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## Feed Preference Tests of Carbohydrate, Protein, and Rodenticide to Tree Rat, House Rat, and Rice Field Rat

Guruh Satria Wibowo<sup>1</sup>, Swastiko Priyambodo<sup>2\*</sup>

<sup>1,2</sup> Department of Plant Protection, Faculty of Agriculture, IPB University

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**ABSTRACT:** Rats are wild animals that coexistence with humans. The losses caused by these rats are damage to house building, agriculture, and plantation. Knowledge of the feed as bait and lure is very important in rat control, particularly with rodenticides and traps. The aim of this research was to find out feed preference to three species of rats and effect of feed availability toward rodenticide consumption. Tree rat (*Rattus tiomanicus*), house rat (*R. tanezumi*), and rice field rat (*R. argentiventer*) were tested with four stages i.e. (a) type of carbohydrate sources, (b) type of protein sources, (c) combination of carbohydrate and protein, and (d) carbohydrate, protein, and rodenticide. The method used was choice test. Carbohydrate tested were corn, rice, unhulled rice (grain), and oat. Protein were cricket, frog, fish, and mouse. Rodenticide (brodifacoum 0.005%) was tested to the rat. Each feed and rodenticide are weighed before and after testing to analyze the preference of rats. The data analyzed using *SAS program for windows* version 9.0, a further test with Duncan multiple range test with  $\alpha = 5\%$  and  $1\%$ . Unhulled rice (grain) and cricket were the kind of feed that most preferred or highest palatable by test animals. The existence of feed causes the lower consumption of brodifacoum rodenticide by three species of rats.

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**Corresponding Author:**  
**Swastiko Priyambodo**

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### INTRODUCTION

Rats are wild animals that live side by side with humans. The existence of mice around other living creatures can be a relationship of parasitism or mutualism (Meehan 1984). The relationship between rat parasitism and humans is demonstrated by several losses, such as rats destroying crops, animal feed, human foodstuffs and household goods, as well as the role of rats as carriers of disease for humans and livestock (Dickman 1988). In rice cultivation, rats are one of the most important pests, as stated by the Center for Forecasting Plant Pest Organisms (*Balai Besar Peramalan Organisme Pengganggu Tanaman* 2014) that during 2014 the pests that attacked rice fields in Karawang Regency were rice field rats 6,635 Ha, rice stem borers 2,958 Ha, brown plant hoppers 1,184 Ha, bacterial leaf blight 1,168 Ha, and grassy stunt virus 43 Ha.

Sudarmaji and Herawati (2009) stated that damage caused by rice field rats (*R. argentiventer*) in Asia reaches 10-15% per year. If calculated, a loss of just 5% is equivalent to 30 million tons of rice and is enough to feed 180 million people for 12 months. Apart from food crops, rats also cause a lot of damage to plantation crops such as oil palm and sugar cane. Losses due to rats in oil palm plants can reach 5% of the total crude palm oil (CPO)/Ha/year in mature plants and can reach 80% in young plants (Adidharma 2009). Losses due to rat attacks on sugar cane commodities reached more than 1,000 hectares with a yield loss of more than IDR 300 million (Ditjenbun 2013).

There are several methods of controlling rats, namely sanitation, technical culture, physical-mechanical, biological, and chemical. Sanitation is carried out by cleaning places where rats hide. Technical culture is carried out by setting plant distances such as rows (*jajar legowo*) and simultaneous planting. The physical-mechanical method is by using barriers (proofing) and traps to prevent rats from entering a location, for example the trap barrier system (TBS). The biological method is to use natural enemies or rodent predators such as owls, and finally the chemical method is to use rodenticides and fumigant (*emposan*) (BB Padi 2015).

There are several types of rodenticides used to control rats. Based on how they work, rodenticides are divided into two, namely acute and chronic poisons. However, currently chronic poison is used more often than acute poison, this is due to the fast-acting nature of acute poison, i.e. less than 24 hours, so it can raise suspicion and deter rats (bait shyness). One type of chronic rodenticide active ingredient that has the most potential for controlling rats is brodifacoum. According to Buckle and Smith (1984)

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brodifacoum at a concentration of 0.005% can cause 100% death of mice and rats three days after treatment, both susceptible and resistant to warfarin.

