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FOOD RESTRICTION INCREASES LIFE SPAN OF HYPERTENSIVE ANIMALS

Tom Lloyd

The Milton S. Hershey Medical Center
The Pennsylvania State University
P. O. Box 850
Hershey, Pennsylvania 17033

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Summary

Food restriction was used to increase the life span of normotensive (WKY) and Spontaneously Hypertensive Rats (SHR). When SHR's were maintained on 40% of an otherwise typical lab rat diet, their mean life spans increased from 18 months to over 30 months. The mean life times of normotensive rats which were similarly food restricted were expanded from 24 months to over 32 months. Histological examination of heart, adrenals, kidneys and brain showed that freely fed hypertensive rats died of end-organ damage associated with high blood pressure. In contrast, deaths of food restricted hypertensive rats appeared due to changes associated with old age, rather than specific lesions due to hypertension. Thus, food restriction allows a genetically hypertensive animal to reach a normal life span and to die of age-related rather than hypertension-related events.

Caloric restriction is a well established technique for increasing the life span of several inbred strains of rats and mice (1-5). The Spontaneously Hypertensive Rat (SHR) has been thought to have a significantly shorter life span than its normotensive progenitor, the Wistar Kyoto Rat (WKY) (6). The present study was designed to examine the effect of food restriction upon life span in the SHR. The SHR/WKY model system was chosen since the development and maintenance of hypertension in the SHR closely resemble the course of essential hypertension in humans (6,8). We previously documented that neither food restriction nor modest changes in salt intake significantly affected development or maintenance of hypertension in SHR's (7).

Methods

Since sodium intake is known to influence hypertension in SHR, we established dietary regimens to examine the effect of food restriction and variation of salt intake on life span. A typical mature (500 g) lab rat consumes 15 to 22 g per day of commercial rat chow containing 0.75% sodium chloride by weight. We therefore constructed the food regimens used in this study on a "normal" daily sodium intake for rats of about 0.45 mEq/day/100 g animal weight. This level of daily sodium intake is comparable to the usual daily ingestion of 10-12 g NaCl in typical American diets (9).

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Three specialized diets which contained 0.3, 0.75 or 1.9% sodium chloride were prepared by Bio-Serve, Inc., Frenchtown, NJ, 08825. Each diet contained normal ratios of protein (24.1%), carbohydrate (55.5%), fiber (5%), fats (5.5%). All diets contained the same standard vitamin supplementation (Vitamin Mix 740) and were manufactured as pellets which weighed 3.5 to 4.0 g each. Each experimental group contained 12 control normotensive (WKY) and 12 hypertensive (SHR) 30 day old male rats which were obtained from Charles River Labs. All animals were maintained in a controlled environment (23° C, 50% humidity, alternate cycles of 12 hr. light and 12 hr. dark), fed the assigned diet once daily and given free access to water. Animals on restricted diets received 7.0 g/day and those on freely fed diets received 16 g/day. This amount of food insured near maximal growth and complete consumption of food each day.

All animals were individually housed for 9 months during the first year of the study and group housed (6/cage) during 3 months of the first year. After the first year of the study all animals were group housed (4-6/cage) although separated by dietary regimen and strain (WKY vs SHR). It was possible to continue the restricted feeding with group housing by delivering one pellet (3.5 g) per rat initially, waiting approximately 20 minutes, and then feeding the second 3.5 g pellet per rat. This technique avoided hoarding or unequal food consumption among the food restricted animals. The group housed, freely fed rats did not hoard either, as determined by their behavior and weight gains. Blood pressure measurements were made weekly on animals from each dietary group(10,11). Microscopic histopathological examinations were performed by Alexander H. Walsh, DVM, Ph.D., Diplomate, American College of Veterinary Pathologists, Ariconn Corporation, Killingworth, CT 06417. Dr. Walsh was unaware of the food or salt history of all animals.

Results

The four dietary regimens, daily sodium intake, and mean individual weights of the animals after one year of study are presented in Table 1. Animals on food regimens of 16 g/day continued to grow, whereas those on 7 g food/day remained in the 200 g range after reaching it in their sixth month of life.

