

EFFECTS OF CHRONIC EXCESS SALT FEEDING*
ENHANCED HYPERTENSOGENIC EFFECT OF SEA SALT OVER SODIUM
CHLORIDE

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Experimental hypertension can be induced in the chick (1), rabbit (2), and rat (3, 4) by feeding excess sodium chloride. A comparable role of dietary salt in the etiology of human hypertension is less clearly established, but evidence has been brought forward (5-8) to support this possibility.

All these studies have emphasized the importance of the sodium ion to the exclusion of other factors. However, the salt actually consumed by various peoples is often quite impure. For example, most of the table salt in Japan is derived by processing sea water, and usually contains a mixture of substances (9). Hypertension is common among the Japanese (7, 10) and average salt consumption is high (10). It is thus possible that a hypertensogenic effect of sodium might be increased by additional elements in the table salt.

The present study was undertaken to explore this possibility. Rats were fed high salt diets for about 1 year. One group was fed a diet containing chemically pure sodium chloride, while the other consumed a food with approximately the same amount of sodium chloride in a mixed salt derived by evaporating sea water. After 1 year on these regimens, the animals on "sea salt" had significantly higher blood pressures than those consuming sodium chloride alone. This suggests that some factors in sea salt can potentiate the hypertensogenic effect of sodium chloride.

EXPERIMENTAL

Animals; Care; Food.—All animals were Sprague-Dawley females. "Sea Salt Colony I" consisted of 37 animals, of which 25 were salt-fed and 12 were controls; "Sea Salt Colony II" had 76 animals, of which 12 were controls.

Animals were kept in air-conditioned rooms with from 2 to 6 animals per cage with free access to drinking (tap) water and food. Since analyses of the tap water revealed it to have only 0.5 to 0.7 m.eq. of sodium per liter, this small source of sodium has been disregarded in our calculations. Control animals received unmodified Ralston purina fox chow pellets, which contained 0.50 to 0.75 per cent NaCl by analysis (11).

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The salt-fed animals received the same chow to which one of the two salt supplements was added by the manufacturer. In one diet ("8 per cent NaCl food") the supplement was c.p. sodium chloride; our analyses of the pellets showed an average of 8.09 per cent NaCl. The other diet ("sea salt food") had 11.6 per cent "sea salt" added, an amount which had been calculated to give a final sodium concentration equal to that in the 8 per cent food; our analyses, however, showed this food to contain an average of only 7.28 per cent NaCl. The manufacturer of this special sea salt stated that it was obtained by evaporating sea water and drying at an air temperature of 1100° F.¹ A comparison of the composition from data obtained by analysis at Brookhaven² with that received from the manufacturer is summarized in Table I. No significant differences are apparent between the two analyses for the ions present in larger concentrations, but differences were found among some of the others; strontium, copper, lithium, manganese, molybdenum, nickel, zirconium, silicon, and tin, were present in significantly higher concentrations according to the Brookhaven analysis, whereas boron, aluminum, and fluorine were significantly lower than the corresponding analyses supplied by the manufacturer.

Regimens.—The 25 salt-fed members of Sea Salt Colony I received the unmodified chow from weaning to the age of 2½ months, when they were started on the food containing sea salt. This feeding was continued throughout the ensuing 14 months, at the end of which period 18 animals remained alive. Sea Salt Colony II was started 11 months after Sea Salt Colony I. Of the 64 salt-fed members of this colony 36 were fed the sea salt food and 28 the 8 per cent NaCl food beginning at 1 month of age and thereafter for 13 months without change. At this time, 27 sea salt and 19 NaCl-fed animals remained alive.

Food Consumption and Weights.—Food consumption of 6 to 20 animals from each group was measured for 3 to 30 consecutive days (most commonly about 20 animals for 4 successive days). In order to minimize variation, different cages of animals on the same regimen were studied from month to month. The weight of powdered pellets, offered in special cups, consumed during a 24 hour interval constituted the measurement. This was expressed as grams per rat per day.

All animals were weighed at the time blood pressure measurements were taken, usually at monthly intervals.

Blood Pressure: Measurement and Values.—The method of Friedman and Freed (12) was used. Measurements were carried out in the air-conditioned room in which the animals were housed with the animals lightly anesthetized with a flowing oxygen-ether mixture and held in a special box thermostatically controlled at 38°C. A minimum of 4 systolic readings was made and the average recorded.

