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# Protein preference in protein-malnourished monkeys

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PROTEIN PREFERENCE IN PROTEIN-MALNOURISHED MONKEYS

By

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B. A., University of Montana, 1970

Presented in partial fulfillment of the requirements for the degree of

Master of Arts

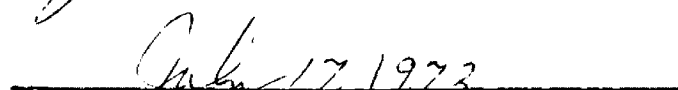
UNIVERSITY OF MONTANA

1972

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## CHAPTER I

### INTRODUCTION

The existence of specific hungers, or of appetites for particular foods, has been postulated for many years. Supporting evidence includes early cafeteria-type studies with pigs (Evvard, 1916), chickens (Pearl & Fairchild, 1921), dairy cattle (Nevins, 1927), human infants (Davis, 1928; Manciaux, Derby & Conoy, 1968), and rats (Richter, Holt & Barelare, 1938). In summary these studies found that animals could choose an adequate diet when presented with an array of foodstuffs. However, this type of experiment has been criticized because the animals were offered only nutritional foods that would probably have provided an adequate diet if sampled at random (Hall, 1961; Cofer & Apley, 1964). Nevertheless, the idea of specific hungers is appealing. Young (1941) for example, argued that animals in the wild were able to find suitable diets long before synthetic diets became available.

Specific hungers for many substances have reportedly been found. Rats were shown to prefer food with large amounts of calcium when their parathyroid glands were removed, and to reduce their sugar intake, thus preventing diabetes, following removal of the pancreas (Morgan & Stellar, 1950). Rats were also shown to prefer salt upon removal of the adrenal glands (Bolles, Sulzbacher & Arant, 1964), and to be able to pick flavored food containing pantothenic acid when they had a deficiency in this substance (Harris, Clay, Hargreaves, & Ward, 1933).

Evidence concerning a specific hunger for protein has been negative. There appeared to be no particular appetite for protein in rats that received diets adequate in protein (Scott & Quint, 1946). A recent study by Hillman & Riopelle (1971) also showed that protein-deprived rhesus monkeys do not evidence a specific



hunger for protein, and that preference does not increase with increasing protein content. Furthermore, these authors reported that lowering the level of protein in the diet decreases the threshold of the palatability of food in general, while raising the acceptability of all foods tested.

The support for the existence of specific hungers in other animals for a variety of food substances made negative results seem surprising and contradictory. The purpose of the present experiment was to ascertain if developing monkeys deprived of normal quantities of protein have a specific hunger for food containing protein, and to determine if these animals would avoid selecting non-food items when food is available, as compared to control animals that were fed an adequate amount of protein.

## CHAPTER II

### METHOD

#### Subjects and Diets

The subjects were 18 laboratory-born rhesus monkeys which were approximately three years of age at the beginning of the experiment. All animals were separated from their mothers at 90 days of age and weaned to a diet containing 25% protein (Casein) by weight. Between 120 and 210 days of age ten monkeys (low-protein group) were transferred to a diet equivalent in calories to the previous diet, but containing 3.5% protein. The remaining eight monkeys were designated as controls (high-protein group) and were retained on the 25% protein diet. Weight gain was minimized in the low-protein monkeys by substituting a 2% protein diet for the 3.5% protein diet between two and three years of age. The 25%, 3.5%, and 2% protein diets were color coded green, white, and red, respectively, with non-nutritive food coloring. Details of diet preparation, feeding procedure, and weight gain are given in Geist, Zimmermann, & Strobel (1972).

#### Apparatus

The apparatus for testing preference (Figure 1) consisted of a 54.6 cm. wide x 58.4 cm. high, .64 cm. plywood board replacing one wall of a steel cage measuring 54.6 cm. wide x 54.6 cm. long x 58.4 cm. high. Mounted on the board were eight bins, 2.5 cm. apart and arranged in two rows separated by 5.1 cm. Each bin consisted of a 10.2 cm. square ledge extending horizontally away from a 10.2 cm. square clear plexiglas window. The window and ledge combination were hinged at the bottom. Reinforcements were placed on the ledges and were visible to the monkey through the window. The monkey made a

response by pulling a knob located at the top of a window causing the reinforcement to drop from the ledge onto the level window where the subject had access to it. A .32 cm. thick opaque masonite partition was interposed between the animal and the preference wall for the "baiting" of the bins, and raised to allow for the presentation of one minute discrete trials.

