Ann. Anim. Sci., Vol. 18, No. 1 (2018) 17–37 DOI: 10.1515/aoas-2017-0025

DE GRUYTER OPEN

FRESHWATER TURTLE NUTRITION – A REVIEW OF SCIENTIFIC AND PRACTICAL KNOWLEDGE*

Mateusz Rawski[1,2♦], Christoph Mans[3], Bartosz Kierończyk[1] Sylwester Świątkiewicz[4], Aneta Barc[1], Damian Józefiak[1]

[1]Department of Animal Nutrition and Feed Management, Poznań University of Life Sciences, Wołyńska 33, 60-637 Poznań, Poland
[2]Division of Inland Fisheries and Aquaculture, Institute of Zoology, Poznań University of Life Sciences, Wojska Polskiego 71C, 60-625 Poznań, Poland
[3]Department of Surgical Sciences, School of Veterinary Medicine, University of Wisconsin-Madison, 2015 Linden Drive, WI 53706, Madison, United States of America
[4]Department of Nutrition Physiology, National Research Institute of Animal Production, Balice n. Kraków, Poland
[♦]Corresponding author: mrawski@up.poznan.pl

Abstract

Freshwater turtles are commonly kept in captivity as pets, bred in zoos for conservation programs, and commercially farmed for pet markets and human consumption, but their nutrition can be challenging. However, based on practical experience, two main strategies may be identified: the use of non-calculated raw diets and the use of balanced commercial feeds. Raw diets are based on fresh, frozen and dried components including invertebrates, fish, rodents and plant matter; they imitate the variety of foods that are accessible to turtles in the wild and are considered most useful when turtles are bred for reintroduction into their natural habitat as part of conservation programs. Granulated, pelleted or extruded commercial diets are frequently used for farmed and pet turtles; they contain animal- and plant-based materials supplemented with vitamin and mineral premixes and calculated to reach the nutrient levels assumed to be optimal for most species. Until more species-specific information on the nutritional requirements of freshwater turtles is available, the Chinese softshell turtle (*Pelodiscus sinensis*), a commonly commercially farmed species for human consumption, may be used as a reference for other species in terms of suggested nutrient levels. Based on experimental data, the most important nutrients and their levels that should be included in turtle diets are crude protein (39.0–46.5%), crude fat (8.8%), Ca (5.7%), P (3.0%), methionine (1.03%), and cysteine (0.25%). The diet composition for freshwater turtles should be based on scientific knowledge and practical experience, so this paper aimed to present and discuss the available data on the nutrient requirements of turtles and the characteristics of the feed materials used in their nutrition.

Key words: freshwater turtles, turtle nutrition, nutrient requirements, metabolic diseases, *Pelodiscus sinensis*

* The current study was supported by the grant NCN PRELUDIUM: UMO-2013/11/N/NZ9/04624, financed by National Science Centre (Poland).

Freshwater turtles are widely distributed in almost all types of aquatic habitats (Bonin et al., 2006), so their diets and feeding strategies in the wild vary substantially. However, most are opportunistic carnivores or omnivores, consuming invertebrates, small vertebrates, and aquatic vegetation (Bouchard and Bjorndal, 2006; Gibbons, 1990; Luiselli et al., 2011; Ottonello et al., 2005; Rhodin et al., 2008; Spencer et al., 1998). The wide spectrum of feeding strategies among freshwater turtles and their slow metabolism may explain their high tolerance for unbalanced diets, but their longevity and the energetic expense required for shell mineralization make them vulnerable to nutritional deficiencies in captivity (McWilliams, 2005). Moreover, the nutrient requirements for most species, especially those not routinely used in large-scale turtle farming, are poorly documented. It should be highlighted that the nutritional needs of freshwater turtles are affected by numerous factors such as the species, environmental conditions, digestion and assimilation efficiency, sex, age, health status and history of specimen (Figure 1), but diet composition should be both species-specific and suitable for raising turtles in captivity. Therefore, diets used in commercial turtle farming may not be adequate for non-commercial purposes in terms of feed ingredients or physical form of the feed which are optimized in terms of feed utilization and economic results. However, the available data on the nutritional requirements of turtle species that are commonly raised commercially (i.e., the Chinese softshell turtle (*Pelodiscus sinensis*) may be an important source of general information for other turtle species kept as pets, in zoological institutions or as part of conservation programs. This paper aimed to present and discuss the available scientific data on the nutrient requirements of turtles and the characteristics of the feed materials used in their nutrition.

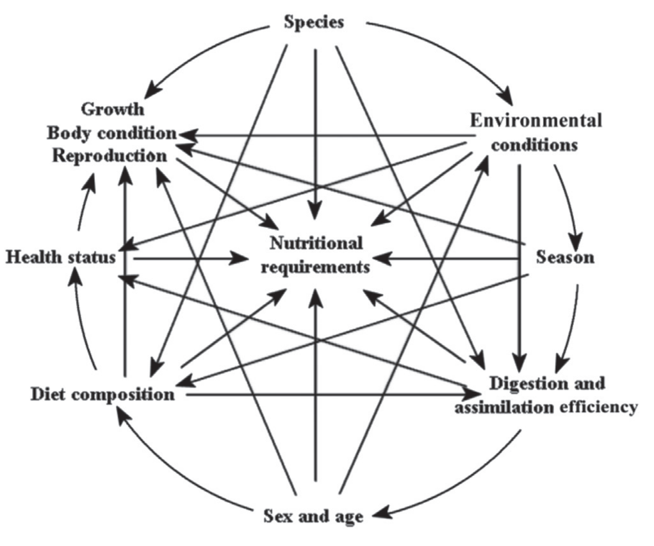


Figure 1. Extrinsic and intrinsic factors and their interactions that affect the nutritional requirements of turtles (Bouchard, 2004; Gibbons, 1990; Luiselli et al., 2011; McCauley and Bjorndal, 1999; Seebacher et al., 2004; Spencer et al., 1998; Zhang et al., 2009)

Gastrointestinal tract anatomy and physiology of freshwater turtles Turtles are monogastric animals, and their gastrointestinal tract (GIT) begins with an oral cavity that has no lips or teeth (Mitchell and Tully, 2009). Therefore, they are unable to chew their food but instead swallow entire prey or large bites. In some species, feed cutting and shredding is performed with the use of a beak-like keratin layer on the upper and lower jaw (*rhamphotheca*) as well as the claws. The shape and function of the *rhamphotheca* is very similar among omnivorous turtle species, such as pond turtles (*Emydidae*), but in highly specialized turtles, the structure reflects their feeding habits. The *rhamphotheca* may be very well developed as in the alligator snapping turtle (*Macrochelys temminckii*), which has powerful jaws, or reduced as in the matamata turtle (*Chelus fimbriata*), which has a limited bite force (Lemell et al., 2010). Due to the large feed intake by a turtle during a single meal, the pharynx, esophagus and stomach are highly flexible. In the stomach, the low pH and enzymatic activity starts the digestion process. The stomach is curved to the left, shorter and wider than the esophagus, and its mucosal surface is divided into the proper gastric and cardiac glandular mucosa regions (Stevens and Hume, 1998). There are two kinds of gastric glands in the stomach, peptic cells and oxyntic cells, which indicates that turtles are well adapted for omnivory (Rahman and Sharma, 2014).

