DECLARATION

I hereby declare that the whole thesis, unless specifically indicated to the contrary in the text, is my own original work and has not been submitted for a degree at any other university.

S HOOSEN

1987

A STUDY OF URINARY AND INTRACELLULAR SODIUM AND POTASSIUM, RENIN, ALDOSTERONE AND HYPERTENSION IN AFRICANS AND INDIANS IN NATAL

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DEDICATION

TO THE MEMORY OF MY FATHER, MOHAMED HOOSEN.

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CHAPTER 1

INTRODUCTION

BACKGROUND OF HYPERTENSION IN BLACKS

Numerous studies have shown that the prevalence of hypertension is higher in Blacks compared to Whites. Seedat et al (1982) found the prevalence of hypertension to be 25% in urban Zulus as compared to 17,2% in Whites. Seftel et al (1980) found a similar prevalence of hypertension in Johannesburg Blacks. Blacks in the United States have a higher prevalence of hypertension and higher average blood pressure readings compared to Whites (Gillum 1979, McDonough 1964, Cruickshank 1982). Negroes from the Virgin Islands (Saunders 1942), the Bahamas (Johnson 1961) and St. Kitts (Schneckloth 1962) have higher blood pressures compared to caucasians living in the same area. Blood pressures in Negroes of Dar-es-Salaam were also higher than in the White population (Cole 1959, Turner 1962). Sever et al (1979) in a study of factory workers in Britain found that Blacks had higher blood pressures compared to Whites.

Of interest is that Blacks living in rural areas have a much lower prevalence of hypertension as compared to their urban counterparts. Hypertension was unusual in Easter

Islanders living on their native land as compared to those who migrated to the South American continent (Cruz-Coke 1964). Rural Zulus in Natal have a much lower prevalence of hypertension (10%) as compared to urban Zulus (Seedat 1982). Sever et al (1980) found higher blood pressures in urban Xhosa as compared to rural Xhosa. Truswell et al (1972) studied !Kung Bushmen in Northern Botswana and found the entire community to be normotensive. Ten years later Kloppers et al (1981) studied a different tribe of Bushmen also from northern Botswana and found the prevalence of hypertension to be 21%. The !Kung Bushmen were a completely isolated community whereas the Bushmen studied by Kloppers had access to army rations and a medical clinic implying an element of urbanization. Kaminer et al (1960) studied 2 groups of Kalahari Bushmen and found that blood pressures were higher in Bushmen prisoners and farm labourers as compared to nomadic Bushmen. Akinkugbe et al (1969) in a study of urban and rural Nigerians found that blood pressure levels were similar in urban and rural women whereas they were much higher in urban men compared to rural men.

Essential hypertension was regarded as being a rare disease in Kenya until 1940. It is now a common cause of heart failure and a major cause of cerebrovascular disease. In 1941 Williams et al described borderline hypertension in 13 Tesot men and in 1944 he recorded undoubted cases of essential hypertension. Samburu warriors recruited into the army had

normal blood pressures on entry and during the first year, however during the 2nd year systolic blood pressure rose significantly and diastolic levels rose slightly.

Not only do blacks have a higher prevalence of hypertension and higher blood pressure levels, but they also have higher mortality rates compared to Whites (Heyman 1971; Kuller 1967). Hypertension is the commonest cause of death in Johannesburg Black adults after violence (Seftel 1980) and hypertension also occurred at a younger age in Blacks compared to Whites (Isaacson 1962). Autopsy studies show that hypertension is the commonest cause of heart disease at Baragwanath hospital (Isaacson 1977).

The pathogenesis of hypertension is multifactorial, and the role of genetic and environmental factors is not clear. Environmental factors must play a major role in the pathogenesis of hypertension in Blacks, as subjects from rural areas have a much lower prevalence of hypertension compared to their urban counterparts (presumably from the same genetic stock). The effect of an environmental factor in a genetically susceptible individual is an unanswered question.

BACKGROUND OF HYPERTENSION IN INDIANS

The prevalence of hypertension in migrant Indians is much higher than in native Indians. Punjabi Indian women living in London are reported to have a prevalence rate of 23% as compared to 1,4% in native Punjabis. Seedat et al (1978) found

the prevalence to be 14,19% in Durban Indians and Valiallah et al (1983) found an identical prevalence rate in Transvaal Indians. These figures are surprisingly high when compared to studies done in India where prevalence rates vary from 3 to 15% for rural areas and 4 to 15% in urban or industrial areas (Chauduri 1978). Verma et al (1983) studied 16 rural villages and found a prevalence rate of 3,6/1000 in adults over 30 years. The prevalence rate in the Simla Hills population was 1,3% (Ghosh 1983), that of the adult population of Agra was 4,3% (Mathur 1963) and the prevalence in Bombay was 10% (Chauduri 1978). Padmavati et al (1959) studied 2 groups in Delhi and found the prevalence of hypertension to be 0,17% in the low socio-economic group (rural) and 2,5% in the high socio-economic group (urban).

Autopsy data of Durban Indians show that the high mortality in Indians compared to Whites was related to the high incidence of hypertensive vascular disease in addition to ischaemic heart disease. Diabetes mellitus was also a significant factor (Wainwright 1969). Seedat et al (1978) also found a significantly higher prevalence of diabetes mellitus in hypertensives as compared to normotensives.

The large difference in the prevalence of hypertension between migrant and native Indians implies an important environmental factor in the pathogenesis of hypertension. Once again the question of an environmental factor in a genetically susceptible individual remains unanswered.

Work on the pathogenesis of hypertension in this ethnic group, related to renin, aldosterone, dietary sodium and potassium and intracellular sodium and potassium is virtually non existent.

DIETARY SODIUM AND POTASSIUM:

In 1904 Ambard and Beaujard found that an increase in dietary sodium chloride caused an elevation in blood pressure and salt restriction caused a drop in blood pressure. Many epidemiological, clinical and animal studies have since been done in an attempt to prove or disprove the "salt hypothesis". Despite this there are still two schools of thought on the association between sodium and hypertension. Evidence for such an association stems mainly from epidemiological and animal studies (Dahl 1962; Dahl 1967). Studies of Greenland Eskimos by Thomas (1927), Australian Aborigines by Hick and Matters (1933), Highland tribes in China by Morse and Beh (1937) and the Cuna Indians of Panama by Kcan (1944) suggest that when salt was introduced into the diet the prevalence of hypertension increased. However the introduction of sodium chloride in the diet was associated with many social and cultural changes which may account for the increased prevalence of hypertension. Frank Lowenstein studied two tribes of Brazilian Indians (1961). He found that the Carajas who lived "primitively" had no increase in blood pressure with age, whilst the Mundurcus who were relatively "accultured" had the

usual increase in blood pressure with age. An important observation was that the Mundurcus used table salt and the Carajas used plant ashes (potassium chloride) for flavouring their food. Ian Maddocks (1967) studied rural and urban Melanesian communities and his findings were similar to those of Lowenstein. The rural communities had no increase in blood pressure with age and their urinary potassium/sodium ratio was 24,2. The urban community on the other hand had a urinary potassium/sodium ratio of 1,9 and the blood pressure rose with age. Prior and Grimely studied sodium intake and blood pressure in three Pacific populations (1969). The mean daily sodium intake of rural Polynesians was 50-70 mmol, that of urban Polynesians was 120-140 mmol and that of a rural European settlement in New Zealand was 156-180 mmol. Urban Polynesians and Europeans had a rise in systolic and diastolic pressure with age while blood pressure only rose in the fifth and sixth decade of female rural Polynesians. There was no rise in blood pressure with age in rural males. Dahl and Love (1957) assessed dietary salt intake in 1346 adults. They found that the group with a low salt intake had a significantly lower incidence of hypertension whilst the group with a high salt intake had significantly more hypertensives than would be expected from a random distribution. Despite the circumstantial evidence presented above, many within population studies have failed to show a direct association between dietary sodium and hypertension.

A study of a Welsh mining valley by Miall et al (1959) revealed that women who habitually added salt to their food had lower systolic pressures than those who did not add salt. He also studied four groups of post menopausal women viz hypertensive and normotensive women who either had high or low salt intakes, and found that hypertensive women had a lower urinary sodium excretion than normotensive women whether their salt intake was low or high. An interesting finding was that hypertensives with a high salt intake had a similar sodium excretion to the low salt group. However normotensives with a high salt intake excreted more sodium than normotensives with a low salt intake. Beevers et al (1980) found a lower urinary sodium excretion in hypertensive subjects whilst Berglund et al (1976) found a positive correlation between diastolic blood pressure and urinary sodium excretion in normotensive subjects and a negative correlation in hypertensive subjects. Hsiao et al (1986) found a positive correlation between sodium/potassium ratio and blood pressure, and a negative correlation between potassium and blood pressure. Simpson et al (1978) in a population survey of 1200 people found no significant relationship between blood pressure and urinary sodium or potassium excretion.

In 1928 Addison postulated that hypertension was due to a low potassium diet and excess added salt. However a deficiency of potassium intake has only recently been highlighted as a causal factor in the pathogenesis of hypertension. Sasaki et al (1978) studied 2 villages in northern Japan and found that

potassium intake was much higher in the village with lower blood pressures although sodium intake was similar. Dahl et al (1972) found dietary sodium chloride to be hypertensinogenic and potassium chloride to be antihypertensinogenic in genetically hypertension-prone rats. Iimura et al (1981) have shown a significant reduction in mean arterial pressure in patients with essential hypertension after a high potassium intake (175 mmol/day) with a moderately high sodium intake (260 mmol/day). This reduction in mean arterial pressure was accompanied by decreases in body weight and body fluid volume and an increase in urine volume, urinary sodium and potassium excretion. They suggest that the beneficial effect of potassium may be due to an enhancement of sodium excretion. Grim et al (1980) in a study of black and white subjects in Evan county, Georgia found that Blacks who generally had higher arterial pressures and a higher prevalence of hypertension, had lower urinary potassium excretion rates than Whites although the sodium excretion was similar in the two groups. A closer look at Kempners (1948) rice and fruit diet for the treatment of hypertension reveals that although it has a very low sodium content it was also rich in potassium.

THE RENIN-ALDOSTERONE SYSTEM:

Robert Tigerstedt discovered renin in 1898 (Marks, 1979), and this discovery lay dormant for 40 years before its importance was appreciated. Renin is a proteolytic enzyme with

a molecular weight of approximately 40 000. It is secreted by the juxtaglomerular cells of the renal cortex and it acts on angiotensinogen (an alpha globulin substrate secreted by the liver) to form angiotensin I, which is rapidly metabolized by converting enzymes to angiotensin II, a potent vasoconstrictor and aldosterone stimulating hormone. Renin is a major factor regulating extracellular fluid volume (Guyton, 1986).

Low plasma renin activity is common in essential hypertension. The incidence of low plasma renin activity in an unselected hypertensive population is between 20-30% (Ganguly, 1979). It has been suggested that suppressed renin levels are due to a change in the adrenal sensitivity to angiotensin II; this enhanced sensitivity modulates a negative feed back loop to generate lower levels of renin. However the role of this abnormality in the pathogenesis of hypertension is uncertain (Kisch, 1976).

Low renin hypertension was initially thought to be a benign type of disease and it was suggested that these patients did not require early treatment (Laragh, 1972). Patients with normal or high renin levels were thought to be more prone to cardiovascular complications (Brunner, 1972). These findings have since been rejected by other workers. Gulati et al (1975) found no evidence for a protective effect in low renin hypertension. Meade et al (1983) have suggested that low levels of plasma renin activity may in fact be associated with an increased risk of cardiovascular disease.

Aldosterone is a mineralo-corticoid secreted by the zona-glomerulosa of the adrenal cortex. Its regulation is controlled by the potassium ion concentration of extra cellular fluid, the renin angiotensin system, the quantity of body sodium and adrenocorticotropic hormone (Guyton 1981).

Several workers have demonstrated increased aldosterone levels in hypertensive patients (Genest 1975; Ljungman 1982; Walker 1979). Khokhar et al (1976) found no difference in the urinary aldosterone excretion rates of hypertensive and normotensive subjects, however after expansion of extracellular fluid volume hypertensive patients showed much less suppression. Parfrey et al (1981) found a decreased responsiveness of the renin-angiotensin - aldosterone system in patients with essential hypertension.

INTRACELLULAR SODIUM:

Studies on intracellular sodium concentration and those on sodium and potassium transport across cell membranes in hypertensive patients are more convincing than those on the role of dietary sodium in hypertension, but are not conclusive. Although much attention has been focused on this aspect in recent years, work done as early as 1943 by Eichelberger showed an increase in muscle sodium in hypertensive dogs. Tobian and Binion in 1952 found an increase in sodium content of medial and intimal layers of renal arteries and the psoas muscle of hypertensive patients. They also found increased sodium in

brain cells suggesting a generalized abnormality. However the sodium content of the right atrium and bladder wall showed no such difference. A major problem with this type of study is contamination with extracellular sodium. Erythrocytes and leucocytes are useful for the determination of intracellular sodium as extracellular sodium is easily removed by washing the cells. The red cell although accessible and easy to work with, is not an ideal cell model as it is a highly specialized cell with no nucleus and it has a slow and almost entirely anaerobic metabolism (Hilton, 1973). In this respect the white blood cell is physiologically a better representative of other cells in the body. Zidek et al (1983) found no correlation between lymphocyte sodium values and sodium activity in red cells. Hilton and Patrick (1973) found the intracellular sodium of white blood cells to be higher than that of red blood cells and the rate constant for sodium efflux was much higher in white cells (4,2) than red cells (0,3). Boon et al (1985) also found the intracellular sodium concentration to be higher in leucocytes but not in erythrocytes of hypertensive patients. It is for this reason that we decided to use lymphocytes in our study of intracellular sodium and potassium.

Sodium passes continuously into the cell by passive diffusion and the relatively low intracellular sodium concentration is maintained by the active extrusion of sodium. A defect in the active transport of sodium out of the cell (sodium-potassium pump) and an increase in passive entry will cause the cell sodium concentration to rise. Many other

sodium-potassium transport systems (Blaustein, 1984) have been defined; however their role in the pathogenesis of hypertension is not clear. The sodium - lithium countertransport system under physiological conditions operates as a sodium exchanger and cannot account for an increase in cell sodium concentration. This system has been suggested as a genetic marker in essential hypertension (Canessa, 1980), however results published are conflicting. Canessa et al (1980), Brugnara et al (1983) and Weder et al (1984) found an increase in lithium - sodium counter transport in patients with essential hypertension. Smith et al (1984) found this increase only in 26,5% of hypertensives whilst 4,8% of normal subjects (without a family history of hypertension) also had an elevated counter transport system. This overlap in results prevents the use of this transport system as a genetic marker. Weder et al (1984) and Canessa et al (1980) found no difference in the lithium - sodium counter transport of hypertensive and normotensive black subjects implying a possible racial difference in the pathogenesis of hypertension. The sodium-potassium cotransport system transports sodium and potassium ions in the same direction in a 1:1 ratio and does not influence the concentration of intracellular sodium unless the concentration of intracellular sodium rises above a critical level (Brand 1984). Its role as a genetic marker in hypertension is debatable. Tuck et al (1984) found lower values for red blood cell cotransport in hypertensive blacks compared to normotensive blacks while cotransport was higher in

caucasian hypertensive subjects compared to normotensive controls. Canessa et al (1984) also found a decreased sodium potassium cotransport system in black hypertensives. In a study of three South African ethnic groups Davidson et al (1982) found no ethnic difference in sodium potassium cotransport. Hypertensives of all three ethnic groups had lower cotransport values; however there was a large overlap in results.

It appears then that the sodium-potassium pump which actively exchanges intracellular sodium for extra-cellular potassium in a ratio of 3:2 is probably responsible for intracellular sodium homeostasis under normal conditions. Both stimulation and inhibition of active sodium-potassium transport and the presence of circulating sodium pump inhibitors have been described (Brock, 1982; Hout, 1983, Hamlyn, 1982; Poston, 1982).

Studies on the actual concentration of intracellular sodium are not as numerous as those on transport mechanisms. Those on red cell sodium show conflicting results. In 1958 Guiseppe D' amico found an increase in red cell sodium in hypertensive patients but as he only studied 5 patients, results are difficult to interpret. Aderounmu and Salaka (1979) in their study of 1008 Nigerians (100 hypertensives and 908 normotensives) found a significantly higher red cell sodium concentration in hypertensive subjects. Tedde et al (1983) found similar results in Italian patients although they studied a smaller number of patients. Wessels et al (1980) found an

increased concentration of sodium in red cells of hypertensive patients only when large numbers were studied. Walter and Distler (1982) and Canessa et al (1980) found no difference in the red cell sodium concentration of hypertensive and normotensive subjects. Munro-Faure et al (1971) also found no difference in the red cell sodium concentration of hypertensive and normotensive subjects but they found higher red cell sodium values in Negro patients compared to Caucasians. Tuck et al (1984) showed a similar racial difference but this was evident only in hypertensive patients. Wessels et al (1980) suggest that in studies which show no difference in the red cell sodium concentration of hypertensive and normotensive patients, the number of patients studied were too small. Simon et al (1986) on the other hand found a decrease in red cell sodium concentration in hypertensives compared to normotensives.

Studies on white blood cell sodium concentration are consistent in that all studies in the literature to date show an increase in intracellular sodium concentration in hypertensive subjects. Ambrosioni et al (1979) studied intralymphocytic sodium concentration in 134 patients and found lower sodium levels in normotensive subjects. They also found a significant correlation with mean blood pressure in hypertensive patients. Edmonson et al (1975) found a higher leucocyte sodium concentration in hypertensives in a study of 34 Jamaican patients. A study of leucocyte sodium by Araoye et al (1978) showed similar results.

It should be borne in mind that an increase in intracellular sodium (red cell sodium) has been described in other conditions such as hyperthyroidism, digitalis therapy, extracellular hypernatraemia, advanced renal insufficiency and psychosis (Zumkley, 1980).

Based on the above facts the aims and objectives of this study were:

(1) To assess the intake of sodium and potassium in urban Zulus, rural Zulus and Indians in Natal (by means of urinary sodium and potassium excretion rates).

(2) To determine whether there is any association between blood pressure, sodium and potassium intake/excretion, plasma renin activity and serum aldosterone levels.

(3) To measure the intralymphocytic sodium and potassium of Zulus and Indians.

(4) To establish whether there are differences in the intracellular sodium and potassium of hypertensive and normotensive subjects.

(5) To assess the relationship (if any) between blood pressure and intracellular sodium and potassium.

(6) To assess the effect of

(i) intravenous furosemide

(ii) diuretic and beta-Blocker therapy

on intralymphocytic sodium and potassium.

CHAPTER 2

PATIENTS AND METHODS

The study was divided into two sections. Part (a) deals with urinary sodium and potassium, plasma renin and serum aldosterone (chapters 3,4,5). Part (b) deals with intralymphocytic sodium and potassium (chapters 6, 7, 8).

a) Urinary sodium and potassium, renin and aldosterone.

This study was carried out over a period of 20 months from March 1981 to October 1982. Five hundred and eighty five patients were assessed. Urban Zulus (240) were studied in Durban at 2 light industrial firms, the Lamontville Township and the outpatient department of King Edward VIII Hospital. Indians (176) were drawn from a satellite clinic of King Edward VIII Hospital, R K Khan Hospital and from the 2 light industrial firms mentioned above. Rural Zulus (169) were studied at Bethesda Hospital in Ubombo (population 43,292) and at Benedictine Hospital in Nongoma (population 75,603) (Department of Statistics). Both these districts are in Northern Zululand with adjacent boundaries.

Hypertensive patients were referred to the above mentioned hospitals by satellite clinics, community workers and general practitioners. These patients presented with minor

complaints or were found to be hypertensive on pre-employment medicals and insurance medicals. Patients with complications of hypertension and those with a major concomitant illness were excluded from the study. In the rural areas blood pressures were measured on all adults attending the outpatient department. Those who were suitable were accepted for the study. None of the patients were on any form of medication. Patients with a past history of hypertension had stopped all medication for at least 4 weeks prior to the study. Controls were chosen on a voluntary basis from the industrial firms mentioned previously and from Lamontville Township. Rural controls were chosen from the same community as the hypertensives on a voluntary basis. Selection of Indian controls was similar to that of urban Zulu controls.

Consent was obtained from all subjects after an explanation of the purpose of the study. Subjects were examined under similar conditions between 8 a.m. and 12 noon. Blood pressure was recorded 3 times with an anaeroid sphygmomanometer at intervals of 20 minutes after the subject had been seated for at least 5 minutes. The fifth phase was taken as the diastolic blood pressure and hypertension was defined according to World Health Criteria (1962). A mean of 3 readings was taken. Blood pressure was retaken after 1 week to exclude those who were erroneously diagnosed as being hypertensive.

Blood was drawn for estimation of urea, electrolytes, plasma renin activity and serum aldosterone after blood pressures were recorded (ie after the patient had been seated

for 1 hour). Blood for the estimation of plasma renin activity was drawn into pre-chilled tubes containing potassium EDTA as an anti-coagulant. This was determined on frozen samples ($-20^{o}C$) by the radio-immunoassay of generated angiotensin 1. (Clinical assays, division of Travenol laboratories). Plasma aldosterone levels were measured by quantitative radio-immunoassay using the antibody coated tube method (CIS). Seven timed overnight urine samples were collected for estimation of sodium, potassium and creatinine. The method of collection was carefully explained to subjects and this explanation was repeated with each sample collected to ensure consistency. Urinary sodium and potassium were measured on an ILC 243 flame photometer.

b) Intralymphocytic sodium and potassium.

Hypertensive patients for this study were selected from the hypertension clinics of King Edward VIII Hospital and Beatrice Street Clinic (a satellite clinic of King Edward VIII Hospital). The majority of patients were newly diagnosed hypertensives. A few were known hypertensives who had been off treatment for at least 2 months. Patients with a blood pressure equal to or more than 160/100 on 3 separate occasions were accepted for the study. These patients had no other illness and were not on any form of medication.

All patients had a thorough physical examination. Urinalysis, urea and electrolytes, serum creatinine, chest radiograph and an electrocardiogram were done on all patients. Patients with an abnormal urine or a serum creatinine of more than 140 umol/ℓ were excluded from the study. Other investigations to exclude secondary hypertension were done where clinically indicated e.g. urinary catecholamines, ultrasound of kidneys and adrenal glands, serum calcium and phosphate. Patients with secondary hypertension or malignant hypertension were excluded from the study.

Normotensive controls were chosen on a voluntary basis from hospital and clinic staff and from a light industrial firm. They were healthy men and women and were not on any form of medication.

Blood pressure was recorded twice in the sitting position (seated for at least 5 minutes) and twice in the standing position (standing for 2 minutes) with a mercury manometer. Blood was taken for the measurement of ILNa and ILK in the sitting position. Patients were on an unrestricted diet.

This study on intracellular sodium and potassium was sub-divided into 3 sections.

i) ILNa and ILK were measured in 88 Zulus and 88 Indians (chapter 6).

ii) The effect of intravenous furosemide (20 mg) on ILNa and ILK was assessed in 47 Zulus and 52 Indians (chapter 7).

iii) The effect of antihypertensive drugs on ILNa and ILK were assessed only in hypertensive subjects. The effect of 25 mg hydrochlorothiazide was assessed in 51 patients (24 Zulus and 27 Indians) and the effect of 160 mg Sotalol was assessed in 36 patients (16 Zulus and 20 Indians). Informed consent was obtained from all subjects.

The study was approved by the Ethics Committee of the University of Natal.

DEFINITION OF HYPERTENSION

(WHO - CRITERIA - 1962)

1) Less than 30 years

Systolic $\geq$ 150 and/or

Diastolic $\geq$ 90

2) 30-65 years

Systolic $\geq$ 160 and/or

Diastolic $\geq$ 95

3) Over 65 years

Systolic $\geq$ 165 and/or

Diastolic $\geq$ 95

CHAPTER 3

URINARY SODIUM AND POTASSIUM EXCRETION RATES IN URBAN ZULUS, RURAL ZULUS AND INDIANS

INTRODUCTION

Major differences exist in the prevalence of hypertension between black and White subjects (Gillum 1979). The prevalence of hypertension in Natal is 25% in urban Zulus as compared to 17,2% in Whites, 14,2% in Indians and 10,5% in rural Zulus (Seedat 1982). One postulated pathophysiological mechanism is the difference in renal handling of sodium and water. Black hypertensives are said to have a greater plasma volume than White hypertensives (Lilley 1976) and black subjects also have a delayed excretion of sodium and potassium following a saline load (Luft 1977). This may be the result of blacks having evolved more efficient mechanisms for renal sodium conservation to cope with a low sodium intake in a semitropical environment and exposure to the high sodium intake of western diets places blacks at a disadvantage (Grim 1980). Luft et al (1977) studied the effect of a saline load on normotensive blacks and Whites, and found that blacks excreted less sodium and potassium following the salt load than Whites. Blacks also have significantly higher blood pressures at lower sodium intakes (800 mmol/day) than Whites (1200 mmol/day) (Luft 1979). Modlin

et al in 1963 found the urinary sodium excretion of young Basuto men to be higher than that of White controls and postulated that the high intake of sodium was associated with the higher incidence of hypertension in the Basuto. Barlow et al (1982) in a similar study found urinary sodium excretion to be lower in normotensive blacks compared to normotensive Whites. Cohen et al (1982) on the other hand found no significant differences in the urinary sodium excretion of normotensive Whites, normotensive blacks and hypertensive blacks. The urinary potassium excretion however, was significantly lower in black subjects in all three studies while the urinary sodium/potassium ratio was higher in blacks. Potassium is said to have an anti-hypertensive effect by causing vasodilation and by increasing natriuresis (Bulpitt 1981).

Patients and Methods:

A total of 585 patients were assessed. One hundred and thirty eight were hypertensive urban Zulus (62 females and 76 males); 96 were urban normotensive Zulus (50 females and 46 males); 87 were rural hypertensive Zulus (44 females and 43 males); 85 were rural normotensive Zulus (30 females and 55 males); 94 were hypertensive Indians (47 females and 47 males) and 83 were normotensive Indians (30 females and 53 males). Further details on methodology are given in chapter 2(a).

Diet

Patients were on an unrestricted sodium diet.

Environment

The average temperature in Durban is 23,8°C in summer and 16,1°C in winter and in the rural area it is 24,5°C in summer and 15,1°C in winter. Subjects were not assessed in midsummer when temperatures could be as high as 31°C in the urban area and 36°C in the rural area. None of the subjects worked under conditions of extreme heat or cold.

Statistics

Data were analysed on the IBM 4331 computer at the institute for biostatistics of the South African Medical Research Council and use was made of BMDP packages and the SAS program (Dixon, 1982; Ray 1982). The SAS program was used where data had to be corrected for age and Quetelet's index (Khosla 1967).

Age and Quetelet's Index

The mean ages and Quetelet's index $\pm$ S.D. for Zulus and Indians are given in Table 1. Rural Zulus were on the average 7 years younger than urban Zulus and 5 years younger than Indians. Normotensive subjects in all groups were 10 years

younger than hypertensive subjects. Hypertensive subjects were heavier than normotensive subjects. Data were corrected for age and Quetelet's Index during statistical analysis.

Blood Pressures

Systolic and diastolic (phase 4 and 5) blood pressures are given in Table 2.

RESULTS

Three hundred and sixty three of the 585 patients (63%) collected 7 overnight urine specimens; 89 (15%) collected 6 specimens; 23 (4%) collected 5 specimens; 17 (3%) collected 4; 23 (4%) collected 3; 17 collected 2 specimens and 20 (6%) collected 1 specimen.

Sodium Excretion

Age had no effect on sodium excretion, but Quetelet's index (weight (kg)/Height $(m)^2$) was positively associated with sodium excretion ($r = + 0,41$; $p < 0,05$); however this effect was not identical in all groups and the data on sodium excretion were therefore analysed at three different levels of

Quetelet's index ie at normal body weight (22) and obesity (27 and 31). Analysis of data at one level would give incorrect results.

At Quetelet's index 22 there was no significant difference in the urinary sodium excretion rates of urban and rural Zulus. Hypertensive urban Zulus and Indians had similar sodium excretion rates to their normotensive counterparts. Hypertensive rural Zulus on the other hand excreted significantly less sodium than normotensive subjects ($p = 0,02$) (Figure 1).

At Quetelet's index 27 there was also no significant difference in the sodium excretion of urban and rural Zulus. The sodium excretion of hypertensive patients in all three groups was not significantly different from normotensive subjects (Figure 1).

At Quetelet's index 31 urban Zulus excreted significantly more sodium than rural Zulus ($p = 0,02$), and urban hypertensive patients had a lower urinary sodium excretion than normotensive patients ($p = 0,01$) (Figure 1).

