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Nutritional effect of feeding enrichment using bamboo *Pleioblastus* spp. in zoo-kept Asian elephants *Elephas maximus*

Yuma Tsuchiya[1], Masato Yayota[1,2], Yukari Kashima[1] and Yukihiro Shiota[3]

[1]Faculty of Applied Biological Sciences, Gifu University, Gifu 501-1193, Japan.
[2]Zoo Biology Research Center, Gifu University, Gifu 501-1193, Japan.
[3]Kyoto City Zoo, Kyoto 606-8333, Japan.

Correspondence: Masato Yayota, email; yayo@gifu-u.ac.jp

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Abstract

Many zoos use browse and other roughage as feed ingredients and enrichment tools for elephants. Amongst these are bamboo species (e.g. *Pleioblastus* spp.), which belong to the family of grasses. Bamboo is used in zoos worldwide as a dietary item for many herbivores. The fibrous attributes of bamboo are potentially beneficial in reducing diet digestibility and overnutrition in captive elephants. This study aimed to determine the effect of feeding bamboo on nutritional intake, digestibility and blood condition in Asian elephants *Elephas maximus*. Four Asian elephants aged 4 to 8 years, fed in two groups of two animals, received a conventional diet (CON) or a diet with bamboo (BAM; 4.5 kg bamboo/animal as fed). In CON, animals were fed sudangrass and timothy hay, rice straw, fresh Italian ryegrass, zoo pellets, carrots, sweet potatoes, steamed potatoes and apples. In BAM, a part of the sudangrass hay, accounting for approximately 20% of the diet on a dry matter basis, was replaced with bamboo *Pleioblastus* spp. Dry matter, crude protein, and neutral and acid detergent fibre digestibility were similar between the treatments. The concentrations of serum components, including total cholesterol, albumin, glucose, Ca and P, did not differ between the treatments and were almost within the range of previously reported values. These results suggest that feeding bamboo has no negative impacts on the nutritional status and health of captive Asian elephants.

Introduction

Asian elephants *Elephas maximus* ingest a variety of foods in the wild, including grasses, legumes, browse, fruit and roots (Baskaran et al. 2010; McKay 1973; Roy 2009; Sukumar 1990). Elephants inhabiting deciduous and dry thorn forests in southern India eat 59 browse and 29 grass species, although grass is predominant over browse in their annual diet (84.6% versus 15.4%, respectively; Baskaran et al. 2010). However, elephants in the tropical moist forests of West Bengal eat 111 plant species, and browse is the main component of their diet in the dry season (93%), while grass is predominant in the wet season (78%; Roy 2009).

In contrast, captive Asian elephants are generally provided a limited number of foods, such as hay, fruit, vegetables and commercial pellets. Of these, fruit, vegetables and commercial pellets contain higher energy and lower fibre levels than wild food sources (Baer et al. 2010). These nutritious diet compositions often lead to overnutrition, induce obesity (Ange et al. 2001; Morfeld et al. 2016; Schiffmann et al. 2018) and can cause health problems such as dental disorders and colic (Hatt and Clauss 2006). Thus, appropriate diet composition and amount should be considered for captive elephants.

Asian elephants eat substantial amounts of browse species in their wild habitat; thus, the addition of browse with twigs and branches to the captive diet is assumed to be useful for increasing the dietary fibre content (Hatt and Clauss 2006). Recently, many zoos have begun feeding browse and some other forage species as diet ingredients and enrichment tools, and several Asian countries have also provided browse and bamboo to their captive elephants (Das et al. 2015a; Vanitha et al. 2008).

