An animal restraint for the study of high-resistance operants in the rat

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An animal restraint system and response operandum suitable for the study of operant behavior in the rat is described. This system combined with an inexpensive Commodore microcomputer and Psychronix interface is well-suited for the study of response parameters. Rats voluntarily entered the restraint, showed no observable signs of excess stress, and were easily extricated at the end of the experimental session. A lever-pressing operant with an adjustable force requirement was readily shaped using automated procedures. Although this system was designed for the study of high-resistance operants, it has potential advantages for other operant applications, including studies of visual discrimination and experiments requiring restraint due to physiological recording.

With few exceptions, an unrestrained animal model has been used in studies of operant behavior with the rat. The vast majority of these studies have been conducted in experimental chambers that are similar in concept if not in details of design to the original operant chambers utilized by B. F. Skinner (e.g., Skinner, 1938). Although the original apparatus design may have been primarily influenced by the desire to save labor (Skinner, 1979), this unrestrained animal approach has advantages that have left it virtually unchanged for over 50 years. In addition to reduced experimenter labor, these include the selection of the leverpress operant that has sufficient spontaneous occurrence to be readily conditioned, yet with not so high a frequency as to obscure the effects of reinforcement (Skinner, 1938). In the confines of the operant chamber, competing behaviors are limited, and stimuli are readily controllable.

The use of a restrained animal model has at least three potential advantages over the standard operant chamber for general studies of operant behavior: enhancement of stimulus control, elimination of competing behaviors, and restriction of response topography. The major advantage of the restraint in our laboratory lies in the ability of the experimenter to study operants having a specific topography. In our research, we have been concerned with the development of an animal model for studying the effects of high-resistance exercise. In addition to being interested in the variables controlling rate, duration, magnitude, and intensity of high-resistance operants, one of us (B.F.T.) is an exercise physiologist interested in cellular mecha-

nisms of muscle hypertrophy produced by high-resistance exercise.

Exercise-physiology research in this area has suffered for lack of an animal model needed to do controlled experiments. Research with human subjects has been compromised by lack of control over motivational variables and potential confounding by variables affecting muscle growth and development that occur outside of the experimental setting. Lack of an adequate animal model has also placed ethical limits on variables that can be studied. The use of steroids in conjunction with various exercise prescriptions cannot ethically be studied, because of the health risk associated with steroid use. Analysis of the effects of the variables that can ethically be manipulated in man are limited to the analysis of body fluids and biopsy tissue; entire muscles cannot be removed for cellular analysis.

Animal models of high-resistance exercise have been developed using the cat (Gonyea & Ericson, 1976) and the rat (Ho et al., 1980; Klitgaard, 1988; Wong & Booth, 1988). These models have had limited success in producing muscle enlargement of the magnitude normally seen in human power lifters and body builders. An important factor in the study of muscle enlargement is the use of an intraanimal control to eliminate the genetic effect on muscle fiber number as a confounding variable. This requires that the high-resistance operant be limited to only one limb, so that the contralateral limb can be used as a control. The models of Ho et al. (1980) and Klitgaard (1988) require the rat to exercise with both limbs and are therefore limited in their ability to control genetic variables. A major limitation of the cat model (Gonyea & Ericson, 1976) is the cost of animal care and housing.

We have developed a procedure and apparatus that eliminate some of these problems. The restraint combines the design concepts of an early device developed by Hall, Clayton, and Mark (1966) and a commercially available Acrylic Restraint marketed by Harvard Bioscience (cat

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52-0916). Development of our restraint incorporating a response operandum and computer control system has yielded a rat model for the study of high-resistance exercise as an operant and its effects on muscle tissue. In the present paper, we describe the restraint, operandum, and microcomputer control system designed for the study of high-resistance operants.

APPARATUS

The apparatus consists of an animal restraint system that can be housed inside an animal chamber designed for a standard operant chamber. A BRS/LVE liquid solenoid valve mounted on the outside of the enclosure can be readily programmed to deliver 50 μ l of 10% sucrose solution, which serves as an adequate reinforcer. To eliminate the delay of reinforcement associated with transit time of the liquid from the valve to the dispenser, we attached a hypodermic needle to the "animal" end of the tube, and filled the tube with liquid at the beginning of each day. Opening the valve reliably results in a drop of liquid

being dispensed from the needle. The expedience of inserting the needle into a standard drinking tube with a bearing in the end allows the animal to receive the reinforcer by licking the end of the tube.