Indian patients had significantly lower urinary sodium excretion rates when compared to urban and rural Zulus at all levels of Quetelet's index ($p = 0,0001$) (Figure 1).

There was no correlation between urinary sodium excretion and blood pressure in all groups.

Potassium Excretion

Age had no effect on urinary potassium excretion. Quetelet's index was positively associated with potassium excretion ($r = 0,41$; $p = 0,02$) and had a similar effect on all groups. Data were therefore analysed at one point only. There was no difference in the potassium excretion of urban and rural Zulus (figure 2). Indians excreted significantly less potassium than urban and rural Zulus ($p = 0,0001$). There was no difference in the potassium excretion of hypertensive and normotensive subjects in all 3 groups.

There was a weak but significant negative association between urinary potassium excretion and systolic blood pressure in rural Zulus ($r = -0,16$; $p = 0,04$) and Indians ($r = -0,19$; $p = 0,01$).

Sodium/Potassium Ratio

Quetelet's index had no effect on the urinary sodium/potassium (Na/k) ratio. The effect of age was significant ($p = 0,01$) and was different in the various groups and data were therefore analysed at 3 different ages (figure 3). At age 52 there was no significant difference in the urinary Na/k ratio of urban and rural Zulus. At age 43 rural Zulus had a lower Na/k ratio than urban Zulus ($p < 0,05$). At age 33 rural Zulus again had a lower Na/k ratio than urban

Zulus ($p = 0,03$). There was no difference in the Na/k ratio of Indians and Zulus. There was also no difference in the urinary Na/k ratio of hypertensives and normotensives in all groups.

There was no correlation between the urinary Na/k ratio and blood pressure.

Socio Economic Background

Socio economic background was assessed by means of a questionnaire on occupation and housing.

Zulus from a poor socio-economic background i.e. labourers and the unemployed had a higher urinary sodium excretion and a higher Na/k ratio than Zulus from a higher socio-economic background i.e. professionals and artisans ($p < 0,05$). However Indians from a higher socio-economic background had higher urinary sodium values compared to those with a lower socio-economic status.

DISCUSSION

Sodium intake has been assessed by means of dietary history, 24 hour urine collections and casual urine specimens. However reliable 24 hour urine specimens are difficult to obtain in a free living population and dietary recall histories seem unreliable in predicting sodium excretion. Spot urine samples show no correlation with 24 hour urine samples (Watson, 1970). Single 24 hour urine estimations are not representative as the day to day variation can be high. The variation was found to be to 25% in the study done by Milne et

al (1980). The decision to use several overnight urine samples was based on the experience of Watson and Langford (1970), Pietinenn et al (1976) and Liu et al (1979). These workers found good correlations between overnight urine specimens and 24 hour samples.

There was a tendency toward lower sodium excretion in hypertensive patients but this only achieved statistical significance in urban Zulus at Quetelet's index 31 and in rural Zulus at Quetelet's index 27. There have been other reports of lower urinary sodium excretion in hypertensive subjects by Beevers et al (1980), Bergland et al (1976) and Miall et al (1959).

There was also a tendency toward higher sodium excretion in urban Zulus but this was only statistically significant in the more obese patients. This is unlike other studies of urban and rural communities of similar origin which showed that urban dwellers had a higher salt intake compared to their rural counterparts (Lowenstein, 1961; Maddocks, 1967; Prior, 1969). Sever et al (1980) studied urban and rural Xhosa in Southern Africa and also found a higher urinary sodium excretion in urban Xhosa compared to rural Xhosa. Variation from our results may be due to differences in methodology and the population studied was also different. The most likely explanation is a change in dietary habits. Like most within population studies, we found no association between dietary sodium and blood pressure.

Numerous studies have shown that potassium supplements given to hypertensive patients cause a small but significant drop in blood pressure (Overlack, 1983; Iimura, 1981; MacGregor, 1982; Khaw, 1982; Matlou 1986). Potassium supplementation however did not lower blood pressure in normotensive subjects (Barden 1986). Richards et al (1984) and Zocalli et al (1985) on the other hand found no fall in blood pressure with potassium supplementation in hypertensive patients on a normal sodium diet. Smith et al (1983) have shown that when potassium supplements are given in addition to sodium restriction there is no further drop in blood pressure with potassium supplementation. Sasaki et al (1979) studied 2 villages in Northern Japan and found potassium intake to be much higher in the village with lower blood pressures although sodium intake was similar.

Voors et al (1983) have shown that potassium supplementation in blacks caused natriuresis, a negative sodium balance and a cumulative potassium balance more positive than Whites. In some black subjects natriuresis was associated with weight loss and/or a drop in systolic blood pressure.

This study showed no difference in potassium excretion of urban and rural Zulus and also no difference between hypertensive and normotensive subjects in all groups. However urinary potassium excretion was negatively associated with blood pressure in rural Zulus and Indians but not in urban

Zulus. Walker et al (1979), Staessen et al (1981) and M'Buyamba-Kabangu et al (1986) have also described a negative association between potassium and blood pressure.

More important than the absolute amounts of sodium and potassium may be their ratio. Barlow et al (1982) found that although both the sodium and potassium excretion rates were lower in blacks, their Na/k ratios were higher than in Whites. A study of dietary intake in 1928 Black and 9739 White subjects (NHANES I) also showed that Blacks had lower sodium and potassium intakes compared to Whites and they also had a higher sodium/potassium ratio than Whites (Frisancho 1984).

In a recent study by Barlow et al (1986) the faecal potassium concentration of blacks (15 mmol) and Whites (20 mmol) was not significantly different. This was a surprising finding as one would expect a lower faecal potassium in black subjects. More studies with bigger numbers are needed to confirm this. Rural Zulus had a lower Na/k ratio compared to urban Zulus. The prevalence of hypertension as mentioned previously is much lower in rural Zulus (10,5%) as compared to urban Zulus (25%) and the difference in Na/k ratio may in part explain this difference in prevalence. There was however no correlation between the urinary Na/k ratio and blood pressure. Hsiao et al (1986) on the other hand found a positive correlation between the urinary sodium/potassium ratio and blood pressure. M'Buyamba-Kubangu et al (1986) also found a positive correlation between the urinary sodium/potassium ratio

and blood pressure in urban blacks of Zaire. Indians had a slightly lower Na/k ratio than urban Zulus (although not statistically significant) with a corresponding lower prevalence of hypertension.

Postulating from mean hourly excretion rates, the 24 hour sodium intake of Zulus is about 160 mmol and that of Indians is about 100 mmol. Current sodium consumption in the western world is 150-250 mmol/day i.e. the sodium intake of Zulus is on par with that of the western world. Potassium intake of Zulus, on the other hand, is about 36 mmol/24 hours which is about half that of western societies. Indians have an even lower potassium intake of 20 mmol/day.

This study has shown no clear-cut association between sodium and hypertension. A potassium deficient diet may play a role. A lower Na/k ratio in rural Zulus may be protective against the development of hypertension.

MEAN AGES ± SD, QUATELETS INDEX (QI) ± SD

TABLE 1

	FEMALES (n)	MALES (n)	MEAN AGE ± SD	QI ± SD
Urban Hypertensive Zulus	62	76	49,7 ± 12,0	30,7 ± 7,2
Urban Normotensive Zulus	50	46	39,6 ± 13,9	28,5 ± 13,9
Rural Hypertensive Zulus	44	43	43,6 ± 11,6	28,5 ± 6,3
Rural Normotensive Zulus	30	55	33,72 ± 12,0	25,5 ± 4,7
Hypertensive Indians	47	47	50,4 ± 11,9	26,8 ± 4,7
Normotensive Indians	30	53	37,7 ± 11,6	22,6 ± 4,5

MEAN BLOOD PRESSURE ± SD

TABLE 2

	SYSTOLIC	DIASTOLIC 4	DIASTOLIC 5
Normotensive Urban Zulus	126,4 ± 13,2	85,4 ± 7,6	78,3 ± 8,2
Hypertensive Urban Zulus	162,56 ± 25,3	108,3 ± 11,6	102,2 ± 12,1
Normotensive Rural Zulus	121,5 ± 12,1	79,9 ± 8,6	75,7 ± 9,4
Hypertensive Rural Zulus	159,7 ± 19,2	104,1 ± 7,2	101,8 ± 7,6
Normotensive Indians	119,3 ± 15,0	75,2 ± 9,9	72,4 ± 10,4
Hypertensive Indians	168,6 ± 19,9	106,2 ± 9,0	102,9 ± 9,2

FIGURE 1

URINARY SODIUM EXCRETION (mean ± SEM) IN HYPERTENSIVE AND NORMOTENSIVE SUBJECTS

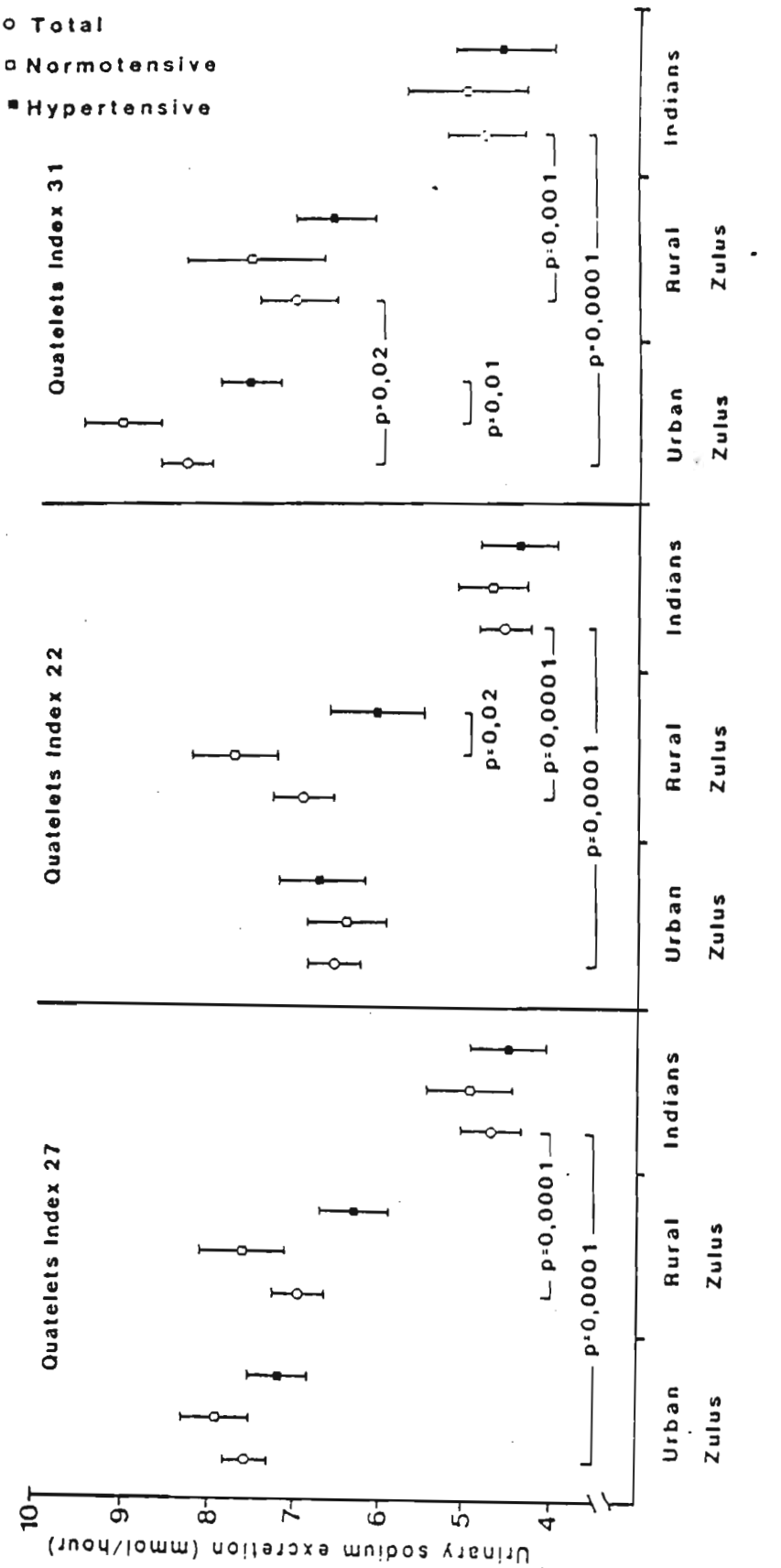


FIGURE 2

URINARY POTASSIUM EXCRETION (mean ± SE
IN HYPERTENSIVE AND NORMOTENSIVE SUBJECTS

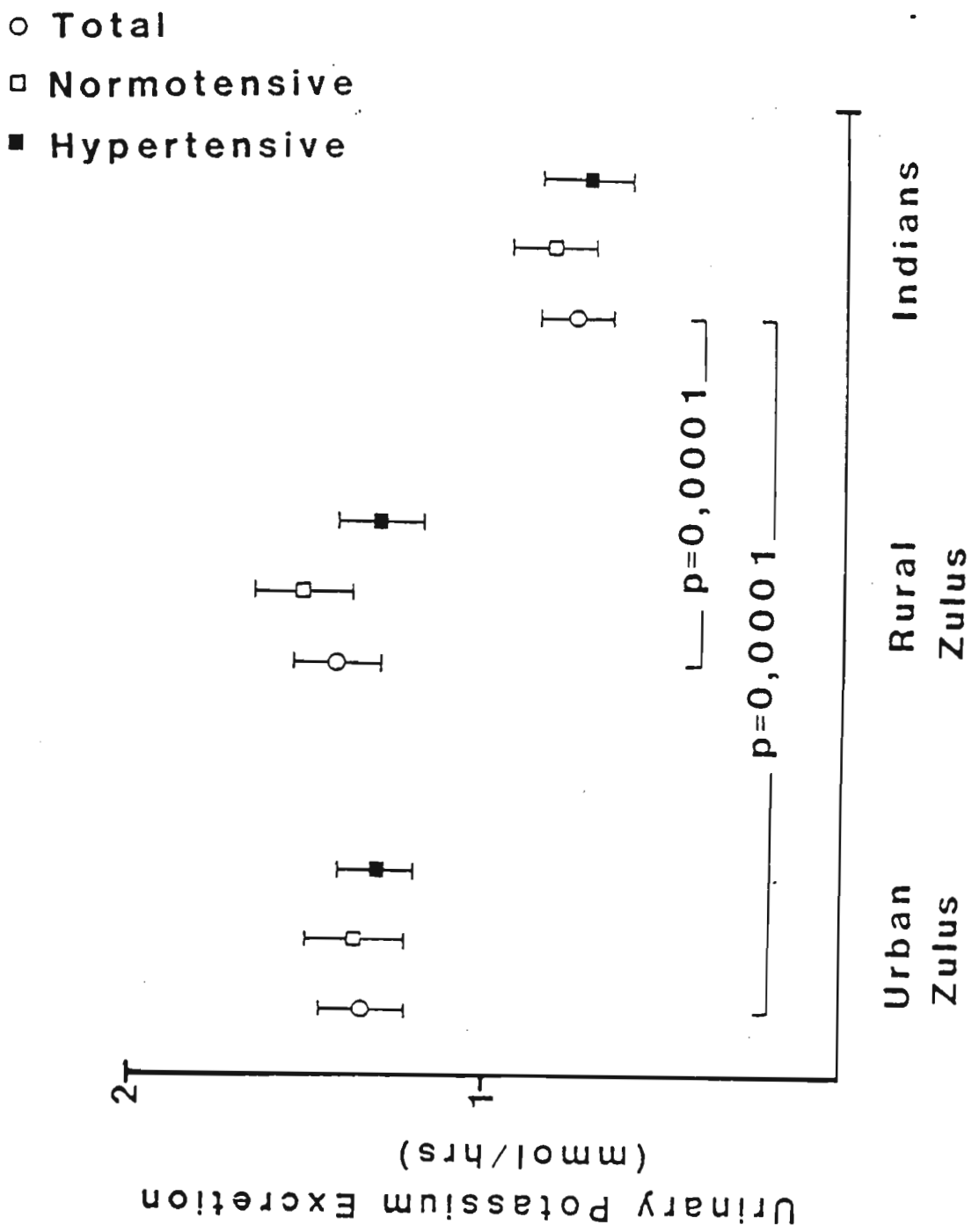
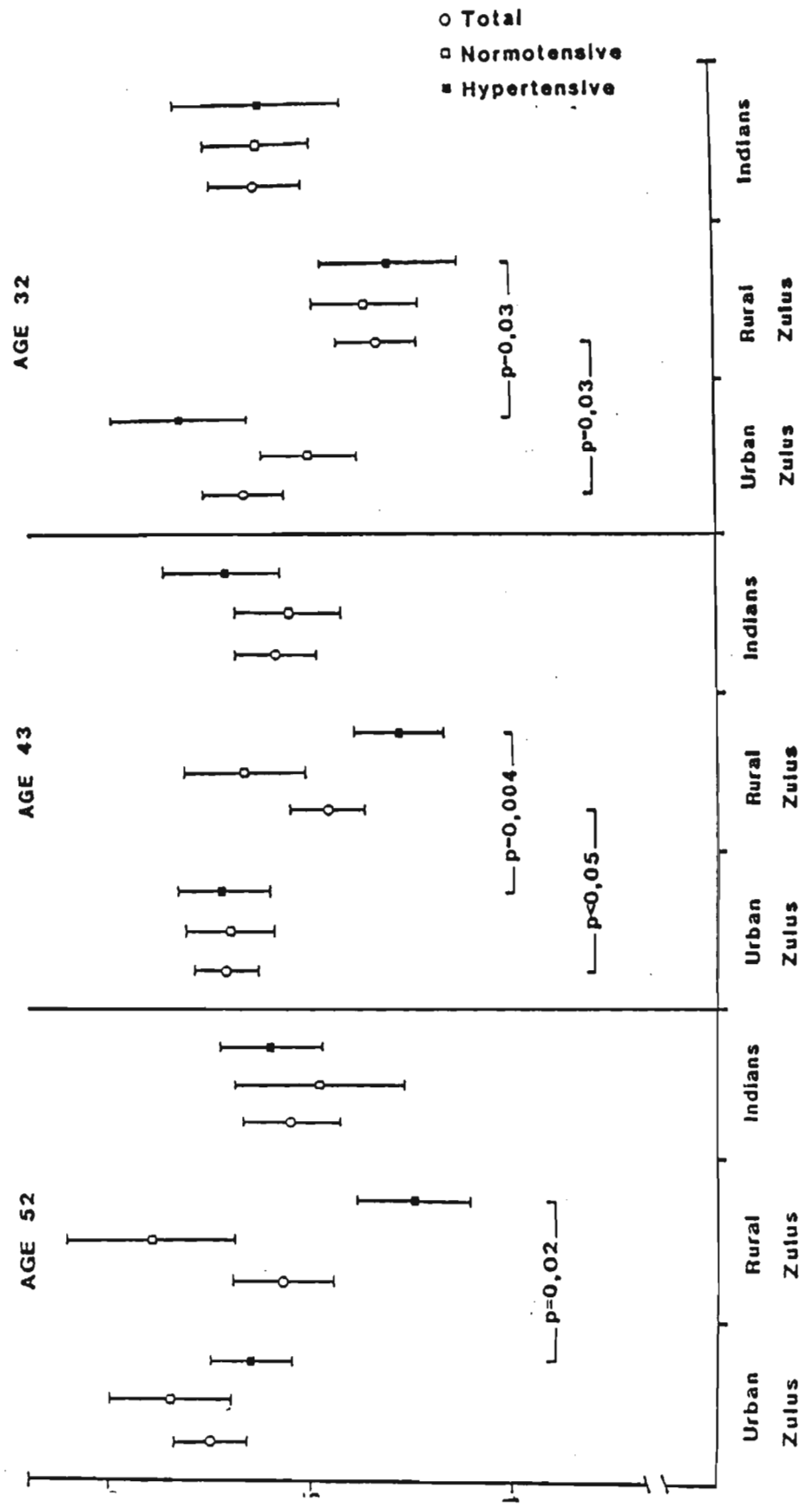


FIGURE 3

THE URINARY SODIUM/POTASSIUM RATIO (mean ± SEM)
IN NORMOTENSIVE AND HYPERTENSIVE SUBJECTS



CHAPTER 4

RENIN AND ALDOSTERONE

INTRODUCTION

Plasma renin levels are reported to be lower in Blacks than in Whites (Gulati 1975; Kaplan 1976; Tuck 1973; Brunner 1972; Lijnen 1985). This is postulated to be due to a true ethnic difference on a genetic basis (Iwai 1973); other explanations offered are a high salt intake and an increase in plasma volume in blacks (Creditor 1967; Creditor 1968; Lilley 1976). Black patients respond better to diuretics than to Beta-blockers and this has been attributed to the high incidence of low renin hypertension in this ethnic group (Holland 1979; Karlberg 1976). Holland and Fairchild (1982) confirmed this therapeutic response but found that it was unrelated to renin status.

Numerous studies have shown that plasma aldosterone levels are increased in patients with essential hypertension (Ljungman 1982; Genest 1975; Kloppenborg 1982). This may be due to decreased metabolic clearance (Nowaczynski 1975) or to an inability of the adrenal gland to "switch off" aldosterone production in response to physiological stimulation such as a high salt intake. Experimental evidence suggests that

aldosterone enhances transepithelial sodium transport and this may lead to an increase in intracellular sodium and calcium concentration thus causing an increase in vascular smooth muscle contractility (Ljungman 1982).

METHOD

A total of 585 patients were assessed. One hundred and thirty eight were hypertensive urban Zulus (62 females and 76 males); 96 were urban normotensive Zulus (50 females and 46 males); 87 were rural hypertensive Zulus (44 females and 43 males); 85 were rural normotensive Zulus (30 females and 55 males); 94 were hypertensive Indians (47 females and 47 males); 87 were normotensive Indians (30 females and 53 males). Further details are given in Chapter 2 (a).

DIET

Patients were on an unrestricted sodium diet.

RESULTS

Mean ages and Quetelet's indices are given in Table 1 and mean blood pressures in Table 2.

RENIN

Plasma renin activity (PRA) was inversely associated with age in urban Zulus ($r = 0,24$; $p = 0,002$) and Indians ($r = 0,16$; $p = 0,04$) but not in rural Zulus. Quatelets index had no effect on plasma renin activity. Age was corrected for during statistical analysis.

The mean PRA $\pm$ SEM (Figure 1) was $4,88 \pm 0,81$ in urban Zulus as compared to $6,57 \pm 0,74$ in rural Zulus. This difference even when corrected for age, was highly statistically significant ($p = 0,001$). The mean PRA for Indians was $4,48 \pm 0,81$. This was not significantly different from urban Zulus but was significantly lower than the PRA of rural Zulus ($p = 0.0001$).

Urban hypertensive Zulus had a significantly lower PRA than their normotensive counterparts ($p = 0,02$). Although the PRA of hypertensive rural Zulus and Indians was lower than normotensive subjects this difference was not statistically significant. The mean PRA of urban hypertensive Zulus was significantly lower than that of rural hypertensive Zulus.

The prevalence of low PRA was 39,3% in urban Zulus as compared to 21,3% in rural Zulus. This difference was highly significant ($p = 0,0005$). The prevalence in Indians was 31,3% and this was not significantly different from the PRA of urban Zulus. Hypertensive urban Zulus and hypertensive Indians had a higher prevalence of low PRA compared to normotensive controls

(Table 3). Low plasma renin levels were found in 52,7% of hypertensive urban Zulus and 28,2% of hypertensive rural Zulus ($p < 0,005$). Frequency histograms of plasma renin activity are shown in addendums 1 and 2.

Figures (2,3,4 and 5) show a statistically significant inverse association between PRA and blood pressure (systolic and diastolic) in urban Zulus and Indians. In rural Zulus this correlation was not statistically significant when corrected for age (Figures 6 and 7). PRA accounts for only 9% of the variation in blood pressure in Zulus and 7% in Indians. There was no correlation between urinary sodium and potassium excretion and PRA.

ALDOSTERONE

Age and Quetelet's index had no effect on serum aldosterone levels. The mean serum aldosterone levels $\pm$ SEM are given in Table 4. Although urban Zulus tended to have lower levels than rural Zulus this difference was not statistically significant. There was no significant difference in the aldosterone levels of hypertensive and normotensive patients in all groups. With the exception of hypertensive urban Zulus and hypertensive Indians there was a significant positive correlation between PRA and aldosterone in all other groups as shown in Table 5. Frequency histograms of serum aldostrone are shown in addendums 3 and 4.

Serum aldosterone and the urinary sodium/potassium ratio were inversely associated in Zulu patients ($r = 0,16$; $p = 0,004$) whilst in Indian patients this association was not

statistically significant ($r = -0,14$; $p = 0,07$). There was no correlation between urinary sodium, urinary potassium and serum aldosterone in both the total Zulu and the total Indian group. However urinary sodium excretion correlated negatively with aldosterone in rural Zulus ($r = -0,21$; $p = 0,01$) and urinary potassium correlated positively with aldosterone in urban Zulus ($r = + 0,21$; $p = 0,005$).

DISCUSSION

The high prevalence of low plasma renin levels in hypertensive patients is in keeping with many other studies (Ganguly 1979; Gulati 1975; Kaplan 1976; Tuck 1973; Iwai 1973; Creditor 1967). The reason for the difference in the prevalence of low renin hypertension between urban and rural Zulus is not clear. If PRA is genetically determined as suggested in the literature, urban and rural Zulus should in theory have similar values. PRA falls with increasing age, and urban Zulus were older than rural Zulus, but even when allowance is made for this factor, the difference persists. This leads to the question of an environmental factor influencing a genetic predisposition to low PRA. An increase in dietary sodium in urban Zulus as a cause has been excluded (Chapter 3). A possible environmental factor such as diet or stress was beyond the scope of this study. Sever et al (1980) found no difference in the plasma renin levels of urban and rural Xhosas. Unlike

Sever et al, this study showed no correlation between PRA and urinary sodium excretion. However, Sever et al looked at casual urine samples whereas we looked at overnight urine samples. Walker et al (1979) also found no correlation between PRA and urinary sodium excretion.

The figures for PRA in this study may be falsely low as these patients were on an unrestricted diet and PRA was not stimulated. The nature of the study and the number of subjects assessed made it impossible to use any form of stimulation e.g. dietary sodium restriction. A separate study of stimulated PRA has been done (Chapter 5).

The negative correlation between blood pressure and PRA described in this study has previously been reported (Brunner 1973; Ganguly 1979).

High serum (Genest 1975) and urinary aldosterone (Thulin 1978; Tolagen 1978) levels have been noted in hypertensive patients. Other workers have found no differences between hypertensives and normotensives (Walker 1979; Biglieri 1979; Laragh 1966). This study also found no difference in the serum aldosterone levels of hypertensive and normotensive subjects of both ethnic groups.

A positive correlation between renin and aldosterone is to be expected. Its absence in urban hypertensives (Indians and Zulus) is suggestive of a defect in the renin-aldosterone

system in these patients. The nature of this abnormality is not clear. It may be related to the high prevalence of low renin hypertension in these two groups.

The finding of a negative correlation between aldosterone and the urinary sodium/potassium ratio is similar to the findings of Ljungman et al (1982). However unlike them we found no correlation between blood pressure and aldosterone.

Future studies should look at factors affecting renin release and the effect of stress on this enzyme. The effect of salt loading on aldosterone should also be assessed as this will answer the question of a possible abnormality in the negative feedback loop of aldosterone excretion.

