FROM REARING TO FEEDING: A NUTRIONAL ASSESSMENT OF IN-HOUSE FEEDER INSECTS AT THE CINCINNATI ZOO & BOTANICAL GARDEN

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Abstract

Along with weekly purchasing of feeder insects the Cincinnati Zoo and Botanical Gardens (CZBG) raises a variety of insects to be used in the diets of zoo animals. The use of feeder insects allows for a natural and sustainable way of providing essential nutrients animals need to maintain proper health and wellness. Feeder insects can offer several nutritional benefits including high quality protein, essential amino acids, fats, calcium, iron, and phosphorus. The nutritional value of insects can vary based on species, life stage, and the environment they are raised in. This is why it is essential to analyze their composition before including them in diets. The goal of this study was to analyze the nutritional content and overall quality of the insects being raised and used as feeder insects at the CZBG. The species that will be examined in this paper include dubia roaches, Madagascar hissing cockroaches, flesh flies, flesh fly larvae, mealworm larvae, and wax moths. The analysis aims to provide data that can guide dietary planning for insectivorous and omnivorous animals that are housed in zoos. To evaluate the nutritional content of these feeder insects, the zoo collaborated with Midwest Laboratories (MWL).

Introduction

The CZBG is currently involved in the conservation of the American Burying beetle (ABB), a species that is at risk for extinction. The zoo takes part in the breeding and reintroduction program to help restore the beetle's population in Ohio through a collaboration between CZBG, The Wilds, Nebraska Game and Parks, Ohio Division of Wildlife, and The Fish and Wildlife Service. ABB production decreased from year to year while the beetles were in human care. This difference from wild populations led to comparisons between the diets of captive and wild beetles. Beetles in human care have traditionally been fed mealworms, but natural diets consist primarily of maggots. While there is published information on mealworms, flesh fly larvae nutrient analysis has not been published. We also wanted to analyze the mealworms we were feeding the ABBs as this may differ from previously published information. Four other insects used to feed insectivorous and omnivorous animals at the zoo were also analyzed.

Materials & Methods

Insect Rearing and Feeding Protocols

This study focuses on six key feeder species at the CZBG for which nutritional analysis has not been performed. The feeding and husbandry practices for each species were documented and standardized as much as possible to ensure consistency of nutrient values. Each species is listed below with its specific rearing and feeding protocol.

Dubia Roaches

Dubia roaches were maintained in colony groups and fed Mazuri® Cockroach diet, which is a commercially available, nutritionally balanced, powdered diet. To supplement their diet and provide additional micronutrients, fresh produce was offered three times per week (Sunday, Tuesday, and Thursday). While the total quantity of produce varied depending on colony size and the number of roaches being used in diets, the types of produce offered remained consistent and included kale, romaine lettuce, endive, escarole, blueberries, strawberries, carrots, etc. Due to fluctuating colony sizes, precise daily feeding weights were not recorded but are estimated to be about 60 g of fruit, and 100 g of greens.

Madagascar Hissing Cockroaches

Madagascar hissing cockroaches were housed in colony enclosures and fed a standardized amount of fresh produce three times per week (Monday, Wednesday, and Friday). Each feeding consisted of approximately 30 g of sweet potato and 25 g of kale per colony. In addition to the fresh produce, the cockroaches were provided with continuous access to the Mazuri® Cockroach diet.

Flesh Fly Adult

Adult flesh flies were housed in a large Black Reptibreeze® enclosure that was modified to prevent escape and facilitate access. Each of the habitats is fitted with an 8-inch hole cut into the main door, to which an 18-inch insect screen sleeve is secured. This sleeve allowed for the addition and removal of materials while preventing escapes. The base of each enclosure contains a tray filled with kitty litter to capture waste, and shallow black containers filled with pine shaving and water are placed inside to provide safe drinking water. Feeding stations included small dishes of dry sugar and, during oviposition or larval collection periods, 4-ounce deli cups half filled with wet cat ZuPreem® Feline cat food were placed in the enclosure on Saturday and Wednesday. These dishes were placed on top of the pine shaving water containers to prevent ghost ant infestation. Sixteen-ounce deli cups were added as pupation chambers when exclusion was anticipated. A halogen light above the enclosure provided lighting and ambient conditions were maintained at 23°C with 50% relative humidity.

The daily care involved topping off water and sugar dishes, transferring pupae into the adult enclosure, and adding or removing wet cat food based on a regular feeding and collection schedule. Weekly maintenance included replacing pine shavings, water dishes, cleaning sugar dishes, and removing dead flies from the kitty litter. Long term care includes replacing the kitty litter approximately every six months and deep cleaning the enclosure as needed. The cat food schedule was structured to support both larva and adult production. With the larvae from the groups reared to adulthood to maintain colony sustainability. This routine ensures a continuous overlap of generations and reliable larval availability for feeding and analysis.