### Procedure

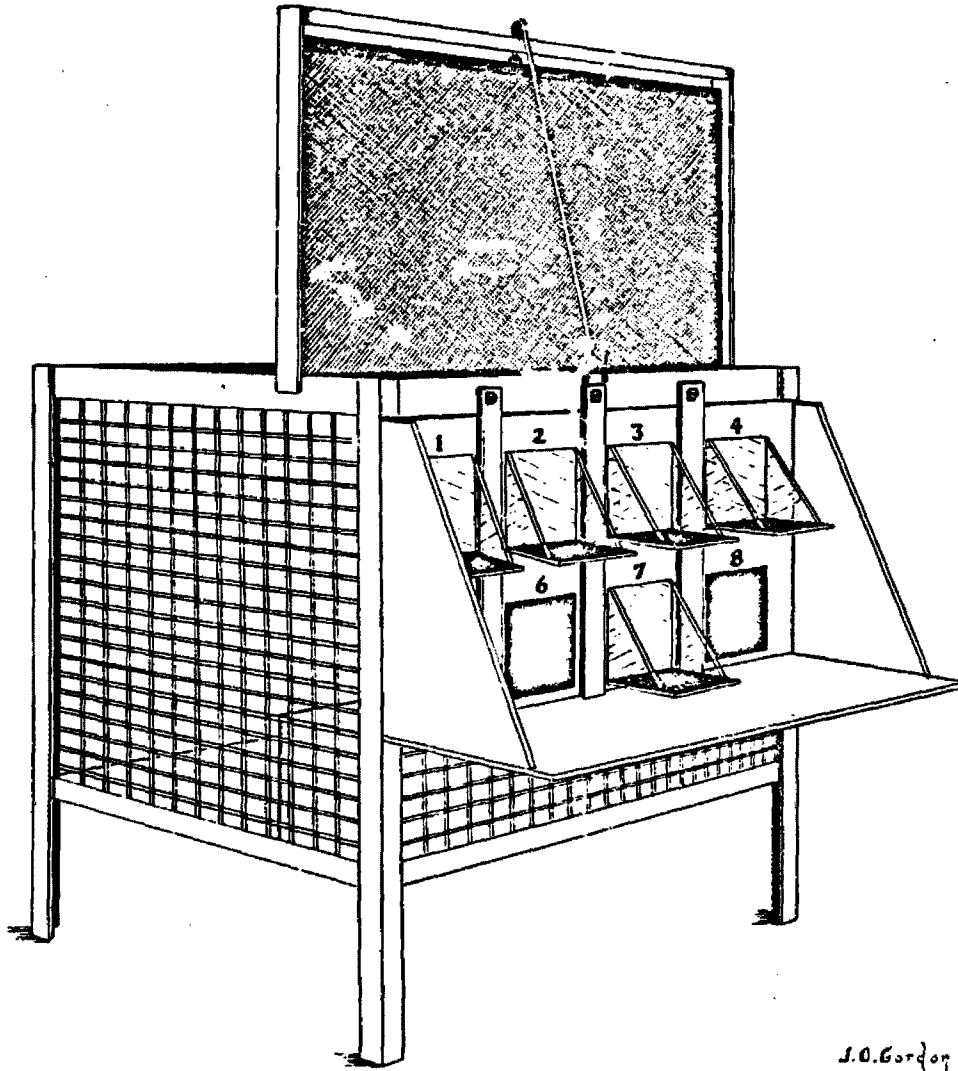
Each monkey was trained to open the reinforcement bins for a piece of sugar-coated cereal. The training continued until the monkeys opened all eight bins spontaneously during a one-minute trial.

