L-Triiodothyronine and Dinitrophenol–Induced Hypertension (34176)

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Thyroxine has been shown to accelerate the production of NaCl hypertension (1, 2). Recently Dahl *et al.* (3) using L-triiodothyronine (T_3) outlined a method of accelerating production of a sustained salt hypertension in rats. In addition, inbreeding successive generations of these animals developed a large susceptibility to this type of hypertension.

The most obvious activity of T_3 is enhancement of oxidative metabolism. Dinitrophenol (DNP) also increases cellular metabolism however, without relief from the symptoms of thyroid insufficiency (4). The purpose of the present study was to determine if a sustained hypertension could also be produced with DNP, and if so to evaluate the accelerated induction and maintenance of salt hypertension using these two drugs.

Methods. Male stock Sprague-Dawley rats were used in all groups, except for female animals used when the effect of age was studied. The procedure for producing T_3 and NaCl hypertension was an adaptation of the method of Dahl *et al.* (3). Animals with blood pressures greater than 160 mm Hg were considered hypertensive. The data were analyzed for the level of significance ($p < 0.05$) by Student's t test on an unpaired basis.

T_3–NaCl hypertension. Ten weanling rats (21 days) were fed Purina laboratory chow to which had been added 8% NaCl. During each of the initial 4 days, 50 μg/rat of L-3,-3′-5-triiodothyronine Na salt (T_3) was injected subcutaneously. On alternate days thereafter for 20 days, T_3 was administered at 5 μg/rat. The animals were 45 days old when the treatment was completed and feeding with the regular laboratory diet was resumed. Tap water was given *ad libitum*. At 17 weeks of age, the animals were treated with methimazole (Tapazole) by adding 0.2% to the diet for 2 weeks (5). This dose was selected because in normotensive rats it was found to affect the appetite for NaCl (6). Blood pressures were taken in all groups before treatment and at weekly intervals thereafter by a modification of the indirect tail-cuff method (7). Results were plotted as means of each group in all studies.

Another group of 18 rats was treated in a similar manner except they were maintained on 8% NaCl throughout the course of the study. They also were fed methimazole (0.2%) in the diet for 2 weeks when 17 weeks old. One untreated group of 17-week-old rats was fed a diet containing 0.2% methimazole for 2 weeks. Another untreated group of 10 rats was given neither salt and T_3 nor methimazole, however blood pressures were taken throughout the period.

Five rats were treated by the T_3–NaCl regimen. The animals were thyroidectomized at 10 weeks of age to elucidate further the effect of the thyroid on maintenance of hypertension. Because of simultaneous parathyroidectomy, 1% calcium gluconate was added to the drinking water to maintain the tissue calcium level. An untreated control group was given injections of saline at 3–6 weeks of age and were thyroidectomized at 10 weeks of age. Blood pressures were taken from 9 through 13 weeks of age inclusive.

DNP–NaCl hypertension. Doses of α-2,-4-dinitrophenol (650 μg/rat, ip) were given to 10 weanling rats each day for the first 4 days and then on alternate days for the following 20 days. During this period 8% NaCl was given in the diet. One week after return to regular diet, the blood pressure returned to relatively normal values. The DNP–NACl regimen was then repeated over the next 24 days. Following the second administration of

