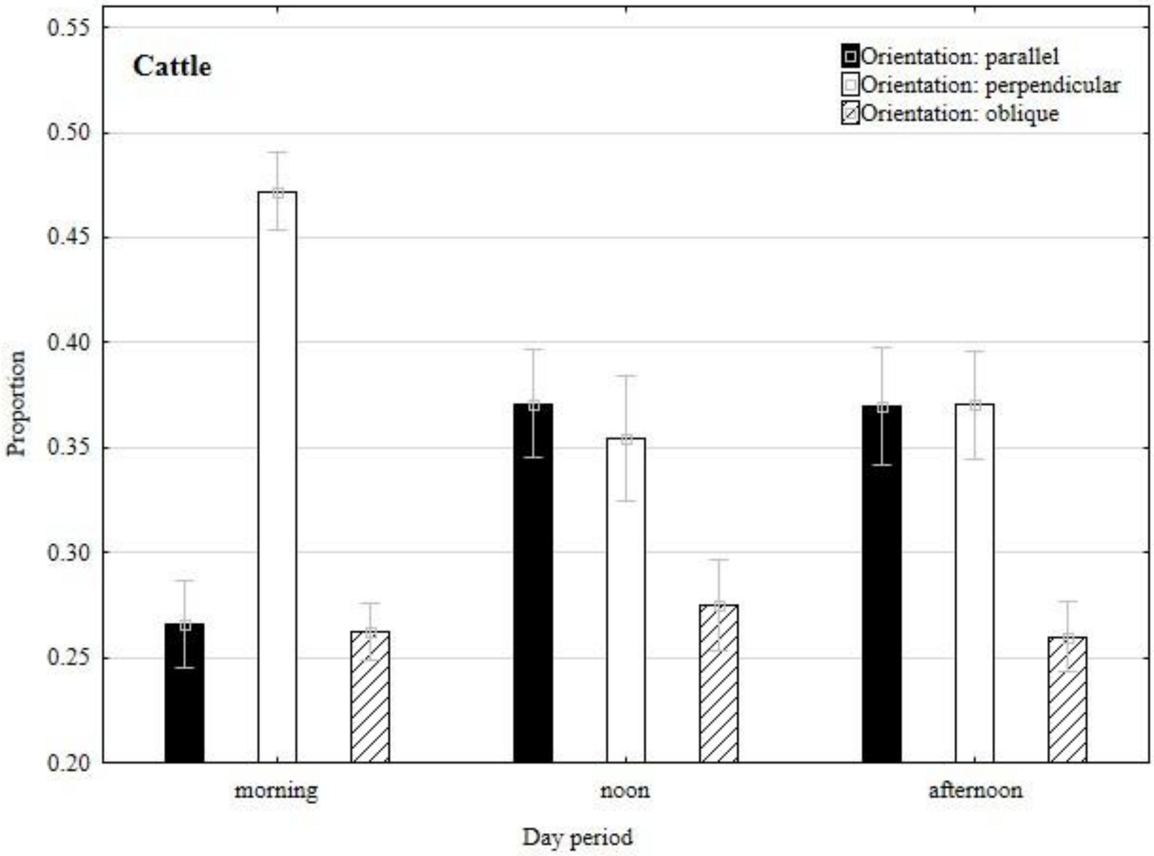


**Figure 6. Distribution of cattle orientation mean ± SE relative to solar orientation in different day periods**

### 4.1.2. Impact of temperature

Mean temperature was significantly different during individual observations of buffalo and cattle ($F_{1,844}$ = 18.749, P < 0.0001). Mean dry bulb temperature during the observation period of buffalo was 29.4°C ($T_{min}$= 20.9°C and $T_{max}$= 34.6) in comparison with the mean dry bulb temperature during observation in cattle reaching 28.6°C ($T_{min}$= 21.8°C and $T_{max}$ = 32.4). For more specific overview of mean temperatures see following Table 2.

**Table 2. Overview of mean, minimum and maximum air temperatures for the period of observation**

| Air temperature(°C) | | mean | min | max |
|---|---|---|---|---|
| **morning** | buffalo | 26.63 | 20.93 | 32.89 |
| **morning** | cattle | 26.40 | 21.78 | 32.30 |
| **midday** | buffalo | 31.49 | 26.97 | 34.40 |
| **midday** | cattle | 30.17 | 27.83 | 32.30 |
| **afternoon** | buffalo | 30.09 | 27.17 | 34.64 |
| **afternoon** | cattle | 29.28 | 25.12 | 32.40 |

Both buffalo and cattle showed preference for PAR orientation to incident solar radiation with increasing dry bulb temperatures. However, the relationship among variables was very weak ( buffalo: $r = 0{,}033$, $p = 0{,}021$) and (cattle: $r = 0{,}193$, $p < 0{,}0001$), based on a linear regression analysis.

In case of buffalo, there was significant but very weak correlation between the temperature and OBL solar orientation ($r = 0.041$, $p = 0.011$) and no significant association between ambient temperature and PER orientation ($P > 0.05$).

Proportion of PER orientation in cattle decreased with increasing temperature, although the correlation was very weak ($r = 0.161$, $p < 0.0001$) and no significant association ($p > 0.05$) was found between OBL orientation and temperature in case of cattle.

### 4.1.3. Impact of age, sex and individual animals

No relationship between proportion of different solar orientations and age of animals was found (Multiple regression, $P > 0.05$). Solar orientation was not significantly affected by sex of observed animals ($P > 0.05$) and neither significant difference in solar orientation was revealed between individual animals ($P > 0.05$).

## 4.2. Magnetic alignment

Based on the statistical analyses, buffalo and cattle were oriented their body axis relative to compass directions with a significant interspecies difference ($F_{7,2511}$ = 12.838, P < 0.0001). Proportions of body axis alignments in both species relative to compass directions are visualized in Figures 7 and 8.

Cattle significantly more often employed N magnetic alignment compared to buffalo (Tukey HSD, P < 0.05). On the other hand, buffalo employed SW and W alignment significantly more often than cattle did (Tukey HSD, P < 0.05). No other significant cardinal preference was revealed between the species (Tukey HSD, P > 0.05). GLM analysis of individual species revealed some interesting results. Significant differences were revealed in preference for magnetic alignment of both cattle ($F_{7,1240}$ = 28.722, p = 0.0000) and buffalo ($F_{7,1271}$ = 25.627, p = 0.0000) as presented in following graphs (see Figures 7 and 8).

