

An Evaluation of the Nutritional Adequacy of the Feeding Program of the Black-Footed Ferret (*Mustela nigripis*) at the Metropolitan Toronto Zoo

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Experimental data or performance record of black-footed ferret (BFF) in captivity is needed to evaluate nutrient levels or adequacy of feeding programs carried out with this endangered species. Diets fed to BFF in three places were compared: Metropolitan Toronto Zoo (MTZ), Sybille Wildlife Research Center (SWRC) and at Omaha's Henry Doorly Zoo (OHDZ). Some reference will be made to prairie dogs (Pd), the primary feed source in the wilderness. Both the GE and the estimated ME values were significantly higher for the MTZ diet compared to the others. Thus, additional fat as recommended by SWRC was considered unnecessary. The high level (5.97% DM) of PUFA in the MTZ diet most likely requires higher vitamin E levels than the 86 IU/kg found in it. Vitamin E in the OHDZ diet was lower and data from the SWRC diet on vitamins was missing. In contrast, the OHDZ contained exaggerated vitamin A levels, about 6-fold the amount found in the MTZ diet. The protein content was found similar in all diets and was above the average 42.5% protein/DM found in Pd. The amino acid profile of the MTZ diet appeared to be adequate. The three diets reviewed seemed to be too high in Ca and the MTZ diet was perhaps too low in P. Overall, an excess of trace minerals was found particularly Fe. Though empirical, it seems some adjustment in the nutrient content of the BFF's diet would be recommended.

Key words: nutrition, black-footed ferret, mustela, feeding, captivity

INTRODUCTION

The black-footed ferret (*Mustela nigripis*) is one of the rarest mammals in North America (Hall, 1981) and is classified by the U.S. Fish and Wildlife Service as an endangered species. Black-footed ferrets (BFF) are small, nocturnal carnivores of the family Mustelidae, in many respects similar (Miller et al. 1988) to other members of this family, the European ferret (*M. putorius*), domestic ferret (*M. putorius furo*) and Siberian polecat (*M. eversmanni*), and mink (*M. vison*). The BFF exists in close cohabitation with the prairie dog (*Cynomys* spp) which should provide its primary source of food (Sheets et al. 1972; Campbell et al. 1987; Richardson et al. 1987; Miller et al. 1988). However, the widespread eradication of the prairie dog and disease outbreak appears to be the primary causes of the BFF decline (Miller et al. 1988).

The BFF Species Survival Management Group has established specific protocols and guidelines regarding diets and feeding strategies for this species recovery program. Initially, all participant institutions were required to strictly adhere to and follow the basic guidelines. Lately, participant institutions have been allowed to introduce minor modifications to the original diet formulation referred to as the Sybille 60/40-R diet. The Metropolitan Toronto Zoo (MTZ) has been participating in this program since 1992 when a group of BFF breeders was received.

This report is an evaluation of the nutritional adequacy of this official Sybille 60/40-R diet and a revision of the feeding program established for this endangered species. Since the nutrient requirements of BFF have not been established, comparison with close relatives, with better known nutrient requirements and the chemical composition of natural feed sources will provide the bases of this nutritional evaluation for the BFF in captivity.

Black-footed Ferret Natural Diet

Black-footed ferrets are highly specialized predators depending primarily on prairie dogs as their main feed source (Sheets et al., 1972; Campbell et al., 1987; Richardson et al., 1987; Miller et al., 1988). A study of the feeding habits of BFF in South Dakota (Sheets et al., 1972) found that 91% of all the scats examined contained remains of black-tailed prairie dogs (*Cynomys ludovicianus*). This represents 86% of the recognizable animal matter by weight, mice was identified as the remainder. Another study conducted in Wyoming (Campbell et al., 1987) found similar results, 87% of the 86 BFF scats examined contained white-tailed prairie dogs (*C. leucurus*) remains. These studies tend to suggest that the natural diet of the BFF consist almost exclusively of prairie dogs.

Nutritional Data-base Evaluated

Feeding programs for BFF in captivity, conducted at two different wildlife centers, the Sybille Wildlife Research Center (SWRC) and the Omaha's Henry Doorly Zoo (OHDZ), were compared to the MTZ feeding program. The complete chemical composition of the main ingredients included in the MTZ diet for BFF was determined. These include Gro-FurDarks 10 and 20% fat mink feed, rabbit carcass and blood meal. The nutritional merit of the MTZ diet was assessed by comparison to commercial pellets and natural ingredient diet for domestic ferrets (McLain et al., 1988). When appropriate the MTZ diet was also contrasted with the nutrient requirement of mink (National Research Council, 1982). Some reference to the prairie dog mineral contribution (Dierenfeld, E.S. & J. T. McGuire, 1989) to the BFF diet was also evaluated. Original data presented in this report on chemical composition of diets or feed ingredients were obtained from representative samples collected at the MTZ and analyzed at the Crampton Nutrition Laboratory (CNL) of McGill University, Montreal. All samples were prepared for analysis in duplicate according to official methods (OAC, 1980) or the most recent method available for specific analysis (CNL Current Methods, 1995).

Chemical analyses of the nutrients content in the National Complete Mink Pellets with 10 and 20% fat (Gro-Fur Darks) manufactured by Milk Specialists Co. (New Holstein, WI 53061, USA) were obtained from their own commercial information reports. Chemical composition of the ground whole dressed carcass of the rabbit and blood meal purchased at the MTZ was conducted at the CNL during 1993. Samples of the MTZ diet fed to the BFF corresponded to formulation #2 effective from September 30, 1993 was identified as MTZ 495-93 for the laboratory. Analysis data for the SWRC diet was adapted from the SSP Sybille Black-footed Institution Nutrition Report (1993). The laboratory report provided in this document indicates that the analysis corresponds to the regular 60/40-R diet with or without the "optional additives". Analysis data on the diet fed to BFF at the OHDZ was adapted from information on ingredient composition and chemical analysis of the diet done at the Nebraska Testing Corporation on samples dated January 1, 1993. Original analysis data was converted to dry matter basis.

