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USING BEHAVIOURAL OBSERVATIONS TO ASSESS THE WELFARE OF RED-NECKED POND TURTLES (*MAUREMYS NIGRICANS*) KEPT IN A ZOO

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Abstract

The aim of this study was to use behavioural observations to assess the welfare of the red--necked pond turtles (*Mauremys nigricans*) kept in a zoo. In 2000, red-necked pond turtles were put on the critically endangered list. Today, the species appears to be extinct in the wild. The welfare of captive populations of the species will have an important impact on their survival. Due to unusual aspects of reptile biology and a lack of monitoring standards, the main criteria available for welfare assessment for these animals may be behavioural. Based on the results of this study, it can be inferred that the welfare of the observed turtles has been moderately well-preserved; however, the artificial conditions created by humans are not able to fully satisfy the behavioural needs of the studied animals.

Introduction

Welfare studies are usually restricted to "higher" vertebrates (mammals, birds) and are primarily referred to in the context of farm animals (pigs, cattle, poultry) and companion animals (dogs, cats, rodents) (KALETA 2013). Over the years, there has been a lot of concern regarding the welfare of cattle or pigs, but "lower" vertebrates are increasingly popular today (WABNITZ et al. 2003). Reptiles, amphibians and fishes have become very common among zoological gardens, private breeders and industrial farms, in which they are raised for hides and meat (KALETA 2013). Assess-

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ment of the welfare of fishes, amphibians and reptiles can be challenging compared with that of mammals and birds, due to relatively limited availability of guidance criteria, although some multi-taxa scientific sources are now available (WARWICK et al. 2018). The unique biology of reptiles and the existence of specific adaptive features make the task even harder. However, it is certain that both reptiles and other "lower" vertebrates are capable of experiencing stress, which manifests as unusual, altered behaviour (MOBERG 1985). Contrary to popular belief, reptiles exhibit a number of abnormal behaviours that are the result of excessive stress and are either atypical in nature and/or occur out of the context under stress conditions (WARWICK et al. 2013). Therefore, behavioural criteria should be the primary foci of research assessing the welfare of these animals. Ensuring adequate animal welfare and reducing the stress that animals experience should be the aim of every private person or institution that maintains living animals (BAYS et al. 2006).

Mauremys nigricans is a medium-sized turtle from the Geoemydidae family. These turtles are characterized by daily activity and strong sexual dimorphism. Females are much larger than males and can grow to 30 cm in length, while males do not exceed 20cm and have longer tails. The colour of the carapace of these turtles can vary from chestnut to black, and the tympanum region is decorated with several irregular, narrow, creamy or yellow strips stretching across the neck. The pharynx is covered in dark brown to black stripes interrupted by red streaks in males and cream streaks in females. The exposed parts of the forearms and hind legs may be characterized by creamy pigment, although in males these spots may be red (ANDERS and IVERSON 2012). The name of red-necked pond turtle was first used by IVERSON (1985) and is still valid today. Breeders also call the species the Kwantung River turtle.

The general range of this species is defined by the Pearl River in Guangdong and Guangxi Provinces in the People's Republic of China (IVERSON 1992). Current knowledge about the natural distribution of the red-necked pond turtle is derived from the reports of Mell (1922, 1929), who claimed that it was an upland species inhabiting mountain streams. The water temperature of the described streams was 16–17°C in August. MELL (1922) also reported a decline in activity of the species from November to December and a rebound in activity in the spring. The above information were obtained through observations of captive specimens that were kept in external enclosures at latitudes consistent with the natural range of these turtles (ANDERS and IVERSON 2012). The natural diet of *M. nigricans* is not known, but the construction of the head and jaw suggests that it consists mainly of molluscs and crustaceans. Reproductive

behaviour has been observed only in captivity, but MELL (1929) reports that females of this species lay two eggs per year. The mating season begins in spring, when the males become intensely red in colour. According to ARTNER (2009) the female accepts the male by retracting her forearm and sliding the hind limbs so that the back of the shell and tail are raised. Some copulations can take aggressive character, evidenced by the male biting the neck of the female.

Mauremys nigricans is a species that is desirable by collectors around the world, and this combined with its very limited range has led to a significant decline in the species's population. Poachers trapping these turtles for the needs of Chinese folk medicine have contributed to the heavy decline in the number of *M. nigricans*. GAILLARD et al. (2017) reported that 500 grams of the red-necked pond turtle in 2015 reached a price of 80 000 dollars for males and 50 000–60 000 dollars for females. Another factor responsible for the population decline is the degradation of the turtle's natural habitat, in particular via deforestation, construction of hydroelectric power plants and stream liming. In 2007–2009, attempts to find *M. nigricans* in the provinces of Guangdong and Guangxi failed, suggesting that the species may have disappeared entirely from its natural habitat; no wild populations of these turtles are known today (ANDERS and IVERSON 2012).