Knowledge of the biology and ecology of rats determines the success of controlling them, especially for control with traps and rodenticides, namely knowledge of the type of food preferred and when this food is needed. These two controls really depend on the type of feed used. It is hoped that the appropriate type of food will attract and make rats consume more. Rats really need carbohydrate and protein food for their daily activities. According to Robeson *et al.* (1981) rats in America require feed of 15 g/rat/day during growth or adult rats during maintenance, 15-20 g/rat/day during pregnancy, and 30-40 g/rat/day during lactation. Carbohydrate feed can be obtained from food plants such as cereals and tubers. One type of plant source of carbohydrate food for rats is rice. Rats can attack rice at various growth stages. At the seedling stage, rats damage rice plants by pulling out seeds that have started to grow (seedlings) to eat the remaining parts of the seeds (endosperm). In the vegetative stage, rats cut the base of the stem to eat the stem. In the generative stage, rats can attack the panicles or grains of rice crops (Priyambodo 2009). Most rats source protein in nature from consuming insects, worms, eggs, dead animals, frogs, fishes, reptiles and birds (Debbi 2015).

### Research Purposes and Benefits of Research

This research aims to determine the consumption levels of tree rat, house rat, and rice field rat for several types of carbohydrate and protein feed. Apart from that, to see the interest of rats in rodenticides when carbohydrate and protein feed is available. It is hoped that the results of the research will provide information regarding the type of feed preferred by tree rat, house rat, and rice field rat. Thus, it can be taken into consideration in efforts to control rats, especially control with traps and rodenticides.

## MATERIALS AND METHODS

### Place of Research

The research was carried out at the Vertebrate Pest Laboratory, Department of Plant Protection, Faculty of Agriculture, IPB University (Bogor Agricultural University).

### Materials and Tools

The materials used in this research were tree rat (*Rattus tiomanicus*), house/roof rat (*R. tanezumi*), rice field rat (*R. argentiventer*), rice, un-hulled rice (grain), wheat (oat), corn, fish, cricket, frog, mouse, rodenticide containing the brodifacoum active ingredient 0.005%, and chloroform. The tools used were a single rat cage measuring 50 cm x 40 cm x 20 cm, feed container, bamboo roof, glass, spoon, tweezers, and electronic scales (Figure 1 - 4).

The scales used to calculate the weight of rats and feed in testing are electronic scales (electronic top-loading for animals). This scale is used to obtain the weight of the rats before and after testing, as well as the feed before and after being consumed by the test animals.



Figure 1. Electronic Scales



Figure 2. Bamboo Roof

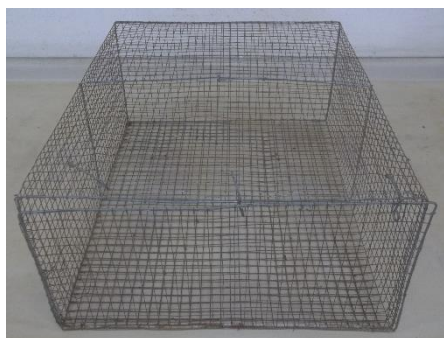


Figure 3. Rodent Individual Cage



Figure 4. Feed Container

## Research Methods

### Preparation of Test Animals

The test animals used were tree rats, house rats, and rice field rats. Tree rats and house rats were obtained from captures in Dramaga District, Bogor Regency. The rice field rats were obtained from catching them at rice plantation, in Subang Regency. The rats used were adult rats, healthy, without defects, and weighing more than 70 g. The number of each species of rats used in each test was 10 heads.

### Feed Preparation and Rodenticide

The feed used is divided into two types, namely carbohydrate and protein feed. Carbohydrate feed consists of rice, un-hulled rice (grain), wheat (oat), and corn (Figure 5). Protein feed consists of fish, cricket, frog, and mouse (Figure 6). The rodenticide used is brodifacum 0.005% in block form with blue colour, which is a ready-to-use rodenticide (Figure 7).