The nutrient requirements of mink (NRC, 1982), the composition of the Prairie dog carcass as BFF's natural feed source (Dierenfeld & McGuire, 1989) and the nutrient composition of commercial pellets and natural ingredient diets for domestic ferret (McLain et al., 1988) in which nutrient concentrations seem to provide for acceptable animal performance in captivity are the best reference values for nutritional evaluation of the MTZ diet for BFF.

Nutritional Characterization of the Basic Ingredients Incorporated in the Diet for Black-footed Ferret

Since no controlled research has been conducted to determine the nutrient requirements of BFF, formulations are tentative, on the basis of the best available information on nutrient requirements of a close relative, the mink (*Mustela vison*), complemented with data on chemical composition of the prairie dogs (Dierenfeld & McGuire, 1989), the primary feed source of free-ranging BFF. Data from commercial formulations for domestic ferrets (*Mustela putorius furo*) in which dietary nutrient concentrations seem to provide for acceptable animal performance (McLain et al., 1988) are used.

The mink pellet (Table 1) represents the main ingredient in the formulation of BFF diets (Table 3). As dry matter, it contains high protein (> 38%), high fat with a choice of two levels (> 10% or > 20%). The protein profile includes a relatively high level of arginine, leucine and lysine among the essential amino acids, characteristic of the profile of amino acid requirement of domestic carnivorous animals (dogs, NRC-1985; cats, NRC-1986). The presence of reducing sugars limits the availability of some amino acids, particularly lysine. Processed by-products and/or during pelleting of the compounded feed, use of excessive heat reduces the protein quality. The high fat content of these pellets provides the BFF diet with high energy density ingredients. The mineral requirements of mink are not well defined. The high levels of Ca and P make these pellets adequate for BFF. The trace mineral concentrations are present in much higher levels than required for dogs and cats. Therefore it appears to be more than sufficient for BFF. These pellets are also good source of vitamin A but the low content of vitamin E suggests the need for supplementation of this vitamin.

The chemical composition of the rabbit's carcass, another main ingredient in the diet of the BFF and blood meal a frequent minor one in the diet of BFF (Table 1) indicates that the former is a good source of protein, fat and minerals, while the latter is mainly a good source of protein and iron. Blood meal is also an exceptional source of leucine and histidine.

TABLE 1. Chemical analysis of the nutrient content in the ingredients included in the diet formulated for Black-footed Ferrets

Analysis	Gro-Fur Darks/Mink Feed ²			
	20% Fat	10% Fat	Rabbit Carcass ³	Blood Meal
Dry Matter, %	90.98	91.69	26.16	89.42
Crude Protein, %	38.16	41.72	65.20	98.41
Crude Fat, %	23.96	11.23	15.78	0.06
ND Fibre, %	10.99	19.96	0.10	0.18
AD Fibre, %	5.17	5.76	0.10	0.18
Ash, %	2.22	2.33	3.36	0.25
Gross Energy, kcal/g	5.68	4.68	5.30	5.73
Arginine, %	2.83	3.00	4.70	5.25
Histioine, %	1.18	1.23	2.60	8.06
Isoleucine, %	1.39	1.63	2.41	1.42
Leucine, %	3.29	3.71	4.67	12.37
Lysine, %	3.24	3.71	5.40	3.06
Methionine, %	0.84	0.75	1.13	0.52
Phenylalanine + Tyrosine, %	2.95	3.34	2.36	8.99
Threonide, %	1.94	2.15	3.05	4.28
Valine, %	2.15	2.43	2.92	8.21
Calcium, %	1.84	2.29	5.93	0.04
Phosphorus, %	1.07	1.11	3.43	0.19
Magnesium, %	0.17	0.16	0.18	0.02
Potassium, %	0.51	0.59	0.72	0.27
Sodium, %	0.66	0.55	0.26	0.31
Iron, ppm	800	556	100	1877
Zinc, ppm	137	183	84	9
Manganese, ppm	90	124	2.4	1.3
Copper, ppm	23	34	4.6	7.8
Selenium, ppm	1.27	1.69	1.18	1.39
Vitamin A, IU/g*	20.16	39.33	6.20	10.00
Vitamin E, IU/g	0.04	0.05	n.d.	0.05

¹ Nutrient content on D.M. Basis

² National Complete Mink-Food Pellets, Manuf. Milk Specialties Co, New Holstein, WI, USA

³ Ground whole dressed-carcass

* Vitamin units 1 I.U. Vitamin A = 0.3 μ g retinol

The fatty acid profiles of the fat content in the ingredients (Table 2) included in the diet formulated for BFF indicates that the mink feed diets are highly unsaturated (>40%), while rabbit carcass is highly saturated (> 80%) and blood meal is more balanced but less important because of the low fat contribution. This high unsaturation of the mink diets most likely indicates supplementation with vegetable oils.

Comparative Nutritional Merits of Different Black-footed Ferret Diets

Major Apparent Nutritional Differences

Since the three BFF's diet presented in Table 3 have been formulated with similar ingredients, their nutritional value should be considered very similar except for some variation due to specific supplements such as Bio-liver for vitamin A in the OHDZ diet. This diet has about a 6 fold higher vitamin A content than the MTZ diet. The lower ash content of the MTZ diet compared to the other two diets, due mostly to lower ground rabbit content, could explain the much higher gross and calculated metabolizable energy (ME) value. However, it is important to point out that the energy value of the MTZ was actually determined at the lab by bomb calorimetry while the energy value for the other two diets was calculated as indicated in the table footnote. The factors used in this calculation appear to underestimate actual energy value because, when applied to the MTZ diet, it gave a low estimate of 5,517 kcal ME/kg DM, closer to the values calculated for the other two diets (Table 3).