TABLE I: Dietary Groups and Mean Body Weights After One Year

Dietary Group	Food g/day	Sodium mEq/Day/100g	Weight at one year in Grams ± S.D.	
			WKY	SHR
1. Freely Fed/Low Salt	16	0.20	410 ± 27	426 ± 22
2. Freely Fed/Normal Salt	16	0.54	373 ± 16	394 ± 20
3. Restricted Food/Normal Salt	7	0.43	222 ± 8	204 ± 3
4. Restricted Food/Elevated Salt	7	1.15	211 ± 17	195 ± 8
N = 12 per group				

The present report provides exact data on abbreviation of life span of the freely fed SHR. Survival curves for the four dietary regimens are presented in Fig. 1. The shape of the survival curves for freely fed WKY and SHR animals (Groups 1 and 2) is typical of previously published survival curves for inbred, similarly maintained animals (12,13). On both low salt or normal salt with normal calories, the mean life time of the SHR (18-19 months) was approximately 33% shorter than the mean life time of its normotensive progenitor, the WKY (23-26 months). When calories were restricted to about 40%

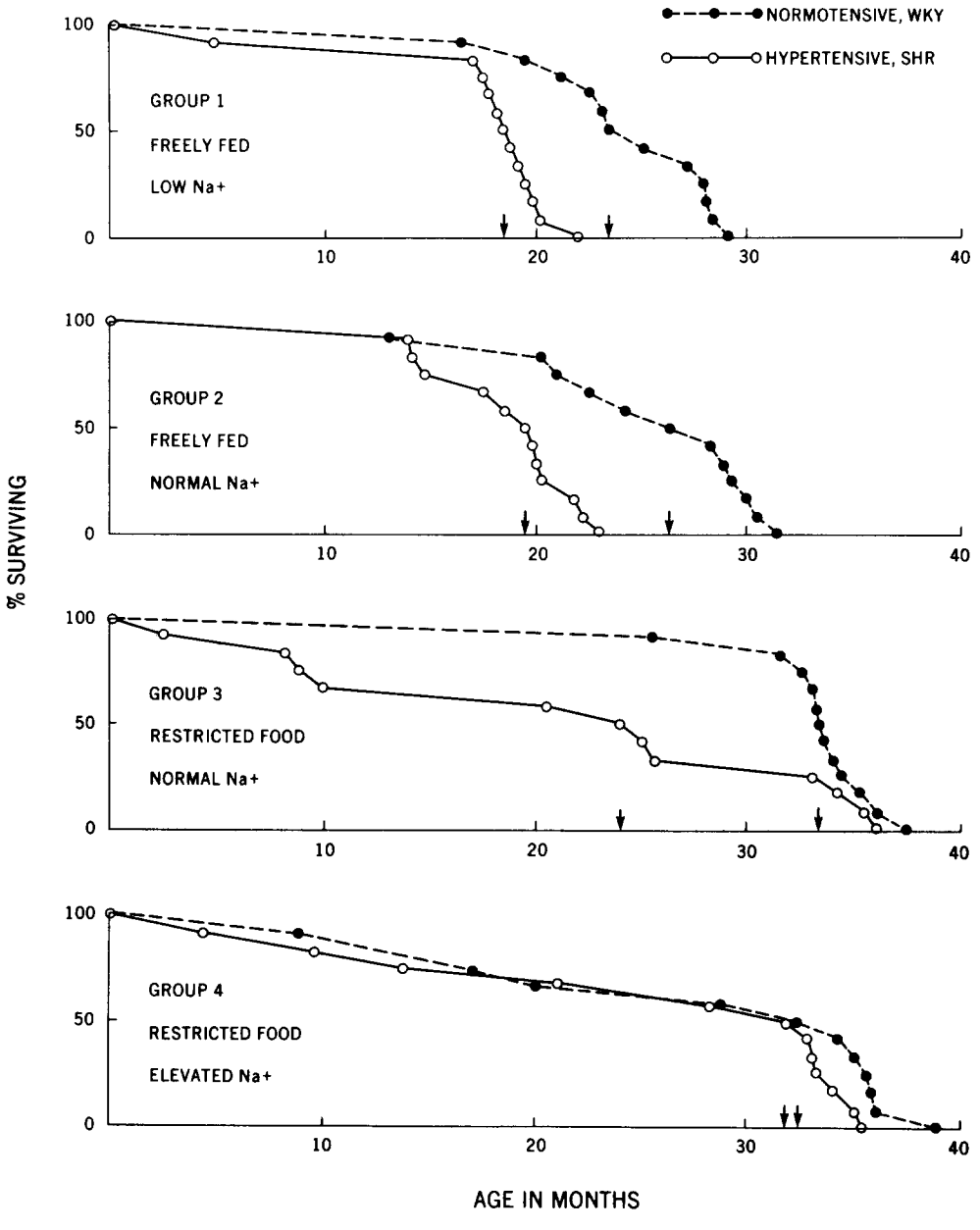

FIG. 1

Survival curves of freely fed and food restricted SHR and WKYs. n = 12 SHR and 12 WKY in each group. The arrows indicate the mean life time for each strain in each group.

TABLE II: Lesion Frequency in Kidney, Heart, Adrenal and Brain Tissue of SHRs and WKYs

Tissue	Lesion	Incidence, % Freely Fed WKY n = 16	Freely Fed SHR n = 18		Restricted SHR n = 15		Restricted WKY n = 15
Kidney							
	Dilatation (Tubules)	94	100	**	47	ns	80
	Hypertrophy/Hyperplasia	--	89	**	40	**	--
	Glomerulosclerosis	94	100	**	33	ns	67
	Fibrosis (Interstitial)	69	67	**	13	ns	27
Heart							
	Hypertrophy/Hyperplasia	--	17	ns	27	*	--
	Edema (Myocardium)	13	50	*	13	ns	--
	Fatty Infiltration (Myocardium)	--	56	**	--	ns	--
	Fibrosis (Myocardium)	63	28	ns	33	**	80
	Thrombosis (Atrium)	6	56	**	--	ns	--
Adrenal							
	Congestion	6	67	**	13	ns	27
	Pigmentation (Zona Fasciculata)	56	33	ns	13	*	53
	Vacuolization (Zona Fasciculata)	44	67	**	13	*	47
Brain							
	Spongiosis	19	22	*	--	ns	7