The small intestine is the longest organ in the turtle GIT (Figure 2), but the duodenum, jejunum and ileum are not well distinguished and difficult to identify (Rahman and Sharma, 2014). The length and capacity of the intestines is diet-dependent and may vary significantly between species and different diets (Bouchard, 2004). The mucosa of the small intestine is composed of a single columnar epithelium, and the lining of the intestinal villi includes three types of cells: simple columnar cells, goblet cells and endocrine cells (McArthur et al., 2008; Rahman and Sharma, 2014; Wurth and Musacchia, 1964). The large intestine seems to be a site for water absorption, microbial fermentation and short-chain fatty acid (SCFA) production (Bouchard, 2004); its proximal part is the caecal extension of the colonic wall (Bouchard, 2004; McArthur et al., 2008). The colon is typically divided into three parts: ascending, transverse and descending. The reptile GIT ends with the cloaca, the site where the terminal parts of the GIT, urinary and reproductive tracts join, which is subdivided into the coprodeum, urodeum and proctodeum (McArthur et al., 2008; Mitchell and Tully, 2009). Turtle saliva does not contain digestive enzymes, which are instead secreted by the stomach, pancreas and intestines. Chelonian stomachs secrete amylase, pepsin, trypsin, chitinase and chitobiase; the pancreas secretes amylase, ribonuclease, trypsin, chymotrypsin, carboxypeptidase A and chitinase; and the intestines secrete proteinase, invertase, amylase, maltase, chitobiase, trehalase, isomaltase and sucrase (McArthur et al., 2008). Protease and amylase are considered the two main digestive enzymes in the turtle GIT (Sun et al., 2007), and the optimal conditions for their specific activities vary among GIT segments. In the pond slider (*Trachemys scripta*), pancreatic protease shows the highest activity (36 U/mg of protein), and in the stomach, maximal protease activity (24 U/mg of protein) was recorded at pH 2.5 and 40°C. In contrast, amylase had the highest activity (12 U/mg of protein) in the anterior intestine under neutral conditions (Sun et al., 2007). The liver is, to some extent, divided into triangular lobes (Rahman and Sharma, 2014), and it plays a key role in vitamin D_3 synthesis and storage and the transformations of lipids, proteins and glycogen. Another important GIT gland is the pancreas, which is situated along the proximal segment of the duodenum (McArthur et al., 2008). Because turtles are ectothermic, their metabolic rate and digestion mainly depends on the temperature of the environment and external heat sources.

For most turtle species, feed intake and enzyme secretion and activity, as well as the absorptive capacity of the intestinal mucosa, are highest and feed passage is shortest in the preferred optimal temperature zone (POTZ), above which these parameters decrease (Figure 3) (McArthur et al., 2008; Seebacher et al., 2004; Sun et al., 2007). In most species, this zone is between 25 and 34°C (Table 1) (Gibbons, 1990; Mitchell and Tully, 2009), and turtles can even reach their POTZ in lower air temperatures through basking behavior. However, there are exceptions, especially in species that naturally inhabit cold mountain creeks such as the big-headed turtle (*Platysternon megacephalum*), whose POTZ is 22–25°C (Jianwei et al., 2013; Zhang et al., 2009). Due to the above-mentioned behavioral mechanism and the lack of energetic expenses for heat production, the average reptile energy expenditure is only 25–35% that of mammals (Mader, 2005). In reptiles, energy utilization mainly depends on feeding strategy and diet composition. Carbohydrates are a source of 75% of the metabolizable energy for herbivorous and 50% for omnivorous reptilian species. In carnivores, carbohydrates provide only 5% of the dietary matabolizable energy, while protein provides 50% and fat 45% (Hand et al., 2000; Mader, 2005). The function of the GIT microbiota has not been well studied in reptiles, but it seems to play an important role in the secretion of bacterial enzymes and the immunological response. Similar to other animals, microbial homeostasis in reptiles may improve gut health, while disturbances in composition may lead to depressed growth and subclinical and clinical infections (Lei and Yaohong, 2010; Zhang et al., 2014; Rawski et al., 2016). The microbial GIT symbionts in turtles significantly support plant matter digestion and produce SCFAs, which may be an important energy source for omnivorous or herbivorous animals (Bouchard and Bjorndal, 2005). Based on its SCFAs concentrations, the anterior large intestine should be considered the main site of microbial fermentation of carbohydrates of plant origin in *T. scripta*. In the Florida red-bellied cooter (*Pseudemys nelsoni*), microbial fermentation may also occur in the small intestine (Bjorndal and Bolten, 1990; Bouchard and Bjorndal, 2005). It has been suggested that the importance of SCFAs as energy source increases with an increase in the amount of plant matter in the turtle diet, but it may also depend on fermentation capacity (Bouchard, 2004; Bouchard and Bjorndal, 2005). The pattern of the relative proportions of SCFAs in *T. scripta* was described as acetate > propionate > butyrate > valerate (Bouchard and Bjorndal, 2005). During development, several species undergo an ontogenetic diet shift from carnivorous hatchlings to omnivorous adults (Bouchard, 2004; Bouchard and Bjorndal, 2006; McCauley and Bjorndal, 1999); this occurs in *Emydidae* and *Chelidae* as well as in other reptile species (Bouchard, 2004; Bouchard and Bjorndal, 2005, 2006; Kennett and Tory, 1996). Both juvenile and adult turtles digest animal and plant matter, but animal matter has a higher digestibility compared to plant matter in juvenile *T. scripta* (97.2% vs. 89.4%) and results in greater growth (0.2 vs. 0.6 g/week). In adults, plant matter is more digestible (Bouchard and Bjorndal, 2006). However, in most cases, adult turtles do not become predominately herbivorous; in *T. scripta*, the animal to plant matter ratio in the diet was recorded as 77:23 (Gibbons, 1990). This diet composition may by highly nutritive for GIT microbiota, and the nutrients supplied from animal matter may support plant matter microbial fermentation (Bjorndal, 1991). It is assumed that adult turtles can meet their metabolic demands on a plant-based diet, which would be insufficient to meet juvenile growth requirements due to the low concentration of protein and energy (Bouchard and Bjorndal, 2006).

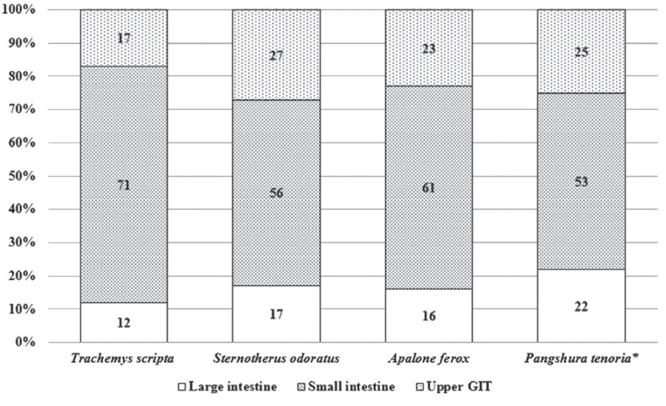
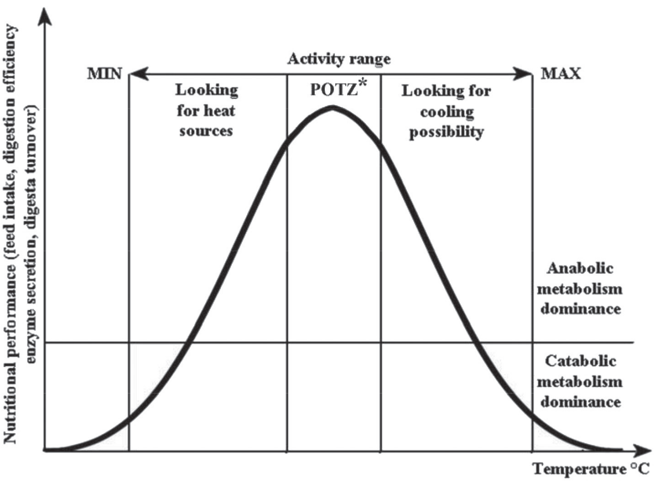


Figure 2. Gastrointestinal tract (GIT) segment proportions (% of the entire GIT length) in selected turtle species. Data for *Trachemys scripta* (n=40, 1 year old non-sexed specimens), *Sternotherus odoratus* (n=36, 1 year old non-sexed specimens) and *Apalone ferox* (n=40, 1 year old non-sexed specimens), M. Rawski unpublished data. *Data for *Pangshura tenoria* (n=20, adult specimens, both sexes) based on the literature (Rahman and Sharma, 2014)



*POTZ – preferred optimal temperature zone.