Flesh Fly Larvae

Flesh fly larvae were reared in controlled conditions designed to support optimal growth and development prior to feeding or nutrient analysis. Larvae were collected twice weekly from adult rearing habitats after a 48-hour oviposition period on wet cat food. For each rearing setup, one to two frozen meatballs of approximately 1.5 inches in diameter were made from ground beef or meat “sawdust” collected from the zoo commissary band saw residue. These were thawed and placed

into a 1-gallon glass jar. Each jar was pre-filled halfway with sifted coconut fiber substate, which facilitates sanitation and allows for easier separation of the larvae or pupae later in development. A 4-ounce deli cup containing larvae and remaining cat food was embedded into the substrate with the lip flush to the surface. The meatballs are placed directly on top of the larvae, creating a warm, clustered feeding zone that allows for rapid development.

The jars were sealed with tightly fitting lids equipped with fine mesh screens to prevent contamination by phorid flies. Maggots were fed additional raw meatballs daily, typically after thawing, though frozen meat was used successfully in high-demand situations due to larvae's body heat rapidly thawing the food. Larvae were monitored daily for feeding activity and signs of pupation. Once the larvae began to migrate into the coconut fiber, feeding ceased, and meat remnants were removed to prevent spoilage. Pupating jars were set aside and labeled to avoid accidental feeding. When the larvae are intended for feeding, they are shifted just before wandering to pupate, using 0.25-inch mesh screen over a bucket or waste bin. This allowed for separation of larvae from the substrate, although manual removal was often required to prevent clogging. In some cases, maggots were separated by exploiting their wandering behavior through a container with an exit hole, this allowed for them to drop into the collection vessel. All maggots were fed out within several hours of collection to ensure gut content was full and fresh, thereby enhancing nutritional value to the receiving animals.

Mealworms

Mealworms were placed in a 10-gallon tank with about 4 cups of cornmeal, one apple slice, three slices of sweet potato, and three pieces of kale. The stocking density is limited to 10,000 mealworms per tank, so multiple tanks may need to be set up depending on the demand for mealworms to be used in different diets at the Insectarium. The veggies were refreshed three days a week.

Waxworms

The waxworms were set up in a critter keeper with approximately 1-inch of wheat bran on the bottom. One side has quarter pieces of egg carton stacked. Placed on the other side is a ball made from ¼ cup of wheat bran, and 2 tablespoons of honey placed on top of the wheat bran. The larvae were received from a commercial vendor, so their exact age was unknown. At a temperature of 26.67° C wax moths appear approximately two weeks later.

Midwest Laboratories Test

A composite sample of each species was then collected and sent to MWL. At MWL, each insect sample was analyzed using multiple chemical analysis techniques. All procedures followed established analytical standards based on Association of Official Analytical Chemists (AOAC) or American Oil Chemists Society (AOCS) methods, as outlined below.

Inductively Coupled Plasma (ICP) analysis. The sample preparation followed the MWL ME 069 wet ash procedure, which involves breaking down the sample using a combination of acids, minerals, and heat. Once the sample is prepared the sample extract is placed into the ICP machine to be nebulized and exposed to high temperature plasma. This process energizes the emissions of the dissolved minerals and metals, which then causes a release of energy as light when it's returning

back to its natural state. The emitted light's wavelength and intensity are used to identify and quantify specific minerals and metals within each sample.

Crude Fat analysis was measured by using acid hydrolysis following the MWL method FD 027. The samples were treated with ethanol and hydrochloric acid to hydrolyze and release fat-bound material. Fat extraction was performed using ethyl ether followed by petroleum ether. The extracted solvents were collected in pre-weighed beakers, evaporated, and dried to remove residual moisture and ether; the remaining residue was recorded as total crude fat.

Crude Fiber analysis was determined using MWL method FD 039. A sample portion was weighed and sealed in a membrane- based bag. The sealed bag was treated sequentially with acid and alkali solutions, dried, and reweighed. The sample and bag were then combusted to determine ash content. Crude fiber was calculated by subtracting the ash weight from the post-digestion residue weight.

Ash content analysis following MWL method FD 019. The samples are weighed and placed in a muffle furnace at 600°C. After combustion, the remaining inorganic residue was weighed. The result represented total ash, with moisture and organic material driven off during heating.

Crude Protein analysis measured using MWL method FD 070. Samples were combusted to determine total nitrogen content. The nitrogen value was multiplied by a factor of 6.25 to estimate crude protein concentration.

Mineral analysis (ICP-MS) was conducted using inductively coupled plasma mass spectrometry to find trace elements and minerals. Samples underwent acid digestion to dissolve solids and convert them into a measurable solution. The digested samples were introduced into a plasma source to generate elemental ions. The mass spectrometers separated the ions by mass-to-charge ratio, allowing for identification and quantification of individual elements. Values were then compared to standard reference materials and adjusted to show the original samples concentration.

Acid Detergent Fiber was analyzed using MWL method FD 021, based on Ankom Technology protocols. The sample was sealed in specialized filter bags and submerged in an acid detergent solution. The bags were then washed, dried, and weighed. The remaining material was recorded as acid detergent fiber.