Data were compared by chi square contingency tables using the Apple II Stats Plus program. Levels of confidence are as follows: **, $p < .01$; *, $p < .05$; ns, $p > 0.20$ and the symbols are placed between the groups which were compared. Autopsies were performed on all animals after their natural deaths, except carcasses partially destroyed by substantial autolysis or by other animals. Tissue specimens were collected from 64 of the 96 animals involved in the study. Heart, kidneys, adrenals and brain were removed at death, fixed in phosphate buffered (pH 7.0) 10% formalin and examined by a veterinary pathologist for gross and microscopic pathologic lesions after appropriate sectioning, fixing and staining (13,14). The incidence and distribution of the following lesions was not statistically significant between any of the groups. The number of lesions seen in each of the four classes: Freely Fed WKY, Freely Fed SHR, Restricted SHR and Restricted WKY is reported in the parentheses following each lesion. Within kidney tissue: pigmentation of macrophages (2,4,1,0); infarction (0,1,0,0); pigmentation of tubules (2,0,0,2); pelvic calculi (0,0,0,1). Within heart tissue: epicarditis/pericarditis (0,1,1,0); degeneration of the myocardium (2,0,0,2). Within adrenal tissue: telangectasia (4,2,1,5); hypertrophy/hyperplasia (0,2,2,0); fibrinoid arterial necrosis (0,2,1,0); medullary adenoma, pheochromacytoma (4,2,1,2); nodular hyperplasia in the cortex (6,1,4,5); degeneration of the zona fasciculata (0,0,0,1). Within brain tissue: hypertrophy/hyperplasia (0,2,0,0).

of ad libitum feeding, a level which has been most frequently used in previous studies (1-5), we observed significantly longer mean life spans in both normotensive and hypertensive animals on either normal or elevated salt intake. The mean life time of the food restricted normotensive WKY's was lengthened by about 40% (to 32-33 months) and that of the food restricted hypertensive SHR's by 33-68% (to 24-32 months). Of particular importance is the fact that a subpopulation of the food restricted hypertensives lived as long as the food restricted normotensive rats. In addition, three of the four food restricted subgroups had peculiar survival curves, which suggested bi- or tri-modal populations. For example, the food restricted normotensive rats given normal salt (Group 3) did not experience deaths in their group until about the 32nd month, whereas 50% of their normotensive littermates who were food restricted (Group 4) but who also received a slightly elevated sodium intake were dead by the 32nd month.