Bamboos (e.g. *Phyllostachys* spp. and *Pleioblastus* spp.) are semi-woody, fast-growing plants widely distributed in Asian regions. Although they are in the grass Poaceae family, they are a comparable forage to browse species (Halvorson et al. 2011; Lefeuvre et al. 2020; Wood et al. 2020) and are used in zoos worldwide as a diet item for many herbivores (Finley et al. 2011; Smith et al. 2014; Wood et al. 2020). Bamboo is not recommended for animals susceptible to tooth wear, such as browsing ruminants (Kaiser et al. 2009), due to its high content of abrasive silica bodies (phytoliths) (Martin et al. 2019). This is not a concern in elephants. Wild Asian elephants ingest bamboo if it is available in their habitat (Baskaran et al. 2010; Joshi 2009), and captive elephants are fed bamboo in some Asian countries (Vanitha et al. 2008). Although the leaves of bamboo include similar or slightly less crude protein (CP) and similar or more fibre (Christian et al. 2015; Wang et al. 2007; Wood et al. 2020) than common forage, sheep (Yayota et al. 2009) and guinea pigs (De Cuyper et al. 2022) show lower dry matter (DM), CP and neutral detergent fibre digestibility of these feeds than common grasses. Moreover, the culms of bamboo contain considerably more fibre and less CP than the leaves (Christian et al. 2015; Dierenfeld et al. 1982), suggesting that feeding bamboo with culms has the potential to reduce feed digestibility and avoid overnutrition in captive Asian elephants.

Thus, it is hypothesised that feeding bamboo with culms has a beneficial effect on elephant nutrition by reducing energy and nutrient concentrations, as well as overall diet digestibility. The objective of this study was to determine the effect of replacing part of the sudangrass hay ration with bamboo on feed and nutrient intake, nutrient digestibility and blood variables in Asian elephants.

Materials and methods

All animal experimental procedures were approved by the Kyoto City Zoo (approved on 20 June 2016). Animal experimental procedures were conducted following the Guidelines for Proper Conduct of Animal Experiments (Science Council of Japan 2006) and the Guidelines of Animal Research and Welfare (Gifu University 2008).

Study site and animals

This study was conducted at Kyoto City Zoo, Kyoto, western Japan (135°47’ E, 35°00’ N; 53 m altitude). Four Asian elephants *Elephas maximus* (one male and three females, aged 4 to 8 years and mean body weight [BW] 1,496±192 kg) were used for this experiment. These elephants were born in captivity in Laos and transferred to the zoo in 2014. The elephants were kept as a group in an outside paddock (approximately 34 m × 17 m) with a pool (approximately 7.5 m × 13.5 m; Figure 1) during the daytime (0930–1530) and housed individually in indoor pens (12.2 m × 6.0–7.5 m) at night (1530–0930).

Experimental period and treatments

This experiment was conducted from 29 July to 25 September 2016, and the period was divided into four investigation periods (Table 1). Each period consisted of 13 days; the first 7 days were for acclimation to the experimental conditions, and the remaining 6 days were for sampling. Animals were allocated to conventional feeding (CON) or bamboo feeding (BAM) groups (Table 1). Animals were divided into two groups (A: 1 male and 1 female, and B: 2 females), and data were taken from only one group in each period because feeding trials of Asian elephants are laborious.

Nutrient content in both diets was designed to satisfy the recommendations of the Nutrition Advisory Group (Ullrey et al. 1997). In the CON treatment, animals were fed sudangrass hay, timothy hay, rice straw, fresh Italian ryegrass, zoo pellets (Elephant supplement 5666, Mazuri, Missouri, USA), carrots, sweet potatoes, steamed potatoes and apples (Table 2). Sudangrass hay (2.3 kg DM/day/animal) was placed at the outside paddock at 1300, and the remaining diet components were placed in each indoor pen at 1600. In the BAM treatment, a portion of the sudangrass hay was replaced with bamboo *Pleioblastus* spp., so that the proportion of bamboo in the diet was approximately 20% on a DM basis. As the bamboo contained approximately twofold higher CP than sudangrass hay, the amount of zoo pellet was reduced in the BAM condition to keep the same CP content in the whole diet. All bamboo (2.8 kg DM/day/animal) was placed on the walls and posts at approximately 0930 (before the exhibition opened), and the sudangrass hay (0.9 kg DM/day/animal) was placed in the outside paddock at 1300. The remaining diets, as well as the CON treatment, were placed in indoor pens at 1600. The authors are aware that this diet does not comply with the recommendation to ensure constant access to low-energy diet items such as straw or branches for elephants; this was, however, necessary for the logistics of the experiment. It was regularly confirmed that this diet did not impact elephant health. Animals had free access to water and mineral blocks (Powerblock, Feedone Co., Ltd., Kanagawa, Japan) in indoor pens and outside paddocks. As sudangrass hay was not provided individually in the outside paddock, all animals (within groups A or B) were allocated to the same treatment in each period.

