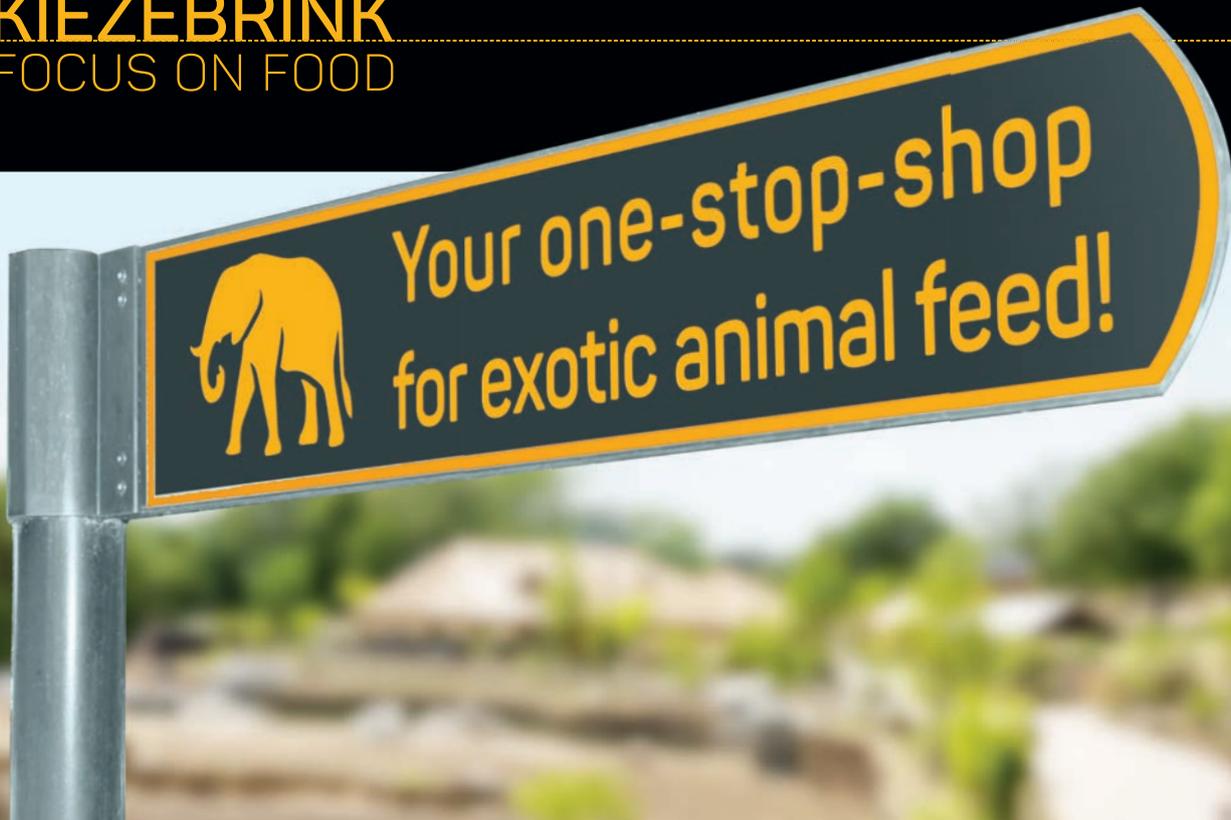




THE EUROPEAN ASSOCIATION OF ZOOS AND AQUARIA

EAZA Nutrition Group News



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INTRODUCTION

Dr Ollie Szyszka, Marwell Wildlife, UK, Chair EAZA Nutrition Group

In this special nutrition edition you will find some very interesting articles based on the presentations given at the last EAZA Zoo Nutrition Conference, which took place in Liberec earlier this year. I would like to take this opportunity to thank the organising committee and specifically those from Liberec Zoo who worked tirelessly to make this conference the success that it was. I would also like to thank Joeke Nijboer and Kristina Johansen for the time and effort that they put into the making of this special issue. I am sure you will find it a valuable addition.

As many of you will be aware, there have been some changes within the EAZA Nutrition Group (ENG) after the resignation of Dr Andrea Fidgett as the Chair of the group, who is moving on to pastures new, and considerably further away! Andrea's knowledge of animal nutrition is exemplary and she can be immensely proud of her achievements with the ENG. She was a valued and well-respected Chair, and we would like to thank her for her leadership and to wish her the very best for the future. I have taken on the position of Chair and am grateful for the continuing and generous support of the committee members with their wealth of knowledge and experience, and in particular Anouk Fens, who has been providing direct assistance with the chairing duties.

We relish the opportunity to make a difference and to continue, and hopefully expand upon, the tasks already ongoing. These, of course, include the organisation of the biennial conferences as well as sourcing and distributing relevant materials, updating the website and liaising with different zoo nutrition experts across Europe to ensure good communications and collaborations. Big steps have already been made regarding the ENG online presence; the website has been updated and a great deal of useful material has been uploaded. Furthermore, the ENG is now also active on Facebook, providing regular posts regarding zoo animal nutrition, and I would recommend this as part of your news feed. As the ENG sits within the research committee, this is a great opportunity for additional result sharing and, where possible, joint research to fill some of the gaps in zoo animal nutrition knowledge. This will of course reach beyond the ENG members, and we aim to encourage close links and communications between all interested professionals.

As a personal introduction, my background is in Animal Science, with a focus on livestock. I completed my PhD in 2012, studying sickness behaviour in cattle. After this I continued to work as a researcher, changing subjects to pig nutrition. I then made the jump from livestock nutrition to zoo animal nutrition when I started working as an Animal Nutritionist at Marwell Zoo (UK) in 2014. With my experience as an animal keeper and my animal science background, the step to zoo animal nutrition seemed a logical one, and one with which I have felt very much at home.

Finally, I would like to take this chance to notify you that the next EAZA Zoo Nutrition Conference will take place at Marwell Zoo on 17–20 January 2019. More information can be found on page 5. We do hope to see you there.

Dr Ollie Szyszka
EAZA Nutrition Group Chair

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Editorial team: Ollie Szyszka, Anouk Fens,

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Editorial assistance: David Williams-Mitchell
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Designer: Louise Tait
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WHO ARE WE?

Our motto **“Animal welfare is our business”** covers feeds and environmental enrichment as well as services by the well educated professionals staffing our nutrition group. Brogaardens nutrition group offers feed planning and nutritional advice to private customers and professionals. The group is staffed with a mix of veterinarians, agronomists and a biochemist ready to help you with your special needs.

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Granovit provides feed and ration recommendations for most of your animals based on the latest nutrition research findings. Recently the product line changed name from **Protector Zoofeeds** to GRANOVIT - but the quality and expert service remains unchanged.

Dodson & Horrell Ltd. was founded in 1939 by Claude Horrell. The company has grown to become a respected authority on animal nutrition and is now one of Europe's leading feed manufacturers. Still a family business to this day the company is still based in Northamptonshire in England and is run by Samuel Horrell, grandson of co-founder Claude Horrell.

Contact Karen Singers Johansen, DVM, for further information at brogaarden@brogaarden.eu



INTRODUCING THE ENG

THIS SPECIAL SUPPLEMENT BEGINS WITH AN INTRODUCTION TO THE EAZA NUTRITION GROUP

Dr Ollie Szyszka, Marwell Wildlife, UK, Chair EAZA Nutrition Group

The EAZA Nutrition Group (ENG) is a working group that functions within EAZA as part of the research committee. The aims of the group include:

- to have an advisory role in nutrition best practice for the EAZA community;
- to have a close working relationship with the TAGS, EEPs, ESBs and other EAZA working groups;
- to contribute to the developments in nutritional analysis and recording;
- to direct enquiring institutions towards relevant resources, training opportunities and specialists.

The current members of the ENG fulfil various roles within the working group, including managing social media, conference organisation, liaising with TAGS and providing and updating relevant resources. The members are made up of a range of Zoo Animal Nutrition specialists from across Europe, working both in zoological collections and in academia, all of whom are actively involved in exotic animal nutrition.

We regularly receive applications for people who want to join the ENG. We welcome all serious applications and each one will be considered independently. Before applying, please consider what you could bring to the ENG. As a member you can be asked to provide guidance, and our members often act as nutritional advisors to TAGS. Within the ENG, members collaborate on nutritional questions and research. Furthermore, we organise the nutrition conference and various publications, such as this special nutrition issue. In addition to this we are always looking to expand and get involved with a range of activities. Members therefore have to have a good working knowledge of Zoo/Exotic Animal Nutrition and need to be active within this field.

ESSENTIAL RESOURCES

As part of our aim to provide and direct towards relevant resources, this is a good opportunity to share with you some of my favourite links on topics related to animal nutrition.

The ENG website at www.eaza.net/conservation/research/eaza-nutrition-group/ is currently being updated and includes the proceedings of past conferences and other publications as well as providing a great many links to different resources.

The ENG Facebook page at www.facebook.com/EAZAnutrition/ offers regular updates on nutrition-related articles and links that would be of interest.

The *Journal of Zoo and Aquarium Research* at www.jzar.org/jzar provides open access journal articles, including those on zoo nutrition topics.

Our American counterpart, AZA Nutrition Advisory Group, has a very good website that provides a lot of information and files, from animal care documents at nagonline.net/tag/animal-care-manuals/ to body condition scoring at nagonline.net/3877/body-condition-scoring/ amongst others.

For an overview of exotic animal nutrition, including more taxon-specific details, the MSD vet manual is an excellent link, and coincidentally is written by an ENG member – www.msdevetmanual.com/management-and-nutrition/nutrition-exotic-and-zoo-animals/overview-of-nutrition-exotic-and-zoo-animals.