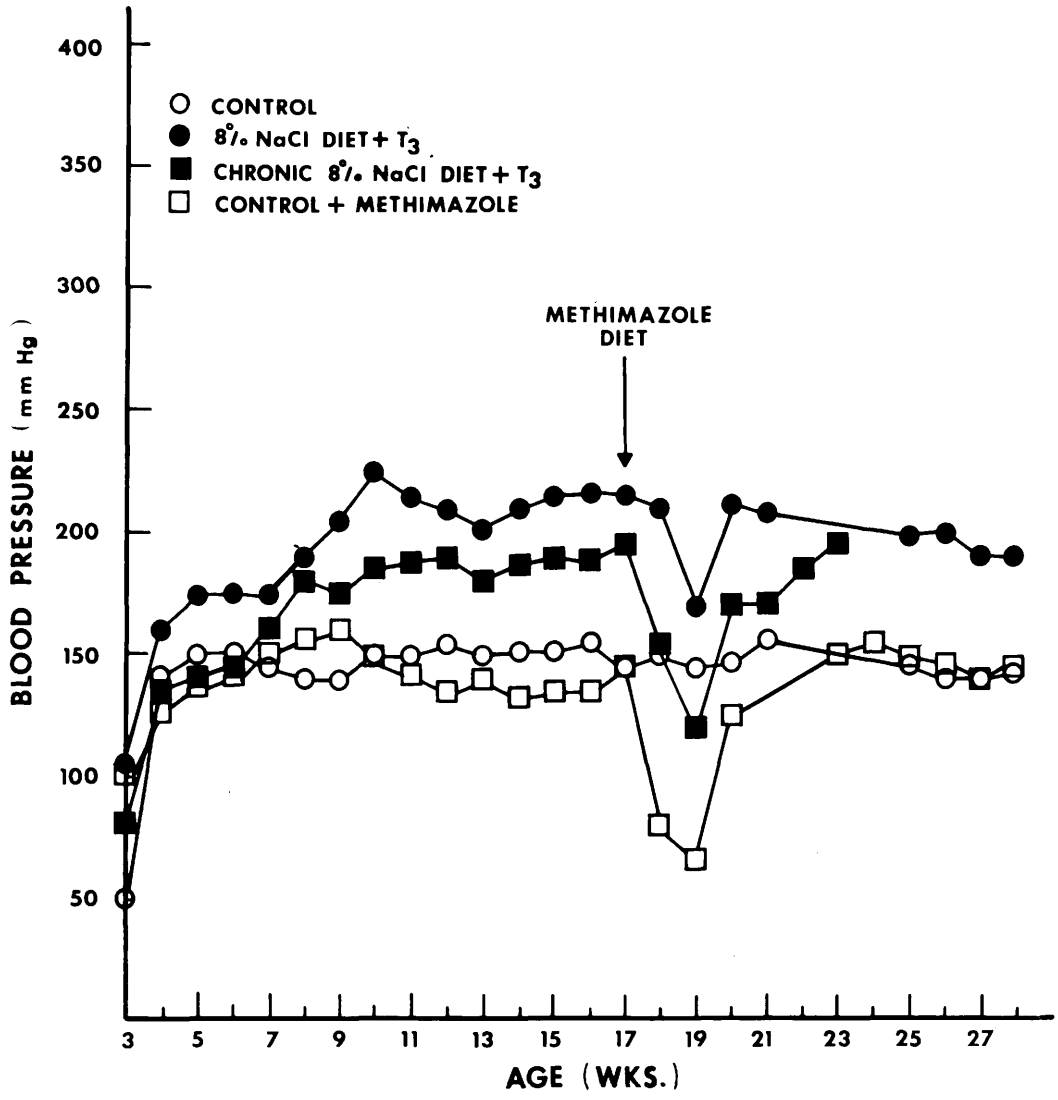

FIG. 1. Effect of L-triiodothyronine treatment along with sustained dietary salt administration (■); and 3 weeks' dietary salt administration (●) on blood pressure. Untreated (○) control rats and methimazole-treated (□) control animals. Methimazole was given to the three groups in the diet for 2 weeks starting at the age of 17 weeks.

DNP and salt, the blood pressure returned to normal. After a period of 4 weeks when the animals were 14 weeks old, T_3 and 8% NaCl were introduced to determine the susceptibility of the animals treated with DNP and salt to sustained hypertension.

In an attempt to elucidate further the mechanism whereby DNP produces hypertension, dinitrophenol and salt, DNP alone, and NaCl alone were administered to three groups of 10 weanling rats for 12 weeks. A fourth group was treated with a normal diet along with intraperitoneal injections of isotonic saline.