Mauremys nigricans was listed on the Red List of Endangered Species in 2000 (IUCN, 2017). In 2011, this turtle was included on the IUCN's list of the 40 most endangered freshwater turtles (*Turtles in trouble...* 2011). Field trials and studies conducted on captive specimens may be useful for *M. nigricans* protection. It is also necessary to educate human societies, especially local ones, about *M. nigricans* conservation. The welfare of the individual turtles of this species that are kept in captivity must also be safeguarded. General welfare principles state that captive animals should be provided with five freedoms: freedom from hunger and thirst, freedom from discomfort, freedom from pain, injuries and diseases, freedom from fear and stress, and the ability to express normal behaviour (FAWC 1992). Unfortunately, in the case of reptiles it is believed that it can be very difficult to evaluate and ensure the conditions conducive to these five freedoms, particularly with regard to behaviour (KALETA 2013, WARWICK et al. 2013). The physiology and morphology, life history, and diversity of living environments of these animals makes it extremely difficult to identify consistent behavioural patterns for them (KALETA 2013). In addition, captive reptiles are usually restricted to small and often poorly-constructed enclosures, leading to physical and behavioural problems. Examples of behavioural disorders are given in Table 1.

Table 1

Abnormal behaviours of turtles kept in captivity (Morgan and Tromborg 2007, Warwick et al. 2013, Kaleta 2013, Arena et al. 2014, Silvestre 2014)

Behaviour	Possible cause
ITB (interaction with transparent boundaries)	an inability to detect invisible barriers, stemming from adaptive limitations of these animals
Hyperactivity	excessive density of animals and poorly designed premises, stress, fear, lack of shelter, hunger
Hypoactivity	cold temperatures in the enclosures where animals are kept, infections, injuries, harassment by other occupants
Hyper-alertness	fear
Head-hiding	fear, excessive lighting
Co-occupant aggression	hunger, overcrowding of animals, lack of ability to escape in poorly designed enclosures
Human-directed aggression	fear
Freezing	fear
Grating of jaw	pain, fear, poorly designed enclosures, nutrient deficiencies
Prolonged retractions of head, limbs or tail	fear, pain, illness
Cloacal evacuations when handled	fear
Pseudovocalisation	fear, pain, illnesses
Atypical location of animals	diseases, injuries, discomfort, co-occupant aggression, hyperthermia, hypothermia
Accelerated body movements	stress, fear
Boundary exploration	excessive animal density, lack of shelter
Accumulation of individuals at the surface	excessive animal density, poorly designed enclosures
Anxiety behaviour	stress, fear, co-occupant aggression, pain
Cannibalism	excessive animal density, hunger, poorly designed enclosures

The aim of the present study was to assess the welfare of captive *M. nigricans* specimens based on observations of their behaviour. Observations consisted of documenting the turtles' time budget and defining individual behaviours. Observational data were used to establish hierarchy in the observed animal group. Observations also focused on the occurrence of abnormal behaviour, which may be caused by stress or disturbance, and additionally aimed to determine the nutritional preferences of the observed animals.

Materials and Methods

This study was conducted at the Wroclaw Zoo (Poland) in September and October of 2017. The focal group of turtles consisted of five individuals aged four years, whose gender has not been determined. The turtles averaged 10 cm in length. The animals were kept in an aquaterrarium with separate water and land portions; the "land" area consisted of numerous stones and roots placed in the water. The average water temperature in the aquaterrarium was 21°C and the average land temperature was 24°C. In the autumn and winter, the entire terrarium building must be heated, with the result that the temperature in the individual animal enclosures also increases. The turtle aquaterrarium was designed to provide numerous hiding places for the animals. The aquaterrarium had glass walls and overall dimensions of 120/50/50 cm. The rear wall was covered with mortar in imitation of rocks. A photograph of a turtle aquaterrarium is included in the supplementary materials.

The turtles were fed on Tuesdays, Thursdays and Fridays. Their diet consisted of dead suckling mice, fish, shrimp, earthworms and plants (chicory, plantain, dandelion). Food was dropped into the water. On Tuesdays the turtles were fed in the afternoon but on Thursdays and Fridays in the morning.