Finally, the National Research Council publishes various nutrient requirement books, mainly for livestock and other farmed animals (for example, fish). These can prove useful as a general reference, as quite a lot of detail is provided.



There is also, however, an edition focusing on primates at www.nap.edu/read/9826/chapter/1, which presents an even more comprehensive reference.

NUTRITION CONFERENCE

A large part of ENG's responsibilities is the organisation of the biennial European Zoo Nutrition Conference. The 10th edition will be hosted by Marwell Zoo in the United Kingdom from 18–20 January 2019. This biennial conference is usually attended by around 150 delegates from about 20 countries, representing zoo nutritionists, veterinarians, curators, animal keepers, staff and students from higher education institutes and private individuals.

The conference is a major event for information exchange between individuals involved in zoo animal nutrition in Europe. We already have some exciting guest speakers confirmed for this event and it looks as if it will be a very interesting programme. The conference will be preceded by an optional one-day EAZA Academy workshop, which will take place on 17 January. Topics for the workshop are currently being confirmed and will be announced shortly. Abstracts are welcomed on all topics related to zoo animal nutrition; the deadline for submission is 30 September. For more information on how to register or submit an abstract please go to the EAZA website: www.eaza.net/events/nutrition-conference. I hope that we will see many of you there!



MORE FIBRE, LESS SUGAR

HOW SIMPLE DIETARY CHANGES CAN HAVE A HIGH IMPACT ON PRIMATE HEALTH AND WELFARE

Amy Plowman, Director of Conservation and Education, Paignton Zoo Environmental Park, UK; Francis Cabana, Wildlife Nutritionist, Wildlife Reserves Singapore; Anouk Fens, Nutritionist, Apenheul Primate Park, the Netherlands; Kristina Johansen, Zoo Nutrition Consultant, UK; and Martine Verheij, Deputy Head Keeper, Apenheul Primate Park, the Netherlands

In recent years the evidence that primates need more fibre and less sugar/starch in their diets has mounted considerably. We also now know that the nutrient content of orchard-grown fruits, often provided to primates in zoos, differs very much from the fruits that they eat in the wild; especially that they are much higher in sugar and lower in fibre. We know that browse is the opposite, high in fibre and low in sugar, and is absolutely vital for folivorous primates. Yet in many zoos most primates are still fed a diet containing large amounts of fruit, starchy pellets (or even worse - high sugar/starch human foods) and very little browse, gum or insects. These high sugar/low fibre diets contribute to a range of health issues such as obesity, poor dental health and diabetes, and can

adversely affect behaviour. Many of these problems can be avoided by changing diets to reduce sugar/starch and increase fibre, which can be readily achieved by removing fruit and increasing browse provision. Other measures can also enhance the impact of these diet changes (Fig. 1).

For temperate zoos, provision of large quantities of browse all year round can be difficult. Apenheul Primate Park holds many folivorous primate species, and making sure there is adequate browse in winter is a high priority. Optimal freeze/thawing techniques have been developed for more than 30 leaf species. Leaves must be packaged for freezing to prevent drying out and pulverisation during thawing. Two packaging techniques work well: stripped leaves in plastic bags and

whole browse sealed in plastic film. Stripping leaves is intensive work but is used for willow and *robinia* to avoid feeding bark (the long fibre structure of willow bark can cause stomach impactions and there are potential toxins in *robinia* bark). Plastic bags are filled with leaves, air is squeezed out and more leaves added but not compressed. When defrosting, the leaves are emptied into a tub so they defrost evenly. All other browse species are frozen as whole branches with a maximum diameter of 1.5cm and length 50cm. This avoids pushing all the leaves together and makes defrosting better, and the animals are able to pluck their own leaves. The branches are sealed in the plastic film used to seal pallets (Fig. 2). The thawing method depends on the species. Some thaw well in the foil

and some are better if the foil is removed. Browse intake is best if fed immediately after thawing.

Thanks to its tropical location, at Wildlife Reserves Singapore large amounts of browse are given all year to the most folivorous primates and as regular enrichment to other species. However, the rest of their diet was typical for zoo primates: very large amounts of fruit, some vegetables, some bread, pellets and vitamin supplements and, less typically, chicken egg rice balls. Despite the browse, nutrient profiles for most diets were still low fibre and high sugar/starch, so they were all changed. Chicken egg rice balls, bread and fruit were removed (small amounts of fruit were left in the orang-utan



diet due to the high energy demands of their free-range enclosure) and replaced with germinated pulses and whole grains, vegetables and more browse. For those species that consume them in the wild, amounts of insects and gum were increased. These changes increased fibre, reduced sugar/starch and enhanced many other nutrients, eliminating the need for vitamin and omega 3 supplements.

The new diets resulted in many health improvements: better faecal quality and fewer parasites in callitrichids; improved weights, faecal quality, coat condition, dental health and infant survival in slow lorises; lower blood glucose levels in chimpanzees and orang-utans (reducing their risk of diabetes); better infant survival, improved coat condition and weight management in a large group of hamadryas baboons (107 individuals), with much less difference between the weights of dominant and subordinate individuals. Even with the colobines who had previously had very little fruit in their diet, its total removal resulted in improved weights of 'thin and lanky' individuals, reduced dental disease and increased breeding and infant survival, indicating that even

small amounts of sugary food can cause dysbiosis in those species highly adapted to a folivorous diet. At Apenheul Primate Park, similar diet changes also resulted in improved health for Alaotran bamboo lemurs (*Hapalemur alaotrensis*). Following the removal of fruit from their diet, the male gradually lost weight until a healthy body weight was achieved (from 1465g to 1150g). In addition, two obese females, weighing 2120g and 1510g arrived at the zoo and were successfully introduced to the fruit-free diet with *ad libitum* access to fresh bamboo plants. Over the next seven months the older female lost almost 700g and gave birth to a healthy young baby. Improved body condition and faecal quality was also seen following fruit removal for four New World primate species at Shaldon Wildlife Trust.

At Wildlife Reserves Singapore positive behavioural changes were also recorded following diet change, including: reduced aggression in chimpanzees and increased feeding/foraging and/or travelling time and reduced abnormal/stereotypic behaviour in slow lorises and orang-utans. For the orang-utans this was a reduction in the performance of regurgitation and reingestion (R/R)

which has also been seen in other zoos as a result of diet change. In Kristiansand Zoo in Norway and Furuviik Zoo in Sweden four orang-utans were showing R/R, one male several times a day. The diets fed were already low in fruit and high in fibre so the aim was to reduce the frequency of R/R by further changes in the diet: removing fruit completely, increasing the time when food was available, increasing amount and availability of browse and green vegetables and changing the pellet to one higher in fibre and lower in sugar/starch (Fig. 3). After implementing these changes, the frequency of R/R dropped dramatically in both groups. One orang-utan completely stopped displaying

the behaviour, whereas the others still displayed the behaviour on occasion. The keepers, who were very involved in the diet changes and provided valuable feedback, also reported a decrease in aggressive behaviour and other stereotypical behaviour such as faecal smearing. Stereotypical behaviour (head twist) also decreased significantly in squirrel monkeys at Shaldon Wildlife Trust, and other behavioural effects have been reported in a range of species, especially reduced aggression and self-directed behaviour (SDB) in lemurs at Paignton and Newquay Zoos. However, the results were not so clear in ring-tailed lemurs (*Lemur catta*) at Apenheul Primate Park. Removal of fruit from the diets did result in less aggression (not significant) but increased SDB rates. This difference in effect may be due to the higher amount of sugar in the original diet.

Overall, our combined experiences of increasing fibre content and reducing sugar/starch levels clearly demonstrate that these measures, combined with a real focus on diet monitoring and weight management, can have hugely positive effects for health, welfare and breeding in zoo primates.

NUTRITION FOR HEALTHY CLAWS

A RESEARCH PROJECT CONDUCTED AT THE TIERGARTEN IN NUREMBERG DISCOVERED THAT DIET CAN IMPROVE CLAW HEALTH IN TAKINS

Dr Angela Gimmel, ECVCN resident, Institute of Animal Nutrition, Vetsuisse Faculty, University of Zurich, Switzerland; Dr Katrin Baumgartner, Head Veterinarian, Tiergarten Nuremberg, Germany; and Professor Dr Annette Liesegang, Director of Department, Institute of Animal Nutrition, Vetsuisse Faculty, University of Zurich, Switzerland

In zoos, the feeding of rare and exotic herbivores often represents a challenge. This is also true for zoos that keep takins. The takin (*Budorcas taxicolor taxicolor*), which belongs to the sub-family of goat-antelopes (*Caprinae*), is native to India, Myanmar and the People's Republic of China and lives at altitudes between 1,000 and 4,500m above sea level. Takins are mainly browsers, although they also consume fresh herbs and grass when seasonally available (Schaller *et al.*, 1986).

The takin herd in the Tiergarten Nuremberg experienced problems with claw health. Claw growth was excessive and, over time, the claw horns cracked and fissures appeared. These changes were mainly observed in the bull, but other individuals in the herd were affected, too. Regular hoof trims under full anaesthesia were necessary for animal health and welfare reasons. The bull and some females were recently moved to Nuremberg from another zoo, where these problems did not occur. The head veterinarian therefore requested the help of the nutritional

consultation service of the Institute of Animal Nutrition, Vetsuisse Faculty, University of Zurich, to determine whether nutrition could be one of the reasons for poor claw health and excessive claw growth in their takins.