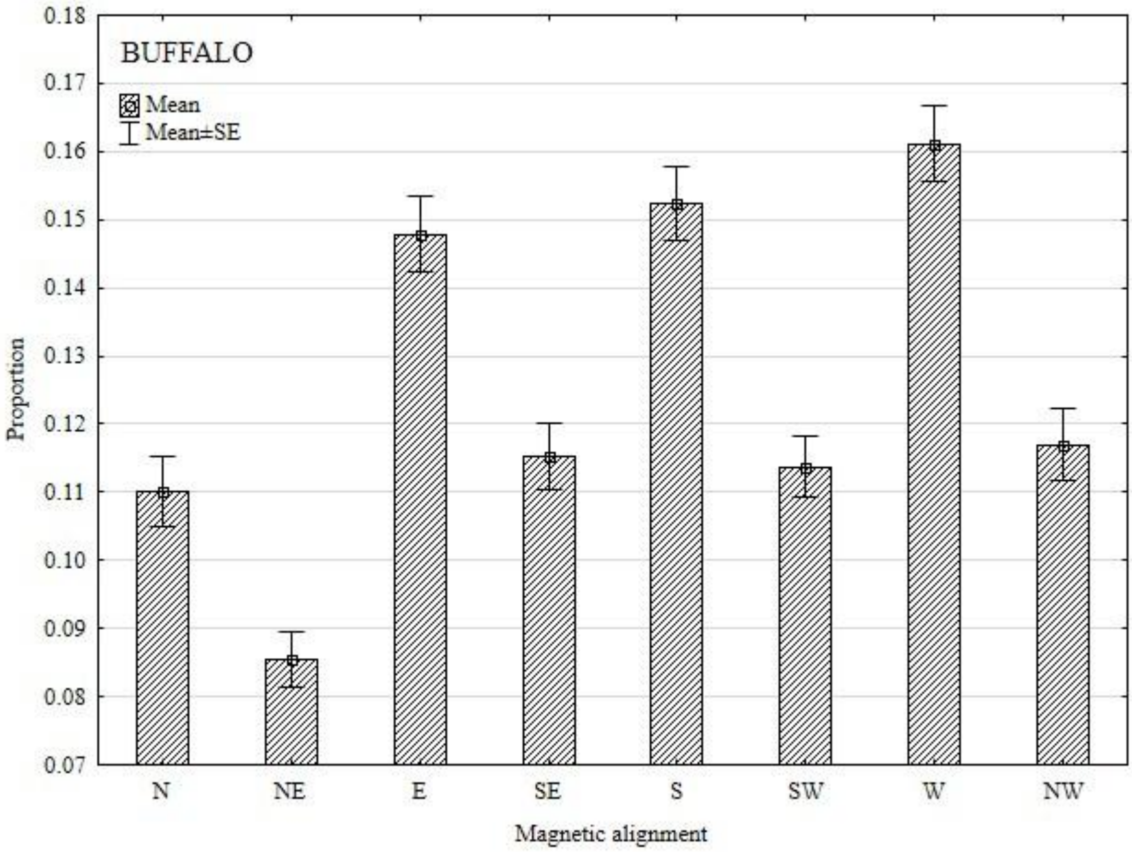


**Figure 7. Proportion of body axis alignment in buffalo relative to compass groups**

Cattle in general significantly preferred E magnetic alignment (Tukey HSD, P < 0.01) and S alignment (Tukey HSD, P < 0.05) compared to other compass groups. No significant difference was found between preferences of E and S alignment (Tukey HSD, P > 0.05).

Buffalo also preferred E alignment (Tukey HSD, P < 0.001) compared to other compass groups except for S and W alignments that were also significantly preferred by buffalo (Tukey HSD, P < 0.0001 for S) and (Tukey HSD, P < 0.001 for W). No significant difference was found among preferences for these three compass groups (Tukey HSD, P > 0.05).

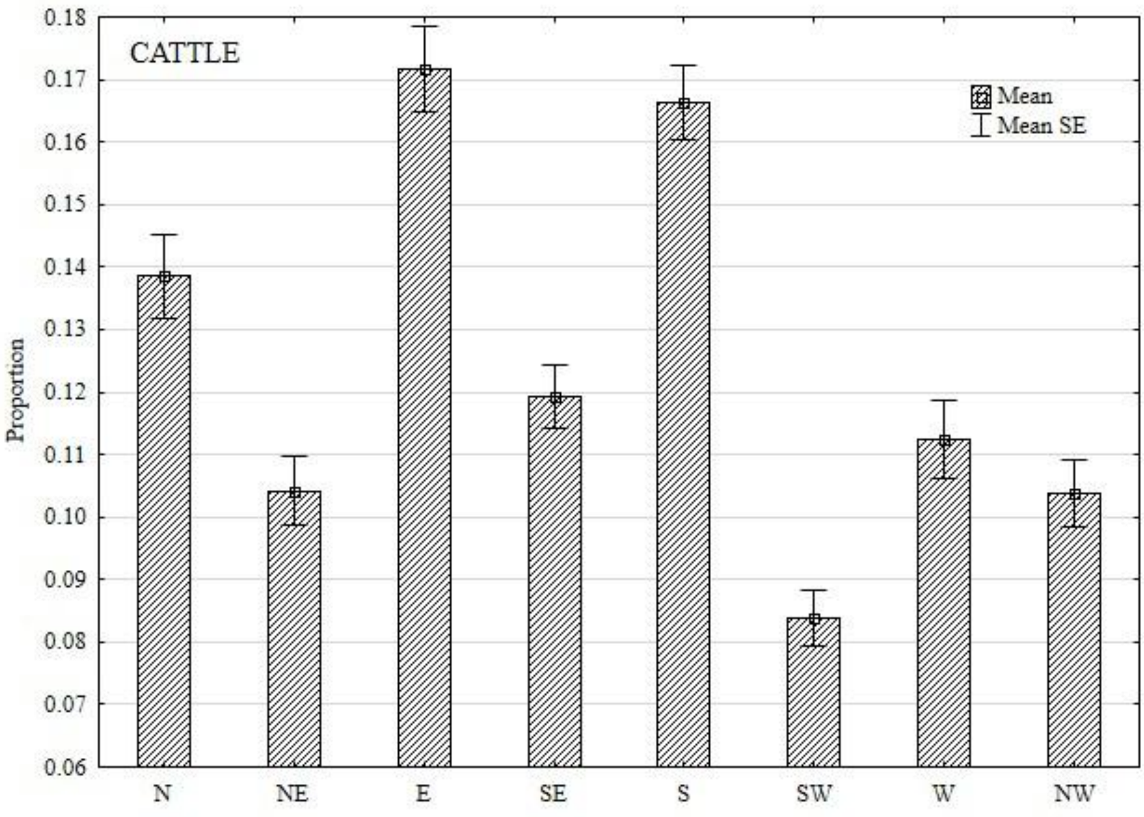


**Figure 8. Proportion of body axis alignment in cattle relative to compass groups**

### 4.2.1. Impact of day period on magnetic alignment

Although GLM analysis revealed statistically significant difference among magnetic alignments and three different day periods in case of cattle ($F_{14,1224}$= 2.420, p = 0.0024), the pos-hoc tests pointed out that the differences among magnetic alignments in cattle have no statistical significance (Tukey HSD, P > 0.05).

Neither magnetic alignment of buffalo was significantly influenced by three different parts of day ($F_{14,1255}$= 1.688, p = 0.0523).

### 4.2.2. Impact of day time on magnetic alignment

As in the previous case, magnetic alignment of both species was not significantly dependant on time of the day (hourly groups). Even though GLM pointed out significant influence of day time on magnetic alignment in buffalo ($F_{49,1215}$ = 1.679, p = 0.0026), further analysis by post-hoc tests did not verify the significant impact of day time on orientation of buffalo due any of compass directions (Tukey HSD, P > 0.05).

### 4.2.3. Impact of weather on magnetic alignment

Significant impact of weather on magnetic alignment of cattle was pointed out by GLM, however following post.hoc testing found no significant difference between the proportion of magnetic alignment in cattle relative to individual compass groups compared to magnetic alignment in cattle with regards to three different weather groups ($F_{14,1224}$ = 1.969, p = 0.0171). It means that for instance the proportion of N alignment did not vary significantly depending on whether it was sunny, cloudy or changeable weather (Tukey HSD, P > 0.05). For specific results see Figure 9. In case of buffalo, no significant association between individual weather groups and orientation relative to cardinal directions was revealed ($F_{14,1255} = 0.877$, p = 0.584).

