# Marmoset Husbandry and Nutrition

NEAL K. CLAPP, DVM, PhD, and SUZETTE D. TARDIF, PhD

The experimental use of small New World primates such as the tamarin and marmoset has provided new challenges in nutrition and husbandry for primate colony managers. Because Saguinus oedipus oedipus is an endangered species, successful captive breeding will be vital to continued experimental use of these animals. Unlike rodent populations which can be maintained satisfactorily in more or less routine and easily prepared and maintained animal facilities, the marmoset presents a variety of problems that must be understood and which, in most instances, have not been addressed. Issues pertinent to successful maintenance and production of these species include type of facilities for housing and breeding, nutritional requirements, behavioral patterns, breeder selection and climatic sensitivity and controls. In addition, costs have escalated both in animal purchase price and in maintaining a suitable colony facility. Whereas Old World monkeys have been maintained and bred in captivity for some time, only a few colonies of Callitrichidae exist. Colony personnel have focused on research investigations with little emphasis on husbandry and nutritional needs; these have been often addressed by word of mouth between colony managers and the trial-and-error approach.

Consequently, many different approaches in colony management are used with varying degrees of success. The annual informal meeting of marmoset colony managers at the American Association for Laboratory Animal Science meeting has provided a sounding board for problems and concerns; the compilation of information regarding the various colonies and their practices by Morin (personal communication) has served as a clearinghouse of information regarding the operation of marmoset colonies around the world. This paper describes the operation of the Oak Ridge Associated Universities (ORAU) Marmoset Research Center which was begun and operated for more than 20 years by Dr. Nazareth Gengozian (current address: Oklahoma Medical Research Foundation, Oklahoma City, Oklahoma); the new marmoset breeding facility was designed by Dr. Conrad Richter (current address: NIEHS, Research Triangle Park, North Carolina), and a number of the husbandry and nutritional practices were initiated by him during his recent tenure at ORAU.

#### **ORAU MARMOSET RESEARCH CENTER**

Three species of Callitrichidae, Saguinus oedipus oedipus, Saguinus fuscicollis illigeri, and Callithrix jacchus, are housed in the ORAU Marmoset Research Center. Approximately 415 tamarins and marmosets comprise the colony; 267 of these are S. o. oedipus. The saddle-back tamarin, S. f. illigeri, was the principal species used by Gengozian for immunological studies, but recognition of high incidences of spontaneous colonic cancer in the cottontop tamarin, S. o. oedipus, has prompted an increase in their numbers and use. C. jacchus, the common marmoset, has been a relatively recent addition to the colony. The two species that are not susceptible to colon cancer are being used as controls in colon cancer studies; C. jacchus, probably the easiest to raise and propagate, and S. f. illigeri, the species with which we have had the most experience.

The Marmoset Research Building (Figure 1), completed in 1969, has 5900 square feet of space; rooms include a medical room, a washing and service area, a food preparation room, a storage room, two large laboratories, a glassware-washing and -sterilizing room, and supporting areas including a shower and clothes-change facility, mechanical equipment room, an office, and a conference room. Each of the 12 animal rooms are  $16 \times 10$  ft

Address for correspondence and reprint requests: Dr. Neal K. Clapp, Marmoset Research Center, Oak Ridge Associated Universities, P.O. Box 117, Oak Ridge, Tennessee 37831-0117.

This report is based on work currently supported by Oak Ridge Associated Universities Corporation and National Cancer Institute, DHHS, contract N01 CP 21004.

# CLAPP AND TARDIF



Fig 1. Oak Ridge Associated Universities Marmoset Research Building.

and house 24 individually caged animals. Temperature is maintained at  $80 \pm 4^{\circ}$  F and humidity is controlled at 55  $\pm$  5% relative humidity. Rooms are supplied with 100% fresh air, which is prefiltered and then refiltered through 90% efficient (DOP) HEPA filters. Animal room light cycles are timercontrolled at 12 hr on and 12 hr off, and rooms are equipped with an automatic watering system. The cage washing area is designed for unidirectional flow of material. Cages, cage pans, and food pans are emptied, scraped, and prewashed on the "dirty" side of the area, washed in an industrial pass-through rack and cage washer, and unloaded on the "clean" side. The food preparation room is equipped with a large food mixer, food grinder, balances, sink, refrigerator, and other necessary equipment. A separate room is set aside for a medical unit, and the necessary examination tables, medical supplies, and equipment are maintained there. This room is kept scrupulously clean because of the nature of the operations conducted there.

