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Some Analyses and Recommendations on Diet Formulation for Conservation Breeding of the Galapagos Rice Rat of Isla Santiago, *Nesoryzomys swarthi*

Dan Wharton,^{1*} Robert Dowler,² and Jennifer Watts³

¹ Conservation Science, Chicago Zoological Society, Brookfield, Illinois

² Department of Biology, Angelo State University, San Angelo, Texas

³ Nutrition Services, Chicago Zoological Society, Brookfield, Illinois

Nesoryzomys swarthi, the most endangered of the three surviving, endemic Galapagos “rice rats,” was only discovered in the early 20th Century and was considered extinct until its rediscovery in 1997 at a north-central coastal location on Isla Santiago. Potential threats to the entire genus include invasive rodent species, feral cats, new diseases, and climate change. These threats have been the basis for conservation breeding recommendations (as yet unmet) by several observers during the last several decades. This paper considers likely dietary requirements of *N. swarthi* in light of recent studies on the ecology of this species plus new data on the nutrient composition of *Opuntia galapageia* (a “resource refuge” for this species) and circulating vitamin values of animals sampled on Isla Santiago. It is concluded that, despite some unusually high mineral values for *O. galapageia*, a diet for *N. swarthi* under human care should be the same as it is for most other rodents, noting some caution in regard to possible needs for mineral and/or protein adjustment. Zoo Biol. 31:498–505, 2012. © 2012 Wiley Periodicals, Inc.

Keywords: *Nesoryzomys swarthi*; *Opuntia galapageia*; conservation breeding; diet formulation

INTRODUCTION

Nesoryzomys swarthi, the most endangered of the endemic Galapagos “rice rats,” was only discovered in the early 20th century on Isla Santiago and was considered

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*Correspondence to: Dan Wharton, Department of Conservation Science, Chicago Zoological Society, 3300 Golf Road, Brookfield, IL 60513. E-mail: dan.wharton@czs.org

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extinct at approximately the same time it was described to science in 1938 [Heller, 1904; Orr, 1938]. Until recently, it has been assumed that the introduction of non-native rodents to Galapagos islands automatically caused rapid population decline and extinction of the native rodents [Key and Munoz, 1994]). However, in 1997, a small population of the Santiago rice rat was discovered still living at a north-central coastal location (locally known as “La Bomba”) on Isla Santiago, coexisting as a relict population with introduced rodents, i.e. the black rat, *Rattus rattus* and the common house mouse, *Mus musculus* [Dowler et al., 2000]. It is suggested by experimental evidence that continued survival of *N. swarthi* in a 14-km² stretch of the north central coast of Isla Santiago is its superior adaptation to the desert conditions of the Galapagos, magnified in the rain shadow of the north central coast of Santiago [Harris and Macdonald, 2007]. High density of the endemic cactus, *Opuntia galapageia*, in this same 14 km² appears also to be a major factor in *Nesoryzomys*' critical survival advantage in this microhabitat. Fruits and pads in the wet and dry seasons, respectively offer a “resource refuge” that ameliorates *Rattus* foraging interference by aggressive encounter that typifies the relationship of *Rattus* with *Nesoryzomys* [Harris and Macdonald, 2007]. *Rattus rattus* is apparently unable to consume *Opuntia* cactus fruits at all, and little of the pads [Gregory and Macdonald, 2009; Harris et al., 2006]. *Rattus rattus* regularly experiences population crashes in this arid coastal zone, offering periodic respite for *N. swarthi*.

Historically, the Santiago rice rat could have existed in all parts of Isla Santiago, as is more or less the case with two other members of the genus, *N. narboroughi* and *N. fernandinae*, on Isla Fernandina [Dowler et al., 2000]. If *N. swarthi* previously had island-wide distribution, it most likely experienced a dramatic reduction in range and numbers as soon as *R. rattus* was introduced as early as the 1600s [Patton et al., 1975]. Current threats to the survival of *N. swarthi* include a shift in ecology, particularly climate change and a higher frequency of wet El Nino years that would favor the invasive, common Old World rats and mice [Harris, 2006]. Other potential threats include new diseases, introduction of additional rodent species, or new predators, feral cats being among one of the biggest threats now resident in several of the Galapagos islands [Dexter et al., 2004; Harris et al., 2006]. Furthermore, any island-wide eradication attempts of invasive rodents have potential risk implications for endemic rodents. *Rattus* eradication programs are already in place or in planning for several islands [Harper and Cabrera, 2010; Platt, 2011].