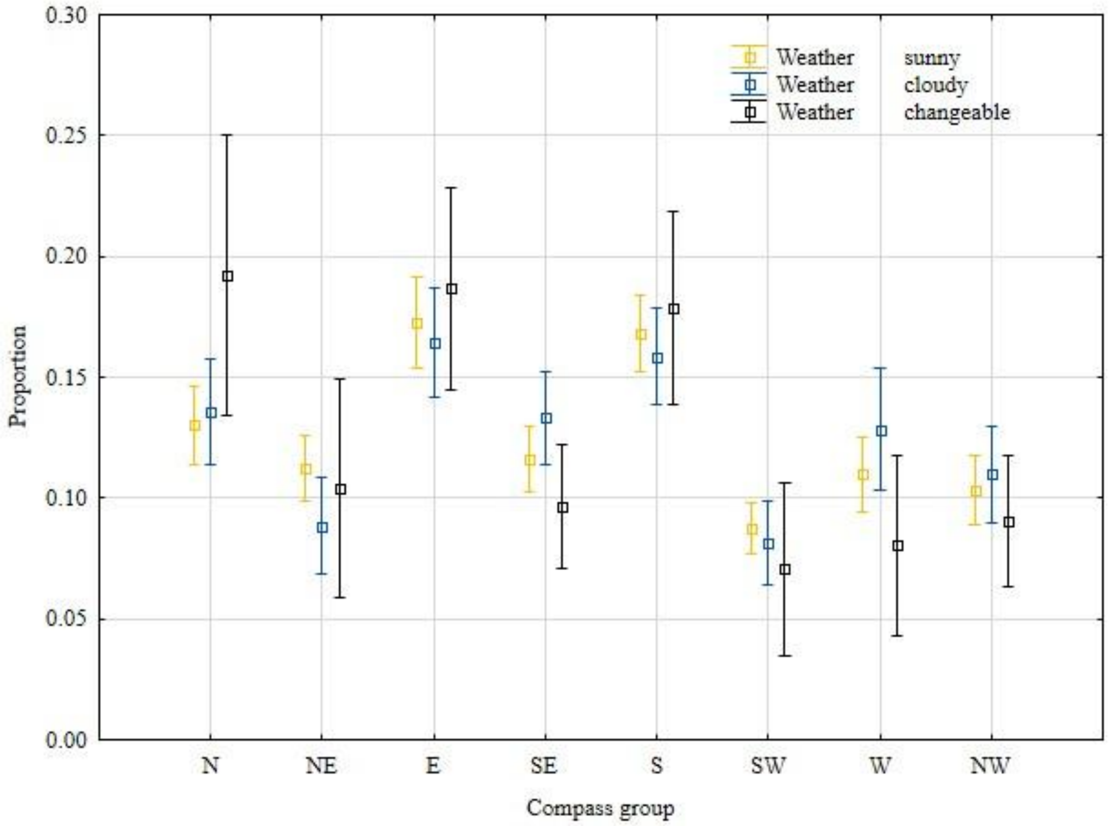


**Figure 9. Proportion of mean ± SE body axis orientation in cattle relative to cardinal directions and different weather conditions**

### 4.2.4. Impact of temperature on magnetic alignment

Proportions of magnetic alignment were not significantly influenced by dry bulb temperature in both species according to GLM (buffalo: $F_{7,1263}$ = 0.698, p = 0.674 and cattle ($F_{7,1232}$ = 1.751, p = 0.0958).

Although GLM pointed out that combination of these factors had significant impact on proportion of magnetic alignment of cattle ($F_{14,1233}$ = 3.982, P < 0.0001), further data analysis revealed only very weak association between air temperature, cloudy weather and N alignment. Meaning that the cattle preferred N alignment with increasing temperature (p = 0.0125, $R^2$ = 0.128) and no other compass groups very significantly influenced by combination of "weather" and "temperature" (P > 0.05). Impact of cloudy weather on northern alignment of cattle is shown in Figure 9.

Although proportions of magnetic alignments in buffalo relative to individual compass groups were significantly influenced by combination of "weather" and “temperature” according to GLM ($F_{14,1264}$ = 9.757, P < 0.0001), significant association among individual weather category, individual compass groups and dry bulb temperature in case of buffalo was not detected by multiple regression P > 0.05).

### 4.2.5. Impact of individuals and their sex on magnetic alignment

Statistical analysis revealed no significant association between the sex and magnetic orientation of neither cattle ($F_{7,2384}$ = 1.034, p = 0.405) and buffalo ($F_{7,2864}$ = 1.658, p = 0.115) . Magnetic alignment was not found to be significantly different among individual animals within herds of both species (P > 0.05).

### 4.2.6. Impact of age on magnetic alignment

GLM analysis revealed significant association between age of cattle and their magnetic alignment ($F_{7,2384} =$ 2.475, p = 0.0157), however further testing with regression analysis pointed out only very weak relationship between age and northerly alignment of cattle (p = 0.00258, $R^2$ = 0.03007). Orientation of cattle relative to the remaining compass groups was not influenced by age of animals (P > 0.05).

Compass orientation of buffalo was significantly influenced by combination of two independent variables and that is age and magnetic alignment , however the association with age of animals was very weak and only in case of NW alignment ($R^2$ = 0.0207, p = 0.00629) and SW alignment ($R^2$ = 0.0123, p = 0.0356).

# 5. Discussion

Our behavioural study provides the first comparison of body axis orientation between cattle and buffalo living in the same environmental conditions. The data allowed us to asses preferences of both species for directional body orientation and compare its major influences, especially the solar radiation and the magnetic field lines.

## 5.1. Solar orientation and thermoregulatory behaviour

Some remarkable findings and interspecies differences were revealed in this analysis compared to previously conducted studies devoted to various animal species referring to thermoregulatory behaviour (Maloney *et al.*, 2005; Hetem *et al.*, 2011).

We hypothesized that the frequency of orientation parallel to incident solar radiation increases with ambient temperature and perpendicular and oblique orientation is more frequent in both species at midday when the sun is at zenith.

It is evident in Figure 4 that daily proportions of orientations relative to solar radiation differed significantly between the species. Buffalo showed significant preference for PAR orientation whilst cattle oriented significantly more often PER relative to solar radiation.

Remarkable finding to emerge from this study is that buffalo significantly preferred PAR orientation in morning and in the afternoon compared with other directions (see Figure 5). That would mean that buffalo significantly preferred PAR orientation except for the midday period when they the sun was directly over their head and thus prevented them from employment of solar orientation (Lease et al.,2014).

Maloney et al. (2005a) reported that the black wildebeest can absorb by their dark skin 30% less radiant heat through PAR orientation when exposed to the sun for the whole day compared with PER orientation of their body axis relative to the sun (Maloney et al., 2005a). Also Lease et al. (2014) confirmed that Black wildebeest exhibits PAR solar orientation during the hottest parts of the day. This might be the reason for preference of PAR orientation in buffalo during the whole day as presented in Figure 5. Our previous study on two-humped camels (*Camelus bactrianus bactrianus*) revealed their preference for PAR orientation when exposed to higher ambient temperatures above 20 °C (Sedláček, 2014), which is in agreement with the orientation of buffalo found in present study.