Protein and Amino Acid Profile

The protein requirements of mature milk, expressed as a percentage of the diet dry matter, are estimated to be between 21.8% and 26.0% for maintenance, 38% for gestation, and 45.7% for lactation (NRC, 1982). The protein requirements for growth are listed at 38% from weaning to 13 weeks of age, and between 32.6 and 38% from 13 weeks to maturity. These values represent the minimum protein requirements of mink for the different phases of the life cycle and it is advisable to use higher protein levels to provide a margin of safety (NRC, 1982). The protein content of the BFF's diet presented in Table 3 is 44% higher than the protein requirement of mink during lactation, which is when the highest demand is present during the life of the mink. Consequently, these diets seem to have an adequate level of protein for the BFF. The MTZ diet has a higher protein content than either commercial pellet or natural ingredient diet for domestic ferrets (Table 4).

Dierenfeld & McGuire (1989) reported a marked seasonal variation in the protein concentration of prairie dogs. Combined average for adult male/female black-tailed prairie dog carcass concentration was highest in spring (57.7% of DM), decreasing through summer (43.6%) and fall (34.3%) to a low of 30.3% in winter. White-tailed prairie dogs follow a similar pattern with the highest protein concentration in spring (55.9%) and 35.5% in summer. No data was presented for fall and winter. The combined mean protein content of male/female prairie dogs considering both species and all seasons is approximately 42.5% of DM, slightly lower than the protein concentration in the MTZ diet for BFF .

TABLE 2. Fatty acid profiles of the fat content in the ingredients included in the diet formulated for Black-footed Ferrets

Analysis ¹ , %	Gro-Fur darks/Mink feed ²		Rabbit Carcass ³	Blood Meal ⁵	
	20% Fat	10% Fat			
C. Fat	23.96	11.23	15.78	0.06	
Fatty Acid					
Myristic	14:0	3.1	3.0	4.3	0
Myristoleic	14:1	0	0	1.2	0
Palmitic	16:0	38.4	30.6	57.1	29.9
Palmitoleic	16:1 (n-7)	5.7	5.4	3.5	0
Stearic	18:0	16.2	9.2	19.1	21.3
Oleic	18:1 (n-9)	4.4	3.4	2.1	31.7
Linoleic	18:2 (n-6)	26.8	37.5	11.3	17.1
Linolenic	18:3 (n-3)	1.0	1.0	0	0
Gadoleic	20:1 (n-11)	1.5	2.2	0	0
Arachidonic	24:4 (n-6)	0.9	1.4	1.4	0
Cetoleic	20:1 (n-11)	0	1.4	0	0
Lignoceric	24:0	1.9	4.8	0	0
Fraction⁴					
Saturated		59.6	47.6	80.5	51.2
Monounsaturated		11.6	12.4	6.8	31.7
Poliunsaturated		28.7	39.9	2.7	17.1

¹ Fatty acid content expressed as % of total methyl esters.

² National Complete Mink-Food Pellets, Manufactured by Milk Specialties Co., New Holstein, WI, U.S.A.

³ Ground whole dressed-carcass

⁴ Sat. = 14:0 + 16:0 + 18:0 + 24:0; Mono = 14:1 + 16:1 + 18:1 + 20:1 + 22:1; PUFA = 18:2 + 18:3 + 20:4

⁵ United Coop of Mississauga, Ontario, Canada

TABLE 3. Composition and chemical analyses of the regular 60/40 black-footed ferret diets used at the Metropolitan Toronto Zoo, Sybille Wildlife Research Center and Henry Doorly Zoo

Ingredients	Sybille Wildlife		
	Metro Toronto Zoo ¹	Research Center ²	Henry Doorly Zoo ³
Gro-Fur Darks 20%-fat mink Pellets ⁴	29.78	30.62	27.79
Ground rabbit carcass	19.85	22.05	20.10
Lard	--	1.35	--
Blood meal	0.60	--	0.54
Bio-liver	--	--	0.54
Vitamin E supplement powder	0.15 ⁵	--	0.09
Water	49.62	45.98	50.94
Chemical analysis on dry matter basis:			
Dry matter, %	32.82	35.70	31.30
Crude protein, %	44.40	47.34	44.09
Crude fat, %	21.67	24.09	21.41
Ash, %	10.73	15.97	11.02
Gross energy, kcal/kg	6,290	5,486 ⁶	5,491 ⁶
CME ⁷ , kcal/kg	4,843	4,224	4,228
Calcium, %	2.18	2.24	2.20
Phosphorus, %	0.92	1.40	1.60
Iron, mg/kg	793	--	649
Zinc, mg/kg	188	--	138
Vitamin A, IU/kg	5,834	--	37,270
Vitamin E, IU/kg	86	--	39

¹ Formulation #2, effective from 1993-09-30. Sample MTZ 495-93.

² Adapted from Sybille Wildlife Research Center SSP Institution Nutrition Report (1993).

³ Adapted from information provided by the Henry Doorly Zoo.

⁴ National Complete Mink-Food Pellets, Manufactured by Milk Specialties Co., New Holstein, WI, U.S.A.

⁵ Vitamin E concentration = 50 IU/g. Hoffmann-La Roche Ltd., 2455 Meadowpine Blvd., Mississauga, Ontario, Canada L5N 6L7.

⁶ Estimated calculated values as GE = grams of each nutrient per 100 g dry matter x kcal per gram (protein = 5.7 kcal/g; fat = 9.5 kcal/g; carbohydrate = 4 kcal/g). Carbohydrates calculated by difference: 100% minus (% protein + % fat + % ash) = % carbohydrate (NRC, 1982).