This technique has been used in our laboratory for several years with highly reproducible readings. Based on our experience with nearly 100 control animals the blood pressures of which were measured at successive intervals, a "control colony standard blood pressure" for ages in months has been drawn up. Although this standard did not eliminate the need for a matched set of control animals, it did provide an additional comparison for both the salt-fed and control animals in the current experiments. Successive monthly measurements on control animals up to 21 months of age in the standard series and in the control groups of the present study have shown no animal with systolic pressures consistently in excess of 130 mm. Hg. Therefore, we regard a systolic pressure regularly in excess of 140 mm. Hg as "hypertension." A value of 180 mm. Hg, which exceeds the control mean systolic pressure by almost 5 standard devia-

¹ The authors are indebted to the Trace Elements Corporation, Houston, for sufficient sea salt ("Admiral Brand Trace Element Sea Salt") to initiate these experiments.

² We wish to thank the members of the staff of the Hot Lab at Brookhaven who carried out both the wet and spectrographic analyses: Dr. H. L. Finston, D. Leahy, E. Selleck, R. Wilson, and S. J. Tassinari.

TABLE I
Analysis of Sea Salt

	Analysis by supplier*	Analysis by ourselves
Chlorine	50.5 per cent	48.2 per cent
Sodium	27.5 per cent	28.1 per cent
Magnesium	3.4 per cent	3.03 per cent
Carbon	2.7 per cent	N.D.†
Sulfur	1.83 per cent	N.D.
Potassium	1.13 per cent	1.0 per cent
Calcium	0.86 per cent	2.0 per cent
Bromine	1580 p.p.m.§	N.D.
Strontium	220 "	1000 p.p.m.§
Iron	100 "	100-200 "
Boron	97 "	± "
Aluminum	55 "	± "
Fluorine	35 "	<5 "
Copper	20 "	500 "
Rubidium	5 "	± "
Lithium	3 "	50 "
Iodine	1.5 "	<5 "
Barium	1.5 "	± "
Nitrogen	0.3-20 "	± "
Arsenic	0.3-0.6 "	± "
Phosphorus	0.3 "	<5 "
Zinc	0.15 "	± "
Manganese	0.13-0.3 "	50-100 "
Lead	0.11 "	± "
Selenium	0.11 "	± "
Cobalt	0.001-0.1 "	± "
Cesium	0.06 "	± "
Uranium	0.04 "	± "
Molybdenum	0.013 "	100 "
Thorium	0.013 "	± "
Silver	0.009 "	± "
Vanadium	0.009 "	± "
Lanthanum	0.009 "	± "
Yttrium	0.009 "	± "
Nickel	0.003 "	80-100 "
Scandium	0.0012 "	± "
Mercury	0.0009 "	± "
Gold	0.0002 "	± "
Zirconium	N.R.¶	300 "
Silicon	N.R.	300 "
Tin	N.R.	300 "

* Approximate analysis supplied by Trace Elements Corp., Houston, Texas.

† N.D., not done.

§ p.p.m., parts per million.

|| ±, none detected, or too low to measure by spectrographic analysis.

¶ N.R., not reported.

tions, has been associated with drastically shortened life expectancy in the present and previous similar studies.

As a rule the blood pressures were recorded at monthly intervals. For technical reasons, no measurements were made on Sea Salt Colony I from the 5th through the 8th month of age.