PAR orientation was more likely to be employed by both species, as the dry bulb temperatures increased while proportion of PER orientation in cattle decreased with increasing ambient temperature. These findings agrees with part of our hypothesis, although the relationship among variables was very weak.

Altering of solar orientations in black wildebeest was indicated to be a well-developed adaptation to regulate the heat gained from the sun in severe environments (Maloney et al., 2005a). The ability of some ungulates to preferentially orient their body axis relative to the sun rays, temperature and season was confirmed by Hetem et al. (2011) after investigation of thermoregulatory behaviour of impala, eland and Blue wildebeest.

Our study provides the evidence that even cattle exhibited significant preference for PER orientation in morning compared to significant preference for PAR and PER orientation in the afternoon as visualized in Figure 6. The mean midday and afternoon temperatures were significantly lower during observations of cattle compared to buffalo observation (see Table 2). Based on the fact that all three ungulates in study of (Hetem et al., 2011) and camels in our previous study (Sedláček, 2014) oriented PER in cold temperature, we presume that cattle exhibited significant preference for PER orientation in morning to gain energy from the solar radiation (Berry al.,1984). Solar behaviour of black wildebeest was found to be a well-developed adaptation to regulate radiant heat in severe environments (Maloney et al., 2005a).

Pelt colour plays a significant role in relation to incident solar radiation. The black pelt conveys the highest proportion of incident solar radiation compared to the low proportion in white pelt (Hetem et al., 2009). Considering that the Donggala cattle were of light colour compared to the dark skin of buffalo, they might tend to employ PER orientation in morning to warm up after cold night (Berry al.,1984; Maloney et al., 2005a; Hetem et al., 2011) even in case when the environmental conditions would be exactly the same during both separate observations of cattle and buffalo which is not our case (see Table 2). Another argument supporting this assumption is the fact that the temperatures dropped lower at night at the period of observation of cattle as it was closer to monsoon season (December to April) resulting in higher proportion of rainy days that could have affected behaviour of cattle (see Figure 10).

Maloney et al. (2005a) state that preferences for solar orientation changed with different behaviours but it could be affected by thermal stress, wind velocity and other such factors.

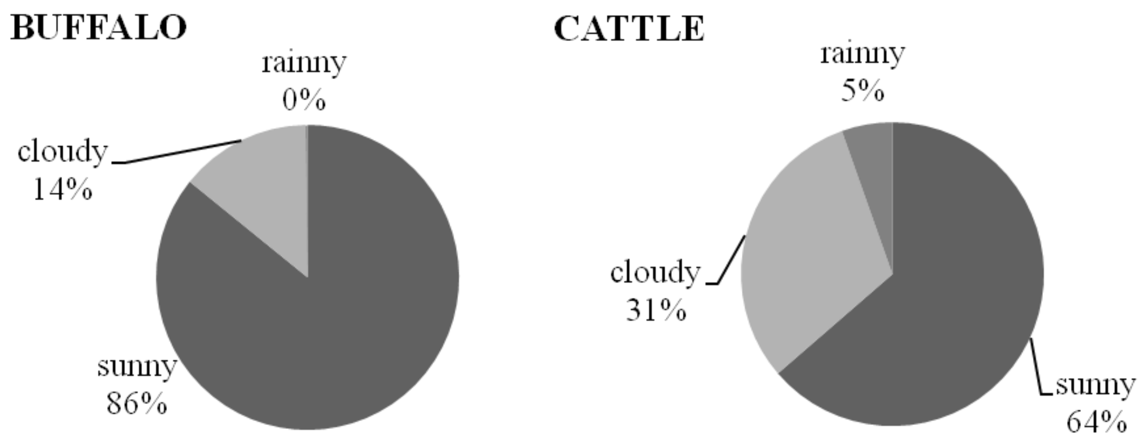


**Figure 10. Distribution of climatic conditions  within period of field research**

Already Walsberg and Wolf (1995) reported that sudden increase in wind speed by 3.5 m/s reduced the energy that squirrels gained from the sun by 10-13 %. Therefore, wind speed could have affected our results as it exceeded 3 m/s in 19 % of scan samples for cattle and 16 % of buffalo scan samples as presented in Figure 11. On the other hand Stelzner and Hausfater (1986)  reported that wind direction influenced trunk orientation in baboons, whereas body posture was primarily affected by ambient temperature and solar radiation Berry et al. (1984) indicated in their study that body position of wildebeest is influenced by sector winds the most in midday when the sun's altitude is high and the animals can not employ solar orientation.

Maloney et al. (2005a) did not detect effects of wind speed on orientation of the black wildebeest and states in their study that thermoregulatory behaviour of wildebeest would be affected only by very strong winds. The wind had impact on behaviour of wildebeest when the wind carried rain. Under these circumstances, the animals were facing away from the rain and the wind (Maloney et al., 2005a). However the analyses of our data revealed no significant influence of weather on body orientation in both species ($P > 0.05$) due to the fact that the observation was stopped if the raining persist for longer period than half an hour. Given that, the buffalo observation was carried out a month earlier when both ambient temperatures and proportion of sunny weather during the observation were higher as demonstrated in Figure 10, the first hypothesis $H_1$ was accepted but only in case of buffalo.

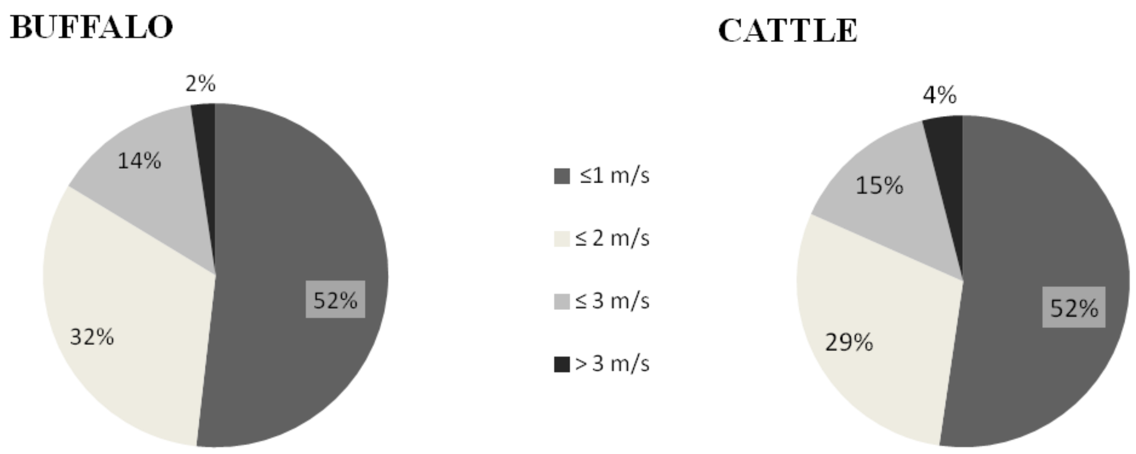


**Figure 11. Proportion of different wind velocities throughout the separate observations for buffalo and cattle**

## 5.2. Magnetic alignment

The data allowed us to examine the influence of different factors affecting orientation of buffalo and cattle relative to geomagnetic field of the Earth and to compare the outcomes with studies from the same scientific field.