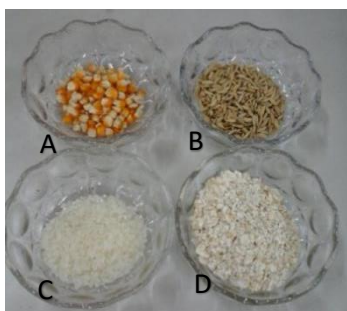


Figure 5. Carbohydrate feed: Corn (A), un-hulled rice (B), rice (C), oat (D)

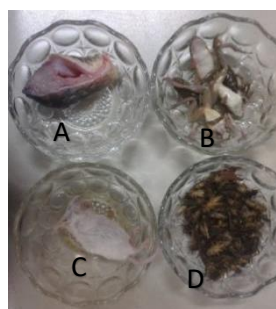


Figure 6. Protein feed: Fish (A), frog (B), mouse (C), cricket (D)

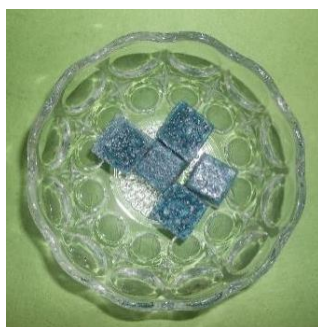


Figure 7. Rodenticide brodifacoum 0.005%

### Test Implementation

The test was carried out using a choice test, namely by giving several food choices to the rats. Each feed and rodenticide is given in 20 g containers or in abundant quantities (*ad libitum*). The container used has the same shape and size for each replication. Tests were carried out for seven consecutive days and the position of the feed in the cage was changed every day. After completing one test the rats were rested for three days and at the end of the final treatment the rats were rested for six days. Observations were made every day on the amount of feed consumed by reducing the initial weight and final weight feed given, including the feed that was scattered at the bottom of the cage.

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## Carbohydrate Feed Preference Test

The test was carried out by giving four food choices to rats. The food used is rice, un-hulled rice (grain), wheat (oats) and corn.

## Protein Feed Preference Test

The feed used is fishes, crickets, frogs, and mice. Feed is given to rats in a dead condition. Crickets, frogs, and mice were killed using chloroform. Then leave it for three to four hours to remove the smell from the chloroform.

## Carbohydrate and Protein Feed Preference Test

Testing was carried out by testing the two best feeds from each carbohydrate and protein test. The two best feeds are the two types most consumed by rats in each carbo-hydrate and protein test.

## Carbohydrate, Protein, and Rodenticide Preference Test

This test was carried out by providing the best feed from carbohydrates, protein, and rodenticide with the brodifacoum active ingredient 0.005%.

## Observations After the Last Treatment

Observations were made by observing the effect of giving rodenticides to rats. Observations were carried out for six consecutive days with un-hulled rice (grain) as feed.

## Feed Conversion

All data obtained from testing was converted to 100 g of rat body weight, with the following formula:

$$\text{Feed consumption (g/100 g body weight)} = \frac{\text{Average feed consumed (g)} \times 100}{\text{Average rat weight (g)}}$$

## Observed Variables

The variables observed in this test were the amount of feed and rodenticide consumed, the rat's preference for feed, the rat's weight before and after treatment.

## Data Analysis

Data analysis was carried out using a completely randomized design with 10 replications for each rat species. Data were processed using Microsoft Office Excel 2013 software. Statistical analysis used the SAS for Windows version 9.0 program, further testing using the Duncan multiple range test with  $\alpha=5\%$  and  $1\%$ .

## How to Calculate Lethal Dose (LD)

The lethal dose of rodenticide for rats is obtained from the following formula:

$$\text{LD (ppm)} = \frac{\text{Total poison consumption (mg)}}{\text{Average body weight of rats (kg)}} \times 100$$

## RESULT AND DISCUSSION

### Carbohydrate Feed Preference Test

The test results show that the highest consumption of tree rats is un-hulled rice (grain), followed by corn, rice, and wheat (Table 1).