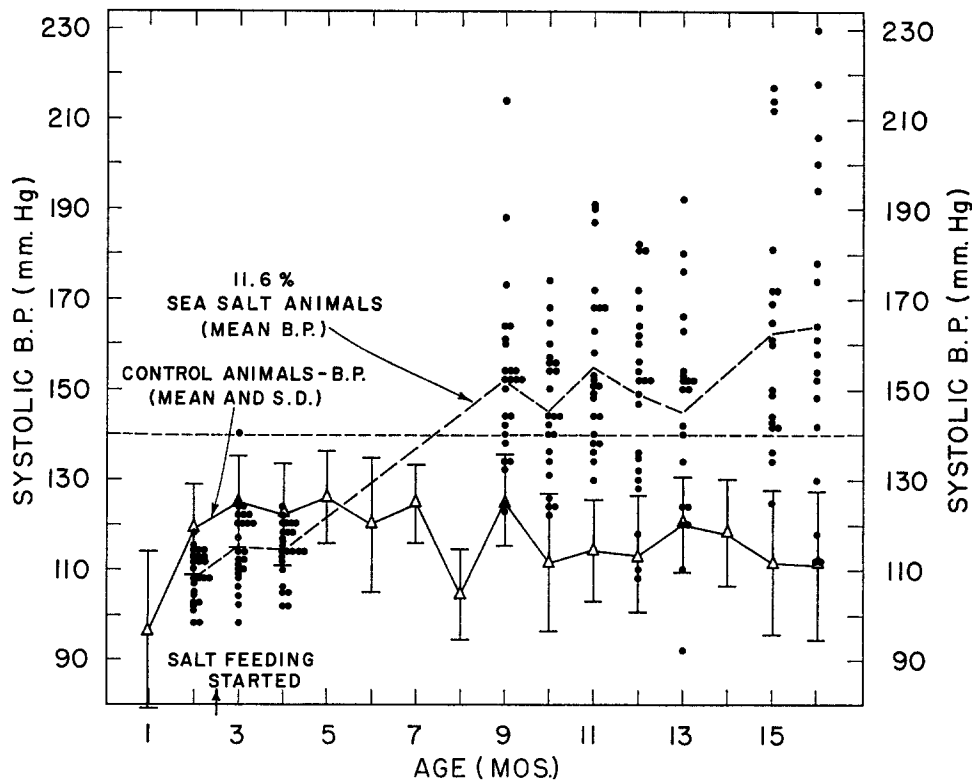


FIG. 1. Effect of 11.6 per cent sea salt food on blood pressure of female rats in Sea Salt Colony I. Dashed line through 140 mm. Hg indicates level at which "hypertension" is considered to begin. No measurements on these salt-fed animals were made from the 5th through the 8th months, for technical reasons.

Respiratory tract infections occasionally occurred in individual animals and under such circumstances, measurement of blood pressure was omitted for that month.

RESULTS

Sea Salt Colony I (Fig. 1).—The hypertensogenic potential of the sea salt food is evident from Fig. 1. From our previous experience, with similar animals on NaCl feeding, the incidence and degree of hypertension seemed significantly greater in this group that had been fed sea salt. A more detailed study was therefore made with a second group of animals.