Our hypothesis was that both water buffalo and cattle intend to orient their longitudinal body axis in N and S direction more frequently compared to other cardinal directions.

Significant finding to emerge from this study is that both species showed significant preference for magnetic alignments towards S and E compared to other compass groups . Although buffalo also oriented significantly more often towards W as well as towards E and S, the differences among preferences of buffalo for these three compass groups were not of any significance (see Fig 7 and 8).

A significant interspecies difference in magnetic alignments in terms of daily proportion is evident from Figures 7 and 8. Cattle exhibited N magnetic alignment significantly more often compared to buffalo, however the proportion of N alignment was not significant which in agreement with results of Hetem et al. (2011) that did not proved N orientation in none of all three ungulates observed. The only preference found in the study was the NW alignment in the blue wildebeest (Hetem et al., 2011)

On the other hand, buffalo aligned their body axis towards SW and W significantly more often than cattle did. Also our previous study pointed out that the two-humped camels oriented the most in the W direction (Sedláček, 2014) which was one of the compass groups significantly preferred by buffalo in the present study.

However, none of these findings were in agreement with those of Begall et al. (2008) who revealed that the roe deer, red deer and cattle intentionally oriented their body axis in the NS direction the most often. The same study brings into discussion another interesting point and that is the influence of behaviour or activity of animals on orientation of their body axis relative to the magnetic field. Begall et al. (2008) found significant proportion of NS alignment in resting roe deer and grazing red deer. Despite the fact that even Slaby et al. (2013) report in their study that cattle was grazing predominantly in the NS direction, we did not find cattle nor buffalo to orient significantly in NS direction in the present study. Neither our previous study proved significant N preference of lying camels (Sedláček, 2014). Begall et al. (2013) presented a preference of horses for NE/SW axial orientation which is also not in agreement with our findings.

Considering the fact that activities throughout the day affect orientation of animals, Bohorórquez-Alonso et al. (2011) described that body orientation of *Gallotia galloti* lizards most possibly depends on either solar radiation but secondly on social communication. This circumstance should be considered even in our case, as buffalo have been seen to orient in a direction so they could see the individual from the herd close to them. For mean daily distribution of all the different kinds of behaviour or activities observed in buffalo and cattle see Figure 12.

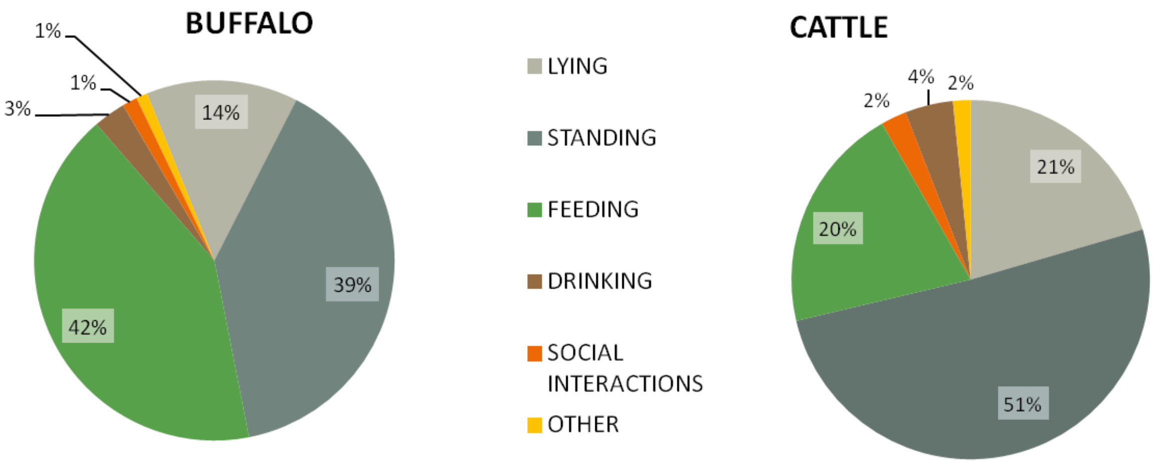


**Figure 12. Ethograms of both species in all weather conditions. Category OTHER contains: movement, excretion, suckling and hygiene**

Further factors that could affect the magnetic alignment of animals are presumably the ones mentioned in chapter above and those are wind, weather and solar radiation itself. Although our results did not revealed any significant influence of day period, day time, weather, temperature, sex or age of observed animals; the results might be different if the different daily activities of animals were considered in the analysis.

Although, neither of observed species oriented relative to the magnetic field in a way that was described in previous studies devoted to the topic, there was a significant preference for S alignment revealed in both species. Furthermore, cattle showed a high frequency of orientation towards N which might together with the preference for S alignment be in agreement with findings of Begall et al. (2008). However our hypothesis $H_2$ must be rejected and further investigation is required to test the influence of daily activities on magnetic alignment in ruminants.

# 6. Conclusions

Buffalo responded predictably and showed significant preference for parallel orientation ($P < 0.0001$) compared to significant perpendicular orientation of cattle relative to the sun ($P < 0.0001$). Buffalo significantly preferred parallel orientation in morning ($P < 0.0001$) as well as in the afternoon ($P < 0.05$) when the sun was not directly over their head to prevent them from employment of solar orientation. Parallel orientation was more likely to be employed by both species, as the dry bulb temperatures increased while proportion of perpendicular orientation in cattle decreased with increasing ambient temperature. These results were in agreement with our hypothesis. Cattle further exhibited significant preference for perpendicular orientation in morning ($P < 0.0001$), presumably to gain energy from the sun after cold night. Both species significantly preferred S alignment (cattle: $P < 0.05$; buffalo: $P < 0.0001$) and E alignment (cattle: $P < 0.01$; buffalo: $P < 0.001$). Cattle further showed a high frequency of orientation due N whereas buffalo were found facing W significantly more often ($P < 0.001$).

Further research in the field is required to test the influence of daily activities on solar orientation and magnetic alignment in ruminants. Whether the preference for S and E alignment in buffalo and cattle is only based on a magnetoreception or not, needs to be investigated in further studies. Such research should be ideally conducted under conditions which would prevent the animals from employment of solar behaviour and impact of wind.

# 7. References


Aarif O and Aggarwal A. 2015. Evaporative cooling in late gestation Murrah buffaloes potentiates immunity around transition period and overcomes reproductive disorders. Theriogenology 84 (7): 1197-1205.

Algers B and Hennichs K. 1985. The effect of exposure to 400 kV transmission lines on the fertility of cows. A retrospective cohort study. Preventive Veterinary Medicine 3(4):351-61.

Altmann J. 1974. Observational study of behaviour: sampling methods. Behavior, 49(3): 227-267.

Baker MA, Reeve N, Mohedano I, Conkey AA, Macdonald DW, Yamaguchi N. 2016. Caught basking in the winter sun: Preliminary data on winter thermoregulation in the Ethiopian hedgehog, *Paraechinus aethiopicus*, in Qatar. Journal of Arid Environments 125:52-5.

Balmori A. 2015. Anthropogenic radiofrequency electromagnetic fields as an emerging threat to wildlife orientation. Science of the Total Environment 518: 58-60.

Barker JS, Tan SG, Selvaraj OS, Mukherjee TK. 1997. Genetic variation within and relationships among populations of Asian water buffalo (Bubalus bubalis). Animal Genetics 28(1):1-3.