Figure 3. Dependence of turtle nutritional performance (including feed intake, digestion efficiency, enzyme secretion, digesta turnover) on environmental temperature (Seebacher et al., 2004; Zhang et al., 2009)

Table 1. A summary of published reports on body temperature in free-ranging turtles and the preferred body temperature

Species	Field body temperature (ºC)	Preferred body temperature (ºC)	References
Chelydra serpentina	22.7 (SD=2.8)	27–30 (hatchlings) 27–33 (yearlings)	Brown et al., 1990; Bury et al., 2000
Macroclemys temmincki	19.96 (12.21–27.76)	NA*	Fitzgerald and Nelson, 2011
Platysternon megacephalum	19.3–22.2	25.3 (juveniles)	Jianwei et al., 2013
Chrysemys picta	25–32 (basking temperature)	34 (juveniles)	Grayson and Dorcas, 2004; Tamplin and Cyr, 2011
Glyptemys insculpta	23.2 (SD=3.9) 5–30 (basking temperature)	30 (juveniles)	Ernst, 1986; Tamplin, 2009
Pseudemys nelsoni	NA*	30 (hatchlings)	Nebeker and Bury, 2000
Terrapene ornata	28.0 (15.3–35.3)	28.3 (fasted) 29.8 (recently fed)	Gatten Jr, 1974; Legler, 1960
Trachemys scripta	27.2–38.0	30 (hatchlings)	Bury et al., 2000; Gibbons, 1990
Ocadia sinensis	NA*	25.4–29.2 (juveniles)	Pan et al., 2002
Apalone spinifera	NA*	30 (juveniles)	Feltz and Tamplin, 2007
Pelodiscus sinensis	NA*	30.3 (juveniles)	Sun et al., 2002
Chelodina longicollis	20.2–24.4	NA*	Seebacher et al., 2004

*NA – data not available.

Nutritional requirements

In contrast to domesticated animals, no standardized nutritional requirements are available for most freshwater turtles, which makes the provisioning of a proper diet in captivity challenging. There are general rules for feeding different age groups of turtle species that inhabit a wide range of ecological niches, but it should be emphasized that in the case of highly specialized species, such as the matamata (*Chelus fimbriata*), only species-specific diets that reflect their feeding ecology in the wild should be provided. Nutritional requirements are best known for commonly farmed freshwater turtle species, and they may be used as a reference and by analogy for other turtles until better information is published. The Chinese softshell turtle (*Pelodiscus sinensis*) is the best-known species due to large-scale farming in Asia, and many studies on its nutritional requirements have been published (the results are summarized in Table 2). In the first days after hatching, turtles may not consume any food due to yolk sac absorption, which satisfies some of the nutritional needs of hatchlings (Mitchell and Tully, 2009). The hatchlings of most freshwater turtle species are almost strictly carnivorous (Bouchard, 2004; Bouchard and Bjorndal, 2006), so their growth performance is correlated with the concentration of crude protein (CP) in the diet (Gibbons, 1990). The optimal CP level for young *Pelodiscus sinensis* is assumed to be as high as 39–46.5% (Jia et al., 2005; Nuangsaeng and Boonyaratapalin, 2001; Zhou et al., 2013) and is likely dependent on the energy content of the diet (Nuangsaeng and Boonyaratapalin, 2001). A study of *P. sinensis* suggests that the optimal CP to energy ratio of the diet should be at the level of 32–36 mg/kJ^{-1} (Zhou et al., 2013). This ratio should be considered an important factor in diet suitability; when the ratio is too low, it may not only limit growth but feed intake as well. Crude protein characteristics, such as the quantity, ratios and bioavailability of essential amino acids, are key factors in animal nutrition (Ei and Kavas, 1996). Methionine and cysteine seem to be limiting amino acids in *P. sinensis* with estimated optimal levels of 1.03% and 0.25% of the diet, respectively (Huang and Lin, 2002). No experimental information is available for lysine. Exogenous taurine also seems to be essential and should constitute 0.9% of the diet, especially when CP of plant origin is used (Hou et al., 2013). The CP concentration in captive turtle diets may be lowered when the animals reach sexual maturity and their growth rate decreases, and plant matter should be provided to achieve this goal. A nutritional experiment using the scorpion mud turtle (*Kinosternon scorpioides*) indicated that slow-growing adults may be fed CP at a lower level than young turtles (26%), but in the case of breeding stock females, diets containing 61–66% animal-derived CP increased laying performance and egg quality compared to 26% dietary CP (da Costa Araújo et al., 2013). Furthermore, in the case of slow-growing, non-breeding stock adult males or females, the balance of protein and energy should be only slightly above zero to avoid obesity (Rawski and Józefiak, 2014).

Nutritional recommendations for *P. sinensis* diets are optimized to maintain high growth performance, so when turtles are not maintained under commercial farming conditions, energy and protein concentrations may be lowered to prevent too-rapid growth and poor skeletal system development. Due to the low energetic expenses of turtles and the high availability of feed in captivity, no additional source of dietary fat is needed in most cases. However, in fast-growing *P. sinensis*, the optimal fat content in the diet is estimated to be 8.8%, and there is no apparent effect of fat source on growth performance (Lin and Huang, 2007). Turtles probably have the highest skeletal mass to body weight ratio among all vertebrates. Calcium and phosphorus should be given at a ratio of approximately 2:1, i.e., 5.7% and 3.0% of the diet, respectively, according to studies of *P. sinensis* (Huang et al., 2003); a lower Ca:P ratio may cause shell malformations or lower growth rates. Other minerals such as Mg, Fe, Zn and Cu also seem to be important for turtle metabolism and shell mineralization (Chen et al., 2014; Chu et al., 2007; Huang et al., 2003; Huang et al., 2010; Wu and Huang, 2008). Vitamins are a group of complex organic compounds that are present in small amounts in plant and animal matter. Their ingestion is essential for normal metabolism, and deficiencies can lead to various diseases (McDowell, 1989). Vitamin C has been shown to have an important role in the ability of turtles to withstand stress (Zhou et al., 2002). The diet of *P. sinensis* should contain 2500 mg/kg of vitamin C and 88 IU/kg of vitamin E (Huang and Lin, 2004; Zhou et al., 2002). For vitamin A, 2000–8000 IU/kg of the diet on a dry matter basis seems to be adequate (Mader, 2005), but this vitamin may be synthesized by turtles using β-carotene, of which 50–90 mg/kg should be provided in the *P. sinensis* diet (Chen and Huang, 2011). In contrast, lutein and canthaxanthin may be equally or more effective forms of provitamin A in reptiles, which may selectively absorb carotenoids (Raila et al., 2002). Vitamin D_3 synthesis occurs in reptilian skin and is stimulated by UVB radiation; in carnivorous and omnivorous species, dietary sources of this vitamin seem to play an important role (Hoby et al., 2010). However, despite being the key for shell mineralization, the vitamin D_3 requirements of turtles are still poorly known (McArthur et al., 2008).

Table 2. Summary of the published reports on the nutritional requirements of young Chinese softshell turtles (*Pelodiscus sinensis*)

Item	Unit	Optimal level	References
Protein to energy ratio	mg/kj^{-1}	32–36	Zhou et al., 2013
Protein	%	39.0–46.5	Jia et al., 2005; Nuangsaeng and Boonyaratapalin, 2001; Xie et al., 2012; Zhou et al., 2013
Fat	%	8.8	Huang et al., 2005
Calcium	%	5.7	Huang et al., 2003
Phosphorus	%	3.0	Huang et al., 2003
Methionine	%	1.03	Huang and Lin, 2002
Methionine	% of protein	2.48	Huang and Lin, 2002
Cysteine	%	0.25	Huang and Lin, 2002
Cysteine	% of protein	0.60	Huang and Lin, 2002
Taurine	%	0.90	Hou et al., 2013
Magnesium	mg/kg	970–980 650–750 (phytic acid free diet)	Chen et al., 2014
Iron	mg/kg	266–325	Chu et al., 2007
Zinc	mg/kg	35–46	Huang et al., 2010
Copper	mg/kg	4–5	Wu and Huang, 2008
β-carotene	mg/kg	49–89	Chen and Huang, 2011
Vitamin C	mg/kg	2500–5000	Zhou et al., 2002
Vitamin A	mg/kg	2.58–3.84	Chen and Huang, 2014
Vitamin E	IU/kg^{-1}	40	Huang and Lin, 2004