A summary of autopsy findings is presented in Table 2. Certain lesions characteristic of aged experimental animals occurred commonly in all groups, e.g. dilatation of renal tubules, renal glomerulosclerosis, myocardial fibrosis, telangectasia and pigmentation of the adrenal zona fasiculata. The freely fed hypertensive rats had a high frequency of end organ damage often associated with hypertension, e.g. renal interstitial fibrosis, myocardial edema, fatty infiltration of the myocardium, atrial thrombosis, adrenal congestion, and vacuolization of the adrenal zona fasciculata. In contrast, the incidence of these hypertension-associated lesions in the food restricted SHR was dramatically lower, approximating the frequency seen in normotensive WKY's.

Discussion

Limiting the daily salt intake by 60% did not significantly alter life spans of either normotensive or hypertensive rats if they received normal calories (Groups 1 and 2). Thus, from these studies, it appears that neither halving nor tripling the usual daily salt intake had any major effect on expansion of life span caused by food restriction of normotensive or hypertensive rats. Unlike the Dahl rat, SHR's are thought not to be remarkably sensitive to modest changes in salt intake. We conclude from our data that a subpopulation of hypertensive animals may be sensitive to even modestly elevated salt levels ingested over a long period of time. Similarly, a recent prospective study by Holden and co-workers showed that in an unselected human population variation of dietary salt had no discernable effect upon blood pressure, but these workers also concluded that subpopulations of salt sensitive individuals are likely within the general population (19).

Other workers have shown that addition of massive amounts of salt (often 10% of total food consumed or about 7 mEqNa/day/100 g) to the diets of SHR's increases both the rate of development and magnitude of hypertension (14-18). Since that level of sodium intake is 3-4 fold greater than the highest human consumption, those studies bear little relationship to human salt intake, and the present study was designed to cover the likely normal range of salt consumption in Western culture. On one extreme; salt intake was reduced to 40% of the usual level, equivalent to the lowest palatable sodium restricted diet for humans. On the other extreme, salt intake was nearly tripled, equivalent to the highest sustained intake in man (Northern Japanese)(20-21).

Histological evaluation of brain, heart, adrenal and kidney removed after the natural deaths of the animals led to the following conclusions: 1) The lesions in freely fed normotensive rats were those commonly seen for aged rats. 2) The tissues from freely fed hypertensive rats had a high incidence of organ damage due to high blood pressure as previously reported (22). 3) However, when hypertensive rats were food restricted, a low incidence of hypertension associated lesions was found. Indeed, food restriction caused a significant change in the incidence of 6 specific lesions likely to be involved in the deaths of freely fed SHR's: (1) interstitial fibrosis in the kidney, (2) myocardial edema and (3) fatty infiltration of myocardium, (4) atrial thrombosis in the heart, and (5) congestion and (6) vacuolization in the adrenal. The frequency of these lesions in food restricted SHR's was not significantly different from that observed in food restricted WKY's. In each case, this frequency was significantly different from the frequency observed in freely fed SHR's. It appears that food restricted SHR's have expanded life spans because they tend to die of causes generally related to aging, rather than from end organ damage due to hypertension.

It must be cautioned that there is no proof that the implications of these studies can be directly extrapolated to humans, or other species. It is possible, in fact, that only those species unable to migrate in the event of food shortage or famine can experience a prolonged life span due to food restriction. This mechanism would allow survival of some individuals until food was again available and the species could be propagated. In a similar fashion, when lactating wood rats are food restricted they will favor the female pups, and allow many (but not all) males to die (23).

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