MEAN AGES ± SD, QUATELETS INDEX (QI) ± SD

TABLE 1

	FEMALES (n)	MALES (n)	MEAN AGE ± SD	QI ± SD
Urban Hypertensive Zulus	62	76	49,7 ± 12,0	30,7 ± 7,2
Urban Normotensive Zulus	50	46	39,6 ± 13,9	28,5 ± 13,9
Rural Hypertensive Zulus	44	43	43,6 ± 11,6	28,5 ± 6,3
Rural Normotensive Zulus	30	55	33,72 ± 12,0	25,5 ± 4,7
Hypertensive Indians	47	47	50,4 ± 11,9	26,8 ± 4,7
Normotensive Indians	30	53	37,7 ± 11,6	22,6 ± 4,5

MEAN BLOOD PRESSURE ± SD

TABLE 2

	SYSTOLIC	DIASTOLIC 4	DIASTOLIC 5
Normotensive Urban Zulus	126,4 ± 13,2	85,4 ± 7,6	78,3 ± 8,2
Hypertensive Urban Zulus	162,56 ± 25,3	108,3 ± 11,6	102,2 ± 12,1
Normotensive Rural Zulus	121,5 ± 12,1	79,9 ± 8,6	75,7 ± 9,4
Hypertensive Rural Zulus	159,7 ± 19,2	104,1 ± 7,2	101,8 ± 7,6
Normotensive Indians	119,3 ± 15,0	75,2 ± 9,9	72,4 ± 10,4
Hypertensive Indians	168,6 ± 19,9	106,2 ± 9,0	102,9 ± 9,2

TABLE 3

PLASMA RENIN ACTIVITY (%)

	LOW	NORMAL	HIGH
ZULUS:			
TOTAL URBAN (n = 163)	39,3*	38,0	22,1*
TOTAL RURAL (n = 169)	21,3	40,2	38,5
NORMOTENSIVE URBAN (n = 72)	23,6 @	44,4	31,9 @
NORMOTENSIVE RURAL (n = 84)	14,3	41,7	44,0
HYPERTENSIVE URBAN (n = 91)	52,7**	33,0	14,3**
HYPERTENSIVE RURAL (n = 85)	28,2	38,8	32,9
INDIANS			
TOTAL (n = 166)	31,3	51,2	17,5
NORMOTENSIVE (n = 87)	24,1	51,7	24,1
HYPERTENSIVE (n = 79)	39,2+	50,6	10,1+

* Urban vs Rural $p < 0,0005$

** Urban vs Rural $p < 0,005$

@ Hypertensive vs Normotensive $p < 0,001$

\+ Hypertensive vs Normotensive $p < 0,05$

MEAN SERUM ALDOSTERONE ± SEM

TABLE 4

	ALDOSTERONE (pg/ml)
Urban Hypertensive Zulus	89,8 ± 5,1
Urban Normotensive Zulus	86,0 ± 6,5
Rural Hypertensive Zulus	113,1 ± 7,8
Rural Normotensive Zulus	95,7 ± 9,6
Hypertensive Indians	109,8 ± 7,0
Normotensive Indians	111,2 ± 7,0

CORRELATION BETWEEN PLASMA RENIN ACTIVITY AND ALDOSTERONE

TABLE 5

ZULUS

Urban Hypertensive	r	=	0,08	(N/S)
Urban Normotensive	r	=	0,32	($p < 0,01$)
Rural Hypertensive	r	=	0,24	($p < 0,05$)
Rural Normotensive	r	=	0,43	($p < 0,0005$)

INDIANS

Hypertensive	r	=	-0,12	(N/S)
Normotensive	r	=	0,24	($p < 0,005$)

FIG. 1 PLASMA RENIN ACTIVITY (mean ± SEM) IN NORMOTENSIVE AND HYPERTENSIVE SUBJECTS

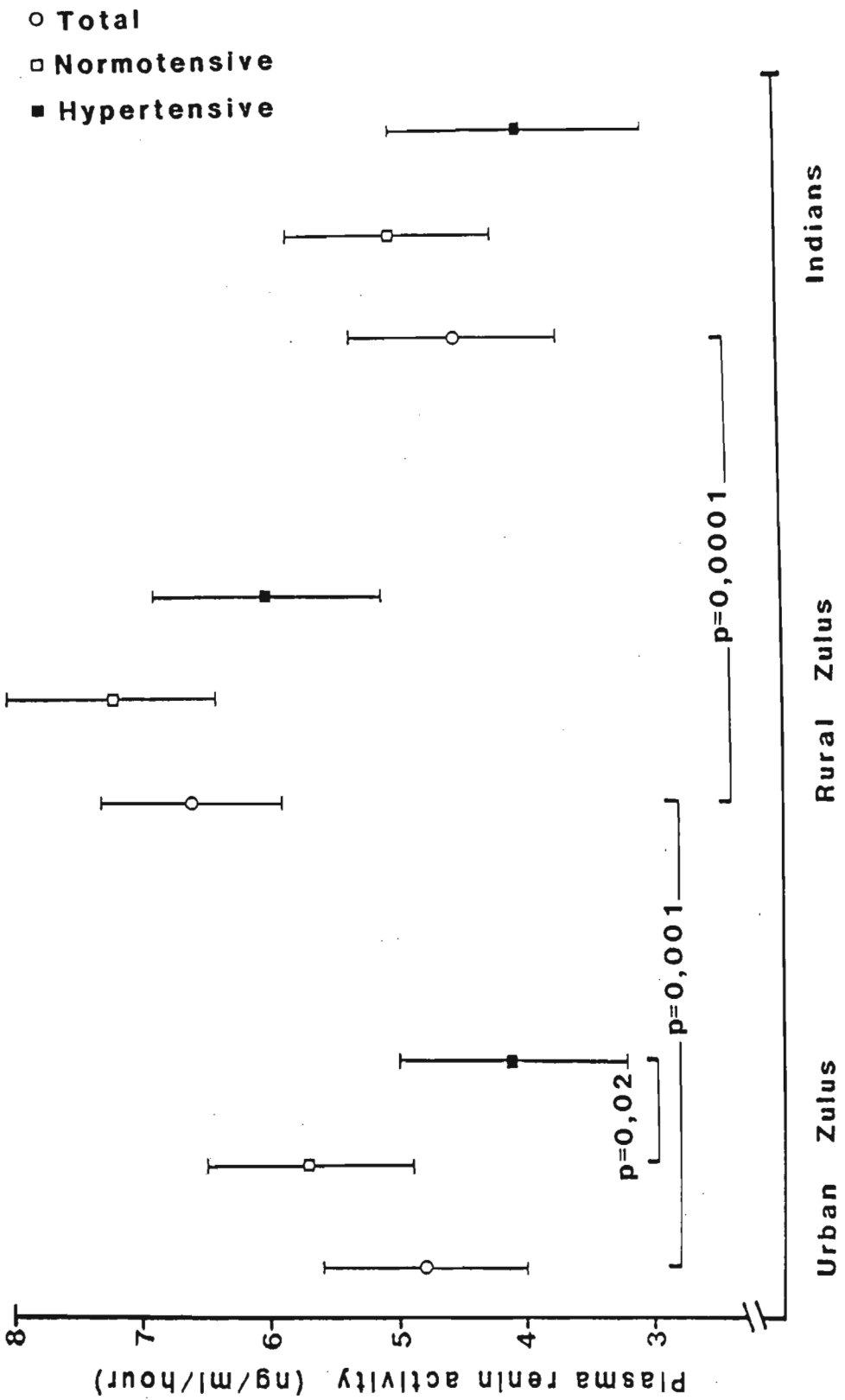


FIG.2

CORRELATION BETWEEN RENIN AND SYSTOLIC BLOOD PRESSURE IN URBAN ZULUS

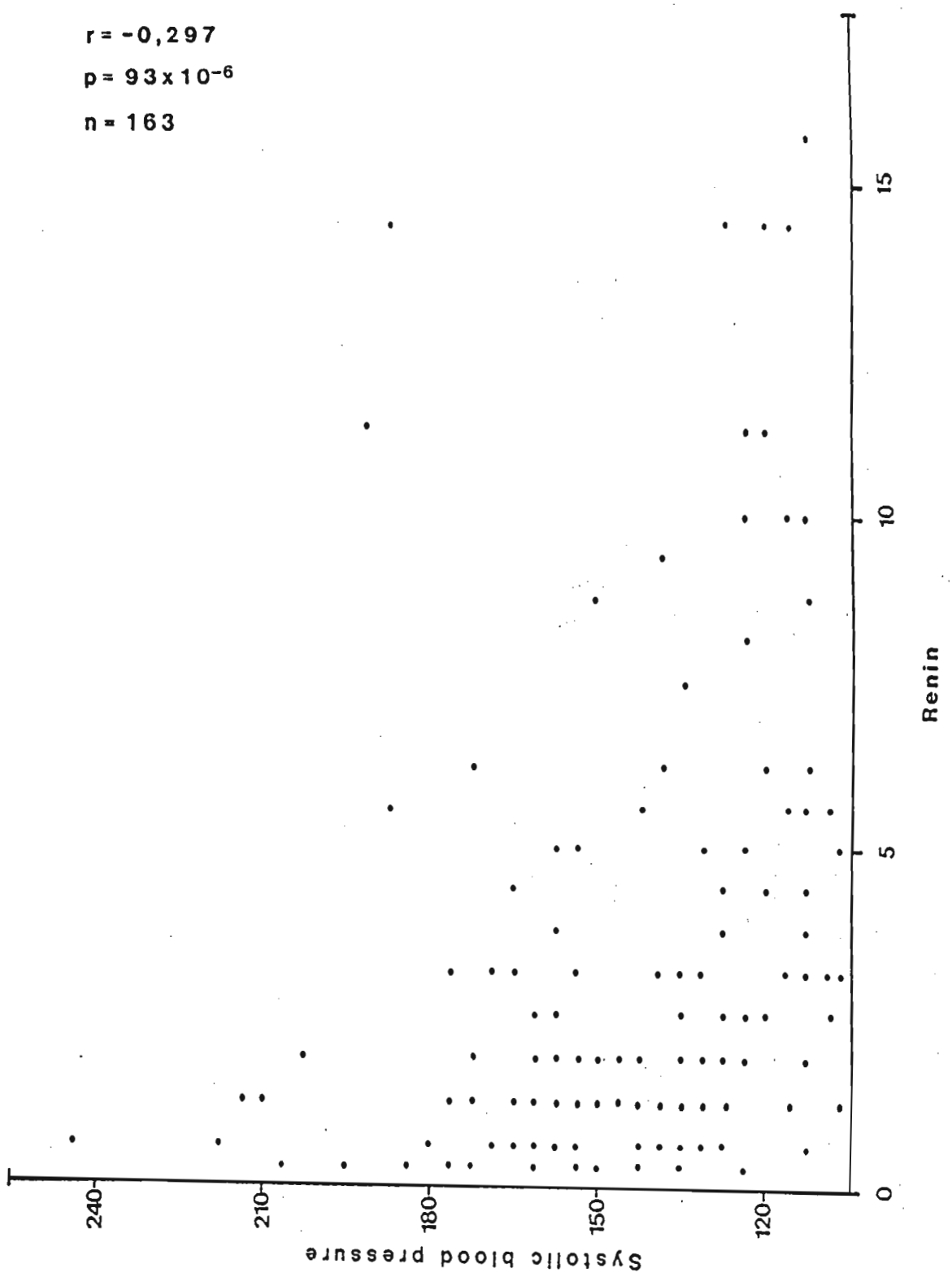


FIG.3

CORRELATION BETWEEN RENIN AND DIASTOLIC BLOOD PRESSURE IN URBAN ZULUS

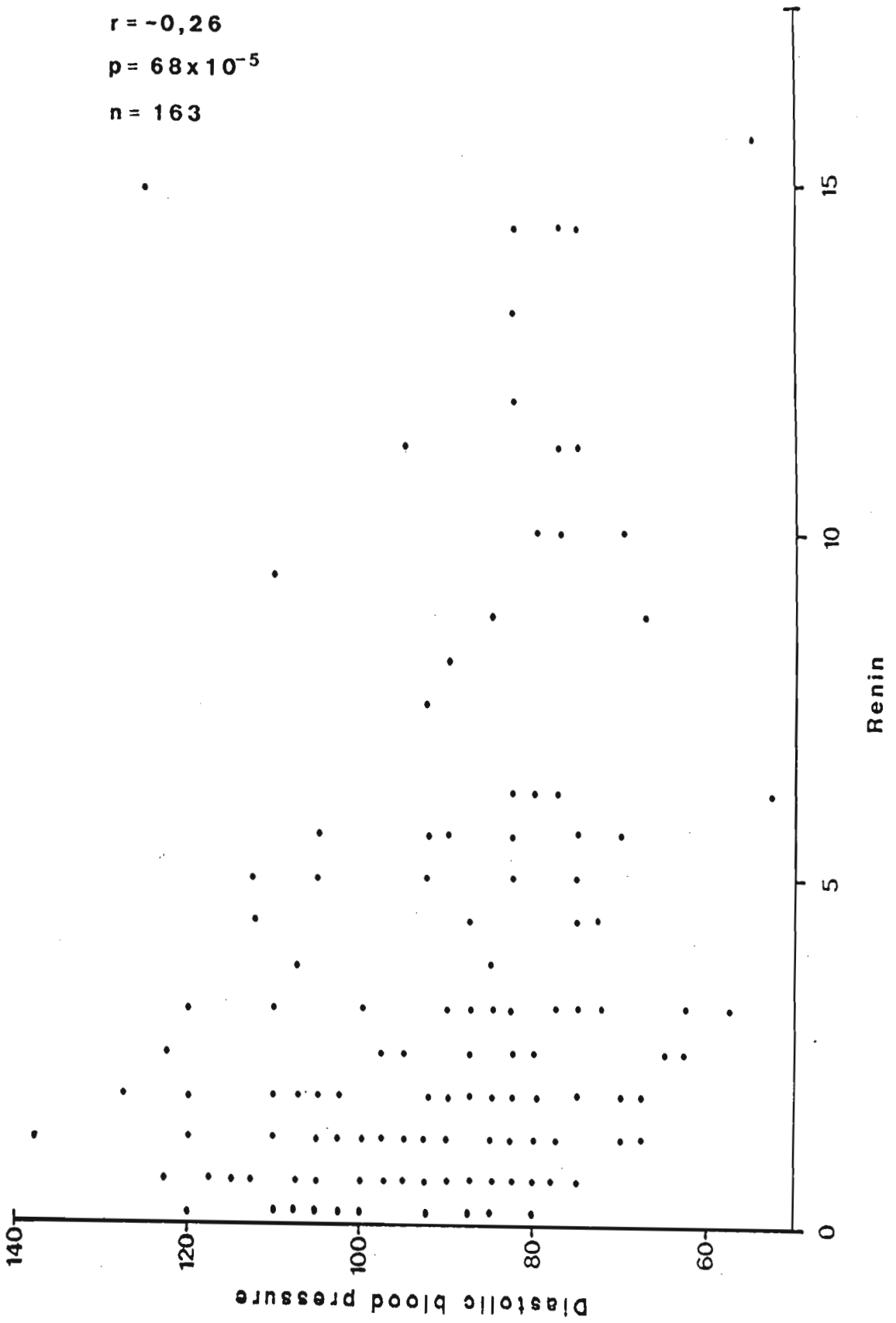


FIG.4

CORRELATION BETWEEN RENIN AND SYSTOLIC BLOOD PRESSURE IN INDIANS

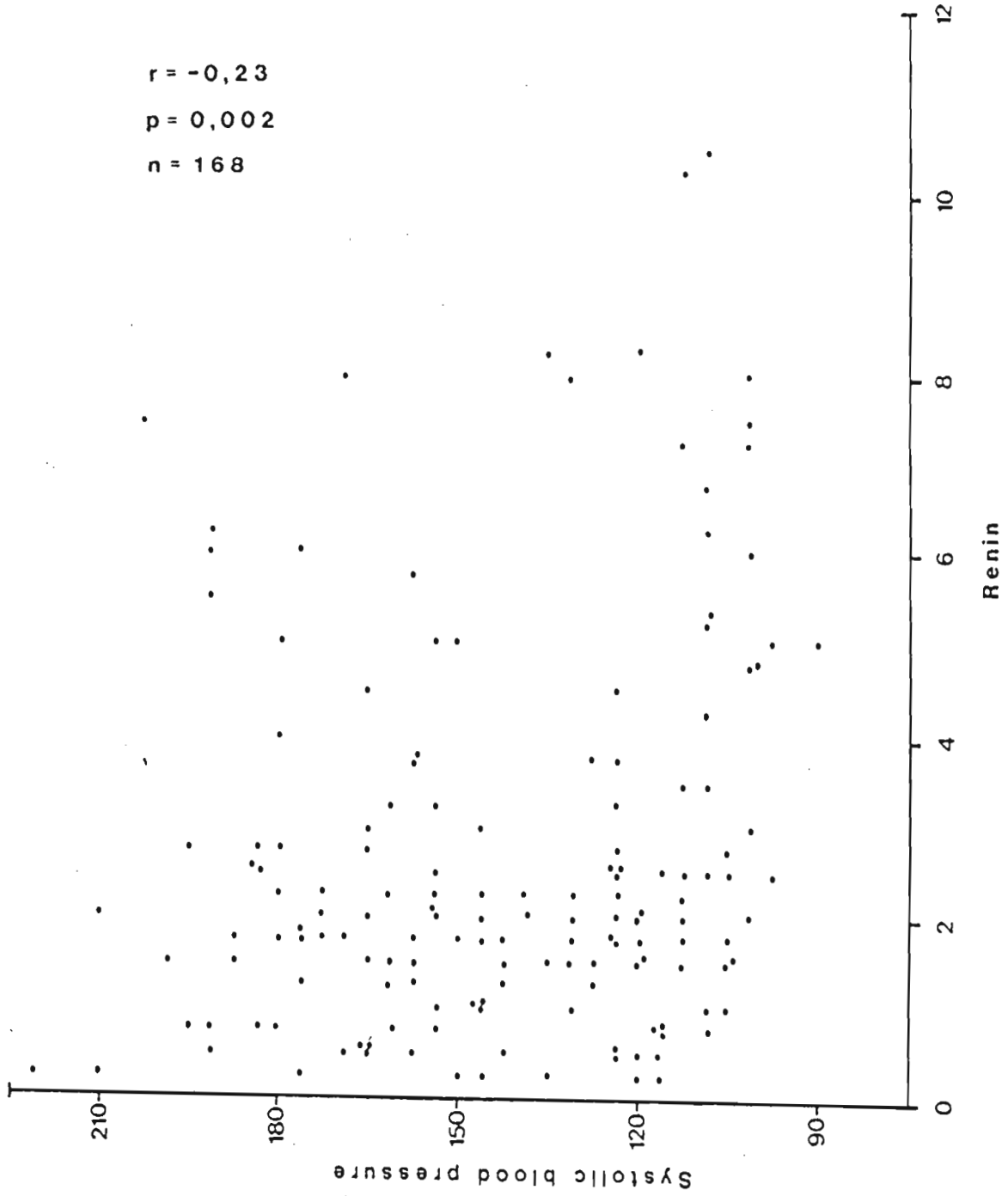
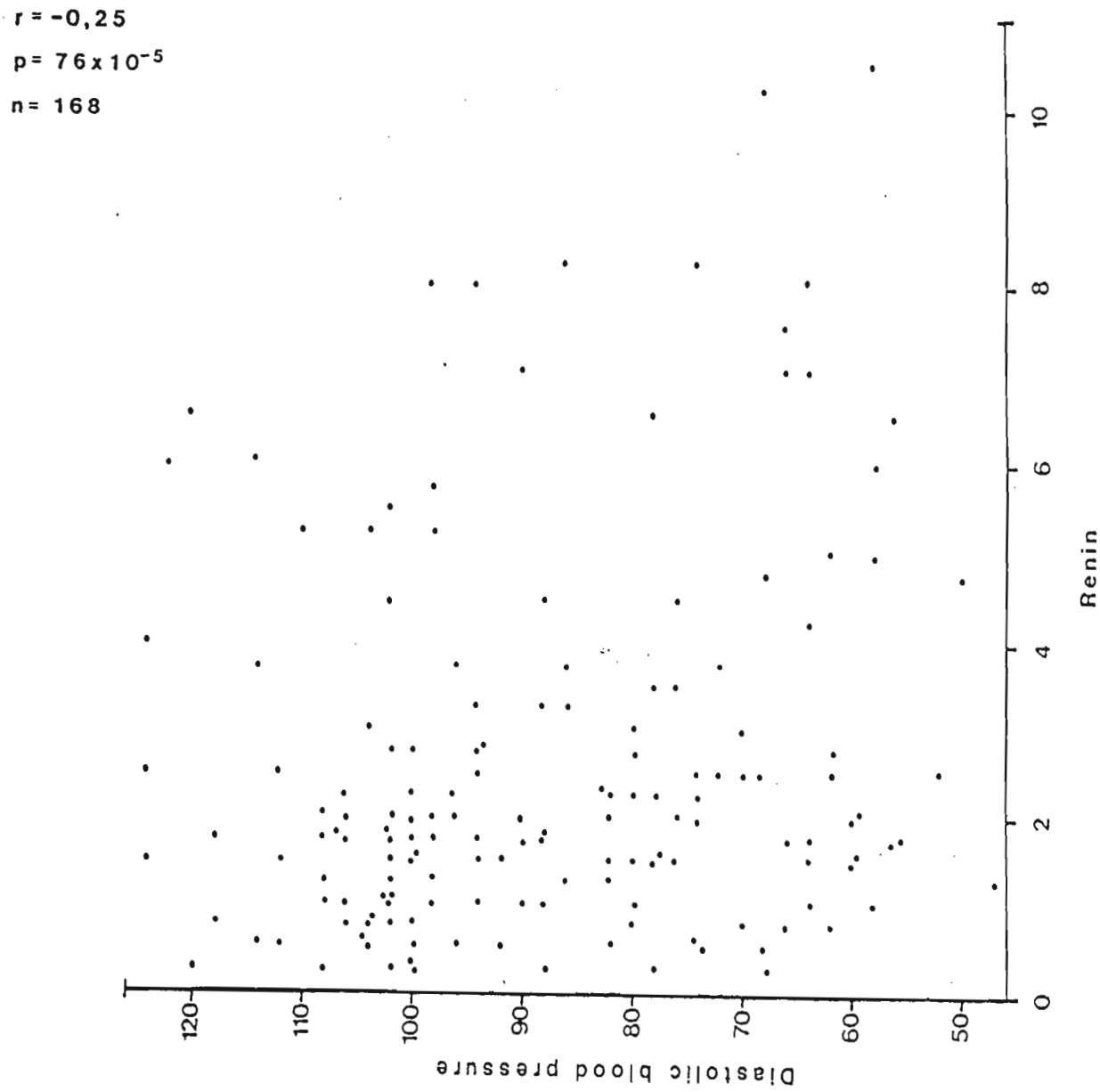


FIG.5

CORRELATION BETWEEN RENIN AND DIASTOLIC BLOOD PRESSURE IN INDIA



THE RELATIONSHIP BETWEEN RENIN AND SYSTOLIC BLOOD PRESSURE IN RURAL ZULUS

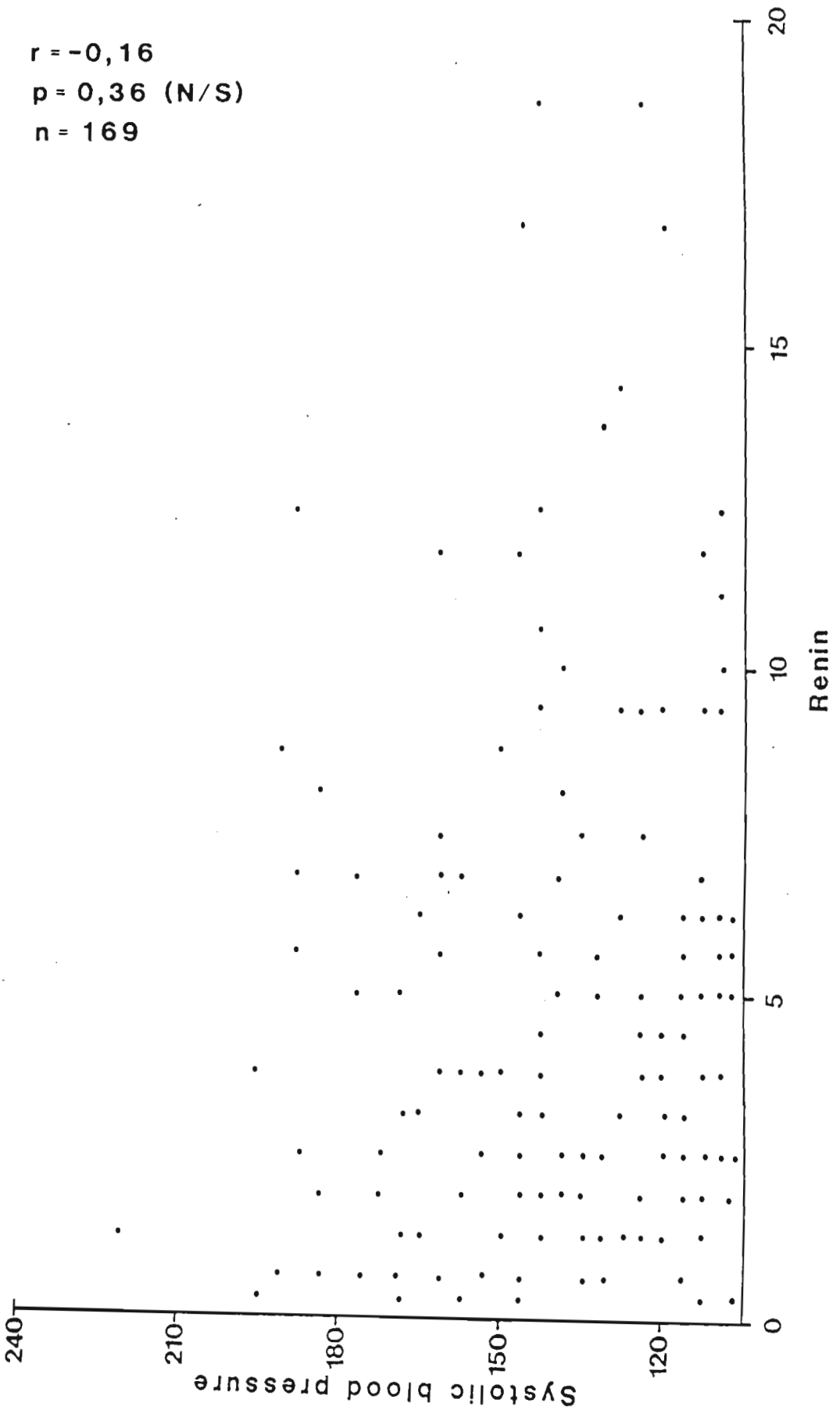
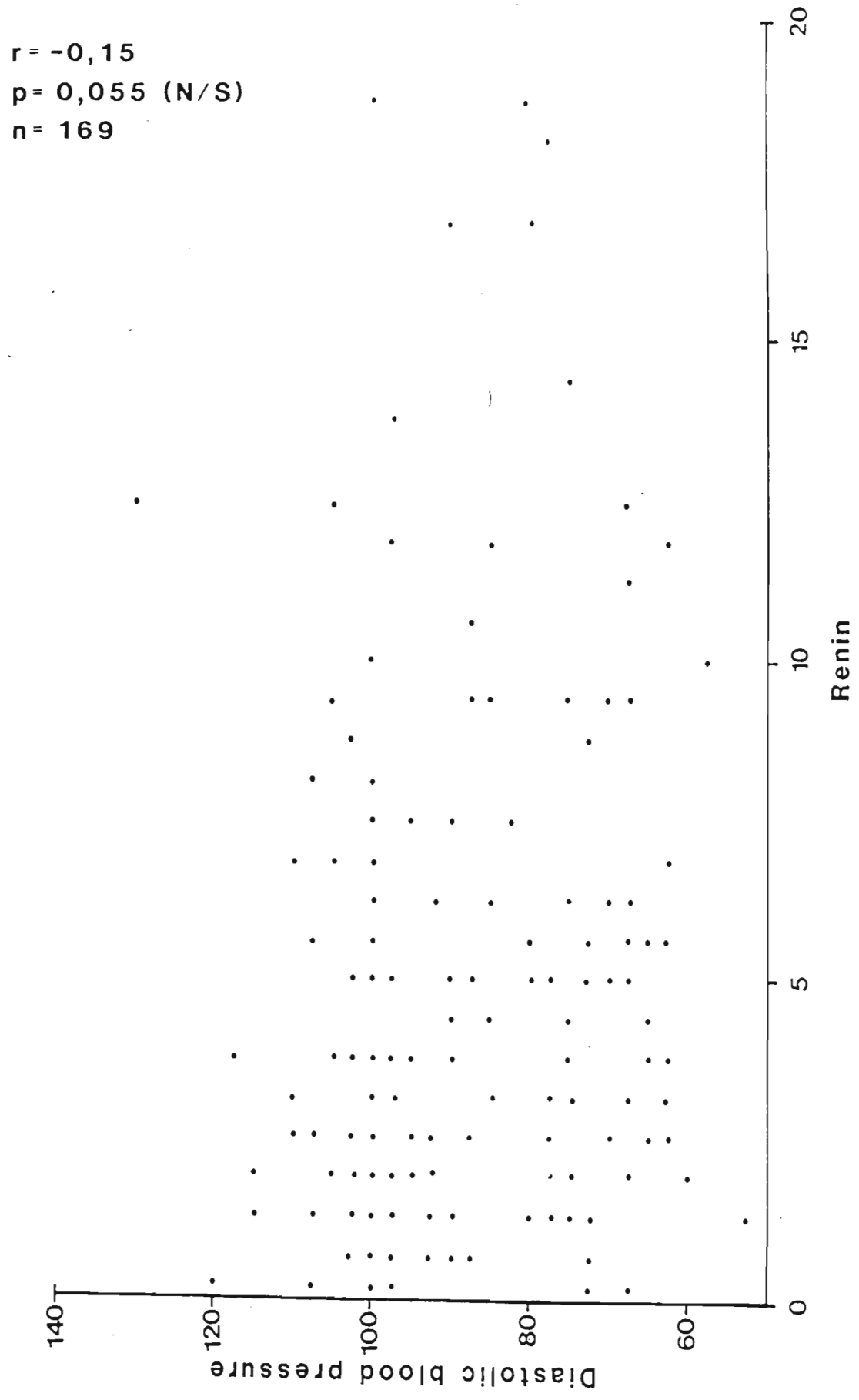


FIG. 7

THE RELATIONSHIP BETWEEN RENIN AND DIASTOLIC BLOOD PRESSURE IN RURAL ZULUS



CHAPTER 5

STIMULATED PLASMA RENIN ACTIVITY IN ZULUS AND INDIANS

INTRODUCTION

A true low renin state in an individual can only be identified when the plasma renin level fails to rise in response to a stimulus. This stimulus can be a low salt diet, upright posture or diuretics. Kaplan et al (1976) found that PRA 30 minutes after intravenous furosemide correlated closely with levels after 4 hours of upright posture or 3 days of low salt diet. Wallach et al (1975) suggest that PRA measured 5 hours after 60 mg of oral furosemide is a suitable screening test, however the duration of the test is too long and problems could arise from absorption and compliance. The intravenous furosemide test was used in this study to assess renin responsiveness.