## **OPERATION AND SANITATION**

Animals are checked twice daily for general health. Cages are constructed of zinc-coated expanded metal ( $18 \times 20 \times 48$  in.) and have floor screens and drop pans that are filled with Sani-chips (P.J. Murphy Corp., Rochelle Park, New Jersey). The cages can be divided into two units, each unit housing one animal. Screens and drop pans are washed twice weekly and cages changed and washed weekly. Feed cups are changed and washed daily. Animal room floors are cleaned daily with detergent, and rooms are cleaned monthly with quaternary ammonium disinfectant. Access to the facility is restricted. Clothing and safety shoes are issued to animal technical staff, and clothing is changed daily.

The control of roaches, a problem in most animal facilities, is an integral part of the sanitation and husbandry procedures and consists of removal of hiding and breeding places, boric acid treatment on

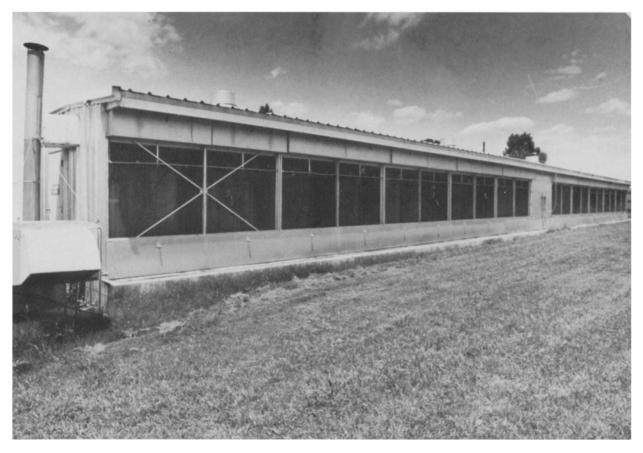


Fig 2. ORAU's breeding facility.

the baseboards of the hallways, roach traps placed in the animal rooms, and routine Diazinon (Pioneer Pest Control, Oak Ridge, Tennessee) spraying in areas where animals are not housed (laboratories, restrooms, offices, etc). Cleaning of floors in storerooms and maintaining refrigerators, etc, on rollers so they can be moved for frequent cleaning are also very important. As an example, a simple change in type of cage identification from a paper card and holder to a solid steel plate with gum label removed one common hiding and breeding place for roaches.

The ORAU Marmoset Research Center is operated as a closed animal colony. The animal-to-man risk from infectious diseases must always be considered, especially when the experimental animal is a primate. Conversely, of concern regarding the health of the marmosets is the possibility that personnel might infect the colony with some human or animal disease. Staff personnel with upper respiratory infections or "cold sores" are not allowed in the facilities.

# MARMOSET BREEDING FACILITY

Facility Description. This area is physically separated from the main marmoset research building and consists of an all-weather insulated aluminum cage enclosure on a concrete pad (Figure 2). The outer walls of the enclosures are made of vertically folding doors so the facility can be opened during warm months. The entire area is screened and protected with a security fence. The 1950-squarefoot area contains 40 family-unit apartments composed of two interconnected  $3 \times 3 \times 5$ -ft wire mesh cage sections suspended two feet from the floor on the framework of the cage enclosure. Each section can be detached and removed separately for cleaning without removing the family from the other section. Each subdivision is visually screened from other subdivisions by means of solid partitions or opaque screens (Figure 3). Heating is provided in the cooler months (folding doors closed) by two gas heaters, one providing backup in the event of the



Fig 3. Oak Ridge Associated Universities Marmoset Breeding Facility. Solid partitions and opaque screens separate animals from visual contact.

other's failure. Because of the semioutdoor nature of the facility, no air conditioning is required, although exhaust fans in the roof provide air movement in very hot, humid weather. Humidity is maintained in dry weather by a sprinkler system which is mounted above the family unit apartments and which serves as a manually operated "rainmaker." Incandescent light fixtures are located throughout the cage enclosure, and a skylight is located in the roof crown providing a natural light cycle. All family unit apartments are provided with automatic watering using city-provided chlorinated drinking water. A  $7 \times 8 \times 16$ -in. removable nest box with sliding door is also used to remove animals from the apartment without the need to use a net for catching them. The breeding facility is operated with even a more stringent policy of restricted access for all personnel because pregnant females and newborns must be protected from stress.