Observations were carried out during daylight hours for three hours per day over 20 days. Thus, the total time devoted to turtle observation was 60 hours. Observations consisted of documenting the turtles' time budget and identifying individual behaviours. Data included the quantity of time spent in water and on land, feeding, in quiescent rest (eyes closed, relaxed limbs), basking, and in aggressive behaviour. Chasing other specimens, biting, and fighting for food were classified as aggressive behaviours. Observations also focused on the occurrence of behaviours commonly regarded as abnormal, such as: interaction with transparent boundaries, hyperactivity, hypoactivity, hyper-alertness, freezing, grating of jaw, prolonged retraction of head and pseudovocalisation. Observations additionally aimed to determine the dietary preferences of the observed animals.

Behavioural observations assessing the time budget devoted to individual behaviours were analysed using *Statistica* ver. 13.1. A nonparametric Spearman's rank order correlation analysis was performed.

Evidence of abnormal behavioural data was examined to determine in what situations and with what intensity the behaviour manifested.

Animal nutritional preferences were determined on the basis of percentage of total food intake. The turtles were fed on a different type of food on each feeding day, and it was determined which type of food was taken most frequently.

Results and Discussion

The turtle's diet was varied, and the contribution of various elements to the diet is presented in Table 2.

Table 2

Percentage of individual components out of the total diet of red-necked pond turtles (*Mauremys nigricans*)

Component	Percentage [%]
Suckling mice	40
Fish	20
Shrimp	15
Earthworms	10
Plants (chicory, plantain, dandelion in equal proportions)	15

The most commonly consumed food was suckling mice, shrimp and earthworms, which were always completely eaten. When fish was offered, 75% of it was eaten. Greatest feeding interest occurred in the early stages of each feeding; after a few minutes, the turtles stopped hunting. The turtles were reluctant to eat the plants that were offered, consuming only 10–15% of the total plant feed available.

Table 3 presents the correlation coefficient and time budget devoted to each individual behaviours.

Table 3

Time budget and correlation coefficients for individual behaviours

Specification	Mean ± SD	Time spent in water	Time spent on land	Time spent in quiescent rest	Time spent on basking	Time spent on feeding	Time spent in aggressive behaviour
Time spent in water	138 min 15 s ± 7.86	1	-1*	0.43	-0.97*	0.07	0.13
Time spent on land	41 min 45 s ± 7.86	-1*	1	-0.43	0.97*	-0.07	-0.13
Time spent in quiescent rest	58 min 50 s ± 3.85	0.43	-0.43	1	-0.45	0.41	0.02
Time spent on basking	35 min 30 s ± 6.68	-0.97*	0.97*	-0.45	1	-0.17	-0.15
Time spent on feeding	10 min 40 s ± 1.61	0.07	-0.07	0.41	-0.17	1	0.7*
Time spent in aggressive behaviour	1 min 31 s ± 0.45	0.13	-0.13	0.02	-0.15	0.7*	1

*significant differences at $P < 0.05$

The observed turtles spent 76.75% of their time in the water and 23.25% of their time on land. Time spent feeding was positively correlated with the time spent in water. This was to be expected, due to the fact that many species of turtles are unable to swallow food while on land. Time of day did not affect food intake; turtles consumed food willingly in the morning and afternoon. The test animals always fed together, since individual animals were not separated during feeding time.

Rest was quantified as the combination of quiescent rest and basking. The animals rested both as a group and individually. Time spent in quiescent rest was positively correlated with the time spent in water and negatively with time spent on land; this may be due to the fact that turtles resting this way also tried to hide from the light, which was impossible on land. Turtles often spent quiescent rest in hiding places that also occur in the wild. Time spent basking was positively correlated with time spent on land and negatively with time spent in water, since the temperature on land was higher than the water temperature. Quiescent rest was usually observed in the mornings, and after some time the turtles went ashore to bask and then returned underwater again. Additionally, the animals basked after feeding. Time devoted to aggressive behaviours was positively correlated with time spent in water and time spent feeding. Turtles exhibited aggression against each other only at feeding times. This aggression was demonstrated to other individuals as a consequence of turtles competing for food. No aggressive behaviour was observed in other situations. Turtles investigated the environment regardless of the time of day, and in the vast majority of cases they did so individually. Investigation of the environment usually consisted of calmly swimming to the gaps between the rocks and examining them. Sometimes turtles tried to swim against the glass or attempted to climb walls. These were symptoms of interaction with transparent boundaries. No hierarchy structure was observed within the group. None of the turtles initiated group activities such as foraging or basking together.