Effect of T_3 and NaCl on sexually mature rats. A group of 15 male and a group of 15 female rats 9 weeks of age were treated with T_3 and 8% NaCl. Blood pressures were tak-

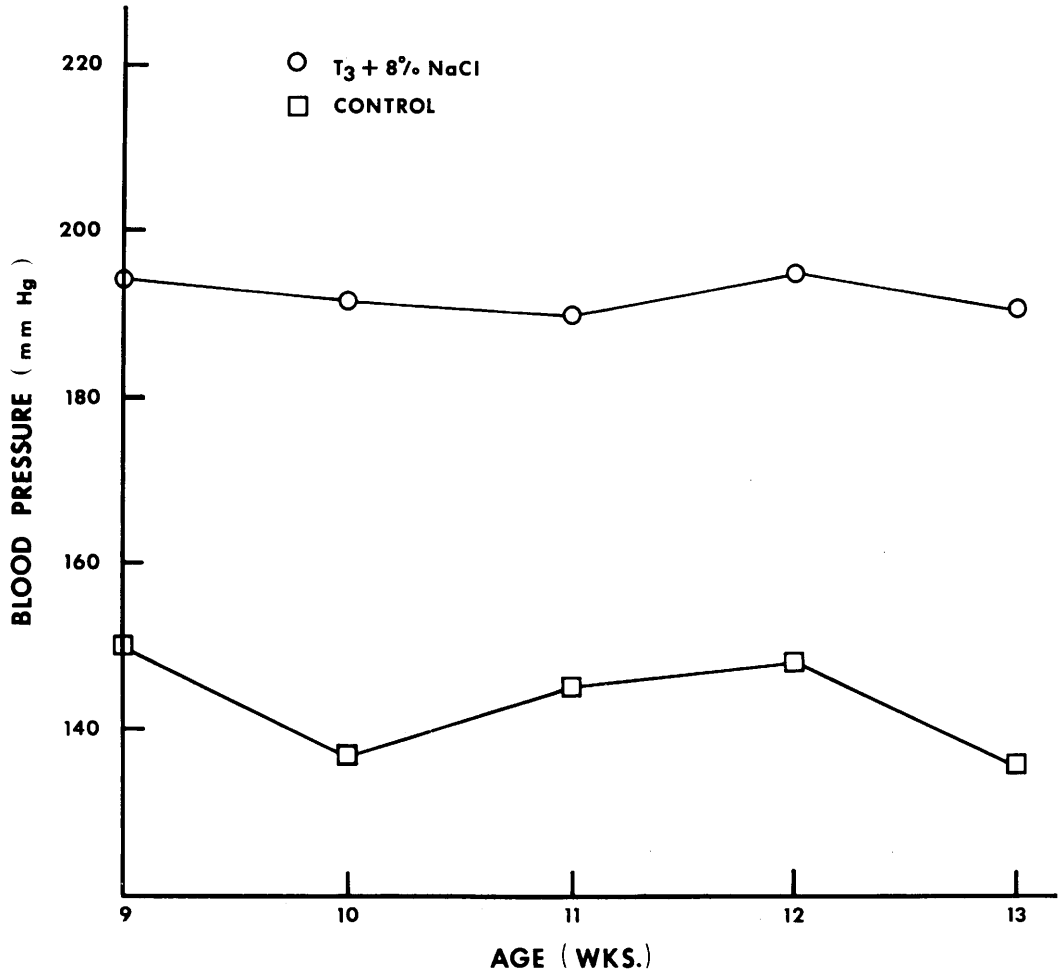

FIG. 2. Blood pressure of T_3–NaCl-treated rats (○), and control animals (□). Both groups of rats were thyroidectomized at 10 weeks of age.

en on both groups at weekly intervals over the next 10 weeks.

