

TWO NEOTROPICAL RODENTS AS BIOASSAY TEST ORGANISMS

Thesis for the Degree of M. S. MICHIGAN STATE UNIVERSITY ANN C. UNDERHILL 1972

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ABSTRACT

TWO NEOTROPICAL RODENTS AS BIOASSAY TEST ORGANISMS

Ву

Ann C. Underhill

Weanlings of three rodents, <u>Sigmodon fulviventer</u>, <u>S. hispidus</u>, and <u>Akodon azarae</u>, were evaluated to determine their usefulness as bioassay test animals for nutrition studies and the findings compared with those for <u>Microtus pennsylvanicus</u> which has previously been used.

Experiment I determined that 7% protein, using casein as the standard, was most efficiently utilized by the animals as shown by the Protein Efficiency Ratio (PER). Experiment II tested the qualities of grain proteins from two wheat-rye hybrids (Triticale-MSU #358 and Triticale-Rockefeller PC70) in comparison with that of casein, all at the 7% level.

It appears that both species of cotton rats would be highly suitable as bioassay test mammals. They both responded to protein quality in a manner similar to M. pennsylvanicus. Akodon azarae also followed much the same pattern, except the PER values were somewhat lower in Experiment I and significantly lower (P>.05) in Experiment II. This could be partly due to its much smaller size.

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A THESIS

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PURPOSE

The object of this study was to determine certain nutritional characteristics of two species of cotton rats, <u>Sigmodon hispidus</u> and <u>S. fulviventer</u>, and of the South American field mouse, <u>Akodon azarae</u>, and to compare these with comparable nutritional characteristics of the meadow vole, <u>Microtus pennsylvanicus</u>. It is hoped that the null hypothesis, H_o: The three species tested are not different from <u>M. pennsylvanicus</u> or from each other in the characteristics studied, will be disproved.

INTRODUCTION

Nutritionists have long been interested in determining food qualities by measuring growth responses in selected organisms. As bioassay test organisms, rodents have proven highly useful in studies of various kinds. They take up little space and are easy and fairly inexpensive to maintain. The gestation period of rodents is relatively short, the litters are often large, and the young mature quickly. Colonies can be built up rapidly and, if using wild rodents, can be replenished with fresh stock as needed.

Voles of the genus <u>Microtus</u> (<u>M. pennsylvanicus</u>, Elliott, 1963) have proved excellent bioassay test mammals. These rodents are easily bred in captivity and weanlings respond in a highly sensitive manner to food stuffs containing different levels of selected nutrients (Shenk <u>et al.</u>, 1970). Species of <u>Microtus</u> occur naturally in northern parts of North America, Europe and Asia, but are rare and only locally distributed in remote mountain sectors of northern Latin America (Hall & Kelson, 1959).

To test some common Neotropical rodents for suitability as bioassay test animals, cotton rats of the genus <u>Sigmodon</u> and South American field mice of the genus <u>Akodon</u> were studied. Cotton rats are common grassland rodents ranging from central United States southward to Peru (Baker, 1969) and the field mice are common in many parts of South America (Cabrera, 1961). If selected animals representing these genera proved successful in bioassay test trials, these Neotropical groups might prove valuable to Latin American nutritionists desiring to establish breeding colonies of locally adjusted and easily-captured test animals. These animals might also prove useful as laboratory stocks in related biomedical investigations.

Table 1 compares gestation and maturation of the young for the two genera studied here and 4 other commonly used experimental rodents. Although the gestation period for <u>Sigmodon</u> is somewhat longer and the litters smaller than the white rat, it can be weaned and reach breeding age in about half the time of <u>Rattus</u>. <u>Akodon</u> is somewhat comparable to <u>Rattus</u> except sexual maturity is reached at an earlier age and litters are not so large. Akodon is also much smaller than the white rat.

One chief draw-back in using wild rodent species rather than domesticated strains of Mus or Rattus is they are much more excitable and therefore more difficult to handle (Harris & Thimann, 1948). Unlike Microtus, this wildness has not yet been successfully bred out of Sigmodon in captivity.

Protein Evaluation Tests:

The three most commonly used protein evaluation tests according to Sheffner (1967) are as follows:

Reproductive and Developmental Characteristics of Some Commonly Used Experimental Rodents. Table 1.