**Table 1. Average consumption (g) of three species of rats on carbohydrate feed treatment**

Type of feed	Species of rat as test animal		
	<i>R. tiomanicus</i>	<i>R. tanezumi</i>	<i>R. argentiventer</i>
Rice	0.562 bB	1.938 aAB	0.473 bB
Unhulled rice	5.135 aA	3.328 aA	7.023 aA
Oat	0.226 bB	2.078 aAB	0.003 bB
Corn	0.669 bB	0.139 bB	0.038 bB
Total	6.592	7.483	7.537

Note: Numbers in the same column followed by the same letter show that they are not significantly different based on Duncan's multiple range test at the level  $\alpha=5\%$  (lowercase letters) and  $\alpha=1\%$  (capital letters)

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According to Supatmi (2009), the factors that influence rice are less preferred than unhulled rice (grain) and corn, namely that rice is often found in housing, while in their natural habitat tree rats often consume grain and corn. In carbohydrate testing on house rats, it was seen that the highest consumption was grain. However, based on the results of the Duncan multiple range test, both at  $\alpha=5\%$  and  $1\%$ , consumption of grain by house rats was not significantly different from that of rice and wheat. This is because house rats are used to find and consuming these three types of food in their natural habitat.

The highest consumption of field rats showed results that were no different from tree rats and house rats, namely grain. Rice field rats' consumption of un-hulled rice (grain) is significantly different from that of rice, oat, and corn. The low consumption of rice and oat by rice field rats can be caused by the nature of rats which are easily suspicious of all objects, including new types of feed they encounter (Meehan 1984). Low consumption on corn in rice field rats is caused by the nutritional content of corn being less than grain and rice (Suharjo and Kusharto 1998).

The nutritional content of rice and grain is relatively the same, while the factor that causes rice to be less liked by rats is because rice does not have a hard outer layer, so rats tend to consume more grain (Aryata 2006). Another factor that causes grain to be preferred by rats is their chewing behavior to reduce the growth of their incisors, which continue to grow, and the aroma of carbohydrates in grain is more pungent than in rice (Permada 2009). The three types of rats have different consumption variations. House rats have a more diverse diet compared to tree rats and rice field rats. The consumption of tree rats and rice field rats tends to be only on grain. This is because in the natural habitat of these two types of rats, grain is more common than other types of food.

### Protein Feed Preference Test

The test results showed that tree rats, house rats, and rice field rats liked crickets (Table 2). The high consumption of three types of crickets by rats is caused by the highest protein content in crickets, namely  $67.77\%$  (Aziz *et al.* 2012). According to Geischa *et al.* (2015) crickets have a strong aroma like shrimp, which is why rats are attracted to consume crickets. Rats preferred crickets compared to other types of test feed because in the natural habitat of rats, crickets are insects that are often found. When crickets were used as test food, the rats' suspicion or neophobic behaviour towards crickets was low.

**Table 2. Average consumption (g) of three species of rats in the protein feed treatment**

Type of feed	Species of rat as test animal		
	<i>R. tiomanicus</i>	<i>R. tanezumi</i>	<i>R. argentiventer</i>
Fish	0.000 bB	3.895 bB	0.392 bB
Cricket	7.679 aA	7.542 aA	7.434 aA
Frog	0.000 bB	0.768 cC	0.000 bB
Mice	0.000 bB	3.059 bBC	0.000 bB
Total	7.679	15.264	7.826

Note: Numbers in the same column followed by the same letter show that they are not significantly different based on Duncan's multiple range test at the level  $\alpha=5\%$  (lowercase letters) and  $\alpha=1\%$  (capital letters)

The consumption of tree rats and rice field rats for crickets was significantly different from the consumption of fishes, frogs, and mice. In fact, tree rats only consumed crickets from the four types of feed tested. Rats will choose to consume the type of food they like most if there is a lot of it, even if there are other types of food around them (Brooks and Rowe 1979). Apart from consuming crickets, rice field rats also consume small amounts of fish and there's no different from frogs and mice which are not consumed.

Apart from consuming crickets, house rats also consume almost the same amount of fish and mice. The high total consumption of protein food by house rats is due to the fact that this type of food is wet. According to Priyambodo (2009) rats can consume wet food of around  $20\%$  of their body weight. In protein test, house rats have more varied consumption compared to tree rats and rice field rats. The consumption of tree rats and rice field rats tends to be only on crickets, this is due to the high level of suspicion of rats towards fishes, frogs, and mice, so that rats avoid contact with these foods.