Restraint System

The restraint, shown in Figure 1, consists of three Plexiglas parts mounted on an Alice Chatham King sling suit frame. The *animal support* is a flat sheet of 4-mm-thick Plexiglas, 625 $\times$ 150 mm. A 90° bend was made in the Plexiglas 380 mm from the end, to provide a vertical surface 150 mm wide and 245 mm high, which was suitable for mounting a houselight and cuelight. A 20 $\times$ 52 mm rectangular hole was cut 48 mm from one side and 100 mm from the vertical surface to permit one foreleg access to the response operandum. A second rectangular hole, 55 $\times$ 104 mm, was cut 67 mm from the rear edge. This opening was covered with hardware cloth to permit passage of feces and urine, a feature necessary to conduct 60-min experimental sessions. The *animal restraint* was fashioned from 3-in. (73-mm inside diameter) Plexi-

Figure 1. Restraint system for the study of operant behavior in the rat. All dimensions are in millimeters.

glas tubing by making the longitudinal slice in the tube shown in Figure 1.

Longitudinal slots were cut in the tube to provide ventilation and cross-sectional slots to receive the *tail stop* (not shown). A Plexiglas piece with horizontal ventilation slots and a 7-mm hole, drilled at a 30° angle to accept a standard stainless steel drinking tube (not shown), was glued to the front of the restraining tube, closing it on that end. The tube was then glued to a 176 mm wide × 355 mm long rectangular Plexiglas frame.¹ Drilling four holes in the animal restraint and animal support allowed both to be mounted on a rodent sling frame (from Alice King Chatham Medical Arts) and held together with the screw clamps that are supplied with the frame. Removing the top clamps allowed separation of the animal restraint and animal support for easy removal of the rat and for periodic cleaning of the apparatus. The tail stop consisted of a 67×76 mm Plexiglas rectangle with a 16×57 mm slot to accommodate the rat's tail. It was inserted into one of the restraint slots to prevent the rat from backing out of the restraint system during the experimental session.

Operandum

The response operandum, shown in Figure 2, consisted of a 70×40 mm rectangular Plexiglas "paddle" glued to one end of a 3.2-mm-diam steel rod 89 mm from a pivot (to center of paddle). A 100-kΩ linear potentiometer, mounted in a Plexiglas frame, served as the pivot. The steel rod extended for 108 mm on the other side of the pivot, thus allowing the force necessary to depress the paddle to be varied by attachment of lead weights of varying mass at different distances from the pivot.

Depressing the paddle with sufficient force rotated the potentiometer, altering its resistance. The response operandum was attached to the base of the rodent sling frame. It was positioned so that the paddle was directly below the access hole in the animal support and the lead

weight was accessible in front of the vertical portion of the animal support.

Microcomputer Control

The apparatus is controlled by a Commodore 64 microcomputer and Psychronix Interface (Crossman, 1984). Resistance from the operandum's potentiometer was applied across Pins 8 and 9 of Joystick Port 1 on the Commodore 64 microcomputer. A digital value corresponding to the paddle position (resistance of the potentiometer) was obtained using a machine language subroutine (see lines 5,000–7,040 in Appendix A) that was originally described in the *Commodore 64 Programmer's Reference Guide* (Commodore, 1982). This digitized index of paddle position can be utilized to measure excursion (change in paddle position over time) of the paddle-press operant, and it enables differential reinforcement of specific excursions. Control of houselights, cuelights, and liquid reinforcement is accomplished with the Psychronix interface. Data are written directly to disk. This permits alternative off-line analyses of a session—for example, several interresponse time distributions with differing bin widths are easily obtainable from a single session's data. Writing data directly to disk also permits using this system within the 5K memory limitations of a less expensive VIC 20 microcomputer should one be available. All programming, other than the machine language subroutine to read paddle values, was done in BASIC. Appendix A shows a sample program listing for an alternative fixed-time 20" fixed ratio 1 (alt FT20" FR1) reinforcement schedule that we used to establish the paddle-press operant. Documentation in the listing should be removed to speed program execution. Lines 10 through 120 initialize the interface and create the disk file for the session; the program assumes that each animal has its own data disk. As events occur, a string variable consisting of a character identifying the event and the clock reading (1/60-sec resolution) is created and written to disk (cf.