Begall S, Burda H, Červený J, Gerter O, Neef-Weisse J, Němec P. 2011. Further support for the alignment of cattle along magnetic field lines: reply to Hert et al. Journal of Comparative Physiology A 197(12):1127-33.

Begall S, Červený J, Neef J, Vojtěch O, Burda V. 2008. Magnetic alignment in grazing and resting cattle and deer. PNAS 105:13451–13455.

Begall S, Malkemper EP, Červený J, Němec P, Burda H. 2013. Magnetic alignment in mammals and other animals. Mammalian Biology 78:10-20.

Benhamou S, Bonadonna F, Jouventin P. 2003. Successful homing of magnet-carrying white-chinned petrels released in the open sea. Animal Behaviour 65(4):729-34.

Berry HH, Siegfried WR, Crowe TM. 1984. Orientation of wildebeest in relation to sun angle and wind direction. Madoqua 13(4):297-301.

Bohórquez-Alonso M, Font E, Molina-Borja M. 2011. Activity and body orientation of *Gallotia galloti* in different habitats and daily times. Amphibia-Reptalia 32: 93-103.

Boissere JR, Lihoreau F, Brunet M. 2005. The position of Hippopotamidae within Cetartiodactyla. Proceedings of the National Academy of Sciences of the United States of America 102 (5): 1537–1541.

Borghese A. 2011. Situation and perspectives of buffalo in the World, Europe and Macedonia. Macedonian Journal of Animal Science 1(2):281-96.

Brown-Brandl TM, Eigenberg RA, Nienaber JA, Hahn GL. 2005. Dynamic response indicators of heat stress in shaded and non-shaded feedlot cattle, Part 1: Analyses of indicators. Biosystems engineering 90(4):451-62.

Burchard JF, Nguyen DH, Block E. 1998. Effects of electric and magnetic fields on nocturnal melatonin concentrations in dairy cows. Journal of dairy science 81(3):722-7.

Burchard JF, Nguyen DH, Richard L, Block E. 1996. Biological effects of electric and magnetic fields on productivity of dairy cows. Journal of dairy science 79(9):1549-54.

Burda H, Begall S, Červený J, Neef J, Němec P. 2009. Extremely low-frequency electromagnetic fields disrupt magnetic alignment of ruminants. Proceedings of the National Academy of Sciences 106 (14): 5708-13.

Burton, J, Wheeler, P, Mustari, A. 2016. *Bubalus depressicornis*. The IUCN Red List of Threatened Species 2016: e.T3126A46364222. Available at http://dx.doi.org/10.2305/IUCN.UK.2016-2.RLTS.T3126A46364222.en /: Accessed 2017-04-05.

Červený J, Begall S, Koubek P, Nováková P, Burda H. 2011. Directional preference may enhance hunting accuracy in foraging foxes. Biology letters. 7: 355-357.

Chaiyabutr N, Komolvanich S, Sawangkoon S, Chanpongsang S. 1997. Alterations in potassium homeostasis during heat stress in swamp buffaloes (*Bubalus bubalis*). Journal of Thermal Biology 22(3): 207-212.

Clapperton JL, Joyce JP, Blaxter KL.1965. Estimates of the contribution of solar radiation to the thermal exchanges of sheep at a latitude of 55° north. The Journal of Agricultural Science 64:37-49.

Coe M. 2004. Orientation, movement and thermoregulation in the giant tortoises (Testudo (Geochelone) gigantea) of Aldabra Atoll, Seychelles. Transactions of the Royal Society of South Africa 59(2):73-7.

Das SK, Upadhyay RC, Madan ML.1999. Heat stress in Murrah buffalo calves. Livestock Production Science 61: 71–78.

de la Cruz-Cruz LA, Guerrero-Legarreta I, Ramirez-Necoechea R, Roldan-Santiago P , Mora-Medina P , Hernandez-Gonzalez R , Mota-Rojas D. 2014. The behaviour and productivity of water buffalo in different breeding systems: a review. Veterinarni Medicina 59 (4): 181-193.

De Rosa G, Grasso F, Pacelli C, Napolitano F, Winckler C. 2009. The welfare of dairy buffalo. Italian Journal of Animal Science 8: 103-116.

Deutschlander ME, Freake MJ, Borland SC, Phillips JB, Madden RC, Anderson LE, Wilson BW. 2003. Learned magnetic compass orientation by the Siberian hamster, *Phodopus sungorus*. Animal Behaviour 65(4):779-86.

Dommer DH, Gazzolo PJ, Painter MS, Phillips JB. 2008. Magnetic compass orientation by larval *Drosophila melanogaster*. Journal of Insect Physiology 54(4):719-26.

FAO. 2005. Buffalo production and research. Borghese A  and Mazzi M editors. Buffalo population and strategies in the world. Rome: FAO regional office for Europe, p 1.

Fischer JH, Freake MJ, Borland SC, Phillips JB. 2001. Evidence for the use of magnetic map information by an amphibian. Animal behaviour 62(1):1-10.

Freake MJ, Muheim R, Phillips JB. 2006. Magnetic maps in animals: a theory comes of age?. The Quarterly Review of Biology 81(4):327-47.

Freire R, Munro UH, Rogers LJ, Wiltschko R, Wiltschko W. 2005. Chickens orient using a magnetic compass. Current Biology 15(16):R620.

Frilot C, Carrubba S, Marino AA. 2014. Sensory transduction of weak electromagnetic fields: Role of glutamate neurotransmission mediated by NMDA receptors. Neuroscience 258:184-91.

Gould JL. 2008. Animal navigation: the evolution of magnetic orientation. Current Biology 18(11):R482-4.

Gu Z, Yang S, Leng J, Xu S, Tang S, Liu C, Gao Y, Mao H. 2016. Impacts of shade on physiological and behavioural pattern of Dehong buffalo calves under high temperature. Applied Animal Behaviour Science 177: 1–5.

Hansen PJ. 2004. Physiological and cellular adaptations of zebu cattle to thermal stress. Animal Reproduction Science 82–83: 349–360.

Hart V, Kušta T, Němec P, Bláhová V, Ježek M, Nováková P, Begall S, Červený J, Hanzal V, Malkemper EP, Štípek K. 2012. Magnetic alignment in carps: evidence from the Czech Christmas fish market. PLoS One 7(12):e51100.

Hart V, Malkemper EP, Kušta T, Begall S, Nováková P, Hanzal V, Pleskač L, Ježek M, Policht R, Husinec V, Červený J. 2013. Directional compass preference for landing in water birds. Frontiers in zoology 10(1):38

Hedges, S., Sagar Baral, H., Timmins, R.J. & Duckworth, J.W. 2008. *Bubalus arnee*. The IUCN Red List of Threatened Species 2008: e.T3129A9615891. Available at http://dx.doi.org/10.2305/IUCN.UK.2008.RLTS.T3129A9615891.en /: Accessed 2017-05-04.

Hetem RS, de Witt BA, Fick LA,Fuller A. 2009. Body temperature, thermoregulatory behaviour and pelt characteristics of three colour morphs of springbok *(Antidorcas marsupialis)*. Comperative Biochemistry and Physiology 152A: 379-388.

Hetem RS, Maloney SK, Fuller A, Mitchell D. 2016. Heterothermy in large mammals: inevitable or implemented?. Biological Reviews 91: 187–205. doi: 10.1111/brv.12166.