Feeding strategies in captivity

Due to the practical experience of zoos and breeders, two main strategies may be distinguished for providing adequate nutrition for turtles in captivity. The first is the use of non-calculated, raw diets based on unprocessed or minimally processed components, such as live, fresh, dried or frozen food items; the main aim is to imitate the natural diets of freshwater turtles with undetermined nutritional requirements (examples of natural diet compositions are shown in Table 3). The second strategy is based on commercial diets, which may be considered suitable for the most commonly kept turtle species (in the *Emydidae* and *Pelomedusidae* families). Another important issue is feeding frequency and quantity, which are restricted under natural conditions by prey availability, season and predation success. These factors lead to periodic starvation that results in the use of body energy reserves, which tends to result in lower growth and breeding performance than would have been possible based on the genetic potential of the animals. This is in contrast to the situation in captivity, where regular access to an appropriate diet will lead to the fulfillment of the genetic potential. However, it should be emphasized that food restriction seems to have health benefits and promotes longevity in animals (Lawler et al., 2005). Additionally, high nutrient and energy availability may result in accelerated growth and poor bone mineralization in turtles, as observed in fast-growing poultry and other animals (Julian, 1998), and it may also cause obesity in adult animals, including turtles (Mader, 2005; Mitchell and Tully, 2009; Rawski and Józefiak, 2014). Turtles can achieve high feed intake; *T. scripta elegans* may consume up to 12% of its body weight during one meal (Rawski, unpublished data). Under experimental conditions, 4% of body weight was reported to be an optimal meal size for suitable growth performance in *P. sinensis* since higher feed intake may decrease nutrient digestibility (Lei, 2006). In contrast, commercial feed producers frequently advise that turtles be fed *ad libitum* or during restricted time periods, sometimes even more than once daily. However, in the opinion of this author, restricting the amount, not feeding time, of commercial feeds containing a high amount of dry matter may be more effective at preventing overfeeding, excessive growth and obesity.

Table 3. Diet composition of selected turtle species in nature

Species	Diet composition	Sampling method	References
Trachemys scripta	Animal matter: *Gastropoda: Physidae, Insecta: Coleoptera, Diptera, Hymenoptera, Odonata: Anisoptera, Zygoptera, Orthoptera: Locustidae* Fish, unknown claws and bones, crayfish, shrimps Plant matter: Algae, *Bacopa caroliniana, Brasenia schreberi, Najas guadelupensis, Nymphaea odorata, Potamogeton* spp., *Sagitaria* spp., *Utricularia* spp., *Lemna spp.*	stomach flushing	Gibbons, 1990
Emys orbicularis	*Gastropoda: Bithyniidae, Lymnaeidae, Physidae, Planorbidae*, *Arachnida: Acarina, Araneae*, *Crustacea: Conchostraca, Decapoda* *Insecta: Coleoptera, Diptera, Heteroptera, Hymenoptera, Odonata Trichoptera* Vertebrata and plant matter	fecal samples	Ottonello et al., 2005
Emydura macquarii	*Arthropoda: Arachnida, Decapoda* *Insecta: Hemiptera, Coleoptera, Diptera, Trichoptera Hymenoptera* Vertebrates, filamentous algae, plant detritus	stomach content	Spencer et al., 1998
Pelomedusa subrufa	Rodents, Birds, Lizards, Snakes, Tadpoles, Frogs, Fish, *Gastropoda, Bivalvia, Anellida, Arachnida, Chilopoa, Crustacea*, *Odonata* larvae, *Rhynochota, Coleoptera* adult, *Coleoptera* larvae, plant matter, fungi	stomach flushing, fecal samples	Luiselli et al., 2011
Chelodina mccordi	Fish, tadpoles, insects, freshwater gastropods, water weeds	observations	Rhodin et al., 2008

Table 4. Nutritional composition of a whole prey used in captive turtle diets

Prey species	Notes	DM*	CP*	EE*	Ash	Ca*	P*	References
			% as feed			% on a DM basis		
Blood worm	-	9.9	53	9.7	12	0.38	0.85	Bernard et al., 1997
Black soldier fly	Larvae	30	38–60	9–26	3–17	0.30–0.80	0.90–2.4	Józefiak et al., 2016
American cockroach	-	39	54	28	3.3	0.20	0.50	Bernard et al., 1997
American cockroach	Nymph	37	54–73	18–26	4.6–5.4	0.02	0.06–0.07	Józefiak et al., 2016
Domestic cricket	Imago	27–38	40–68	14–44	2.7–5.7	0.14	0.99	Bernard et al., 1997; Mader, 2005
Domestic cricket	Larvae	33	40–50	10	9.1	0.1–0.2	0.8	Mader, 2005
Domestic cricket	Imago, high Ca diet	30	65	13	9.8	0.90	0.92	Bernard et al., 1997
Jamaican field cricket	Imago	31	56	24	6.4	0.80	0.99	Józefiak et al., 2016
Earthworm	-	20	62	18	5.0	1.7	0.90	Bernard et al., 1997; Mader, 2005
Night crawler	Wild	15–26	31–81	6–13	9–46	0.97–1.5	0.79–0.96	Bernard et al., 1997; Mader, 2005
Night crawler	Commercial	16–24	50–81	11–13	25	1.2	0.86	Mader, 2005
Mealworm	Larvae	38–43	53	31–60	3.0–7.0	0.04–0.12	0.83–1.4	Bernard et al., 1997; Mader, 2005
Superworm	Larvae	41–43	40–50	41–44	2.9–3.5	0.03–0.12	0.6–0.8	Mader, 2005
Tubifex worm	-	12	46	15	6.9	0.19	0.73	Bernard et al., 1997
Wax moth	Larvae	34	42	46	2.7	0.11	0.62	Bernard et al., 1997
Wax moth	Larvae, high Ca diet	40	NA†	NA†	2.5	0.50	0.33	Bernard et al., 1997
Domestic mouse	Neonatal, <3 g	19–26	51–64	17–34	8.0–9.7	1.2–3.5	1.6	Crissey et al., 1999; Dierenfeld et al., 2002; Douglas et al., 1994
Domestic mouse	Juvenile, 3–10 g	18–29	44–59	24–30	8.5–10	1.5–3.0	1.4	Crissey et al., 1999; Dierenfeld et al., 2002; Douglas et al., 1994

Domestic mouse	Adult or > 10 g	33	56	24	11-12	2.6-3.0	1.7–1.9	Clum et al., 1996; Crissey et al., 1999; Dierenfeld et al., 2002; Douglas et al., 1994
Domestic rat	Neonatal, <10 g	21	65	16	12	1.9	NA[†]	Dierenfeld et al., 2002; Douglas et al., 1994
Domestic rat	Juvenile, 10–50 g	23–30	58–60	24–27	12–15	2.1	NA[†]	Dierenfeld et al., 2002; Douglas et al., 1994
Domestic rat	Adult or > 50 g	34	56	12	9.8	2.6	1.72	Clum et al., 1996; Dierenfeld et al., 2002; Douglas et al., 1994
Chicken	One-day-old	26	65	22	6.4	1.7	1.2	Dierenfeld et al., 2002
European smelt	Dried	91	47	34	11	NA[†]	NA[†]	Declared by producer (Katrinex, Poland)

[*]Abbreviations: DM – dry matter, CP – crude protein, EE – ether extract (crude fat), Ca – calcium, P – phosphorus, NA – data not available.

Raw diets

Raw diets are considered most suitable for turtles designated for reintroduction into the wild as well as for those used as breeding stocks in conservation programs. The diets should be as similar as possible to the variety of food resources accessible to specific turtle species in their natural environment, including live prey, and in most cases, these diets are based on invertebrates, insects and their larvae, as well as small vertebrates to maintain foraging and hunting abilities of animals at optimal level. Many turtle breeders and zoos use gelatin-based diets (puddings) that fall somewhere between raw and commercial diets. These diets are multi-ingredient mixtures solidified by gelatin and represent the easiest way to maintain a diverse diet based on fresh and frozen ingredients. The main advantage of these diets is the possibility for modifying recipes according to changes in scientific knowledge, experience and the available components. All raw components should be used fresh or after a single freezing; prolonged storage or refreezing may promote microbial contamination and nutrient degradation, which can result in negative side effects for the animals. Gelatin-based diets should be offered at a temperature similar to that of the turtles' environment and not frozen. If raw diets are aimed at imitating natural ones, invertebrates, fish, rodents and aquatic plants should be used. The nutritional values of materials of animal origin that are commonly used in turtle nutrition are given in Tables 4 and 5. It should be suggested that even in the case of raw, nature imitating diets use, their nutritive value should be calculated and at the diet composition optimized according to current knowledge about nutritional requirements of turtles.