The first part of the experiment used four categories of reinforcement: green 25% protein diet, white 3.5% protein diet, red 2% protein diet, and approximately 200 objects constructed of miscellaneous toys ranging up to 7.6 cm. in diameter. The monkey was placed in the cage with the opaque door blocking his view of the preference wall. Two 1/2 gram portions of each of the three diets, and two of the objects, were randomized and individually loaded into the eight bins. The partition was raised for a one-minute interval, then lowered. If the monkey opened all of the bins in less than one minute, the partition was lowered after the last bin was opened. Each monkey was given five sessions consisting of nine one-minute trials.

For each trial, the following information was recorded by the experimenter: 1) the type of reinforcement in each bin; 2) the order in which the bins were opened by the monkey; 3) whether or not the reinforcement was removed from the bin; 4) the total time spent opening the bins, if all bins were opened in less than one minute.

In the second part of the experiment, the color of the 2% protein reinforcer was reversed from red to green, while the color of the 25% protein reinforcer was

FIGURE 1



Artist's conception of the preference wall apparatus

changed from green to red. The color of the 3.5% protein reinforcer remained white. Each monkey was run as before, but twice as many trials were given, to allow stabilization of preference behavior. The subjects were given 10 sessions, consisting of nine one-minute trials each. The animals were fed 150-200 grams of diet each day late in the afternoon. All food remaining was removed from the cage and drop pan the following morning. Testing was conducted just prior to the afternoon feeding.

## CHAPTER III

### RESULTS

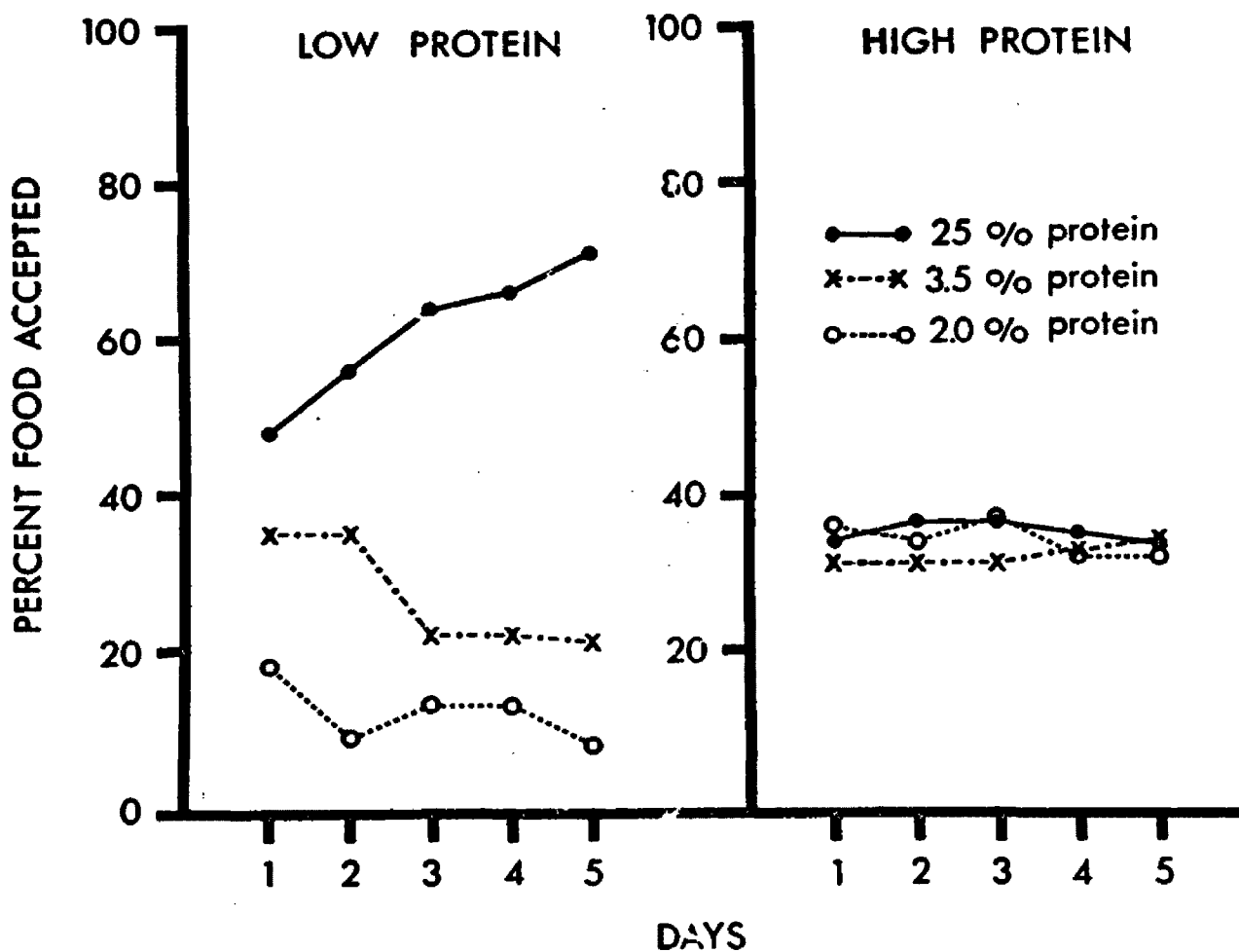
The amount of each diet accepted was compared to the total number of food reinforcements accepted. This ratio was calculated for each monkey in a group, then the values were averaged to obtain the points on the graphs (Figure 2).