Reasons for reduced claw health in ruminants are numerous, and it is often a multifactorial disease. In the literature, diets with high starch or high sugar contents increase the risk of lameness via ruminal acidosis (Lean *et al.*, 2013). This problem is exacerbated when not enough roughage or browse are offered. Also, high-protein diets are implicated in inducing lameness in cattle (Westwood *et al.*, 2003). Regarding other dietary components, deficiencies in minerals such as calcium, phosphorus, selenium, copper and zinc negatively impact claw health in dairy herds (Lean *et al.*, 2013). Vitamin D deficiency also could play a role in the pathogenesis of claw problems (Lean *et al.*, 2013).

In the Tiergarten Nuremberg, the takins are kept in an enclosure with different substrates. A large part

of the enclosure is earth or grass paddock, with a small concrete area where the feeding takes place. The daily ration in summer consists of fresh grass, second-cut hay, carrots, beetroot, wheat bran, oats, pellets, vitamin and mineral supplements; crisp bread is also given, but only for training. In addition, the animals are offered fresh branches on a daily basis and have access to multiple salt licks. The whole group is fed together, so methods for intake reconstruction of the individual animal are needed. There are mainly two methods used. In method 1, the total weight of the group is calculated and the amount of the feed materials is divided by the total weight of the group and then multiplied with the weight of the bull (Table 1). In method 2, the weights are calculated with metabolic bodyweight (body weight^{0.85}). That means the metabolic body weight of each individual animal has to be added to receive the total group metabolic body weight. The amount of the feed materials is then divided by the total metabolic body weight of the group and finally multiplied with

Table 1: Example of intake reconstruction using method 1 with body weight (BW)

Food item	Group intake [g/day]	Total group weight [kg]	Intake [g/kg BW/day]	Individual intake bull 255kg BW [g/day]
Grass	38000	1445	26.3	6706
Hay	7000		4.8	1235
Beetroot	5000		3.5	882

Table 2: Example of intake reconstruction using method 2 with metabolic body weight (BW^{0.85})

Food item	Group intake [g/day]	Total group BW ^{0.85} [kg]	Intake [g/kg BW ^{0.85} /day]	Individual intake bull 111.1kg BW ^{0.85} [g/day]
Grass	38000	660.3	57.5	6391
Hay	7000		10.6	1177
Beetroot	5000		7.6	841

Table 3: Metabolisable energy (ME), crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre (NDF), iron (Fe), copper (Cu), zinc (Zn) and selenium (Se) content of diet per day calculated with different reconstruction methods (RM) and compared to recommended diet and nutrient recommendations.

	ME	CP	ADF	NDF	Sugar	Fe	Cu	Zn	Se
	[MJ]	[g]	[g]	[g]	[g]	[mg]	[mg]	[mg]	[mg]
Diet RM 1	42.4	661	651	1530	807	125.1	9.1	60.3	0.2
Diet RM 2	39.8	633	623	1104	771	119.7	8.7	56.7	0.2
Recommendations (NRC ¹ or Supplemente ²)	31	260				380	30	76.5	0.32
Recommended diet	32.6	435	945	1530	273	402	31	206	0.8



the metabolic body weight of the bull (Table 2).

Depending on the method of intake reconstruction (method 1 or method 2), the bull with an approximate body weight of 255kg had an estimated daily energy intake between 42.1 and 39.8 MJ metabolisable energy (ME) respectively, which is markedly higher than recommendations for a hypothetical 255kg goat ram according to the NRC 2007 (31 MJ ME/day). Also, crude protein intake was high and trace elements (iron, zinc, copper and selenium) were

low compared to goat and sheep requirements. Vitamin D intake was calculated to be adequate for the takin bull. Regarding fibre, the structure of the individual feedstuff seemed too low. Also, neutral detergent fibre (NDF) and acid detergent fibre (ADF) were relatively low and sugar was relatively high compared to the recommendations for goats (Table 3).

Recommendations were made to reduce the energy and highly

fermentable carbohydrate content (starch and sugar) of the daily ration. Also mineral supplementation was augmented to cover the requirements suggested for a hypothetical 255kg goat ram. The recommendation for the herd was as follows: grass, a late first cut of hay, lucerne hay, whole plant corn pellets, wheat bran, carrots, mineral supplement, a selenium supplement and crisp bread for training. Branches should continue to be offered in high quantities, such as hazel or linden, to provide the takins with sufficient browse. Also, enough easily accessible salt licks should be provided.

After the nutritional changes were implemented, an improvement of claw health in the bull and the two highest ranking females was observed. Prior to the changes, claw growth was excessive and the claws had to be clipped often. After the diet change, no additional clipping has been necessary to date.

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GOING FRUIT-FREE

HOW A FRUIT-FREE DIET CAN IMPROVE BEHAVIOURAL PROBLEMS IN LEMURS

Anouk Fens, nutritionist, and Thomas Bionda, behavioural biologist, Apenheul Primate Park, the Netherlands; Ilse Sijtsma and Robin Dapper, students, Van Hall Larenstein

In human care, many lemurs face nutrition-related health issues such as obesity, poor dental health, diabetes, hepatic iron storage disease and reproductive failure. Knowledge of the feeding ecology of wild primates is crucial for understanding their nutrient needs while in human care and, ultimately, helpful for appropriate nutritional management. Optimal dietary husbandry of lemurs in an *ex situ* setting will most likely prevent nutrition-related health problems.

Diets provided to lemurs in human care differ nutritionally from their natural diets. In the wild, lemurs show both frugivorous and folivorous feeding strategies and can adapt to seasonal variations. In zoos, lemur diets contain mainly commercial fruits, (starchy) vegetables, pellets and seeds. Due to the substitution of cultivated fruits for wild fruits, zoo diets are not comparable in nutrient composition to wild diets.

Cultivated fruits for human use are generally higher in easily digestible carbohydrates and lower in fibre, protein and calcium than wild fruits.

In the last few years, attention has been paid to the diets of primates in human care, and lemurs in particular. A number of cases reported beneficial effects on physical health after commercial fruit was removed from the diet, such as improved weight management and faecal consistency. Recent studies investigated the effect on behaviour of removing fruit from lemurs' diets, as high sugar levels are closely associated with undesirable behaviours in humans.

This study examined the effect of a fruit-free diet on nutrient composition and the behaviour of ring-tailed lemurs in order to gain more insight into the relation between nutrition and aggressive behaviour and self-directed behaviour (SDB).

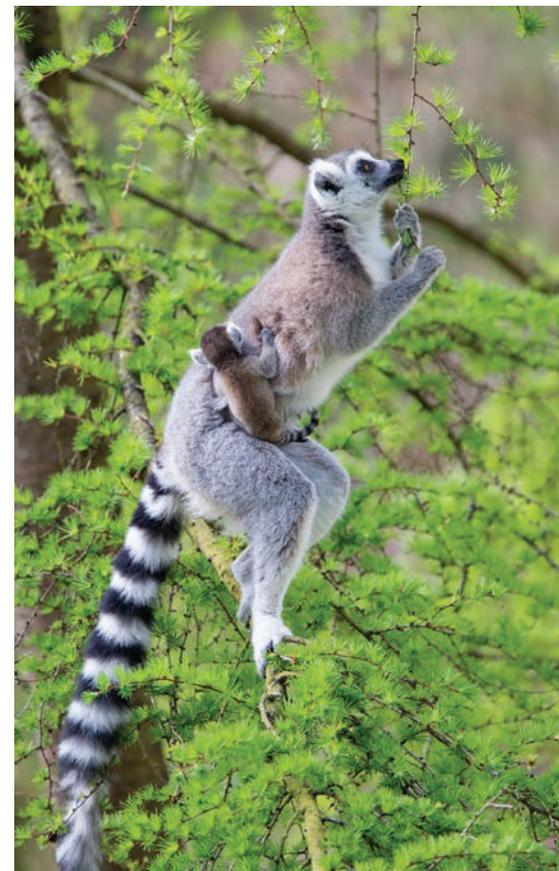


Table 1: Nutritional composition of original and fruit-free diets for ring-tailed lemurs on dry matter basis.

Nutrient	Original	Fruit-free
Energy ME (kJ/day)	1495	1010
Crude protein	17.2	22.9
Crude fat	6.7	4.9
Neutral detergent fibre	11.2	25.7
Acid detergent fibre	7.2	16.6
Non-structural carbohydrate*	29.7	20.8

*amongst others, sugars and starch

METHODS

The study was conducted at Apenheul Primate Park in the Netherlands. Study subjects were nine ring-tailed lemurs (*Lemur catta*), one male and eight females. The nine lemurs constituted a combined family group and were housed together in an indoor enclosure with optional access to their outside enclosure. The lemurs' original diet consisted of fruit, vegetables, pellets (Leaf-Eater Primate Diet – Nutrazu; Trio Munch Rings – Mazuri; Primate PT1 – Arie Blok), and boiled eggs, and browse was available *ad libitum*. Weighed food items were offered three times a day for seven days in a row. Uneaten food items were removed and weighed before the next feeding to determine feed intake. The original diet was evaluated and compared to published nutritional requirements in order to develop a fruit-free diet. Diet evaluation was conducted with Zootrition software (version 2.6; 2005). The new diet was introduced gradually over a week in the middle of the data collection period.

Data on activity and social interactions of individual lemurs were collected from November 2015 to January 2016. A new breeding male was introduced a month before data collection started. Data were collected using a combination of *ad libitum* observations (aggression) and focal animal sampling (SDB), comprising 64 h and 113 h of observations respectively. Lemur activities were classified in the following categories: aggression, SDB and affiliative social behaviour. Resting and other behaviours were also recorded.