### Carbohydrate and Protein Feed Preference Test

Based on previous test results, it is known that tree rats, house rats, and rice field rats prefer grain for carbohydrate food, and crickets for protein food, so these two ingredients were chosen for the carbohydrate vs protein test. All three species of rats consumed more carbohydrate food than protein food (Table 3). According to Ratclub (2006) rats generally need  $75-80\%$  of carbohydrates,  $12-20\%$  of protein, and around  $4-6\%$  of fat.

**Table 3. Average consumption (g) of three species of rats on carbohydrate and protein feed treatments**

Type of food	Species of rat as test animal		
	<i>R. tiomanicus</i>	<i>R. tanezumi</i>	<i>R. argentiventer</i>
Rice	-	-	0.392 (4.30%) cC
Grain	5.209 (73.63%) aA	3.944 (41.78%) aA	6.101 (66.97%) aA
Oat	-	2.101 (22.25%) bB	-
Corn	0.349 (4.93%) cB	-	-
Fish	-	1.304 (13.81%) bB	0.672 (7.38%) cC
Cricket	1.517 (21.44%) bB	2.092 (22.16%) bB	1.945 (21.35%) bB
Total	7.075	9.441	9.110

Note: Numbers in the same column followed by the same letter show that they are not significantly different based on Duncan's multiple range test at the level  $\alpha=5\%$  (lowercase letters) and  $\alpha=1\%$  (capital letters). The sign (-) indicates that the food was not given during the test.

The test results showed that the average consumption of tree rats, house rats, and rice field rats for carbohydrate food reached 5.56 g (78.56%), 6.05 g (64.03%), and 6.49 g (71.27%) of total consumption. Meanwhile, for protein food, tree rats, house rats, and rice field rats only consumed 1.52 g (21.44%), 3.40 g (35.97%), and 2.62 g (28.73%) of the total consumption. The percentage of this consumption shows that tree rats consume more carbohydrates than house rats and rice field rats, then followed by rice field rats. This indicates that tree rats need more carbohydrate nutrition than house rats and rice field rats. For protein, the highest percentage of consumption is house rats, then rice field rats, and tree rats.

There is consistency of rats in consuming food in carbohydrate and protein testing with carbohydrate testing. This shows that the amount of food needed by rats is the same, both when only carbohydrates are available and when protein is available. The opposite results were shown for protein consumption by the three types of rats. In the protein test, the three types of rats consumed more than 7.5 g of crickets, but when carbohydrates were available their consumption decreased to only around 1.5 - 2.1 g. This shows that even though crickets have high protein nutrition and a strong aroma, rats choose to consume carbohydrates rather than protein.

According to Goldberg (1971) rats consume more carbohydrates because carbohydrates are needed for growth. Meanwhile, the protein consumed by rats functions for the development process. Another factor causing the high consumption of carbohydrates by rats is that carbohydrates are an important nutrient that rats need for the maturity of their reproductive organs (Taylor *et al.* 1983).

**Carbohydrate, Protein, and Rodenticide Preference Test**

The results of testing food and rodenticides showed that the three species of rats tested preferred consuming food rather than rodenticides (Table 4). The presence of food that is preferred by the three species of rats causes only a small amount of brodifacoum rodenticide to be consumed. Priyambodo and Nazarreta (2013) stated that the presence of bait causes rats to have a choice in consuming and indirectly prevents rats from consuming poisoned bait.

**Table 4. Average consumption (g) of three species of rats on carbohydrate, protein, and rodenticide treatments**

Type of food and rodenticide	Species of rat as test animal		
	<i>R. tiomanicus</i>	<i>R. tanezumi</i>	<i>R. argentiventer</i>
Un-hulled rice	5.924 aA	5.584 aA	4.929 aA
Cricket	2.517 bB	1.858 bB	2.031 bB
Brodifacoum 0.005%	0.057 cC	0.419 cC	0.429 cB
Total	8.498	7.861	7.389

Note: Numbers in the same column followed by the same letter show that they are not significantly different based on Duncan's multiple range test at the level  $\alpha=5\%$  (lowercase letters) and  $\alpha=1\%$  (capital letters).

