Operant Discrimination Learning and Operant Bar-Pressing Rates in Inbred and Heterogeneous Laboratory Mice

By: Benjamin Padeh, Douglas Wahlsten, and J. C. DeFries

Abstract:

Mice from four inbred strains and a heterogeneous line were first reduced to 90% of their ad libitum body weight and were then trained to press a bar to obtain food pellets. After 9 days of training with a continuous schedule of reinforcement (CRF), mice were required to discriminate between conditions of reward and nonreward as indicated by a tone and a light; discrimination training continued for 21 days. Considerable genetic variation was apparent for asymptotic pressing rates under CRF as well as discrimination performance. However, the highest discrimination ratios were achieved by mice with the lowest pressing rates. Results are discussed in terms of associative and nonassociative processes which may contribute to genetic variation in performance on learning tasks.
KEY WORDS: operant conditioning; strains; mice; learning.

Article:
INTRODUCTION

In the genetic analysis of learning, it is frequently assumed that genotypes which lead to the highest levels of performance also possess the greatest degree of learning ability. There is abundant evidence, however, that a host of factors independent of associative learning may determine which genotypes are superior on a particular task (Wahlsten, 1972b). The problem is especially complicated because the factors most important in determining genetic differences in performance on one task may not be involved at all in genetic variation on another task (e.g., Wahlsten, 1972a, 1973).

Many of these problems may be circumvented to a large extent by the use of a proper test of learning which compensates for nonassociative influences on performance level. The vast literature on the psychology of learning has revealed several ways to accomplish this (Kimble, 1961; Teitelbaum, 1966; Bitterman and Schoel, 1970). Perhaps the most important conclusion reached by psychologists is that tasks which require the subject to make a choice or discriminate clearly defined alternatives are not influenced to so great a degree by differences in motivation, activity, or operant levels as are unitary tasks which require a simple running or operant response. Measures of response speed or operant rate in unitary tasks are especially sensitive to differences in both primary and incentive motivation (Bolles, 1967), and are therefore not to be taken as direct indices of learning ability.

These problems are clearly seen in the situation used in the present study, namely operant training of bar pressing in mice. It is easy to imagine that strains which have operant levels of pressing when no reward is given would also acquire higher pressing rates when reward is present, quite apart from any differences in learning ability. A positive relation between operant level and rewarded response rate has in fact been reported in two previous studies with mice (Goodrick, 1967; Southwick and Clark, 1968), although a third study reported no clear relation (Smart, 1970). Even the fixed-ratio and delayed-reward schedules employed by Smart (1970) did not clearly separate activity or performance factors from learning.

In the present study, operant activity levels were rendered less important by requiring mice of several genotypes to discriminate between conditions of reward and nonreward within a training session. Whereas general operant levels should influence pressing rates under both conditions similarly, the difference between rates in the two conditions should reflect the subject's ability to adapt or change its behavior to suit variations in its environment, i.e., to learn.

MATERIALS AND METHODS
Subjects
Mice were from the four highly inbred strains A/Ibg (24 female, 20 male), BALB/clbg (18 female, 19 male), C57BL/6Ibg (17 female, 18 male), and DBA/2lbg (24 female, 19 male) and from the outbred line HS/Ibg (42 female, 42 male). The HS line is a heterogeneous population of mice originally derived by crossing eight highly inbred lines (see McClearn *et al.,* 1970).

Apparatus
The apparatus consisted of ten identical Skinner boxes each 17 by 14 by 14 cm. The long side walls were made of black plexiglas, the ceiling and the front and rear walls were built of clear plexiglas, and the floor was covered with perforated aluminum. On the front wall, 5 cm from the right wall and 3 cm above the floor, the lever, a 4- by I .5-cm black plexiglas rectangle, was attached to a microswitch; it required a force of at least 6g to operate. For visual signals, a small pilot lamp (Sylvania 28ESB) was connected to a 220-ohm series resistor operating from a 28-v supply and was mounted 3 cm above the bar. For auditory signals, a loudspeaker (Speco u260, 5 cm diameter, 8 ohm, 0.1 w) was mounted in the middle of the ceiling; it delivered a pure tone of 11 kHz.

