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MORRO BAY KANGAROO RAT CAPTIVE BREEDING PROJECT

Final Report

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The Morro Bay kangaroo rat (Dipodomys heermanni morroensis) is found in an area that covers less than 84 acres (34 ha) on the south side of Morro Bay, San Luis Obispo County, California (Villablanca, 1987a). The problems surrounding the decline of this species have been investigated and reviewed by numerous people (Stewart and Roest, 1960; Congdon and Roest, 1975; Roest, 1973, 1977; Toyoshima, 1980; Gambs and Holland, 1988). The principal factors responsible for the decline of this kangaroo rat have been the human modification and destruction of its habitat (U.S. Fish and Wildlife Service, 1982). The Lompoc kangaroo rat (D. h. arenae) is geographically the closest subspecies to D. h. morroensis. Both forms are located near the coast, but they are allopatric. The Lompoc kangaroo rat is not threatened or endangered with extinction.

The Morro Bay kangaroo rat was listed as an Endangered Species by the U.S. Fish and Wildlife Service in 1970 (Federal Register 35(199):16047) and by the State of California in 1971. The Recovery Plan was approved in August 1982 (U.S. Fish and Wildlife Service, 1982). One of the actions called for in the Recovery Plan was to "artificially increase the Morro Bay kangaroo rat population" (Task 25). In 1984, Aryan Roest of the California Polytechnic State University (Cal Poly), San Luis Obispo, initiated a captive breeding program in cooperation with the U.S. Fish and Wildlife Service and the California Department of Fish and Game (Roest, 1984). The objective of the program was to produce animals for reintroduction to their natural habitat (Recovery Plan Task 253). The colony was started with 10 wild-caught individuals, and by June 1988, the colony numbered 22 (not including 3 in an outdoor exclosure near Morro Bay) (Aryan Roest, pers. comm.). However, the number in the colony was still not large enough to carry out a reintroduction by June 1988, and efforts to continue breeding at Cal Poly had all but ceased by then.

Kangaroo rats are difficult to breed in captivity (Hatch, et al., 1971; Daly, et al., 1984), and D. h. morroensis has proven to be no exception (Roest, 1988). Until recently, most attempts to breed captive Dipodomys have involved trying to replicate their natural habitat in the laboratory (Day, et al., 1956; Chew, 1958; Butterworth, 1961). Daly, et al. (1984) developed a method of breeding D. merriami and D. microps in small laboratory cages. Roest (1988) used a similar technique for D. h. morroensis, where animals were kept individually in 10 gal. aquaria and were paired when external signs of estrus (Pfeiffer, 1960; Hatch, et al., 1971; Villablanca, 1987b) indicated females might be receptive.

Several factors contributed to our decision to try and breed the captive D. h. morroensis: 1) The limited

reproduction that had occurred at Cal Poly, 2) the cessation of breeding attempts at Cal Poly, and 3) a "natural" approach to breeding Morro Bay kanagaroo rats, involving long-term and undisturbed pairings in large cages that replicated natural conditions, had not been attempted.

In setting up our breeding protocol we emphasized two factors that have proven to be important in other attempts to breed animals with poor reproductive performances in captivity. The most important was an evaluation of the species' life history, especially its social structure and behavior. The objective was to maintain the kangaroo rats in a manner that was as consistent with their natural situation as possible (Eisenberg and Kleiman, 1977; Rathbun, et. al 1981). Secondly, some mammals do not tolerate disturbances well, with the result that they become stressed, which in turn may affect their reproductive performance (Eisenberg and Kleiman, 1977). By reducing human disturbances, we hoped to improve the rat's productivity.

Although little is known about the social organization of the Morro Bay kangaroo rat (U.S. Fish and Wildlife Service, 1982; Gambs and Holland, 1988), considerable field research has been completed on the life history of other species of Dipodomys (Behrends, et al., 1986a; 1986b; Randall, 1987). We felt that some of the published data on other kangaroo rats were especially relevant to our plans:

1. Females may be receptive during estrus for less than 24 hours (Daly, et al., 1984; Wilson, et al., 1985), and possibly for less than 6 hours.

2. In some species of Dipodomys, adults are solitary, with little home range overlap during periods of non-breeding. There is little evidence that D. h. morroensis is as strongly territorial as some other species of Dipodomys (Roest, pers. comm.; Behrends, et al., 1986a; Ward and Randall, 1987).

3. In D. merriami, females move relatively long distances from their normal home ranges during estrus, as do males during testicular enlargement. Wilson, et al. (1985) and Behrends, et al. (1986b) suggest that these female movements are related to the females taking an active role in choosing mates.

4. Not all males are good breeders (or females will not have some males?)(Daly, et al., 1984; Roest, 1988).

5. Diet (and day-length?) may be an important factor in initiating and maintaining reproductive condition (Reichman and Van de Graaf, 1975).

Most of the above factors make it imperative that a captive colony be composed of a large number of individuals (i.e., > 50), because not all of the individuals will be reproductively active. A large colony is also desirable to reduce the negative effects of inbreeding (Ralls and Ballou, 1982). Unfortunately, little could be done to overcome the small size of the captive Morro Bay kangaroo rat colony, because there probably were (and are) no more than 50 - 100 individuals left in the wild (Aryan Roest, pers. comm.). Even if it was decided to augment the colony with wild-caught animals, it has not been possible to set traps because of problems in gaining access to the occupied remaining habitat, which is privately owned (Carl Benz, pers. comm.). Because of the small size of the captive colony that we inherited and its aged condition, we realized our chances of success were small (see original research proposal). If something were not tried, however, failure was assured because attempts to breed the animals had ceased.

Period Covered:

This report covers the period from 13 January 1989, when the Morro Bay kangaroo rat colony was transferred to the Piedras Blancas Research Station, to 28 February 1990, when attempts to breed the kangaroo rats were stopped. The design of the breeding facility, the care and status of the animals, and the results of our breeding attempts are summarized. Information on the fate of animals is reviewed through 19 November 1990, when the colony was transferred to the National Zoological Park.

Breeding Facility:

Six breeding cages were constructed from plywood in a 30 x 30 ft. room at Piedras Blancas Research Station (Fig. 1). All cages were 4 ft. high with a plywood floor. Four cages were 6 X 12 ft., one was 6 X 10 ft., and a sixth cage was L-shaped, with arms 8 and 10 ft. long. Cages were divided into 2 or 3 sections by 2-feet-high Masonite partitions, and a 2-inch-square hole in the Masonite partition provided access between sections. A cement brick in front of each hole served as a door. The bricks were moved aside to allow animals to move between sections. In April 1989, each brick was replaced with galvanized hardware-cloth (0.25-inch-mesh), which was held in place with a brick. Seven-inch-high, Masonite baffles were placed in a maze-like configuration in each section. Approximately 1 in. of sand, taken from the Los Osos area, was placed in the bottom of each cage. One or two 7.5 or 14 watt red light bulbs were installed in each cage to provide illumination for night observation. These lights remained on 24-hours per day.

Nest boxes, 12 in. long, 4 in. wide, and 5 in. deep, were fitted to the outside of each cage, one near each corner. The top of each nest box could be opened to capture or check on the kangaroo rats. Originally, two 20 to 25-inch-long, 2.5-inch-diameter PVC pipes were fitted to each nest box from the inside of the cage. On 1 and 2 March 1989, these entrance tunnels were removed so that the animals entered each nest box by the two 2.5 in. diameter holes through the sides of the cages.