Breeding Program. Animals are selected for breeding based on genealogical relationships and

breeding performance of parents. Every attempt is made to enhance outbreeding, and occasionally new genetic stock is introduced into the colony, preferably with laboratory-bred animals from other closed pools. Breeding pairs are established, examined closely for compatibility, and incompatible pairs are remated. Once daily the colony nurse makes a visual examination for health, pregnancy, behavioral signs of stress or aggression, and other specified observations or examinations. Family units are left intact through the second, and sometimes third, consecutive litter. In the case of Saguinus species, third-litter birth occurs at approximately two years. Because this time roughly reflects the age of sexual maturity, animals removed from the family unit then are suited for mating as well as for experimental use. Where younger animals are required for experimental purposes, they can be removed earlier. This procedure not only increases the survival of live births by providing new mothers who have had experience in assisting

Year	Surviving/total born	Percent surviving* 23.4	
1976	11/47		
1977	23/95	24.2	
1978	16/44	36.4	
1979	31/70	44.3	
1980	33/63	52.4	
1981	30/59	50.8	
1982	15/28	53.6	
1983	36/58	62.1	
1984 (1-6)	16/23	69.6	

 TABLE 1. REPRODUCTIVE PERFORMANCE OF S. o. oedipus,

 OAK RIDGE ASSOCIATED UNIVERSITIES

\*Number live young surviving to one week of age.

parents in care of younger siblings but also obviates space having to be allocated to juvenile stock animals. Table 1 illustrates the continued improvement in reproductive performance of S. o. oedipus in the ORAU colony. Survival figures represent survival at one week of age. At ORAU, 85% of deaths before one year of age occur in the first week postpartum.

Sanitation. Due to breeder cage construction, feces and urine fall directly through the cage section bottom to the concrete pad which is hosed down daily. Cages are hosed *in situ* daily and removed periodically and washed in a rack washer in the Marmoset Research Building. This caging design results in a much-improved breeding performance and much-reduced labor cost.

### MARMOSET NUTRITION

Despite the time of several years during which marmosets have been used as experimental animals, nutritive and dietary requirements of the marmoset are not well understood. The compilation of information on marmoset colony management by Morin (personal communication) indicates that a wide variety of feedstuffs are fed at the different colonies throughout the world. Most colonies feed some kind of commercially available formulated diet (pelleted, dry, or canned) with additives which include vitamin and mineral supplements, fruit portions, insects, meal worms, baby mice, marshmallows, vegetables, etc. The diets have presumably been formulated to simulate those dietary entities which are naturally consumed in the wild. With a dearth of scientific information to provide insight into the "proper" diets, trial and error has been the rule, and a wide variety of so-called delicacies and supplements have been given; the presumed intent was to assure that the animals eat an adequate

caloric level for survival. Tardif and Richter showed that behavioral characteristics affect competition for desired food within family groups (1), and, with the need to raise and breed callitrichids for long-term studies, further steps are being undertaken to better understand the nutritive requirements for marmosets.

At least three research efforts have been reported with somewhat different interests and endpoints but also with some interesting similarities. Flurer and coworkers have focused their efforts on energy metabolism and natural ingredient diets (2). Knapka (personal communication) has likewise utilized natural ingredients to formulate a pelleted diet which has been successfully used in a S. mystax colony; this diet is intended to meet the total nutrient requirements of the animal without the additives (or supplements) which are difficult to evaluate and control or to manipulate. Further, he has developed a gluten-free diet because of the possible adverse effects of gluten etiologically in some inflammatory bowel disease in humans. At the New England Regional Primate Research Center, Escajadillo and coworkers (3) have formulated semipurified diets for both adults and infants.

The so-called BAM diet, developed by Richter in the late 1970s for the ORAU colony, is shown in Figure 4. The basic dietary ingredient is High Protein Monkey Chow 5045 (Ralston Purina Co., St. Louis, Missouri) with the addition of bananas and applesauce plus vitamins D<sub>3</sub>, C, multiple B, folic acid, and copper. This diet is mixed with Karo water in a large food mixer to a slurry consistency and is fed in the morning. The afternoon feeding consists of approx 1/2 ounce of canned marmoset diet (Zupreen, Marmoset Diet, Hill's Pet Products, Topeka, Kansas) plus a variety of supplements [boiled eggs, small marshmallows, peanuts, mealworms, raisins, coconut, lettuce, and sweet potatoes (usually one given per day)]. The major nutrient analysis of BAM diet including the supplements is shown in Table 2. A significant difference between BAM and an ordinary pelleted diet is that BAM contains 55% water as it is fed to the animals. This feed is available to the marmosets for 3-4 hr but is removed before it becomes a media for bacterial growth.

The major nutrient analysis of four experimental diets is listed by dry weight in Table 3. Whereas some have questioned the marmosets' need for high protein levels, the protein level in the diets is nearly identical, approx 20%. The percent of fat and crude

# MORNING BAM Applesauce (Basic Diet) Monkey Chow + Vitamins D<sub>3</sub>, C, Multiple B, Folic Acid and Copper EVENING Small Piece (~ ½ Ounce) Canned Marmoset Diet + Variety of Dietary Supplements (Boiled Eggs, Small Marshmallows, Peanuts, Mealworms, Raisins, Coconut, Lettuce, Sweet Potatoes [one item per day])

ORAU MARMOSET DIET

Fig 4. ORAU marmoset BAM diet.