Figure 2. Response operandum used in conjunction with animal restraint. Lead weight is not to scale. Dimensions are in millimeters. Consistent with drafting conventions, zeros to the right of the decimal are indicated as slashed.

lines 160-170). The main portion of the program goes through a loop (lines 150-280), checking for a response (lines 210-220) and 20 sec of elapsed time (lines 230-240). The session ends after 50 reinforcers are delivered (line 270), exiting the loop for an end of session process.

DISCUSSION

We have used this apparatus to train 32 rats, collecting data over more than 50 daily 20-min sessions for each animal, and we have experienced no serious problems related to the equipment. Nor did we observe any physical ill effects on any of the animals. There was no evidence of bleeding, nor of torn claws; nearly all the feces and urine produced in a session did pass through the hardware cloth, and any that remained was easily wiped up after removal of the rat. Adaptation to the animal restraint system and magazine training was readily accomplished in one session in which 50 reinforcers were delivered on a fixed-time (FT20") schedule. During this session, responses had no effect. Following this initial session, the paddle-press operant was established, using an alt FT20" FR1 schedule to deliver 50 reinforcers per day. Force required for the operant was set to a .98 N, a minimal value for the apparatus; this force was applied through a distance required to produce complete extension of the elbow joint in individual animals by software control (the typical distance was approximately 35 mm). The alt FT20" FR1 schedule was in effect until 75% of the reinforcers obtained in a session were delivered as a result of the FR1 component of the schedule. The paddle-press operant was established in a mean of 6.9 sessions, with no animal requiring more than 16 sessions. By the end of the third session, all animals readily entered the restraining system. The handler simply positioned the rat facing the open end of the restraint, allowing the rat to crawl into the restraint. All the handler had to do was center the rat's tail and insert the tail stock. Animals would not, however, voluntarily back out of the restraint at the end of their experimental sessions. Removal of the top restraining part left the rat on the horizontal animal support, where a handler could easily pick him up. This made extrication of the animals a simple task. Taken together, these observations of the voluntary entry and the need to remove the top part of the restraint to extricate the animal suggest that the restraint was not aversive and likely not stressful either; this is probably the result of the use of positive reinforcement.² Studies using aversive control with this apparatus might be expected to experience minor problems in getting subjects to enter the apparatus.

Although such a restraint system was required for our research, the advantages of the restraint should make it a useful apparatus for other studies. It would appear that mounting visual displays in the solid end of the restraint would make this restraint a valuable tool for the study of visual discrimination in the rat. The animal's eyes would always be oriented toward the display, maximiz-

ing the detection of visual stimuli. This restraint may also prove beneficial in situations in which general limitations on movement are necessary, such as physiological studies in which excessive activity could interfere with physiological recording (Hall et al., 1966).

Several restraining devices for rats have been described in the psychological literature (see Hall et al., 1966, for references). In spite of their potential advantages, these devices appear to have been used limitedly, in studies of aversive control (e.g., Azrin, Hopwood, & Powell, 1967; Dallemagne & Richelle, 1970) and in studies involving physiological recording (Hall et al., 1966). One can only speculate that problems of adapting animals to restraining systems, daily placement in and removal from the restraint, concerns that restraint systems would induce stress, and lack of a commercially available restraint system have been major factors in selection of the operant chamber over a restraining device. Our restraint appears to minimize all but the last problem. Though they were not commercially available, we were able to have five restraints constructed for less than \$100 per system (excluding the sling suit frames and liquid solenoid valves, which we already possessed). Although our animals were not examined for internal symptoms of stress, careful daily observation indicated no physical injuries, good health over the duration of the study, and consistent voluntary restraint entry after about 3 days of training. These observations strongly suggest that the restraint was not aversive and probably not more stressful than standard operant chambers.

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NOTES

1. Since the restraint is designed to be removed from the animal support, making this frame wider than the support provides a convenient handle that facilitates removal.
2. An earlier version of this restraint system was constructed with

the restraint permanently attached to the support. In a pilot study with the earlier version and a water reinforcer, removal of the rat at the end of the session became progressively more difficult, eventually requiring more time than was required for the entire experimental session. The removable restraint eliminates this problem and has the added advantage of facilitating cleaning.