Table 5. Nutritional composition of commercial turtle feeds based on producers declarations

Nutrient (%)	Nonspecific feeds[1]	Age specific feeds[2]		
	all turtles	hatchling	growth formula	adult maintenance
Crude protein (min)	38	39	35	25
Fat (min)	7.4	10	5	5
Fiber (max)	3.4	3	5	8
Ca (min)	2.2	ND[3]	ND[3]	ND[3]
P (min)	1.2	1	1	1

[1]Based on average of declared nutritional values of 15 commercial feeds recommended by producers as formulations for all turtles.

[2] Based on declared nutritional values of feeds recommended by producer as age specific formulations.

[3] ND – no declaration.

Invertebrates

Insects are a rich source of high-quality protein, essential amino acids and other nutrients. Additionally, they have short life cycles and are easy to produce and handle (Józefiak et al., 2016; Ramos-Elorduy et al., 2002). Due to a high amount of invertebrates in the natural diets of many turtle species (Table 3), insects are an important dietary component in captivity, but the high fat content in insect larvae, e.g., meal worms (*Tenebrio molitor*) or wax worms (*Pyralidae*), may lead to excessive energy intake. Chitin is the main component of insect exoskeletons, and it seems to be well digested by chitinases and chitobiases produced by the stomach and pancreas in turtles (McArthur et al., 2008). An important disadvantage of feeder insects is that their Ca:P ratio is less than optimal (2:1), and most contain low amounts of vitamin A and D_3. Therefore, supplementation of additional Ca and vitamin A and D_3 is required. Feeding insects a vitamin-and-mineral-rich diet shortly before feeding them to turtles, which is also known as "gut-loading," will improve their nutritional content (Finke, 2003). Earthworms and night crawlers are also suitable turtle feed; they have high mineral contents because of the high volume of soil in their guts (Bernard et al., 1997). Similarly, a variety of shellfish may also be used due to their high nutritive value, but negative effects may result from the long-term use of shellfish-based diets due to the high concentrations of environmental pollutants in shellfish, especially heavy metals (Sivaperumal et al., 2007).

Fish

Fish are a natural and valuable component of captive turtle diets. They contain, on average, 15–20% high-quality protein that is rich in essential amino acids, i.e., lysine, methionine and cysteine. Moreover, fish are a good source of vitamins A, B complex and D_3 as well as minerals, such as Ca, P, Fe, and S, and long-chain polyunsaturated fatty acids (Tacon and Metian, 2013). Whole small fish should be fed frequently to most freshwater turtles and should be the main diet component for *Chelus fimbriata* and other piscivorous species. The presence of live small fish (*Poeciliidae* or *Danio* spp.) in the turtle tank may serve as a food source but also stimulate turtle foraging behavior. However, when a fish-based diet is used, frequent use of fish in the *Cyprinidae* family should be avoided due to their high amounts of thiaminase, a B_1 antivitamin (Mader, 2005).

Mammals and birds

For many years, the main component of captive turtle diets was animal matter derived from commercially raised domestic mammals or birds, which do not account for a significant proportion of natural freshwater turtle diets. However, whole carcasses (e.g., mice and rats or quails and chicks) are frequently used in captive turtle feeding. The skeletons and GIT contents of vertebrates provide valuable vitamins and minerals (Hand et al., 2000), and fur and feathers mechanically stimulate the GIT, similar to the function of the fiber in plant matter. Among the most commonly used rodents, adult mice seem to be most suitable for freshwater turtles due to their high mineral content and adequate Ca:P ratio (Mader, 2005). In contrast, filleted meat, an excellent protein source, should not be fed as one of the main diet components because of its low content of minerals and vitamins. If the abovementioned feeds, such as insects, whole fish or rodents, are not available or they are refused by the turtles, internal organs, particularly the liver and kidneys, are good alternatives due to their high protein quality and high concentrations of fat-soluble vitamins (Acker et al., 1959). However, care should be taken not to feed raw liver in large quantities since hypervitaminosis A, which is often fatal, can develop (Mans and Braun, 2014).

Plant matter

To simulate their natural diet, most adult freshwater turtles should be provided with various amounts of plant matter. In captivity, aquatic plants such as duckweed (*Lemna* spp.), pondweed (*Elodea* spp.), and hornwort (*Ceratophyllum* spp.) are frequently used. Algae such as *Spirulina* spp. are also used in commercial fish and turtle diets, and they are also available in a dried form and may be used as a separate feed component. Aquatic plants may be permanently present in turtle tanks and be ingested between main meals. For some tropical and subtropical turtle species, fruits may also be used, but in the case of *T. scripta elegans*, we observed digestive disturbances after feeding fruits, such as bananas (Rawski, unpublished data).

Commercial diets

Most commercial diets are extruded or hot pelleted and are based on animal and plant materials with dry matter contents close to 90%. The declared average nutritional values of various commercial diets are presented in Table 6, and in many of them, the Ca:P ratio is close to 2:1. However, the levels of these nutrients are low relative to the optimal levels for *P. sinensis*, not exceeding 2.1% for Ca and 1.4% for P. Frequently, the declared nutritional content is not described in detail or given just as minimal or maximal levels, but based on research carried out on *P. sinensis,* the chemical composition of commercial diets appears to meet the main requirements of turtles in terms of the CP and fat contents. In contrast to fresh components, the vitamin levels of commercial diets may be partially decreased relative to the declarations of the producers due to improper storage as well as processing, i.e., extrusion. Practically, the use of commercial feeds supplemented with natural components seems to be a good strategy for maintaining diet diversity in captivity (Mitchell and Tully, 2009), but from the nutritional point of view, this strategy leads to an unknown supply of energy and nutrients. In commercial diets, the main components are meals of animal origin, cereals and soybean meal supplemented with vitamin and minerals, and in the case of plant matter-based diets, the levels of essential amino acids may be not sufficient for strict carnivores. Moreover, the phosphorus in plant matter, and in cereals in particular, is bound in a phytate form, which is unavailable to monogastric animals (Pen et al., 1993). When plant matter is used as an animal protein replacement in diets for mainly carnivorous turtles, Ca, Mg and P supplementation or the use of endogenous phytase may be needed, as was shown in *P. sinensis* (Chen et al., 2014). If commercial diets are offered, the trends of lower CP requirements and ontogenetic diet shifts during different life stages are often not considered, but several commercial diets are now available that provide formulations for hatchlings, growing turtles and adults that contain approximately 40, 35 and 20% CP, respectively (Table 5).

Consequences of improper nutritional practices

In the available literature we can find frequent reports on diet related and metabolic disorders in turtles. Most of them deal with nutritional metabolic bone disease and hypovitaminosis A which are well discussed (Mader, 2005; Boyer, 2006; Donoghue, 2006; Mans, 2013; Mans and Braun, 2014). However in the case of captive turtles issues which are not directly caused by inaccuracy of diet composition seem to be neglected. They involve environmental factors, form and amount of feed as well as feeding frequency which all together affect metabolism and feed acceptance. Most of diet and environmental-related issues are caused by lack of knowledge. In some cases turtle keepers are not able to identify whether the animal is turtle or tortoise and keep freshwater species with no or limited access to water which results in dehydration and malnutrition (Köbölkuti et al., 2016).