⁷ Estimated as follows: ME = GE x 0.77 (NRC, 1982).

The amino acid profile with the appropriate combination of essential amino acids must be evaluated because animals do not have a requirement for protein itself but actually require a series of individual amino acids in different proportions. The "ideal protein" would mimic or match the matrix of amino acids exactly as the animal needs them. Unfortunately, this profile has not been established for the BFF, therefore the essential amino acid profile required by carnivorous animals can be used for evaluation purposes.

The amino acid profile of the protein included in the MTZ diet for BFF is compared to commercial pellet and natural ingredient diet for domestic ferret in Table 5. The content of individual essential amino acids is comparable but the MTZ diet provides at least 8% units more of protein than the diets for the domestic ferret. The essential amino acid profile is also comparable to the one recommended for cats and dogs (NRC, 1985,1986). Hyperammonemia and Reye's syndrome have been described as an arginine deficiency in ferrets (Deshmukh & Thomas, 1985; Thomas & Deshmukh, 1986).

Energy

The energy requirements of mink are listed as 3,600 kcal ME/kg DM for adult maintenance (NRC, 1982), increasing by 9% for gestation to 3,930 kcal ME/kg DM and by 25% for lactation to 4,500 kcal ME/kg DM. The energy requirements for growth from weaning to maturity have been estimated at 4,080 kcal for male kits and 3,930 kcal ME/kg DM for females. This takes into account the differential growth rate between sexes (NRC, 1982).

The analysis of the MTZ BFF diet (Table 3) shows a gross energy concentration of 6,290 kcal/kg DM. Using the average figure of 77% for converting GE to ME values (NRC, 1982) this results in an estimated energy value of 4,843 kcal ME/kg DM. This value exceeds even the most demanding phase of the mink life cycle, lactation with 4,500 kcal. The addition of extra dietary fat (lard) as recommended by the SWRC and included in their diet (Table 3) can be considered unnecessary, and furthermore contraindicated considering the already high PUFA concentration in the diet base on the fatty acid composition of their mayor ingredients (Table 2).

From field data, Powell et al. (1985) estimated that the free-ranging black-footed ferret expended an average of 130 kcal ME/day during the winter (December through March). They also speculated that despite a lower cost of thermoregulation, the requirements during the summer are considerably higher due to increased activity and reproduction.

TABLE 4. Chemical composition of the MTZ black-footed ferret diet as compared to the concentration of nutrients in commercial feed formulations for the domestic ferret

Nutrients	MTZ Black-Footed Ferret Diet ¹	Domestic Ferret	
		Commercial Pellets ²	Natural-ingredient Diet ²
Dry matter, %	32.82		26.10
Crude protein, %	44.40	35.00	36.40
Crude fat, %	21.57	18.20	28.40
Ash, %	10.73	8.20	7.66
NDF, %	14.90		
ADF, %	4.65		
Crude fibre, %		2.90	2.30
G.E., Kcal	6,290	4,470	5,020
M.E. ³ , Kcal	4,843	3,442	3,865
Calcium, %	2.18	1.44	1.34
Phosphorus, %	0.92	1.11	1.11
Magnesium, %	0.16	0.28	2.11
Sodium, %	0.37	0.39	0.46
Potassium, %	0.52	0.83	0.57
Iron, mg	793	289	268
Zinc, mg	188	117	105
Copper, mg	25	18	12
Manganese, mg	110	45	54
Selenium, mg	0.67	0.2	
Vitamin A, IU	5,834	18,164	83,910
Vitamin E, IU	86	111	55

¹ Second formulation, effective from 1993-09-30. Sample MTZ 495-93.

² McLain et al. 1988. Nutrition. In: Biology and Diseases of the Ferret. Pp. 135-152. J.G. Fox (Ed) Lea & Febiger, Philadelphia.

³ Estimated values, calculated as follows: ME = GE x 0.77 (NRC, 1982).

TABLE 5. Amino acid profile of the MTZ Black-footed ferret diet as compared to the amino acid content of two formulations for the domestic ferret (values expressed as a percentage of dry matter)

	MTZ Back- Footed Ferret Diet ¹	Domestic Ferret	
		Commercial Pellet ²	Natural Ingredient Diet ²
Crude Protein	44.40	35.00	36.40
<u>Essential Amino Acids:</u>			
Arginine	2.36	2.23	2.30
Cystine		0.49	0.31
Histidine	1.11	0.63	0.84
Isoleucine	1.15	0.63	0.84
Leucine	2.70	2.34	2.75
Lysine	1.67	1.87	2.06
Methionine	0.44	0.47	0.54
Phenylalanine + Tyrosine	2.54	2.32	2.24
Threonine	2.74	1.30	1.11
Valine	1.72	1.22	1.73
<u>Non-essential Amino Acids:</u>			
Alanine	0.91	--	--
Aspartic acid	3.11	--	--
Glutamic acid	5.01	--	--
Glycine	2.32	3.21	3.24
Serine	1.68	1.67	1.61

¹ Second formulation, in use since 1993-09-30. Sample MTZ 495-93.

² McLain et al. 1988. Nutrition. In: Biology and Diseases of the Ferret. Pp. 135-152. J.G. Fox (Ed) Lea & Febiger, Philadelphia.

Energy

The maintenance energy requirements of captive animals are often below what might be expected under free-ranging conditions because of the lower activity and reduced thermoregulatory

demand (Stromberg et al., 1983). Siberian polecats (*Mustela eversmanni*) kept in cages and exposed to ambient temperature expended about 104 kcal ME/kg BWt/day (Powell et al., 1985).

The well established relationship between digestible energy and protein in the diet of growing domestic animals on feed consumption, weight gain, and nitrogen retention has also been documented for mink (Sinclair et al., 1962).