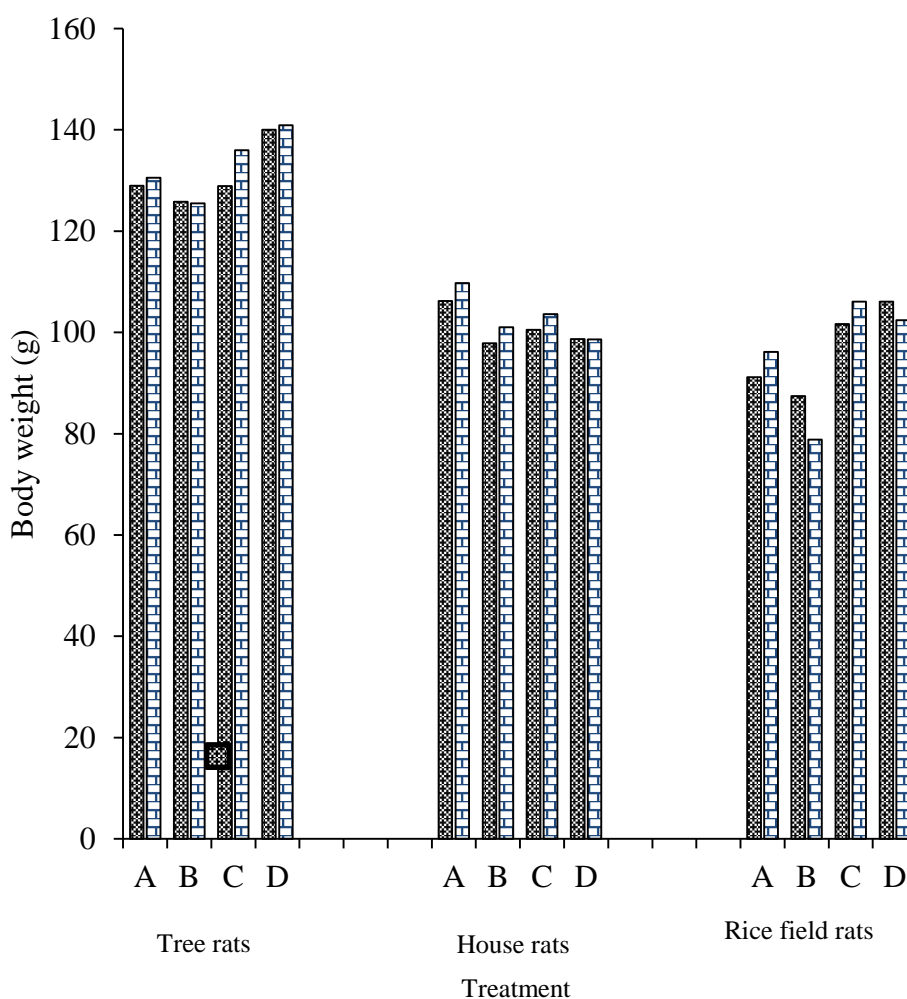
Tree rat consumption of the rodenticide brodifacoum was only 0.057 g (0.675%) among the total consumption. House rats 0.419 g (5.33%) and rice field rats 0.429 g (5.81%). Consumption of the rodenticide brodifacoum shows that tree rats still have high suspicion of the rodenticides tested. This is due to the nature of neophobia in rats, namely the nature of rats that are easily suspicious of objects they have just encountered. Apart from that, rodenticide that contain poison tend to be avoided by rats.

Treatment of house rats and rice field rats showed almost the same consumption of the rodenticide brodifacoum. This is because when controlling house rats and rice field rats, this type of rodenticide is often applied, so that both species of rats are deterred from consuming this rodenticide. Sudarmaji (2005) stated that rice field rats are a major species of agricultural pest and are difficult to control, because these rats are able to learn from control actions that have been carried out previously.

Another factor that causes the low consumption of rodenticides for brodifacoum is that the rodenticide used in this test has a non-pungent aroma (slight odor) (Syngenta 2011), so that when tested with grain and crickets, the three species of rats preferred to consume food with a strong aroma, stronger than rodenticides. Comparison of the consumption of brodifacoum rodenticide in the three species of rats shows different levels of suspicion. The highest suspicion lies in tree rats, which is indicated by the small amount (0.675%) of consumption of rodenticides. The low level of suspicion is shown by house rats and rice field rats.