The temperature in the breeding room (55-70^{0}F) was intermittently monitored and it remained similar to outside ambient temperature, but was buffered from high and low extremes. An artificially extended light cycle maintained the colony on 14 hours of daylight and 10 hours of darkness, which is the same light cycle that occurs naturally during the peak breeding season (April-May). On 8 February 1989, the windows of the facility were blackened and the light cycle was reversed to simulate night conditions during daylight hours (10 hr. dark cycle starting at 1000 hr.). This reversal, which is commonly done in research collections, made observations more convenient for us. Between 6 and 10 August 1990 the light cycle was gradually changed to ambient conditions and the window covers removed.

The feeding schedule for individuals in the captive colony was:

Monday	1/2 large leaf romaine lettuce 1 small slice apple (ca. 0.2 oz.) 1/2 cup seed mix
Wednesday	1/2 large leaf romaine lettuce
Friday	1 large leaf romaine lettuce 1 small slice apple (ca. 0.2 oz.) 1/3-1/2 cup seed mix

Quaker Old Fashioned rolled oats and sunflower seeds were mixed with Special Cockatiel Mix (see below) in a ratio of about 1 part each to 10 parts Special Cockatiel Mix, respectively. Special Cockatiel mix is made by The Seed Factory, P.O. Box 1830, Modesto, California. It is composed of: 20% white proso millet, 25% red proso millet, 15% safflower (whole), 2% steel cut oat groats (red color, human consumption grade), 2% steel cut oat groats (green color, human consumption grade), 12% recleaned whole wheat, 10% recleaned canary seed, and 15% buckwheat.

The manufacturer guarantees the following nutrient composition: >13% crude protein, > 8% crude fat, < 4% crude fiber, <11% ash, and <11% moisture.

Animals:

On 13 January 1989, 21 D. h. morroensis (9 males and 12 females; Table 1) and 14 D. h. arenae (9 males and 5 females; Table 2) were received from Aryan Roest at Cal Poly. The arenae were originally acquired to use in developing techniques prior to applying them to morroensis. We continued to use the arenae as surrogates whenever new procedures were tried.

Some of the kangaroo rats had lost their monel metal ear tags, which had been previously applied, and many of those that retained their metal ear tags showed signs of physical irritation on the pinnae. Before any morroensis animals were removed from their aquaria (each tank was labelled with the individual identification of the animal it contained), new tags were fitted to those that had lost tags. Four of the arenae were also re-tagged. Each new ear tag incorporated a passive implantable transponder (PIT) that is about the size of a rice grain, and when activated with a reader-wand, produces a unique 10-digit alphanumeric code (Tables 1 and 2). Each tag was constructed from nylon monofilament fishing line, a 1/4-inch-diameter, colored disk cut from Dymo labelling tape, and a PIT. The nylon was cut into a 1/2-inch-long post and one end was melted to form a knob. The PIT was then epoxied onto the knob, so that the nylon post was centered and perpendicular to the PIT. A small hole was then pierced through the pinna of a kangaroo rat and the nylon post inserted through the hole from the back of the pinna. A small hole was punched through the center of the colored disk, which was threaded onto the nylon post, resulting in the pinna being sandwiched between the PIT and the disk. The remaining end of the nylon post was melted to form a knob, maintaining the disk in place. These ear tags have not irritated the pinnae, the colored disks enabled us to individually identify animals while free-ranging in their cages, and the PIT could be read without opening the nest boxes.

Major events, such as capture dates, birth dates, and parentage were incorporated onto individual cage cards, along with information on daily events, such as reproductive condition, changes in cage configuration, location, pairings, etc. A master log book, with detailed notes on all activities with the colony, was also maintained.

To reduce the possibility of inbreeding, past breeding records were examined to determine potential male-female pairings. A list of males for each female was made, excluding siblings, offspring, fathers, nephews, and uncles. Male 1540 was also excluded from female 1412's list of potential mates because they were trapped in the same area two days apart, and therefore may have been related.

Presently (19 November 1990) there are 6 arenae (4 males and 2 females) and 7 morroensis (4 males and 3 females) in the colony. The average age of the seven surviving morroensis is 48.5 months, with five of them older than four years of age (Tables 1 and 2). The average age of the 14 morroensis at death at Piedras Blancas was 53 months.

Methods of pairings:

Before trying to breed morroensis, several different techniques were tried first with arenae individuals. Initially, the animals were observed continuously during all introductions, and were separated when not being watched. After a reasonable time elapsed with no serious problems, the kangaroo rats were allowed to remain for greater lengths of time without continuous observation. First, a cage was tested with arenae to ensure that the design would not result in unforeseen problems. Then, on 28 January 1989, three arenae (males 350 and 381, and female 363) were placed in a cage with the passage between the cage divisions open so that all three animals had access to each other. On 1 February, male 381 was found with slight wounds and male 350 was severely wounded; male 350 died the next day. The doors between the divisions were closed before male 350 was replaced with male 344. Although the passage between the cage divisions remained closed, we discovered that the two males were able to cross cage sections by crawling under a small gap between the divisions and the cage floor. The result was fighting between the males. Thereafter, the gap was plugged (and all other cages were checked) and we allowed the female access to only one male at a time.

On 16 February 1989, the passage between female 363 and male 344 was opened for three days. On 17 February both animals were in good health, but on 18 February the female was found moribund from severe lower back and tail injuries and was euthanized later that day. Female 345 was introduced to the cage to replace female 363.

During our observations, we determined that much of the fighting was occurring inside the nest boxes, and despite the two entrances to the boxes, the defending kangaroo rats refused to leave the shelters. Rather, they remained in the boxes and became wounded in the close quarters. We also found that the kangaroo rats spent very little time outside their next boxes, even when paired, which resulted in a great amount of time observing "empty" cages. On 1 March 1989 we removed the PVC pipes leading to the nest boxes and blocked the entrances to the nest boxes with bricks. By excluding the arenae from their shelters, we hoped to minimize wounding by allowing subordinate animals to escape the aggression of dominant animals. We also hoped to encourage animals to move about the cage and interact with

each other more frequently, thereby reducing the time it took us to determine compatibility. On 7 March 1989, the passage between female 345 and male 381 was opened and their nest boxes blocked. They lived harmoniously in the cage for 6 days, after which there were signs of aggression directed at the male by the female. On 20 March, male 381 was found dead in the cage from a severe wound near the dorsal gland.

We next tried (in desperation) a method of pairing that we hoped would allow animals to breed when the female was in estrus, but would separate them at other times. On 18 April we taped a thick, nylon hair-net over the end of a 3-inch-diameter, 6-inch-long PVC pipe, so that it formed a small sack big enough to hold a kangaroo rat. The open end of the pipe was placed over the doorway of the female's cage section and the netted end extended into the male's section. Theoretically, the net would separate the animals (since they are not as avid chewers as many other rodents) but would allow them to breed through the mesh when the female was receptive and solicited copulation. Unfortunately, one of the kangaroo rats chewed through the net within hours after installation of the net, and the male began to chase the female (she was known to be anestrus).