The ten Skinner boxes were enclosed in ten sound-attenuating chambers (20.5 by 33 by 43 cm) within refrigerators. In the middle of the rear wall of each chamber, a 28-v lamp (G.E. 1829) provided diffuse illumination of the entire chamber (house light). A ventilating blower and a pellet dispenser (Davis model No. 104) for each chamber were mounted on the external wall of the refrigerators. Food rewards were 45-mg Noyes "Peanut" pellets.

Detailed descriptions of materials and methods are available in Padeh (1972).

Procedure
Mice were weaned and then housed with like-sexed littermates at 21 days for HS and at 25 days for inbred strains. At 65 ± 6 days of age, food deprivation was begun; each mouse was housed individually and reduced to and maintained at 90% of its *ad libitum* body weight.

The first stage of training began 12 days after the initiation of food deprivation and lasted 3 days. Mice were first trained to approach the magazine to obtain food and were then trained to press the bar to obtain food from the magazine on a continuous reinforcement schedule (CRF). The session on the first day was 4 hr long, while sessions on the next 2 days were 1 hr long. The second stage consisted of six daily 50-min sessions, each a sequence of 55-sec periods of a CRF schedule separated by 5-sec blackout periods during which the house light (HL) was off and reward was not given for bar pressing. The third stage entailed 21 successive days of discrimination training. Each 50-min session had 25 periods of 55 sec with the 11 kHz tone and pilot lamp on (1.1.) and food reward available, and 25 periods of 55 sec with tone and light off ($\overline{\text{TL}}$) and reward unavailable; 55-sec periods were separated by 5-sec blackout periods with the house light out and reward unavailable. Reward for pressing under TL was on a variable-interval schedule (VI 10 sec).

Raw data included the numbers of bar presses during periods of reinforcement, periods of no reinforcement, and blackouts, as well as number of food pellets received.

RESULTS

The rates of bar pressing during the CRF schedule of stages 1 and 2 are shown across days for both sexes and all strains in Fig. 1. As is clearly evident, all groups showed a large increase in response rate across days in stage 1. Repeated measures analysis of variance revealed large strain differences in both stage 1 ($F = 9.7$, df = 4/233, $p < 0.01$) and stage 2 ($F = 10.3$). Sex differences were also significant in both stages 1 ($F = 6.8$, df = 1/233, $p < 0.01$) and 2 ($F = 16.7$), but a large strain-by-sex interaction was present in stage 2 ($F = 8.1$, df = 4/233, $p < 0.001$). Strain rank orders were generally similar for males and females, except for the high rate of DBA males and low rate of DBA females. The bar-pressing rates in stage 2 appeared to be asymptotic for all groups; although the change in rate across days was significant ($F = 3.3$, df = 5/1165, $p < 0.01$), no linear trend was evident, and no interactions with days were significant.
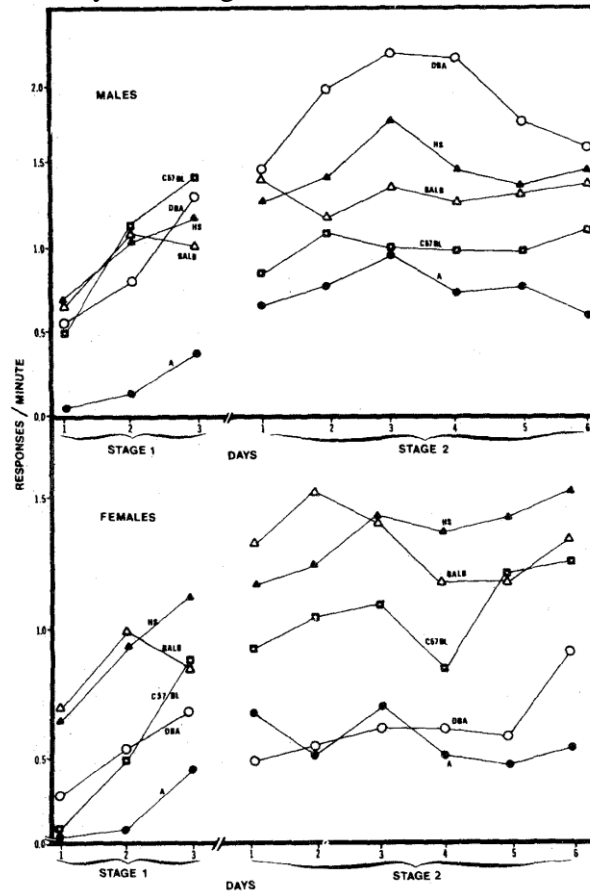

Fig. 1. Daily rate of response to reinforced trials for males and females of each strain during stages 1 and 2.