Hetem RS, Strauss WM, Fick LG, Maloney SK, Meyer LC, Shobrak M, Fuller A, Mitchell D. 2012a. Does size matter? Comparison of body temperature and activity of free-living Arabian oryx (*Oryx leucoryx*) and the smaller Arabian sand gazelle (*Gazella subgutturosa marica*) in the Saudi desert. Journal of Comparative Physiology B 182(3):437-449.

Hetem RS, Strauss WM, Fick LG, Maloney SK, Meyer LC, Shobrak M, Fuller A, Mitchell D. 2012b. Activity re-assignment and microclimate selection of free-living Arabian oryx: responses that could minimise the effects of climate change on homeostasis?. Zoology 115(6):411-416.

Hetem RS, Strauss WM, Heusinkveld BG, Steven de Bie, Prins HHT, Wieren SE. 2011. Energy advantages of orientation to solar radiation in three African ruminants. Journal of Thermal Biology 36 (7): 452-460.

Hsu CY, Ko FY, Li CW, Fann K, Lue JT. 2007. Magnetoreception system in honeybees (Apis mellifera). PloS one.2(4):e395. doi:10.1371/journal.pone.0000395.

Ismail K, Kamal K, Plath M, Wronski T. 2011. Effects of an exceptional drought on daily activity patterns, reproductive behaviour, and reproductive success of reintroduced Arabian oryx (*Oryx leucoryx*). Journal of arid environments 75(2):125-31.

ITIS. 2017. *Bubalus bubalis* Linnaeus, 1758. Available at www.itis.gov/: Accessed 2017-04-05.

Johnsen S, Lohmann KJ. 2005. The physics and neurobiology of magnetoreception. Nature Reviews Neuroscience 6(9):703-12.

Kadzere CT, Murphy MR, Silanikove N, Maltz E. 2002. Heat stress in lactating dairy cows: a review. Livestock Production Science 77 (1): 59–91.

Keren EN and Olson BE. 2006. Thermal balance of cattle grazing winter range: Model development. Journal of Thermal Biology. Jul 31;31(5):371-7.

Keren, EN and Olson BE. 2007. Applying thermal imaging software to cattle grazing winter range. Journal of thermal biology 32(4): 204-211.

Khongdee T, Sripoon S, Vajrabukka C. 2011. The effects of high temperature and wallow on physiological responses of swamp buffaloes (Bubalus bubalis) during winter season in Thailand. Journal of Thermal Biology 36: 417-421.

Kimchi, T. and Terkel, J. 2001. Magnetic compass orientation in the blind mole rat *Spalax ehrenbergi*. Journal of Experimental Biology 204: 751–758.

Kirschvink JL, Gould JL. 1981. Biogenic magnetite as a basis for magnetic field detection in animals. Biosystems 13(3):181-201.

Kirschvink JL, Walker MM, Diebel CE. 2001. Magnetite-based magnetoreception. Current opinion in neurobiology 11(4):462-7.

Kirschvink JL, Winklhofer M, Walker MM. 2010. Biophysics of magnetic orientation: strengthening the interface between theory and experimental design. Journal of the Royal Society Interface.

Kirschvink JL. 1989. Magnetite biomineralization and geomagnetic sensitivity in higher animals: an update and recommendations for future study. Bioelectromagnetics 10(3):239-59.

Lactin DJ, Johnson DL. 1998. Convective heat loss and change in body temperature of grasshopper and locust nymphs: relative importance of wind speed, insect size and insect orientation. Journal of Thermal Biology 23(1):5-13.

Lease HM, Murray IW, Fuller A, Hetem RS. 2014. Black wildebeest seek shade less and use solar orientation behavior more than do blue wildebeest. Journal of Thermal Biology 45:150-6. doi: 10.1016/j.jtherbio.2014.08.008. Epub 2014 Aug 28.

Lohmann K, Pentcheff N, Nevitt G, Stetten G, Zimmer-Faust R, Jarrard H, Boles LC. 1995. Magnetic orientation of spiny lobsters in the ocean: experiments with undersea coil systems. Journal of Experimental Biology 198(10):2041-8.

Lohmann KJ, Luschi P, Hays GC. 2008. Goal navigation and island-finding in sea turtles. Journal of Experimental Marine Biology and Ecology 356(1):83-95.

Maloney SK and Dawson TJ. 1995. The heat load from solar radiation on a large, diurnally active bird, the emu (*Dromaius novaehollandiae*). Journal of Thermal Biology 20 (5): 381-387.

Maloney SK, Moss G, Cartmell T, Mitchell D. 2005b. Alteration in diel activity patterns as a thermoregulatory strategy in black wildebeest (*Connochaetes gnou*). Journal of Comparative Physiology, Part A, 191(11):1055-1064.

Maloney SK, Moss G, Mitchell D. 2005a. Orientation to solar radiation in black wildebeest (Connochaetes gnou). Journal of Comparative Physiology, Part A, 191:1065-1077.

Marai IFM and Haeeb AAM. 2010. Buffalo's biological functions as affected by heat stress - A review. Livestock Science 127 (2-3): 89-109.

Marai IFM, Daader AH, Soliman AM and El-Menshawy SMS. 2009. Non-genetic factors affecting growth and reproduction traits of buffaloes under dry management housing (in sub-tropical environment) in Egypt. Livestock Research for Rural Development 21(30).

Marai IFM, El-Darawany AA, Fadiel A, Abdel-Hafez MAM. 2007. Physiological traits as affected by heat stress in sheep-A review. Small Ruminant Research 71(1–3): 1–12.

Marai IFM, Haeeb AAM, Daader AH, Yousef HM. 1995. Effects of Egyptian subtropical summer conditions and the heat-stress alleviation technique of water spray and a diaphoretic on the growth and physiological functions of Friesian calves. Journal of Arid Environments 30:219-225.

Martin J, López P, Carrascal LM, Salvador A. 1995. Adjustment of basking postures in the high-altitude Iberian rock lizard (*Lacerta monticola*). Canadian journal of zoology 73(6):1065-8.

Mirkena T, Duguma G, Haile A, Tibbo M, Okeyo AM, Wurzinger M, Sölkner J. 2010. Genetics of adaptation in domestic farm animals:A review. Livestock Science 132(1-3):1-12.

Mitchell D, Maloney Shane K, Jessen C, Laburn Helen P, Kamerman Peter R, Mitchell G, Fuller A. 2002. Adaptive heterothermy and selective brain cooling in arid-zone mammals. Comparative Biochemistry and Physiology, Part B, 13:571–585.

Mora CV, Davison M, Wild JM, Walker MM. 2004. Magnetoreception and its trigeminal mediation in the homing pigeon. Nature 432(7016):508-11.

Moran DS, Pandolf KB, Vitalis A, Heled Y, Parker R, Gonzalez RR. 2004. The role of solar and UV radiation in environmental stress assessment. Journal of Thermal Biology 29(7):529-533.

Moritz RE, Burda H, Begall S, Němec P. 2007. Magnetic compass: a useful tool underground. Subterranean Rodents. Springer Berlin Heidelberg:161-174.

Muheim R, Bäckman J, Åkesson S. 2002. Magnetic compass orientation in European robins is dependent on both wavelength and intensity of light. Journal of Experimental Biology 205(24):3845-56.