Stress, improper environmental conditions and diet form

Captive reptiles are subjected to many stressors, however, this is frequently ignored due to common opinion that they are less likely to be negatively affected by them than higher vertebrates. The time to response after stressors including improper environmental conditions vary from several minutes to even weeks after the factor occurs (Silvestre, 2014). A very frequent stress-related issue in turtles is anorexia which may be caused by hypothermia, diseases, injuries, chronic pain or harassment. Particularly prone to anorexia are hatchlings, wild-caught individuals and animals in short period after transfer between facilities. They frequently refuse to ingest food for a long period of time due to poor acclimatization to captivity. Hatchlings may not accept the food until full yolk sack resorption, in adults food refusal up to two weeks after the transfer may be interpreted as normal. In their case, longer periods of starving should be interpreted as a sign of illness or improper environmental conditions. For young and wild-caught animals anorexia may be also caused by improper form of the diet – they may not accept pelleted feeds, and prefer live prey. In adult females 2–4 weeks of decreased feed intake coincidental with increased locomotory activity may be a symptom of egg development and physically decreased capacity of the gastrointestinal tract. In the above case egg binding occurrence should be excluded. Authors' observations suggest that in newly settled turtles presence of the hiding areas, constant temperature in preferred optimal temperature zone, single animal enclosures and 24/24h of light photoperiod supports acclimatization. In turtle enclosure gradient temperature areas should be present to avoid hypothermia or heat stress and allow the animal for selection of its preferred temperature – optimal for metabolism in the moment. It should be underlined that too low temperatures negatively affect energy and nutrient assimilation efficiency, feed intake as well as digestive turnover rates, as it is shown in Figure 1 (Kepenis and McManus, 1974; Parmenter, 1981) Too high temperatures and heat stress when no possibility of cooling is given may also cause anorexia. Long-term nutritional deficiencies such as insufficient energy, CP, minerals or vitamin intake may lead to cachexia. When it is of nutritional origin, improper environmental conditions like low temperatures and underlying disease also contribute to development of cachexia. Treatment of both anorexia and cachexia should focus on an increase in diet energy content and optimizing environmental conditions. The type of food offered should be re-considered for cachexic wild-caught turtles which usually prefer live prey. Suboptimal environmental conditions and protein deficiency may not only reduce growth performance, but may also lead to regression in shell development in hatchlings (Gibbons, 1990). However, turtles have the ability of growth compensation, if dietary imbalances are corrected (Xie et al., 2012). To avoid hypophosphatemia and hypocalcaemia, re-feeding of cachexic animals should not be too rapid, and energy level should be increased by 10–50% only when the animal shows improvement during treatment (Mader, 2005). All together, above-mentioned issues may cause growth depression which is frequent, however, non-specific symptom of improper nutrition. It is present in most cases of non-balanced diet in young turtles.

Stereotypic-like nutritional behavior

In many cases, reptile keepers use one or a few kinds of feed for many years without diversifying the diet of captive turtles. It may be the reason not only for development of metabolic disorders, but may also cause stereotypical-like behavior when animals do not accept other kinds of components than those they were fed for years. Turtles will imprint on food and prefer a diet that they are used to, instead of newly introduced feeds (Burghardt and Hess, 1966). If new food items are not accepted, then the most effective seems to be the use of live feeds – fish, shrimps, bloodworms or other insects larvae, and small vertebrates – to enrich turtle diet and stimulate feed intake.

Obesity

Another issue in the case of captive animals is positive energy balance – higher intake than expenditures of metabolic energy may accelerate growth in young animals and have positive effects if there are no deficiencies in the diet. However, it may lead to obesity in adults, which is defined as an accumulation of excessive amounts of adipose tissue in the body (Hand et al., 2000). More prone to obesity are species that are sedentary "bottom walkers" like snapping turtles (*Chelydra* spp.) or musk turtles (*Sternotherus* spp.) and African sidenecks (*Pelomedusa* spp. and *Pelusios* spp.). The best method of obesity prevention is regular body condition score (BCS) monitoring (Rawski and Józefiak, 2014). In chelonians, BCS is assessed mainly on the basis of comparison of straight carapace length and BW. However, additional visual assessment should be performed, and conditions, which may mimic obesity excluded (Jackson, 1980; Willemsen and Hailey; 2002; Rawski and Józefiak, 2014). Obese turtles store adipose tissue mainly in the coelom and internal organs, which may severely impair their function (Divers and Cooper, 2000). According to screening in *Pelomedusa* spp. and *Pelusios* spp. up to 22% of captive turtles may be overweight and obese (Rawski and Józefiak, 2014). Treatment of obesity should focus on restriction of energy intake. It should be lowered progressively to no less than 60% of usual intake. The body weight loss in that case should not exceed 0.5 to 1% weekly (Mader, 2005). According to practical experience of the authors feeding regime in terms of frequency of feeding may be one of the most effective in obesity prevention. It may be suggested that hatchlings should receive feed 6 times per week, animals between 6 months to 2 years of age 3-4 times per week and older ones 1 or 2 times per week. Additionally, turtles should undergo seasonal environmental stimulation for breeding; lipogenesis which occurs as a part of preparation for folliculogenesis is an important obesity-preventing factor in reptiles (Divers and Cooper, 2000).

Conclusions

Despite the recent increase in our scientific knowledge of turtle nutrition, the statement by Kollias and Gentz from 1996 that "reptile feeding is an art not a science" may still be partially valid. Due to the lack of suitable nutritional guidelines for most turtle species, observations of animal development as well as the experience and knowledge of keepers are still the most important sources of information for feeding freshwater turtles. Diet composition should be verified through long-term experiments, including digestibility studies at each turtle life stage. However, the basic rules for reptilian nutrition may be stated as follows:

1. The diversification of feed sources and their similarity to natural diet components are key to achieving sustainable growth and shell mineralization.

2. Commercial diets, if properly used without overfeeding, provide appropriate nutrition for turtles in captivity. However, in many cases, their formulation should be more specifically tailored to the needs of turtles with similar natural diets.

3. The exact nutritional requirements of turtles are still largely unknown, but large-scale farming and scientific experiments provide an opportunity to gain important knowledge that is applicable to this problem.

References

Acker R.F., Hartman P.A., Pemberton J.R., Quinn L.Y. (1959). The nutritional potential of poultry offal. Poultry Sci., 38: 706–711.

Bernard J.B., Allen M.E., Ullrey D.E. (1997). Feeding captive insectivorous animals: nutritional aspects of insects as food. Nutrition Advisory Group Handbook, Fact Sheet, 3: 1–7.

Bjorndal K.A. (1991). Diet mixing: nonadditive interactions of diet items in an omnivorous freshwater turtle. Ecology, 72: 1234–1241.

Bjorndal K.A., Bolten A.B. (1990). Digestive processing in a herbivorous freshwater turtle: consequences of small-intestine fermentation. Physiol. Zool., 63: 1232–1247.

Bonin F., Devaux B., Dupré A. (2006). Turtles of the World. Baltimore, USA, Johns Hopkins University Press, 416 pp.

Bouchard S.S. (2004). Diet selection in the yellow-bellied slider turtle, *Trachemys sripta*: ontogenetic diet shifts and associative effects between animal and plant diet items. Doctoral dissertation, University of Florida.

Bouchard S.S., Bjorndal K.A. (2005). Microbial fermentation in juvenile and adult pond slider turtles, *Trachemys scripta*. J. Herpetol., 39: 321–324.

Bouchard S.S., Bjorndal K.A. (2006). Ontogenetic diet shifts and digestive constraints in the omnivorous freshwater turtle *Trachemys scripta*. Physiol. Biochem. Zool., 79: 150–158.

Boyer T.H. (2006). Hypovitaminosis A and hypervitaminosis A. Reptile Medicine and Surgery, D.R. Mader (ed). St. Louis, MO, Elsevier: pp. 831–835.

Brown G.P., Brooks R.J., Layfield J.A. (1990). Radiotelemetry of body temperatures of free-ranging snapping turtles (*Chelydra serpentina*) during summer. Can. J. Zool., 68: 1659–1663.

Burghardt G.M., Hess E.H. (1966). Food imprinting in the snapping turtle, *Chelydra serpentina*. Science, 151: 108–109.

Bury R.B., Nebeker A.V., Adams M.J. (2000). Response of hatchling and yearling turtles to thermal gradients: comparison of *Chelydra serpentina* and *Trachemys scripta*. J. Therm. Biol., 25: 221–225.

Chen L.P., Huang C.H. (2011). Effects of dietary β-carotene levels on growth and liver vitamin A concentrations of the soft-shelled turtle, *Pelodiscus sinensis* (Wiegmann). Aquac. Res., 42: 1848–1854.

Chen L.P., Huang C.H. (2014). Estimation of dietary vitamin A requirement of juvenile soft-shelled turtle, *Pelodiscus sinensis*. Aquacult. Nutr., 21: 457–463.

Chen C.Y., Chen S.M., Huang C.H. (2014). Dietary magnesium requirement of soft-shelled turtles, *Pelodiscus sinensis*, fed diets containing exogenous phytate. Aquaculture, 432: 80–84.

Chu J.H., Chen S.M., Huang C.H. (2007). Effect of dietary iron concentrations on growth, hematological parameters, and lipid peroxidation of soft-shelled turtles, *Pelodiscus sinensis*. Aquaculture, 269: 532–537.