RESULTS

The fruit-free diet was developed by removing fruit, eggs, Trio Munch rings and Primate PT1 pellet from the original diet and increasing the amount of vegetables and Leaf-Eater pellets. Nutritional analyses showed increased fibre levels (ADF 7.2 > 16.6%; NDF 11.2 > 25.7%) and decreased non-structural carbohydrates (29.7 > 20.8%) in the fruit-free diet (Table 1). Crude protein levels increased from 17 to 23 per cent. Energy levels in the fruit-free diet were 12 per cent lower compared to the original diet.

The total amount of observed aggression did not differ significantly between the original and fruit-free diet, except for chasing, which was performed significantly less on the fruit-free diet. Two female lemurs showed significantly less individual aggressive behaviour after the diet change. Two SDBs (scratching and shaking) were significantly higher when the fruit-free diet was eaten.

CONCLUSIONS AND DISCUSSION

We found that lemurs that consumed the fruit-free diet showed, in total, less aggressive behaviour, but not to a significant degree. When looking at specific types of aggressive behaviour, we did find a significant decrease in the amount of chasing. Surprisingly, we found significantly more SDBs. In contrast, Britt *et al.* (2015) observed both aggression and SDBs to be significantly lower in several lemur species when they were fed a fruit-free diet. These differences might be explained by the difference in reduction of non-structural

carbohydrates. In the study described here, the fraction of non-structural carbohydrates was reduced by 12 per cent (from 29.7 to 20.8), whereas Britt *et al.* (2015) reported an average reduction of 25 per cent (62.1 and 46.3 for the original and fruit-free diet respectively). Above all, non-structural carbohydrate levels in their fruit-free diet were notably higher compared to levels in the original diet fed in Apenheul. Although the difference in data collection prevents direct comparisons, the rates of agonistic behaviour in our study do not appear to be very deviant from other studies and thus may explain the absence of a significant reduction of this behaviour.

Data collection took place in winter, when the lemurs in Apenheul Primate Park are housed differently compared to summer. In winter, the lemurs are housed in their inside enclosure, which is much smaller than the large outside exhibit used in summer. A smaller enclosure might lead to a decrease in aggressive behaviour, as has been shown in pigtailed monkeys (*Macaca nemestrina*), although other studies show contradicting results.

Additionally, results might have been influenced by the introduction of a new breeding male shortly before data collection took place.

Nutritional analysis showed the fruit-free diet to be more correlated with nutrient requirements and therefore more resembling their natural diet. Once more, behavioural observations showed that a transition to such a diet is possible without evident negative effects during observations. Therefore, the fruit-free diet might be beneficial in preventing nutrition-related problems, and stimulate positive behaviour and overall adequate welfare.

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PREVENTING DIETARY DRIFT

AN ASSESSMENT OF DIETARY DRIFT AT PAIGNTON ZOO ENVIRONMENTAL PARK LED TO SOME SIMPLE BUT VITAL CONCLUSIONS

Louise Cox, undergraduate student, and Brian Evans, Senior Lecturer, University Centre Hartpury, UK, and Amy Plowman, Director of Conservation and Education, Paignton Zoo, UK

Dietary drift, the unapproved and gradual alteration of an animal's diet, is commonplace within zoos. Research at Paignton Zoo found that drift was present in all sections studied (birds, primates and large mammals). A lack of communication between keepers and volunteers feeding the animals, and those formulating diets (called nutritionists in this article but that term includes curators, vets, research staff, etc.) was found to be an important factor contributing to drift. Drift can be caused by swapping feed ingredients due to availability, providing extra for training or medication and creating diets based on a number of items or scoops rather than weight. Due to changes in the nutritional composition of the diet, drift can contribute to unexplained changes in an animal's health. Interview findings indicate that keepers and volunteers would benefit from, and welcome, ongoing nutritional training that would help to prevent drift.

An intake study was conducted over 10 days in July and August 2016, and involved two species per section (Table 1). Diets as per the diet sheet, offered diets and consumed diets were analysed using Zootrition computer software



URAL OWL

for nutrient composition and energy provision. There was a large variation in weights of ingredients offered when diets were specified by number of items rather than weight. Variation was also seen when diet sheets were formulated in grams; however this seemed to be less than other variations. Differences were also seen in the weight of chicks on different sections, whereby the heaviest chick fed to ural owls (*Strix uralensis*) weighed less than any chick fed to coatis (*Nasua nasua*). Therefore, feeding by weight rather than number of items will help to



PIED TAMARIN

Table 1: Examples of minimum and maximum weights of food items offered.

Species	Food item	Min g fed	Max g fed	Diet sheet (number of items or g)
Red river hog	Beetroot	1527	3587	12
Great grey owl	Mice	88	133	4
Ural owl	Chicks	120	146	4
Ring tailed coati	Chicks	156	161	4
Ring tailed coati	Pellet	399	412	400g
Red river hog	Pellet	4982	5109	5000g
Mandrill	Root veg	1278	1932	1280g
Mandrill	Pellet	960	1252	960g
Pied tamarin	Root veg	237	362	360g

RING TAILED COATI



provide more consistent diets. Root vegetable amounts offered to pied tamarins (*Saguinus bicolor*) were often lower than on the diet sheet, thought to be caused by cooking vegetables, as this alters the weight. It is vital that diet sheets state whether items should be cooked or soaked, and if they should be weighed before or after this, to ensure that animals are fed the correct amounts.

ENERGY INTAKE

Mandrills (*Mandrillus sphinx*) were offered, and consumed, an average of 4.8 per cent (307 kcal) extra energy per day compared to their diet sheet, which would lead to 10,220 kcal extra per year if this level of overfeeding continued. This could have significant weight implications, although no mandrills were deemed overweight by keepers, suggesting that this extra feed may be necessary. Communicating this to the nutritionist is important, so that appropriate increases in diets can be monitored. Red river hogs (*Potamochoerus porcus*) were provided similar amounts of energy to their diet sheet (0.8 per cent less on average) but consumed 5.3 per cent (761 kcal) less energy per day, potentially losing 69,000 kcal over a year. Similarly, the condition of these animals was not a concern, perhaps suggesting the river hogs do not require as much energy as calculated. On the other hand, keepers suggested that intake varied

seasonally, so these results, found over two weeks in summer, may be unrepresentative of year-round daily intake. Communicating with the nutritionist is again important, to ensure that seasonal differences are accounted for.

MONITORING DIETS

It is also important to consider what the animal is choosing to eat from the diet that is offered. For example, red river hogs were offered 3.2 and 3.7 per cent less ADF and NDF respectively than the diet sheet sets out, but actually consumed 18.6 and 19.7 per cent less ADF and NDF. It is therefore vital to ensure accurate monitoring of leftover food, and to seek further advice if specific feed items are commonly left uneaten, to ensure that the nutrient composition of the consumed diet is appropriate.

Ensuring that animals are offered the diet that is formulated for them is important, as it allows keepers to accurately monitor diets. With a lack of 'ideal' diet guidelines for many zoo species to begin with, feeding differently to the diet sheet makes monitoring the suitability of a diet more difficult. Any extra food or temporary diet changes should also be recorded to help monitoring.

TRAINING AND COMMUNICATION

Alongside the intake study, interviews were conducted with seven keepers and six volunteers to understand the cause of drift. Keepers highlighted a

lack of understanding around how specific diets had been formulated, and of animal nutrition generally. Ongoing nutritional training would be welcomed by keepers, and would highlight the consequences of providing inappropriate or larger quantities of food. Regular meetings between nutritionists and keepers are fundamental to ensuring good communication, and would provide an opportunity to raise concerns or questions about animal diets.

Two volunteers expressed concern that keepers were too busy to be asked questions regarding diets and that they therefore found it difficult to know what to do in certain situations, such as when ingredients were unavailable. Reassuring volunteers that keepers are available may help to prevent miscommunications and drift. If volunteers work independently, brief guidelines or a protocol on how to prepare diets if problems arise, such as missing ingredients, may be beneficial. Guidelines detailing when to use additional food and how the daily diet should be modified – for example, if giving very unpalatable medication in additional food – could also be beneficial for keepers

TAKE-HOME MESSAGES

Regular meetings between keepers and nutritionists are vital for maintaining communication, addressing concerns regarding diets and allowing diets to be modified by appropriate staff. Creating diets based on the weight of ingredients rather than numbers will also help to reduce drift. Furthermore, a lack of knowledge surrounding how diets have been formulated, and general understanding of nutritional requirements can contribute to drift. Therefore, ongoing training for keepers and volunteers who are managing diets can only be beneficial in reducing drift and will benefit the health of animals in the long term.

We would like to offer a huge thank you to all the keepers at Paignton Zoo for being so accommodating during this study!

INSIDE THE ZOO KITCHEN

LIBEREC ZOO'S NEWLY RENOVATED KITCHEN PROVIDES HIGH-QUALITY NUTRITION IN AN EFFICIENT AND COST-EFFECTIVE WAY

Petra Bolechová, curator and nutritionist, Liberec ZOO, Czech Republic

The Liberec ZOO is the oldest zoological garden in the Czech Republic. In its 13-hectare area, more than 829 specimens from 150 species of animals are kept. A new entrance to the zoo was built in 2014, and a new, multifunctional Zoo Expo building was renovated by returning it to its original location and including a new animal kitchen background.

Our central animal kitchen has several separate work zones to provide hygienic zoo feeding, including a fish area, a meat area, a storage area for dry food pellets, and the kitchen, where all vegetables, fruits and additives are prepared daily according to the diet sheet for each animal species. All diet sheets are developed by the nutritionist and approved by curators.