Results. T_3 and NaCl hypertension. L-Triiodothyronine and NaCl administered simultaneously produced a sustained hypertension, which is in agreement with the work of Dahl *et al.* (3). In the present study, all of the animals exhibited blood pressures over 160 mm Hg(Fig. 1). This was a larger percentage than previously reported for stock or first generation rats (3). Continuous treatment with NaCl following the administration of T_3 and NaCl did not cause any significant difference in hypertensive blood pressure level than in the group treated for only 3 weeks (Fig. 1).

The three groups of animals treated with methimazole had a decrease (50–80 mm Hg) in blood pressure during the treatment period (Fig. 1). A period of about 3 weeks was required for the blood pressure to return to control values. Loss of appetite was apparent in the three treated groups and was followed by weight reduction.

Removal of the thyroid at 10 weeks had no effect on blood pressure of T_3–NaCl hypertensive rats or of normal controls (Fig. 2). These data support the results obtained by thyroid inhibition produced with methimazole. Neither the thyroid nor sustained L-triiodothyronine–NaCl treatment was necessary

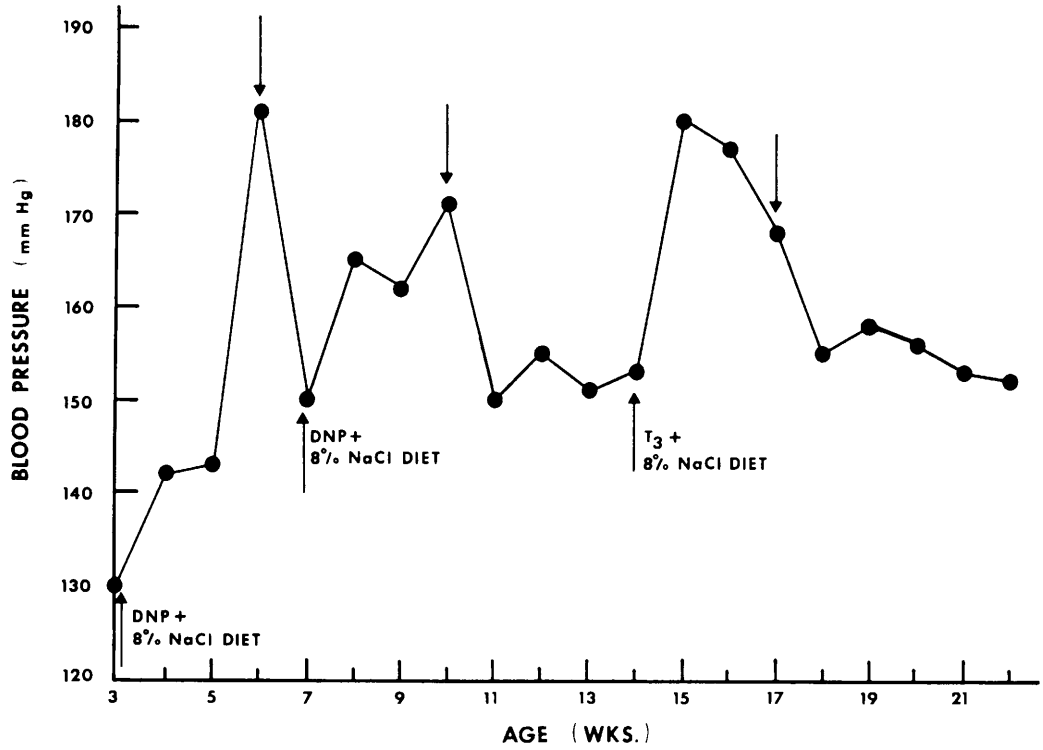

FIG. 3. Dinitrophenol and dietary salt were administered for a period of 3 weeks when the animals were 3 weeks old and again when 7 weeks old. T_3 and NaCl were given for 3 weeks when 7 weeks old; T_3 and NaCl were given for three weeks when the animals were 14 weeks old (↑ = start treatment; ↓ = stop treatment).

for the maintenance of T_3–NaCl induced hypertension.