**Changes in Body Weight of Rats**

Based on the results of weighing the body weight of the rats before and after treatment, it was discovered that there was an increase or decrease in each rat (Figure 8). The carbohydrate feed preference test treatment caused an increase in the three species of rats, the highest increase was shown by rice field rats. This is in accordance with the highest average consumption of rice field rats compared to tree rats and house rats.



**Figure 8.** Initial weight (▨) and final weight (▩) of rats on several treatment. Carbohydrate feed preference test (A), protein feed preference test (B), carbohydrate and protein feed preference test (C), and carbohydrate, protein and rodenticide feed preference test (D).

The protein feed test treatment had different effects on each rat's body weight. Treatment of house rats caused an increase in the rats' body weight. This is due to the highest consumption of house rats compared to the other two species of rats. The opposite effect was shown by tree rats and rice field rats, in both types of rats there was a decrease after protein treatment, the lowest decrease

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occurred in rice field rats. This indicates that protein consumption in rats does not really have an effect on increasing body weight of rats, this is in accordance with Goldberg's (1971) statement that the food that influences the growth process is carbohydrate food.

Testing carbohydrate and protein food preferences showed an increase in body weight in the three species of rats, the highest increase was shown by tree rats. Then testing food and rodenticides showed that there was an increase in body weight in tree rats, this was caused by the very low consumption of rodenticides in tree rats, the opposite was shown by field rats, the highest consumption of rice field rats caused a decrease in body weight in these rats. Supatmi (2009) states that there is a positive correlation between rodenticides vs. bait consumed and a decrease in rat body weight. Fatmawati (2015) also stated that in general, consumption of rodenticides by rats causes a decrease in their body weight.

### Death and Rodenticide Consumption

Rodenticide treatment in carbohydrate, protein and rodenticide tests caused death in several mice (Table 5). According to Fatmawati (2015), death in rats during rodenticide treatment is caused by rats consuming rodenticide at a lethal dose, while rats that remain alive consume rodenticide at a non-lethal dose (sub-lethal dose).

**Table 5. Death and consumption of rats in rodenticides**

Species of rat	Death (head)	Rodenticide consumption (g/100 g)		Lethal dose (ppm)
		7 days	per day	
<i>R. tiomanicus</i>	0	0	0	0
<i>R. tanezumi</i>	3	10.81	1.545	6.756
		11.50	1.643	6.389
		4.28	0.612	2.140
<i>R. argentiventer</i>	3	0.04	0.006	0.022
		2.44	0.406	1.356
		3.63	0.908	2.017

Based on this data, it can be seen that the number of house rats and rice field rats that died was the same, with the average consumption of rodenticides for house rats (8.86 g) being higher than for rice field rats (2.04 g). This shows that the lethal dose for house rats (5.095 ppm) is higher than for rice field rats (1.132 ppm). The factor that causes the number of deaths of house rats to be the same as those of rice field rats, even though the consumption of house rats is higher, is that house rats have a higher resistance to rodenticide than rice field rats. Besides, house rats have better adaptability to the test location (at the laboratory of IPB Univ.) than rice field rats. The rats that died experienced symptoms of bleeding in their anuses, this was due to the way the rodenticide brodifacoum works. Brodifacoum is a chronic poison, namely by inhibiting the coagulation or blood clotting process and breaking down blood capillaries (Priyambodo 2009).

### CONCLUSIONS AND SUGGESTION

Tree rats, house rats, and rice field rats consume more carbohydrates than protein. All three species of rats like unhulled rice (grain) for carbohydrates and crickets for protein. House rats' consumption of protein is far above that of tree rats and rice field rats. The presence of food (unhulled rice and cricket) causes rats' interest in rodenticides to be low. The lowest interest in rodenticides is shown by tree rats, because it had the highest suspicion of rodenticides, while house rats and rice field rats had the lowest suspicion.

It is necessary to test with other types of food, especially for protein testing. It is necessary to test different active ingredients of rodenticide i.e. warfarin, coumatetralyl, and bromadiolone. It is necessary to test feed and rodenticides in the field.

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