Animal nutrition is a part of each zookeeper's daily agenda. All prepared diets are distributed daily to each animal department by electric car. In addition, a large number of rats, mice, rabbits and so on are kept in a separate area inside the zoo in order to feed our huge collection of raptors. Another important item is bulky feed and browse for our ungulates. The collection's diet is a big part of the zoo's expenses, but a good quality product is worth the cost, because it ensures that our animals and their offspring are healthy.

A whole new section of our nutrition department is an abbatoir for farm animals, which we hope will help us to reduce our purchase costs. This comprises several sections, just as in a regular abbatoir: a space for the



IMAGES FROM THE NINTH EUROPEAN ZOO NUTRITION CONFERENCE

temporary animal housing, a butchery hall, chilling rooms, a splitting room and a freezer. This section of the nutrition department operates under full veterinary supervision to ensure that welfare rules are properly followed and that we use the best quality products according to the HACCP regulations.

On 26–29 January 2017 our zoo had the opportunity to host the Ninth European Zoo Nutrition Conference, which became a meeting place for nutrition experts from all over the world. The conference covered many interesting issues and, above

all, provided space for extensive, practical consultations, which were very positively viewed by all our zoo representatives at this event. Practical demonstrations and workshops, which took place in the zoo itself, proved to be extremely popular, and even the cold weather did not deter the delegates from participating by preparing snacks for insects.

We are very glad to have been given the opportunity to host this conference, as the expertise that was shared there will enable us to improve our own zoo's animal nutrition.

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NUTRITION MATTERS

AT THE NINTH EUROPEAN ZOO NUTRITION CONFERENCE IN LIBEREC, EXPERTS FROM ACROSS THE GLOBE SHARED THEIR KNOWLEDGE AND THEIR MOST RECENT RESEARCH

As in previous years, the themes of obesity and lack of dietary fibre were prominent at the Ninth European Zoo Nutrition Conference, held in Liberec in January 2017. Yet many other topics were also raised, which demonstrated the variety of angles that make nutrition important in zoos, as the examples below will show. In particular, aspects of nutrition that go beyond just physiological needs were tackled: for example, the lack of sustainability of certain diet items (e.g. palm oil), where one can encounter the paradox that a food might be beneficial for individuals in a zoo, but its production may harm the same species in the wild. It is also clear that food and feeding strategy can determine animal welfare; for example, the provision of dietary fibre and structure as a tool to ensure normal behaviour in, for instance, primates, and even for animals with particular feeding habits, such as exudativores (gum eaters) and myrmecophages (ant and termite eaters).

It was good to see that the presentations dealt with a wide variety of animals from very different taxonomic groups, including mammals, birds, fish, amphibians, reptiles and insects. Even though the amount of information presented at the conference was considerable, there is still a demand for more scientific knowledge. For instance, the problem of iron overload disease in black rhinoceros (see page 25) has still not been resolved, and is hampering the conservation of this species. Many talks also demonstrated how the science can be applied in practice, or what the challenges may be in doing so. These challenges include the use of food to administer medicine (e.g. contraception to giraffe), which toxic plants to avoid, how to store fish properly, how to analyse the nutritional value of insects, and how

to avoid pests in stored food. The conference demonstrated that there is still a lot of work to do in optimising nutrition, but that the science is being applied in zoos, and more exciting data will emerge over time.

OIL PALM PRODUCTS IN ANIMAL FEED: AN UNDERESTIMATED ETHICAL PROBLEM – *Stanislav Lhota, primatologist, Usti nad Labem Zoo, Czech Republic*

Many zoos have recently started thinking about their ecological footprint. Environmentally responsible and sustainable practices of animal care are becoming part of zoo conservation strategies. This includes, among other factors, environmentally responsible building, energy use, waste management and animal feeding. However, this change often lags behind the conservation messages that the zoos pass on to the visitors and the wider public. Palm oil can be one such example.

Palm oil production represents the main cause of deforestation in southeast Asia, and it is becoming a serious problem in tropical Africa and America as well. The ongoing expansion of palm oil plantations is threatening the survival of charismatic animals such as orang-utan, tigers, Asian elephants, clouded leopards and sun bears, as well as countless smaller and less well-known species. It is causing soil degradation and water pollution, which also affects the economy of local communities.

Zoos were among the first institutions to expose these serious issues to the broader public via campaigns that caused a huge international response. Today, many zoos continue advocating the boycott of palm oil, while others chose a more positive campaign, supporting the sustainable production of palm oil and its certification according to the RSPO (Roundtable on Sustainable Palm Oil)

Principles and Criteria. The zoos focus their campaign on the palm oil that is being used in human food and, to a lesser extent, in cosmetics, house care products and biofuels.

However, a significant proportion of palm oil, palm kernel oil and palm kernel meal is used to feed animals, which may include zoo animals. Some products, such as the virgin red palm oil, which is used to supplement the diet of specialised parrots, may be available in sufficient supply in organic quality, and with RSPO certificates. However, the majority of the palm oil products used for animal feed is not RSPO-certified and cannot be traced to the plantation, so it is likely that these are not deforestation-free products. On the other hand, most of the palm oil products in zoo animal feed can be replaced by more sustainable non-tropical products. They are more widely available than the RSPO-certified sustainably produced palm oil products traceable to plantation.

Given this situation, avoiding palm oil products in zoo animal feed and replacing them with non-tropical alternatives is probably the most feasible, and immediately available, solution.

GUM-EATERS: A REVIEW OF THE NUTRITION OF EXUDATIVOROUS MAMMALS – *Francis Cabana, wildlife nutritionist, Wild Reserves Singapore*

Gum is widely available and composed mainly of soluble structural carbohydrates. No mammalian enzymes can digest gum; therefore a mammal ingesting gum must rely on microbial fermentation in order to access the energy it possesses. Gums are known to be relatively poor in nutrients, but in spite of this, some mammals have evolved to exploit this food resource. I aim to review the literature for all mammal species that have been recorded to ingest gum,

Table 1: Clutch size in depending of the offered feed amount

Eggs/clutch	n	offered feed amount	
		ad libitum	saisonal
budgerigars	58	5.5 ± 1.5	6.8 ± 1.9
lovebirds	60	4.9 ± 1.3	5.8 ± 1.2
grey parrots	10	2.9 ± 0.9	3.8 ± 1.1

whether quantified or not, and discuss this in the context of their evolutionary adaptations. I also investigated the recommended diets for these species being kept in human care to examine whether gum is recommended. I conducted a literature search on the ISI Web of Knowledge to tabulate all mammal species observed ingesting gum and classify them as obligate, facultative or opportunistic feeders. I encountered 92 mammal species that eat gum in the wild (26 obligate feeders, 35 facultative feeders and 31 opportunistic feeders). Obligate feeders have entirely evolved to exploit this resource but were found not to be given gum in captivity, which may explain why they are failing to thrive, as opposed to facultative feeders, which have fewer issues. Gum may be necessary for the health of obligate feeders in captivity, and I present data to this effect, specifically for slow lorises (*Nycticebus spp.*), callitrichids (*Cebuella and Callithrix spp.*) and marsupial gliders (*Petaurids*). Wasting syndromes, dental disease, cannibalism, abnormal behaviour patterns and dysbiosis of gut microbes have all been linked with inappropriate diets given to obligate gum feeders.

NUTRITION AND FERTILITY IN PET BIRDS

– *Petra Wolf, European Chair of Nutrition Physiology and Animal Nutrition, AUF, University of Rostock, Germany*

During egg production and rearing of nestlings, a lot of health problems occur (e.g. disturbances of egg production, egg binding, skeletal disorders in nestlings etc.) which sometimes depend on the diet eaten before and during the reproduction period.

A lot of special diets are offered for this time. Moreover, secret

formulae circulate amongst breeders to optimise the supply of breeding parents and hatched young. But all formulae are based on experience and tradition or are transferred from recommendations for poultry. Investigations in pet birds are rare.

But as is known from other species, the fertility in pet birds depends also – amongst other factors – on the nutrition of the male and female birds. The sperm quality of the cock depends on the zinc, methionine and cysteine level of the diet. Pumpkins, hemp and wheat germs are rich in zinc, whereas egg yolk, brazil nuts and striped sunflower seeds deliver methionine. Arginine is a precursor of spermin and spermitin and increase the number and quality of spermatozoa. Pine nuts and peanuts present arginine-rich feedstuffs. Moreover, the fertilisation rate depends on the body mass of the cock. The higher the body condition score (BCS) the lower the rate.

Egg quality is influenced by energy, protein, vitamin E and selenium level as well. Fourteen days before the first egg is laid, the feed intake doubles. In general the offered feed amount influences the laying activity in pet birds. A luxury consumption over the whole year decreases the clutch size (see Table 1). Hatchability can be influenced by the quality and strength of the eggshell and depends on the calcium and vitamin D supply. The death of the embryo might be caused by several vitamin deficiencies. A death at the beginning of the incubation period is seen in cases of deficiency in vitamin A, biotin and vitamin E. A lack of riboflavin leads to embryo mortality in the middle of that period and embryonic death at the end can be observed by an insufficient supply of niacin acid or vitamin D.

INSECTS AS FEED FOR ZOO

ANIMALS – *Dennis Oonincx, entomologist, Wageningen University & Research, the Netherlands*

Several dozen insect species are used as a source of feed for amphibians, reptiles, birds and mammals. The nutrient composition differs between species, but also between developmental stages within a species. Furthermore, the diet provided to these insects, but also abiotic factors during rearing, have an effect on their composition.