DNP and NaCl hypertension. Dinitrophenol (650 μg/rat) and 8% NaCl ration produced a hypertensive response only during treatment (Fig. 3). When treatment was terminated, the hypertension ceased spontaneously. An elevated blood pressure was obtained when treatment was resumed. Four weeks following the final week of DNP–NaCl treatment, the T_3 and salt regimen failed to produce a sustained hypertension.

In a second study, DNP and NaCl in the diet produced hypertension that persisted with continuous treatment (Fig. 4). Following 12 weeks' chronic administration the blood pressure returned to normotensive levels 4 weeks after cessation of treatment.

The continuous feeding of weanling rats with a diet containing 8% NaCl alone resulted in hypertension by the age of 9 weeks. There was a latent period before salt hypertension occurred that was not as apparent with stimultaneous DNP–NaCl administration. When the animals were removed from the diet, their blood pressures returned to normal. Dinitrophenol alone did not affect the blood pressure.

Effect of age on production of hypertension. When male and female rats, 9 weeks old, received T_3 and NaCl, only the males showed an insignificant transitory increase in blood pressure (Fig. 5). This is in contrast to weanling rats, which showed a greater susceptibility to the treatment and remained hypertensive once the regimen was terminated.

Discussion. When treated with T_3 and NaCl, all of the weanling animals developed a sustained hypertension of over 160 mm Hg by the end of the treatment period. The effective production of hypertension in this study may be explained by genetic factors