 TABLE 2. MAJOR NUTRIENT ANALYSIS\* OF ORAU MARMOSET

 "BAM" Diet with Additives

Nutrient	Percent by weight	Percent of dry weight
Crude protein	9.0	19.8
Crude fat	3.2	7.0
Crude fiber	2.9	6.4
Ash	2.39	5.3
Carbohydrate	27.9	61.4
Moisture	54.6	_

\*Performed by Lancaster Laboratories, Lancaster, Pennsylvania.

fiber in the Hayes diet (Knapka, personal communication) is nearly twice that of the others, whereas the ORAU diet is about 10% higher in carbohydrate than the other three diets. In comparison, the variety of diets fed to marmosets indicates that several different formulations are reasonably successful in the various colonies. Little is known about minimum requirements of most nutrients, although some progress has been made in reducing obvious nutritional deficiency problems and in reducing the impact of the wasting marmoset syndrome (WMS) which was a major concern to marmoset colony survival only a few years ago (4, 5). Whether this is a nutritional disease is still not unequivocally determined, but the changes made in nutrition and husbandry practices during the late 1970s and early 1980s by Richter at ORAU have controlled, if not completely removed, WMS as a major cause of death in the ORAU colony.

More efforts are certainly needed to determine the minimum nutritional requirements of callitrichids (both adults and young alike) and the effects of certain physiological demands such as pregnancy, lactation, and aging. Species variations probably exist in the area of nutrition just as they occur in behavioral characteristics, breeding, space limitation, etc. The "old wives tales" must be identified and replaced with scientifically deter-

TABLE 3. MAJOR NUTRIENT ANALYSIS OF EXPERIMENTAL DIETS FOR MARMOSETS

Nutrient	Diet source (percent of dry weight)				
	ORAU	Zucker <sup>2</sup>	Knapka*	Hayes <sup>3</sup>	
Crude protein	19.8	23.4	22.2	20.0	
Crude fat	7.0	6.6	8.1	15.0	
Crude fiber	6.4	3.6	4.2	10.0	
Ash	5.3	3.8	7.8	5.0	
Carbohydrate	61.4	53.2	47.2	49.5	

\*Personal communication.

mined information that will make research efforts involving the gastrointestinal tract more meaningful and productive.

#### ACKNOWLEDGMENTS

The ORAU marmoset colony and facility was developed by Dr. Nazareth Gengozian, former director of the Marmoset Research Center and principal investigator on NIH grants ROI AMO 9289 AIB, ROI AI 12007 AI, and ROI HL 16757, concerning the immunology of natural chimerism and thrombocytopenic purpura, on ORAU's prime contract with DOE and its predecessor agencies AEC and ERDA concerning bone-marrow transplantation and EPA.

Basic long-term support for the facility and colony has been provided by the Corporation of Oak Ridge Associated Universities (ORAU); contract DE-AC05-760R00033. From 1975 to 1981, it was under the veterinary medical and pathology supervision of Dr. Conrad Richter; 1981–1982, Dr. Harry Walburg; and from 1982 to present, Dr. Neal Clapp. Since 1982, support has been jointly by ORAU and NCI contract NO1 CP 21004.

Parapathologic support has been under the supervision of Mrs. Gretchen Humason, 1966 to present, along with histologic and necropsy services which have been performed in the past by Gretchen Humason and Eleanor Selle. Presently, support and assistance have been supplied by the core staff of the Marmoset Research facility under Dr. Neal Clapp; Dr. Suzette Tardif (breeding program), Marsha Henke (diagnostic enterology), Barbara Gangaware (histopathology), Jerry Batson (cytogeneticist), Robert Carson (paramedical nurse), Isaiah Caldwell (animal care supervisor), and William Arndt and Roy Rice (animal caretakers).

# REFERENCES

- 1. Tardif SD, Richter CB: Competition for a desired food in family groups of the common marmoset (*Callithrix jacchus*) and the cotton-top tamarin (*Saguinus oedipus*). Lab Anim Sci 31:52-53, 1981
- 2. Flurer C, Scheid R, Zucker H: Evaluation of a pelleted diet in a colony of marmosets and tamarins. Lab Anim Sci 33:264-267, 1983
- 3. Escajadillo A, Bronson RT, Seghal P, Hayes KC: Nutritional evaluation in cotton-top tamarins (*Saguinus oedipus*). Lab Anim Sci 31:161–165, 1981
- 4. Morin M: A different approach in examining a wasting syndrome. Lab Anim 12:36-38, 41, 1983
- Richter CD, Tankersley W, Webb A: Chronic recurrent colitis: A wasting syndrome in marmosets and tamarins. 29th Annual Session, American Association of Laboratory Animal Science, New York, September 24–29, 1978 (abstract)