PEFERENCE	Manual for Laboratory Animal Care, 1961	Manual for Laboratory Animal Care, 1961	Manual for Laboratory Animal Care, 1961	Elliott, 1963 Burt, 1969	Meyer & Meyer, 1944; Goertz, 1965; Svihla, 1929; Jimenez, 1969	Feter Dalby, personal communication
AGE AT SEXUAL MATURITY	100 days	35-50 days	3-5 mos.	25 days 9° 45 days o	40 - 50 days	8 wks.
AŒ AT WEANING (in days)	21	16-21	10	41	<i>5</i> -3 0*	14-21
AVERAŒ NUMBER IN LITTER	8-12	10-12	3.5-4	5.6	3-6	<i>۲•</i> 4
GESTATION (in days)	27	19-20	63	21	27	20-25
GENUS	Rattus	Mus	Cavia	Microtus	Sigmodon	Akodon

*Young are able to leave nest at μ -7 days (Odum, 1955), can be easily weaned in the laboratory at 10 days, but seem to gain weight most readily later in life when weaned at 30 days (Meyer & Meyer, 1944).

- 1. Protein Efficiency Ratio (PER) is used mainly in feeding experiments with small animals and measured by gain in weight/protein intake. However, Block & Mitchell (1946) do not believe that this is a true efficiency ratio because not all of the protein is used for growth, but only that consumed above maintenance.
- 2. Biological Value (BV) is determined by the nitrogen balance and defined as nitrogen retained/nitrogen absorbed. It measures the percent absorbed nitrogen retained for growth and maintenance but does not include the correction needed for maintenance to achieve the maximum efficiency of utilization.
- 3. Net Protein Utilization (NPU) expresses both digestibility and BV. It is expressed by the coefficient of digestibility X BV and represents the proportion of nitrogen retained.

It was decided to use the PER method for several reasons: (1) It is the simplest and most convenient method of measuring nutritive value of proteins; (2) PERs correlate highly with NPUs (Bender, 1956); and (3) this was the method used by Shenk, Elliott, & Thomas (1970) in their Microtus studies, with which this investigation is compared.

The efficiency with which protein is utilized is diminished if the caloric intake is too low or if the protein is fed in excess. It is usual to employ diets which provide more than the quantity of protein needed for maintenance but less than that required for optimum performance (Woodham, 1968). This accentuates the differences between proteins thus making it easier to discriminate between samples.

EXPERIMENTAL

Test Mammals Used:

Rodents used in this study were descendants of individuals livetrapped on MSU Museum field trips and maintained in laboratory colonies.

Sigmodon hispidus Say and Ord. -- The wide-ranging hispid cotton rat is distributed in grassy and brushy habitats from Nebraska to Peru. Although it is adapted to a variety of hot, cool, moist and dry climates at both high and low elevations, one sample was selected for study from 13 km ENE of Pichucalco, elevation 61 m, in Chiapas, México (18° N latitude). Adults weigh, on the average, 110-115 grams. The parental stock was captured in July 1969, in disturbed grassy areas between mixed jungle and sugar cane fields. The annual rainfall at this locality is heavy, approximately 3,500 mm.

Sigmodon fulviventer Allen. -- The tawny-bellied cotton rat inhabits the mesquite-grassland on the east side of the western Cordillera from southern New Mexico and Arizona south to the Mexican state of Michoacán. Adults weigh, on the average, 175-200 grams. The parental stock was captured in July 1968, 2 km N of Gallego, elevation 1366 m, in Chihuahua, México (30° N latitude), on a shortgrass plain with a low annual rainfall, approximately 150 mm.

Akodon azarae (Fischer). -- Azare's field mouse lives in grassy habitats in southern South America (parts of Bolivia, Brazil, Uruguay, and Argentina). Adults weigh, on the average, 19-21 grams. Parental stock came from INTA, 20 km W of Balcarce, elevation 100 m, Province of Buenos Aires, Argentina, and was captured in June 1970. This grassland locality receives approximately 860 mm of rainfall annually.

The test animals included, therefore, samples of Neotropical grass-land rodents from a semi-arid temperate habitat (Sigmodon fulviventer), from a moderately-humid temperate habitat (Akodon azarae), and from a humid, hot habitat (Sigmodon hispidus).