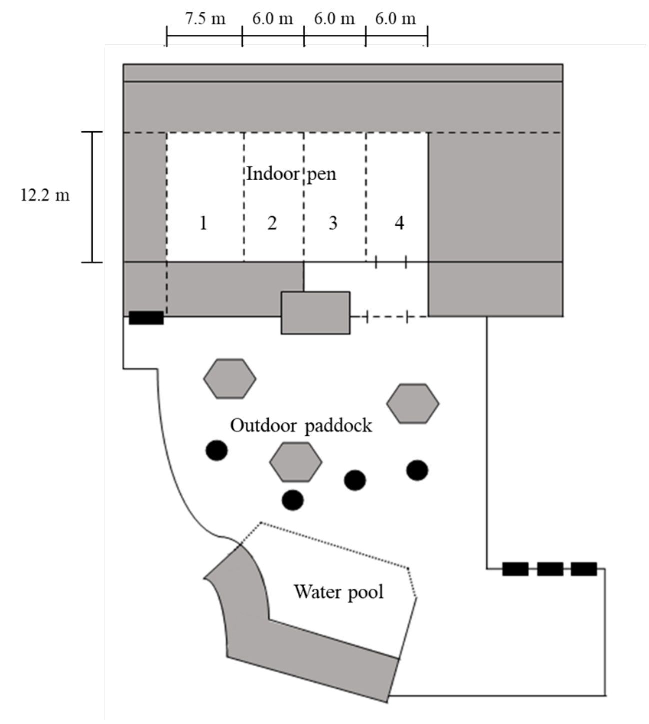

Figure 1. Asian elephant facility at Kyoto City Zoo. Sudangrass hay was placed at the outside paddock at 1:00 pm (●), and bamboo was placed on the walls and posts (squares) at approximately 9:30 (before the exhibition). Elephants did not enter the grey areas. The sizes of the outdoor paddock and water pool were approximately 34.0 m × 17.0 m and 7.5 m × 13.5 m, respectively.

Table 1. Elephants used in this study, experimental layout and sampling schedule. [a]Age of the elephants at the start of the experiment. [b]Body weight on 12 Sep 2016.

Group	Animal	Sex	Age (year)[a]	Body weight (kg)[b]	Metabolic body weight (kg$^{0.75}$)	Treatment and sampling period	
						Conventional feeding group	Bamboo feeding group
A	1	Male	4	1195	203	First period	Third period
	2	Female	6	1390	228	5-10 Aug	5-10 Sep
B	3	Female	5	1340	221	Fourth period	Second period
	4	Female	8	2060	306	20-25 Sep	22-27 Aug

During the sampling period, the leftovers in the indoor pens were collected at approximately 1000 after the elephants were put in the outside paddock for exhibition, and the leftovers in the outside paddock were collected at approximately 1630 after the elephants were housed in the indoor pens. The leftovers were separated by feed ingredients and then weighed and sampled to determine the dry weight; note, however, that the amounts of leftovers were small and mainly composed of bamboo and fresh grass.

Sample collection

A 100 g sample of each feed, except for carrot, sweet potato, steamed potato and bamboo, was taken every day during the sampling period. Approximately 200 g of carrot, sweet potato and steamed potato was taken, and two branches of bamboo were also collected every day during the sampling period. The carrot, sweet potato, steamed potato and apple samples were kept in a freezer (–20°C), and the other feed samples were kept in a refrigerator (4°C) until the end of each experimental period. At the end of each experimental period, feed samples were mixed

Table 2. Ingredient composition of the experimental diet. Mean with (standard deviation). FM: fresh matter, DM: dry matter, DE: digestible energy, CP: crude protein, aNDFom: α-amylase-treated neutral detergent fibre without residual ash, ADFom: acid detergent fibre without residual ash. [a]Bamboo: *Pleioblastus* spp., [b]Zoo pellet: Elephant supplement 5666, MAZURI. [c]The DE content in the diets was derived from the Nutrient Requirements of Horses (NRC 1989) and the Japanese Feeding Standard for Horses (Equine Research Institute, Japan Racing Association 2004). The DE content in zoo pellets was derived from the website of the supplier (https://info.mazuri.com/BusinessLink/media/Mazuri/ProductSheet/5666.pdf). Values of CP, aNDFom and ADFom were from our own analysis.