The low protein monkeys were found capable of learning to select the high protein diet from an array of items including low, medium, and high protein diets, along with various non-food objects (Figure 2).

These animals started to select the 25% protein diet during the first session. By the end of the fifth session, 71% of the food accepted by the malnourished monkeys was 25% protein, while only 21% was 3.5% protein, and 8% was 2% protein. They accepted significantly more pieces of the 25% diet than either the 2% or 3.5% diet during the fifth session ( $p < .001$ , by correlated t-test). No significant differences were found between the selection of the 2% and the 3.5% protein diets. The high protein monkeys accepted about the same number of each of the three protein diets; no significant statistical differences were found among the acceptance values of the three diets during the final session.

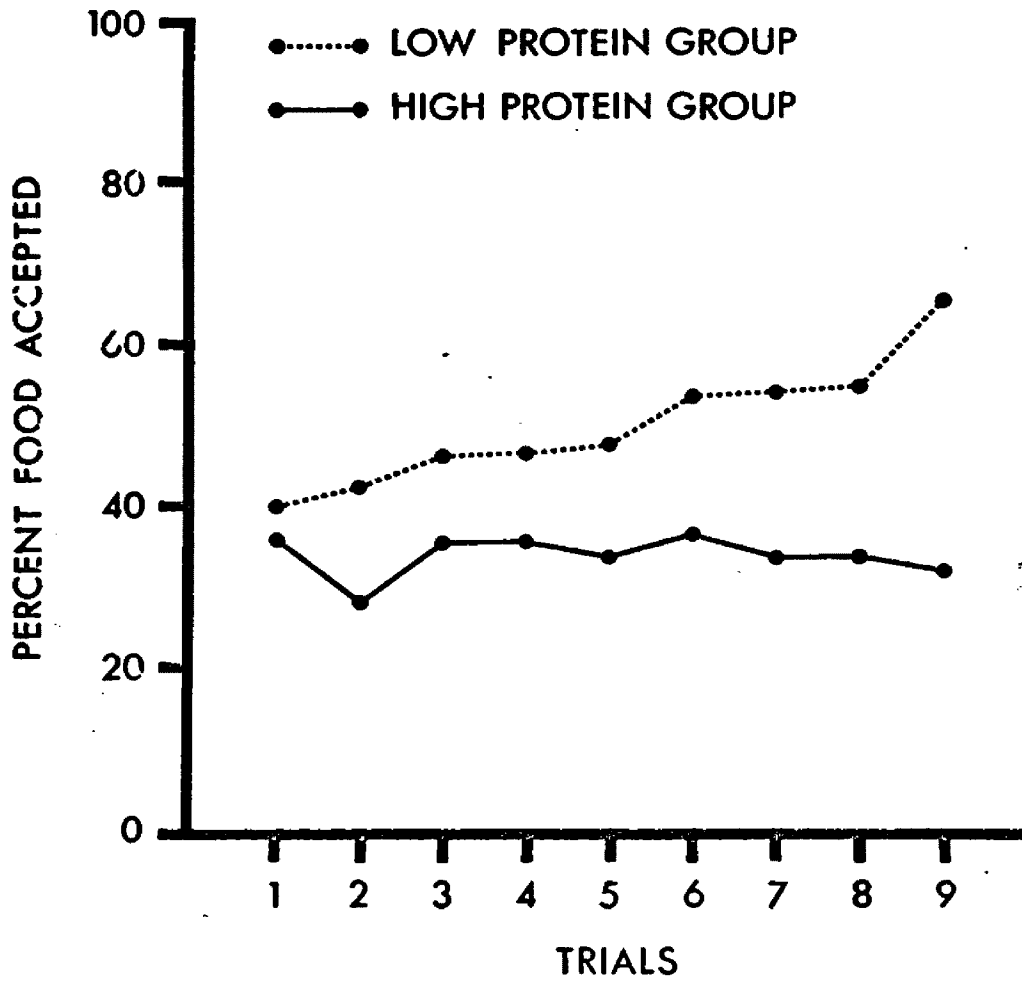
The low protein monkeys rapidly learned to discriminate between the protein values within the nine one-minute trials of the first session (Figure 3). The high protein diet accounted for 29% of the total food accepted during the first trials, but rose to 66% of the food accepted during the last trials. The malnourished monkeys did not accept more high protein reinforcements as the trials progressed; they accepted almost all diet pieces offered during the first trial, and took fewer 2% and 3.5% protein pieces toward the end of the session. The high protein monkeys chose about the same number of each diet reinforcement throughout the first session.