Since most domestic animals eat to satisfy their energy demands and will stop eating when this driving force is abolished, the concept of energy density of diets has a practical implication when considering that it is the main factor determining the quantity of feed intake. An animal will require and consume less of a high-energy density diet and more of a low-energy density one. Due to this direct relationship between energy density and feed intake, it is important to keep in mind that all dietary nutrients should be expressed on a basis that relates to the energy concentration or the daily requirements, regardless of the quantity of feed consumed daily. The well established relationship between digestible energy and digestible protein in the diet of growing domestic animals on feed consumption, weight gain and nitrogen retention has also been documented for mink (Sinclair et al., 1962).

Fat and Fatty Acid Profile

Fat, as a component of animal diets, could be mostly considered as an optional supplement since most ingredients mixed in the compounded diet contain enough essential fatty acid to provide the minimum requirement by animals. However, some evidence suggests that some additional fat, added as part of the ingredients, is desirable for species that are strictly carnivorous (NRC, 1982; NRC, 1986). Fat appears to exert a positive influence on the palatability of the diet and is an unavoidable component of the natural diet consumed by carnivorous in the wild.

Dietary fat levels reported that have been used satisfactory in mink diets range up to 40% (NRC, 1982), whereas dry, commercial mink and domestic ferret feeds usually contain anywhere from 16 to 28% fat (Table 4).

Dierenfeld & McGuire (1989) reported a significant seasonal variation in the fat content of the prairie dog carcasses. Carcass fat increased consistently from spring through summer and fall reaching a maximum at the beginning of winter. Their data has been adapted to provide a combined average for adult male/female black-tailed dog carcasses which gives a value of 29.45% fat on DM at the beginning of the spring, 52.15% beginning of summer, 45.75% beginning of fall and a high of 60.7% at the beginning of winter. Thus the energy expenditure during winter time reduced body fat from 60.7% to 29.45%. A similar pattern was found in juveniles, with average high fat levels of 49.2% at the beginning of winter dropping to 35.9 % at the end of spring. Analyses of carcasses from male and female white-tailed prairie dogs reported by the same researchers, showed a low of 34.15% fat at the beginning of the spring and a high of 60.15% in summer. In general, adult female black-tailed prairie dogs had higher fat content than adult males; the opposite was found in white-tailed prairie dogs. Powell et al., (1985) reported a fat content of 32.34% of the DM in black-tailed prairie dogs collected in the fall without reference to the sex or age class of the animals.

This information is not sufficient to recommend an optimal fat level in the diet of the BFF in captivity or to conclude whether or not there would be an advantage to seasonal changes in the fat level in the diet to simulate the natural variation in the fat content in the prey food. Since fat is the diet component that most significantly affects the energy density and the nutrient:energy density the main attention should be directed towards the level of feed consumption by the animals. High-energy density diets should also increase the concentration of other nutrients. Nutrient density should also be increased if, due to heat stress, animals reduce their feed consumption. Although the MTZ diet and the others shown in Table 3 contain lower levels of fat than those observed in the prairie dogs it should be considered adequate because the calculated ME value is within a high energy density range and above values for domestic ferret's diet (Table 4).

The fatty acid (FA) profile of the major ingredients (Table 2) included in the diets for BFF compared in Table 3 permit to estimate that their FA profile is mostly saturated (> 66%) and the rest is mainly polyunsaturated (PUFA > 24%). Thus the mono-unsaturated fraction is small (< 10%). Among the saturated FA, palmitic acid (16:0) is the major component; among the PUFA, linoleic acid (18:2 n-6) is the major fraction while linolenic (18:2 n-3) and arachidonic (20:4 n-6) made small contributions. Nonetheless these three PUFA defined as essential FA appear to be in sufficient amounts to satisfy this role in the BFF. Although arachidonic acid could be synthesized from linoleic acid, they belong to the same family (n-6), strict carnivorous, such as the cat, cannot convert linoleic acid into arachidonic or alpha-linolenic acid to eicosapentaenoic acid (20:5 n-3) because the σ -5-desaturase activity is very low or absent (NRC, 1986). Thus, the diets for cats must include both linoleic (0.5% DM) and arachidonic acid (0.2% DM) as minimum requirements (NRC, 1986; Robbins, 1993). No information is available in this respect for *mustelids* to indicate the essentiality for dietary arachidonic acid.

We can expect that BFF have a similar essential FA requirement as mink, a minimum of 0.5% for adult maintenance and 1.5% for young growing kits and pregnant/lactating females (NRC, 1982).

The essential FA levels calculated from analysis on individual ingredients (Table 2) indicated that on DM basis the MTZ diet contains about 5.57% linoleic acid, 0.19% linolenic acid and 0.21% arachidonic acid. With a total essential FA concentration of 5.97% of the DM, this diet exceed by 10-fold the essential FA requirements of cats and by almost 4-fold the estimated requirement of the mink. Among the BFF's diet ingredients, Gro-Fur Darks 20% fat mink food provides approximately 95% of the total essential FA and the rabbit carcass provides the small remaining portion.

The relatively high level of PUFA in the MTZ diet for BFF considered together with relatively low level of vitamin E (86 IU per kg DM) as shown in Table 3 should perhaps be regarded as a matter of concern. Nutritional steatitis or yellow fat disease has been described in both mink (Lalor et al., 1951) and domestic ferrets (Brooks et al., 1985) as a result of feeding high PUFA diets with inadequate vitamin E supplementation. This seems to indicate that Mustelidae are particularly sensitive to diets containing high levels of PUFA. Within a species, young growing animals are the most susceptible to the disease with mortality rates ranging from 50 to 75% in minks. The etiology of the disease in all the species susceptible has been reported to be associated to diets high in PUFA and/or deficient in vitamin E (McLain et al., 1988). Vitamin E is important since it functions as a protective antioxidant in the lipid fraction of cell membranes and helps to prevent this disease.