Another technique that we tried had been successfully implemented by Mr. Potter of Creston, California, now deceased. Mr. Potter housed 7-8 male and female D. h. jolonensis in a single 10-gal. aquarium. Although the animals were very crowded, apparently little fighting occurred and several females gave birth. We theorized that the crowded conditions might break down the normal social structure of the kangaroo rats, which we were learning was characterized by high levels of intraspecific aggression.

In an attempt to duplicate Mr. Potter's methods, on 25 April 1989 we placed five arenae (females 343, 345 and 385, and males 335 and 344) into a neutral 10-gal. aquarium containing three PVC tubes. Initially, the kangaroo rats ran wildly around the cage and fought over possession of the tubes. When the tubes were removed, all the animals frantically ran and jumped around the cage in a haphazard manner, bouncing off the sides and top of the aquarium. We quickly put the tubes back in, which were immediately claimed by the three females. After 30 to 40 minutes, it became apparent that maintaining a group of kangaroo rats in an enclosure of this size would probably result in mortalities; therefore the animals were returned to their respective individual aquaria.

On 3 May 1989, we duplicated the above technique in a larger area. We placed five arenae (males 335, 336 and 391 and females 343 and 385) in one quarter of a breeding cage (3 X 6 ft.). Initially, no nest boxes or tubes were provided; the kangaroo rats wandered around the cage looking

for shelter and fought violently when they contacted one another. After five PVC tubes were added to the cage, the frequency of aggressive interactions temporarily decreased. The animals were observed intermittently for about 5 hr. Periods of intense aggression were followed by lulls with few interactions and little fighting. Four hours after the kangaroo rats were placed in the cage, male 391 was removed and returned to his aquarium because he continually followed, attempted to mount, and bit the back of another individual (possibly female 343). The remainder of the animals were separated one hour later when aggressive behavior among the individuals increased in frequency and intensity.

The reasons for Mr. Potter's success are unknown. Perhaps he placed his animals in a single aquarium while they were young and therefore they adjusted to the close quarters more effectively, or perhaps he had initial fighting and mortality before the animals adjusted. It is also possible that D. h. jolonensis, which is found inland compared to D. h. arenae and morroensis, behaves differently. Because any mortality in morroensis individuals is unacceptable, the Potter method was considered too risky and not tried with any morroensis.

Based upon the results of these initial pairing trials, we concluded that intraspecific aggression will likely prevent pairs or small groups of Morro Bay kangaroo rats from living harmoniously in cages of our size (6 x 12 ft) over any meaningful length of time.

When the initial "natural" pairing techniques with arenae proved to be unsuccessful, we began to visually check for estrus in morroensis females every Monday, Wednesday, and Friday, starting on 27 March 1989. Starting in early May 1989, we used the same criteria developed by Villablanca (1987b; Table 3). The females were blocked in their nest boxes and then removed and examined while in the hand. When genital swelling was noted, regardless of the perforate condition of the vagina, females were paired with one or more males. Up to four males were introduced to the females, one at a time. Initially, we paired individuals in either of their home cages. We also paired animals in two neutral, unoccupied large cages, which were connected with two PVC-pipe tunnels. At first, 2-inch-diameter PVC pipes were used to connect the cages. However, when animals began to nest in the tunnels, we inserted 1-1/2-inch diameter PVC pipes into the 2 inch pipes, which made them less desirable as shelters. Each pair was left together until external signs indicated the female was anestrus. This arrangement created a larger breeding area and, when necessary, allowed the animals to escape from each other through the tunnels.

On 17 May 1989, a pre-estrous arenae female (345) and male 386 were placed in a cage complex, as described above, and remained there until the morning of 20 May. The animals appeared to adjust fairly well to each other's presence and no serious injuries resulted. We then began to pair morroensis animals in this manner, using females that were nearing estrous. When animals could not be observed continuously, they were separated from each other within the cage complex to prevent injury.

After several pairings using the entire cage complex described above, we observed that the morroensis were establishing territories and were avoiding contact with each other. We increased the likelihood of contact between animals by decreasing the size of the breeding arena by using sections of single cages. Occasionally, we also attempted to pair females with males in adjacent sections of the same cage by opening the doorway between their respective cage sections.

Attempts to assess estrus and pair animals were discontinued on 28 February 1990.

Results of pairings

The outcome of contact between individuals varied, and unlike Roest's findings, where females primarily directed aggression towards males (Roest, 1988), we found that most often the males chased the females in an aggressive or investigatory manner (Table 4). If a female was not receptive during the first few minutes after the introduction of a male, breeding did not occur. Fewer females cycled during the fall and winter months, and those that did cycle, were in estrus less frequently than in the spring and summer (Figs. 2-5). Morroensis were paired up to 480 minutes (Table 5). When their interactions became too aggressive, when the female did not appear to be receptive to the male, or after a period of time when the animals avoided each other without breeding, they were separated.

Between 9 May 1989 and 19 January 1990, only two arenae females (343 and 345) showed signs of estrus. During this period, 32 attempts were made to breed these two females, one of which resulted in a copulation. On 10 July 1989, arenae female 345 and male 382 copulated, and she gave birth to one stillborn pup on 11 August 1989.

Between 9 May 1989 and 19 January 1990, 40 pairings were attempted with morroensis females 323, 326, 327, 348, 368, 375, and 1412. Four copulations resulted from these pairings, but no pups were born. Female 348 and male 372 copulated on 19 May 1989 and 18 July 1989. Female 327 copulated with male 322 on 25 May 1989 and on 5 April 1989

she was forced to copulate with male 370, although she was not receptive to him.

Vocalizations

Morroensis and arenae females typically emit a soft "whirring" vocalization when receptive to a male (Villablanca, pers. comm.). On 30 October 1989 we noticed that arenae male 382 emitted a similar vocalization when paired with female 345. After this date, he was paired nine more times with females 345 and 343, and emitted this vocalization during four of those pairing attempts. None of these pairing attempts resulted in copulations. However, male 382 did successfully copulate with female 345 on 10 July 1989. A "whirring" vocalization was heard at that time, however it was thought that the female was vocalizing and not the male.

Health and mortality

Since January 1989, 8 arenae (3 females and 5 males) and 14 morroensis (9 females and 5 males) have died. As outlined in the research study plan, all carcasses were sent on dry ice to the National Zoological Park for storage in an ultra-cold freezer and eventual necropsy, and genetic and contaminant analyses. Tom Williams from Monterey and John Truax from Morro Bay were the consulting veterinarians for this project. The following is a chronological account of the health and mortality problems encountered in the colony, excluding the three arenae deaths described in the Methods of Pairing section of this report.

On 28 January 1989, morroensis female 1391 was found dead in her aquarium, 15 days after her arrival at Piedras Blancas. There were no external signs indicating the cause of death. The carcass was sent to the National Zoo on 31 January 1989.

Morroensis male 1579 was found dead in his nest box on 13 March 1989. His body was too decomposed for necropsy, so it was stored in a normal freezer at the Piedras Blancas Research Station before being transferred to the ultra-cold freezer at the National Zoological Park on 13 June 1989.