Item	Conventional feeding group		Bamboo feeding group	
	kg FM/day/animal	kg DM/day/animal	kg FM/day/animal	kg DM/day/animal
Sudangrass hay	3.5 (0.4)	3.2 (0.4)	1.0 (0)	0.9 (0.0)
Bamboo[a]	–	–	4.5 (0.4)	2.9 (0.5)
Timothy hay	3.0 (1.4)	2.7 (1.3)	3.0 (1.4)	2.7 (1.2)
Italian rye-grass (fresh)	15.0 (4.1)	2.4 (0.6)	15.0 (4.1)	4.3 (1.2)
Rice straw	2.8 (0.5)	2.5 (0.5)	2.8 (0.5)	2.4 (0.5)
Zoo pellet[b]	3.3 (0.5)	2.8 (0.4)	2.3 (0.5)	1.9 (0.4)
Carrot	1.6 (0.3)	0.2 (0.0)	1.6 (0.3)	0.2 (0.0)
Sweet potato	0.6 (0.3)	0.2 (0.1)	0.6 (0.3)	0.3 (0.1)
Apples	1.6 (0.3)	0.2 (0.0)	1.6 (0.3)	0.2 (0.0)
Steamed potato	1.0 (0)	0.4 (0.0)	1.0 (0)	0.4 (0.0)
Total	32.4 (6.5)	14.6 (2.4)	33.4 (6.4)	16.2 (3.0)
DE (Mcal/kg)[c]		2.2		2.1
CP (%)		10.9		10.3
aNDFom (%)		45.7		50.5
ADFom (%)		26.6		28.6

and approximately 200 g of representative samples for each feed were collected. Apples and steamed potatoes were freeze-dried (DC400/800, Yamato Co., Ltd., Tokyo) after measuring their fresh weight. Fresh Italian ryegrass, fresh bamboo, carrots and sweet potatoes were oven dried at 60°C for 48 hr (DKM 600, Yamato Co., Ltd., Tokyo) after measuring their fresh weight. All dried samples were left at room temperature for 24 hr, weighed and ground to pass through a 1 mm screen using a Wiley mill and stored in plastic bags until chemical analysis.

All animals were group fed in the outside paddock. Thus, hard-husk seeds (maize and sunflower seeds) were provided as an indigestible marker to the sampled animals (two animals/period). The animals ate either of these seeds (maize or sunflowers). These seeds were not digested and were excreted directly in the faeces; thus, all faeces that were excreted by the animals selected for sampling could be identified. One hundred grams of the seeds were mixed with 500 g of steamed potatoes and rolled into a ball shape, and then a zoo keeper fed this food ball to the animals during morning and evening husbandry training.

All faeces of each animal were collected and weighed every day during the sampling periods. Faeces in the inside pens and the outside paddock were collected at approximately 1000 and 1630, respectively. Faeces were loosened by hand, marker seeds were removed to the extent possible, and faeces were well mixed using a small concrete mixer (AMZ-30Y, Alumis Co., Ltd., Saga) for 10 min; then, 4% of daily faeces weight was sampled and stored in a freezer (–20°C) until the end of each experimental period. All daily collected samples were composited and mixed thoroughly, and approximately 500 g of representative faecal sample was collected from each animal in each period. The representative samples were stored in a freezer (–20°C) and then oven dried at 60°C for 48 hr (DKM 600, Yamato Co., Ltd., Tokyo) after measuring their fresh weight. All dried samples were treated similarly to the feed samples.