Whereas for certain zoo animals insects are primarily a form of enrichment, for others their complete diet may consist of insects. Especially in the latter situation, insect composition is vital for the health and wellbeing of the consuming animal.

Commercially available insects are a good source of most nutrients for captive insectivores. However, certain nutrients are present at low concentrations, and could lead to deficiencies. This in turn can affect the health, wellbeing and productivity of the consuming animal. Some examples of these potentially limiting nutrients are calcium, vitamin A and carotenoids, vitamin D and E and omega 3 fatty acids.

An overview of the most commonly available feeder species and their nutrient composition was provided. The nutrient composition of the feeder species can be altered to better suit the specific nutrient requirements of the consuming animal. Alterations in the diet of the insect can have a large effect, although other factors such as development stage also need to be considered. The best feeder insect depends partially on the nutrient profile, but also on the purpose and on available time. Combining uses of feeder insects, for instance, by using them as display animals or using them for waste management can help decrease costs and increase sustainability. Lastly, some information on the rearing of a selection of insect species was provided.

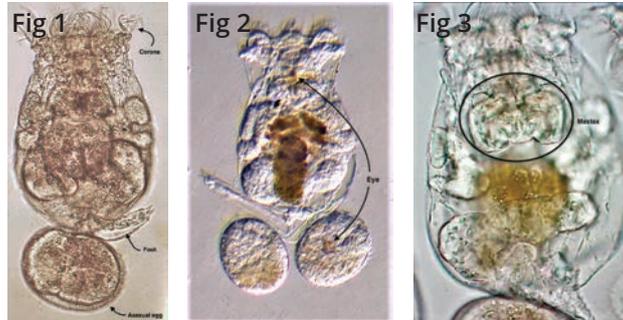
THE ROTIFER SOLUTION

HOW LARGE-SCALE PRODUCTION OF NUTRITIOUS ROTIFERS CAN ENHANCE THE REPRODUCTIVE CAPACITIES OF MARINE AQUARIUM FISH

Olivier Detournay, Chief Scientific Officer at Planktovie, France, and Eric Henry, Chief Scientific Officer at Reed Mariculture, California, USA

While more than 90 per cent of freshwater ornamental fish species are currently produced in human care, the vast majority of marine aquariums are populated with wild-caught specimens. In addition, less conscientious traders continue to support the use of destructive fishing techniques, namely the use of cyanide, to anaesthetise highly priced fish species; this severely damages natural ecosystems, particularly coral reefs, where most of these fish are from. It is estimated that during the journey from ocean to aquarium, up to 80 per cent of the traded animals die either during capture (especially due to the use of poisons during collection) or during shipment, due to poor handling practices and diseases. To reduce this pressure on wild populations, there has been an increased focus recently on supplying aquariums with fish bred in human care. An additional advantage is that such fish are more robust and far better adapted to live in aquariums.

To date, successful rearing has been scientifically reported for only a few species, and less than one per cent of marine aquarium fish are commercially produced, highlighting the importance of improving larval culture at first feeding, as well as the nutritional profiles of the larval feeds. Live feeds are an essential component in the diet of many reared fish species, especially during the first feeding stages of development. Because larval fish are well-adapted to feed on small zooplankton in the water column, use of live prey items such as copepods, rotifers and *Artemia* has resulted in dramatic improvements in the rearing success of several fish species, enabling larvae to reach the juvenile stage when they are able to



feed efficiently on processed diets. However, several challenges still limit the use of live feeds, in particular commercial availability, expense and nutritional profile.

Among live feeds, *Artemia* is widely used because it is commercially available as canned resting eggs (cysts), but it is labour-intensive to prepare, expensive and nutritionally highly variable. Moreover, its large size and rapid swimming speed make it difficult for first-feeding stage larvae to capture. Copepods still remain quite difficult to culture at high densities, requiring large tanks with a large footprint to provide the quantities sufficient to rear fish at a large scale. However, rotifers present many advantages that have been extremely beneficial for rearing a growing number of marine fish species among 10 genera (Table 1). They are used extensively in aquaculture and aquariums because of their very high reproductive rates (as great as doubling or better every 24 hours), ease of culturing, optimal size for many larval fish and a nutritional profile that can be tailored to the needs of prey species by use of special feeds (such as Reed Mariculture's RotiGrow and N-Rich feeds).

BIOLOGY OF ROTIFERS

Rotifers are small (50–1000 µm) zooplankton that occur in freshwater, brackish and marine environments. Rotifers feed on particulate matter

such as microalgae and are consumed by a wide variety of fish, shellfish, corals and other organisms. The most commonly used saltwater rotifers are species of *Brachionus*, *B. plicatilis* (large, 'L-type') and *Brachionus rotundiformis* and several poorly circumscribed species (smaller and super-smaller, 'S-type and SS-type'). They swim and feed by beating of cilia that are grouped into the corona, surrounding the mouth. The 'foot' has adhesive glands, for attachment (Fig. 1). The single red eyespot is visible (Fig. 2) in the right-hand specimen and in one developing embryo. Rotifers are equipped with a 'mastax' chewing mechanism that can break walls of algae, enabling them to digest such foods much more efficiently than other small aquatic animals and protozoa (Fig. 3). The food passage time through the gut is extremely fast (only ~45 min), giving them the capacity to consume 10 per cent of their dry weight per hour with a biomass conversion exceeding 30 per cent. Their lifespan is about one to two weeks depending on temperature. Their lifecycle time is about 18 hours from egg to producing eggs. During the first two to five days of their life, female rotifers mostly produce eggs by parthenogenesis, giving birth to genetically identical offspring. In the domesticated strains used in aquaculture, only in very stressful conditions will males be produced and the rotifers reproduce sexually and create resting eggs. Parthenogenetic eggs are normally carried by females until they hatch, but eggs are physiologically independent (Fig. 1). 'Saltwater' rotifers are native to estuaries and inland saline environments, and can thrive in a wide range of salinities. All together,

Table 1: Genera that have been successfully reared in human care using rotifer for early stage feeding

Family	#spp
Acanthuridae	2
Pomacentridae	12
Pseudochromidae	3
Gobiidae	8
Apogonidae	8
Syngnathidae	12
Blenniidae	2
Pomacanthidae	1
Labridae	4
Plesiopidae	2
Scianidae	2

these features make *Brachionus* rotifers particularly versatile and easy to produce in large quantities to meet the needs of larvae of a wide range of species.

CULTURE

A vigorous culture consists of exclusively females carrying eggs that are produced by parthenogenesis. Only when a culture is severely stressed (temperature, salinity, pH, low oxygen, high ammonia) are males and sexual females produced. Males lack a digestive system and remain small. They mate with sexual females, which produce special eggs, 'resting cysts', that can survive for several years. Sexual reproduction is extremely rare in well-managed cultures and indicates that something has gone seriously awry.

A significant daily harvest (25–50 per cent) is critical to maintain a highly productive culture. This prevents accumulation of less nutritious, older rotifers, which reduce the average reproductive rate. Optimal culture conditions should be followed to keep high yield of production (Table 2).

Table 2: Parameter values for rotifer culture

Parameter	Optimal range	Tolerance
Temperature (°C)	26–27	10–30
Salinity (g/L)	15	3–40
Oxygen (mg/L)	2–8	>1
pH	7.0–8.5	6.5–9.0
Ammonia (NH ₃) (mg/L)	0–<1	<1

CULTURE SET-UP

A 20-litre bucket is a typical set-up for a small system. They are inexpensive, easy to move around and easy to clean. Such a bucket, containing 15 litres of water at 1 million rotifers/L will hold roughly 15 million rotifers, and yield five million rotifers per day with a 33 per cent daily harvest. This quantity is enough to feed approximately 1000 fish larvae.

Whatever type of vessel you use, from 20 litres up to 1000 litres, you will need an aeration system, a waste trap to get rid of debris, and an algae feed distributor (i.e. a peristaltic pump controlled by a timer). A list of culture equipment for both large- and small-scale cultures can be downloaded at <http://www.jove.com/pdf-materials/53629>.

Rotifers do not intrinsically possess high nutritional value – rather they act as 'nutrient carriers' for conveying the high-value essential fatty acids (ARA, EPA and DHA) and other nutrients from the microalgae to the target species. Although you can feed rotifers a variety of feeds such as yeast, the rotifers will only be as nutritious as the feed they have ingested – in other words, they are what they eat. A rotifer that is fed insufficient or low-quality feed will provide little nutritional value to your larvae.

Marine microalgae are widely recognised as the best feed for growing and enriching rotifers. Microalgae are what rotifers naturally feed on in the wild, and provide the complete chemical composition that larval fish need for proper neural development. Microalgae are also the easiest feed to work with. Yeast and emulsion products rapidly foul a rotifer culture, creating high levels of bacteria and ammonia and causing the rotifers to stick together. Microalgae such

as *Nannochloropsis* have a cell wall that resists bacterial breakdown so there is no fouling, excessive bacterial proliferation or stickiness.

FEEDING LARVAE

The literature is quite vague regarding the number of rotifers to give to larvae as soon as they can feed on exogenous preys, ranging from 1000 fish per day to 5000 fish per day at a density around 20 rotifers/ml. But whatever the protocol used, the results are quite spectacular in term of growth, survival and sexual maturation for more than 84 marine fish species.

CONCLUSION

Rotifer is ideal zooplankton for first-feeding larval fish that require enriched feed to support their needs for HUFAs and other critical factors. The possibility of culturing them at a very high density combined with the small footprint and ease of culture make them very economical for meeting the large demand for larval feeding, and even post-larvae and adult fish.