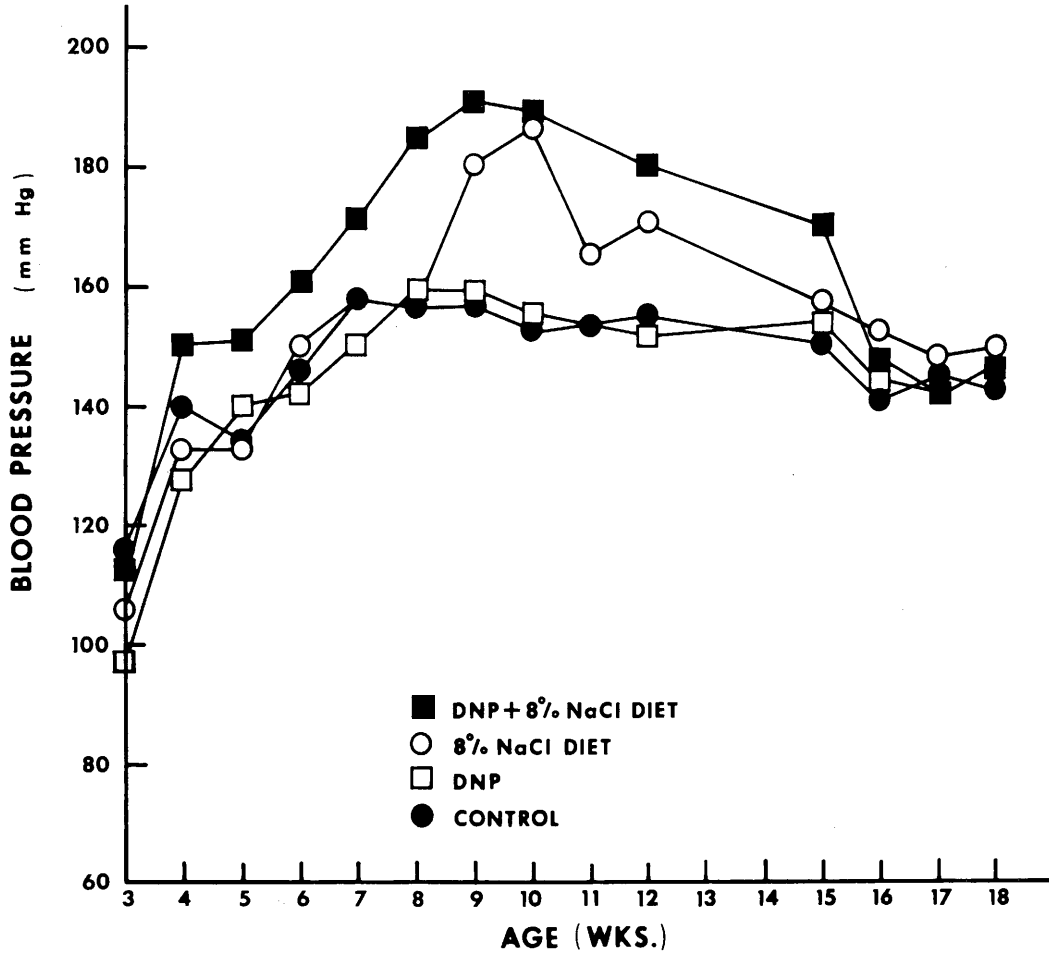

FIG. 4. Blood pressure during and following continuous administration of DNP and salt (■); salt alone (○); DNP alone (□); and in untreated control animals (●).

which contributed to a greater susceptibility to hypertension (3). The inability to enhance T_3 + NaCl hypertension with continued NaCl in the diet suggests that T_3 modified a specific mechanism which resulted in production and maintenance of elevated blood pressure. Once this modification took place, it was not spontaneously reversible. This alteration could have been related to the tissue NaCl concentration for it has been shown that the NaCl concentration in the aorta is elevated during the sustained phase of T_3–NaCl hypertension (8).

Aoki (9) reported that goitrogens inhibited the induction and maintenance of elevated blood pressure in spontaneously hypertensive rats. In the present study, methimazole temporarily inhibited the sustained phase of hypertension, but did not produce a prolonged normotensive blood pressure. A concurrent reduction in weight was observed as previously reported by McCarthy *et al.* (10), which may be the cause of the transient decrease in blood pressure. However, sexually mature T_3–NaCl hypertensive rats did not respond with a return to normotensive blood pressure following thyroidectomy. Thus, the intact thyroid was not necessary for the maintenance of T_3–NaCl-induced hypertension following cessation of treatment. This would seem to indicate that a thyroxine imbalance was not responsible for maintaining the sustained hypertensive phase.