Blood was taken via the ear vein of the elephants. The sampling was conducted once during each sampling period considering the health management schedule in the zoo; the sampling time was set at approximately 0900. The blood of one female was not taken because this elephant did not reach a satisfactory training status. Thus, three blood samples were collected in each treatment during the whole experimental period.

Feed intake, faecal output and nutrient digestibility

Feed intake in the indoor pen was determined by weighing the amount of feed offered and the leftovers the next day. The intake of sudangrass hay and bamboo in the outside paddock was estimated by calculating the feeding amount according to the eating time for each animal because the elephants fed in a group in the outside paddock. Eating time was recorded by direct observation using 2 min intervals and the instantaneous scan sampling method. Observation of the eating time was generally conducted on the second and last days of each sampling period. However, the observations in Period 4 were conducted on the third day due to poor weather conditions on the second day. The average eating time on two observation days was used to calculate intake in the outside paddock. Intake in the outside paddock was estimated as follows:

$$\text{Intake in the outside paddock (kg/animal)}$$

$$=(\text{Feed}-\text{Leftov})\times \text{Eat time}_i / \left(\sum_{(i=1)}^{n} \text{Eat time}_i\right)$$

where *Feed* is the amount of feed offered to all elephants, *Leftov* is the amount of leftovers of all elephants and $\textit{Eat time}_i$ is the eating time of the *i*th individual. Total feed intake was the sum of feed intake in the inside pen and the outside paddock.

Nutrient intake, including DM, CP, neutral detergent fibre assayed with heat stable α-amylase excluding residual ash (aNDFom) and acid detergent fibre excluding residual ash (ADFom), was estimated from the diet intake and nutrient content of the diet. Nutrient intake per metabolic body weight (MBW) was also calculated by dividing daily nutrient intake by MBW. The MBW in each treatment was estimated by calculating the MBW for each animal and then summing these values in the treatment (Clauss and Hummel 2017). Nutrient digestibility was estimated by daily nutrient intake and faecal nutrient output over the sampling period: Nutrient digestibility (%) = (Nutrient intake – Faecal nutrient output)/Nutrient intake × 100.

Chemical analysis

DM, crude ash (CA), crude protein and ADFom in feed and faeces were determined according to AOAC methods (AOAC 2007: protocol nos. 930.15, 942.05, 990.03 and 973.18, respectively). aNDFom was measured following Van Soest et al. (1991). For leftovers, DM content was analysed to determine the dry weight. The chemical composition of the feed ingredients is shown in Table 3.

The blood samples were incubated at room temperature until clotting and centrifuged at 1,000g and 4°C for 15 min to collect serum. Serum glucose (GLU), total cholesterol (TCHO), albumin (Alb), calcium (Ca) and phosphate (P) concentrations were measured using the dry chemistry method (Fuji Drychem NX500i, Fuji Film Co., Ltd., Tokyo).

Statistical analysis

Due to the small sample size (n=4 or n=3 in blood variables), a general linear (mixed) model that considers the (random) effect of period and individual elephants was not applied. Instead, feed intake, faecal output, diet digestibility and blood variables were analysed by paired sample t-tests to compare the simple treatment effects. All statistical procedures were performed using R version 3.6.3 (R Core Team 2020). The level of significance was set at $P<0.05$.

Results

The daily DM intake of Asian elephants in the BAM treatment (mean±standard deviation: 16.0±3.1 kg DM/day/animal) did not differ from that in the CON treatment (14.5±2.4 DM/day/animal; Table 4). The elephants in BAM ate 2.8 kg DM of bamboo per day. The slight numerical difference in the DM intake between the treatments was caused by the unexpected difference in the DM content of the fresh grass between the treatments (BAM: 28.6% and CON: 15.9%), as the same amount of fresh grass was provided on an as-fed basis. Daily CP, aNDFom and ADFom intake (kg/day/animal) also did not differ between the treatments (Table 4). Similarly, faecal output did not differ between the treatments (BAM: 7.1±2.1 and CON: 6.8±1.3kg DM/day/animal).