Rates of pressing during the 5-sec blackout periods in stage 2 were very similar to those during the CRF periods for each group, and they showed no significant changes across days or interaction with days. In general, all groups showed slightly higher response rates during the CRF than during blackout from the onset. Thus no evidence of discrimination learning was present in stage 2.

From the results of stages 1 and 2, large genetic differences in response rate are clear, but they could represent the influence of different levels of motivation or general motor activity as easily as the effect of different associative learning abilities.

During discrimination training in stage 3, response rates under the TL condition increased two to four times above the CRF level of stage 2, as would be expected on a VI schedule. The rates during TL showed very little change after the third day of discrimination training (see Fig. 2). The rates of responding during the unreinforced $\overline{TL}$ condition also increased initially, but they gradually declined over days (see Fig. 3). Strain and sex differences were highly significant for pressing rates under both TL and $\overline{TL}$, but the strain-by-sex interaction was significant only during the unrewarded $\overline{TL}$ period ($F = 3.8$, df = 4/233, $p < 0.01$). Females in general were better able to inhibit responding during $\overline{TL}$, but only A females attained a truly low level of responding during

$\overline{TL}$. Among males, declines in rates to $\overline{TL}$ were generally small, while DBA males actually increased their unrewarded response rate slightly.
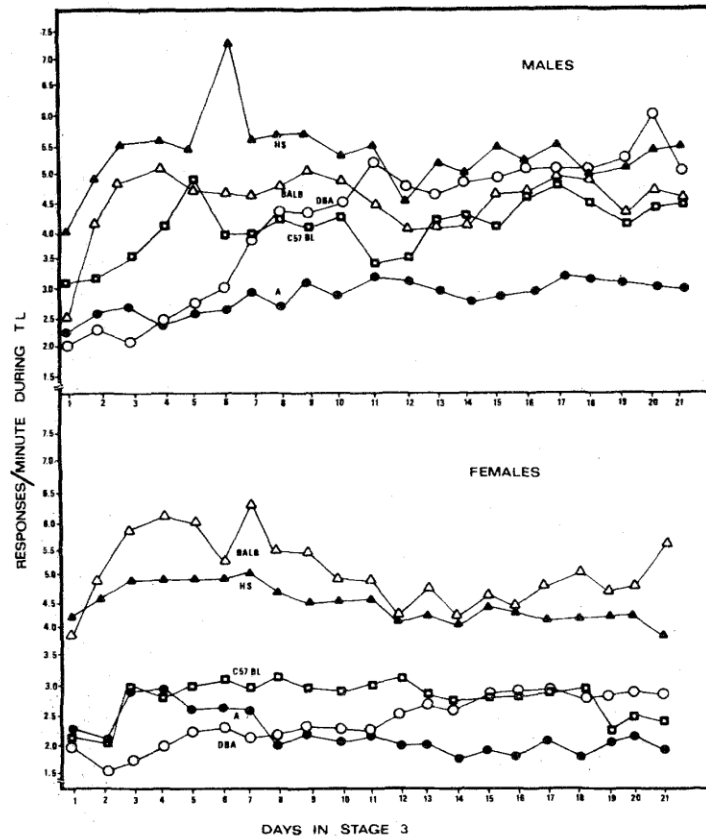

Fig. 2. Daily rate of response to reinforced trials for males and females of each strain during stage 3.

Although all groups of mice showed a higher rate of pressing during TL than $\overline{TL}$ after a few days of training, there appeared to be a strong correlation between rates of pressing under the two conditions across strains. A mice had the lowest pressing rates under both conditions, while HS and BALB mice generally were highest. Pressing rates during the blackout period also showed the same strain ranking. As in the case of stage 2, response rates during discrimination training strongly suggest the importance of nonassociative motivational or activity factors for genetic variation in bar-pressing performance.