Brooks et al. (1985) reported an outbreak of nutritional steatitis in farmed ferrets (*Mustela putorius furo*) in New Zealand. The disease was caused by feeding a diet containing high PUFA (7.7% DM) and a relatively low vitamin E concentration (90 mg alpha-tocopherol = 134 IU vitamin E activity/kg DM). Young growing kits (13-15 week old) were the most affected with a total of 793 cases of which 183 animals died. That is a 23% mortality rate.

Experimental work has also shown that PUFA levels as low as 3% of DM induce nutritional steatitis in vitamin E deficient rats (Danse & Verschuren, 1978; Danse & Steenberg, 1978). This fact causes more concern about the MTZ diet for BFF since rats are known to be much less sensitive than mink to excess dietary PUFA. Therefore, as a precautionary measure it would be advisable to lower PUFA concentration in the MTZ diet and at the same time increase vitamin E to a minimum of 150 IU/kg DM. This is a matter that requires prompt attention to prevent any risk of a sudden outbreak of nutritional steatitis in the BFF in captivity at the MTZ. The same advice is valid for the BFF's diet at SWRC and OHDZ.

Minerals

In general, all minerals in the MTZ diet, as shown in Table 6, exceed the recommended levels for mink (NRC, 1982) and with the exception of phosphorus, are higher than the concentrations reported in prairie dogs, the BFF primary prey food source. In addition, some of the elements (Ca, Fe, Zn, Mn and Se) exceed the practical levels used in commercial and natural ingredient formulations for the domestic ferret (Table 6).

Despite the fact that all minerals in the MTZ diet for BFF are at concentrations well above the normal levels for mink, a few may still pose a risk. These will be discussed for each mineral.

Calcium and Phosphorus

The Ca (2.18-2.24%) and P (0.92-1.60%) concentrations on DM basis in the three diets for BFF (Table 3) are excessive when compared to the 1.0% maximum dietary recommendations for mink (NRC, 1982). These concentrations also exceed the reported level for the mineral in prairie dogs (1.54%). Calcium level in the MTZ diet was also found to be 51% and 61% higher than the Ca content in the commercial and natural ingredient diet for domestic ferret, respectively. Considering the Ca requirement for other well known carnivores, it is safe to state that this mineral should not exceed 1.2% on DM basis the BFF diet.

Due to digestive mechanisms of absorption and metabolic interrelationships of minerals, excessive dietary Ca affects the utilization of P, Zn, Cu primarily but indirectly of many other nutrients, including protein and fat which may have a detrimental effect on the animal's performance when imposed for extended period of time. It is well established that excess dietary Ca affects growth and feed efficiency in swine (NRC, 1988) and poultry (NRC, 1994).

TABLE 6. Mineral profiles of the MTZ black-footed ferret diet, black-tailed prairie dog carcass and domestic ferret diets, as compared to the mineral requirements of mink

Nutrient (D.M. basis)	MTZ Ferret Diet ¹	Requirements of mink ²	Black-tailed Prairie dog Carcass ³	Domestic Ferret ⁴	
				Commercial Feed	Natural Diet
Calcium, %	2.18	.3 - 1	1.54	1.44	1.34
Phosphorus, %	.92	.3 - .8	1.14	1.11	1.11
Magnesium, %	.16	.04	.07	.28	2.11
Sodium, %	.37	.2	.26	.39	.46
Potassium, %	.52	.3	.47	.83	.57
Iron, mg	793	20 - 30	215	289	268
Zinc, mg	188	59 - 66	75	117	105
Copper, mg	25	4.5 - 6	5	18	12
Manganese, mg	110	40 - 44	3.5	45.3	54
Selenium, mg	.67	.1		.2	

¹ Sample #495-93 Black-footed ferret formulation #2, in use since September 30, 1993.

² National Research Council (NRC). 1982. Nutrient requirements of mink and foxes. Washington, D.C. National Academy of Sciences.

³ Dierenfeld, E.S. and J.T. McGuire. 1989.

⁴ McLain et al., 1988. Nutrition. Pp. 135-152. In: Biology and Diseases of the Ferret. J.G. Fox (Ed.), Lea & Febiger, Philadelphia.

Furthermore, the MTZ diet not only contains excessive levels of Ca and P but their ratio (2.37: 1.00) is somewhat out of the most commonly recommended range for other species including mink (1.5/2.1: 1.0). It would be advisable to reduce the Ca and P content of the BFF's diet to a maximum of 1.2% for Ca and 0.8% total or 0.5% available P. The two main ingredients used in the current formulation of BFF diets are very high in Ca content, particularly the rabbit carcass, therefore both need to be reduced somewhat to provide space for other ingredients with low Ca content such as blood meal, horse meat, livers, or even natural ingredient diet for domestic ferret. This could be the beginning of a new formulation for BFF diets based on more basic ingredients. This would give more flexibility for the formulation and would allow more easy adjustments of the nutrient profile according to the expected nutritional needs through the life cycle of the BFF in captivity.

Zinc

In general, the estimated Zn requirements for wild and domestic animal species range from 10 to 70 ppm in the DM of the diet (Robbins, 1993). As shown in Table 6, the concentration of Zn in the MTZ diet was 188 ppm which exceeds the recommended levels for mink (59-66 ppm) by almost 3-fold. Although this is still below the toxic levels (1500 ppm) reported for the domestic ferrets (Straube et al., 1980; Straube & Walden, 1981).

The recommendation by the SWRC for adding 20 ppm of supplemental Zn in the form of Zn-gluconite tablets can be considered unnecessary as this mineral is already present in excess. Furthermore, even though Zn is relatively nontoxic in a wide range of intake, one should exercise caution as high dietary concentrations may interfere with Cu and Fe utilization by the animals.