FIGURE 2



Percent accepted values for the high protein and the low protein monkeys during the original experiments.

FIGURE 3



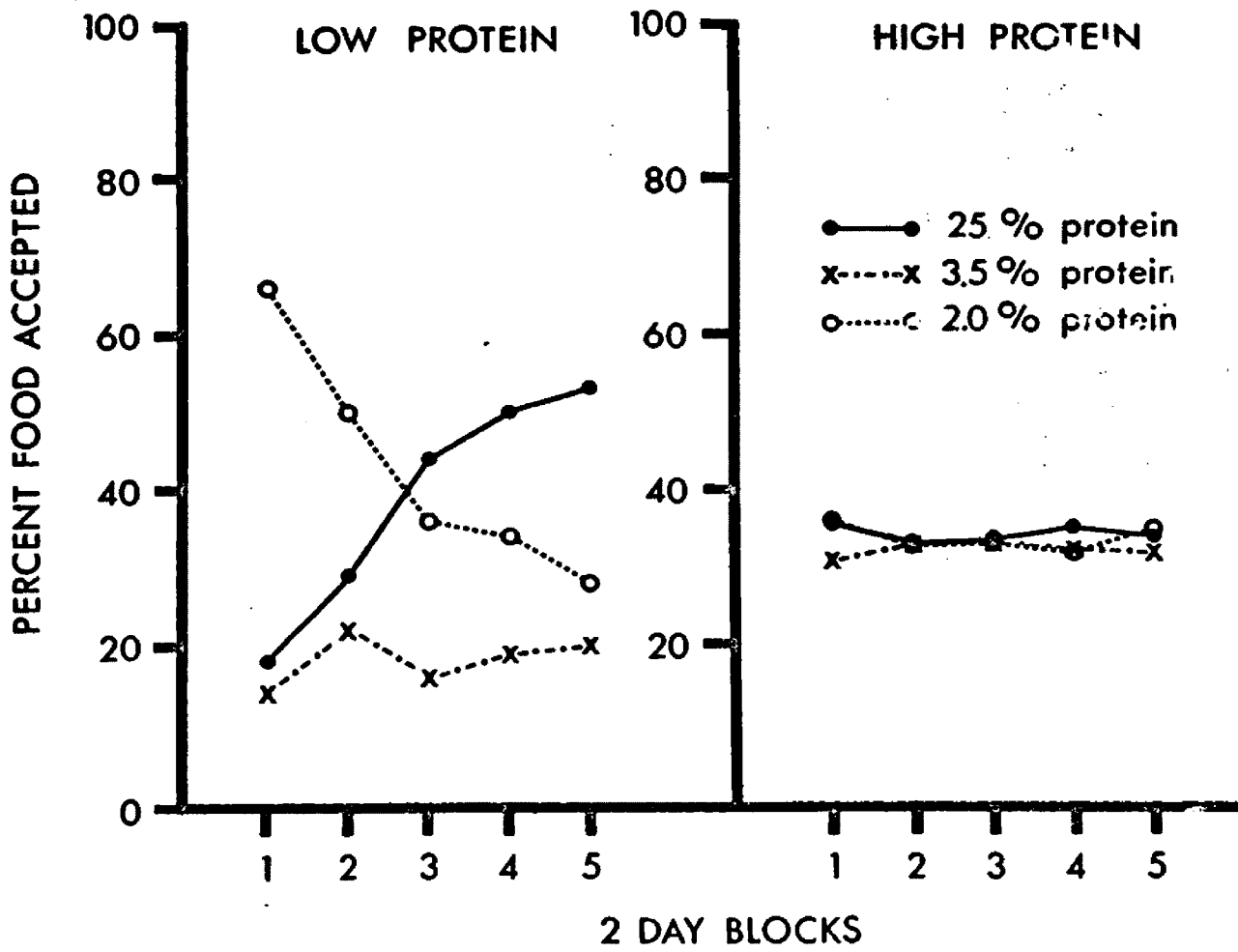
Percent high protein diet accepted during the nine trials of the first session.