This study was done in response to the findings in the previous chapter of a large number of hypertensive and normotensive subjects with low plasma renin levels (unstimulated).

METHOD

Seventy one patients were assessed. Sixteen were

hypertensive Zulus (12 males and 4 females); 12 were normotensive Zulus (10 males and 2 females); 24 were hypertensive Indians (6 males and 18 females) and 19 were normotensive Indians (8 males and 11 females). Details of patient selection are given in chapter 2 (b). Patients were on an unrestricted salt diet. Twenty milligrams of furosemide (Lasix, Hoechst) were given intravenously after blood had been collected for a basal plasma renin level. A blood sample was collected again after ½ hour for PRA. Patients remained seated for the duration of the test and blood was taken in the sitting position. PRA was measured by radio-immunoassay as outlined in appendix I.

STATISTICS

Data on PRA were skewed and were therefore expressed as $\log_{10}$. Paired Students t test was performed on the transformed data. Median values are used instead of mean because of the wide scatter of results with an uneven distribution and the relatively small number of patients.

RESULTS

The mean age in years $\pm$ SD of hypertensive Zulus was

41,7 ± 11,6; that of normotensive Zulus was 46,1 ± 10,9; that of hypertensive Indians was 46,0 ± 8,3 and that of normotensive Indians was 42,7 ± 6,7. Mean systolic and diastolic blood pressures are given in Table 1.

Hypertensive Zulus had a basal median PRA of 1,04 ng/ml/hour with a range of 0,037 to 17,4 and a median stimulated PRA of 0,98 ng/ml/hour with a range of 0,008 to 18,06. There was no significant difference in the PRA before and after stimulation in this group of patients.

Normotensive Zulu controls on the other hand had a significant increase in PRA after stimulation with furosemide ($p = 0,001$) (Figure 1). The median basal PRA of these subjects was 1,5 ng/ml/hour with a range of 0,29 to 4,4 and the median stimulated PRA was 2,1 ng/ml/hour with a range of 0,53 to 6,8.

The median unstimulated PRA of hypertensive Indians was 1,24 ng/ml/hour with a range of 0,29 to 12,9 as compared to a stimulated median of 1,49 ng/ml/hour and a range of 0,16 to 9,8. There was no significant increase in PRA after stimulation.

Normotensive Indians had a basal median PRA of 1,63 ng/ml/hour and the range was 0,54 to 7,5. The median PRA after stimulation was 3,12 ng/ml/hour with a range of 0,68 to 15,5. This increase was highly significant statistically ($p = 0,0001$) (Figure 2).

There was no ethnic difference in the renin response to furosemide.

There were no major side effects with the usage of intravenous furosemide. One normotensive Indian subject developed postural hypotension. She recovered very quickly without any treatment, but the test was abandoned in her. All subjects needed to void before the end of the test period.

DISCUSSION

Kaplan et al (1976) used a renin level of 0,5 ng/ml/hour or more as a normal value in response to intravenous furosemide for Blacks, and a value of 1 ng/ml/hour for Whites. In this study only 3 normotensive subjects (1 Indian and 2 Zulus) had plasma renin levels of less than 1 ng/ml/hour after stimulation with furosemide and none of them had a level of less than 0,5 ng/ml/hour.

Fifty percent of hypertensive Zulus and 33% of hypertensive Indians had a PRA of less than 1 ng/ml/hour after stimulation. If a cut off point of less than 0,5 ng/ml/hour is used, 25% of hypertensive Zulus and 21% of hypertensive Indians have low plasma renin levels.

This study clearly shows that hypertensive subjects have a hypo-responsive renin angiotensin system, as demonstrated by the failure of renin levels to rise in response to furosemide (natriuresis). This study also shows no ethnic difference in renin responsiveness in the groups studied.

Sagar et al (1982) like us, found no difference in the basal PRA of hypertensive and normotensive Indians (North India). However the increase in PRA after stimulation was greater in hypertensive than in normotensive subjects, whereas we found a significant increase in stimulated PRA in normotensive subjects only.

The lack of correlation between renin and aldosterone in hypertensive Indians and hypertensive urban Zulus (as discussed in chapter 4) is in keeping with the suggestion of an abnormality of this system.

It would be interesting to do this study in rural Zulus, as they appear to behave differently (as regards PRA) compared to urban Zulus. This was beyond the scope of this study.

TABLE 1

MEAN BLOOD PRESSURE ± SD (mmHg)

	SYSTOLIC	DIASTOLIC
Hypertensive Indians	175 ± 15	112 ± 6
Normotensive Indians	121 ± 9	81 ± 7
Hypertensive Zulus	169 ± 23	115 ± 9
Normotensive Zulus	121 ± 12	81 ± 5

FIGURE 1

The effect of stimulation on
plasma renin activity in Zulus

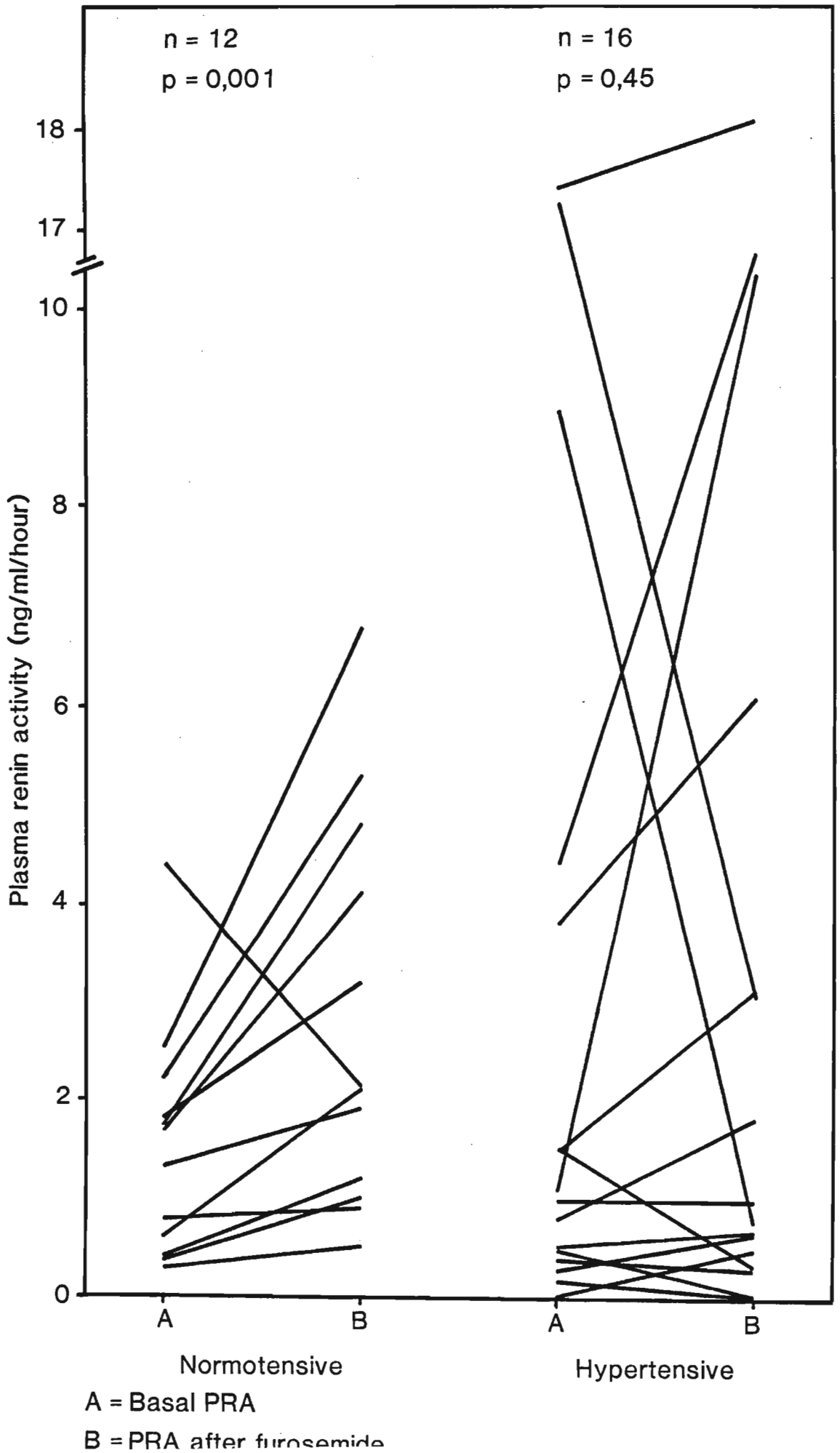
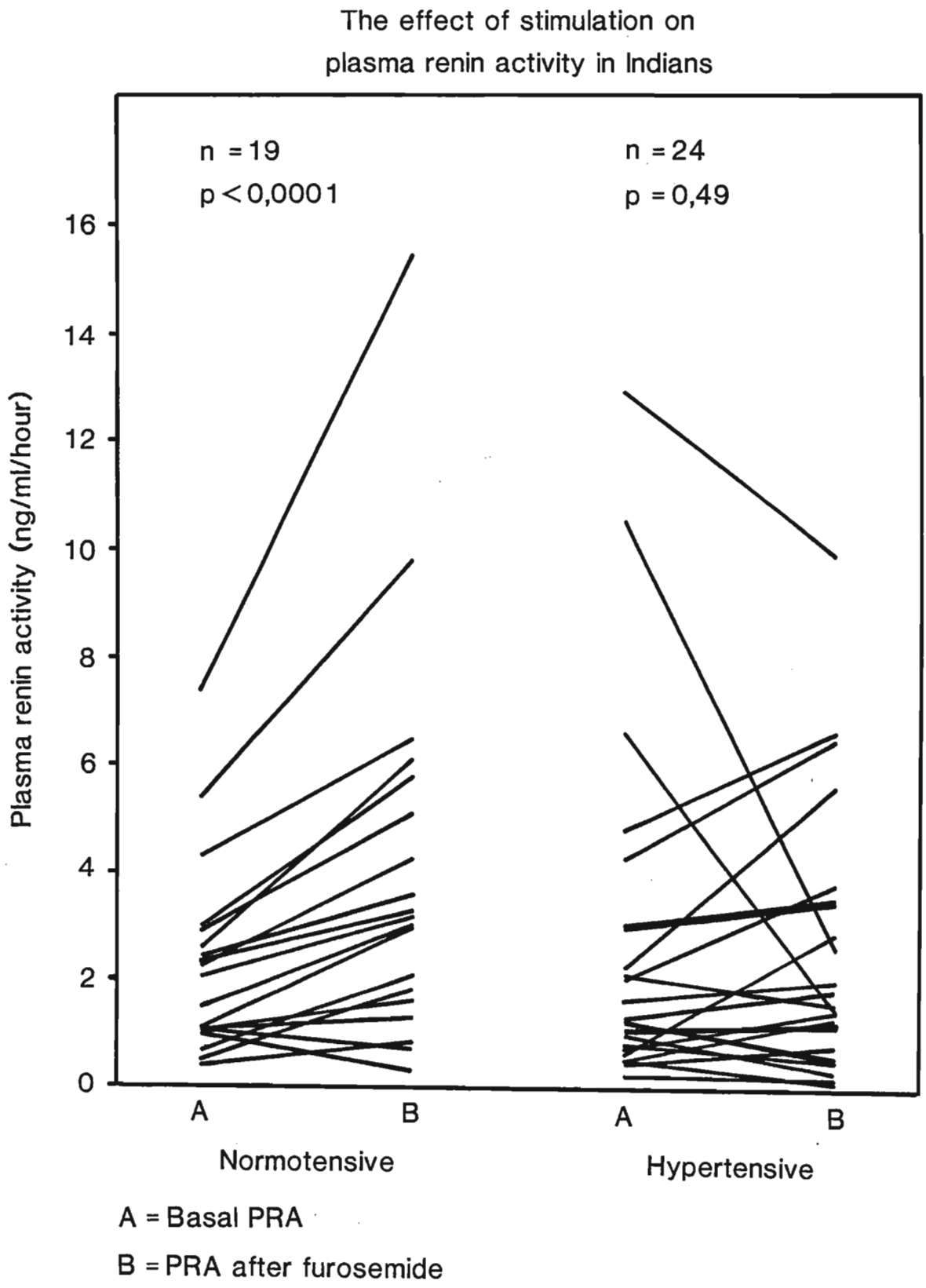


FIGURE 2


The effect of stimulation on
plasma renin activity in Indians
n = 19
p < 0,0001
n = 24
p = 0,49
Plasma renin activity (ng/ml/hour)
0
2
4
6
8
10
12
14
16
A
B
A
B
Normotensive
Hypertensive
A = Basal PRA
B = PRA after furosemide

CHAPTER 6

INTRALYMPHOCYTIC SODIUM AND POTASSIUM IN ZULUS AND INDIANS

INTRODUCTION

An association between sodium and hypertension is widely accepted but the mechanism by which sodium causes hypertension is not clear. Blaustein's hypothesis (1980; 1984) is that an increase in dietary sodium causes an increase in plasma volume which leads to an increase in extracellular fluid volume. This volume expansion acts on the hypothalamus to produce a natriuretic hormone which causes an increase in urinary sodium excretion and also an increase in intracellular sodium concentration. This, via sodium-calcium exchange, leads to an increase in intracellular calcium concentration and thereby an increase in vascular tone and peripheral resistance.

Although dietary sodium appears to have little effect on blood pressure in the population being studied, an abnormality in the handling of sodium at a cellular level remains a possibility.

PATIENTS AND METHODS

One hundred and seventy six patients were studied. They

comprised 39 normotensive Zulus (15 females and 24 males), 39 normotensive Indians (27 females and 12 males), 49 hypertensive Zulus (21 females and 28 males) and 49 hypertensive Indians (35 females and 14 males).

Further details are given in Chapter 2(b).

DIET

All subjects were on an unrestricted sodium diet.

STATISTICS

Data were analysed on an IBM 4331 computer. BMDP packages were used. Statistical methods used were one way analysis of variance, paired Student's t test and Pearson's correlation coefficient.

The test level was adapted according to Bonferrori (Neter J 1974).

RESULTS

Mean ages and Quetelet's indices are given in Table 1. Mean sitting and standing systolic, diastolic and mean arterial blood pressures are given in Tables 2 and 3.

INTRALYMPHOCYTIC SODIUM (ILNa)

The mean ILNa $\pm$ SD of hypertensive Zulus was 80,1 $\pm$ 17,3

mmol/kg dry cell weight compared to 71,1 ± 12,4 mmol/kg dry cell weight in normotensive controls. This difference was statistically significant (p = 0,007). There was however no difference in the ILNa of hypertensive and normotensive Indians. The mean ILNa ± SD was 73,5 ± 10,7 mmol/kg dry cell weight and 75,1 ± 18,3 mmol/kg dry cell weight in normotensive and hypertensive Indians respectively (Figure 1). There was no difference in the ILNa of the total Zulu group compared to the total Indian group.

There was a significant positive correlation between ILNa and the sitting and standing systolic, diastolic and mean arterial blood pressure in the total Zulu group only (Figures 3-8).

There was no association between ILNa and PRA (basal and stimulated).

INTRALYMPHOCYTIC POTASSIUM (ILK)

There was no difference in the ILK of hypertensive and normotensive subjects of both ethnic groups. There was also no difference between the total Indian group and the total Zulu group.

The mean ILK ± SD was 369 ± 42 mmol/kg dry cell weight in hypertensive Zulus; 372 ± 30 mmol/kg dry cell weight in normotensive Zulus; 362 ± 38 mmol/kg dry cell weight in hypertensive Indians and 366 ± 34 mmol/kg dry cell weight in normotensive Indians (Figure 2).

There was no correlation between ILK and blood pressure in all groups.

SERUM SODIUM

The mean serum sodium ± SD for normotensive Indians was 138,8 ± 2,3 mmol/l compared to 140,5 ± 2,1 mmol/l in hypertensive Indians. This difference was statistically significant ($p = 0,007$). The significance level becomes 5% when adapted to Bonferroni. The mean serum sodium of hypertensive Zulus was 140,1 ± 2,5 mmol/l compared to 139,0 ± 3,2 mmol/l in normotensive Zulus ($p = 0,01$). This difference was not statistically significant according to the Bonferrori test.

Serum sodium was inversely associated with ILNa in normotensive Zulus only ($r = -0,45$; $p < 0,05$).

SERUM POTASSIUM

There was no significant difference in the serum potassium of hypertensive and normotensive subjects of both ethnic groups. The mean serum potassium ± SD was 3,9 ± 0,4 mmol/l in hypertensive Zulus; 3,9 ± 0,4 mmol/l in normotensive

Zulus; 3,6 ± 0,4 mmol/l in hypertensive Indians and 3,7 ± 0,3 mmol/l in normotensive Indians. The serum potassium of hypertensive Indians was significantly lower than that of hypertensive Zulus ($p = 0,005$; significance level is 5% according to the Bonferrori test).

There was no correlation between serum potassium and ILK in all groups.

DISCUSSION

An increase in lymphocyte sodium concentration in hypertensive Zulus described in this study is similar to the findings of other studies which have reported an increase in white blood cell sodium in hypertensive patients (Ambrosioni 1979; Edmonson 1975; Araoye 1978). This has been described in black and White subjects. Black subjects are also reported to have higher cell sodium levels compared to Whites (Munro Faure 1971).

The method used for determining ILNa and ILK was that described by Ambrosioni et al, there are however many differences in the results. Ambrosioni et al (1979) found little overlap in the ILNa of hypertensive and normotensive patients whereas this study showed a very large overlap in results (Figure 9 and 10). Other workers have also described a wide overlap in the range of cell sodium in hypertensive and normotensive subjects (Tedde 1983; Losse 1981). It was not possible to separate the normotensives into those with and

those without a family history of hypertension as the majority of subjects were unable to answer this question (due to socio economic factors). This factor could explain high values in normotensive subjects but not low ILNa values in hypertensive subjects.

This study showed a positive correlation between ILNa and blood pressure in the total Zulu group. Although the correlation coefficient was small and the confidence interval only 95%, the association was fairly consistent in that it was statistically significant for both sitting and standing systolic, diastolic and mean arterial blood pressures. Ambrosioni et al (1979) also found a positive correlation between ILNa and blood pressure, and in contrast to this study, they found this association only in hypertensive subjects. Tedde et al (1983) also found a positive correlation between blood pressure and intracellular sodium (red cell) in hypertensive subjects. Ringel et al (1984) however found no association between individual blood pressures and red cell sodium concentration.

A surprising finding was that in Indians there was no difference in the ILNa of hypertensive and normotensive subjects and there was also no association with blood pressure. To date studies on subjects of Indian origin are not available for comparison. A difference in experimental conditions cannot explain these observations as both groups were studied

concurrently using the same technique and the same laboratory. This ethnic difference is probably genetic in origin and may be related to differences in transport systems.

ILK was similar in hypertensive and normotensive subjects of both ethnic groups. This finding is similar to that of other workers (Tedde 1983; Aderounmu 1979).

Serum sodium was higher in hypertensive subjects compared to normotensive controls. This difference was greater in Indians compared to Zulus. Aderounmu et al (1979) described higher serum sodium and lower serum potassium levels in hypertensive Africans. Unlike them we found no difference in the serum potassium concentration of hypertensives and normotensives of both ethnic groups. Cohen et al (11) on the other hand found no difference in the serum sodium and potassium of hypertensive and normotensive blacks in Johannesburg.

The findings in summary are that hypertensive Zulus have a higher ILNa concentration compared to normotensive Zulus, and the hypertensives also have a slight increase in serum sodium levels (difference not significant according to Bonferrori), whereas hypertensive and normotensive Indians have similar ILNa concentrations and the hypertensives have significantly higher serum sodium levels.

TABLE 1

AGE AND QUETELET'S INDEX

(RANGE AND MEAN ± SD)

	AGE		QUETELETS INDEX	
	RANGE	MEAN ± SD	RANGE	MEAN ± SD
HYPERTENSIVE ZULUS	21 - 63	43,7 ± 11,2	22 - 53	30,7 ± 7,8
NORMOTENSIVE ZULUS	26 - 64	41,2 ± 9,2	17 - 40	27,6 ± 6,1
HYPERTENSIVE INDIANS	29 - 63	46,8 ± 9,1	17 - 40	26,4 ± 4,9
NORMOTENSIVE INDIANS	30 - 65	42,7 ± 8,4	18 - 35	25,9 ± 4,1

TABLE 2

MEAN ± SD SITTING SYSTOLIC, DIASTOLIC AND MEAN ARTERIAL BLOOD PRESSURES

	SYSTOLIC	DIASTOLIC	MEAN ARTERIAL
HYPERTENSIVE ZULUS	166,3 ± 21,5	107,3 ± 9,2	126,9 ± 12,1
NORMOTENSIVE ZULUS	117,8 ± 10,1	73,5 ± 7,1	88,2 ± 7,3
HYPERTENSIVE INDIANS	170,1 ± 21,0	107,3 ± 7,1	128,2 ± 9,6
NORMOTENSIVE INDIANS	118,8 ± 9,6	73,8 ± 7,6	88,8 ± 7,5

TABLE 3

MEAN ± SD STANDING SYSTOLIC, DIASTOLIC AND MEAN ARTERIAL BLOOD PRESSURES

	SYSTOLIC	DIASTOLIC	MEAN ARTERIAL
HYPERTENSIVE ZULUS	168,4 ± 21	113,7 ± 9,1	131,9 ± 11,6
NORMOTENSIVE ZULUS	121,1 ± 10,4	79,2 ± 6,2	93,2 ± 6,6
HYPERTENSIVE INDIANS	169,9 ± 19,9	112,1 ± 7,2	131,4 ± 9,9
NORMOTENSIVE INDIANS	121,3 ± 10,6	79,5 ± 7,2	93,5 ± 7,7

FIGURE 1

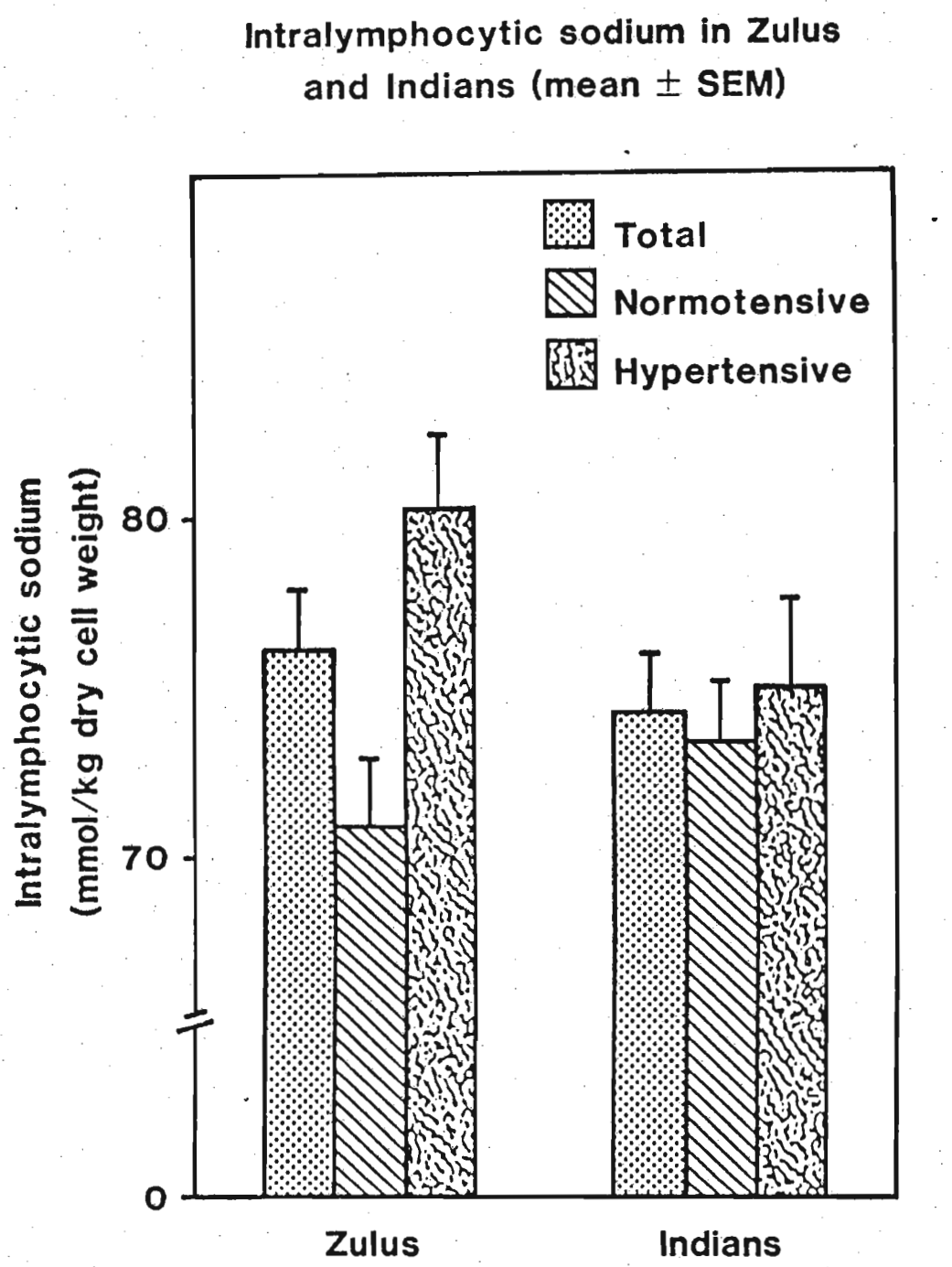

Intralymphocytic sodium in Zulus and Indians (mean ± SEM)
Total
Normotensive
Hypertensive
Intralymphocytic sodium (mmol/kg dry cell weight)
80
70
0
Zulus
Indians