Conservation breeding has been recommended by several observers for the current status of, and threats to, *Nesoryzomys* [Dowler and Carroll, 1996; Dowler et al., 2000; Trillmich, 1986]. While conservation biologists often label conservation breeding as the strategy of “last resort,” this is too often translated to mean “last minute” when chances of success are often reduced by a concomitant lack of animals and the necessary experience in their breeding management [Wharton, 2006]. However, the list of species brought back from the brink of extinction via conservation breeding continues to grow and includes several species with fairly complex captive needs such as the black-footed ferret, California condor, and Guam kingfishers [Grenier et al., 2007; Kessler and Haig, 2004; Walters et al., 2008]. Endangered rodent species have been the subject of captive breeding in several instances with good results [Alligood et al., 2011; Roest, 1991]. Other Oryzomine rodents have also bred successfully in captivity [Hamilton, 1946; Voss et al., 1992].

Oryzomys bauri, of Isla Santa Fe, is the only Galapagos rodent that has been brought into captivity to date. A total of 14 individuals of *O. bauri* were transferred to Isla Santa Cruz and all died within a week of a “mystery illness” [Brosset, 1963; Harris, 2009]. It has been presumed that they died from disease contracted via observed contact with black rats although inadequate husbandry is an alternate explanation.

Whether or not relatively permanent, scientifically managed captive populations of *Nesoryzomys* are deemed desirable or necessary, it would appear that, at the minimum, temporary captive populations ought to be established to secure a body of literature describing captive management in great detail. Should *N. swarthi* or the other two surviving members of the genus on Fernandina (*N. narboroughi* and *N. fernandinae*) experience population crashes, well-documented experience in the captive management and breeding of at least one member of the genus will be invaluable in maximizing the likelihood of success of conservation breeding as a species rescue strategy.

It is the objective of this paper to offer insight on captive diet formulation for *N. swarthi* through (1) a brief review of existing published studies on the foraging ecology of the *N. swarthi*; and (2) some new information on nutrient composition of *O. galapageia* and circulating vitamin levels found in wild living *N. swarthi*.

MATERIALS AND METHODS

Reports on the preferred and selected foods of *N. swarthi* as compared to *R. rattus* are found primarily in the work of Gregory and Macdonald [2009].

The *O. galapageia* (one ~100-g leaf sample) was collected in the month of August (Galapagos dry season is June to December) and held under refrigeration for 3 days followed by freezing at -80°C for 4 months. Although this was a single sample, it was taken from a mature plant standing well within the very arid coastal range of all known *N. swarthi*. While soil and moisture conditions for this particular plant sample source are not likely to represent those of all *O. galapageia*, they are typical for all those that fall within the very limited range of the single, small population of *N. swarthi*.

Rodents were collected using Tomahawk and Sherman live traps baited with rolled oats and peanut butter. Animals were handled using a rat restraining device and blood samples were collected from the ventral tail artery [Staszuk et al., 2003], refrigerated for 3 days, and then centrifuged by hand prior to freezing for 4 months at -80°C . Other samples and measures were taken for other studies. All rodent samples were collected during the Galapagos dry season (August), the period in which selected diet is somewhat broader than in the wet season January to May [Gregory and Macdonald, 2009].

The *O. galapageia* and blood samples were sent to Michigan State University Diagnostic Center for Population and Animal Health (Lansing, MI) for analysis. All feed analyses followed established Association of Official Analytical Chemists (AOAC) guidelines for wet chemistry methodology; macro- and microminerals in both cactus and blood samples were determined using atomic absorption spectrophotometry. Vitamin A and E concentrations were determined by High Pressure Liquid Chromatography (HPLC) using butylated hydroxytoluene (BHT) and ascorbic acid as oxidant stabilizers and a mobile phase of acetonitrile:methylene chloride:methanol. The amount of blood collected from each animal was very small; one pooled sample (three

TABLE 1. Nutrient composition of selected domestic produce, domestic *Opuntia* (nopales; from USDA Nat'l nutrient Database), and *Opuntia galapageia* from the home region of *Nesoryzomys*.*

Nutrient	Unit	Spinach	Kale	Broccoli	Domestic nopales	<i>Opuntia galapageia</i>	ProLab ratPellet ^b
ME ^a	kcal/g	2.61	3.22	3.01	2.72	2.64 ^b	3.04
Crude fiber	%	11.10	11.8	13.80	37.41	16.00	5.30
Crude fat	%	4.16	4.50	3.76	1.53	2.90	6.0
Crude protein	%	33.97	21.24	32.01	22.44	5.50	24.0
Vitamin A	IU A/g	93.77	572.7	165.6	77.72	2.28	22.0
Vitamin E	mg/kg	224.47	51.48	178.3	0.0	69.14	52.0
Ash	%	20.43	9.85	9.88	19.39	21.60	6.9
Calcium	%	1.18	0.87	0.52	2.79	7.40	0.95
Copper	mg/kg	15.44	18.66	4.83	0.52	42.00	17
Iron	mg/kg	321.9	109.4	94.5	100.34	78.00	290
Magnesium	%	0.94	0.22	0.27	0.88	1.57	0.25
Manganese	mg/kg	106	49.8	24.6	7.7	26	110
Phosphorus	%	0.58	0.36	0.71	0.27	0.06	0.69
Potassium	%	6.63	2.88	3.49	4.37	0.73	1.20
Selenium	mg/kg	0.12	0.06	0.32	0.1	Insufficient sample	0.48
Zinc	mg/kg	62.95	28.31	42.96	42.52	50.00	110

*All values are on a dry matter basis.