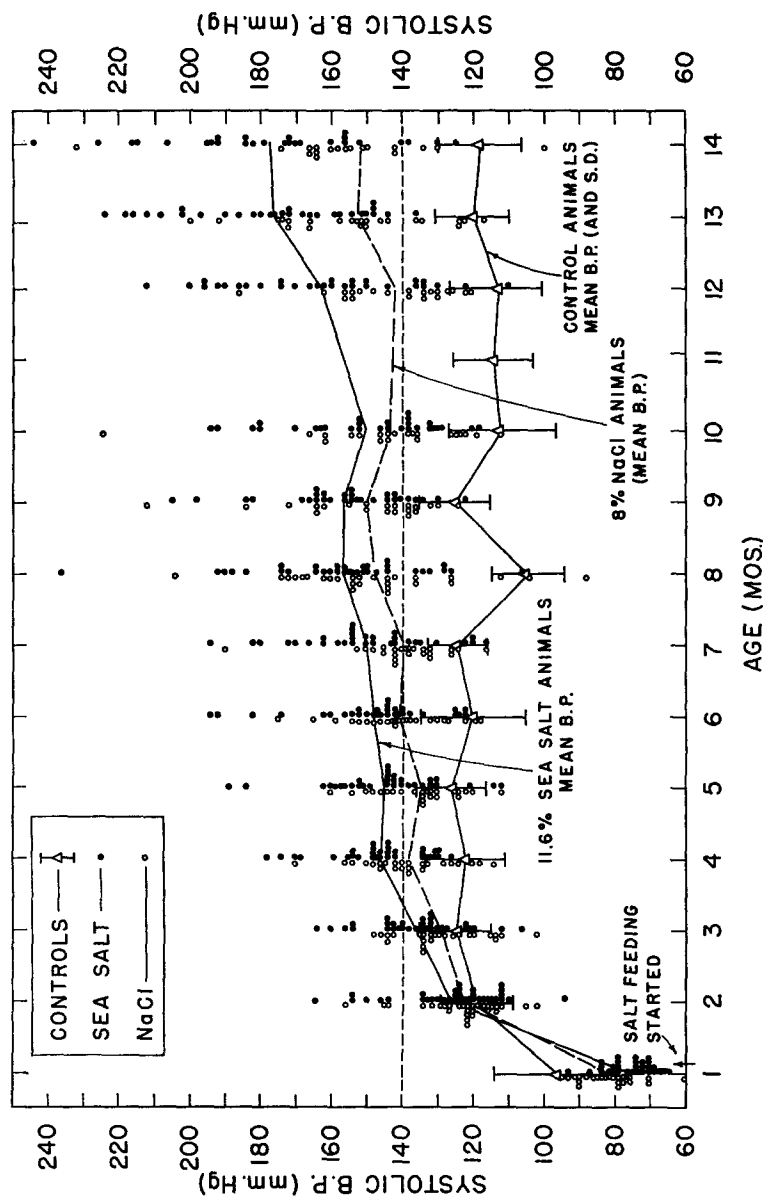


FIG. 2. Effect of 11.6 per cent sea salt *versus* 8 per cent NaCl food on blood pressure in Sea Salt Colony II. For dashed line at 140 mm. Hg, see Fig. 1. By "Student's" *t* test, the mean blood pressure at 12, 13, and 14 months was significantly higher in the group fed sea salt (at 12 months, *t* = 3.16, *p* < 0.01; at 13 months, *t* = 3.45, *p* < 0.01; and at 14 months, *t* = 2.49, 0.05 > *p* > 0.01).

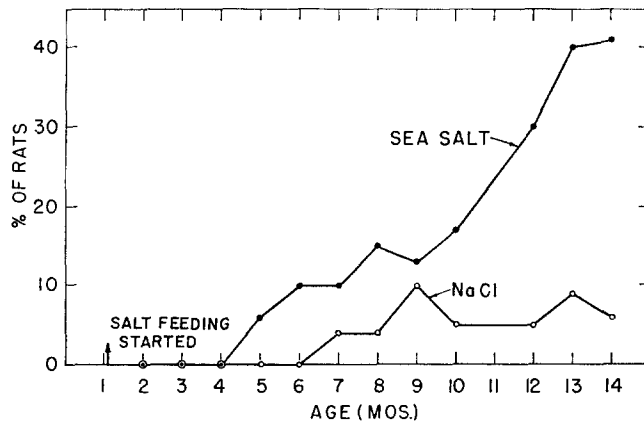


FIG. 3. Per cent of rats with systolic blood pressures in excess of 180 mm. Hg. Animals were given food containing either 11.6 per cent sea salt or 8 per cent NaCl. Sea Salt Colony II only. By the chi-square technique, the prevalence of this degree of hypertension was significantly greater in animals fed sea salt at 12, 13, and 14 months (at 12 months $\chi^2 = 4.69$, $0.05 > p > 0.01$; at 13 months $\chi^2 = 6.68$, $p < 0.01$; at 14 months $\chi^2 = 7.96$, $p < 0.01$).

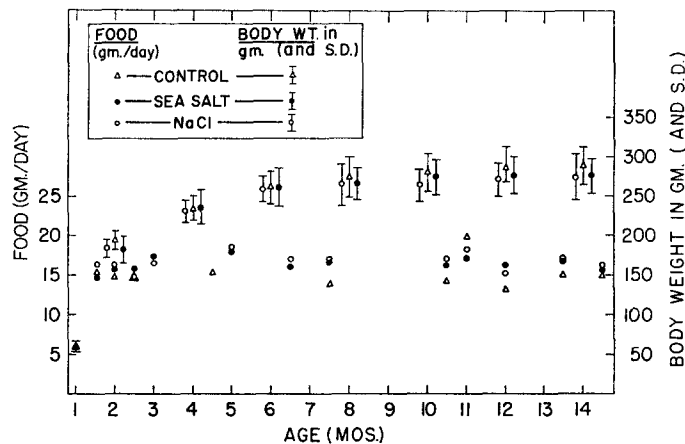


FIG. 4. Average daily food consumption and body weights on salt-feeding regimens compared with similar items for control animals.