FIGURE 2

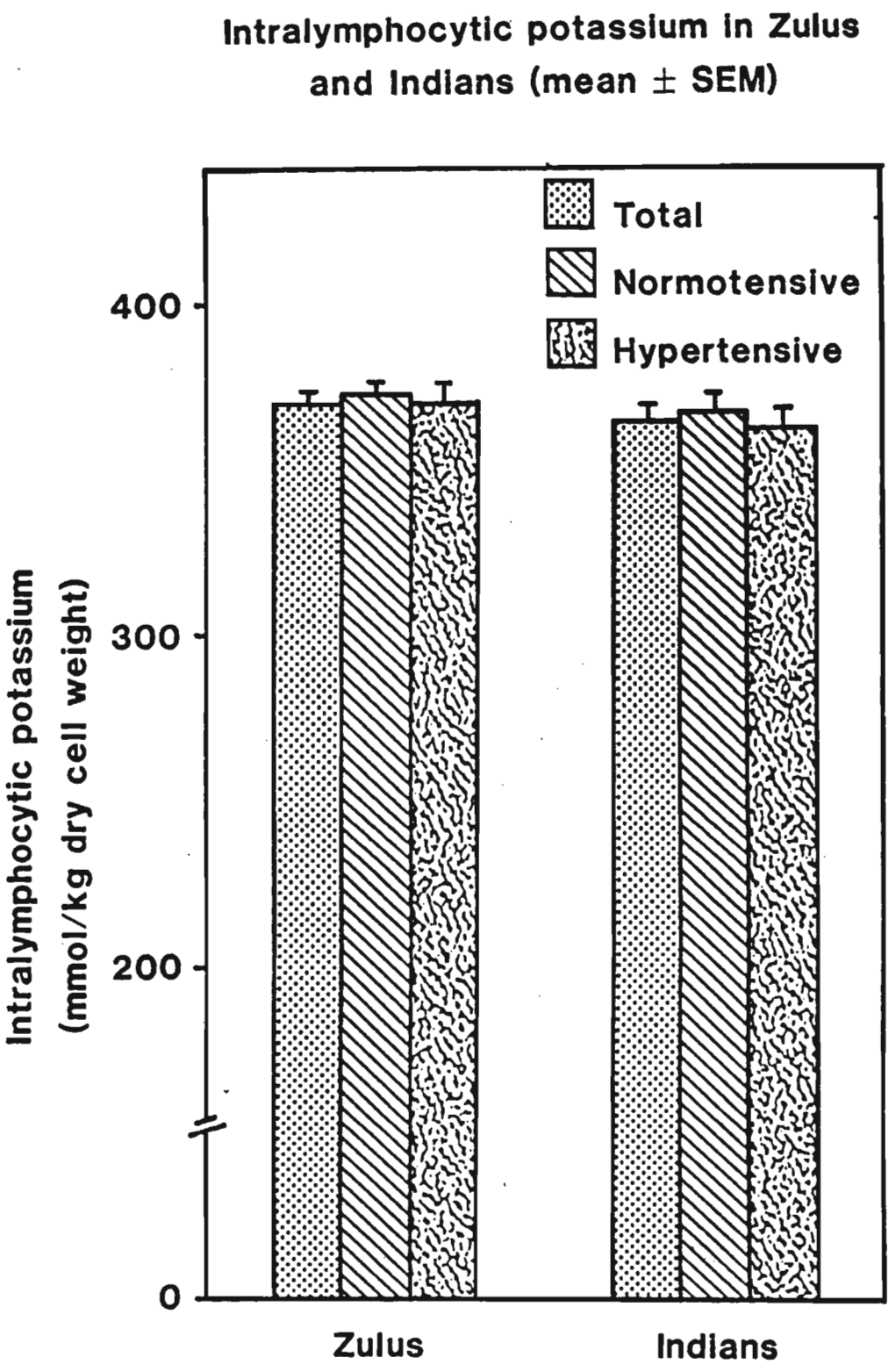

Intralymphocytic potassium in Zulus and Indians (mean ± SEM)
Total
Normotensive
Hypertensive
Intralymphocytic potassium (mmol/kg dry cell weight)
400
300
200
0
Zulus
Indians

FIGURE 3

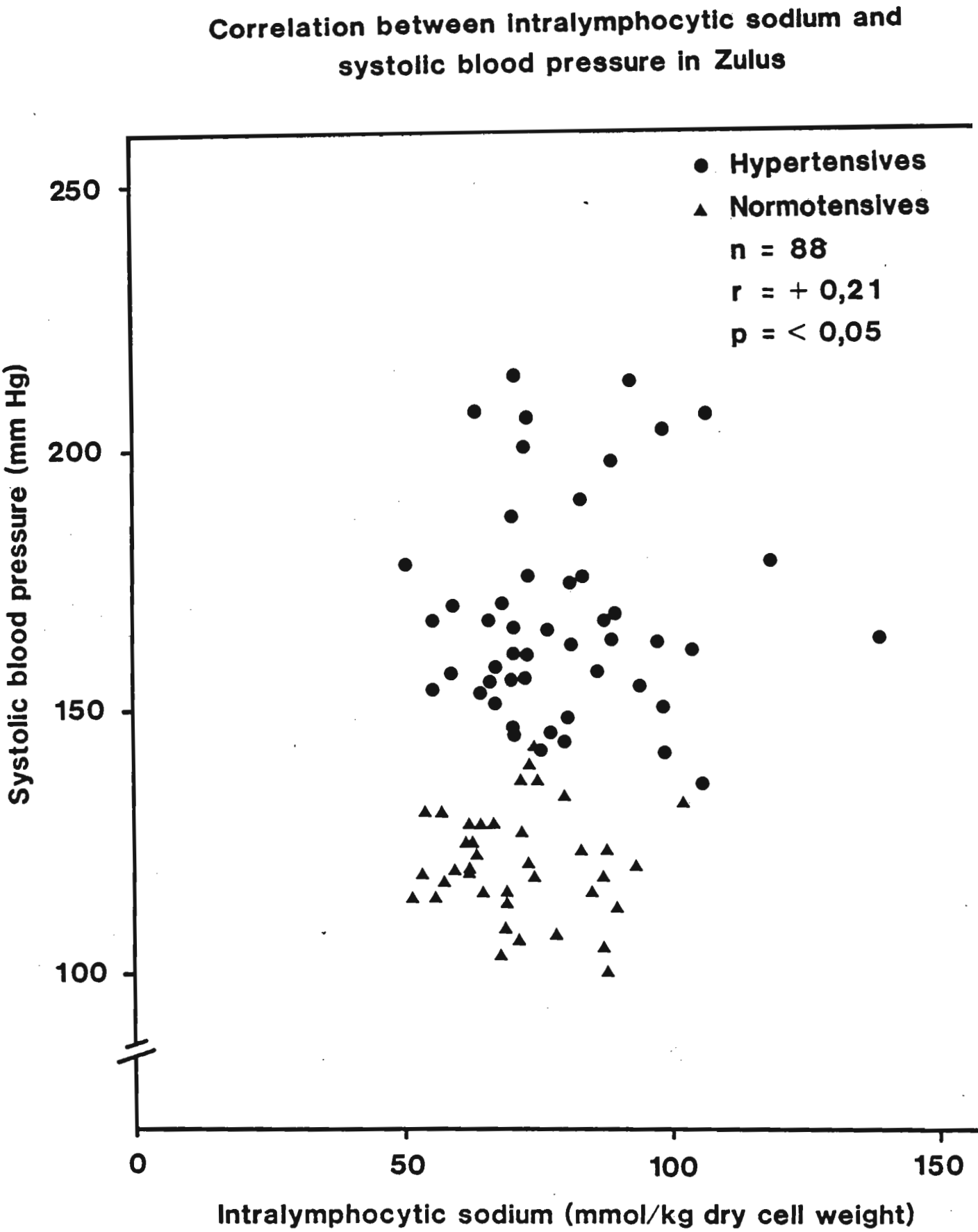

Correlation between intralymphocytic sodium and systolic blood pressure in Zulus
Hypertensives
Normotensives
n = 88
r = + 0,21
p = < 0,05
Systolic blood pressure (mm Hg)
250
200
150
100
0
50
100
150
Intralymphocytic sodium (mmol/kg dry cell weight)

FIGURE 4

Correlation between intralymphocytic sodium and diastolic blood pressure in Zulus

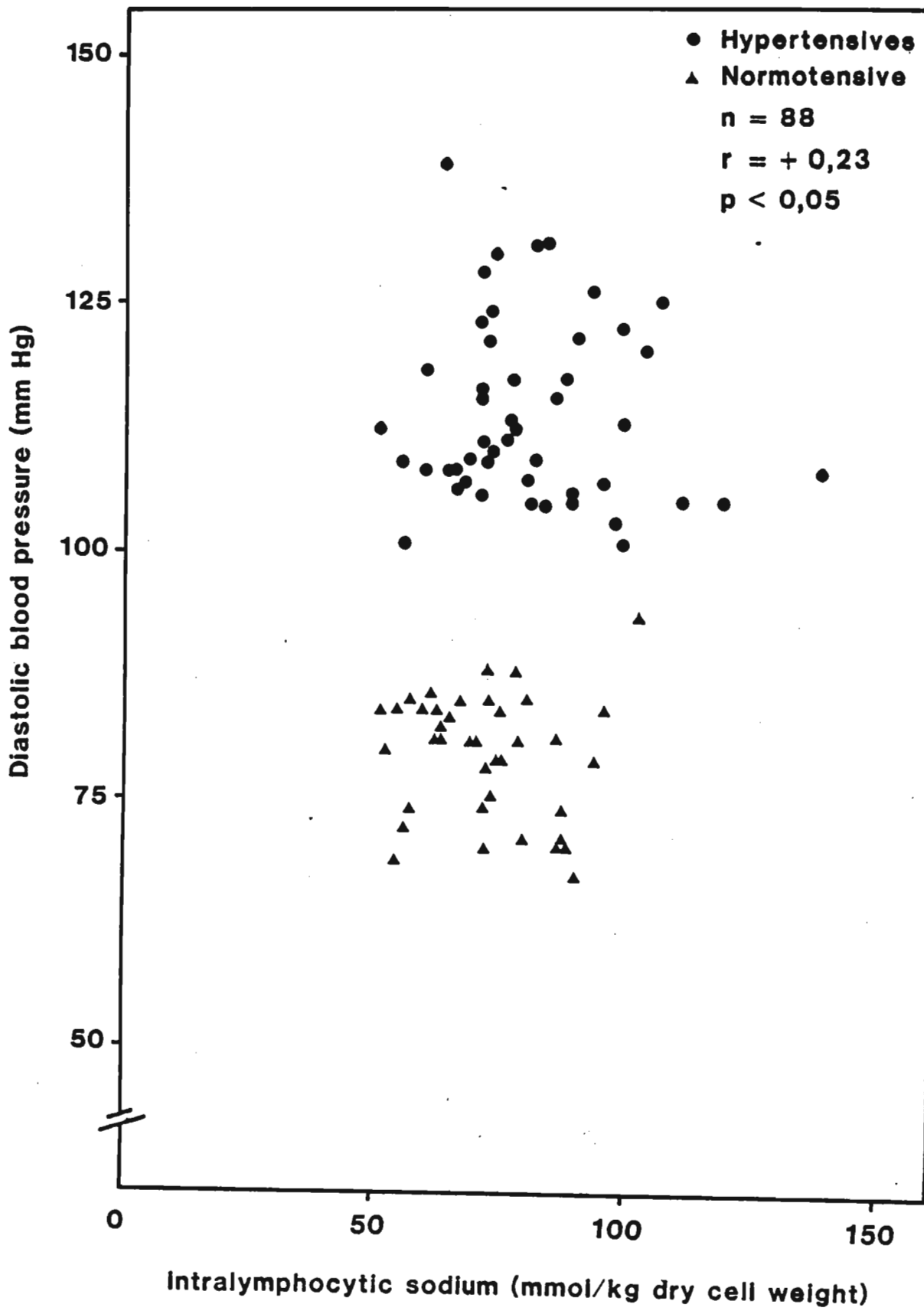


FIGURE 5 **Correlation between intralymphocytic sodium and mean arterial pressure in Zulus**

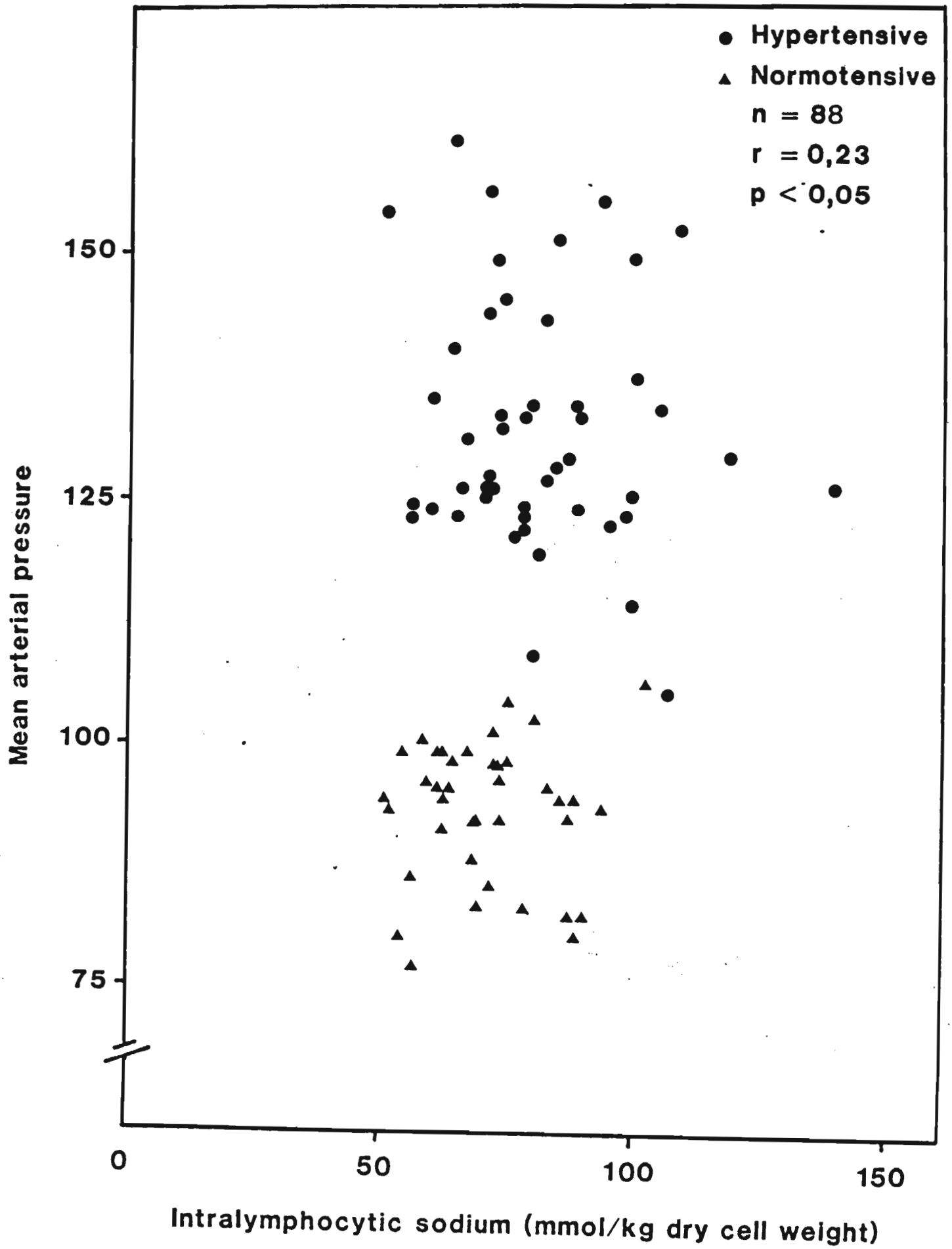


FIGURE 6

The relationship between intralymphocytic sodium and systolic blood pressure in Indians

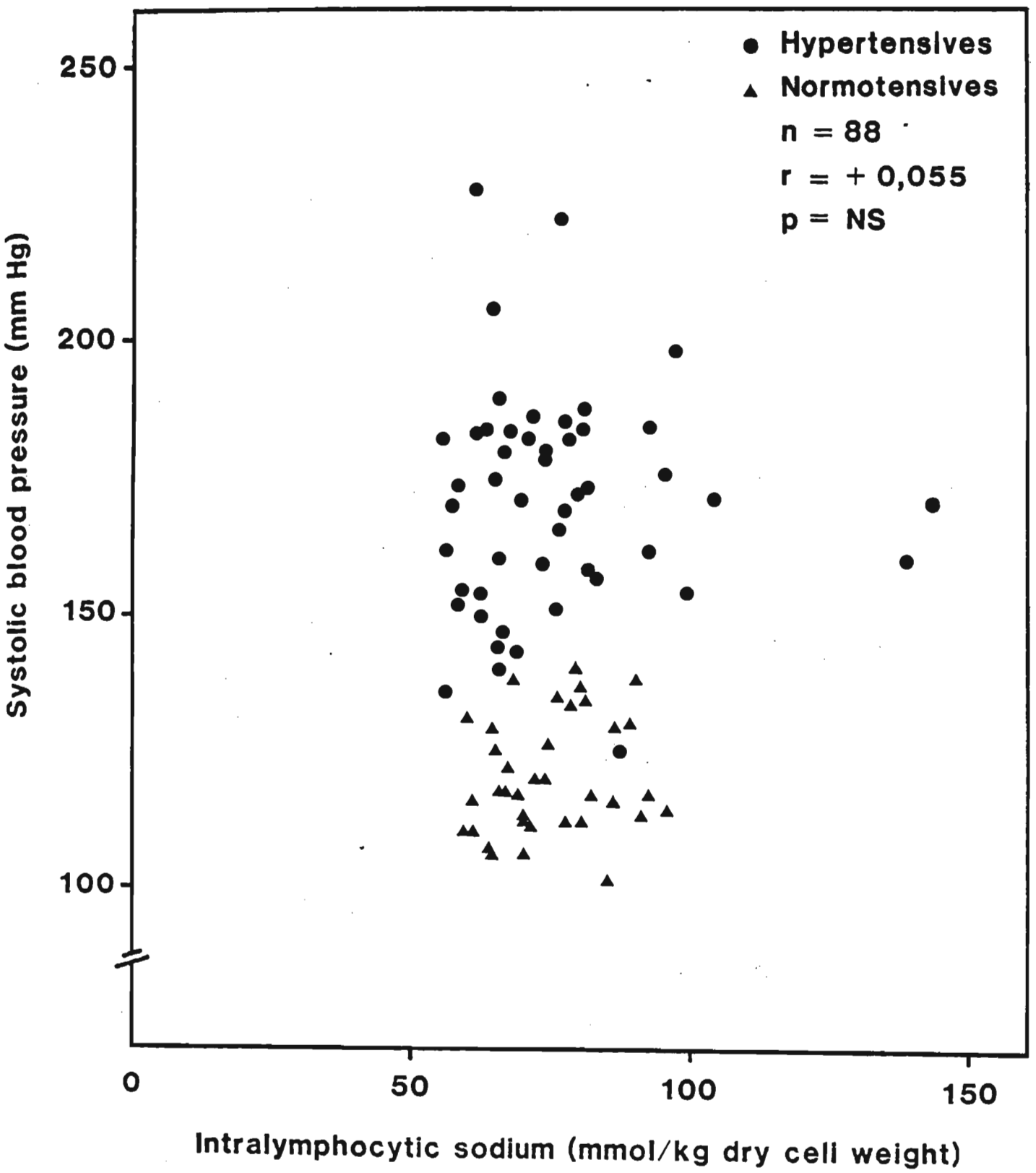


FIGURE 7

The relationship between diastolic blood pressure and intralymphocytic sodium in Indians

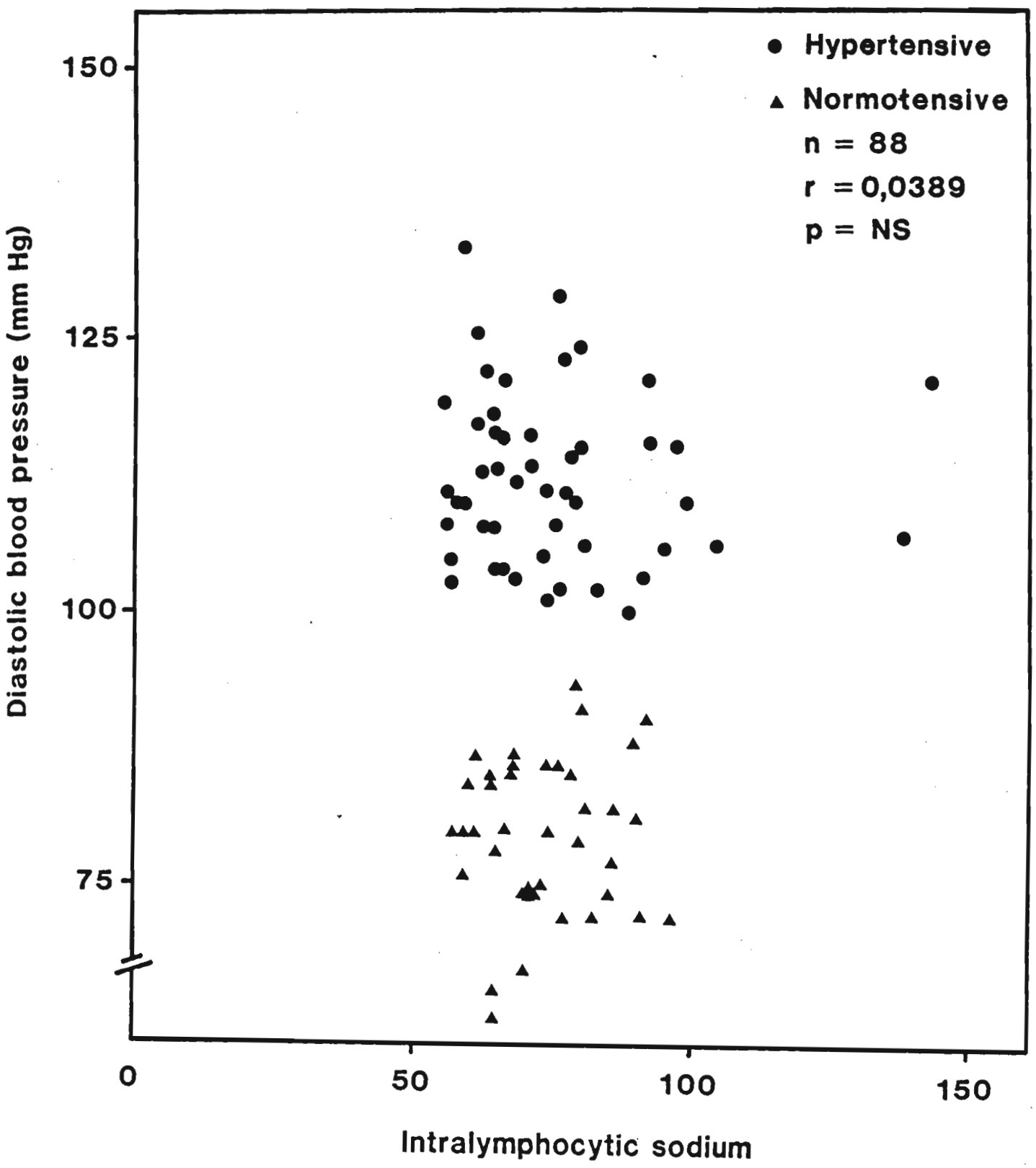


FIGURE 8

The relationship between mean arterial blood pressure an intralymphocytic sodium in Indians

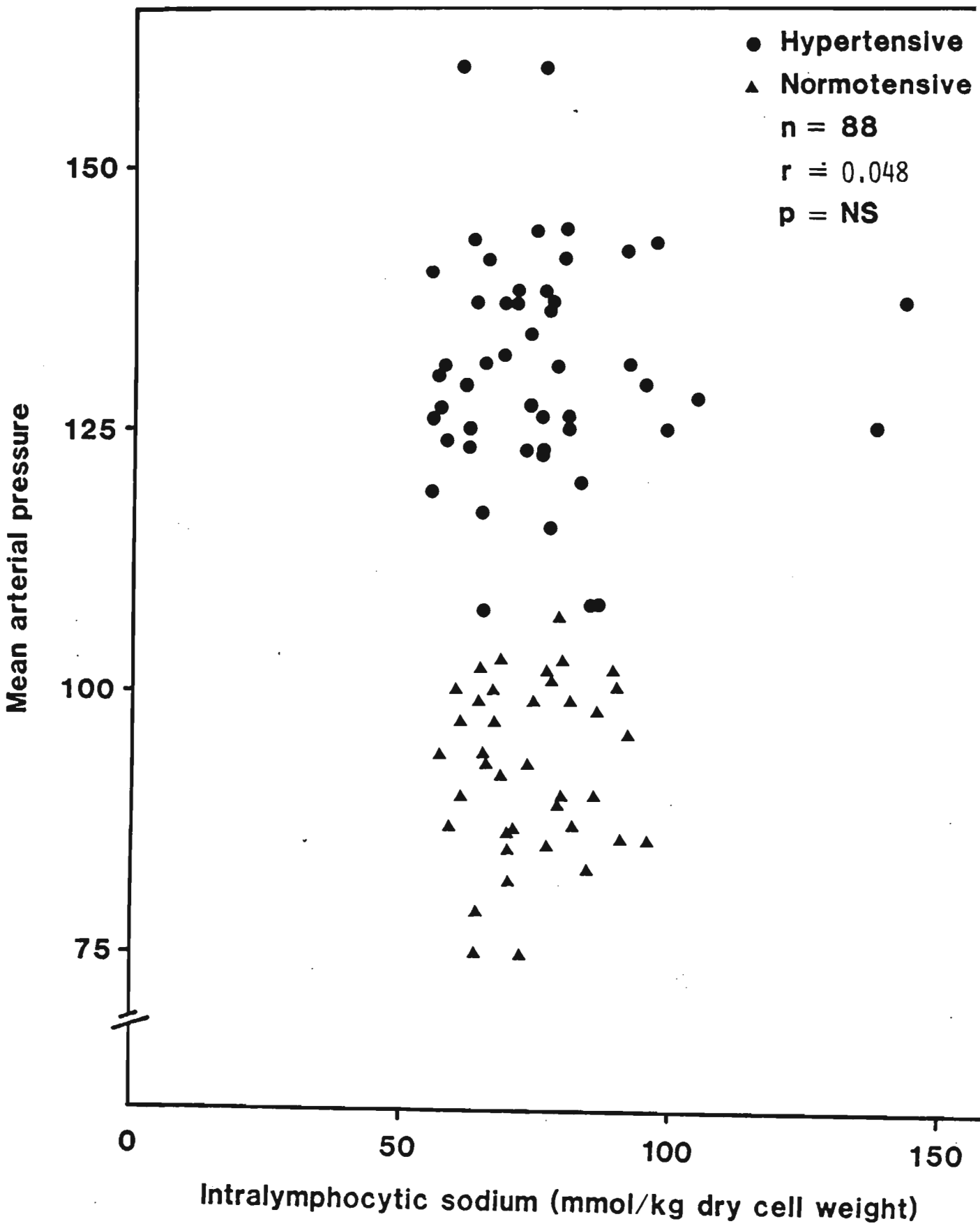


FIGURE 9

Individual values of intralymphocytic sodium in hypertensive and normotensive Indians