^aME, metabolizable energy; primate energy values are used as the default energy value for domestic food items.

^bThe energy value for the *O. galapageia* was calculated by hand using the proximate nutrient values. The energy value and all nutrients listed for the commercial diet were as reported on the analytical information provided with the diet.

animals) was sent for analysis. It was noted by visual appearance that the hematocrit was between 50% and 60% and the serum was moderately hemolyzed.

Trapping, sampling, and rodent re-release methods were conducted in accordance with the American Society of Mammalogists guidelines for the use of wild mammals in research [Gannon et al., 2007] and the Institutional Animal Care and Use Committee (IACUC), Chicago Zoological Society. Permits for field collection of samples were provided by Parque Nacional Galápagos, Ecuador.

RESULTS

The nutrient composition of *O. galapageia* as compared to raw, domestic *Opuntia* (nopales), spinach, kale, broccoli (all from the USDA National Nutrient Database) and commercial rat pellets (ProLab RMH 2500 (product #5P14)) is reported in Table 1. The *O. galapageia* was lower in crude protein, vitamin A, phosphorus, and potassium compared to other domestic produce, but higher in calcium, copper, and magnesium. The composition of domestic nopales compared to the *O. galapageia* showed remarkable differences in several nutrients such as, crude fiber, crude protein, vitamin A, calcium, copper, phosphorus, and potassium. Crude fiber was significantly higher in the nopales compared to any other diet item listed.

The energy value and all nutrients listed for the commercial lab diet pellets were as reported on the analytical information provided with the diet.

TABLE 2. Circulating values for serum vitamins A, E, and β -carotene in a pooled sample from three wild *Nesoryzomys swarthi*.

Nutrient	Unit	<i>N. swarthi</i>	Reference range*
Vitamin A	ng/ml	76.4	30–60
Vitamin E	μ g/ml	1.95	2–4
Beta-carotene	μ g/ml	0.240	0.2–0.3

*Reference range is from the laboratory rat, *Rattus norvegicus*.

Circulating serum vitamin levels were determined in the sample of blood collected and are represented in Table 2. The volume and quality of the sample was not conducive to evaluating mineral concentrations.

DISCUSSION

Although the study by Gregory and Macdonald [2009] was focused on the dynamics of niche partitioning and resource competition between *N. swarthi* and *R. rattus*, another very useful finding from their work is that the two species of rodent appear to have “shared preferences.” Food selection by the behaviorally subordinate *N. swarthi* was uncorrelated with preference. Selected diet broadened during the dry season when the far less arid-adapted *R. rattus* population crashed and presumably lifted a constraint on ability to forage. Both species feed on insects as represented by mean percent of insect matter in feces, ranging from about 38% to 30% (wet to dry season) for *N. swarthi*. The biggest contrast was, of course, in the use of *Opuntia* fruit and pads by *N. swarthi*, selecting *Opuntia* more than any other plant material during the most *Rattus*-competitive wet season. *Opuntia* was also selected at high frequency by *N. swarthi* during the dry season, perhaps indicating continued interference by *Rattus* or other drivers not yet documented [Gregory and Macdonald, 2009].

Altogether, it is suggested that the prepared diet for *N. swarthi* could be similar to that of other New World rodents. The estimated nutrient recommendations for rodents are: 5–15 % crude protein, 10–30 % crude fat, 0.5–1.0% Ca, 0.3–0.4% P, 3.5–8.0 mg/kg iron, 1.2–2.5 mg/kg zinc, 230 IU/g vitamin A, and 2.7 IU/g vitamin E [NRC for Laboratory Rodents, 1995]. Generally, captive diets consist primarily of scientifically formulated, nutritionally complete rodent pellets designed for the laboratory rat (*Rattus norvegicus*), supplemented with chopped fruits and vegetables (often a mixture of chopped kale, sweet potato, carrot, and apple) and commercially available insects such as house crickets (*Acheta domesticus*) and mealworms (*Tenebrio molitor*). If conservation breeding is pursued where laboratory rodent pellets are not readily available (e.g. Charles Darwin Research Station, Galapagos), commercial, dry dog food is a reasonable substitute, supplemented with locally available produce and occasional, light-trapped insects uncontaminated with insecticide. Three sample diets are presented in Table 3 in order of dependence upon a commercial product.