Methods:

Two separate experiments were run: (1) To determine what level of casein is most efficiently utilized by the test animals; and (2) to determine whether the test animals utilize Triticale, a wheat-rye cross, as a protein source comparable to Microtus pennsylvanicus (Elliott, 1963; Shenk, Elliott, & Thomas, 1970). Conditions for each of the experiments were similar with only the diets differing. Several sets of weanling siblings were subjected to each of three diets in both experiments conducted.

The diets in each experiment were isocaloric, with casein (Hammerstein quality) being the chief variable and the amount of carbohydrate, consisting of 2 parts cornstarch: 1 part dextrin: 1 part sucrose, changing to compensate for the difference in calories. Two experimental strains of triticale, MSU #358 (15.94% protein) and PC70 (15.25% protein), were then compared to the casein. Mazola corn oil, Vitamin Diet Fortification Mixture, Salt Mix W, and Alphacel were then added in specified quantities to supplement the diets.

Each diet was mechanically mixed and then pressed into wafers to fit into spillproof feeders (Shenk & Elliott, 1969). These wafers were dried in an oven at approximately 65.5° C for 8-12 hours. They were then left in the experimental room for 48-72 hours in order for the moisture content to equilibrate relative to that of the air.

Animals in post-weaming stages gain the greatest amount of weight in the shortest amount of time, so their sensitivity to small dietary differences would be greatest. Also, in short term assays with weamlings emphasizing growth very little nutrient material is laid down as fat reserves.

Test animals of the genus <u>Sigmodon</u> were weaned at 10-12 days, then housed with siblings and fed Purina Mouse Breeder Chow for 2-3 days, until they weighed at least 15 gms. After this short acclimation period, they were fed the experimental diets for a test period of 10 days.

Occasionally older rats were used, but protein was utilized most efficiently by the younger rats. Test animals of the genus <u>Akodon</u> were weaned at 12-14 days, fed Purina Mouse Breeder Chow for 2-3 days until they reached a weight of at least 6-7 gms., and then fed the experimental diets for 6-7 days.

Whitmoyer (1956) found there were significant differences in growth rates of Microtus pennsylvanicus attributable to sex during the first 4 weeks. Although there is a slight difference in growth rates of male and female Sigmodon hispidus, Jimenez (1969) found the difference was not highly significant in young animals, and there is no significant difference between sexes of S. fulviventer. Peter Dalby (personal communication) reports there are no significant differences in growth rates between sexes in Akodon. Therefore, both males and females were used in this study.

During each test, animals were individually housed in small disposable plastic cages with metal tops. Ground corn cob was used for bedding, a piece of cotton was used for nest material. Water and food were offered ad libitum. The tests were run in a room that housed a

colony of meadow voles (<u>Microtus</u>) with a relatively constant temperature of 15.6° C, 75-85% humidity, and with 24 hours of light per day.

Weights of the animals and of the food were determined at the beginning and at the end of the test periods. Per cent of dry weight of the food was determined by standard oven drying at 100° C for 3 hours.

Protein Efficiency Ratios (PER) were determined by using the formula:

PER = weight gained by the animal amount of dry protein consumed

Experiment I: Determination of most efficiently utilized level of casein.

Siblings from each of 8 litters of weanling <u>Sigmodon hispidus</u>, from each of 11 litters of <u>S. fulviventer</u>, and from each of 9 litters of <u>Akodon azarae</u> were fed isocaloric diets containing 7% (Diet 1), 14% (Diet 2), or 21% (Diet 3) casein, as shown in Table 2.

Experiment II: Comparison of qualities of 3 proteins.

From Experiment I it was determined that test animals of all three species utilized protein best at the 7% level, so all diets in Experiment II contained 7% protein. Using the same techniques as above, siblings from each of 16 litters of S. hispidus, from each of 15 litters of S. fulviventer, and from each of 16 litters of A. azarae were fed isocaloric diets that contained either casein (Diet 1), Triticale-MSU #358 (Diet 2), or Triticale-Rockefeller PC70 (Diet 3) as shown in Table 3. The PERs calculated from this test were then compared with findings of known PERs of Microtus pennsylvanicus and other animals.

