

agreement with that recently found by the counting method (5).

For the TIV experiments the virus was suspended and centrifuged down several times at low centrifugal force in 0.075M KCl containing potassium phosphate to give a final ionic strength of 0.1 and a pH of 7.0. The washed virus was resuspended and dialyzed against this solvent for 24 hours before insertion into the gravity cell. The concentration was estimated from refractive measurements in a differential refractometer at 20°C with the 5461 Å Hg line. Dry-weight determinations were carried out on both solution and equilibrated solvent from which a value of 0.199 ml/g was found for the specific refractive increment. The nitrogen content of this sample was 16.0 percent. No extraneous material nor aggregates were observed in schlieren photographs during velocity sedimentation in a double sector cell; the sedimentation coefficient,  $s_{20}$ , was  $22 \times 10^3$  S.

The principal drawback to the gravity cell is the long time required for equilibrium to be established. Unfortunately, the equilibrium time cannot be shortened by some of the simple devices used in ultracentrifuge experiments (2, 6), but short cells can be used and layering techniques could be employed. However, the cell requires virtually no care once it has been set up. The time required for equilibrium to be established can be estimated by the theory of Mason and Weaver (7). Also the effective range over which the particle

weights can be determined is limited by the precision with which  $(c_2 - c_1)$  can be determined and upon sedimentation of the solute on the bottom of the cell. While experimental tests for the latter should always be carried out, its effects can be estimated from the relation of the velocity of sedimentation to the velocity of back diffusion (3). In general the apparatus should be quite useful in the molecular weight range from  $10^8$  to  $10^{10}$ , depending somewhat upon the partial specific volume and density (8).

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## Potency of Conditioned Reinforcers Based on Food and on Food and Punishment

**Abstract.** *Pigeons peck more frequently on the left key of two keys when food is presented more frequently during the stimulus that reinforces pecks on that key than during the stimulus that reinforces pecks on the right key. This preference can be annulled and reversed by punishing each peck on the left key during the stimulus that reinforces pecks on the left key.*

Much has been learned recently about the effects of punishment on an organism's tendency to emit a single reinforced response (1). In this experiment (2), punishment alters the pigeon's tendency to emit each of two responses, each of which is reinforced with presentations of a stimulus associated with a different frequency of intermittent positive reinforcement.

Two pigeons at 80 percent of their free-feeding weights were reinforced daily on two, concurrent, chained

schedules of reinforcement, a procedure studied extensively by Autor (3). The experimental chamber contained two response keys. One chain composed of two links was programmed on each key. Pecks on a key during the first link of the chain were reinforced on a 3-minute, variable-interval (VI) schedule with the presentation of a second color of key signaling the start of the second link. Pecks on the key in the second link were reinforced with 4 seconds of access to mixed grain, accord-

ing to a variable-interval schedule of a different value for each key. The first links of the chains were programmed concurrently; the left key was lighted orange and the right key was simultaneously lighted green. However, during the second link on a key, only that key was lighted; either the left key was red and the right dark, or the right key was yellow and the left dark. Each second link lasted for 30 seconds. When the second link ended, both keys were again lighted with the colors appropriate to the first links.

Pecking during the concurrent first links of both chains was maintained by a conditioned reinforcer, the presentation of the second color, on which pecks were reinforced with grain. A preference for pecking the left key in the first link was established by reinforcing 72 pecks per hour (on the average) in the second link on the left key and only 30 pecks per hour in the second link on the right key.

Punishment was introduced in order to determine how much current following each peck during the second link on the left key would annul and reverse the preference for pecking the left key during the first link, when both keys were lighted. Each peck on the left key during the second link passed an electric current through the pigeon for approximately 30 msec (4). The current was varied between 0 and 2.9 ma by adjusting a resistance in series with the pigeon and the source (120 volt a-c). Each intensity was in effect for a minimum of seven sessions and until the responding did not change systematically from session to session. After the intensity of punishment that annulled the preference based on 72 and 30 reinforcements per hour was determined, the experiment was repeated with 43 and 18 reinforcements per hour.

Figure 1 shows the rate of pecking during the two links of the chain on each key as a function of the intensity of the punishment following each peck during the second link on the left key. The upper graphs show the rates of pecking during the concurrent first links. The lower graphs show the rates during the second links. Circles and triangles represent different pigeons. Filled points and solid lines show the rates of pecking maintained by 72 and 30 reinforcements per hour. Unfilled points and dashed lines show the rates maintained by 43 and 18 reinforce-

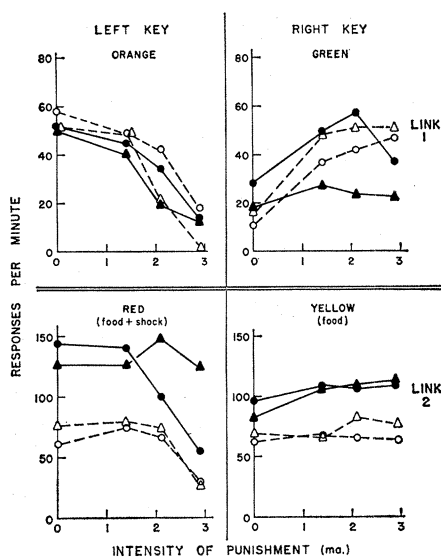


Fig. 1. Rate of pecking on the left and on the right keys during link 1 (upper graphs) and during link 2 (lower graphs) as functions of the intensity of punishment (milliamperes of electric current) following each peck on the left key during link 2. Filled and unfilled points and solid and dashed lines distinguish the effects of different frequencies of positive reinforcement; triangles and circles distinguish the two pigeons.

ments per hour. Each point is the median of three or five successive sessions.