On 24 April 1989 we noted that arenae male 388 had two large lumps on his back and exudate on his pinnae. He was taken to Truax, who diagnosed the swellings as mass cell tumors. The animal was euthanized the next day.

On 2 June 1989, morroensis male 1540 was found dead in his aquarium; there were no external signs indicating a cause of death.

On 3 March 1989, a 2-inch-diameter swelling was found in the perianal region of morroensis female 333. When the swelling continued to enlarge, she was sent to Williams on 6 March, who diagnosed the swelling as a serona (serum deposit after a hematoma). The fluid from the swelling was drained by Williams, but no tumor cells were found. She was returned to Piedras Blancas on 13 March 1989, and housed in an aquarium so that the swelling could be drained of fluid every one-to-two days. Because the swelling did not improve, we obtained a second opinion from veterinarian John Truax on 8 May 1989. He diagnosed a malignant fibrosarcoma. The animal died on 11 June 1989. On 13 June 1989, the carcasses of #1579, #1540, and #333 were shipped to the National Zoological Park.

Morroensis male 324 died in his nest box between 7 and 10 July 1989. His carcass was sent to the National Zoological Park on 11 July 1989. There were no external signs indicating cause of death.

Morroensis female 332 was noted to have a small, hard tumor-like lump in the left urogenital region on 24 March 1989. The lump increased to about 3/8 inch in diameter and a second swelling developed anterior to it. The lumps were examined by Truax on 2 May 1989 and diagnosed as cystic fibrosarcomas, most likely malignant (no laboratory tests were done). Truax suggested that removal of the tumors would probably result in the formation of new tumors. Since this type of tumor is possibly caused by a virus, they could be passed on to progeny. Truax suggested that the tumors probably would become fibrous and interfere with the kangaroo rat's urogenital system and left rear leg function and most likely the animal would have to be euthanized. The sarcoma continued to enlarge until we decided on euthanasia. On 20 July 1989 Frances Villablanca, from the University of California at Berkeley, collected the animal. He euthanized her and used a femur for chromosome analysis. The rest of the carcass was sent to the National Zoological Park.

During handling, arenae female 343 struggled excessively and injured a hind leg on 23 October 1989. She appears to be in good health, although she walks with a slight limp (see Muths, 1990).

On 20 November 1989, arenae male 386 was found dead in his aquarium. He died between 16 and 20 November and his body was moderately decomposed.

On 13 December 1989 morroensis female 1584 was observed shaking her body in a wobbly manner, as she wandered around her cage during daylight. The next day she was found dead in her cage with seeds in her cheek pouches. The carcasses of arenae 386 and morroensis 1584, as well as a stillborn

pup from arenae female 345, were sent to the National Zoological Park on 20 December 1989.

In January 1990, a hard pea-sized tumor was observed on the ventral side of morroensis female 326 near her left hind leg. The tumor appears to be enlarging and looks very similar to the cystic fibrosarcoma seen in morroensis female 332.

On 5 March 1990, morroensis female 368 was found dead in her nest box. She died between 2 and 5 March. There were no externals signs indicating cause of death. She was shipped to the National Zoo on 6 March 1990.

Morroensis female 375 had a permanently swollen genital area associated with a clear or cloudy discharge. At times the discharge hardened into a greenish plug. This condition was chronic since 21 March 1989. The condition may have adversely affected her estrous cycle, or at least obscured external signs of estrus, because obvious signs of estrus were never seen. On 7 March 1990, she was found dead in her aquarium. Truax performed a necropsy on 9 March 1990 and concluded that a crushed pelvis, hepatic carcinoma, and purulent inflammation of the intestinal tissue contributed to the death of the animal. We do not know how the animal's pelvis was broken. However, captivity is known to weaken bone tissue in kangaroo rats (Muths, 1990).

Morroensis female 320 was found dead in her nest box on 9 April 1990. She had broken a hind leg during handling on 9 May 1989 (see Muths, 1990). She moved with a pronounced limp, and was consistently thin in appearance until her death. She was sent to the National Zoo on 10 April 1990.

Arenae female 385 was found dead in her nest box on 30 April 1990. She died between 27 and 30 April and was slightly decomposed when found.

Morroensis female 1412 was found dead in her nest box on 22 August 1990. She had a bean-sized lump surrounding her urogenital area that first appeared as a small white lump lateral to the vagina in mid-April 1989. The hard lump gradually enlarged and spread so that it covered the entire urogenital area. It obscured signs of estrus, except for the occasional appearance of small drops of fresh or dried blood, or a thin crusty discharge.

Morroensis female 323 was found dead in her nest box on 18 June 1990. She died between 14 and 18 June from unknown causes.

Arenae female 345 was found dead in her cage on 20 July 1990. The cause of death was wounds inflicted by arenae female 343. The two rats were able to come into contact

with one another when the brick covering the hole in the partition between the cages was accidentally dislodged.

Morroensis male 372 was found dead in his nest box on 3 August 1990. He died between 1 and 3 August from unknown causes.

Morroensis male 370 was found dead in his nest box on 20 August 1990. He died between 15 and 20 August and cause of death was related to a large tumor that developed on his right side.

The carcasses of morroensis female 375 and arenae females 363 and 385 and males 350, 381, and 388 were sent to the National Zoo on 27 June 1990.

In summary, most of the kangaroo rats that died have not undergone necropsy. Half of the 8 arenae deaths were caused by intraspecific aggression during our initial attempts at pairing or during accidental encounters (Table 6), while tumors and carcinomas were identified as contributing to 4 of the 14 deaths of morroensis.

Consultations:

Several biologists have been consulted about kangaroo rat housing, diet, determination of reproductive condition, breeding techniques and behavior. These biologists include Aryan Roest and Roger Gambs from Cal Poly, Martin Daly of McMaster University in Canada, Jan Randall from San Francisco State University, Frances Villablanca from the University of California at Berkeley, Dick McMillan from the University of California at Irvine, Miles Roberts from the National Zoological Park, and Bill White and Alan Metzler from the Charles Paddock Zoo in Atascadero.

Summary:

1. Several configurations of the large breeding enclosures were tried unsuccessfully.

2. Between 27 March 1989 and 28 February 1990, we visually assessed estrus of all females 3 times/week and paired those that showed signs of estrus.

3. One arenae pairing and three morroensis pairings resulted in copulations (total pairings = 109).

4. Arenae female 345 gave birth to one stillborn pup 31 days after mating with male 382.

5. Our failure to successfully breed D. heermanni may be attributed to several factors, including the advanced age and small size of the colony, male impotency, high levels of intraspecific aggression, male\female incompatibility, and the need for a more intensive program of monitoring for estrus.

6. Six arenae (4 males and 2 females) and 7 morroensis (4 males and 3 females) remain in the colony.

7. Two arenae and one morroensis were euthanized and three morroensis died from cystic fibrosarcoma tumors. Three arenae died from wounds inflicted by conspecifics. Three arenae and 10 morroensis died from as yet undetermined causes, but presumably related to their old age.

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Table 1. Status of Dipodomys heermanni morroensis while at the Piedras Blancas Research Station (13 January 1989 through 19 November 1990). * =still alive. L=left. R=right. Dash=unreadable number on tag. ?=tag unconfirmed before death and shipment to National Zoo.