Ingredients of Diets Used In Experiment I (Grams/100 Grams of Feed) Table 2.

DIET 3	21%	56%	3%	2%	3%	15%	320
DIET 2	ጀተፒ	63%	3%	2%	3%	15%	320
DIET 1	%2	%02	38	%7	%E	<i>¥</i> 51	320
	CASEIN (HAMMERSTEIN QUALITY)	CA RBOHYDRA TE*	CORN OIL	VITAMIN FORTIFICATION MIX - NBC	SALT MIX W-NBC	ALPHA-ŒLLULOSE	TOTAL CALORIES

*Carbohydrate consists of 2 parts corn starch: 1 part dextrin: 1 part sucrose.

Table 3. Ingredients of Diets Used In Experiment II (Grams/100 Grams of Feed)

	ן אַדַּתַ	DIET 2	DIET 3
PROFETIV	7% CASETN	MSU #358	ROCKEFELLER PC70
NOTE TO			
CA RBOHYDRA TE	70%	30%	, 29%
COEN OIL	3%	-	
VITAMIN FORTIFICATION MIX - NBC	%7	2%	2%
SALT MIX W - NBC	3%	3%	3%
ALPHA_CELLULOSE	15%	20%	. 20%
TOTAL CALORIES	320	320	320

RESULTS

Table 4 shows the average weight gain, amount of dry protein (casein at levels of 7%, 14% and 21%) consumed, PER, and sample size (n) for each of the three samples of the species studied, Sigmodon fulviventer, S. hispidus, and Akodon azarae, and also includes values for Microtus pennsylvanicus obtained by Dr. Fred Elliott and his associates at Michigan State University in the last few years. Standard errors for each value are also included. These results are compared in Figure 1.

Table 5 shows similar values found for each of the species in Experiment II and the results are graphed in Figure 2.

Weight Gain, Dry Protein Consumption, and PER for μ Species of Rodents at Different Levels of Casein. Table 4.

	Weight Gain*	Dry Protein Consumption	Per	ជ
7% CASEIN:				
S. fulviventer	8.98 ± 1.86**	3.61 ± .41	2.61 ± .31	6
S. hispidus	8.11 ± 3.04	2.18 ± .05	3.07 ± .31	9
A. azarae	2,30 ± .12	1.07 ± .01	2.19 # .31	6
M. pennsylvanicus		tte dag maj pag Cas and	2.84 ± .18	5
14% CASEIN:				
S. fulviventer	10.14 ± 4.72	6.25 ± .31	1.57 ‡ .26	8
S. hispidus	13.57 ± .20	6.19 ± .13	2.27 ± .09	7
A. azarae	3.18 ± .07	5.26 ± .09	1.51 ± .15	8
M. pennsylvanicus	20 00 00 00 00 00 00 00 00 00 00 00 00 0		1.75 ± .16	5
21% CASEIN:				
S. fulviventer	10.08 ± 5.57	8.92 ± .99	1.06 ± .24	6
S. hispidus	10.51 ± 5.39	9.25 ± .22	1.15 ± .22	7
A. azarae	3.29 ± .18	3.30 ± .12	1.06 ± .15	7
M. pennsylvanicus				
*per 10 days for Sigmodon,	per 6 days for Akodon	** Standard error	rd error	

Figure 1. Average Protein Efficiency Ratio (PER) of 4 Species of Rodents at Different Levels of Casein.