The mink-food (Gro-Fur Darks, 20% fat) as ingredient of the BFF diets made the major contribution to the total Zn content (88.7%). The dietary level of Zn for BFF should not exceed 70 ppm. This figure is based on other comparable species with recommended levels better defined such as 59-66 ppm for mink (NRC, 1982), 50 ppm for domestic cat (NRC, 1986) and 36 ppm for domestic dog (NRC, 1985). The second assumption made in this suggested level of Zn is that at least 80% is available to the animal.

Iron

The level of Fe in the BFF diets (Table 3) seems rather extreme (649- 793) when compared to the dietary requirements of other carnivorous, 20-30 ppm for mink, 80 ppm for the domestic cat, and 32 ppm for the domestic dogs from data sources as above. The concentration of Fe is also well above the practical levels used in domestic ferret diets, 268-289 ppm (Table 4), and is almost 4-fold the level found in the prairie dog carcass at 215 ppm (Table 6). Although blood meal is a very rich source of Fe at the % level of its inclusion in the MTZ diet does not create the problem but rather the mink-food which contributes with more than 93% (658 ppm) of the total Fe found the MTZ BFF diet.

Under normal conditions, the level of Fe uptake by the body is regulated by absorption. That is, the homeostatic mechanisms for Fe depends primarily on absorption rate which is normally low (10-15 %) and adjusts in response to body needs. Body Fe remains in the body for very long time and the mechanisms for excretion are limited (Linder, 1985). Nonetheless, Fe toxicity exists, hemosiderosis and hemochromatosis, both are relatively frequent problems in many zoo animals (Robbins, 1993). Iron overloading is a dangerous debilitating condition that results in damage of the liver, heart, pancreas, and possible other organs (Linder, 1985). The causes of this Fe overload are as yet to be clarified although, excessive long term dietary Fe intake is a promoter. Since the maximum Fe tolerance level of the BFF is unknown, as a precautionary measure, it would be safe to reduce its level to the NRC recommendations for other carnivora, to a maximum of 80 ppm in the DM of the diet.

Fat Soluble Vitamins

No data are available concerning the vitamin requirements of the BFF or the domestic ferret. The specific vitamin requirements of mature mink for maintenance, gestation or lactation have not

been established, and the only information for this species is limited to the needs for the early growth period, from weaning to, 3 weeks of age, all of which adds to the difficulties for assessing the vitamin adequacy of the MTZ diet for the BFF. Nonetheless, it is possible to make some inferences about vitamins from other domestic species with well defined requirements.

Vitamin A

The vitamin A requirements for a couple of domestic species with feeding habit similarities indicate that 3,710 IU/kg DM in the diet for dogs (NRC, 1985) and 3,333 IU/kg DM for cats (NRC, 1986) provide for their nutritional needs. For the mink maximum vitamin A recommended is 5,930 (NRC, 1982) for early growth.

Dierenfeld & McGuire (1989) reported a marked seasonal variation in the vitamin A content of prairie dogs. Black-tailed prairie dog carcass during the spring had 3.5 times more vitamin A than during the summer, and levels continue to decline through the fall to a low in winter. Their original data was converted and adapted to result in the following averages in adult male/female dog carcass: 55,160 IU/kg DM in the spring; 17,660 IU/kg DM in summer, 8,830 IU in the fall and to a low 6,500 IU/kg in winter. A similar pattern was found in the white-tailed prairie dog where combined male/female carcasses were 2.5 times higher in spring with 23,300 IU/kg than in summer with 9,000 IU/kg of DM.

Assuming that the BFF in the wild in order to meet their daily energy requirements, approximately 200 kcal for a 700 g ferret, a free-ranging BFF must consume between 40-60 g of prairie dog meat (DM basis). Thus, this animal will have a daily vitamin intake of 2,758 IU during the spring and a low of 325 IU during winter. Since excess vitamin A intake can be stored in the liver then these BFF could be able to balance their needs during the entire year. For the same reason vitamin A has the potential to act as a cumulative toxicant in most species. In most species, more than 90% of the vitamin A in the body is stored in the liver. Chronic toxicity usually results from intakes 100 to 1000 times their nutritional requirements for a prolonged period. The estimated safe level for cats is 100,000 IU/kg diet (NRC, 1987). Mink tolerate 40 IU of vitamin A per gram of body weight without disturbance over periods of 3-4 months. Fully grown animals could tolerate 200-300 IU/g body weight for 6-8 weeks (NRC, 1982).

The vitamin A concentration of the MTZ diet for BFF as shown in Table 3 provides 5,834 IU/kg DM equivalent to 137 IU/100 kcal ME. Although exceeding the requirements of cats is slightly deficient and does not meet the minimum dietary needs of the growing mink. However, considering some additional information available at the MTZ concerning poor reproductive performance during 1993 and 1994 season may indicate the need for increasing the dietary vitamin A to 6,000 IU/kg. This would be undoubtedly a good nutritional decision without running into a risk of toxicity.

A laboratory report concerning the ferret diet used at the OHDZ indicates a vitamin A concentration of 3,500 µg/kg but it does not specify if this is expressed on a as fed or DM basis. Assuming that this value is on an as fed basis as retinol (1 µg retinol = 3,333 IU vitamin A activity) then the ferret diet at the OHDZ provides 37,270 IU of vitamin A activity per kg DM (Table 3) or 6.4-fold the level in the MTZ diet. Needless to say this is an exaggerated level of vitamin A under any circumstances. An educated guess could be that the inclusion of bio-liver in

the OHDZ diet could explain this elevated concentration of vitamin A in the diet. On the other hand, if this concentration of vitamin A was given on DM basis, the level is at best marginal but most likely deficient.