ID#	Sex	PIT/ear tag no.	Date Born/Trapped	Months in captivity	Condition
*322	M	7F7E664413/Red L	Born 21 Jun 86	52.5	
324	M	7F7E407801	Born 21 Jun 86	36.5	Died 10 Jul 89 of unknown causes.
*325	M	7F7E406C1D/Blue R	Born 21 Jun 86	52.5	
*330	M	7F7E663C71/Red R	Born 6 Oct 86	49.5	
*349	M	349 R	Born 21 Jul 87	39.5	
370	M	7F7E66491F/metal R	Born 15 Apr 85	64.5	Died 20 Aug 90, large tumor right side.
372	M	7F7E407331/Blue R	Born 30 Jul 84	72	Died 3 Aug 90 of unknown causes.
1540	M	?	Trapped 4 Apr 85	50	Died 2 Jun 89 of unknown causes.
1579	M	7F7E663128	Trapped 17 May 85	46	Died 13 Mar 89 of unknown causes.
320	F	?	Born 30 Jul 84	67.5	Died 9 Apr 90 of unknown causes.
323	F	7F7E66441B/Black R metal L	Born 21 Jun 86	47.5	Died 18 Jun 90 of unknown causes.
*326	F	326 L	Born 13 Jun 86	53	Estrous cycling female; tumor-like lump on ventrum.
*327	F	327 L	Born 13 Jun 86	53	Estrous cycling female.
332	F	?	Born 6 Oct 86	34.5	Euthanized 20 Jul 89, fibrosarcoma.
333	F	7F7E40704A	Born 6 Oct 86	32	Died 11 Jun 89, fibrosarcoma.
*348	F	34__ R	Born 21 Jul 87	39.5	Estrous cycling female.

Table 1, continued

ID#	Sex	PIT/ear tag no.	Date Born/Trapped	Months in captivity	Condition
368	F	?	Born 15 Apr 85	58.5	Died 5 Mar 90 of unknown causes.
375	F	7F7E664405	Born 15 Apr 85	58.5	Swollen vulva with discharge. Died 7 Mar 90.
1391	F	?	Trapped 2 Aug 84	54	Died 28 Jan 89 of unknown causes.
1412	F	7F7E663820/Green L	Trapped 6 Apr 85	65	Tumor on vulva masks estrus. Died 22 Aug 90.
1584	F	7F7E406C6F	Trapped 5 Jun 85	54	Died 14 Dec 89 of unknown causes.

Table 2. Status of Dipodomys heermanni arenae while at the Piedras Blancas Research Station (13 January 1989 through 19 November 1990). * =still alive. L=left. R=right. Dash=unreadable number on tag. ?= tag unconfirmed before death and shipment to National Zoo.

ID#	Sex	PIT/ear tag no.	Date Born/Trapped	Months in captivity	Condition
*335	M	7F7E406C7D/Black R	Born 11 Feb 87	41	
*336	M	Torn L	Born 11 Feb 87	41	
*344	M	84__ L	Born 17 Apr 87	39	
350	M	7F7E663822	Born 21 Jul 87	30.5	Died 2 Feb 89 from wounds in pairing attempt.
381	M	7F7E406F04	Born 4 Mar 86	36.5	Died 20 Mar 89 from wounds inflicted in pairing attempt.
*382	M	Torn L	Born 4 Mar 86	52.5	
386	M	?	Trapped 29 Jan 84	70	Died 20 Nov 89 of unknown causes.
388	M	?	Born 8 May 84	59.5	Euthenized 25 Apr 89, mass cell tumors.
391	M	None	Trapped 5 Feb 84	79	Died 22 Aug 90 of unknown causes.
*334	F	484 L	Born 11 Feb 87	38.5	Estrous female
*343	F	Torn L	Born 17 Apr 87	36.5	Estrous female. Broken or injured hind leg.
345	F	__4__ L	Born 17 Apr 87	27.5	Died 20 Jul 90, wounds inflicted by Female 343.
363	F	7F7E663554	Born 8 May 84	57.5	Euthenized 18 Feb 89, wounds inflicted in pairing attempt.
385	F	835 L	Born 4 Mar 86	50	Died 30 Apr 90 of unknown causes.

Table 3. Levels of vulval swelling and vaginal discharges used to assess estrus in captive kangaroo rats (based on Villablanca, 1987b).

Vulval Swelling

1. Flat. The vulva does not rise above the surrounding area and is almost entirely covered by the clitoris.

2. Slightly raised. The vulva is slightly raised above the level of the surrounding area and its diameter is greater than that of the clitoris.

3. Moderately raised. The vulva is noticeably above the surrounding area. It is often conical in shape, or else it will appear as a small cylinder that is as long or longer than it is wide.

4. Large. The vulva resembles the slightly flattened top half of a sphere. The top is still rounded along the edges. The vaginal lips may vary from taut to highly undulating.

5. Fully distended. The vulvar skin is taut, and the vulva appears like a flat or round-topped cylinder that is wider than tall.

Vaginal Discharges

1. No discharge

2. White dried flaky crust

3. Striated vaginal cast (post estrus).

4. Plug: mucus, or black, or post copulatory was noted.

5. Blood fresh or dried.

6. Clear discharge

7. Cloudy discharge

Table 4. Intraspecific interactions resulting from attempts to pair estrous female kangaroo rats with a single male. (See Appendix A for definitions).

	Morroensis	Arenae
1. Male chases female, female flees	39	22
2. Male pursues female, female receptive	1	9
3. Female chases male, male flees	10	0
4. Copulate	4	1
5. Both flee	2	0
6. Fighting	10	1
7. Male ignores female	4	1
Total	70	34

Table 5. Summary of attempts to pair female and male captive kangaroo rats.

	Morroensis	Arenae
Total pairings	72	40
Total pairings of two animals	72	37
Total pairings of two animals, results could be determined	70	34
Total pairings used to determine duration of pairing	69	33
Duration of pairings, x̄±S.D. (min.)	50.6±91.07	82.1±233.7
Duration of pairings, range (min.)	1-480	2-1335

Table 6. Number of D. h. morroensis and D. h. arenae deaths by probable cause of death at the Piedras Blancas Research Station.

CAUSE OF DEATH	Morroensis	Arenae
Wounds from pairing attempts	0	3
Euthanized after wounded while paired	0	1
Euthanized because of tumors	1	1
Fibrosarcomas or tumors	3	0
Unknown	10	3
Total	14	8

FIGURE CAPTIONS

Figure 1. Floor plan of the captive kangaroo rat facility in the old Engine Room at the Piedras Blancas Research Station, San Simeon, CA. Cages are numbered 1 through 6. Cross-hatching represents areas that can not be used for large cages. Aquaria are kept on the shelves. The platform area was used for food storage and preparation. The engine is not functional.

Figure 2. Temporal distribution of Morro Bay kangaroo rats (N = 6) that were in estrus, based on the occurrence of a perforate vagina or a vaginal cast (see Table 3).

Figure 3. Temporal distribution of Morro Bay kangaroo rats (N = 6) that were in estrus, based on the occurrence of vaginal swelling of level 3 or greater, perforate vagina or a vaginal cast (see Table 3).

Figure 4. Temporal distribution of Lompoc kangaroo rats (N = 3-4) that were in estrus, based on the occurrence of a perforate vagina or a vaginal cast (see Table 3).