Whether differences in associative learning exist therefore depends on the *relative* rates of pressing during reward and nonreward conditions. Although the absolute difference between rates during reward and nonreward is sometimes employed as a measure of discrimination in studies using only one strain, it is clearly unsatisfactory when various strains differ so widely in operant levels. A more satisfactory measure for the present purposes is the ratio of rate during TL to the sum of the rates during both TL and $\overline{TL}$. This discrimination ratio is shown across days for all groups in Fig. 4. All groups started near 0.5 and increased substantially over days. The strain × sex interaction was highly significant ($F = 5.9$, df $= 4/233$, $p < 0.001$).

Whereas most groups increased their ratio gradually to about 0.7 after 21 days, the A females rapidly achieved a value of over 0.85, while DBA males required more than a week to rise above chance level.

DISCUSSION

The present results clearly demonstrate that strains achieving the highest response rates during continuous reinforcement (CRF) do not achieve the highest discrimination ratios when distinctive cues signal the availability or impossibility of reward. In fact, the group with the lowest pressing rate during CRF, A/Ibg females, attained the highest discrimination ratio, while the DBA males pressed most rapidly under CRF but showed the poorest discrimination learning. Together with the fact that the elimination of responses to TL took longer than the elevation of responding to $\overline{TL}$, these results suggest that the ability to withhold or inhibit

responses is a very important aspect of discrimination learning, provided that the nature of the task renders response speed unimportant (see also Carlton, 1969).
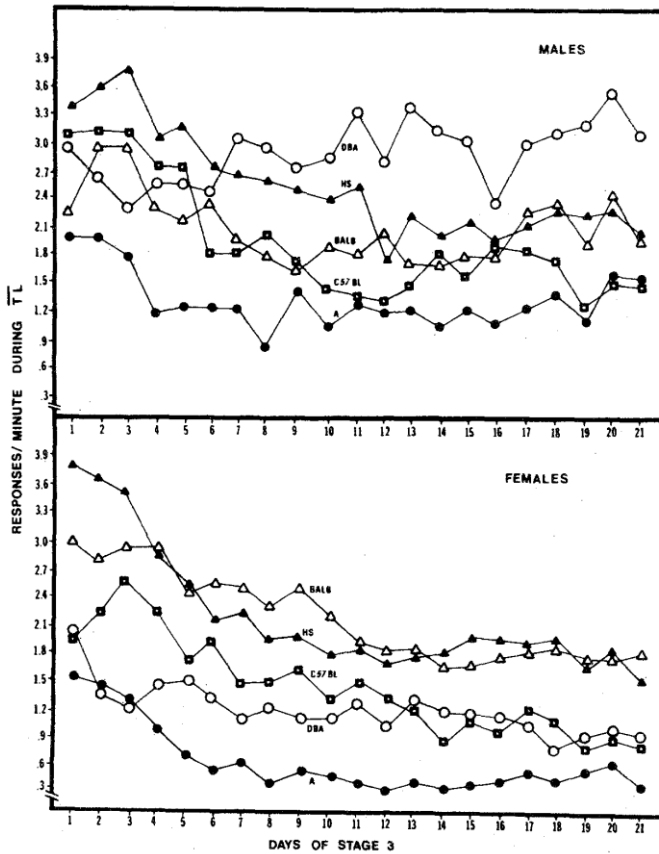