DM intake per MBW (g/day/$BW^{0.75}$) did not differ between the BAM condition and the CON condition (66.9 versus 60.6 g/day/$BW^{0.75}$). Fibre intake per MBW, including aNDFom and ADFom, was higher under BAM than CON (aNDFom: $P=0.038$; ADFom: $P=0.047$). However, CP intake per MBW did not differ between the treatments.

DM digestibility in the elephants was 56.5±5.8% under BAM and 53.2±1.2% under CON, and there was no significant difference between the treatments. Similarly, CP, aNDFom and ADFom digestibility did not differ between the treatments (Table 4).

Serum GLU, TCHO and Alb concentrations did not differ between the treatments (Table 5). There was no significant difference in serum Ca and P concentrations between the treatments.

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Table 3. Chemical composition of feed ingredients. DM: dry matter, CA: crude ash, CP: crude protein, aNDFom: α-amylase-treated neutral detergent fibre without residual ash, ADFom: acid detergent fibre without residual ash. [a]Bamboo: *Pleioblastus* spp. [b]Zoo pellet: Elephant supplement 5666, MAZURI.

	DM (%)	CA (% of DM)	CP (% of DM)	aNDFom (% of DM)	ADFom (% of DM)
Conventional feeding group					
Sudangrass hay	90.6	7.4	5.2	60.2	35.0
Bamboo[a]	–	–	–	–	–
Timothy hay	90.1	6.8	8.7	53.6	31.6
Italian rye-grass (fresh)	15.9	11.9	19.9	50.4	28.0
Rice straw	89.8	16.2	3.4	55.1	33.6
Zoo pellet[b]	86.0	10.3	21.1	23.4	12.4
Carrot	9.3	8.0	8.7	11.4	12.5
Sweet potato	33.7	2.8	2.2	4.9	3.2
Apples	13.1	3.0	1.5	11.0	8.6
Steamed potato	37.3	2.8	2.0	5.7	4.4
Bamboo feeding group					
Sudangrass hay	90.0	7.7	4.8	59.0	32.1
Bamboo[a]	64.1	5.8	10.7	79.2	45.2
Timothy hay	89.3	7.6	7.7	52.0	29.8
Italian rye-grass (fresh)	28.6	13.4	14.0	52.9	29.2
Rice straw	87.9	17.1	3.4	53.4	31.8
Zoo pellet[b]	86.5	10.2	20.4	24.1	12.4
Carrot	8.1	7.7	7.8	10.6	11.3
Sweet potato	41.3	3.3	2.1	4.3	3.3
Apples	13.3	4.7	1.3	9.1	6.7
Steamed potato	38.2	3.6	2.0	6.8	4.6

Table 4. Nutrient intake, faecal output and digestibility in Asian elephants. Mean with (standard deviation). DM: dry matter, DE, digestible energy, CP: crude protein, αNDFom: α-amylase-treated neutral detergent fibre without residual ash, ADFom: acid detergent fibre without residual ash.

Item	Conventional feeding group	Bamboo feeding group	t	P value
Intake per animal (kg DM/day/animal)				
DM	14.5 (2.4)	16.0 (3.1)	0.635	0.571
CP	1.6 (0.3)	1.7 (0.5)	0.232	0.831
aNDFom	6.6 (1.1)	8.1 (1.5)	1.253	0.299
ADFom	3.9 (0.6)	4.6 (0.8)	1.175	0.325
Intake per metabolic body weight (g DM/day/$BW^{0.75}$)				
DM	60.6 (2.6)	66.9 (4.0)	2.076	0.129
CP	6.6 (0.1)	6.8 (1.1)	0.495	0.655
aNDFom	27.7 (1.3)	33.7 (2.3)	3.535	0.038
ADFom	16.2 (0.8)	19.2 (1.2)	3.253	0.047
Faecal output (kg DM/day/animal)	6.8 (1.3)	7.1 (2.1)	0.167	0.878
Digestibility %				
DM	53.2 (1.2)	56.5 (5.8)	1.036	0.376
CP	72.5 (2.6)	73.9 (3.0)	0.839	0.463
aNDFom	51.2 (1.6)	56.0 (6.6)	1.270	0.294
ADFom	47.8 (0.9)	51.0 (7.1)	0.832	0.467

Table 5. Serum parameters of Asian elephants. Mean with (standard deviation).

