Water Deprivation Associated with Operant Conditioning Inhibits Hypertensive Disease in Young Spontaneously Hypertensive Rats

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Abstract—The development of high blood pressure in young spontaneously hypertensive rats (SHR) is inhibited by operant conditioning motivated by chronic water deprivation. The present study showed that the inhibition of hypertension was not due to the operant conditioning, per se, but was a result of chronic water deprivation (23.5 hr schedule) imposed on the rats from five through 12 weeks of age. Blood pressures were measured directly using a carotid artery cannula and light enflurane anesthesia. The behaviorally conditioned SHR and the water-deprived SHR controls had equal blood pressures and were significantly less hypertensive than untreated or enriched environment SHR groups. The antihypertensive action of water deprivation was not detected by weekly indirect blood pressure measurements made in the awake state. Nonetheless, the chronicity of the deprivation-induced inhibition of hypertension was confirmed by a lesser degree of left ventricular hypertrophy in the deprived SHR relative to the nondeprived SHR. Our behavioral results again demonstrated hyperreactivity in the SHR relative to the Wistar Kyoto (WKY). This behavioral hyperreactivity in the SHR may explain the exaggerated increase in drinking in the deprived SHR groups when returned to ad lib conditions. The data of this study and our previous work suggest that arousal differences between SHR and WKY strains are more reliably differentiated by fixed interval schedules of reinforcement than by a fixed ratio schedule. Hyperarousal may precede hypertension in the SHR, but it is undetermined whether hypertension can be found in the absence of hyperarousal in these rats.

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The spontaneously hypertensive rat (SHR) is a genetic model of hypertension developed by Okamoto and Aoki (1963). High blood pressure gradually develops in all progeny of spontaneously hypertensive rat (SHR) matings, usually by four weeks of age in the most recent generations of SHR (Lais et al., 1977).

In our earlier work (Schaefer et al., 1978) we used instrumental conditioning techniques to demonstrate behavioral hyperarousal in the SHR, first in the adult fixed hypertensive and later in the adolescent rat. Although our young
SHR displayed the same behavioral hyperreactivity as the mature SHR, hypertension was apparently prevented or delayed in the young SHR by our operant conditioning procedure.

The present study was an attempt to confirm our earlier results with the adolescent rats and to determine which of the factors involved in the conditioning process might impede the development of high blood pressure. We felt that there were three major aspects of our behavioral protocol that might have countered the hypertensive disease process. These three factors were the chronic water deprivation procedure needed to motivate bar pressing, the regular exposure of the rats to an enriched environment (the behavioral testing chamber), and the operant conditioning process, per se.

**Methods**

**Subjects**

Timed-pregnant SHR and normotensive Wistar Kyoto (WKY) rats were obtained from Charles Rivers Laboratories (Wilmington, MA). The rats were housed in a quiet room with a 12-hr light cycle. At weaning (three weeks of age) pups were selected from the litters of five SHR and four WKY mothers to make up five SHR and two WKY groups of four males and four females each. These pups were then housed by group in plastic cages until five weeks old. From five through 14 weeks of age, all rats were housed individually in suspended metal cages.

**Procedure**

The treatments that each group received are outlined in Table 1. The two SHR_D, C groups were combined as were the two SHR_N groups, since they were indistinguishable in terms of body weights, water intakes, and most importantly, directly measured blood pressures. SHR_D, C includes one group that replicated the original study as closely as possible and a similarly treated group in which we made weekly indirect blood pressure measurements, a procedure not used in our former study. SHR_N includes the untreated control group and the group exposed to the enriched environment (Skinner box).

The operant conditioning chamber was a BRS/LVE Rodent Test Cage (Beltsville, MD) enclosed in a sound attenuating chamber. The Skinner box was equipped with a single response lever, and bar presses were rewarded with 0.02 cc amounts of water presented via a dipper mechanism. The house light and the white noise masking stimulus were on continuously during all sessions.

When the rats were approximately four weeks old an auto-shaping procedure was conducted. Each group of 8 rats to be conditioned or exposed to the Skinner box was placed as a group in the box for 48 hours. Food pellets were available ad lib but water was available only through the dipper mechanism. For the first 24 hours in the box water was delivered only after a bar press except in the case of the enriched environment group where free water deliveries continued every 60 sec.

At five weeks of age all rats were housed individually and the 23.5 hr water deprivation procedure was begun. Body weights and water intakes of all animals were recorded five days per week. After the first full day of water deprivation the rats were trained individually in the Skinner box in 15 min sessions, five days per week. Manual

**Table 1. Experimental Protocol Including Group Size, Combinations, and Treatments Received**

<table>
<thead>
<tr>
<th>Group</th>
<th>Number of Subjects</th>
<th>Tail Cuff Measurements</th>
<th>Water Deprivation</th>
<th>Skinner Box Exposure</th>
<th>Behavioral Training</th>
</tr>
</thead>
<tbody>
<tr>
<td>WKY_D, C</td>
<td>8</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>SHR_D, C</td>
<td>7**</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>(N = 15)</td>
<td>8</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>SHR_N</td>
<td>8</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>(N = 16)</td>
<td>8</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>WKY_N</td>
<td>8</td>
<td></td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
</tbody>
</table>

* The subscripts D and C refer to water deprivation and behavioral conditioning, respectively, while the subscript N signifies that the animals were neither water deprived nor conditioned.
** One male rat in this group was sacrificed after developing abnormal incisors which prevented normal eating and growth.