Clum N.J., Fitzpatrick M.P., Dierenfeld E.S. (1996). Effects of diet on nutritional content of whole vertebrate prey. Zoo Biol., 15: 525–537.

Costa Araújo J. da, Vieira P., Palha M., Rodrigues P.B., de Freitas R.T., da Silva A. (2013). Effect of three feeding management systems on some reproductive parameters of Scorpion mud turtles (*Kinosternon scorpioides*) in Brazil. Trop. Anim. Health. Pro., 45: 729–735.

Crissey S.D., Slifka K.A., Lintzenich B.A. (1999). Whole body cholesterol, fat, and fatty acid concentrations of mice (*Mus domesticus*) used as a food source. J. Zoo Wildlife Med., 30: 222–227.

Dierenfeld E.S., Alcorn H.L., Jacobsen K.L. (2002). Nutrient composition of whole vertebrate prey (excluding fish) fed in zoos. US Department of Agriculture, Agricultural Research Service, National Agricultural Library, Animal Welfare Information Center, 20 pp.

Divers S.J., Cooper J.E. (2000). Reptile hepatic lipidosis. Seminars in Avian and Exotic Pet Medicine, Elsevier.

Donoghue S. (2006). Nutrition. In: Reptile Medicine and Surgery, Mader D.R., Divers S. (eds). Elsevier, pp. 251–298.

Douglas T.C., Pennine M., Dierenfeld E.S. (1994). Vitamins E and A, and proximate composition of whole mice and rats used as feed. Comp. Biochem. Phys. A, 107: 419–424.

Ei S.N., Kavas A. (1996). Determination of protein quality of rainbow trout (*Salmo irideus*) by *in vitro* protein digestibility-corrected amino acid score (PDCAAS). Food Chem., 55: 221–223.

Ernst C.H. (1986). Environmental temperatures and activities in the wood turtle, *Clemmys insculpta*. J. Herpetol., 20: 222–229.

Feltz J., Tamplin J. (2007). Effect of substrate on selected temperature in juvenile spiny softshell turtles (*Apalone spinifera*). Chelonian Conserv. Biol., 6: 177–184.

Finke M.D. (2003). Gut loading to enhance the nutrient content of insects as food for reptiles: a mathematical approach. Zoo Biol., 22: 147–162.

Fitzgerald L.A., Nelson R.E. (2011). Thermal biology and temperature-based habitat selection in a large aquatic ectotherm, the alligator snapping turtle, *Macroclemys temminckii*. J. Therm. Biol., 36: 160–166.

Gatten R.E. Jr (1974). Effect of nutritional status on the preferred body temperature of the turtles *Pseudemys scripta* and *Terrapene ornata*. Copeia, 1974: 912–917.

Gibbons J.W. (1990). Editor. Life history and ecology of the slider turtle. Washington D.C., USA, Smithsonian Institution Press, 368 pp.

Grayson K.L., Dorcas M.E. (2004). Seasonal temperature variation in the painted turtle (*Chrysemys picta*). Herpetologica, 60: 325–336.

Hand M.S., Thatcher C.D., Remillard R.L., Roudebush P., Novtony B.J. (2000). Editors. Small Animal Clinical Nutrition. Mark Morris Institute, USA, 1192 pp.

Hoby S., Wenker C., Robert N., Jermann T., Hartnack S., Segner H., Aebischer C.P., Liesegang A. (2010). Nutritional metabolic bone disease in juvenile veiled chameleons (*Chamaeleo calyptratus*) and its prevention. J. Nutr., 140: 1923–1931.

Hou J., Jia Y., Yang Z., Li Y., Cheng F., Li D., Ji F. (2013). Effects of taurine supplementation on growth performance and antioxidative capacity of Chinese soft-shelled turtles, *Pelodiscus sinensis*, fed a diet of low fish meal content. J. World Aquac. Soc., 44: 786–794.

Huang C.H., Lin W.Y. (2002). Estimation of optimal dietary methionine requirement for softshell turtle, *Pelodiscus sinensis*. Aquaculture, 207: 281–287.

Huang C.H., Lin W.Y. (2004). Effects of dietary vitamin E level on growth and tissue lipid peroxidation of soft-shelled turtle, *Pelodiscus sinensis* (Wiegmann). Aquacult. Res., 35: 948–954.

Huang C.H., Lin W.Y., Wu S.M. (2003). Effect of dietary calcium and phosphorus supplementation in fish meal-based diets on the growth of soft-shelled turtle *Pelodiscus sinensis* (Wiegmann). Aquacult. Res., 34: 843–848.

Huang C.H., Lin W.Y., Chu J.H. (2005). Dietary lipid level influences fatty acid profiles, tissue composition, and lipid peroxidation of soft-shelled turtle, *Pelodiscus sinensis*. Comp. Biochem. Physiol., Part A Mol. Integr. Physiol., 142: 383–388.

Huang S.C., Chen S.M., Huang C.H. (2010). Effects of dietary zinc levels on growth, serum zinc, haematological parameters and tissue trace elements of soft-shelled turtles, *Pelodiscus sinensis*. Aquac. Nutr., 16: 284–289.

Jackson O. (1980). Weight and measurement data on tortoises (*Testudo graeca* and *Testudo hermanni*) and their relationship to health. J. Small Anim. Pract., 21: 409–416.

Jia Y., Yang Z., Hao Y., Gao Y. (2005). Effects of animal–plant protein ratio in extruded and expanded diets on nitrogen and energy budgets of juvenile Chinese soft-shelled turtle (*Pelodiscus sinensis* Wiegmann). Aquacul. Res., 36: 61–68.

Jianwei S., Fanwei M., Yongpu Zhang W.D. (2013). Field body temperature and thermal preference of the big-headed turtle *Platysternon megacephalum*. Curr. Zool., 59: 626–632.

Józefiak D., Józefiak A., Kierończyk B., Rawski M., Świątkiewicz S., Długosz J., Engberg R.M. (2016). Insects – a natural nutrient source for poultry – a review. Ann. Anim. Sci., 16: 297–313.

Julian R. (1998). Rapid growth problems: ascites and skeletal deformities in broilers. Poultry Sci., 77: 1773–1780.

Kennett R., Tory O. (1996). Diet of two freshwater turtles, *Chelodina rugosa* and *Elseya dentata* (Testudines: *Chelidae*) from the wet-dry tropics of northern Australia. Copeia, 1996: 409–419.

Kepenis V., McManus J.J. (1974). Bioenergetics of young painted turtles, *Chrysemys picta*. Comp. Biochem. Phys. A: Physiology, 48: 309–317.

Köbölkuti L., et al. (2016). Effects of malnutrition and improper captive maintenance on European pond turtle (*Emys orbicularis*): a case report. J. Anim. Plant Sci., 26: 874–879.

Lawler D.F., Evans R.H., Larson B.T., Spitznagel E.L., Ellersieck M.R., Kealy R.D. (2005). Influence of lifetime food restriction on causes, time, and predictors of death in dogs. J. Am. Vet. Med. Assoc., 226: 225–231.

Legler J.M. (1960). Natural history of the ornate box turtle, *Terrapene ornata ornata* Agassiz. Kansas, USA, University of Kansas Publications, Museum of Natural History, 157 pp.

Lei G., Yaohong Z. (2010). Effects of *Bacillus subtilis* on growth performance, digestive enzyme activities and blood biochemical indices of *Pelodiscus sinensis*. Chinese J. Anim. Nutr., 1: 42.

Lei S. (2006). Effects of ration level and feeding frequency on digestibility in juvenile soft-shelled turtle, *Pelodiscus sinensis*. J. Zhejiang Univ. Sci. B., 7: 580–585.

Lemell P., Beisser C.J., Gumpenberger M., Snelderwaard P., Gemel R., Weisgram J. (2010). The feeding apparatus of *Chelus fimbriatus* (Pleurodira; Chelidae) – adaptation perfected? Amphib-Reptil., 31: 97–107.

Lin W.Y., Huang C.H. (2007). Fatty acid composition and lipid peroxidation of soft-shelled turtle, *Pelodiscus sinensis*, fed different dietary lipid sources. Comp. Biochem. Phys. C., 144: 327–333.

Luiselli L., Akani G.C., Ebere N., Rugiero L., Vignoli L., Angelici F.M., Eniang E.A., Behangana M. (2011). Food habits of a pelomedusid turtle, *Pelomedusa subrufa*, in tropical Africa (Nigeria): The effects of sex, body size, season, and site. Chelonian Conserv. Bi., 10: 138–144.