Sigmodon fulviventer
 Sigmodon hispidus
 Akodon azare
 Microtus pennsylvanicus

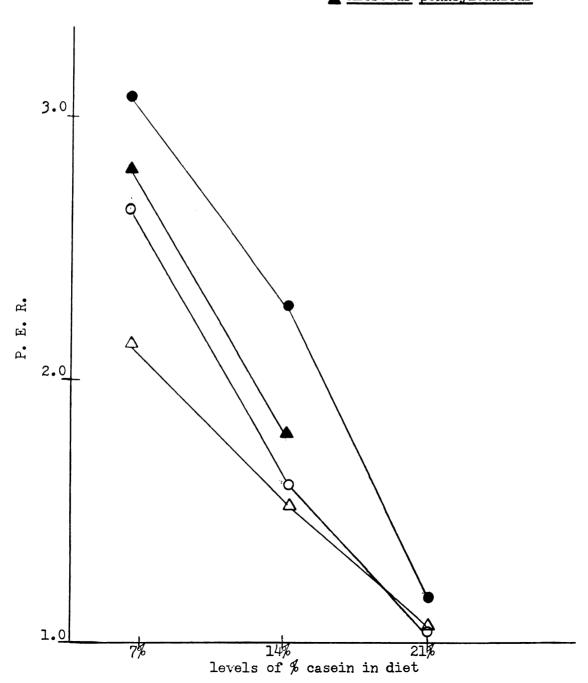
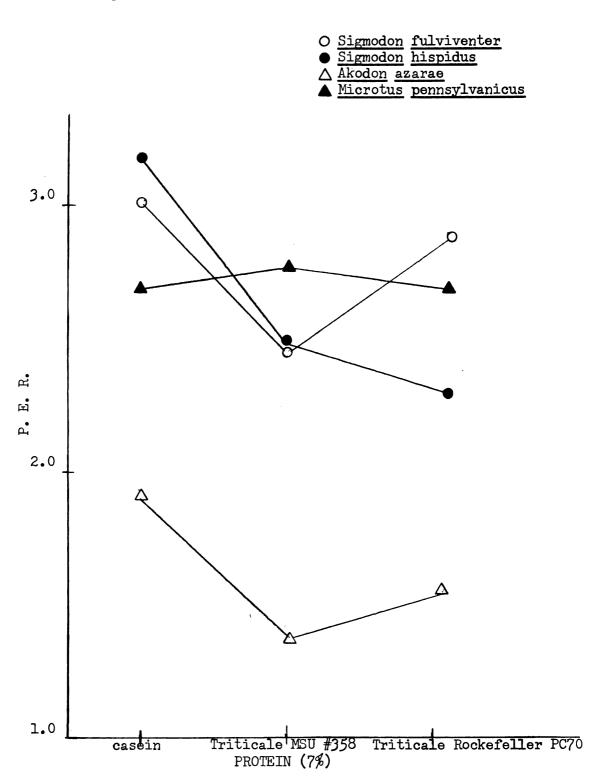


Table 5. Weight Gain, Dry Protein Consumption, and PER for 4 Species of Rodents Using Different Proteins.

	Weight Gain*	Dry Protein Consumption	Per	ជ
7% CASEIN				
S. fulviventer	8.28 ± .30**	2.82 ± .02	2.99 ± .21	15
S. hispidus	10.43 ± 1.40	3.27 ± .03	3.16 ± .22	13
A. azarae	2.07 ± .03	1.09 ± .002	1.90 ± 016	15
M. pennsylvanicus			2.72 ± .11	5
TRITICALE, MSU #358:				
S. fulviventer	68. ± 61.6	3.86 ± .24	2.46 ± .15	13
S. hispidus	9.10 ± .85	3.43 ± .04	2.51 ± .15	13
A. azarae	1.67 ± .03	†0• ∓ 61•I	1.34 ± .14	14
M. pennsylvanicus		-	25.74 ± .92	3
TRITICALE, ROCKEFELLER PC?	70:			
S. fullyiventer	8.99 ± 1.34	3.25 ± .10	2.81 ± .47	11
S. hispidus	7.41 ±97	3.10 ± .04	2.31 ± .22	14
A. azarae	1.66 ± .03	1.18 ± .003	1.46 ± .17	14
M. pennsylvanious			2.64 ± .61	7
*per 10 days for Sigmodon,	per 6 days for Akodon	** Standard	d error	

Figure 2. Average Protein Efficiency Ratio (PER) of 4 Species of Rodents Using Different Proteins.



ANALYSIS OF DATA AND DISCUSSION

As the amount of protein in the diet is increased, both the weight gain and the amount of dry protein consumed increase. However, as can be seen in Table 3, the amount of protein consumed increases at a faster rate than the weight gain, which accounts for the decreased efficiency of utilization.

Table 3 and Figure 1 show that the most efficient use of casein occurs at the 7% level for all 4 species of rodents. As the amount of casein in the diet increases, the efficiency of utilization decreases. This agrees with reports that there is a significant linear trend toward reduced efficiency of utilization of digestible protein with increasing levels of dietary protein (Greeley et al., 1964). When intake exceeds the requirement, efficiency of utilization must decrease rapidly since protein cannot be stored in the body to any appreciable extent (Hegsted & Chang, 1965).