Figure 5. Temporal distribution of Lompoc kangaroo rats (N = 3-4) that were in estrus, based on the occurrence of vaginal swelling of level 3 or greater, perforate vagina, or a vaginal cast (see Table 3).

FIGURE 1

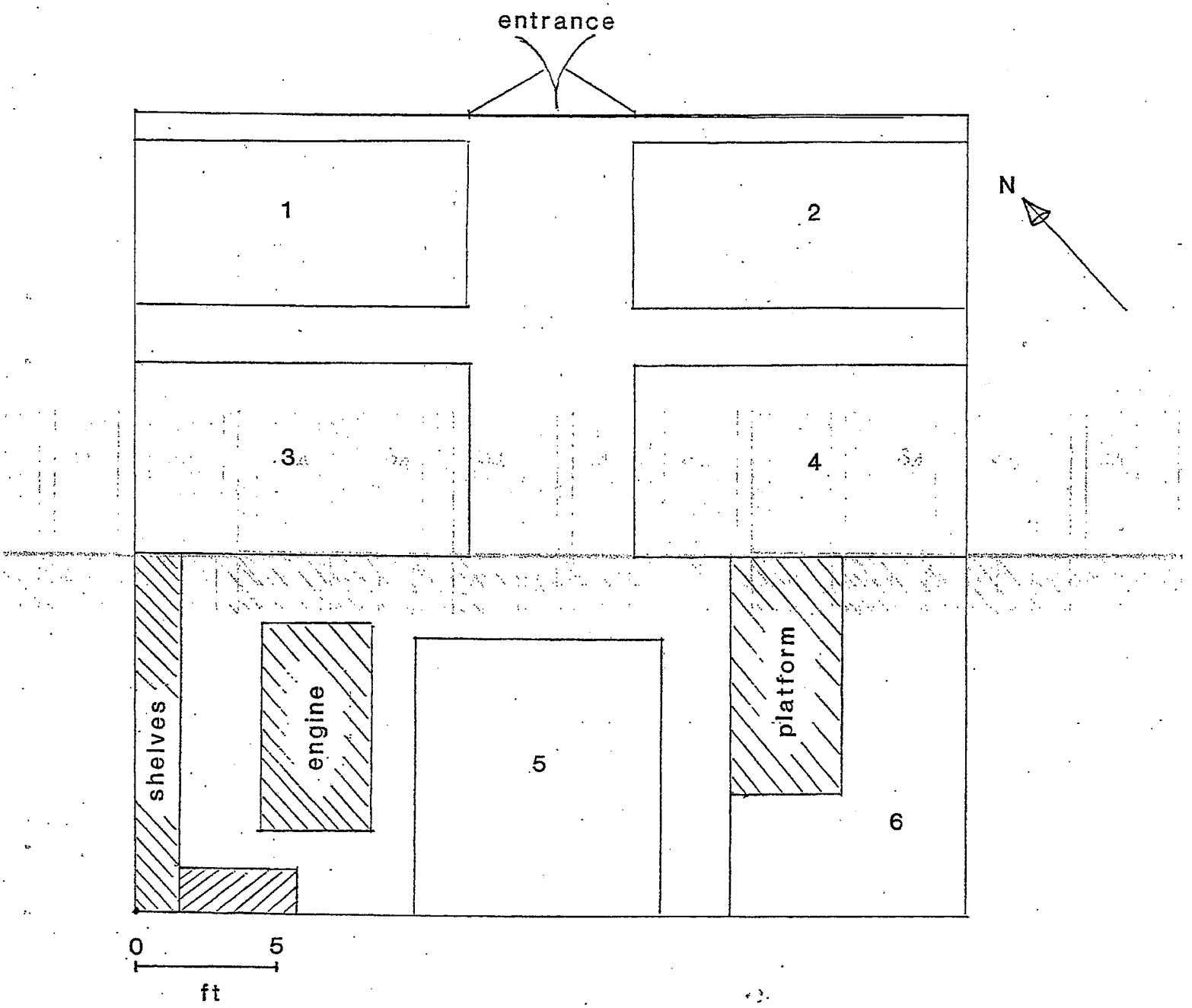


FIGURE 2

MORRO BAY KANGAROO RATS IN ESTRUS (N=6)

March 6, 1989 to February 28, 1990

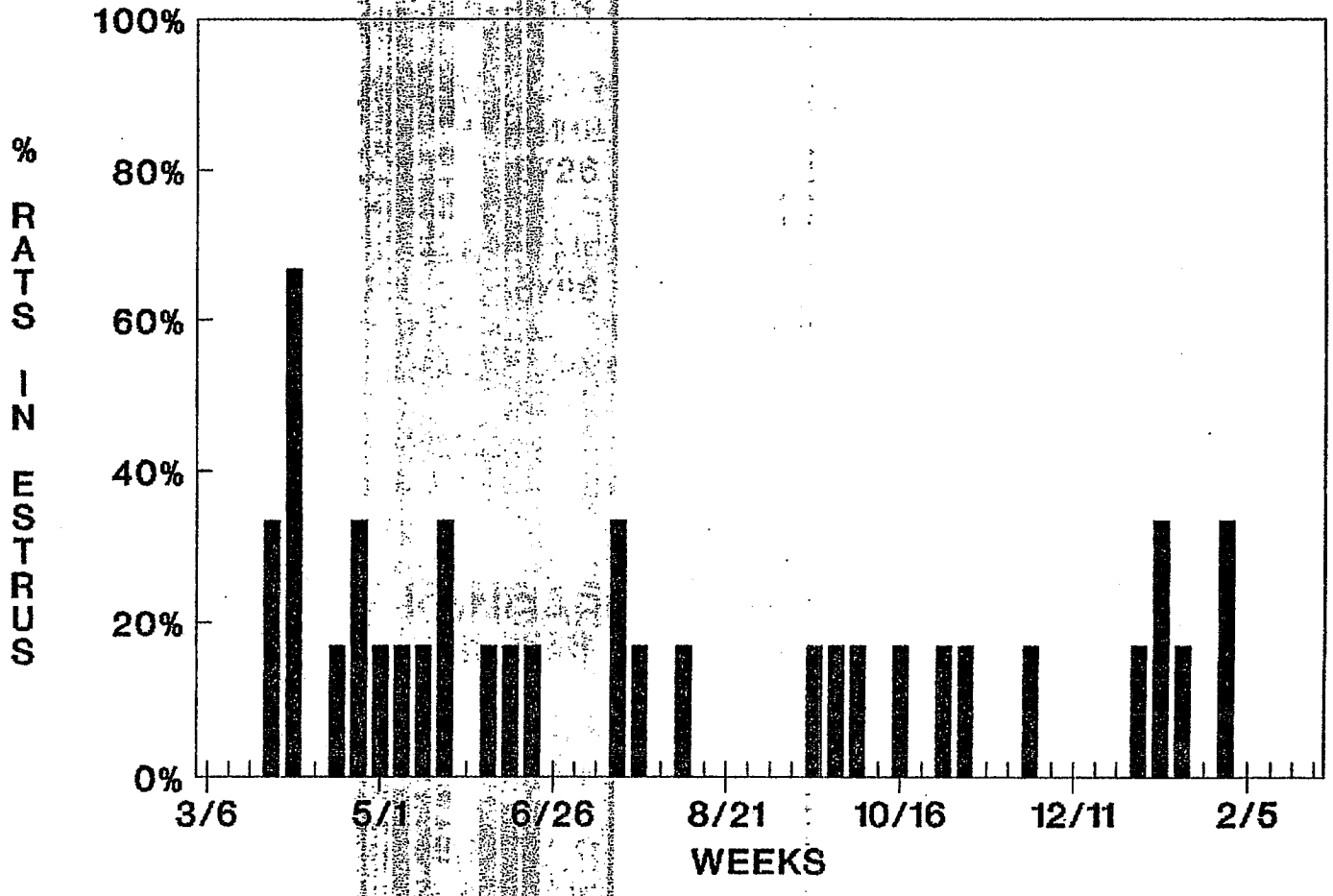


FIGURE 3

MORRO BAY KANGAROO RATS IN ESTRUS (N=6)

