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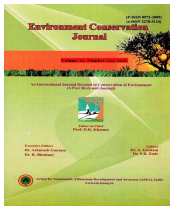

Efficacy of oral probiotics on morphometric measurements and their allometric relationships in Asian elephants

Dharmendra Chharang ✉
Department of Animal Nutrition, Post Graduate Institute of Veterinary Education and Research, Rajasthan University of Veterinary and Animal Sciences, Jaipur, Rajasthan, India.

Sheela Choudhary
Department of Animal Nutrition, Post Graduate Institute of Veterinary Education and Research, Rajasthan University of Veterinary and Animal Sciences, Jaipur, Rajasthan, India.

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ABSTRACT

An experiment was undertaken on 18 Asian elephants to study the effect of oral probiotics on body measurements for two months. Simultaneously, the efficacy of existing prediction equations and allometric relationship of heart girth-body weight (BW), height-forefoot circumference (FFC) and height-body weight were also observed. The animals were divided into three groups, with six each. The experimental probiotics; *Lactobacillus acidophilus* and *Saccharomyces cerevisiae,* were supplemented @ 1 gm 1 × 10^9 cfu/gm for every 50 kg BW/day to the elephants of LACTO (T_2) and SAC (T_3) groups, respectively, whereas no probiotic was given to the control group. Heart girth was measured four times, on days 0, 20, 50 and 60 of the experiment to determine BW. Other morphometric estimations, like length, height, hind girth, and FFC were documented once, at the end of study. The data of heart girth and body weight revealed non-significant effect of the treatment. Irrespective of probiotics treatment, allometric parameters such as heart girth-body weight and height-FFC showed an isometric relationship whereas, the height-body weight relationship wasn't found to yield an equivalent accuracy. The equations involving heart girth and FFC were observed to be most authentic to calculate BW and height, respectively.

Introduction

Several studies have revealed about microbiota's indispensable role in disease control, homeostasis and health promotion (Alayande *et al.,* 2020). Predominantly, dose and duration of the treatment as well as the microbial strains, are among the vital components impacting the competence of probiotics (De Cesare *et al.,* 2017). To the investigators' information, no such research trials have been performed on endangered elephants yet.

The body sizes usually vary in correlation with another related variant by way of exponential scaling, called as allometry (Anzai *et al.,* 2017). The aspect of allometric relationships could be profitably used in various ecological studies, which may involve ageing wild elephants or estimating biomass of the population. Precise computation of body weight (BW) is advantageous for the assessment of well-being, nutritional condition, feeding program, chemical immobilization and medication for the treatment (Kanchanapangka *et al.,* 2007). Nonetheless, it is impracticable to weigh enormous sized elephant due to tremendous BW. Weighing the earth' largest living animal is a challenging work, as it needs a distinctive training plan, proficient drivers and appropriate scales. Hence, the only approach to calculate their BW is by applying prediction equations based on definite body variables (Sukumar *et al.,* 1988; Hile *et al.,* 1997). Therefore, the study was conducted with the objectives to assess the effect of probiotics feeding on morphometric measurements and it also examined the efficacy of existing prediction equations as well as their allometric relationships in Asian elephants.

Corresponding author E-mail: *dchharang@gmail.com*
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Material and Methods

The experiment was organized with the prior permission of the Additional Principal Chief Conservator of Forest and Chief Wildlife Warden, Government of Rajasthan, Jaipur (India). The study protocol was duly approved by the Institute Animal Ethics Committee (PGIVER/IAEC/I9-05) and performed in accordance with relevant guidelines and regulations for care and management during the experiment (MoEF and CC, 2008).

Selection of experimental animals

Eighteen healthy, captive adult female Asian elephants with alike BW (3495 ± 133.34 kg) were divided into three groups of six elephants each. The average age composition in T_1, T_2 and T_3 groups was 44, 42.50 and 48 years, respectively. The group wise details of experimental elephants are given in Table 2. All the elephants were housed in a hygienic and well ventilated individual enclosure, with a separate feeding arrangement.

Experimental feeding

The experiment was planned for 60 days, in which, ten days adaptation period was observed and then elephants were placed on three dietary experimental feeds for 50 days of digestibility trial. During the digestibility trial, experimental probiotics *Lactobacillus acidophilus* and *Saccharomyces cerevisiae* were administered @ 1 gm 1×10^9 cfu /gm for every 50 kg BW per day orally along with basal feed to all the experimental elephants of LACTO (T_2) and SAC (T_3) groups, respectively. The group T_1 was CONT group (control) received no probiotic.