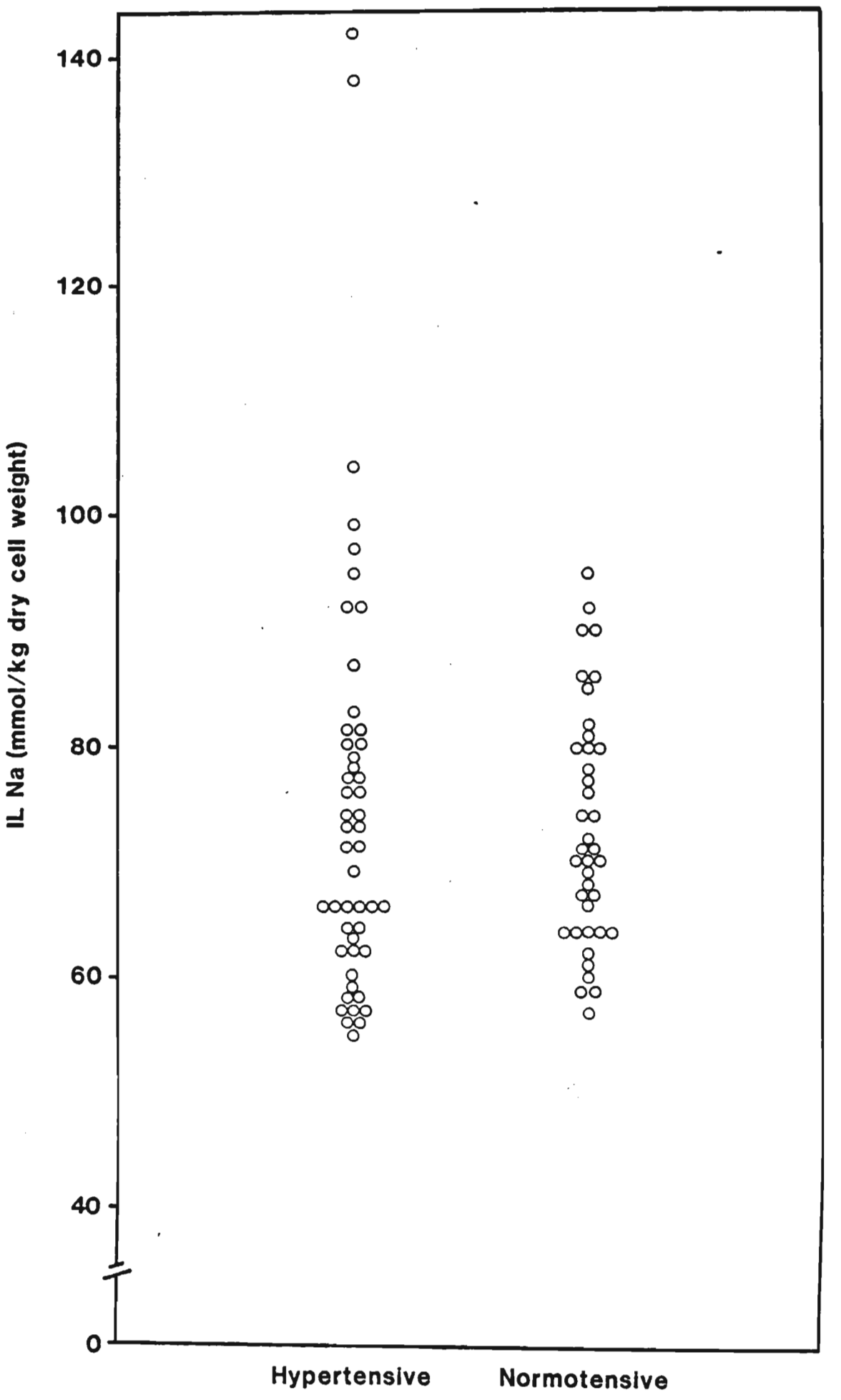
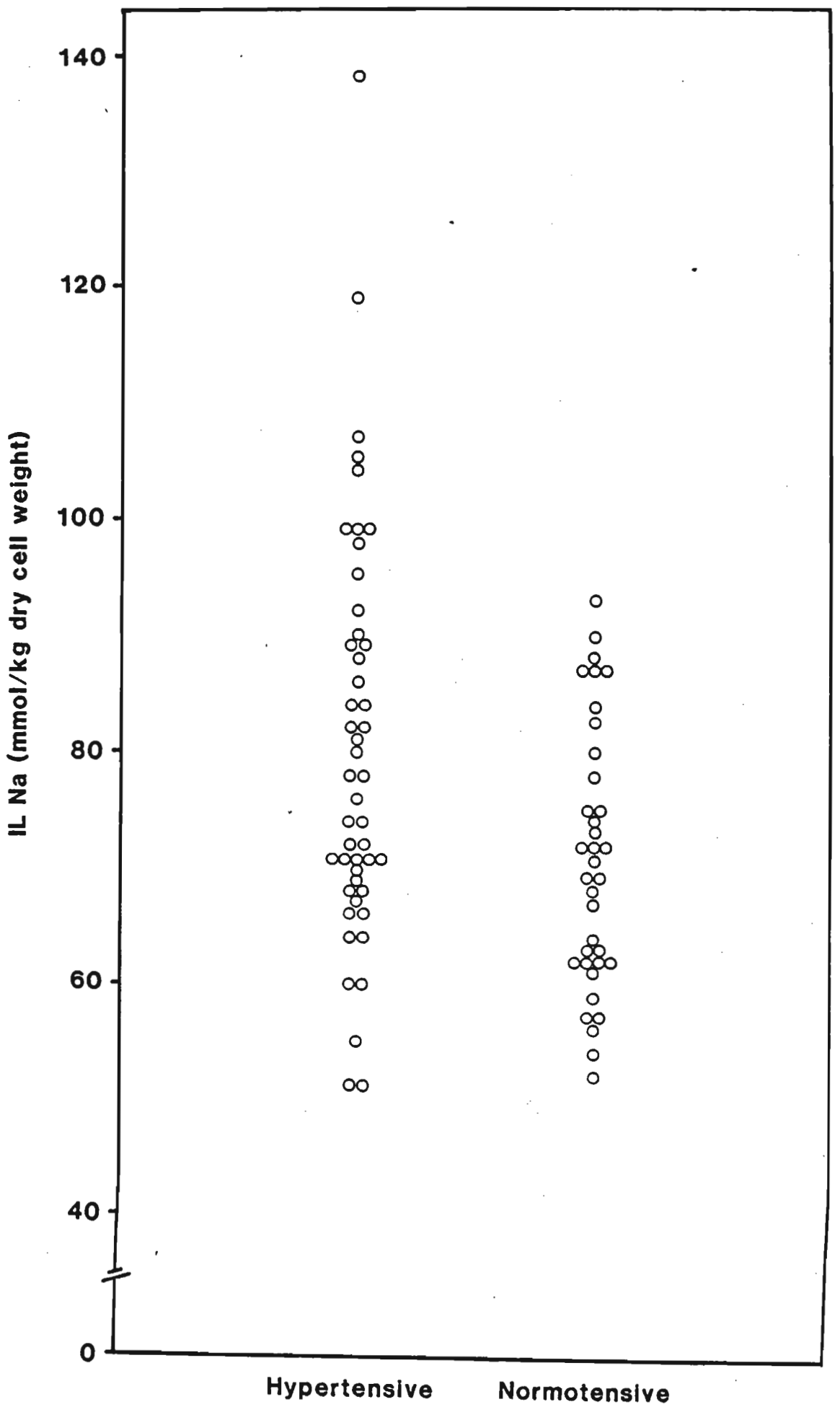


FIGURE 10

Individual values of intralymphocytic sodium in hypertensive and normotensive Zulus



CHAPTER 7

THE EFFECT OF FUROSEMIDE ON INTRALYMPHOCYTIC SODIUM AND POTASSIUM

INTRODUCTION

Furosemide causes a diuresis by inhibiting the reabsorption of sodium and water in the thick ascending loop of Henle and in the proximal convoluted tubule. It was previously believed that furosemide inhibited active Na-K transport in the kidney by reducing aerobic generation of adenosine triphosphate (ATP) and by inhibiting the Na-K ATPase system (Nechay 1977). Recent literature suggests that active chloride transport is the primary event and sodium transport down the electrochemical gradient is passive (Broocks 1978; Burg 1976).

A furosemide sensitive ouabain resistant Na-K cotransport system has been described in human red cells and leucocytes, and various abnormalities of this system have been described. Dagher et al (1980) found the Na-K cotransport system to be virtually absent in subjects with essential hypertension. Tuck et al (1984) on the other hand found red cell Na-K cotransport to be lower in Black hypertensives compared to Black controls, and higher in White hypertensives compared to White controls.

Garay et al (1981) also found cotransport to be reduced in Ivory Coast Blacks. Davidson et al (1982) however found no ethnic difference in red cell Na-K cotransport.

The aim of this study was to assess the effect of furosemide on intracellular sodium and potassium in Zulus and Indians, and to assess the difference in response (if any) between hypertensives and normotensives and between the two ethnic groups.

Method

A total of 99 patients were studied. They comprised 34 hypertensive Indians (26 females and 8 males), 18 normotensive Indians (12 females and 6 males), 27 hypertensive Zulus (11 females and 16 males) and 20 normotensive Zulus (8 females and 12 males).

Blood was drawn for the measurement of intralymphocytic sodium and potassium before and 30 minutes after the intravenous administration of 20 mg furosemide. Details of methodology are given in Chapter 2 (b). Intralymphocytic sodium and potassium were measured as described in appendix III.

RESULTS

Mean ages and Quetelet's indices are given in Table 1.

The mean sitting and standing systolic blood pressure fell significantly after the administration of furosemide in hypertensive Zulus ($p < 0,001$), hypertensive Indians ($p < 0,01$) and normotensive Indians ($p < 0,01$) but not in normotensive Zulus. The mean sitting and standing diastolic blood pressure fell significantly only in hypertensive Zulus ($p < 0,01$).

Sitting and standing mean arterial pressure after furosemide was significantly lower in hypertensive Zulus ($p < 0,001$) and hypertensive Indians ($p < 0,001$) but not in normotensive subjects (figure 1 - 4).

There was no ethnic difference in the blood pressure response to intravenous furosemide.

There was a significant positive correlation between the difference in sitting mean arterial blood pressure and the difference in ILNa before and after furosemide for the total group only ($r = 0,2$; $p < 0,05$).

The mean ILNa of normotensive Zulus increased from 64,3 to 67,6 mmol/kg dry cell weight and that of normotensive Indians increased from 67 to 70,8 mmol/kg dry cell weight after the administration of furosemide. Although the mean increase appears similar in the two groups it was only statistically significant in Indian controls ($p < 0,01$).

The mean ILNa decreased from 80,9 to 79,6 mmol/kg dry cell weight and from 71,8 to 70,5 mmol/kg dry cell weight in hypertensive Zulus and hypertensive Indians respectively. This difference was not statistically significant. Individual changes are shown in figures 5 and 6.

Although ILNa tended to increase in normotensives and decrease in hypertensives, there was no significant difference in the response to furosemide when hypertensives were compared to normotensives. There was also no ethnic difference.

There was no difference in the ILK before and after furosemide in all groups.

DISCUSSION

Furosemide is known to inhibit sodium - potassium cotransport, unidirectional sodium efflux and unidirectional sodium influx under controlled conditions (Dunn 1970; Ellory 1982) but appears to have no effect on net sodium movement.

This study, in effect, studied the effect of furosemide on net sodium movement in vivo and the only significant finding was a net increase in cell sodium in normotensive Indians. It is not possible to speculate from this study whether this net increase was due to an increase in passive diffusion, an increase in influx or a decrease in efflux. Zidek et al (1984) found that piretamide (a loop diuretic) caused an increase in

red cell sodium in hypertensive subjects. They did not study the effect of this drug in normotensive subjects. Etkin et al (1982) on the other hand found that furosemide decreased net sodium influx in hypertensive Whites, while having no effect in normotensive Whites, hypertensive Blacks and normotensive Blacks. Tuck et al (1984) described lower sodium potassium cotransport rates in Black hypertensives compared to Black normotensives and lower values in Blacks compared to Whites. If Zulus do indeed have reduced cotransport rates, then furosemide would have little effect on cell sodium concentration i.e. sodium-potassium cotransport is not available for inhibition by furosemide. Indian hypertensives appear to behave in a similar fashion to Zulu hypertensives in their response to furosemide. The greater mean difference in ILNa before and after furosemide seen in normotensive subjects may be related to higher levels of sodium potassium cotransport being available for inhibition.

The fact that furosemide appears to have a significant effect in normotensive Indians only, implies that this group may have a near normal cotransport system and this may also be responsible for Indians having a "normal" cell sodium concentration. This concept would of course not explain the lack of inhibition in hypertensive Indians. More work needs to be done on the various sodium potassium transport systems in this population.

An interesting observation was that diastolic blood pressure tended to increase in normotensive subjects of both ethnic groups as did the ILNa after administration of furosemide whereas in hypertensives the blood pressure fell as did the ILNa (bearing in mind again that the only statistically significant difference was in normotensive Indians). There was also a significant correlation between the difference in ILNa and the difference in mean arterial pressure before and after furosemide for the total group only.

Individual variation in the response to furosemide can be seen in figures 5 and 6. This is to be expected as studies which have described abnormalities of sodium potassium transport systems have found this only in a certain percentage of patients (Smith 1984).

In summary, ILNa increased significantly after the administration of furosemide in normotensive Indians only. The lack of response in other groups may be related to low levels of the furosemide sensitive sodium potassium cotransport system.

TABLE 1

MEAN (± SD) AGE AND QUETELETS' INDEX

	AGE	QUETELETS INDEX
HYPERTENSIVE ZULUS	43,3 ± 10,9	29,1 ± 5,9
NORMOTENSIVE ZULUS	42,8 ± 9,7	28,4 ± 5,2
HYPERTENSIVE INDIANS	45,0 ± 8,6	27,1 ± 4,2
NORMOTENSIVE INDIANS	43,4 ± 7,8	26,2 ± 4,8

FIGURE 1

Effect of Furosemide on sitting blood pressure in Zulus (mean ± SEM)

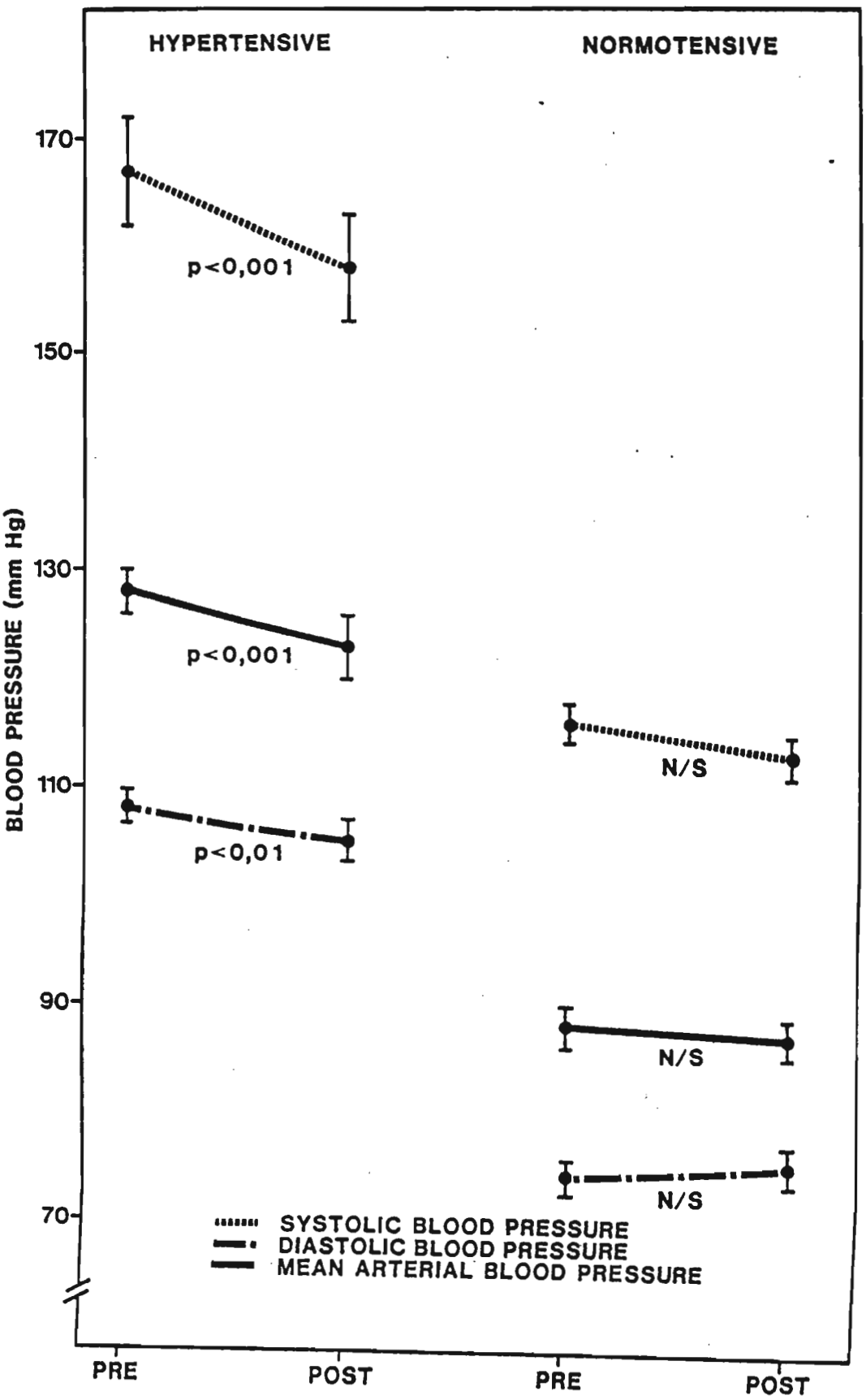


FIGURE 2

Effect of Furosemide on standing blood pressure in Zulus

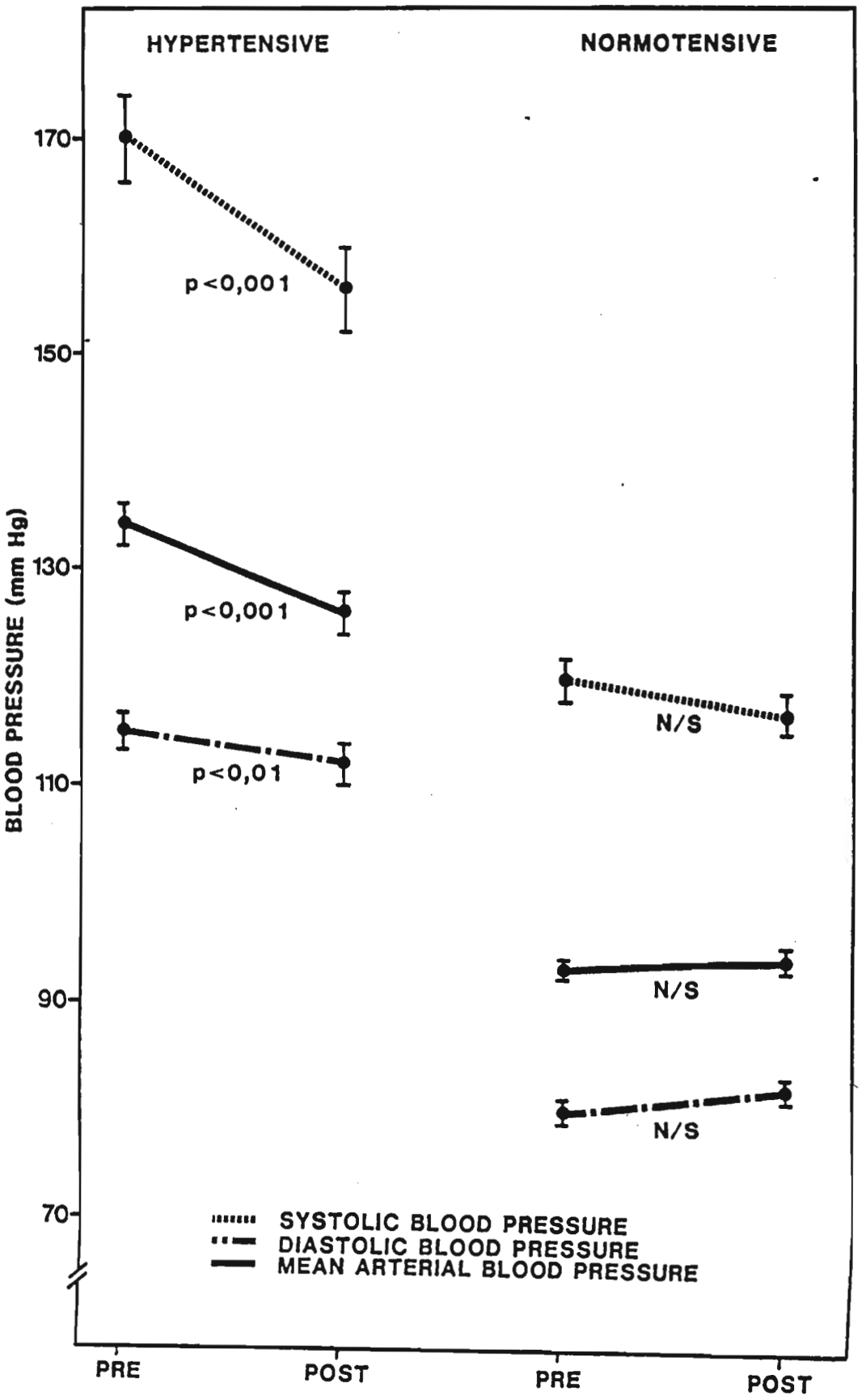


FIGURE 3

Effect of Furosemide on sitting blood pressure in Indians

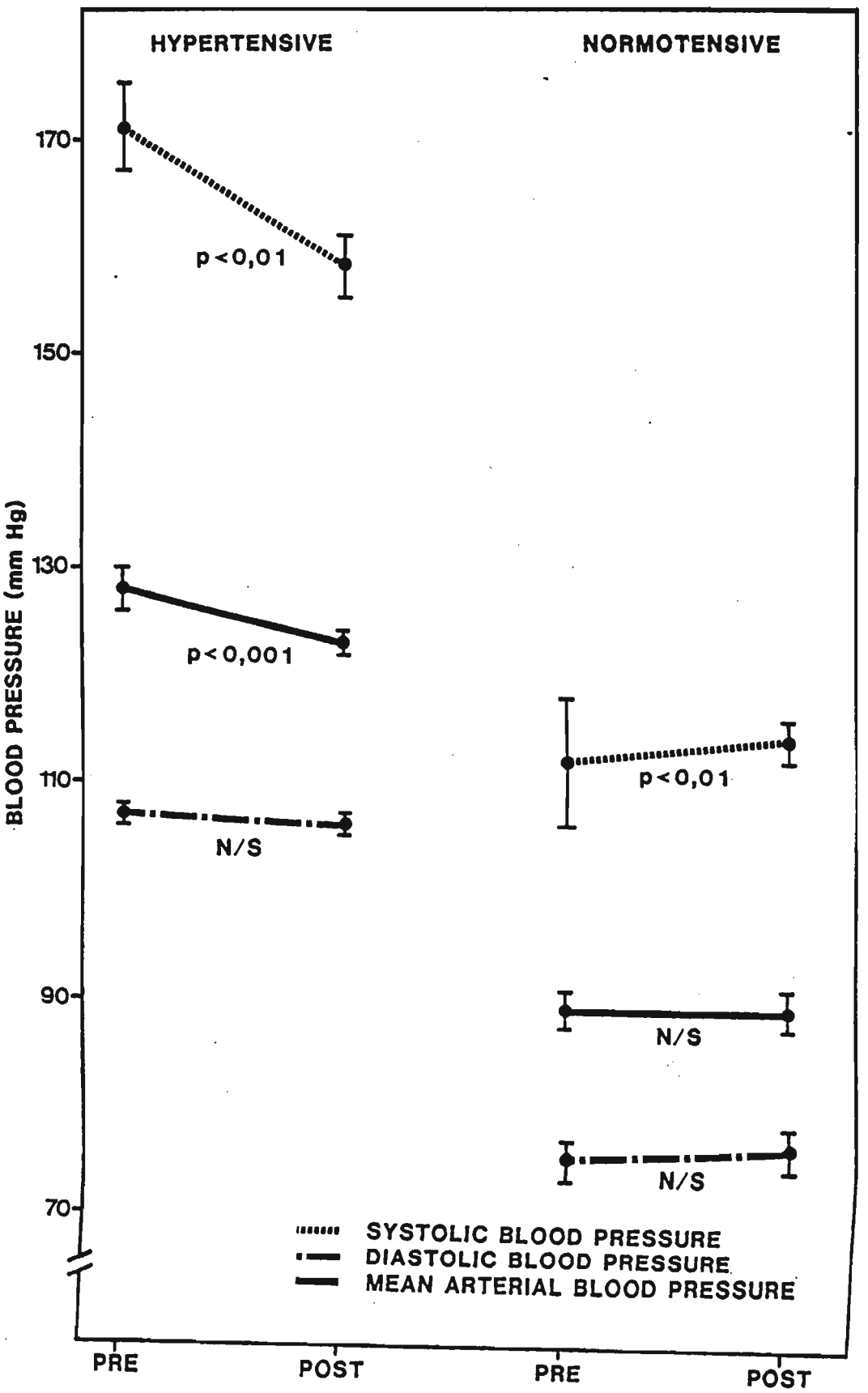


FIGURE 4

Effect of Furosemide on standing blood pressure in Indians

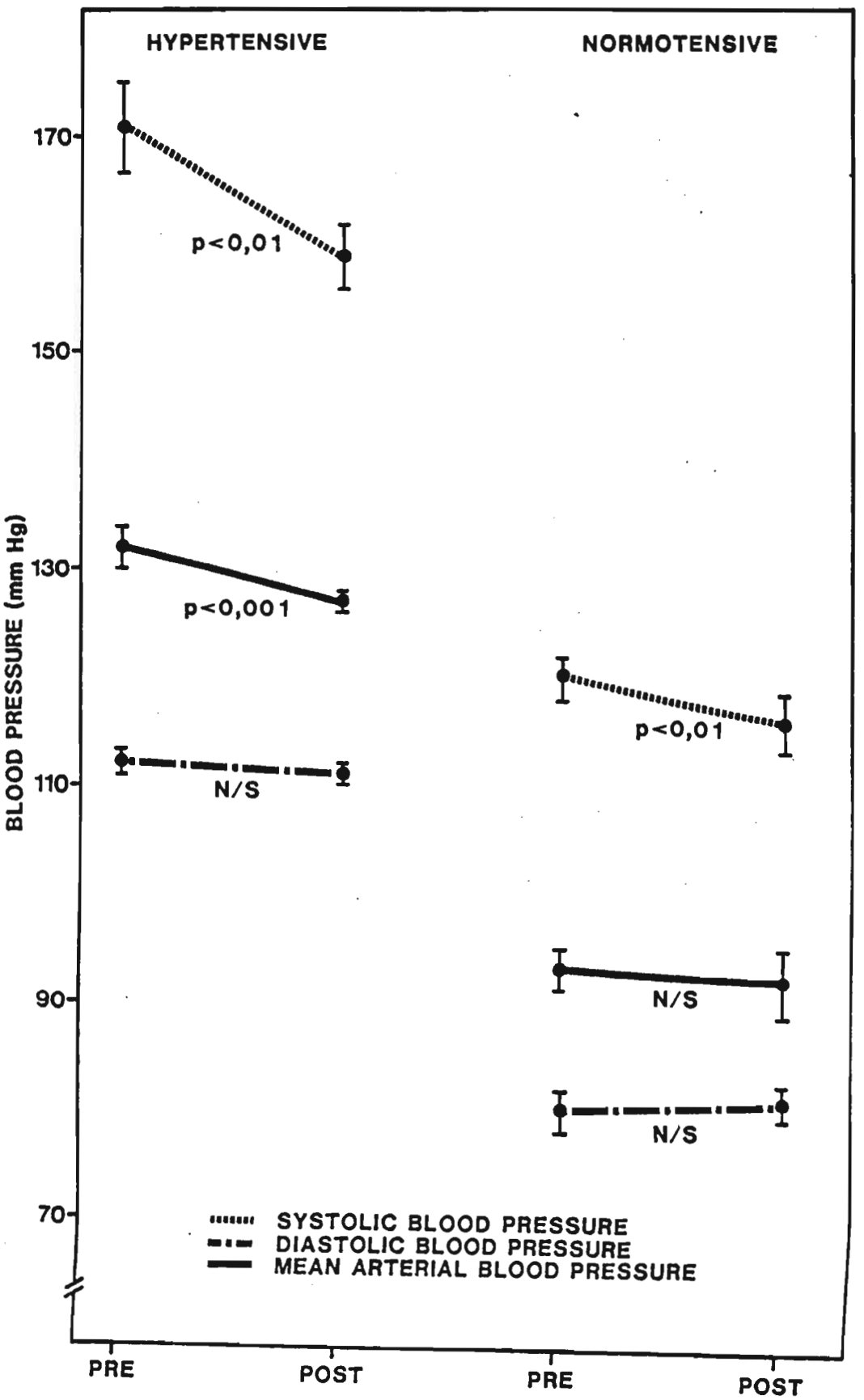


FIGURE 5

Effect of Furosemide on ILNa in Zulus (Individual changes and mean ± SEM)