Because of all these advantages and the versatility of rotifers in feeding a wide number of marine (and fresh) water species, this zooplankton offers considerable hope for improving the breeding protocols that will take the pressure off the wild while also allowing the beauty of such environments to be displayed in public and private aquariums.

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A DIET FOR DRAGONS

HOW REVISING THE DIET OF COMMON SEA DRAGONS HELPED TO REDUCE MORTALITY DURING RELOCATION

Kristina Johansen, nutrition consultant, UK, and Kasper Jørgensen, Den Blå Planet, Kastrup, Denmark

Den Blå Planet, a large aquarium in Denmark, had experienced a high mortality in its newly acquired group of sea dragons (*Phyllopteryx taeniolatus*), close relatives of the seahorse. Sixteen out of 20 sea dragons died within a short period after arriving at the aquarium.

The autopsies revealed infestation with opportunistic parasites *Uronema* spp.. *Uronema* are often seen on recently captured, transported or otherwise weakened hosts. Most aquatic species are very sensitive to changes in their environment; any changes in, for example, temperature or water quality can challenge the immunological defence system and render the fish susceptible to opportunistic parasites and other diseases. Changes in the gut microbiota could play a role in this (Wang *et al.* 2016). The fact that the *Uronema* caused such widespread disease and death in the group could have been indicative of stress and an immunocompromised status.

The relocation had been carefully planned and all measures had been taken to minimise the stress of the sea dragons, but even small changes in environment can cause stress. Consequently, the high mortality was likely to be related to transport and the change in environment for the newly introduced sea dragons, despite the many precautions taken.

OPTIMISING THE DIET

To increase the chances of survival for a new group of sea dragons that was scheduled to arrive at the aquarium,



a decision was made to optimise the diet. Information on the optimal nutrition of sea dragons is scarce. Reduced feed intake had been noted in the first group, as is often the case for seahorses and other fish under stressful conditions. A comparative approach was adapted, using sea horses and other aquatic species as a model. Ringø *et al.* (2015) have collated data on the effect of dietary components for maintaining a healthy gut microbiota in various species. Chang and Southgate (2001) found a significant correlation between mean survival of *Hippocampus* spp. juveniles and dietary n3-highly unsaturated fatty acids (HUFA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) contents. They recommended 9.3 mg DHA g⁻¹ dry weight. Other studies have examined different fatty acid regimes, such as Saavedra *et al.* (2016). The following strategy was adopted to provide increased nutritional support:

- Increase feed intake and prevent energy depletion.

- Support the immune system:
 - support the gut – a healthy gut helps maintain a healthy immune system
 - optimise fatty acid intake and ratio
- Increase level of antioxidants to counter the formation of free radicals.
- Increase available high-quality protein.
- Balance vitamin and minerals in the diet.

FEEDING LIVE PREY

Feeding live prey plays a large role in many public aquariums, as some species of fish and invertebrates are very selective and will eat only live prey. Additionally, live prey is crucial in rearing the early life stages of many species. The nutritional quality of live prey varies significantly. One way of improving the nutritional composition of live prey is to use 'gut-loading', in which the prey is given an enriched diet just prior to feeding them to the target species.

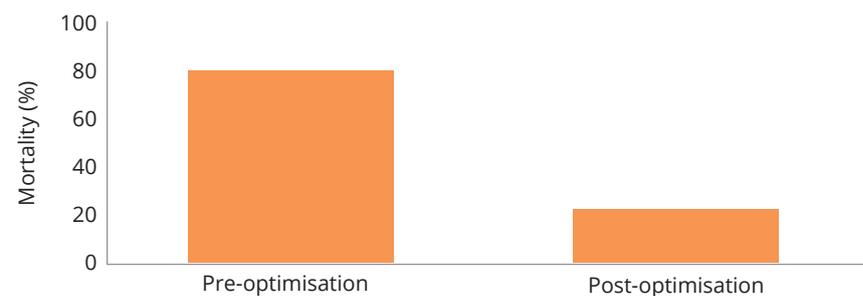
These particular sea dragons were



accustomed to eating only live prey. Their diet in the wild consists mainly of tiny shrimp-like creatures called mysids, of which they consume as many as 1000 per day. For practical purposes the aquarium used live mysids. The new strategy was to gut-load the mysids prior to feeding them to the sea dragons, as this appeared to be the only feasible way of changing the sea dragons' diet. The diet fed to the mysids could be tailored to increase their nutritional value.

The new diet was developed as a joint effort between nutritionists, feed producers, veterinarians and the sea dragon keepers. A gel was considered the best vehicle for the diet, to accommodate the eating habits of the mysids. The gel was formulated to include fish meal, salmon oil and *Spirulina* to achieve high-quality proteins, as well as omega-3 fatty acids such as DHA and EPA. Mannan oligosaccharides (MOS) were included, as studies have shown a beneficial effect on gut flora and villi growth in other aquatic species (such as

Figure 1: Effect of diet optimisation on common sea dragon mortality following relocation to aquariums.



Staykov *et al.* 2007). Selenium levels and vitamin E levels were adjusted for antioxidant properties, as studies have shown that they have beneficial effects, especially under stressful conditions (e.g. Küçükbay *et al.* 2009). Micronutrients were balanced according to the recommended amounts for comparable species where information was available, and taking into account that many minerals are obtained via the water as well. The mysids to be fed out to the sea dragons were removed from their holding tank and placed in a bowl. Here the 'sea dragon gel' was fed to the mysids for gut-loading purposes before the enriched mysids were fed to the sea dragons as their staple diet.

RESULTS

The morbidity and mortality was considerably lower after the new feeding regime was implemented; the loss dropped from 16/20 to 4/20. This is a reduction of 60 per cent mortality when comparing post-optimisation versus pre-diet optimisation (Fig. 1). In addition, the overall health of the sea dragons appeared to improve.

CONCLUSION

This case indicates that optimising diets for aquatic animals could greatly improve health and survival rates. Once the practical implications of creating the gel and encouraging the mysids to eat had been solved, diet optimisation was an easy and simple way of counteracting some of the stress-related challenges of relocation. These results might be relevant for other aquatic species, as

changes in environment affect most animals living an aquatic life.

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KITCHEN SECRETS

THE ADVANTAGES FOR ZOOS OF A CENTRALISED NUTRITION CENTRE

Cora Berndt, nutritionist, Wildlands Adventure Zoo Emmen, the Netherlands

In 1985, Emmen Zoo opened the first centralised zoo kitchen in the Netherlands. A centralised system turned out to have many advantages, and efficiency was greatly increased in numerous ways. Since then, many visitors from all over the world have visited Emmen to learn from the smooth running of the kitchen.

Just over 30 years later, in 2016, we had the opportunity to build an even better version at the new Wildlands Adventure Zoo in Emmen. Many improvements were made, including a more accessible location, kitchen layout and routing, better transport possibilities and improved staff safety and sustainability.

We have found a centralised system to be very efficient and effective, with many advantages and only one disadvantage.

ADVANTAGES

Purchase of products: One person carries out the purchasing and has a good overview as well as control of the budget.

Waste management/cost-efficiency: Leftover feed items are reduced to a minimum.

Hygiene: All food trays are collected and cleaned in a central cleaning system; the dishwasher is situated in such a way that contamination of the rest of the kitchen is prevented.

Staff efficiency: A small number of very focused staff members are in charge of nutrition. Three staff members work seven days a week. They prepare diets (8h/day), distribute feed the next day (2.5h/day), collect fresh grass (1.5h/day), collect browse (3h/day) and clean (4h/day).

Diet and nutrition control: All animals receive exact quantities of their diets. A nutritionist (12h/week) specifically calculates all the diets, in consultation with the veterinarian.

Database: A Microsoft Access database is used for all individual



diets. This database provides final recipes that are projected onto computer screens in the kitchen, making feed preparation very easy.

The only disadvantage is that the physical distance between the kitchen and the animals and their keepers makes good communication essential. This is the only way that changes can be made quickly when they are needed (for example, for the feeding of diseased animals or other diet tweaks).

In 1985, the keepers at Emmen Zoo were initially resistant to the idea of having a centralised kitchen, as preparing food was considered one of the specific tasks of keepers. However, as they became used to the new system, they were able to see its advantages. Nevertheless, good communication between keepers, kitchen staff, nutritionist and veterinarian remains essential to ensure that this way of working remains a success!

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SOLVING THE RHINO PROBLEM

A SUSCEPTIBILITY TO IRON STORAGE DISEASE MEANS THAT THE BLACK RHINOCEROS IS IN NEED OF SPECIAL CARE

Marcus Clauss, Clinic for Zoo Animals, Exotic Pets and Wildlife, Vetsuisse Faculty, University of Zurich, Switzerland

Of the three rhinoceros species typically displayed in zoos, the black rhino (*Diceros bicornis*) is something of a problem child, as it appears susceptible to a variety of demographic and health issues that are not observed in other rhino species, and for many of which the causes are not well understood⁴. These comprise a sex ratio at birth skewed towards males⁵, hemolytic anemia¹, leptospirosis¹¹, vesicular and ulcerative dermatopathy⁹, hypophosphatemia and hyperinsulinemia¹⁶ and abnormal tooth wear¹⁷. Maybe most prominently, black rhinos have long been considered susceptible to iron overload or iron storage disease (ISD)^{3,13}.

It should be noted that these health issues seem to be perceived differently in the United States and

Europe, in particular with respect to ISD. Whereas recent US husbandry guidelines⁷ prominently mention ISD – extensively referencing the workshop on the problem that was hosted by Disney's Animal Kingdom in 2010, and which was published as a complete special issue in the *Journal of Zoo and Wildlife Medicine* in 2012 – the recent EAZA best practice guidelines¹⁴ mention ISD in only two sentences, with a single older reference. This might be caused by an assumption that ISD does not represent a problem in the European black rhino population.