SCHOFIELD et al. (2006) created a simplified ethogram for loggerhead sea turtles (*Caretta caretta*). They divided the investigated behaviours of the turtles into individual and group behaviours. According to that study, among individual behaviours one can distinguish: locomotive behaviour (surface rest, resting at the bottom, swimming vertically, horizontally and near the surface, patrolling), digestive behaviour (searching and eating) and comfortable behaviour (self-care) (SCHOFIELD et al. 2006). Group behaviours, on the other hand, include agonistic behaviours (male conflict, conflict between females) and reproductive behaviours (aesthesia, copulation, copulation with the assistance of another male) (SCHOFIELD et al. 2006).

The observations of the Schofield et al. (2006) study differed slightly from those carried out in the course of this study. The turtles observed in the Wroclaw Zoo exhibited specific individual and group behaviours. Exploration of the environment took place in an individual manner, as was found by SCHOFIELD et al. (2006). Consumption of food among the animals observed in the Wroclaw Zoo also took place in a group, probably as a result of certain feeding times. The turtles could not search for and take food at any time, so they took the opportunity to take food when it was distributed. Self-care behaviours were not observed in this study. Conflicts between males and females in this study were impossible to identify because the sex of the turtles was not determined. There were no conflicts between turtles observed in this study other than those observed during feeding. However, these behaviours were the result of natural co-occupation rivalry. Although they are a natural phenomenon, these behaviours can result in undesirable injuries and infections if left unmanaged. No reproductive behaviour was observed in this study, because of the young age of turtles.

Observations carried out in the Wroclaw Zoo revealed several abnormalities in turtle behaviour. One of these was evidence of interaction with transparent boundaries, resulting from the fact that the walls of the turtle enclosure were made of glass. The turtles did not see transparent barriers and therefore pressed against them, trying to get out of the aquaterrarium. Theoretically, the turtles could also see their reflection in the glass. In this case, pressing on the glass would not be an effect of the interaction with transparent boundaries, but rather an attempt to interact with the turtle's own reflection. However, it is difficult to determine whether this was occurring. It is also abnormal that there was no hierarchy in the group examined in this study. Under normal conditions, animals living in a group usually develop a hierarchical structure. BOICE et al. (1974) stated that hierarchies are established after natural cycles such as hibernation. However, no such natural cycles occurred for this observed group, and this could be responsible for the lack of hierarchy. The resting patterns of the animals in this group are also noteworthy. According to SCHOFIELD et al. (2006), resting should be an individual behaviour. Therefore, resting in a group may be a form of previously unspecified disorder resulting from a limited living space. On the other hand, it may also be that for this species group resting is normal behaviour resulting from an increased tolerance of these turtles towards their co-occupants. Other observed behaviours included turtles hiding their head, limbs and tails in the shell, excreting urine or faeces and hesitating during food catching, but these behaviours are typical in the face of danger, so they were not considered to be abnormal.

Normal behaviour of reptiles associated with a sufficient level of welfare includes environmental exploration, species-specific behaviour (gearing, thermoregulatory behaviour) and behavioural diversity (IZZO et al. 2011, but see BASHAW et al. 2016). According to WARWICK et al. (2013), normal behaviours include quiet environmental exploration, subtle changes in body posture and orientation, calm food intake and quiet breathing.

The turtles examined in this study demonstrated behaviours consistent with these standards. The only exception was the lack of calm feeding, which was probably tied to group maintenance and specific feeding times.

The aim of this study was to determine the welfare of the focal turtles on the basis of observations of their behaviour. It was shown that the observed turtles were characterized by a number of normal behaviours. However, there were also a few behaviours that indicated a reduced level of animal welfare. These behaviours may have resulted from the moderate stress associated with the large number of people visiting the zoological garden in Wroclaw, in combination with the adaptive limitations of the examined animals, as well as a limited and controlled living space. Based on the results obtained here, it can be inferred that the welfare of *M. nigricans* has been moderately well-preserved, but that the artificial conditions created by humans are not able to fully satisfy the behavioural needs of the studied animals.

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References

ANDERS B., IVERSON J.B. 2012. *Mauremys nigricans (Gray 1834) – Red-necked Pond Turtle, Chinese Red-necked Turtle, Kwangtung River Turtle, Black-necked Pond Turtle*. In: *Conservation biology of freshwater turtles and tortoises. A compilation project of the IUCN/SSC Tortoise and Freshwater Turtle Specialist Group*. Eds. A.G.J. Rhodin, P.C.H. Pritchard, P.P. van Dijk, R.A. Saumure, K.A. Buhlmann, J.B. Iverson, R.A. Mittermeier. Chelonian Research Monographs No. 5: 1–9.