All three of these diets meet the requirements for protein and fat, although the dog food contains a much higher amount of fat per gram, so care must be taken to evaluate animals' weights. If a manufactured, nutritionally complete diet item is not available, a variety of produce, seeds, complete mineral supplement, and protein source are recommended to provide a balanced diet (Diet 3).

TABLE 3. Examples of three alternate rodent diets to fulfill daily nutrient requirements, per animal for *Nesoryzomys swarthi*.

Diet 1	Grams	Diet 2	Grams	Diet 3	grams
Rodent pellets	20	Dry dog food	15	Tuber	10
Vegetables	15	Vegetables	15	Vegetables	15
Leafy greens	15	Leafy greens	20	Leafy greens	15
Insects (3 times per week)	2	Insects (3 times per week)	2	Insects (4 times per week)	4
				Nuts/seeds	2
				Mineral supplement	

The nutrient profile of the pads of *O. galapageia* presents some potential “red flags” for *N. swarthi* diet management given a coevolutionary dietary history of *Nesoryzomys* with *Opuntia* that probably spans most of the 3.5 million year history of *Nesoryzomys* in the Galapagos [Patton and Hafner, 1983]. Although it is not a given that diet management problems will, in fact, arise from the unusual composition of *O. galapageia*, pads and fruits of *O. galapageia* are the dominant food items of free-living *N. swarthi*, as noted above. The following *O. galapageia* composition features are the ones to keep in mind if nutritional issues arise: low crude protein, low vitamin A, high calcium, high copper, low phosphorous, and potassium. If *N. swarthi* has adapted to the high levels of calcium and copper, transition to a domestic rat pellet or dog food diet supplemented with domestic produce low in these minerals could possibly cause deficiency. These concerns are easily remedied by dietary supplementation. Clinical signs in young animals for both dietary calcium and copper deficiencies are usually a gradual deterioration of health, followed by poor reproduction as they reach sexual maturity. In adults, the deficiencies are characterized as unthriftiness, decreased cardiovascular function, decreased bone density (Ca), and anemia (Cu) [National Research Council, 1995]. Commercial rat pellets contain nutrient values that satisfy the nutrient requirements for the common domestic rodents and although *N. swarthi* may survive, they may not thrive. The commercial rat pellets contain a level of protein that is potentially much higher than the natural diet of *N. swarthi*. These animals have some adaptations for desert living and it is yet unknown if a consistent, relatively high protein content would affect the kidneys.

Other studies of *Opuntia* sp. generally reveal a very high oxalic acid content of fruits and pads and this is considered the major factor for avoidance of, or adaptation to *Opuntia* foods by desert mammals that concentrate urine as a hydration stability adaptation [Verts and Caraway, 2002]. Oxalic acid is a highly reactive carboxylic acid that strongly binds to divalent metal ions resulting in a salt that is also known as an oxalate. The high levels of Ca and Mg in the *Opuntia* are not surprising considering the oxalic acid concentrations, but the effects of consuming these minerals at such high levels have not been pursued.

The low concentration of vitamin A in the *Opuntia* could indicate that wild *N. swarthi* has an adaptation for coping with low Vitamin A consumption. The vitamin A concentration of the pooled plasma sample from the wild animals was higher than the reference range for domestic rodents, possibly indicating that (a) *N. swarthi* absorbs and retains more dietary vitamin A than typical rodents; or (b) obtains vitamin A from other food sources as yet undocumented. The value for vitamin E is most

likely artificially low due to hemolysis, so it is difficult to comment on the circulating concentration of this nutrient. The concentration of vitamin E in *Opuntia* is similar to domestic food items and the commercial diet, therefore it can be presumed that the commercial diet would be adequate in both of these vitamins; however, 3-day storage of blood samples prior to centrifuging and freezing could have given artificially low values for all vitamins due to their general instability as time from collection increases. The pooled serum sample from three animals is essentially $N = 1$ and results should be considered preliminary.

CONCLUSIONS

1. The existing scientific literature on *N. swarthy* suggests that it is a foraging opportunist and has shared food preferences with the common, invasive *R. rattus* of Isla Santiago.
2. A suitable captive diet for *N. swarthy* would most likely consist of the same items fed to other opportunist rodents, especially New World rats.
3. As a food item, *O. galapageia* has a high ash, magnesium, calcium, and copper content as compared to most domestic food plants.
4. The observed dependence of *N. swarthy* on *O. galapageia* foods in their natural habitat may or may not indicate some nutrition-related idiosyncracies that will require special management of *N. swarthy* when under human care.

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