Inadequate vitamin A intake leads to a deficiency condition adversely affecting reproduction including low conception rates, stillbirths and abnormal semen (NRC, 1987). The low reproductive performance of BFF observed at the MTZ during 1993- 94 could be, to some degree related to the marginal vitamin A level in the BFF diet. Thus, this vitamin should be increased to at least 6,000 IU/kg DM diet to match the requirements specified for mink or perhaps to 6,900 to add a safety margin.

Vitamin E

The dietary requirements of vitamin E estimated for most animal species are in the range of 5 to 50 IU/kg of diet (or 2 to 4 IU/kg of body weight per day, NRC, 1987). The vitamin E requirements of mink during early growth are listed as 27 IU/kg DM, equivalent to 6.6 IU per 1,000 kcal ME. The requirements of mature mink for maintenance, gestation and lactation have not been determined (NRC, 1982) and no information is available for the BFF.

The vitamin E concentration in the BFF's natural diet as determined by Dierenfeld & McGuire (1989) on black-tailed prairie dogs, had a seasonal variation from a low of 63.5 IU/kg DM in the fall (most likely the beginning of the fall), to a high of 101.7 IU/kg DM in the winter (most likely the beginning of the winter). These changes are for the most part associated with changes in the fat content of the body. In contrast, white-tailed prairie dogs had the highest vitamin E content in the spring with 118 IU/kg DM when animals showed the lowest amount of carcass fat, and a low of 59 IU/kg DM in the summer. These values were adapted from the original data presented by Dierenfeld & McGuire (1989) to obtain combined average for male/female carcasses and converted to IU of vitamin E activity (1 mg alpha-tocopherol = 1.49 IU vitamin E).

The analyzed vitamin E level in the MTZ diet for BFF was 86.3 IU/kg DM as shown in Table 3. This value exceeds the requirements of growing mink (27 IU/kg DM) and is very close to the vitamin E concentration found in prairie dogs (84.5 IU/kg DM average for all seasons and both species). However, as previously discussed, the need for vitamin E in the diet is markedly influenced by the dietary composition, especially the level of fat and its degree of saturation, the level of feed antioxidants, and the content of selenium. The latter a close nutritional partner of vitamin E in keeping the cell's body protected from peroxidation reactions and its by-products (Stowe & Whitehair, 1963).

Various researchers have suggested values for the incremental effects of dietary PUFA level on the nutritional requirements for vitamin E ranging from 0.18 to 0.60 mg alpha-tocopherol per gram of PUFA (Combs, 1992). Although there seems to be no consensus among the experts in the field as to the precise quantification of this important dietary relationship, to be safe, it is advisable to accept the upper range recommended when dealing with zoo animals consuming high levels of fat with a significant fraction as PUFA.

Considering that the MTZ diet for BFF contains 59.7 g of PUFA/kg DM, it should have a total of 35.82 mg of alpha-tocopherol or 53.4 IU of vitamin E activity/kg DM. Based on this calculation

it may appear that the MTZ diet provide sufficient vitamin E to the BFF to prevent nutritional steatitis. However, evidence presented by other researchers (Brook et al.,1985) indicate that vitamin E levels of 90 mg of alpha- tocopherol/kg DM, equivalent to 134 IU/kg, were insufficient to protect from an elevated PUFA level (7.7% of the DM) causing an outbreak of nutritional steatitis in ferrets (*Mustela putorius furo*). Consequently, it would be advisable to increase the vitamin E concentration of the MTZ for BFF to a minimum of 150 IU/kg DM as a precautionary measure until more information is available.

CONCLUSION

Knowledge of the black-footed ferret nutrient requirements to date are somewhat limited, as are those of the domestic ferret. Therefore, the recommendations made in this nutritional evaluation of the MTZ diet for BFF are based on the limited data available. Thus, some degree of educated guessing is included which leaves some uncertainty. Long term effects of the current diets are not clear at this time, which means that there is no guarantee of its nutritional adequacy to maintain a good reproductive performance and healthy progeny for many generations to come. The need for future research in this area is evident, therefore, if there is a serious commitment for the preservation of the species, appropriate resources should be made available for research. This research should be conducted at the zoo where the animals are presently in captivity but partnership with universities could be highly beneficial. Universities can provide a valuable scientific and technical support in helping the guardians of these endangered species to develop nutritional recommendations and feeding programs that guarantee their reproduction and healthy life.

In general, the nutrient concentrations in the MTZ diet for BFF meet or greatly exceed the dietary recommendations established for mink (NRC, 1982) as well as nutrient levels reported in the BFF natural diet (Dierenfeld & McGuire, 1989) and commercial formulations for the domestic ferret.

Observations of animal body and hair coat condition, growth performance, as well as clinical records would indicate that the MTZ ferret diet had provided the minimum needs for maintenance and growth. Whether the suboptimal reproductive performance observed in recent years at all participant institutions could be nutritionally related deserves further investigation.

The apparent excessive dietary minerals and PUFA intake must be kept in mind and under close observation to validate the changes recommended. Even though the minerals under consideration, Ca, P, Fe and Zn are not high enough to cause toxicity, excessive intake for a prolonged period of time may cause subclinical problems which are very difficult to detect.

The commercial mink-food included as the main ingredient in the diet of BFF (Gro-Fur Darks 20% fat) seems to be the source of the excessive PUFA and minerals in the final compounded diet. As there is an urgent need to reduce some of these levels, it is imperative to initiate discussions with the manufacturer of this product (Milk Specialties Co.) to make a modified customer product that is more in line with the needs of the BFF. Formulations from basic ingredients and by-products could be an alternative approach, which would provide a greater flexibility for the diet and most likely would also result in economic advantages.

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