Morphometric measurements

Measurements of the heart girth were recorded randomly on days 0, 20, 50, and 60 of the experiment to estimate body weight (Figure 1). Other morphometric estimations, like length, height, hind girth, and FFC were recorded at the end of the experiment before feeding and watering. The chest circumference around the thoracic cavity behind the elbow was considered as girth of the animals. It was measured with care taken to ensure it was not affected by inhalation by the elephant. The length was measured between the base points of the trunk along the curvature of the back to the base point of the tail. The straight-line interval between the rod and the earth was measured as height (Figure 2). The circumference of hind girth was measured in front of the wing of the ilium. The FFC was measured at the widest point of the right forefoot, including nails, at the level of sole. Body weight was calculated as per Hile *et al.* (1997), applying the following formula:

Body weight (kg) = 18.0 × Heart girth (cm)-3336

Height-body weight relationship was calculated as per Sukumar *et al*. (1988), applying the following formula:

Body weight (kg) = $\{(0.06 \text{ height in cm}) - 0.335\}^3$

Height-FFC relationship was calculated as per Sukumar *et al*. (1988), applying the following formula:

Height (cm) = 2.03 FFC

Statistical analysis

All the statistical analysis of data was performed using SPSS 16. The difference among groups was calculated by one way ANOVA. The significant effects of different means were compared by Duncan's Multiple Range Test. Significance was defined at $P < 0.05$. All the values represent mean ± standard errors of the mean (Snedecor and Cochran, 2004).

Results and Discussion

In the present study, the heart girth and body weight were recorded as an ancillary observation to ascertain the effect of feeding probiotics on elephants' physical health. The accomplishment of morphometric variables could be measure of the animal's nutritional condition and considered as an index of an animal's health. The results of heart girth and body weight, as shown in Table 1, revealed non-significant effect of the treatment. Non significant differences regarding the heart girth and body weight were recorded in the Asian elephants of different groups. The body weights recorded at different periods showed more or less similar results. In agreement to this, no significant differences were also observed in the weight changes in probiotics supplemented captive cheetahs (Koeppel, 2004), rats (Hamad *et al.,* 2009); horses (Agazzi *et al.,* 2011); dogs (Marelli *et al.,* 2020). In contrast to the present study, karimi *et al*. (2013) reported a significant reduction in weight gain in animal models. Whereas, significant increase ($P < 0.05$) in the body weight was recorded in the *Saccharomyces cerevisiae* fed rabbits (El-Badawi, 2018; Ahmad *et al.,* 2019).

Table 1: Average values of heart girths and body weights in the Asian elephants

Period	T_1	T_2	T_3	Overall	P-value
Heart Girth (cm)					
0 day	375.17± 12.18	378.83 ± 10.82	384.50 ± 16.90	379.50 ± 7.41	0.887
20 days	376.50 ± 11.37	379.33 ± 10.91	385.67 ± 16.23	380.50 ± 7.14	0.880
50 days	378.33 ± 10.49	378.83 ± 10.52	388.17 ± 15.71	381.78 ± 6.86	0.823
60 days	379.33 ± 10.45	378.33 ± 10.70	388.83 ± 15.35	382.17 ± 6.81	0.806
Body weight* (kg)					
0 day	3417 ± 219.25	3483 ± 194.78	3585 ± 304.12	3495 ± 133.34	0.887
20 days	3441 ± 204.69	3492 ± 196.28	3606 ± 292.19	3513 ± 128.59	0.880
50 days	3474 ± 188.82	3483 ± 189.32	3651 ± 282.79	3536 ± 123.45	0.823
60 days	3492 ± 188.08	3474 ± 192.67	3663 ± 276.30	3543 ± 122.55	0.806

* **calculated body weight as per Hile *et al.* (1997)'s prediction formula**

Table 2: Details of the morphometric measurements of the Asian elephants.

Name of elephant	Reg. no.	Age (yrs)	Heart girth (cm)	BW* (kg)	Actual BW (kg)	Length (cm)	Height (cm)	Hind girth (cm)	FFC (cm)	Height/ FFC ratio
Jaimala	11	41	353	3018	3088	645	230	410	114	2.02
Rajrani	20	56	352	3000	2994	663	255	405	118	2.16
Phoolwanti	116	30	385	3594	3520	725	253	448	128	1.98
Jhomati	53	48	402	3900	3881	798	236	434	130	1.82
Chameli	123	44	370	3324	3341	702	241	415	126	1.91
Jaytara	92	45	414	4116	4066	750	269	449	134	2.01
Laxmi	125	47	352	3000	3089	709	248	405	122.5	2.02
Laxmi	130	52	350	2964	3103	738	258	408	122	2.12
Anno	93	48	365	3234	3268	715	251	418	119	2.11
Tami	109	35	391	3702	3779	730	248	429	121	2.05
Gomati	81	33	400	3864	3791	751	256	443	128	2.00
Shobha	96	40	412	4080	3998	773	270	457	135	2.00
Bhogwati	30	49	333	2658	2722	660	221	375	120	1.84
Champa	105	33	362	3180	3239	682	228	412	120	1.90
Rangoli	43	44	402	3900	3812	790	260	465	124	2.10
Majani	55	50	390	3684	3742	727	253	441	123	2.06
Champakali	52	50	404	3936	3852	799	282	468	132	2.14
Chanchal	9	62	442	4620	4440	780	274	453	134	2.05
Overall Mean ± SEM		44.83± 0.45	382.17 ± 6.81	3543 ± 122.55	3540.28 ± 104.23	729.83 ± 11.24	251.83 ± 3.86	429.72 ± 5.93	125.03 ± 1.45	2.02 ± 0.002