When the colors of the 2% and the 25% protein diets were reversed, the low protein monkeys again learned to select the differently colored 25% protein diet. The results, in terms of percentage accepted scores, are shown for two session blocks in Figure 4. The low protein monkeys' acceptance rate of the 25% protein diet dropped from 71% on the last trial of the original experiment to 18% on the first trial of the reversal ( $p < .001$ , by correlated t-test). Their acceptance rate of the 2% protein jumped from 8% to 65% during the same time ( $p < .001$ , by correlated t-test). The acceptance rate of the 3.5% protein diet did not change.

The preference by malnourished animals for the 25% protein diet took longer to develop during the reversal experiment than it did during the original experiment, but by the 9th and 10th sessions, 53% of the food accepted by the low protein monkeys was 25% protein, while only 28% was 2% protein, and 20% was 3.5% protein. They selected significantly more 25% protein diet reinforcements than either the 2% or 3.5% reinforcements ( $p < .001$ , by correlated t-test).

FIGURE 4



Percent accepted values for the high protein and the low protein monkeys during the reversal experiments.

## CHAPTER IV

### DISCUSSION

The results appear to suggest that the dietary history of the rhesus monkey has an effect on the quality of the food he will accept. The higher acceptance level of 25% protein diet by low protein malnourished monkeys compared to the depressed 2% protein diet acceptance level is compatible with the hypothesis that a specific hunger exists for protein. The low protein monkeys were specifically selecting foods with a high protein content from an assortment of foods containing low, medium, and high protein values. These results appear to contradict Hillman and Riopelle's (1971) conclusions that protein-deprived rhesus monkeys do not evidence a specific hunger for protein, or that preferences do not increase with increasing protein content.

In addition, the present results do not support Hillman and Riopelle's (1971) third conclusion, that lowering the level of protein deprivation decreases the threshold of the palatability of food, in general, and raises the acceptability of these foods. Just the reverse was found in the present experiment. Monkeys receiving diets adequate in protein accepted all three foods in a near random fashion indicating that all food types were equally acceptable, and that the threshold of palatability was low. The low protein monkeys, on the other hand, differentially selected the high protein food, showing a lower overall acceptance rate, and a higher palatability threshold. It appears that protein deprivation increases the threshold of palatability and decreases the acceptability of the foods.