Fig. 3. Daily rate of response to unreinforced trials for males and females of each strain during stage 3.
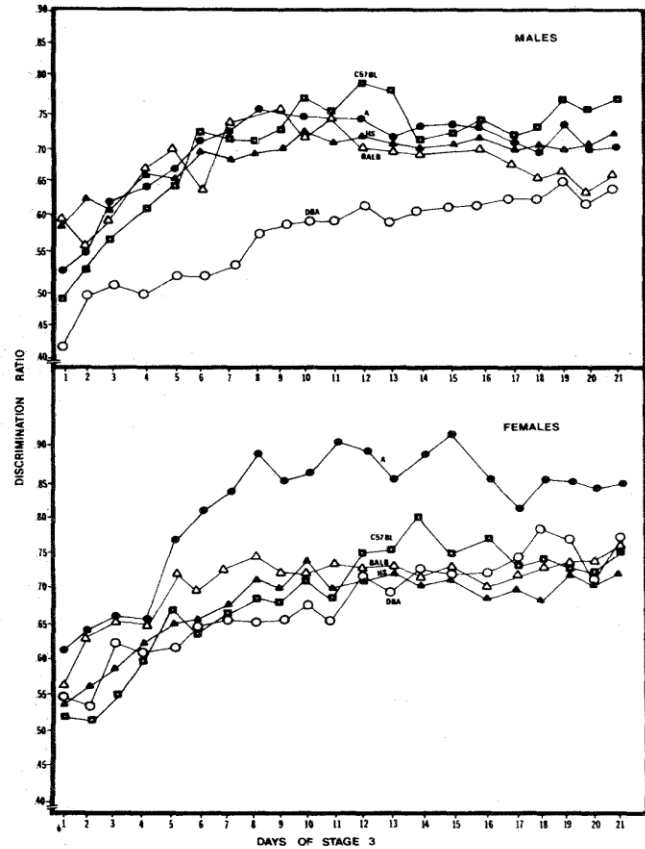

Fig. 4. Discrimination ratio for males and females of each strain.

Whether motivational differences among the strains contributed to variations in performance remains to be established. Although the low pressing rates of A mice suggest that their hunger drive may have been low, their superior discrimination performance reveals no deficiency in their motivation. Nonetheless, it is conceivable that the acquisition of the difficult discrimination of a VI schedule from nonreward proceeds most rapidly at low or moderate levels of motivation. This question can be answered only through direct manipulations of levels of food deprivation or related procedures.

That nonassociative processes may strongly affect results in genetic studies of learning is also demonstrated by recent experiments with avoidance learning. Fuller (1970) reported an inverse rank ordering of four inbred strains on active and inhibitory shuttle avoidance learning; strain differences were primarily produced by differing levels of "kinetic drive." Studies with discriminated avoidance learning in a Y-maze have shown that strain differences in avoidance are more reflections of the ability to initiate a running response in stressful situations where freezing behavior is predominant than of the ability to discriminate safe from dangerous regions (Barrett *et al.,* 1973; Anisman, 1974). All three of the above studies also found that drugs which disrupt freezing facilitate response initiation but have no effect on discrimination ability. Another recent study by Alpern and Marriott (1972) has shown that the AO strain, which is notoriously poor in active avoidance involving a handling procedure (Schlesinger and Wimer, 1967; Wahlsten, 1973), actually acquires a discrimination reversal in T-maze shock avoidance faster than the DBA strain, which is quite good when a rapid response but no choice must be made. Using regression analysis, Oliverio *et al.* (1972) have found that wheel-running activity shows a strong, negative genetic correlation with shuttle avoidance, while activity is not correlated with performance in a Lashley III maze. They also demonstrated that septal lesions, which typically disrupt freezing responses, greatly improved the otherwise poor performance of C57BL/6J mice in shuttle avoidance but disrupted discrimination learning of three strains in a maze (Oliverio *et al.,* 1973). Thus it is becoming apparent that genetic variation in performance on unitary tasks where response speed or vigor is

important has little relation to performance on more complex discriminations which clearly require associative learning ability.

As these issues concerning genetic variation in associative and nonassociative processes are rapidly becoming resolved, it is perhaps fitting to question whether the highly revered associative learning should continue to be regarded as the hallmark of adaptiveness, while activity and inhibition are relegated to the phylogenetic graveyard. It is worthy of note that the A/Ibg female mice, in spite of their wise restraint and careful deliberation, actually received fewer food pellets than any strain but DBA! When responding during nonreward conditions is not actually punished, maybe rapid, indiscriminate responding is the best strategy, especially when the discrimination is very difficult. How foolish we apostles of intelligence may someday appear when it is discovered that our wily subjects have refused to cross the threshold into our mental mousetrap!

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