* **calculated body weight as per Hile *et al.* (1997)'s prediction formula**
FFC- forefoot circumference

The possible reason for not achieving the desired result can be viability of strains and short duration of the intervention. Mechanisms by which probiotics might regulate body weight have not been clearly understood. Irrespective of probiotics treatment provided, the study also examined the efficacy of existing prediction equations for estimating weight and height, as shown in Table 2. The predicted body weights were found to be more or less similar to the actual weights of the elephants. The height was observed as twice the FFC measured at the sole. The height/FFC ratio was observed to be 2·02. The predicting approaches based on morphometric measurements have been applied in many species like black rhino, elephants, and zebu cattle with the aims of nutritional formulation, herd management, and medication in circumstances where factual weighing is not feasible (Freeman and King, 1969; Sreekumar and Nirmalan, 1989; Lesosky *et al.,* 2012). Heart girth has been proved to be authentic

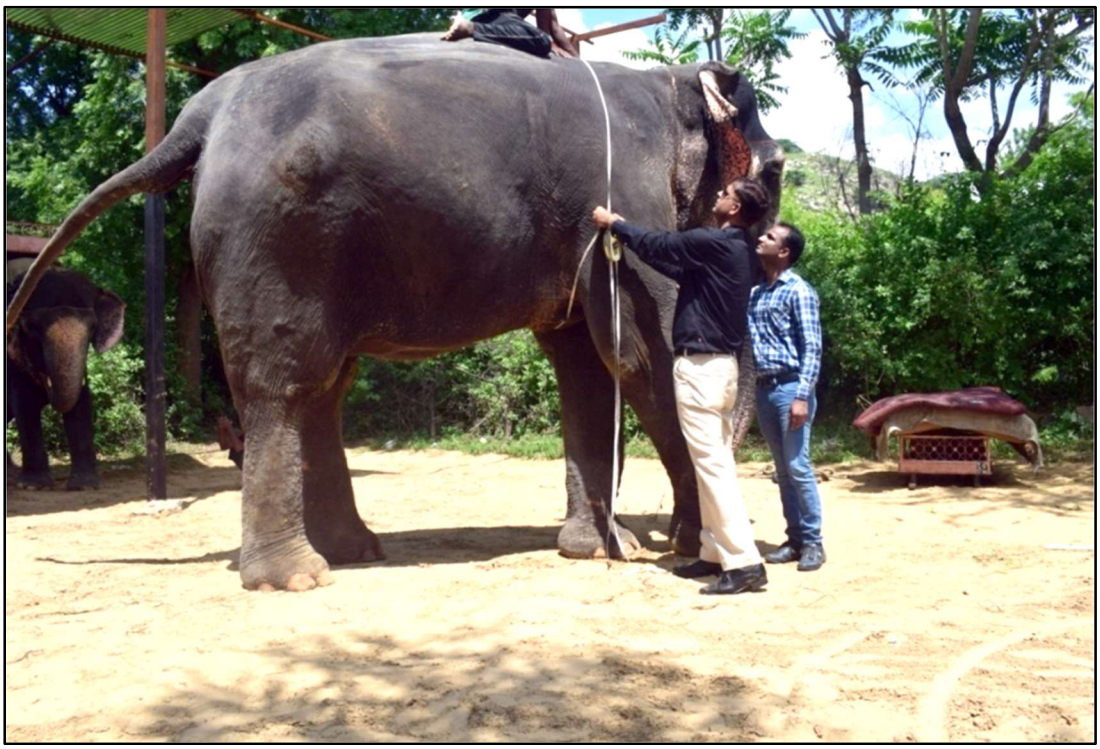

Figure 1: Measurement of heart girth

Figure 2: Measurement of height

predictor of body weight (Hile *et al.,* 1997; Lesosky *et al.,* 2012). In the present experiment, the predicted body weights were found to be more or less similar to the actual weights of the elephants which confirm the existing prediction equation formulated by Hile *et al*. (1997) whereas, the results were not in agreement with Sukumar *et al*. (1988). In addition to this, the data of predicted height was found as twice the forefoot circumference measured at the sole, which also confirm the existing prediction equation for height (Sukumar *et al.,* 1988). Allometric parameters such as heart girth-body weight and height-FFC relationship showed a proportionate isometric relationship in all the elephants, as shown in Table 2, which coincides with the observations of Hile *et al*. (1997) and Sukumar *et al*. (1988), respectively. Contrary to this, the height-body weight relationship was not found to yield an equivalent accuracy in elephants (Hanks, 1972; Sukumar *et al.,* 1988).

Conclusion

No statistical difference due to probiotics treatment regarding the body measurements was observed. Though, the body weights and height of elephants can be authentically estimated from several morphometric measurements. The predicted body weights were found to be more or less similar to the actual weights as well as height was found as twice to FFC which confirm the existing prediction equations. The heart girth and FFC were indicating an isometric relationship with body weights and height of elephant, respectively. The height- body weights relationship was not found to yield an equivalent accuracy in elephants. The results may have significant importance for size estimation of large wild animals in the field, as well as for management in captivity.

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Conflict of interest

The authors declare that they have no conflict of interest.

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