March 6, 1989 to February 28, 1990

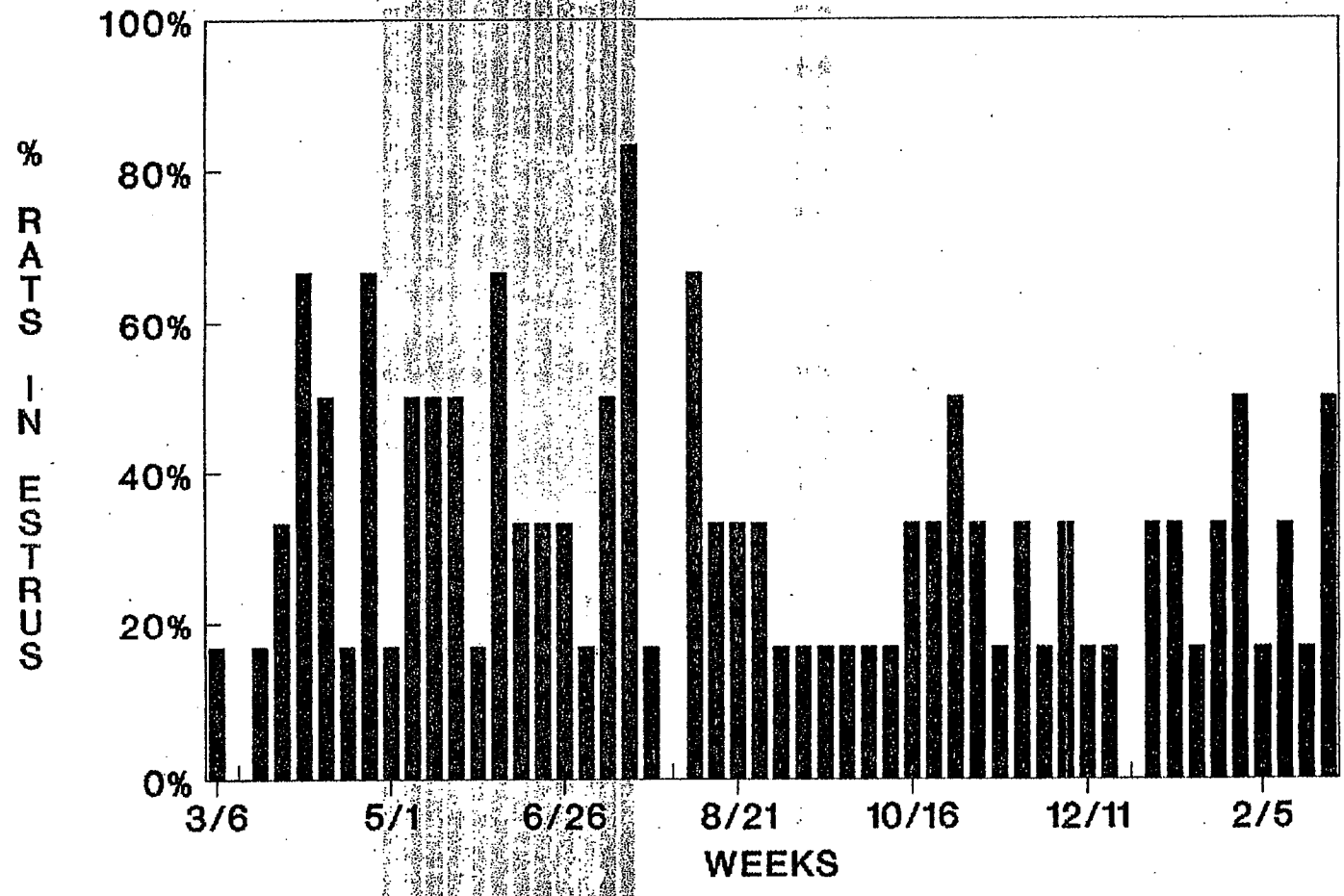


FIGURE 4

LOMPOC KANGAROO RATS IN ESTRUS (N=3-4)