ARENA P.C., WARWICK C., STEEDMAN C. 2014. *Welfare and environmental implications of farmed sea turtles*. Journal of Agricultural and Environmental Ethics, 27(2): 309–330.

ARTNER H. 2009. *Successful breeding of the Chinese red-necked pond turtle* Mauremys [Chinemys] nigricans (Gray, 1834). Emys, 16(2): 4–22.

BASHAW M.J, GIBSON M.D., SCHOWE D.M., KUCHER A.S. 2016. *Does enrichment improve reptile welfare? Leopard geckos (*Eublepharis macularis*) respond to five types of environmental enrichment*. Applied Animal Behaviour Science, 184: 150–160.

BAYS T., LIGHTFOOT T., MAYER J. 2006. *Exotic pet behavior, birds, reptiles and small mammals*. Saunders-Elsevier, St. Louis.

BOICE R., QUANTY C.B., WILLIAMS R. C. 1974. *Competition and possible dominance in turtles, toads and frogs*. Journal of Comparative and Physiological Psychology, 86(6): 1116–1131.

Farm Animal Welfare Council (FAWC), 1992. *FAWC updates the five freedoms*. Veterinary Record, pp. 131–357.

FUSTER J. F., PAGES T., PALACIOS L. 1997. *Effect of temperature on oxygen stores during aerobic diving in the freshwater turtle,* Mauremys caspica leprosa. Physiol. Zool., 70: 7–18.

GAILLARD D., LIU L., HAITAO S., SHUJIN L. 2017. *Turtle soup: local usage and demand for wild caught turtles in Qiongzhong County, Hainan Island*. Herpetological Conservation and Biology, 12(1): 33–41.

IUCN. 2017. *IUCN Red List of Threatened Species*. Version 2017.2, www.iucnredlist.org.

IVERSON J.B. 1985. *Checklist of the Turtles of the World with English Common Names*. Society for the Study of Amphibians and Reptiles, Herpetological Circulars, 14: 1–14.

IVERSON J.B. 1992. *A revised checklist with distribution maps of the turtles of the world*. Richmond, IN: Privately published, p. 363.

KALETA T. 2013. *Zachowanie się niższych kręgowców trzymanych przez człowieka jako wskaźnik ich dobrostanu*. Życie Weterynaryjne, 88(10): 860–866.

MELL R. 1922. *Beiträge zur Fauna sinica*. I. *Die Vertebraten Südchinas; Feldlisten und Feldnoten der Säuger, Vögel, Reptilien, Batrachier*. Archiv für Naturgeschichte, 88: 1–134.

MELL R. 1929. *Beiträge zur Fauna sinica*. IV. *Grundzüge einer Ökologie der chinesischen Reptilien und einer herpetologischen Tiergeographe Chinas*. Berlin, p. 282.

MOBERG G. 1985. *Animal stress*. American Physiological Society, Bethesda.

MORGAN K.N., TROMBORG C.T. 2007. *Sources of stress in captivity*. Applied Animal Behaviour Science. Conservation, Enrichment and Animal Behaviour, 102: 262–302.

SCHOFIELD G., KATSELIDIS K.A., DIMOPOULOS P., PANTIS J.D., HAYS G.C. 2006. *Behaviour analysis of the loggerhead sea turtle* Caretta caretta *from direct in-water observation*. Endangered Species Research, 2: 71–79.

SILVESTRE A.M. 2014. *How to assess stress in reptiles*. Journal of Exotic Pet Medicine, 23(3): 240–243.

Turtles in trouble. The world's 25+ most endangered tortoises and freshwater turtles – 2011. 2011. Turtle Conservation Coalition. Eds. A.G.J. Rhodin, A.D. Walde, B.D. Horne, P.P. van Dijk, T. Blanck, R. Hudson, MA: IUCN/SSC Tortoise and Freshwater Turtle Specialist Group, Turtle Conservation Fund, Turtle Survival Alliance, Turtle Conservancy, Chelonian Research Foundation, Conservation International, Wildlife Conservation Society, and San Diego Zoo Global, Lunenburg, p. 54.

WABNITZ C., TAYLOR M., GREEN E., RAZAK T. 2003. *From ocean to aquarium. The global trade in marine ornamental species*. UNEP.

WARWICK C., ARENA P., LINDLEY S., JESSOP M., STEEDMAN C. 2013. *Assessing reptile welfare using behavioural criteria*. In Practice, 35(3): 123–131.

WARWICK C., JESSOP M., ARENA P., PILNY A., STEEDMAN C. 2018. *Guidelines for inspection of companion and commercial animal establishments*. Front. Vet. Sci., 5: 151.