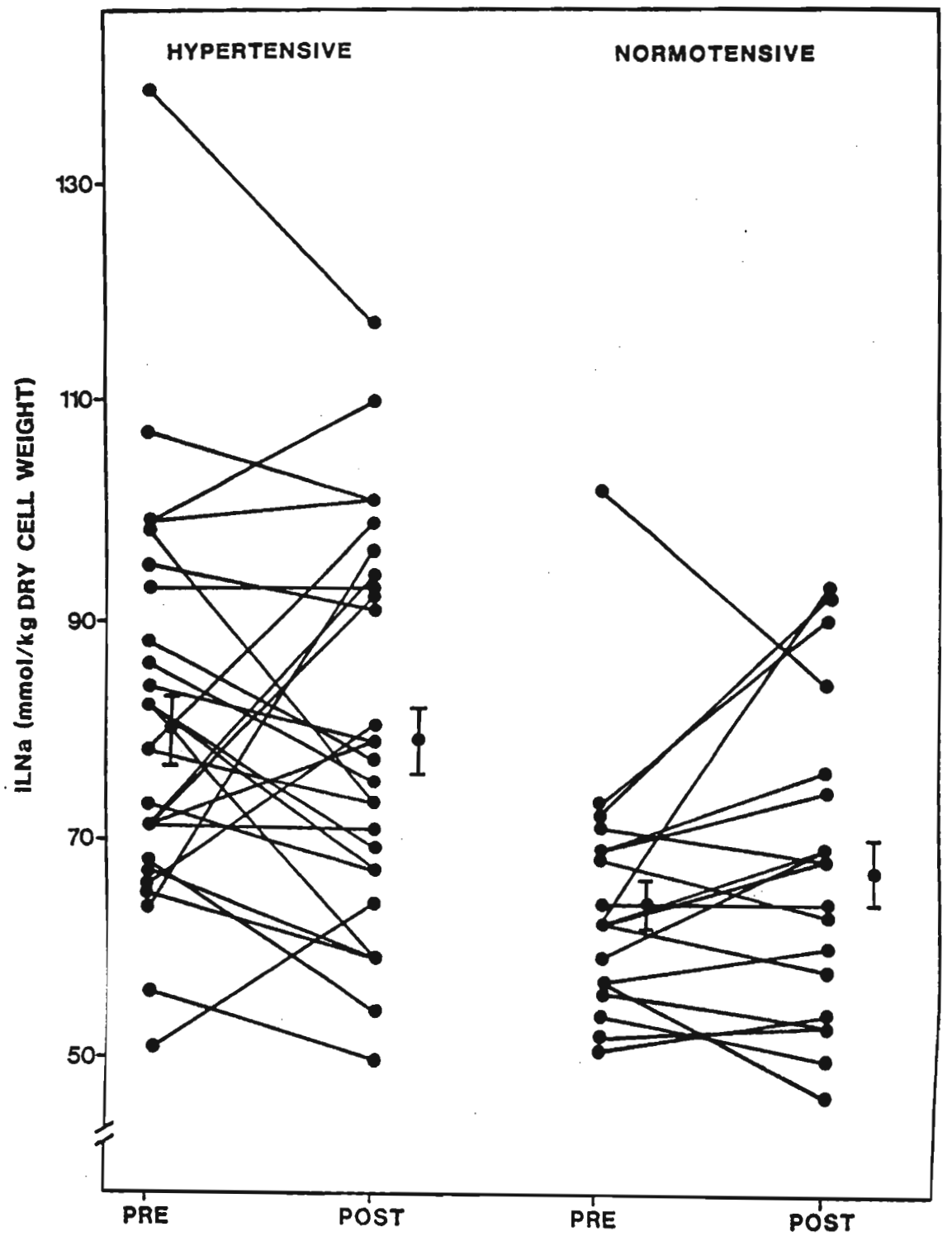
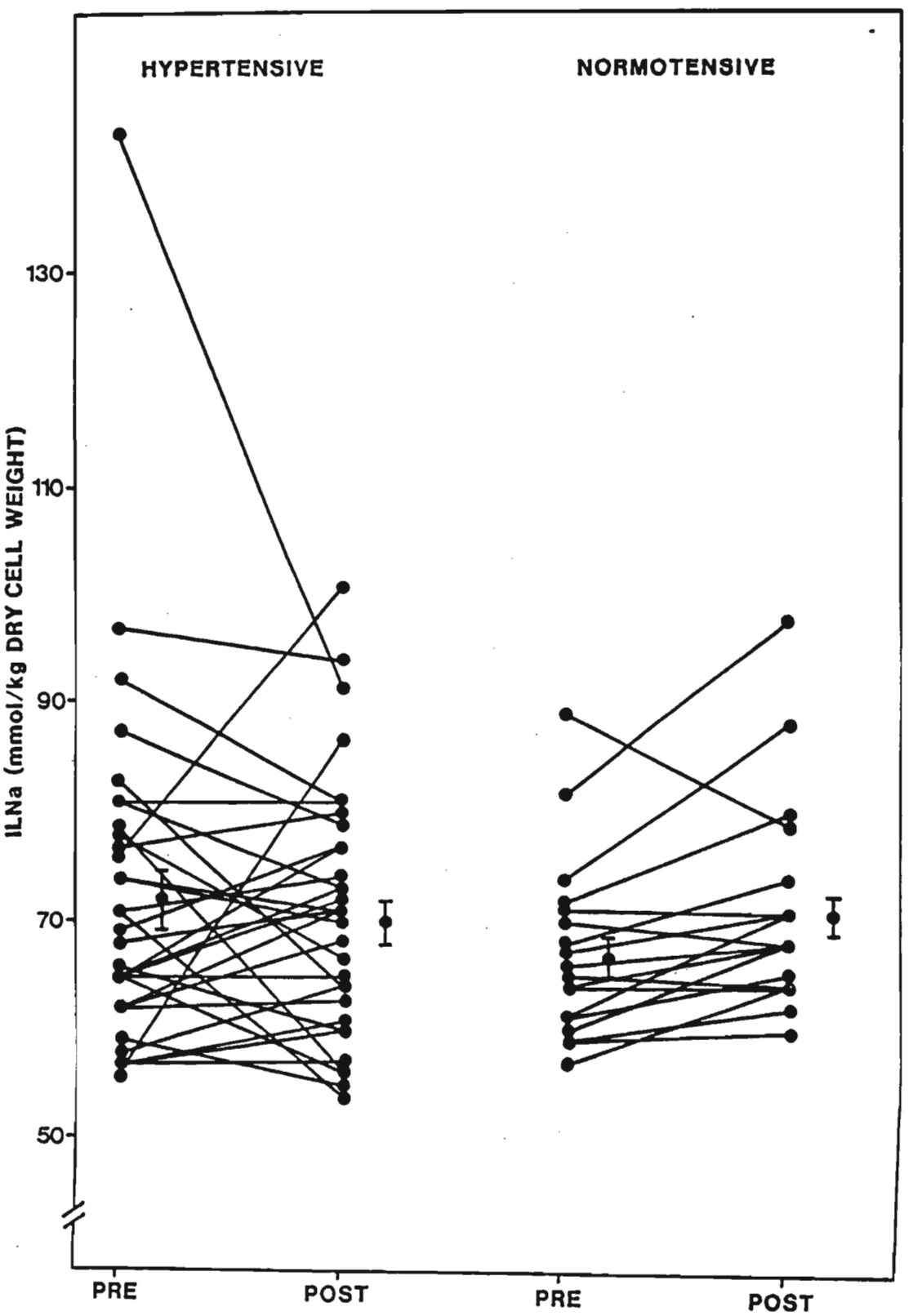


FIGURE 6

The effect of Furosemide on ILNa in Indians (Individual changes and mean ± SEM)



CHAPTER 8

THE EFFECT OF HYDROCHLOROTHIAZIDE AND SOTALOL ON INTRALYMPHOCYTIC SODIUM AND POTASSIUM

INTRODUCTION

The aim of this study was to assess the effect of antihypertensive drugs on ILNa and ILK. Two commonly used drugs with different mechanisms of action were chosen viz a thiazide diuretic (hydrochlorothiazide) and a beta blocker (sotalol). Thiazides exert their hypotensive effect by increasing the renal excretion of sodium and chloride and an accompanying volume of water (Goodman and Gillman 1980). Thiazides cause a net sodium loss of 250 mmol and an average water loss of 2 litres (Fries 1983). The mechanism by which sotalol reduces blood pressure has not been completely elucidated.

PATIENTS AND METHODS

Eighty eight hypertensive patients were studied. Twenty four Zulus (9 females, 15 males) and 27 Indians (16 females, 11 males) were given 25 mg hydrochlorothiazide daily for 1 month; 16 Zulus (6 females, 10 males) and 20 Indians (16 females and 4 males) were given 160 mg sotalol daily for 1 month. Both drugs

were given orally. Blood was taken for the measurement of ILNa, ILK, serum sodium and serum potassium before and 1 month after treatment.

Compliance was assessed by pill count. Details of patient selection and blood pressure measurement are given in chapter 2. Statistical methods used were analysis of variance and Students t test.

RESULTS

Mean ages and Quetelet's indices are given in Table 1.

HYDROCHLOROTHIAZIDE

Intralymphocytic sodium remained virtually unchanged with treatment. The mean ± SD pre treatment ILNa was 78,6 ± 14,3 mmol/kg dry cell wt in Zulus and 73,5 ± 13,2 mmol/kg dry cell wt in Indians. The mean ± SD post treatment ILNa was 78,6 ± 11,5 mmol/kg dry cell weight in Zulus and 74,1 ± 15,2 mmol/kg dry cell weight in Indians. Individual changes are shown in Figure 1.

The mean ± SD ILK prior to treatment was 379,8 ± 44 mmol/kg dry cell wt in Zulus and 361,5 ± 33,5 mmol/kg dry cell wt in Indians, and the ILK after treatment was 380,1 ± 29 mmol/kg dry cell wt in Zulus and 374,9 ± 29,9 in Indians. The

decrease in ILK with treatment was not statistically significant. Individual changes are shown in Figure 2.

As expected the sitting and standing systolic, diastolic and mean arterial blood pressure fell significantly after treatment with hydrochlorothiazide ($p < 0,001$) (Figures 3 and 4).

There was no change in serum sodium and potassium with treatment in both groups. Results are given in Table 2.

SOTALOL

The mean $\pm$ SD ILNa prior to treatment with sotalol was 80,5 $\pm$ 5,5 mmol/kg dry cell weight in Zulus and 78,4 $\pm$ 24,4 mmol/kg dry cell wt in Indians. The mean $\pm$ SD after treatment was 76,9 $\pm$ 10,7 mmol/kg dry cell wt in Zulus and 71,6 $\pm$ 8,5 mmol/kg dry cell wt in Indians. This decrease in cell sodium was not statistically significant in both groups (Figure 5).

Intralymphocytic potassium also, did not change significantly after treatment. The mean $\pm$ SD ILK before treatment was 377,3 $\pm$ 39,8 mmol/kg dry cell wt in Zulus and 363,3 $\pm$ 32,2 mmol/kg dry cell wt in Indians. The mean $\pm$ SD ILK after treatment was 373,2 $\pm$ 23,4 mmol/kg dry cell wt in Zulus and 358,9 $\pm$ 18,0 mmol/kg dry cell wt in Indians (Figure 6).

The mean sitting and standing diastolic and mean arterial blood pressure fell significantly in Zulus and Indians. The mean sitting systolic blood pressure fell significantly only in

Indians. There was no significant fall in sitting and standing mean systolic blood pressure in Zulus and mean standing systolic blood pressure in Indians (figures 7 and 8).

Serum sodium did not change significantly after treatment in both groups. Serum potassium on the other hand increased significantly after treatment in Indian subjects only i.e. from a mean of 3,6 to 4,1 mmol/litre (Table 3).

DISCUSSION

Data on the effect of treatment on intracellular sodium concentration are controversial. Aderounmu et al (1979) found that red cell sodium content and active sodium efflux were unchanged by antihypertensive treatment i.e. alpha methyldopa and debrisoquine. Araoye et al (1978) found that white blood cell sodium fell significantly in patients on hydrochlorothiazide and reserpine but not in patients on alpha methyldopa and hydrallazine. Ambrosioni et al (1980) also found that diuretics caused a significant decrease in ILNa, whereas non diuretic drugs such as beta blockers, clonidine and alpha methyl dopa caused no variation. Walter et al (1980) on the other hand found that red cell sodium was increased after antihypertensive medication which included diuretics. They also found that therapy with 100 mg hydrochlorothiazide for 1 week caused an increase in cell sodium by 30%. Cole et al (1983) studied erythrocyte membrane transport in hypertensive patients who had been on treatment for 5 years and found none of the

transport abnormalities which were found in untreated hypertensive patients, suggesting that antihypertensive treatment "reverses the underlying changes in membrane transport".

The results of this study are similar to those of Aderounmu et al (1979) in that no change in intracellular sodium concentration was noted after treatment with antihypertensive drugs. There was also no correlation between the drop in blood pressure and the change in intracellular sodium concentration with both drugs. The results of the present study could be explained by the findings of Freis et al (1983) i.e. that net sodium loss seen with diuretic therapy occurred from the extracellular fluid compartment.

In conclusion the antihypertensive effects of hydrochlorothiazide and sotalol appear to be unrelated to intracellular sodium in the population studied.

TABLE 1

MEAN ($\pm$ SD) AGE AND QUETÈLETS' INDEX

HYDROCHLOROTHIAZIDE	AGE	QUETELETS INDEX
ZULUS	44,0 $\pm$ 10,6	30,5 $\pm$ 6,9
INDIANS	47,3 $\pm$ 9,4	25,8 $\pm$ 4,4
SOTALOL		
ZULUS	47,9 $\pm$ 9,9	31,9 $\pm$ 7,1
INDIANS	47,7 $\pm$ 8,0	27,3 $\pm$ 4,5

TABLE 2

EFFECT OF HYDROCHLOROTHIAZIDE ON SERUM SODIUM AND POTASSIUM (MEANS ± SD)

	ZULUS	INDIANS
SERUM SODIUM (MMOL/L)		
PRE TREATMENT	140,6 ± 2,6	140,6 ± 2,4
POST TREATMENT	139,5 ± 2,9	139,1 ± 2,4
SERUM POTASSIUM (MMOL/L)		
PRE TREATMENT	3,8 ± 0,4	3,7 ± 0,4
POST TREATMENT	3,7 ± 0,4	3,6 ± 0,4

TABLE 3

EFFECT OF SOTALOL ON SERUM SODIUM AND POTASSIUM

	ZULUS	INDIANS
SERUM SODIUM (MMOL/L)		
PRE TREATMENT	140,7 ± 2,0	140,1 ± 1,8
POST TREATMENT	141,5 ± 3,7	140,2 ± 2,9
SERUM POTASSIUM (MMOL/L)		
PRE TREATMENT	3,9 ± 0,3	3,6 ± 0,4*
POST TREATMENT	3,9 ± 0,5	4,1 ± 0,5

* pre vs post $p < 0,001$

FIGURE 1

Effect of hydrochlorothiazide on intralymphocytic sodium (Individual changes and mean ± SEM)

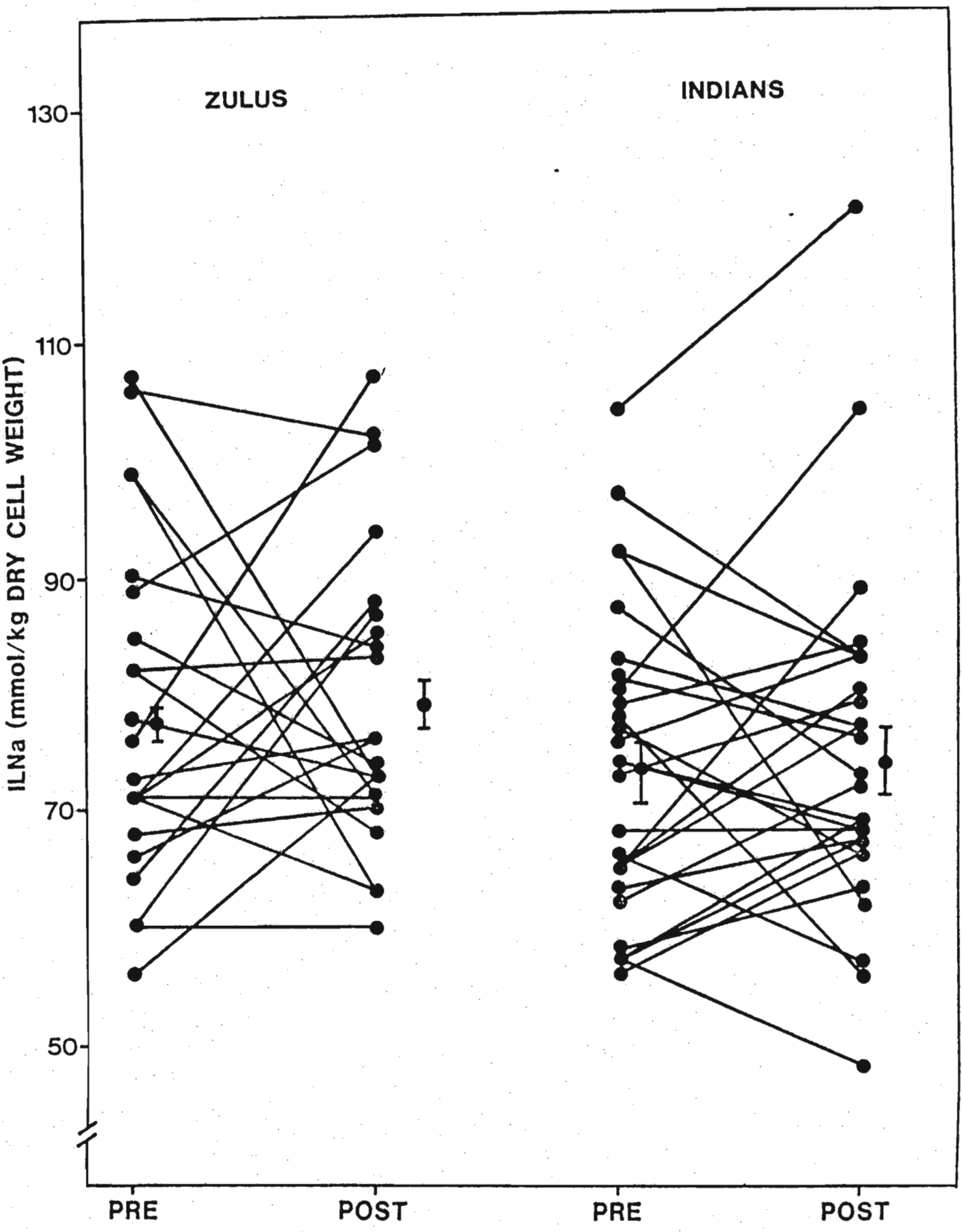


FIGURE 2

Effect of hydrochlorothiazide on intralymphocytic potassium (Individual changes and mean ± SEM)

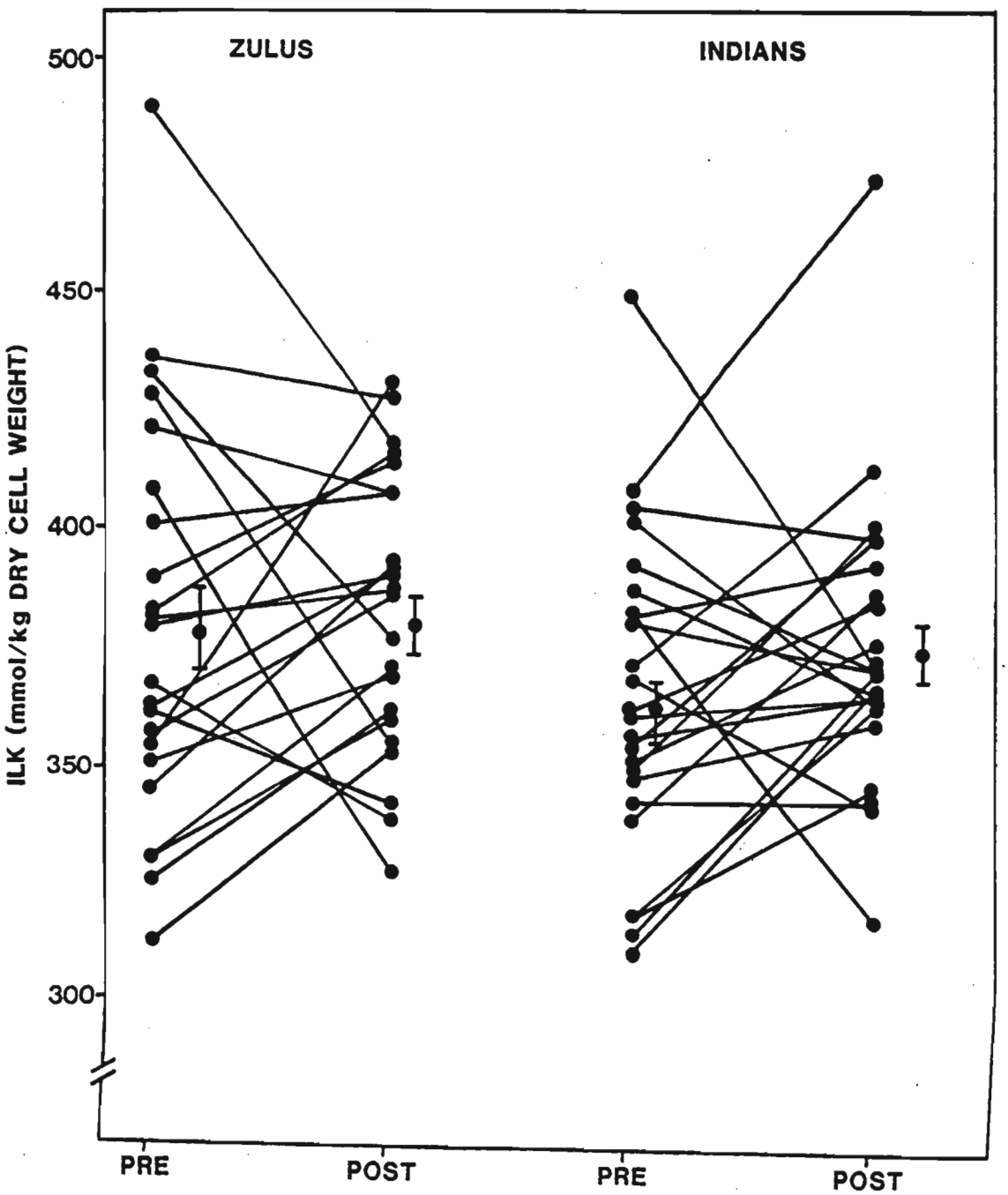


FIGURE 3

Effect of hydrochlorothiazide on sitting blood pressure (mean ± SEM)

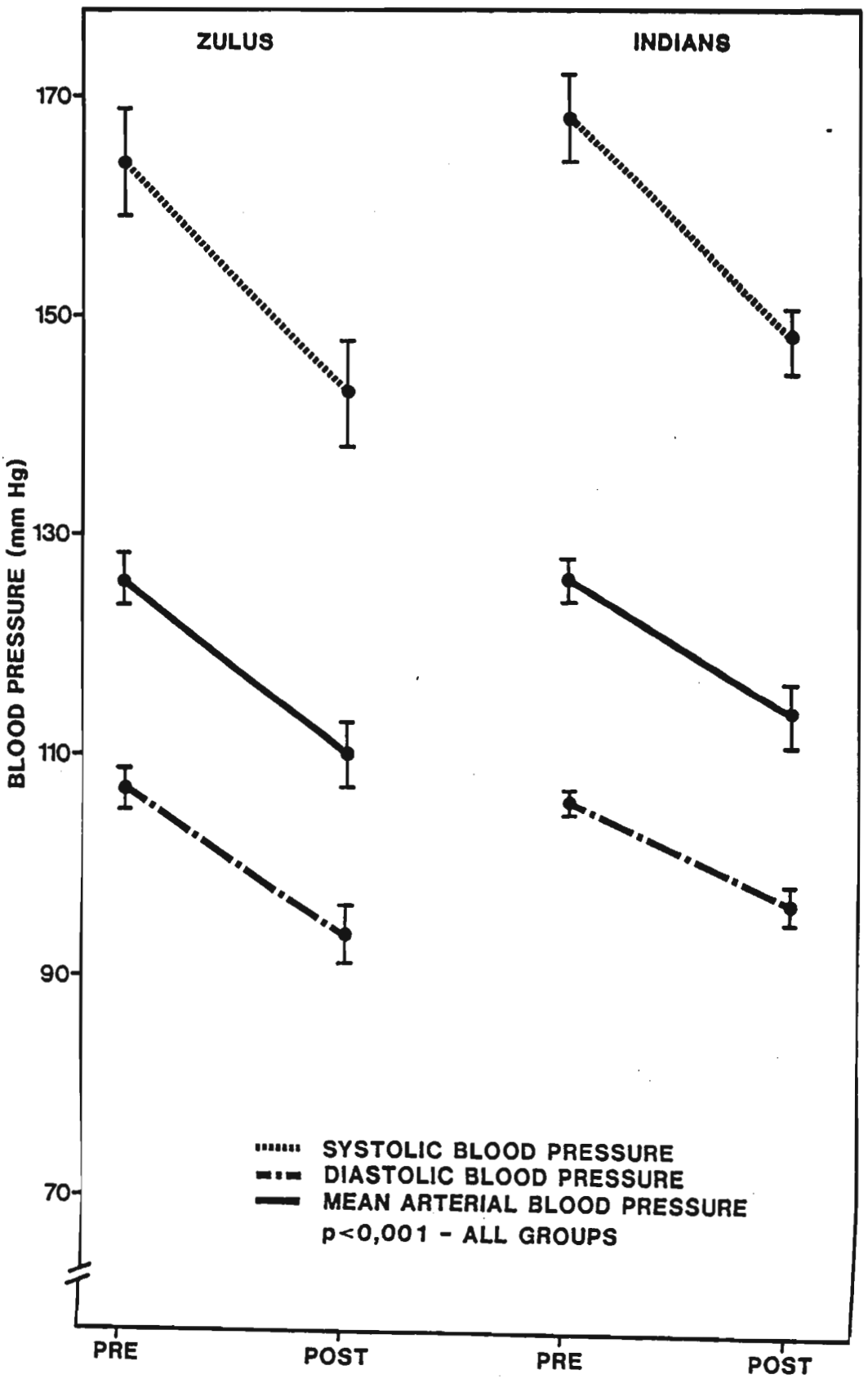


FIGURE 4

Effect of hydrochlorothiazide on standing blood pressure (mean ± SEM)

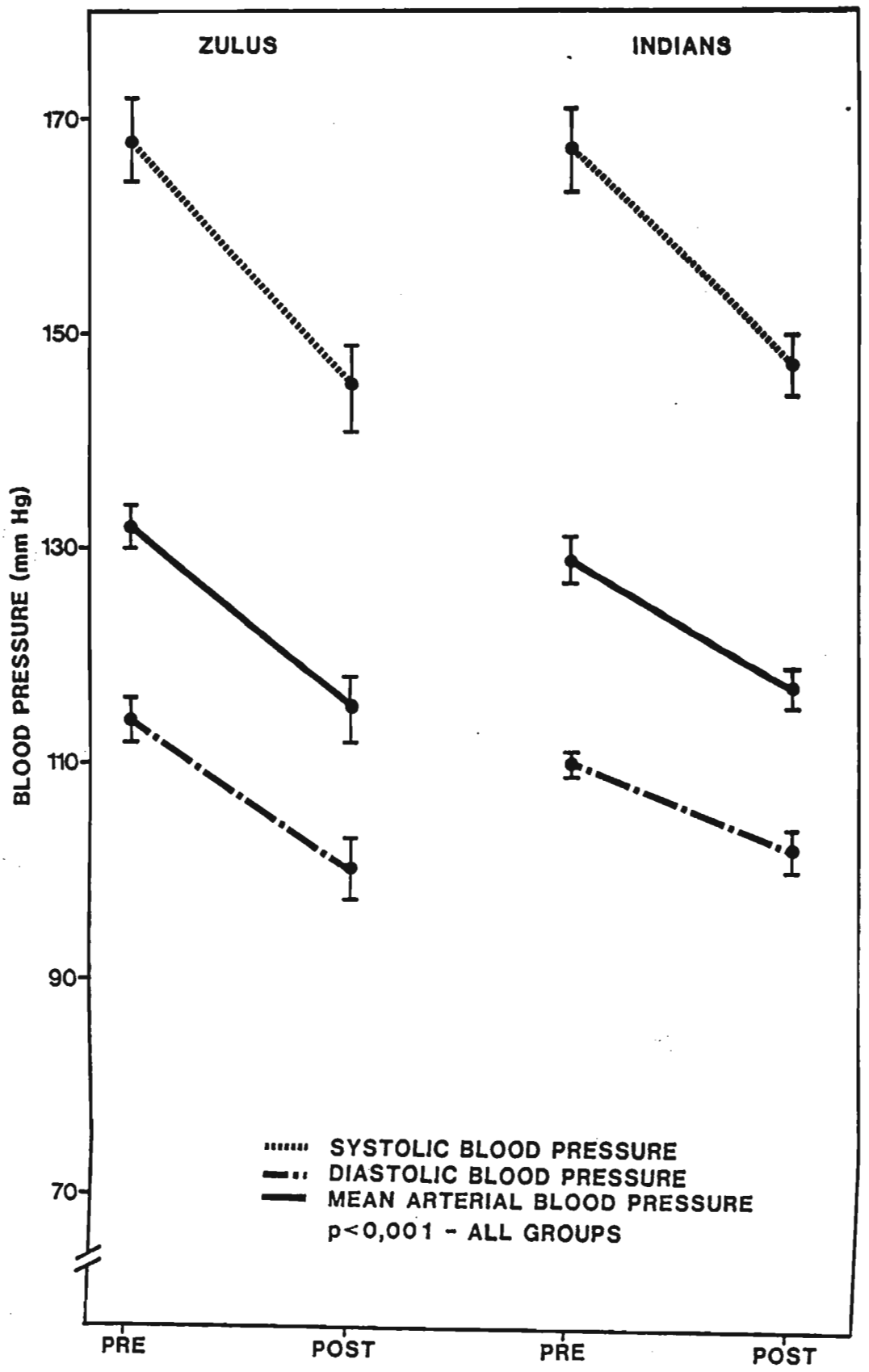


FIGURE 5

Effect of Sotalol on intralymphocytic sodium (individual changes and mean ± SEM)