March 6, 1989 to February 28, 1990

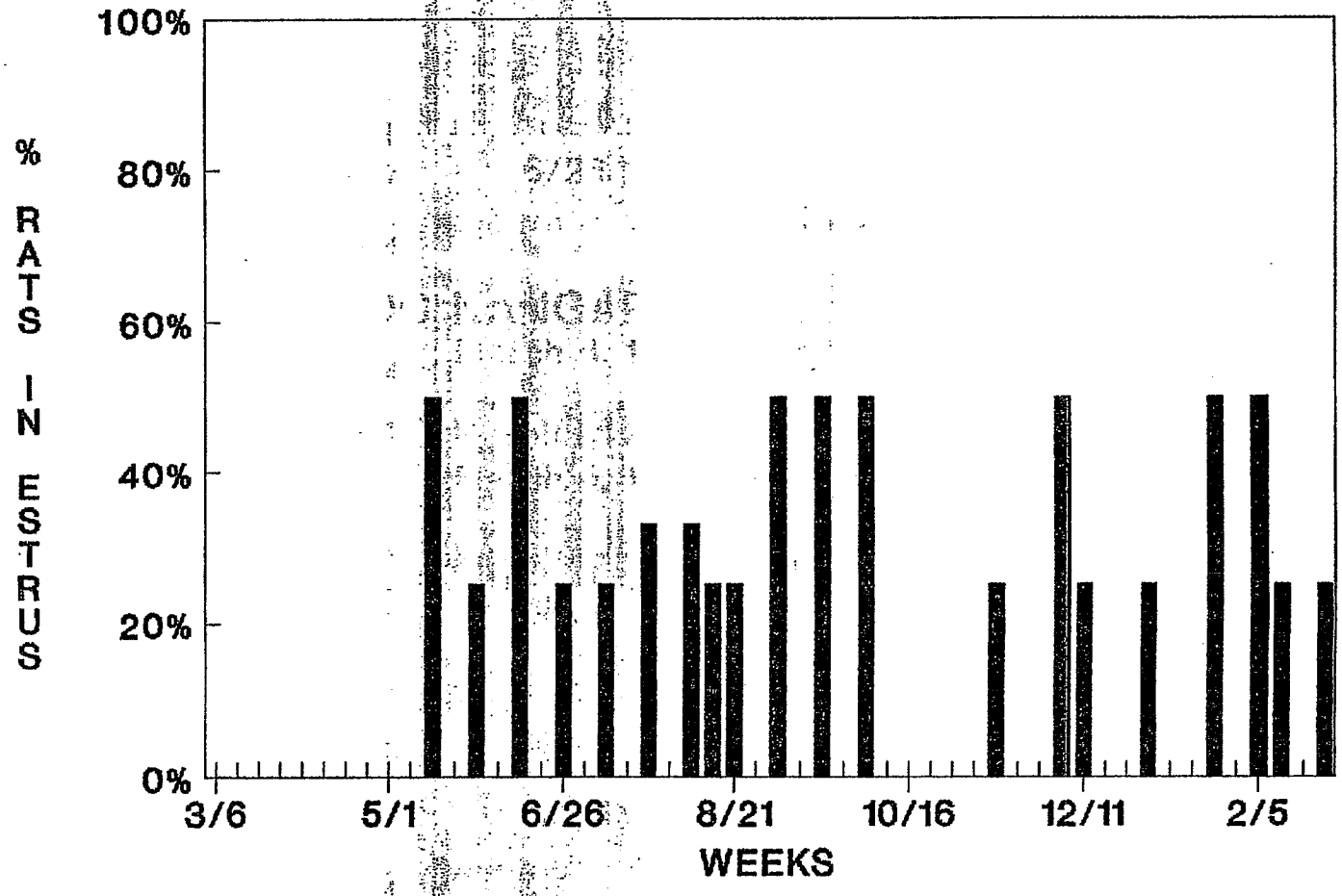
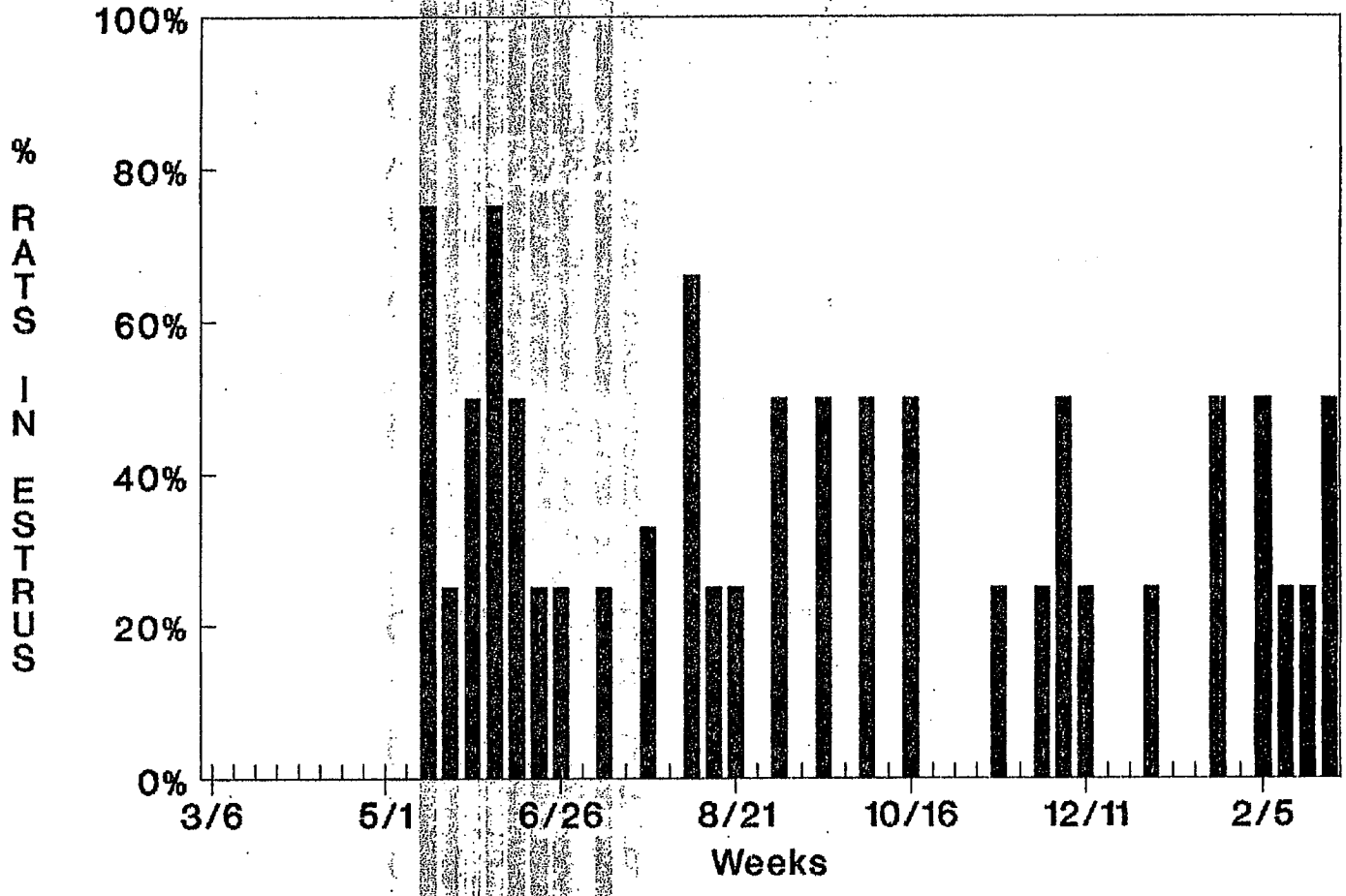


FIGURE 5

LOMPOC KANGAROO RATS IN ESTRUS (N=3-4)

March 6, 1989 to February 28, 1990



Appendix A. Definitions for Pairing Categories (see Table 4)

1. Male chases female, female flees.
   a. Male chases female aggressively, female flees.
   b. Male chases female nonaggressively, female flees or hides.
   c. Male chases female, unknown if chase was aggressive or not.
   d. Male only chases female when she approaches him (territorial behavior)
   e. Includes pairings where other behaviors also occurred, but male chasing female was the predominant activity.

2. Male pursues female, female seems receptive, but does not allow copulation. Female does not flee from male, and may even approach him. Often times circling behavior occurs and the female may allow male to mount her, but does not allow copulation.

3. Female chases male, male flees or hides. Fighting may or may not occur.
   a. Includes pairings where other behaviors occurred, but female chasing male is the predominant behavior.
   b. Female investigated male and chased him when he fled.

4. Copulate - includes all pairings that resulted in copulation.

5. Both flee - includes all pairings where fleeing by both the male and female was the predominant behavior.

6. Fighting
   a. Includes all pairings where aggressive fighting, low intensity boxing matches, or brief scuffles were the predominant behavior, and very little or no chasing occurred.
   b. Includes pairings in which the male is the aggressor, female is the aggressor, both are aggressive, or when the aggressor is unknown.
   c. Includes one pairing where much fighting occurs, both the male and female are aggressive, and both the male and female flee.

7. Male ignores female (no chasing occurs)
   a. Female climbs on the male's back and the male exhibits no response.
   b. Female flees when she comes into contact with the male, but the male exhibits no response.
   c. Female acts receptive or interested in the male, but the male exhibits no response.