Shenk et al. (1970) report that maximum casein efficiency values of 2.5 were obtained for M. pennsylvanicus between 5% and 8% casein in the region of 8% to 23% fiber. Efficiency decreased below this point. Therefore it was decided to not test <u>Sigmodon</u> and <u>Akodon</u> on diets containing less than 7% casein at this time.

The PERs of the experimental species correlate quite well with those of <u>Microtus</u> at the 7% and 14% levels. <u>Microtus</u> was not directly tested at the 21% level, but the PERs of the other three species are quite similar. Using the Student's t-test it was found that there is no difference (P < .05) between any of the species except <u>S. hispidus</u> and <u>Akodon</u>, the highest and lowest values respectively, at 7% and 14% casein levels.

There are no significant differences between PERs of Akodon at the different levels of protein, except between 7% and 21% casein, as was the case in Microtus. It is interesting to note, however, that there is a significant difference between the PERs of S. fulviventer at 7% and 14% levels, but not at 14% and 21% levels, whereas there is no difference between 7% and 14% levels of S. hispidus but there is between 14% and 21% levels. This would seem to indicate that S. fulviventer is more sensitive to lower levels of protein. A larger sample size should be used to confirm this point. However, if indeed this is true, then, even though the PERs for S. hispidus are somewhat higher and there is no significant difference among these two species and Microtus, it would appear that S. fulviventer might be more similar to Microtus, and S. hispidus might actually be slightly more comparable to the albino rat. The latter has been found to utilize 10% casein most efficiently (Chapman et al., 1959). It would be most interesting to conduct a study directly comparing the white rat with these two species of cotton rats. Sigmodon hispidus should also be tested at intermediate levels (between 7% and 14%) of casein to find more accurately which level is most efficiently utilized.

Since the 7% level of casein was found to be most efficiently utilized, this diet was used in all feeding trials in Experiment II.

Using a two-way analysis of variance it was found that there was no difference within each species in the qualities of the three proteins, casein, Triticale-MSU #358, and Triticale-Rockefeller PC70. There was, however, a difference among species. Using Student-Neumann-Keuls test for Least Significant Range (LSR) (Sokal & Rohlf, 1969) it was found that Akodon was significantly different (P>.05) from both species of cotton

rats in all 3 cases and was different from Microtus in all but the 7% casein diet.

Several other animals besides those mentioned thus far have been used for protein quality studies. Some of the most useful animals have been chicks (Woodham, 1968; Miller & Kifer, 1970; Ousterhout & Snyder, 1962; Elamberg, 1971), swine (Greeley et al., 1964; Pond et al., 1966), and chinook salmon (Hastings, 1969). Protein Efficiency Ratios have been determined for these and other animals, but quite often different proteins are used. Procedures vary according to the species and the researcher. Table 6 gives PER values for several animals with casein as the test protein at specified percentages of protein and test periods of particular lengths.

Both species of <u>Sigmodon</u> fit in well with these PER values. <u>Akodon</u> is somewhat low, but no lower than one PER value that has been stated for the albino rat (Block & Mitchell, 1946).

The white rat is chiefly a seed-eater as opposed to Microtus and Sigmodon (Baker, 1971) which seem to be mostly grass-eaters. Thus Rattus feeds on high-caloric but hard-to-get food whereas the other 2 species feed on low-caloric but easily obtained food (Vorontsov, 1960). It would seem reasonable, then, to suppose that grass-eaters should be able to utilize efficiently a lower percentage of protein than seed-eaters since there is a higher percentage of protein in seeds. Unfortunately, close comparisons of Rattus with Sigmodon and Microtus with respect to protein utilization have not yet been made.

The ubiquitous \underline{S} . <u>hispidus</u> appears to be more catholic in its feeding habits than the restricted \underline{S} . <u>fulviventer</u>. This might help explain why the former is not quite as sensitive to lower protein diets as the latter.

Table 6. PERs for Frequently Used Bioassay Test Animals.