However, on a global scale, ISD in black rhinos is most likely a problem of being kept in human care, and is definitely an issue in Europe. It should not be forgotten that one of the first reports of the problem originated from a UK collection⁶. In

addition, a comparison of serum samples from free-ranging animals and from rhinos kept at a UK facility showed a dramatically higher transferrin saturation in the latter group⁸. Cases of ISD as determined at necropsy have been reported in four 'Swiss'¹⁵ and five 'German'¹² black rhinos. Published evidence for an absence of ISD in the European population is completely lacking.

The fact that the causes for ISD in black rhinos are not fully understood should not serve as an excuse not to address the problem from a practical point of view. Feeding recommendations that aim at limiting iron-rich components of black rhino diets, such as compound pelleted feeds of uncontrolled iron levels, have been put forward², and recommendations on how to perform phlebotomy (bloodletting

– the only way to get iron out of the body) on black rhinos have been produced¹⁰ and facilitated by corresponding training programmes. Training black rhinos for bleeding without physical or chemical restraint is labour-intensive but possible; it can also be, one might add, a highly rewarding way to interact with these animals, and an occasion where animal keepers can actually apply their skill in animal interaction.

FUTURE PROGRESS

Progress in black rhino management most likely will depend on viable access to blood: not only for phlebotomies to take out iron, but also to gain insight into the prevalence and progression of ISD and the many other health problems reported in this but not other species. Training black rhinos for blood sampling is important not only for when an individual might become ill, but also for prophylactic screening, and especially – if the animal is clinically healthy – as a control to compare with animals diagnosed with a problem. Studies concerning health issues also need samples of healthy individuals as control. While it is not necessarily the task of zoos themselves to initiate research into the health issues of black rhinos, any research will largely depend on the availability of blood samples. For an *ex situ* population of a species with such diagnostic and therapeutic challenges, zoos are in a position to facilitate such research by ensuring that the necessary material can be easily collected.

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BEAR NECESSITIES

IS THERE A LINK BETWEEN DENTAL HEALTH AND ORAL pH IN BEARS?

Francis Cabana and Claudia Tay, Wildlife Reserves Singapore, and Kirsty Officer, Chuon Vuthy and Nev Broadis, Free the Bears, Cambodia

Asiatic black bears (*Ursus thibetanus*) have a largely herbivorous diet, supplemented by insects and opportunistic mammals such as ungulates (Narita *et al.* 2006). This species has a seasonal diet with periods of high fruit abundance and consumption alternating with periods of nuts and seeds selection (Huygens *et al.* 2003). Oak acorns, stone pine seeds and walnuts are ingested during this period, providing a diet high in protein, lipids and easily digestible starches (Huygens *et al.* 2003; Hwang *et al.* 2006). Sun bears (*Helarctos malayanus*) are known as frugivores due to their dependence on periodic mast-fruiting events when fruits make up almost 100 per cent of their diet (Fredriksson *et al.* 2006). These tropical fruits are particularly succulent and rich in fibre, fat and protein (Voon and Kueh, 1999; Wong *et al.* 2002).

Besides fruit, sun bears also consume a variety of small mammals, insects and a certain amount of honey (Wong *et al.*, 2002). Fruits and seasonality are a common theme amongst these bears' diets, and diets provided to bears in human care have focused on the moderate protein content (10 per cent of diet on a dry matter basis requirements: Cicjnak *et al.* 1987) and assumed high soluble carbohydrate requirement. Interestingly, many of the food items consumed in the wild also contain a moderate to high fibre content and are higher in starches than sugars. Food items deemed to be fallback foods have not been integrated into *ex situ* diets.



In human care, Asiatic black bears and sun bears are minimally affected by the changing seasons compared to their wild counterparts. The amount and type of food is fixed, with water available throughout the day. Bears are often fed high-sugar diets consisting of commercial fruits and sometimes boiled eggs, cooked rice, cooked meat, vegetables and bread (Vickery and Mason, 2004). Fruits given tend to be high in sugar, especially as these fruits are cultivated for human consumption, and tend to make up the bulk of the bears' daily food intake.

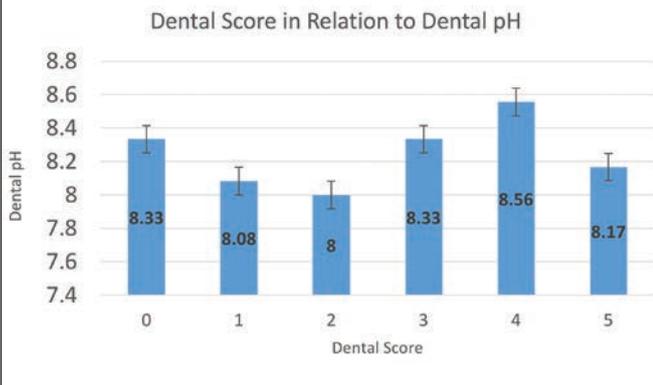
Captive bears with some level of health problems are not unusual in the zoo world. Obesity, diabetes, cancer, systemic hypertension and eye disease have occurred in the captive population. Obesity and dental disease are common, occurring in the majority of zoos for all species of bears. All of these issues can, at least in part, be linked to diet. Bacteria that inhabit the bear's oral cavity feed on sugars in the bear's diet. Naturally a diet higher in sugar is linked with a larger amount of dental disease, which can be initially assessed by the amount of dental plaque (Sheiham, 2001). After digesting the sugars, the microbes excrete organic acids. If there is constant and enough sugar in the diet, bacteria (and plaque) will easily reproduce and

spread across the teeth (Brathall, 1996). When found in large enough amounts, the production of organic acids may be high enough to actually change the pH of saliva. With the protective effects of saliva neutralised, bacteria are free to erode the surface of teeth, expose the inner dentine layer and make entrances for more pathogenic microbes. There is no information yet on whether the change in pH is merely a microclimate surrounding the bacteria, or whether the pH of the entire oral cavity will be more acidic if the bacteria are found in large enough quantities.

As noted, most bear diets contain a large amount of fruit, and some zoos also base their diets on a gruel or porridge mixture. The large concentrations of soluble carbohydrates that these diets contain may be the cause of dental issues. We set out to determine whether there was a link between dental disease in bears and oral pH through a pilot study.

We collected oral pH data from bears in Wildlife Reserves Singapore (WRS) and Free the Bears in Cambodia (FTB). Our population was 1.2 Asiatic black bears and 20.21 sun bears. Each bear was trained to present its open mouth between bars and we used a litmus paper to measure the pH. At WRS, four strips were used per sampling session, one

Figure 1: Dental scores of 46 bears according to their dental pH, showing no trend or correlation between both variables



on each upper canine and one on the upper and lower incisors. Four independent people looked at the colour of each strip and assigned them a matching pH. The average was taken for each strip and the average of the four strips per session was taken for an overall pH score of that session. Each bear was sampled a total of four times before any food was given. At FTB, each bear was tested three times and had the litmus paper read by one person.

Each bear's teeth were given a score by two vets using a developed dental scoring guide. Grade 0 was described as healthy gums with no evidence of gingivitis with white teeth and no evidence of plaque or calculus. The gums and teeth are progressively worse through to grade 6, which was described as severe with extensive plaque and calculus, severe gingivitis with ulceration, extensive gum pockets and recession, furcation exposure and multiple unstable teeth.

The pH of the sun bear mouths sampled varied between 8 and 9, and the Asiatic black bear oral pH varied between 9 and 10. We found no correlation between oral pH and dental score of the bears (Fig. 1). These preliminary results are from a small sample size; we were not able to detect a trend. The decrease in oral pH may be more localised to microclimates surrounding the bacteria as opposed to overall oral cavity pH. In other words, the lower pH which is reported to be a crucial

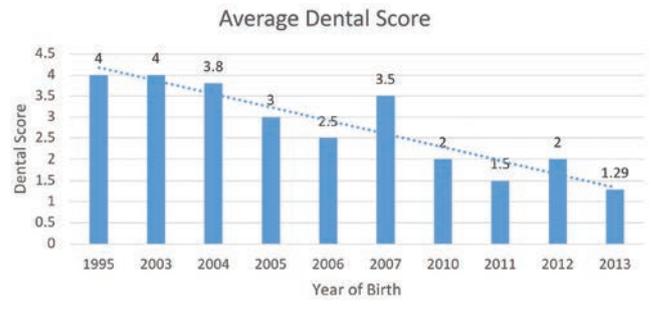
step in the formation of caries may only be in the bacteria's immediate surroundings. This may be more conducive to protecting the bacteria and allowing them to erode the dental enamel. We did notice a relationship between dental score and age of the bear, with older bears having a significantly higher dental score (meaning worse dental health) compared to younger bears (Fig. 2). This is expected since the older the bear, the longer it would have been ingesting a diet high in soluble carbohydrate, allowing bacteria to progressively reproduce and plaque and calculus to build up.

Contrary to popular belief, bears in captivity do not need to have high fruit components in their diets. Diets of the bears at WRS are now free of grains and low in fruit. The diet is mainly vegetables, browse and omnivore pellets, with a daily rotation of meat, eggs, silkworm, mealworms, ant eggs and local jungle fruits. Faecal scores have become ideal and body weights have stabilised. We expect these diets to help prevent dental disease, but monitoring must continue.

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Figure 2: Average dental scores arranged by age of the bears showing a clear trend of worse teeth (higher score) on older bears.



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