Neutral Detergent Fiber was determined using MWL method FD 022, based on Ankom Technology methods. Samples were sealed in filter bags and treated with a neutral detergent solution. After washing and drying, the bags were reweighed. The residue remaining in the container represented neutral detergent fiber.

Gross energy was determined using the MWL FD 072 analysis guide. The samples are weighed and put in a pressurized oxygen bomb and ignition via electrical current. The temperature change in surrounding water can be calculated into BTU and Calories.

Results

The results of the analysis of the feeder insects collected are presented in Tables 1 and 2 on a dry matter basis (DMB). Scientific names of those samples are listed in Table 3. In Table 1, the protein

content ranges from 46.10-68.40 % (DMB). The analysis for protein was performed by combusting the sample to determine total nitrogen content. The nitrogen value was multiplied by a factor of 6.25 to estimate crude protein concentration. Many invertebrates contain non-protein nitrogen from sources such as chitin which may artificially elevate the available protein (Bernard et al., 1997). The ability to digest chitin is sometimes unknown and may vary among species. Data regarding protein and chitin use is not published for four of the species analyzed. The fat analysis ranged from 17.10-38.60% with the some of the higher analyses being larval samples. It is typical for the larval stage of an insect to contain a high percentage of fat. Calcium has been controversial in insects as a food item as many insects have an inverse calcium to phosphorus ratio (Bernard et al., 1997; Finke 2015). Feeder insects can be gut loaded with diets high in calcium before then feeding in the diet to provide more calcium to that animal. Calcium is important for bone development and maintenance; however, for invertebrates like ABB's that rely on a chitin exoskeleton for structural support, calcium needs may be variable. Therefore, it is important to obtain the nutritional content of all food items in animals' diets, especially if the diet is comprised primarily of insects. Using the nutritional content of the feeder insects helps to present a clear picture of the total nutritional value for each diet.

Conclusion

This study highlights the nutritional composition of various feeder insect species received and raised at CZBG and their potential role in supporting the dietary needs of insectivorous and omnivorous zoo animals. By analyzing key nutrients including protein, fat, fiber, minerals, energy, and moisture content, this research provided valuable data to inform evidence based dietary planning. The findings emphasize the importance of controlled rearing practices and consistent insect diets to ensure reliable nutrient profiles. Incorporating these feeder insects into zoo nutrition programs not only supports animal health and naturalistic feeding behaviors but also promotes sustainable, in-house food production systems. Continued monitoring and analysis will further enhance the effectiveness and adaptability of insect-based feeding strategies in zoological settings.

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Table 1. Proximate analysis of dry matter, crude protein, acid hydrolyzed fat, ash, acid detergent fiber, neutral deter fiber, and gross energy content of the feeder insects (DMB).[a]

Item	**DM**	**CP**	**FAT**	**ASH**	**ADF**	**NDF**	**GE**
	----------%----------						kcal/g
Flesh Fly	26.60	68.80	17.10	4.90	46.40	61.60	6.00
Flesh Fly Larvae	23.77	56.50	30.70	6.93	11.40	13.60	6.80
Dubia Cockroach	34.80	53.70	31.60	3.20	25.20	43.50	6.30
Madagascar Hissing Cockroach	35.30	58.60	26.60	2.80	60.30	128.00	6.80
Mealworm Larvae	34.18	46.10	38.60	3.09	5.80	3.09	7.20
Wax Moths	35.50	58.60	30.10	3.10	32.70	89.60	n.d.

[a]Analysis by Midwest Laboratories Inc. 13611 B Street, Omaha, NE 68144.
n.d. = value not determined.

Table 2. Major and trace mineral content of invertebrates (DMB).[a]

Item	Ca	P	Mg	Na	K	Cu	Fe	Zn	Mn	Se
	----------%----------					----------ppm----------				
Flesh Fly	0.11	0.90	0.11	0.41	1.02	13.20	129.00	155.00	11.30	0.45
Flesh Fly Larvae	1.36	1.04	0.10	0.51	1.13	10.30	160.00	104.00	8.10	0.52
Dubia Cockroach	0.14	0.57	0.12	0.34	0.83	20.40	51.40	172.00	10.90	0.49
Madagascar Hissing Cockroach	0.17	0.60	0.14	0.17	0.68	19.00	40.20	149.00	6.20	n.d.
Mealworm Larvae	0.06	0.78	0.19	0.14	0.92	16.60	43.90	106.00	6.40	0.29
Wax Moths	0.08	0.82	0.08	0.06	0.82	20.00	157.00	165.00	7.60	1.35

[a]Analysis by Midwest Laboratories Inc. 13611 B Street, Omaha, NE 68144.
n.d. = value not determined.

Table 3. Scientific names of invertebrate species in Tables 1 and 2.

Common Name	Scientific name
Flesh Fly	*Sacrophaga bullata*
Flesh Fly Larvae	*Sarcophaga bullata*
Dubia Cockroach	*Blaptica Dubia*
Madagascar Hissing Cockroach	*Gromphadorhina portentosa*
Mealworm Larvae	*Tenebrio molitor*
Wax Moths	*Galleria mellonella*