Test Animal	% Protein	Test Period	PER	Reference
Chinook salmon	26.5%*		2.38	Hastings, 1969
Young swine	18%	2 wks.	4.8	Barnes et al., 1966
Albino rats	10%	4-8 wks.	2.5	Block & Mitchell, 1946 Chapman et al., 1959 Barnes et al., 1966
Albino mice	8 . 5%	7-8 days	2.5	Bosshardt et al., 1946
Microtus pennsylvanicus	7%	6-10 days	2.5	Shenk, Elliott & Thomas, 1970
Sigmodon hispidus	7%	10 days	3.1	
S. fulviventer	7%	10 days	2.8	
Akodon azare	7%	6 days	2.0	

^{*}Minimum protein level dependent on water temperature (Mertz, 1969).

Sigmodon hispidus should be better adapted to utilizing protein at a wider range of concentrations. Sigmodon fulviventer, on the other hand, occurs in semi-arid grassland plains where protein levels in forage are generally constant and concentrated.

This difference found between the two species of cotton rats might also depend on the difference in their genetic make-ups. The difference in size alone could have some bearing on the efficiency of utilization of the protein. However, there could also be sufficient genetic difference between subspecies of either of the two species to cause a different degree of efficiency. Drickamer & Bernstein (1972) found that there is indeed a significant inverse relationship between latitude and weight gain of two subspecies of Peromyscus maniculatus during the first two weeks of life. Sigmodon fulviventer shows a decline in overall size from north to south (Jimenez, 1969), but there might be some relation to size, rate of development, and protein utilization.

It should be noted that these samples of cotton rats do not necessarily represent the whole species with respect to growth, litter size, and other characteristics. It would be necessary to test several different subspecies from each species (especially Sigmodon hispidus) from different areas to determine what the variations would be. This information would be useful to both nutritionists and zoologists.

It appears, therefore, that <u>Sigmodon</u> would make a suitable animal for bioassay of food stuffs. It behaves similar to <u>Microtus</u> pennsylvanicus in efficiency of utilization of protein.

Akodon seems to respond somewhat differently, indicating that it does not utilize the protein as well as the other species. This could be due to the fact that it is a much smaller rodent, may have a higher

rate of metabolism, needs more energy to maintain itself, and thus must eat more food in order to gain a comparable amount of weight. Peter Dalby (personal communication) reports that Akodon has a low geometric growth rate for the first few weeks of life compared to the Microtines, which could be another reason why different results were found with this rodent. Also, these animals were quite wild and were only a few generations removed from the wild. A few generations more in the lab might have made a much more manageable group to work with and more typical results might have been obtained. Even though Akodon does have somewhat lower PER values than either Sigmodon or Microtus, it responds in a similar manner, and once a base PER is known for this species, it might also make a suitable bioassay animal.

SUMMARY

The weanlings of three Neotropical rodents were evaluated to determine their suitability as bioassay test mammals for nutrition studies. The findings were compared with those for Microtus pennsylvanicus, which occurs in northern parts of North America. The three species chosen were taken from strikingly different habitats:

(1) A semi-arid temperate habitat (Sigmodon fulviventer); (2) a moderately-humid temperate habitat (Akodon azarae); and (3) a humid, hot habitat (Sigmodon hispidus).

Two experiments were run, using the same methods as Shenk, Elliott, & Thomas (1970). Experiment I determined which level of protein (casein) was most efficiently utilized by weanlings for growth. Three siblings from each of several litters of each species were fed three different isocaloric diets with various percentages of casein (7%, 14%, and 21%) with only the amount of carbohydrate changing to make up the difference in calories. Experiment II tested the qualities of Triticale-MSU #358, and Triticale PC70 (wheat-rye hybrids) and 7% casein by comparing Protein Efficiency Ratios (PER).

Very little difference in PERs was found between casein and the Triticale lines. However, PER values for <u>Akodon</u> were significantly lower than those for the other three species. Possibly because of its small size it has a higher rate of metabolism and needs more protein for a comparable weight gain.

It appears that, in comparison with <u>Microtus pennsylvanicus</u>, <u>Sigmodon</u> would make a suitable bioassay test organism for nutrition studies, and although <u>Akodon</u> is somewhat lower, it also follows the same pattern and with the base PER known, it might also make a useful bioassay mammal.

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