Dinitrophenol with a high-salt diet pro-

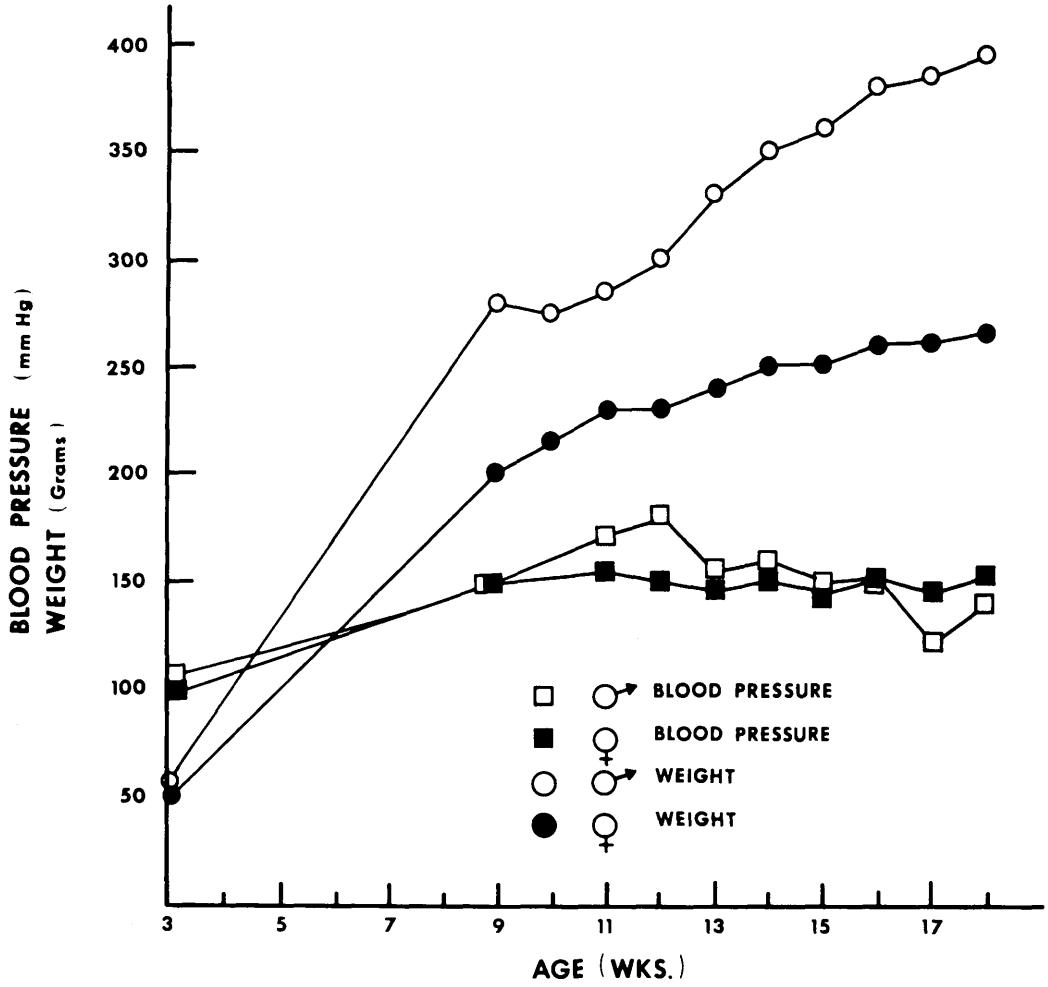

FIG. 5. Blood pressure and body weight response during and following the 3-week regimen of T_3 and NaCl in male and female rats starting at 9 weeks of age.

duces hypertension, which is dependent upon continuous treatment for maintenance of the elevated blood pressure. Also DNP does accelerate the production of hypertension with salt as does T_3. Since DNP and NaCl hypertension is spontaneously reversible, T_3 and DNP are probably not affecting the same mechanism to facilitate the induction of elevated blood pressure.

Summary. The data support previous results showing that L-triiodothyronine (T_3) accelerates the production of sodium chloride hypertension. Dinitrophenol (DNP) with NaCl produces hypertension rapidly, however, it did not result in a sustained elevation of blood pressure. The ability to produce hypertension with T_3–NaCl was dependent upon the use of weanling animals, whereas it was not for DNP–NaCl hypertension. These disparities in results suggest that the two drugs probably aided the induction of hypertension by different mechanisms. In addition, temporary reversal of hypertension with methimazole and lack of effect on hypertension after thyroidectomy indicate the thyroid is not necessary for the maintenance of T_3–NaCl hypertension.

Acute hypertension that resulted from DNP and salt was not dependent upon the age of the animals. A second administration of the two materials starting at 7 weeks of age produced a similar response as when given to weanling

rats. The T_3 hypertension could not be produced in the same group of rats at the age of 14 weeks, which suggested that age was a factor in T_3–NaCl hypertension.

In the study designed to evaluate the effect of age on the ability to produce hypertension with T_3 and salt, rats 9 weeks of age failed to become irreversibly hypertensive. This supports a previous report that sexually immature rats must be used in order to produce hypertension with T_3–NaCl treatment (3). This suggests that T_3 affects an underdeveloped system in the weanling rat. In contrast DNP–NaCl hypertension can be produced in sexually mature animals, which is additional evidence that T_3 and DNP affect different mechanisms in the accelerated production of salt hypertension.

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