The differences are evident between the present study and that of earlier studies and may be due to differences in the conditions of protein deprivation. Hillman and Riopelle's malnourished monkeys were fed 1, 2, or 4 grams of protein

per kilogram body weight per day for 22 weeks before the experiment, with controls getting 7.4 grams of protein per kilogram body weight per day (N. Hillman, Personal Communication, 1971). In the present experiment, the low protein monkeys received about 2 grams of protein per kilogram body weight per day for 2 1/2 years before the experiment, and controls had been fed about 12.5 grams of protein per kilogram body weight per day for the same period. The development of a specific hunger for protein could possibly be dependent on the duration of protein deprivation.

The color reversal was designed to see if the low protein monkeys were responding to the color of the diets, or if they were responding to other cues. The performance decrements in selecting 25% protein during the first session of the reversal indicated that they were indeed using color as a discriminative cue. The reluctance of the animals to change their color preference following a change in protein content demonstrates the degree to which habit influences food preferences. This relationship was previously shown by Weiskrantz & Cowey, (1963); Harriman, (1953); Young & Chaplin, (1945); and Siegel, (1957).

After the first reversal session, the low protein monkeys showed a drop in their acceptance of the green diet, accompanied by a concomitant rise in their acceptance of the red diet. The animals appeared to be responding to factors in the diets other than the immediate color cues. If color were the only cue, the malnourished monkeys would have had difficulty in learning the reversal. By the end of the reversal experiment, the low protein monkeys had again learned to select more of the 25% protein diet reinforcements than either the 3.5% or 2% protein reinforcement. It should be emphasized that during the reversal experiment, the subjects continued to receive food in their living cages that was color coded in the original colors. Thus, the low protein monkeys continued to receive

red 2% diets in their daily ration, but red 25% diet in the test situation. Had the food colors in the living cage been reversed, the learning in this phase of the experiment might have been more rapid. Nevertheless, it is apparent that the low protein monkeys made a discrimination between red diet in the living cage and red diet in the test situation.

Two views are held concerning the mechanism operating in specific hungers. Some experiments show that innate food preferences exist (Dove, 1935; Fay, Miller & Harlow, 1953; Bolles, Sulzbacher & Arant, 1964), while others show that these preferences are learned (Young, 1955; Smith, Poul & Weinberg, 1958; Scott & Quint, 1946; Mensel & Draper, 1965; Weiskrantz & Cowey, 1963). The present study supports the learning theory. The learning is extremely rapid and performance reaches its peak in nine one-minute trials of a single session. In Young's terminology (1968), the rapid preference or adaptive trend implies that the high protein food has a large hedonic intensity for the protein-deprived monkeys.

The acceptance level of the non-food objects was fairly low, and constant within the groups during both parts of the experiment. The high protein animals did, however, accept more of the objects than did the low protein animals. These results are consistent with the findings of Zimmermann and Strobel (1969) that high protein monkeys have a higher rate of manipulatory activity of non-nutritive objects than low protein monkeys in the same situation.

The results of this experiment indicate that monkeys deprived of protein can discriminate diets that are isocaloric yet different in protein content. The diets differed on a variety of dimensions such as sugar content and texture, so the source of the discriminative cue is not known. But the discrimination was learned rapidly when color cues for the various protein contents were available and the persistence of this preference was demonstrated when those cues were reversed.

## CHAPTER V

### SUMMARY

Ten protein-malnourished monkeys and eight dietary controls were run in a preference experiment designed to look at the differences in the way the two groups accepted foods containing various amounts of protein. Each monkey was given choices among diets containing 2%, 3.5% or 25% protein, and non-food objects. The protein-malnourished monkeys showed a marked preference for the high protein food; the adequately nourished monkeys chose all foods at the same ratio. These results indicated that a rapid learning process was involved in the formation of a preference for high protein foods. The colors of the 2% and the 25% diets were then reversed, and the protein-malnourished monkeys showed a temporary disruption in their preference behavior, but they soon established a stable preference for the high protein food. The experimental results are consistent with the hypothesis that protein-malnourished rhesus monkeys have a specific hunger for protein.

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