Mader D.R. (2005). Editor. Reptile Medicine and Surgery. St. Louis, USA, Elsevier Health Sciences, 1241 pp.

Mans C. (2013). Clinical update on diagnosis and management of disorders of the digestive system of reptiles. J. Exotic Pet Med., 22: 141–162.

Mans C., Braun J. (2014). Update on common nutritional disorders of captive reptiles. Vet. Clin. North Am. Exot. Anim. Pract., 17: 369–395.

McArthur S., Wilkinson R., Meyer J. (2008). Editors. Medicine and Surgery of Tortoises and Turtles. Oxford, UK, Wiley-Blackwell, 600 pp.

McCauley S.J., Bjorndal K.A. (1999). Response to dietary dilution in an omnivorous freshwater turtle: implications for ontogenetic dietary shifts. Physiol. Biochem. Zool., 72: 101–108.

McDowell L.R. (1989). Vitamins in animal nutrition: comparative aspects to human nutrition. San Diego, USA, Academic Press, Inc., 496 pp.

McWilliams D. (2005). Nutrition research on calcium homeostasis. II. Freshwater turtles (with recommendations). Int. Zoo Yearbook, 39: 77–85.

Mitchell M.A., Tully T.N. (2009). Manual of exotic pet practice. St. Louis, USA, Elsevier Health Sciences, 546 pp.

Nebeker A.V., Bury R.B. (2000). Temperature selection by hatchling and yearling Florida red-bellied turtles (*Pseudemys nelsoni*) in thermal gradients. J. Herpetol., 34: 465–469.

Nuangsaeng B., Boonyaratapalin M. (2001). Protein requirement of juvenile soft-shelled turtle *Trionyx sinensis* Wiegmann. Aquacult. Res., 32: 106–111.

Ottonello D., Salvidio S., Rosecchi E. (2005). Feeding habits of the European pond terrapin *Emys orbicularis* in Camargue (Rhône delta, Southern France). Amphib-Reptil., 26: 562.

Pan Z., Zhang Y., Ji X. (2002). Diel variation in body temperature, thermal tolerance, and thermal dependence of locomotor performance in hatchling Chinese striped-necked turtles (*Ocadia sinensis*). Acta Zool. Sinica, 49: 45–52.

Parmenter R.R. (1981). Digestive turnover rates in freshwater turtles: the influence of temperature and body size. Comp. Biochem. Physiol. A: Physiology, 70: 235–238.

Pen J., Verwoerd T.C., van Paridon P.A., Beudeker R.F., van den Elzen P.J.M., Geerse K., van der Klis J.D., Versteegh H.A.J, van Ooyen A.J.J., Hoekema A. (1993). Phytase-containing transgenic seeds as a novel feed additive for improved phosphorus utilization. Nat. Biotech., 11: 811–814.

Rahman M.S., Sharma D.K. (2014). Morphometric, anatomical and histological features of gastrointestinal tract (GIT) of freshwater turtle, *Pangshura tentoria*. Int. J. Sci. Eng. Res., 7: 90–94.

Raila J., Schuhmacher A., Gropp J., Schweigert F.J. (2002). Selective absorption of carotenoids in the common green iguana (*Iguana iguana*). Comp. Biochem. Phys. A., 132: 513–518.

Ramos-Elorduy J., González E.A., Hernández A.R., Pino J.M. (2002). Use of *Tenebrio molitor* (Coleoptera: Tenebrionidae) to recycle organic wastes and as feed for broiler chickens. J. Econ. Entomol., 95: 214–220.

Rawski M., Józefiak D. (2014). Body condition scoring and obesity in captive African side-neck turtles (*Pelomedusidae*). Ann. Anim. Sci., 14: 573–584.

Rawski M., Kierończyk B., Długosz J., Świątkiewicz S., Józefiak D. (2016). Dietary probiotics affect gastrointestinal microbiota, histological structure and shell mineralization in turtles. PLOS ONE, 11(2): e0147859.

Rhodin A., Ibarrondo B., Kuchling G. (2008). *Chelodina mccordi* Rhodin 1994 – Roti Island snake-necked turtle, McCord's snake-necked turtle, kura-kura rote. Chelonian Conserv. Bi., 5: 001–008.

Seebacher F., Sparrow J., Thompson M.B. (2004). Turtles (*Chelodina longicollis*) regulate muscle metabolic enzyme activity in response to seasonal variation in body temperature. J. Comp. Physiol. B., 174: 205–210.

Silvestre A.M. (2014). How to assess stress in reptiles. J. Exotic Pet Med., 23: 240–243.

Sivaperumal P., Sankar T.V., Viswanathan Nair P.G. (2007). Heavy metal concentrations in fish, shellfish and fish products from internal markets of India vis-a-vis international standards. Food Chem., 102: 612–620.

Spencer R.J., Thompson M.B., Hume I.D. (1998). The diet and digestive energetics of an Australian short-necked turtle, *Emydura macquarii*. Comp. Biochem. Phys. A., 121: 341–349.

Stevens C.E., Hume I.D. (1998). Contributions of microbes in vertebrate gastrointestinal tract to production and conservation of nutrients. Physiol. Rev., 78: 393–427.

Sun J.Y., Du J., Qian L.C., Jing M.Y., Weng X.Y. (2007). Distribution and characteristics of endogenous digestive enzymes in the red-eared slider turtle, *Trachemys scripta elegans*. Comp. Biochem. Phys. A., 147: 1125–1129.

Sun P., Xu X., Chen H., Ji X. (2002). Thermal tolerance, diel variation of body temperature, and thermal dependence of locomotor performance of hatchling soft-shelled turtles, *Trionyx sinensis*. J. Appl. Ecol., 13: 1161–1165.

Tacon A.G., Metian M. (2013). Fish matters: importance of aquatic foods in human nutrition and global food supply. Rev. Fish. Sci., 21: 22–38.

Tamplin J. (2009). Effect of age and body size on selected temperature by juvenile wood turtles (*Glyptemys insculpta*). J. Therm. Biol., 34: 41–48.

Tamplin J.W., Cyr A.B. (2011). Effects of acclimation and egg-incubation temperature on selected temperature by hatchling western painted turtles (*Chrysemys picta bellii*). J. Therm. Biol., 36: 507–514.

Willemsen R.E., Hailey A. (2002). Body mass condition in Greek tortoises: regional and interspecific variation. Herpetol. J., 12: 105–114.

Wu G.S., Huang C.H. (2008). Estimation of dietary copper requirement of juvenile soft-shelled turtles, *Pelodiscus sinensis*. Aquaculture, 280: 206–210.

Wurth S.M.A., Musacchia X.J. (1964). Renewal of intestinal epithelium in the freshwater turtle, *Chrysemys picta*. Anat. Rec., 148: 427–439.

Xie Q.S., Yang Z.C., Li J.W., Li Y.J. (2012). Effect of protein restriction with subsequent re-alimentation on compensatory growth of juvenile soft-shelled turtles (*Pelodiscus sinensis*). Aquacult. Int., 20: 19–27.

Zhang X., Peng L., Wang Y., Liang Q., Deng B., Li W., Fu L., Yu D., Shen W., Wang Z. (2014). Effect of dietary supplementation of probiotic on performance and intestinal microflora of Chinese soft-shelled turtle (*Trionyx sinensis*). Aquacult. Nutr., 20: 667–674.

Zhang Y.P., Du W.G., Shen J.W., Shu L. (2009). Low optimal temperatures for food conversion and growth in the big-headed turtle, *Platysternon megacephalum*. Aquaculture, 295: 106–109.

Zhou F., Ding X.Y., Feng H., Xu Y.B., Xue H.L,, Zhang J.R., Ng W.K. (2013). The dietary protein requirement of a new Japanese strain of juvenile Chinese soft shell turtle, *Pelodiscus sinensis*. Aquaculture, 412: 74–80.

Zhou X., Niu C., Sun R., Li Q. (2002). The effect of vitamin C on the non-specific immune response of the juvenile soft-shelled turtle (*Trionyx sinensis*). Biochem. Phys. A., 131: 917–922.

Received: 30 V 2017
Accepted: 23 VIII 2017