The effect of increasing intensities of punishment on the rate of pecking the left key during link 2 was variable (lower left graph). One curve (filled triangles) shows almost no effect of punishment on the rate of the pecking directly punished. Despite this, the rate of pecking the left key during link 1

(upper left graph), when both keys were lighted, declined regularly as the intensity of punishment increased. This shows the sensitivity of the present method in measuring the effects of punishment, although the change from 72 to 43 reinforcements per hour during link 2 did not produce an expected (5) decrease in the rate of pecking during link 1 (filled vs. unfilled points).

The rate of pecking the right key during link 2 (lower right graph) was relatively constant, but when both keys were lighted during link 1 the rate of pecking the right key generally increased (upper right graph). Thus punishing pecks during the second link on the left key had two effects on behavior when both keys were lighted during link 1: less frequent pecking on the left key where pecks changed the color to one associated with punishment and positive reinforcement and more frequent pecking on the right key where pecks changed the color to one associated with a lower frequency of positive reinforcement.

The change in preference with increasing intensity of punishment was accompanied by a decreasing tendency to peck the left key twice in succession (Fig. 2a). The ordinate in Fig. 2a is the proportion of pecks on the left key for which the next peck was also on the left key. The symbols are the same as in Fig. 1.

Figure 2b summarizes the approximately linear change in preference as a function of the intensity of the punishment. The ordinate is the number of pecks on the left key divided by the total number of pecks on the two keys. At about 1.7 ma, where this statistic has a value of 0.5, there is no preference for either key. That is to say, a stimulus associated with 72 (or 43) reinforcements per hour and 1.7 ma of punishment maintained a rate of pecking on the left key that was approximately equal to the rate maintained on the right key by a stimulus associated with 30 (or 18) reinforcements per hour and no punishment. Punishing and decreasing the frequency of positive reinforcement may thus have the same effect on the reinforcing potency of a conditioned reinforcer, in the sense of decreasing the rate of pecking maintained by presentations of the conditioned reinforcer.

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## References and Notes

1. See N. H. Azrin and W. C. Holz, *J. Exptl. Analysis Behavior* 4, 343 (1961), which contains additional references.
2. Supported in the Harvard Psychological Laboratories by grant M-5139 from the National Institutes of Health. Expenses of preparation paid by grant B-316 from the National Science Foundation to the University of Chicago.
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## Serum Protein Synthesis by the Fetal Rat

**Abstract.** Injection of  $C^{14}$ -labeled amino acids intraperitoneally into rat fetuses in utero results in greater labeling of fetal serum proteins than when the amino acid is injected into the mother. Maternal partial hepatectomy leads to only minimal changes in serum protein synthesis. Rat fetuses synthesize at least some of their serum proteins during the last 3 days of gestation.

There is both genetic (1) and isotopic (2) evidence that serum proteins are produced by the mammalian fetus. The phenotypes of the haptoglobin and transferrin present in human cord serum may differ from those in the maternal serum, thus indicating their formation by the fetus (1). Labeled amino acids have been shown to be incorporated by slices of human fetal liver and by premature guinea pigs in vivo into serum albumin and the serum globulins with the exception of  $\gamma$ -globulin (2).

We have previously shown by immunoelectrophoresis that the rat fetus gradually acquires most of the serum proteins present in the adult during the last part of gestation (3). This report presents evidence of the production of serum albumin and at least some of the serum globulins by the rat fetus during this period. Effect of maternal subtotal hepatectomy on forming maternal and fetal serum proteins was investigated.

Sprague-Dawley rats in the 20th to 22nd day of gestation were used in all experiments. In one series of experiments, the pregnant rats were anesthetized and the left and median lobes of the liver were removed. The labeled amino acid was injected via the tail vein and the mother and fetuses were bled 6 hours later. Intact and sham-operated rats were injected similarly to

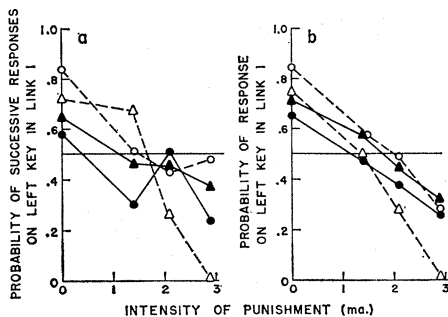


Fig. 2. (a) The proportion of pecks on the left key during link 1 for which the next peck was also on the left key; (b) the number of pecks on the left key during link 1 divided by the total number of pecks on each of the two keys during link 1, both as functions of the intensity of punishment following each response on the left key during link 2. The symbols are the same as in Fig. 1.