Appendix A
Program Listing for alt FT20" FR1*

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10  rem shaping program--alt FT20" FR1
20  gosub 5000:rem subroutine that sets up lever position
    read
30  dr=56579:pp=56577:r=0
40  poke dr,248: rem set interface for 3 inputs & 5 outputs
50  poke pp,0: rem turns all devices off
60  print "{SC}{HM}{CR}{CR}{CR}          SHAPING PROGRAM"
70  print "{CD}{CR}{CR}{CR}{CR}INSERT DATA DISK, TURN ON 28V
    SUPPLY"
80  print "{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}
    {CD}{CR}{CR}{CR}{CR}TO ABORT PRESS RUN/STOP"
90  print "{CR}{CR}{CR}{CR}{CR}ENTER RUN 290 <RET> "
100 input "{HM}{CD}{CD}{CD}{CD}{CD}{CR}{CR}{CR}
    {CR}{CR}{CR}ENTER SESSION NUMBER"; s$
110 open 1,8,12,"0:session "+s$+"s,w":rem opens disk file
120 input"{CD}PRESS ANY KEY AND RETURN TO START";z$:rem
    waits for required keystroke and return
130 print"{HM}{CD}{CD}{CD}{CD}{CD}  SESSION IS RUNNING
    "
140 ti$="000000":rem sets internal clock to zero
150 poke pp,64:rem turns on houselight (lamp 4) and other
    devices off
160 t$=str$(ti):a$="a"+t$
170 print#1, a$:rem writes lamp event to disk
190 t1=ti
200 t2=ti
210 gosub6000: rem checks paddle position
220 if r=1 then 250
230 td=t2-t1
240 if td<1200 then 200
245 goto 260
250 r=0:gosub1000:rem response recording subroutine
260 gosub2000:rem reinforcer subroutine
270 if h=50 then 290
280 goto 150
285 rem *****
290 rem end of session shut-down
300 poke 56577,0: rem turns everything off in box
310 print "SESSION ENDED"
320 t$=str$(ti):print#1,"e"+t$
330 close 1
340 open 15,8,15
350 close 15
360 end
990 rem *****
1000 rem response subroutine
1010 poke pp,72:rem initiates response pulse to cumulative
    recorder
1015 rc=rc + 1:print"{HM}{CD}{CD}{CD}{CD}{CD}{CD}
    {CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}TOTAL RESPONSES =
    "rc
1020 t$=str$(ti):a$="b"+t$
1030 print#1, a$:rem writes response event to disk

```

Appendix A (Continued)

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1040 for i=1 to 9:next
1050 poke pp,64:rem ends response pulse
1060 return
1070 rem *****
2000 rem reinforcer subroutine
2005 t$=str$(ti):print#1,"c"+t$:rem writes reinforcer event
    to disk
2010 poke pp,208:rem turns on liquid solenoid valve for
    reinforcer; cue light on; leaves house light on
2020 for i=1 to 40:next: rem allows pulse duration to
    deliver .5ml reinforcer
2030 poke pp,80: rem turns off reinforcer valve, cue light
    continues
2040 h=h+1:rem increments reinforcer counter
2050 print"{HM}{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}{CD}
    {CD}{CR}{CR}{CR}{CR}{CR}{CR}{CR}{CR}{CR}{CR}{CR}
    REINFORCERS = " h
2060 for i=1 to 957: next: rem continues cue light on
2080 return
2090 rem *****
5000 rem initial subroutine to load machine language program
    for operandum position
5010 c=12*4096:rem set subroutine start location
5020 rem poke in the operandum position subroutine
5030 for i=0 to 63: read a: poke c+i, a:next
5040 return
5050 rem *****
5090 rem response position subroutine
6000 sysc:rem call operandum position program
6010 p1=peek(12*4096 + 257): rem response position #1 value
6015 if p=0 then 6030
6020 if p1 > 46 then p=0
6025 goto 6040
6030 if p1 < 40 then p=1:r=1;rem changes position and sets
    response flag
6040 return
6050 rem *****
7000 data162,1,120,173,2,220,141,0,193,169,192,141,
    2,220,169
7010 data128,141,0,220,160,128,234,136,16,252,173,
    25,212,157
7020 data1,193,173,26,212,157,3,193,173,0,220,9,
    128,141,5,193
7030 data169,64,202,16,222,173,0,193,141,2,220,
    173,1,220,141
7040 data6,193,88,96

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*The program is written in Commodore Basic. Special keys are enclosed in brackets {} as follows:
 {SC}=shift-CLR, {HM}=HOME, {CR}=RETURN, {CD}=CRSR down.

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