Němec P, Altmann J, Marhold S, Burda H, Oelschläger HH. 2001. Neuroanatomy of magnetoreception: the superior colliculus involved in magnetic orientation in a mammal. Science 294 (5541):366-8.

Nguyen DH, Richard L, Burchard JF. 2005. Exposure chamber for determining the biological effects of electric and magnetic fields on dairy cows. Bioelectromagnetics 26(2):138-44.

Papi F, Luschi P, Akesson S, Capogrossi S, Hays GC. 2000. Open-sea migration of magnetically disturbed sea turtles. Journal of Experimental Biology 203(22):3435-43.

Pereira AMF, Titto EL, Infante P, Titto CG, Geraldo AM, Alves A, Leme TM, Flávio Baccari F Jr., Almeidae JA. 2014. Evaporative heat loss in *Bos taurus*: Do different cattle breeds cope with heat stress in the same way? Journal of Thermal Biology 45: 87–95.

Phillips JB, Borland SC. 1992. Behavioural evidence for use of a light-dependent magnetoreception mechanism by a vertebrate. Nature 359(6391):142-4.

Phillips JB, Youmans PW, Muheim R, Sloan KA, Landler L, Painter MS, Anderson CR. 2013. Rapid learning of magnetic compass direction by C57BL/6 mice in a 4-armed 'plus' water maze. PloS one 8(8):e73112.

Phillips JB. Magnetic navigation. 1996. Journal of Theoretical Biology 180(4):309-19.

Ritz T, Adem S, Schulten K. 2000. A model for photoreceptor-based magnetoreception in birds. Biophysical journal 78(2):707-18.

Ritz T, Wiltschko R, Hore PJ, Rodgers CT, Stapput K, Thalau P, Timmel CR, Wiltschko W. 2009. Magnetic compass of birds is based on a molecule with optimal directional sensitivity. Biophysical journal 96(8):3451-7.

Rodda GH. 1984. The orientation and navigation of juvenile alligators: evidence of magnetic sensitivity. Journal of Comparative Physiology A 154(5):649-58.

Rodgers CT, Hore PJ. 2009. Chemical magnetoreception in birds: the radical pair mechanism. Proceedings of the National Academy of Sciences 106(2):353-60.

Rodriguez M, Petitclerc D, Burchard JF, Nguyen DH, Block E, Downey BR. 2003. Responses of the estrous cycle in dairy cows exposed to electric and magnetic fields (60 Hz) during 8-h photoperiods. Animal reproduction science 77(1):11-20.

Rogers LJ, Munro U, Freire R, Wiltschko R, Wiltschko W. 2008. Lateralized response of chicks to magnetic cues. Behavioural brain research 186(1):66-71.

Sedláček R. 2014. The Adaptation of Camels to Extreme Environments [BSc.]. Prague: Czech University of Life Sciences Prague, 45p.

Seebacher F, Grigg GC, Beard LA. 1999. Crocodiles as dinosaurs: behavioural thermoregulation in very large ectotherms leads to high and stable body temperatures. Journal of Experimental Biology 202(1):77-86.

Silanikove Nissim. 2000. Effects of heat stress on the welfare of extensively managed domestic ruminants. Livestock Production Science 67:1–18.

Silva JAR, Santos NFA, Araújo AA, Lourenço Júnior JB, Garcia AR, Viana RB, Nahúm BS. 2010. Effect of the climatic conditions of the Brazilian Eastern Amazon in the tolerance to the heat in buffaloes (*Bubalus bubalis*) created on the shading and sun. Revista Veterinaria 21(1): 777-780.

Slaby P, Tomanová K, Vácha M. 2013. Cattle on pastures do align along the North–South axis, but the alignment depends on herd density. Journal of Comparative Physiology A,199 (8): 695-701.

Stapput K, Thalau P, Wiltschko R, Wiltschko W. 2008. Orientation of birds in total darkness. Current Biology 18(8):602-6.

Stelzner JK, Hausfater G. 1986. Posture, microclimate, and thermoregulation in yellow baboons. Primates 27(4):449-63.

Stevenson RD. 1985. The relative importance of behavioral and physiological adjustments controlling body temperature in terrestrial ectotherms. American Naturalist: 362-386.

Strauss WM, Hetem RS, Mitchell D, Maloney SK, Meyer LCR, Fuller A. 2015. Selective Brain Cooling Reduces Water Turnover in DehydratedSheep. PLOS ONE 10(2): e0115514. doi:10.1371/journal.pone.0115514.

Thalau P, Ritz T, Burda H, Wegner RE, Wiltschko R. 2006. The magnetic compass mechanisms of birds and rodents are based on different physical principles. Journal of the royal Society Interface 3(9):583-7.

Thompson ML, Mzilikazi N, Bennett NC, McKechnie AE. 2015. Solar radiation during rewarming from torpor in elephant shrews: supplementation or substitution of endogenous heat production?. PloS one 10(4): e0120442. doi:10.1371/journal.pone.0120442

Vácha M, Půžová T, Drštková D. 2008. Ablation of antennae does not disrupt magnetoreceptive behavioural reaction of the American cockroach to periodically rotated geomagnetic field. Neuroscience letters 435(2):103-7.

Vislobokova IA. 2013. On the origin of Cetartiodactyla: comparison of data on evolutionary morphology and molecular biology. Paleontological Journal 47(3):321-34.

Walcott C, Gould JL, Kirschvink JL. 1979. Pigeons have magnets. Science 205(4410):1027-9.

Walsberg GE, Tracy RL, Hoffman TCM. 1997. Do metabolic responses to solar radiation scale directly with intensity of irradiance? The Journal of Experimental Biology 200: 2015-2021.

Walsberg GE, Wolf BO. 1995. Effect of solar radiation and wind speed on metabolic heat production by two mammals with contrasting coat colours. The Journal of Experimental Biology 198: 1499-1507.

Walsberg GE. 1983. Coat color and solar heat gain in animals. BioScience 33(2):88-91.

Walsberg GE. Consequences of skin color and fur properties for solar heat gain and ultraviolet irradiance in two mammals. 1988b. Journal of Comparative Physiology B 158(2):213-221.

Walsberg GE. The significance of fur structure for solar heat gain in the rock squirrel, *Spermophilus variegatus*. 1988a. Journal of experimental biology138(1):243-257.

Wiltschko R, Ritz T, Stapput K, Thalau P, Wiltschko W. 2005. Two different types of light-dependent responses to magnetic fields in birds. Current Biology 15(16):1518-23.

Wiltschko R, Wiltschko W. 2003. Avian navigation: from historical to modern concepts. Animal behaviour 65(2):257-72.

Wiltschko W, Wiltschko R, Ritz T. 2011. The mechanism of the avian magnetic compass. Procedia Chemistry 3(1):276-84.

Wiltschko W, Wiltschko R. 1972. Magnetic compass of European robins. Science 176 (4030): 62-4.

Wiltschko W, Wiltschko R. 2005. Magnetic orientation and magnetoreception in birds and other animals. Journal of Comparative Physiology A, 191(8):675-93.

Wolfenson D, Roth Z, Meidan R. 2000. Impaired reproduction in heat-stressed cattle: basic and applied aspects. Animal Reproduction Science 60–61: 535–547.

Yang DY, Liu L, Chen X, Speller CF. 2008. Wild or domesticated: DNA analysis of ancient water buffalo remains from north China. Journal of Archaeological Science 35(10):2778-85.