Sea Salt Colony II (Fig. 2).—The mean pressure of the group fed sea salt was consistently higher than that of the NaCl group. The blood pressure differences became highly significant after 12 months ($p < 0.01$) and remained significantly different until the completion of the experiment after 14 months of salt-feeding (13 months, $p < 0.01$); (14 months, $0.05 > p > 0.01$). The incidence of systolic blood pressures in excess of 180 mm. Hg became particularly striking in the group on sea salt (Fig. 3). After 1 year on the regimen,

about 30 to 40 per cent of the animals on sea salt had pressures above 180 in contrast to only 5 to 10 per cent of those given pure NaCl. The incidence of this severe hypertension was significantly greater in the group fed sea salt (at 12 months $\chi^2 = 4.69$, $0.05 > p > 0.01$; at 13 months $\chi^2 = 6.68$, $p < 0.01$; and at 14 months $\chi^2 = 7.96$, $p < 0.01$).

Average Weights and Food Consumption (Fig. 4).—The higher incidence and greater severity of hypertension in animals on the sea salt regimen were not due to a greater consumption of sodium chloride. Indeed, the quantities of food consumed by both salt-fed groups were almost identical, while the content of NaCl in the sea salt food was somewhat less (7.3 per cent) than in the pure NaCl group (8.1 per cent).

DISCUSSION

The important role of dietary sodium has been demonstrated in experimental hypertension associated with desoxycorticosterone acetate administration (13), adrenal regeneration (14), and salt feeding alone (1–4). The present studies suggest the intriguing possibility that other ions may add to the severity of hypertension induced by excess dietary sodium.

A possible role of other cations in hypertension has been suggested by a number of previous investigations. Schroeder, Perry, and their associates (15–19) noted that, except for the ganglionic blocking agents, all known anti-hypertensive agents had “an affinity for certain trace metals” (15). They postulated that the hypotensive action might be due to this metal-binding capacity. Measurements of the urinary excretion and tissue concentrations of certain trace metals in people with and without hypertension in several geographic areas supported this hypothesis. Among the various elements, vanadium, cadmium, lead, and manganese appeared to be the most suspect in human hypertension. Comparing the mortality rates in the United States from cardiovascular diseases (including hypertension) with hardness of public water Schroeder concluded that something “associated with the nature of public water supplies affects adversely the course of degenerative cardiovascular disease in the United States” (19).

Orbison, Christian, and Peters (20, 21) studied changes in blood pressure and vascular lesions in bilaterally nephrectomized dogs given solutions of NaCl or modifications of Locke's solution intraperitoneally; the complex cation-anion mixtures in the latter were more effective in producing both hypertension and vascular disease than was the simple NaCl solution. In the clinical and experimental studies of Martini and Kaiser (22) and Kaiser (23) both sodium and chloride appeared necessary for the pressor effect. The need for chloride in addition to sodium has, however, not been observed by ourselves or others (22, Discussion).

The present studies do not indicate which ion or ions may be involved in

the enhancement of the hypertensive process in rats. Comparison of sea salt used in the present study with the Locke's solution that enhanced renoprival hypertension (20, 21) shows the following ions in both: Na, K, Ca, Mg, Cl, and P. The data of Meneely and Ball (24) demonstrated that feeding of potassium chloride diminished the hypertensogenic effect of sodium chloride in rats; it therefore seems unlikely that these ions enhanced the hypertension of the sea salt animals. In the cardiac necrosis produced by Selye in rats injected with desoxycorticosterone and certain sodium salts (25), the phosphate salt of sodium as well as some calcium salts caused more severe lesions, whereas magnesium chloride had a protective effect. Therefore it is possible that the presence of calcium and phosphorus (as the phosphate) in sea salt contributed to its increased toxicity in the present studies. The four trace elements held most suspect by Schroeder *et al.* were present in very low concentrations in the sea salt of the present study (Table I) but in the absence of evidence to the contrary, it remains possible that these trace elements could have enhanced the hypertension.

Subtle renal damage by sea salt cannot be excluded as a cause of the increased hypertension. However, this possibility seems unlikely since blood urea nitrogen determinations, made periodically, remained with rare exceptions within normal limits. The histological and histochemical studies of the kidney, which have been made thus far during these two salt-feeding studies, have not revealed significant differences (26).

SUMMARY AND CONCLUSIONS

Female rats were fed diets containing either excess sea salt or excess sodium chloride for periods up to 14 months. The hypertension produced by sea salt was more pronounced than that caused by sodium chloride alone, although the average amount of sodium chloride contained in the sea salt feeding was slightly less. The ions involved in this incremental effect of sea salt were not identified.

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