Item	Conventional feeding group	Bamboo feeding group	t	P value
Glucose (mg/dl)	86.7 (10.1)	84.0 (5.2)	–0.543	0.641
Total cholesterol (mg/dl)	41.0 (6.0)	46.3 (5.5)	0.941	0.446
Albumin (g/dl)	3.9 (0.2)	4.2 (0.2)	1.299	0.324
Ca (mg/dl)	10.3 (0.4)	9.9 (0.4)	–2.600	0.122
P (mg/dl)	5.5 (0.5)	5.6 (0.3)	0.480	0.678

Discussion

Daily DM intake per animal was similar between the BAM and CON conditions. Even when considering the difference in individual MBW, DM intake was not different between the BAM and CON conditions. Thus, replacing hay with semi-woody bamboo did not affect feed intake in the elephants. The digestibility of DM, CP, aNDFom and ADFom did not differ between the treatments, suggesting that feeding bamboo at a proportion of approximately 20% of the total diet (DM basis) does not influence nutrient digestibility in captive elephants when the whole diet contains a similar amount of energy and similar protein and fibre content (compared to the CON diet). The DM and nutrient digestibility, including CP, aNDFom and ADFom, in the present study was approximately 10–20% higher than that in previous studies (Clauss et al. 2003; Das et al. 2015a, b; Katole et at. 2014). Clauss et al. (2003) reported DM, CP and ADFom digestibility of hay-based diets in zoo-housed Asian elephants of 34.4, 52.3 and 26.0%, respectively, although they used internal and external marker methods for estimating nutrient digestibility. They also suggested appreciably low digestibility in Asian elephants according to their summary of past literature data. Das et al. (2015a) showed that DM, CP and aNDFom digestibility in captive Asian elephants fed tree branches and forage with some concentrates was 39.6, 38.3 and 26.6%, respectively. Meissner et al. (1990) found a negative correlation between DM intake and digestibility in wild African elephants. In previous studies (Clauss et al. 2003; Das et al. 2015a, b; Katole et at. 2014), the DM intake of a captive elephant was over 100 g/kg $BW^{0.75}$/d, whereas that in the present study was under 80 g/kg $BW^{0.75}$/d. Some nutritious foods, such as fresh grass, zoo pellets, vegetables and fruit, were fed to the elephants due to the ease of purveyance, stability of supply and preference. This feeding regimen easily satisfies the nutritional requirements of elephants with a relatively small amount of feed offered. Thus, one of the possible explanations for the differences in feed digestibility between the current and past studies was probably due to the difference in feed intake. Moreover, nutritious feeds directly increase feed digestibility. Thus, the results of the current study suggest that feed digestibility in captive Asian elephants is influenced by the quality and quantity of captive feed. The digestibility of bamboo is lower than that of dried grass in sheep (Yayota et al. 2009), horses (Kawai et al. 1995) and guinea pigs (De Cuyper et al. 2022); thus, feeding bamboo at a proportion of approximately 20% in total diets was expected to reduce the digestibility of the captive diet. However, this treatment did not affect feed intake and digestibility in captive Asian elephants.

The concentrations of serum components, including GLU, TCHO, Alb, Ca and P, were similar between BAM and CON elephants. Since the diet regimen was designed to have similar nutrient content following the recommendations of Ullrey et al. (1997) and elephants showed similar feed intake and nutrient digestibility, replacing grass hay with bamboo did not affect serum components. In addition, the concentrations of serum components under both treatments were almost within the range of previously reported values in wild (Silva and Kuruwita 1993), semi-captive (Dos Santos et al. 2020) and captive (EAZA 2020; Fowler and Mikota 2006) elephants. Thus, feeding bamboo has no negative impact on the nutritional status and health of captive Asian elephants and can be used as a substitute for hay with some potential enrichment effects that increase foraging time with longer handling time, as shown in elephants eating browse species or bamboo (Lefeuvre et al. 2020; Saputra et al. 2022; Stoinski et al. 2000).

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