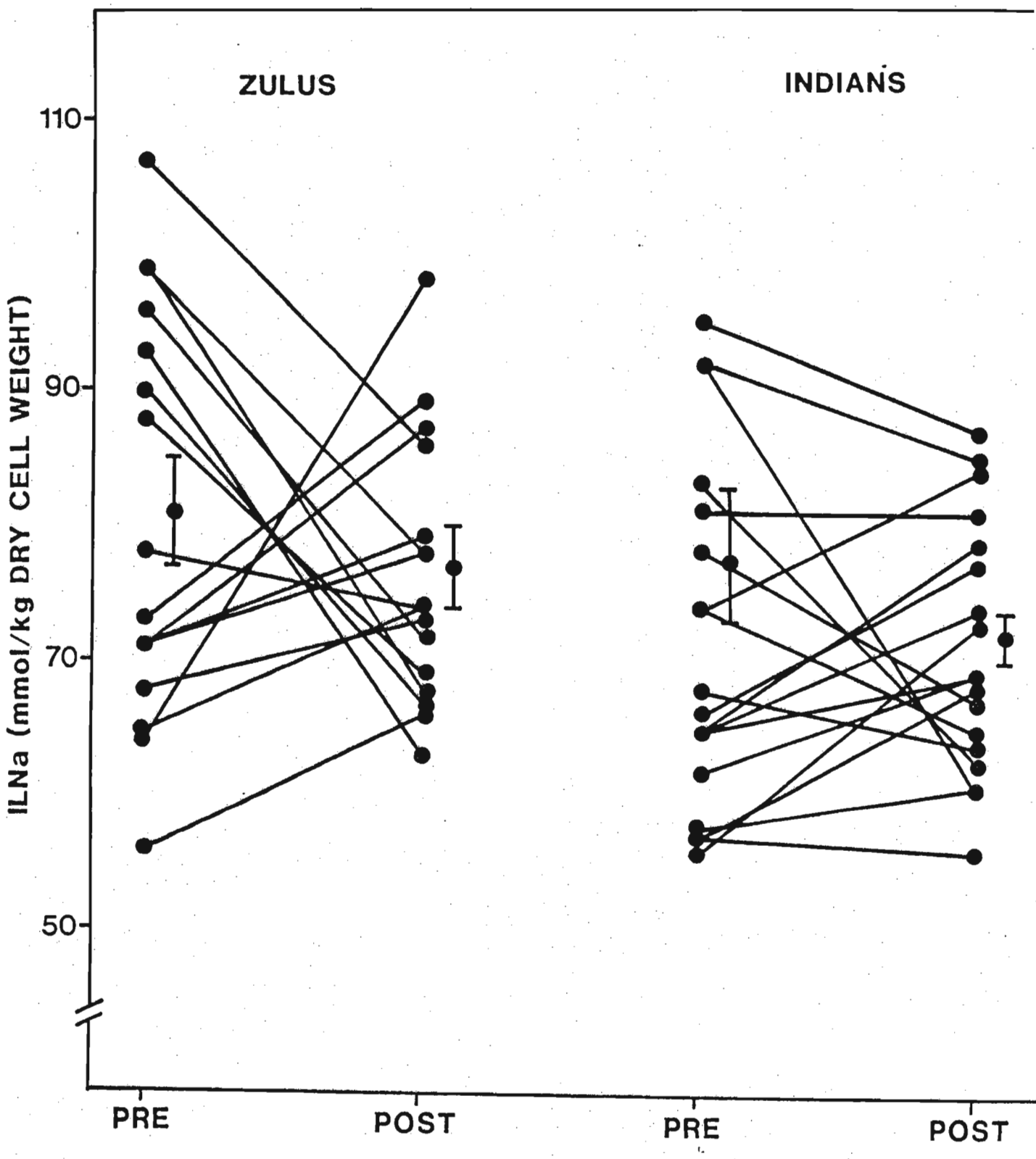


FIGURE 6

Effect of Sotalol on intralymphocytic potassium (Individual changes and mean ± SEM)

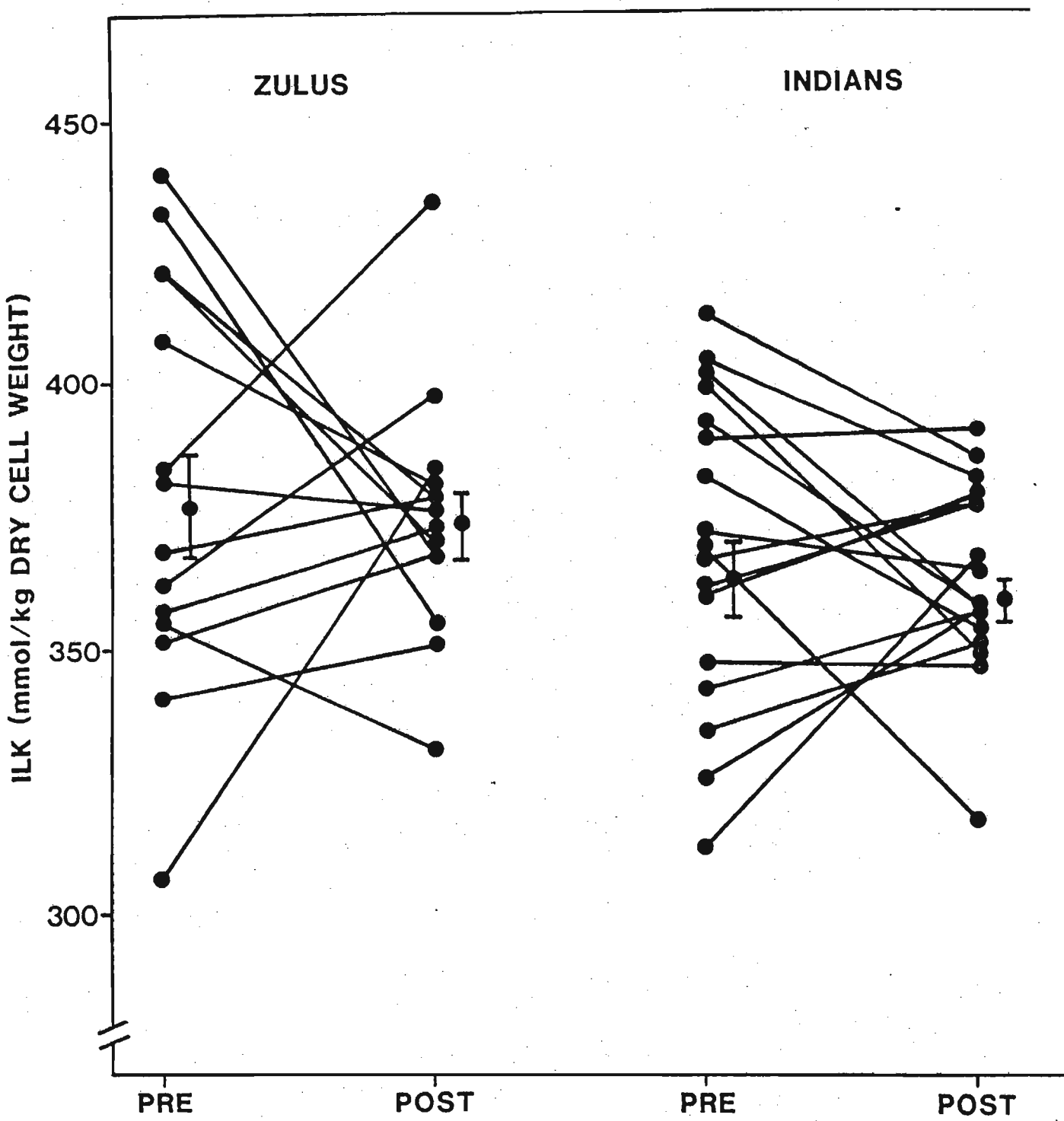


FIGURE 7

Effect of Sotalol on sitting blood pressure (mean ± SEM)

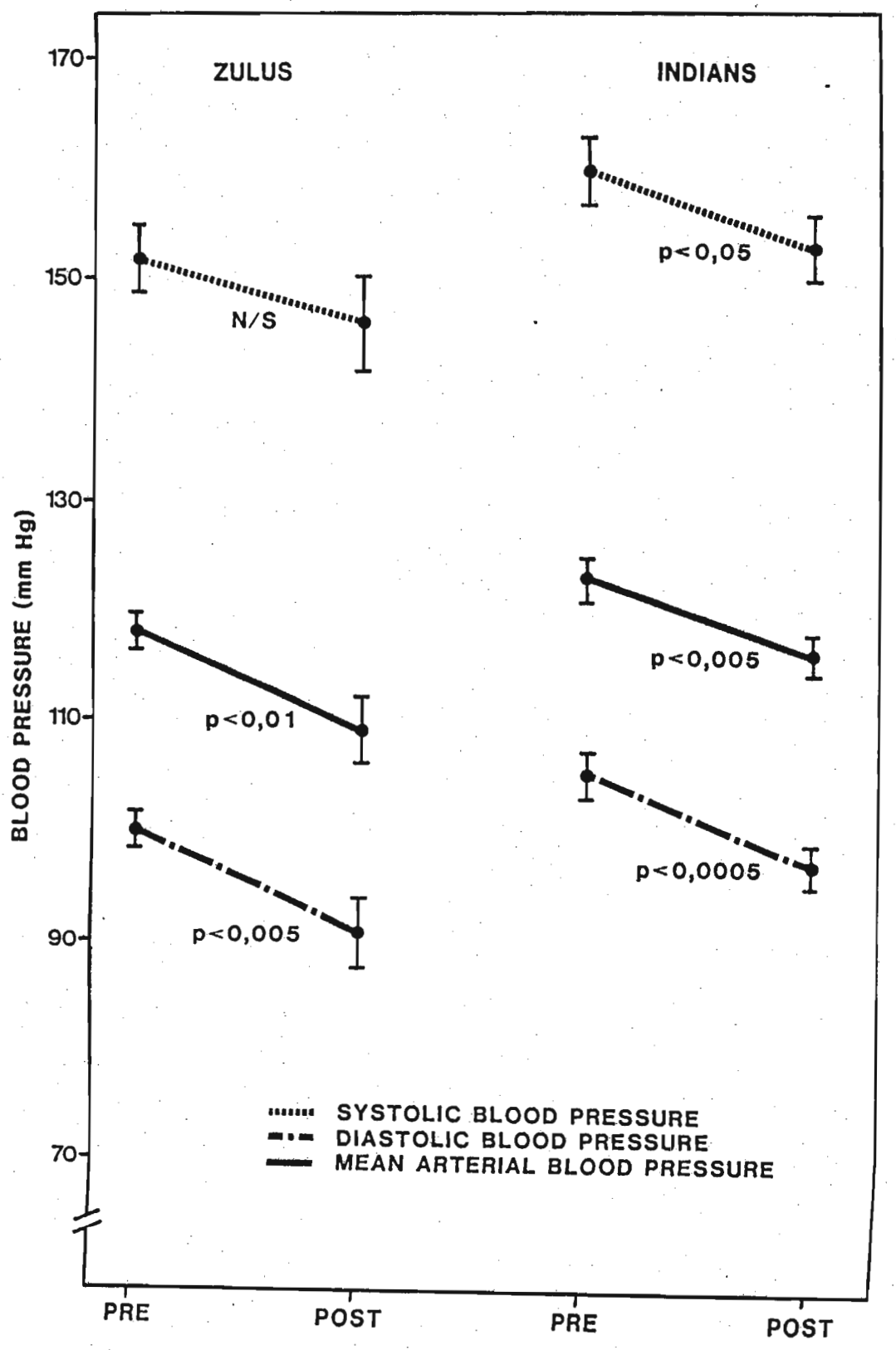
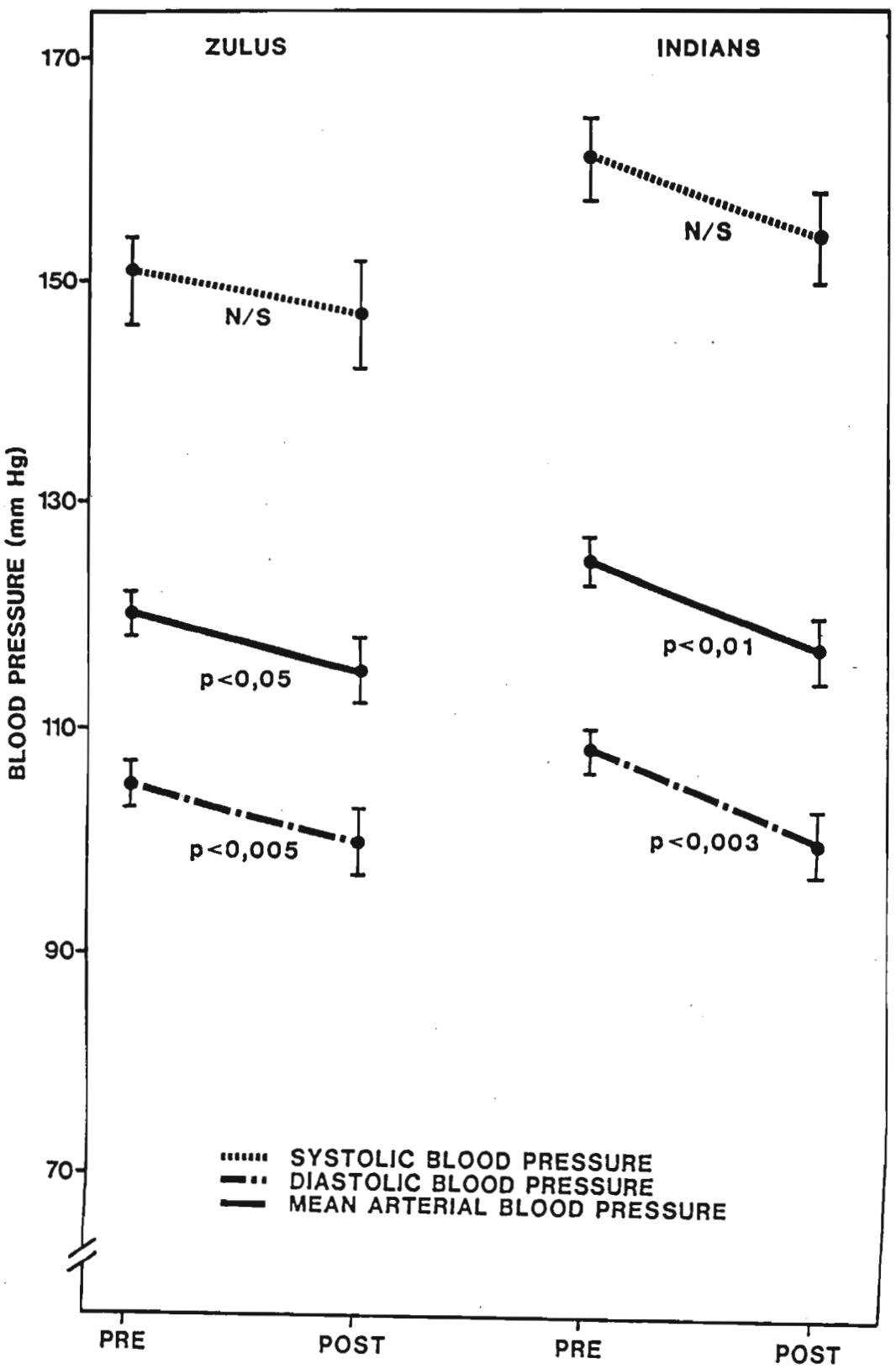


FIGURE 8

Effect of Sotalol on standing blood pressure (mean ± SEM)



CONCLUSIONS

The aim of this study was to evaluate the role of sodium and potassium in the pathogenesis of hypertension in Zulus and Indians. It is widely accepted that hypertension is a multifactoral disease; sodium and potassium were studied not necessarily because they were considered to be the most important, but if found to play even a small role dietary manipulation of the general population is possible. This applies also to other cations such as magnesium and calcium, however unlike salt cellars magnesium and calcium cellars are not the norm at the dinner table.

A prevalence study has shown that 90% of hypertensive Zulus and 58% of hypertensive Indians were undiagnosed or inadequately treated (Seedat 1982). This data suggests that a preventive approach to hypertension may be more successful than a therapeutic one.

Although work on the sodium and potassium status of blacks has been done in South Africa, these studies either looked at normotensive subjects only or depended on inadequate urine samples, viz: casual urine samples or single 24 hr urine samples. Sever et al (1980) studied casual urine sodium and potassium in Xhosas; Modlin et al (1963) studied single 24 hr urine samples in normotensive Basuto men; Cohen et al (1982) studied serum and urinary sodium and potassium in normotensive Whites, normotensive blacks and hypertensive blacks - some of these

patients collected one 24 hour urine sample and others collected casual urine samples; Barlow et al (1982) studied single 24 hour urinary sodium and potassium excretion rates in normotensive blacks and Whites.

No studies on the sodium and potassium status of South African Indians have been done to date.

This study, by using 7 timed overnight urine specimens from 585 subjects, attempts to provide more comprehensive data on the sodium, potassium status of blacks (urban and rural) and new data in Indians. Touyz (1984) has studied red cell sodium, potassium, magnesium and calcium in Johannesburg Blacks and Davidson et al (1982) have studied red cell sodium-potassium cotransport in Blacks, Whites and Coloureds in Cape Town. However no work on white cell sodium and potassium has been done in South Africa in any population group. A survey of world literature shows no studies on intracellular sodium and potassium in native or migrant Indians.

The daily sodium intake of Zulus as calculated from mean hourly excretion rates (160 mmol) is similar to a Western diet (150 - 250 mmol/day). Sodium intake of Indians although much lower (100 mmol/day) is probably related to the lower body weight in Indian subjects (sodium excretion was positively correlated with Quetelet's index). There was a tendency to lower sodium excretion in hypertensive subjects but not significantly so. There was no difference in the sodium excretion of urban and rural Zulus. There was no correlation between sodium excretion and blood pressure in both ethnic

groups. The finding of a similar sodium intake in urban and rural Zulus reflects a change in rural dietary habits toward an urban Western type diet.

Dietary potassium intake of Zulus (40 mmol/24 hrs) on the other hand was half that of a Western diet. Potassium intake of Indians was even lower at 20 mmol/day, this was also related to body weight. There was no difference in the potassium excretion of urban and rural Zulus and between hypertensive and normotensive subjects in all groups. Urinary potassium excretion was negatively associated with systolic blood pressure in rural Zulus and Indians, but not in urban Zulus.

Rural Zulus had a lower sodium/potassium ratio and a lower prevalence of hypertension compared to urban Zulus. There was however no association between the sodium/potassium ratio and blood pressure. The lower sodium/potassium ratio in rural Zulus may be protective against the development of hypertension.

The prevalence of low plasma renin activity (PRA) was high. Whether low PRA is a cause or the effect of hypertension is not known. The higher plasma renin level in rural Zulus compared to urban Zulus was a surprising finding. If PRA is genetically determined, then urban and rural Zulus should have similar values. This was not related to salt intake either, as there was no correlation between PRA and urinary sodium and potassium excretion.

The absence of a correlation between renin and aldosterone in hypertensive urban Zulus and hypertensive Indians suggests a defect in the renin - aldosterone system in these patients.

The lack of stimulation of plasma renin activity by natriuresis confirmed a hyporesponsive renin angiotensin system in hypertensive urban Zulus and hypertensive Indians.

Although dietary sodium was similar in hypertensive and normotensive Zulus, intracellular sodium (using the lymphocyte as a cell model) was significantly higher in hypertensive Zulus compared to normotensive Zulus. The large overlap in results prevents the clinical use of this test. The sodium-potassium pump is known to be stimulated by the addition of potassium ions to the extracellular fluid and inhibited by lowering the extracellular potassium concentration (Nielsen 1986). The high cellular sodium concentration may be related to the "potassium deficient diet" in the patients i.e. some patients have a genetically determined increase in cell membrane "leakiness" to sodium, and dietary potassium is insufficient to stimulate the sodium-potassium pump to compensate for the increased cell sodium concentration. However, Indian patients also have a low potassium intake and no increase in cell sodium in hypertensive subjects. Sodium appears to be more important in the pathogenesis of hypertension in Zulus than in Indians.

Intralymphocytic sodium, after the administration of furosemide, increased significantly only in normotensive Indians. The lack of a significant response in other groups

may be related to low activity of the furosemide sensitive sodium-potassium cotransport system. The positive correlation between the difference in mean arterial pressure and the difference in ILNa before and after furosemide is further evidence that cell sodium plays some role in the pathogenesis of hypertension i.e. change in blood pressure was related to change in cell sodium concentration.

However the anti-hypertensive effects of sotalol hydrochloride and hydrochlorothiazide are unrelated to changes in cell sodium.

It is hoped that this thesis provides some insight into the sodium, potassium, renin and aldosterone status of Zulus and Indians. Further studies on sodium-potassium transport systems are necessary to establish the defect leading to an increase in cell sodium in Zulus. Longitudinal studies on dietary sodium intake are also necessary.

APPENDIX I

DETERMINATION OF PLASMA RENIN ACTIVITY

Plasma renin levels were measured indirectly by generation of angiotensin I by the antibody coated tube method (Travenol laboratories).

Reagents

1) Rabbit Anti-Angiotensin I serum coated tubes.
2) Angiotensin I tracer (I^{125}).
3) Angiotensin I standards.
4) Phenylmethylsulfonyl flouride solution (PMSF).
5) Maleate generation buffer.
6) Renin activity control plasma.
7) Assay buffer concentrate.

METHOD

1) Frozen plasma samples were thawed overnight in a refrigerator.
2) 1 ml of plasma was added to chilled non coated glass tubes placed in an ice-water bath.
3) To this was added 10 microlitres of PMSF solution and 100 microlitres of maleate buffer.

4) 500 microlitres of the above solution was placed in another pre chilled tube (step 3).

5) The first series of tubes were then placed in a bath at 37°C.

6) The second set of tubes were kept in an ice-water bath at 0°-4°C.

7) After 90 minutes the first series of tubes were transferred to an ice-water bath.

8) Four gamma coat tubes were used for each patient and control (i.e. each sample was done in duplicate).

9) 100 microlitres of 37°C plasma was added to duplicate tubes and 100 microlitres of 4°C plasma was added to the next 2 tubes.

10) 1 millilitre of Tracer buffer solution (1 ml buffer + 0,1 ml tracer) was added to the above tubes.

11) Tubes were gently mixed and incubated at room T° for 3 hours.

12) The contents of the tubes were aspirated by water suction.

13) Each tube was counted in a gamma counter for 1 minute.

Calculations were made as follows:

CPM = counts per minute

average CPM of 37° samples - average CPM of 4° samples X 7,4 = PRA ng/ml/hr.

APPENDIX II

ALDOSTERONE ASSAY

Serum aldosterone levels were measured by the antibody coated tube method (Aldoctk-125 C1S).

Reagents

1) ^{125}I - labelled aldosterone.
2) Standard aldosterone
3) Antibody coated tubes (Rabbit antiserum).
4) Phosphate buffer.
5) 8 - anilino - 1 - naphtalene sulphonic acid (ANS).

METHOD

1) 0,2 mls of serum was added to antibody coated tubes.
2) To this was added 0,1 ml I^{125} aldosterone and 0,7 ml buffer (Buffer + ANS mixture).
3) Contents of tubes were then mixed and incubated for 18 hours.
4) Contents of tubes were then aspirated.
5) Tubes were rinsed twice with 2 mls distilled water.
6) Tubes were counted in a gamma counter for 1 minute.

The assay was performed in duplicate.

Calculation of results:

A calibration curve is completed from standards and the unknown is read directly from this curve.

APPENDIX III

DETERMINATION OF INTRALYMPHOCYTIC SODIUM AND POTASSIUM CONCENTRATION

REAGENTS

1. Sodium metrizoate/ficoll solution (lymphoprep - Nyegaard and Co-Oslo).
2. Earles buffer (Bryant 1975).
3. Tissue culture medium reconstituted with earle's buffer (TC 199 - Difco laboratories).
4. Magnesium chloride (112 mmol/ℓ).
5. Carbon dioxide to correct the pH of earles buffer.
6. Phenol red.

METHOD

1. 15 mls heparinised blood (lithium heparin) were centrifuged for 10 minutes at 2000 rpm with a 15,7 cm head.
2. 2 mls of platelet rich plasma was removed and discarded (to eliminate platelet contamination).
3. The blood was then diluted (1 : 1) with Earles buffer (pH 7,2).

4. This mixture was layered gently onto lymphoprep (5 mls lymphoprep to 10 mls blood + Earles buffer mixture).
5. This was centrifuged for 30 minutes at 1800 rpm (this resulted in 4 layers i.e. red cells, lymphoprep, lymphocytes, plasma and buffer).
6. The plasma and buffer were first removed and discarded. The ring of lymphocytes were then gently removed.
7. 10 mls tissue culture medium was added to the lymphocytes.
8. This was centrifuged at 2500 rpm for 10 minutes.
9. The supernatant was discarded and 150 ul of tissue culture medium was added to the cell button.
10. Viability of lymphocytes was assessed by the trypan blue dye method and morphology of unstained specimens.
11. The cell suspension was transferred to a preweighed polythylene tube (weighed three times on a 5 decimal place balance).
12. This tube was then centrifuged for 10 minutes at 2200 rpm.
13. The supernatant was discarded and the inside of the tube was carefully wiped with tissue paper.

All the above steps were carried out at room temperature.

14. The cell button was then washed with 2 mls ice cold magnesium chloride.
15. The specimen was then centrifuged at 2200 rpm for 2 minutes in a refrigerated centrifuge (0°C).

16. The supernatant was discarded and the inside of the tube was again wiped dry with tissue paper.
17. The specimen was then dried in a vacuum desiccator containing phosphoric acid for 36 hours placed in an oven at 37°C.
18. The tubes were re weighed 3 times to obtain the dry weight of the cell button.
19. 1,5 mls of a 15 mmol Lithium solution was added to the cell button.
20. The top of the tube was sealed and the specimen was placed in an ultrasound desiccator for 30 minutes (this caused disruption of all lymphocytes).
21. Sodium and potassium were read on an IL 243 flame-photometer using the internal standard.

Results were expressed as µmol/kg dry cell weight.

REFERENCES

Addison W L T. 1928. The use of sodium chloride, potassium chloride, sodium bromide in cases of arterial hypertension which are amenable to potassium chloride.
Can Med Assoc J. 18 : 281-285.

Aderounmu A F, Salako L A. 1979. Abnormal cation composition and transport in erythrocytes from hypertensive patients.
Eur J Clin Invest. 9 : 369-375.

Akinkugbe O O, Ojo O A. 1969. Arterial pressures in rural and urban populations in Nigeria.
Br Med J. 2 : 222-224.

Ambard L, Beaujard E. 1904. Archives of General Medicine.
1 : 520.

Ambrosioni E, Tartagni F, Montebugnoli L, Magnani B. 1979. Intralymphocytic sodium in hypertensive patients : a significant correlation. Clin Sci. 57 : 325s-327s.

Ambrosioni E, Tartagni F, Montebugnoli L, Costa F V, Magnani B. 1980. Intralymphocytic sodium in hypertensive patients. In: Intracellular Electrolytes and Arterial Hypertension. Edited by Zumkley H and Loose H. 78-86.

Araoye M A, Khatri I M, Linda L Y, Fries E D. 1978. Leukocyte intracellular cations in hypertension : Effect of antihypertensive drugs.
Am Heart J. 96 (6) : 731-738.

Barden A E, Vandongen R, Beilen L J, Margetts B, Rogers P. 1986. Potassium supplementation does not lower blood pressure in normotensive women.
J Hypertens. 4 : 339-343.

Barlow R J, Connell M A, Levendig B J, Gear J S S, Milne F J. 1982. A comparative study of urinary sodium and potassium excretion in normotensive urban Black and White South African males.
S Afr Med J. 62 : 939-941.

Barlow R J, Connell M A, Milne F J. 1968. A study of 48 hour faecal and urinary electrolyte excretion in normotensive Black and White South African males.
J Hypertens. 4 : 197-200.

Beevers D G, Hawthorne U M, Padfield P L. 1980. Salt and blood pressure in Scotland.
Br Med J. 281 : 641-642.

Berglund G, Wikstrand J, Wallentin I, Wilhelmsen L. 1976. Sodium excretion and sympathetic activity in relation to severity of hypertensive disease.
Lancet. i : 324-327.

Biglieri E G. 1979. Effect of posture on the plasma concentration of aldosterone in hypertension and primary hyperaldosteronism.
Nephron. 23 : 112-115.

Blaustein M P. 1984. Sodium transport and hypertension.
Hypertension. 6 (4) : 453-455.

Blaustein M P. 1980. How does sodium cause hypertension? An hypothesis. In: Intracellular Electrolytes and Hypertension. Edited by H. Zumkley and H. Losse. George Thieme Verlag. Stuttgart. New York. 151-157.

Blaustein M P, Hamlyn J M. 1984. Sodium transport inhibition, cell calcium and hypertension. The natriuretic hormone/Na-Ca exchange/hypertension hypothesis.
Am J Med. 77 (4A) : 45-59.

Boon N A, Harper C, Aronson J K, Grahame-Smith D G. 1985. Cation transport function in vitro in patients with untreated essential hypertension : a comparison of erythrocytes and leucocytes.
Clin Sci. 68 : 511-515.

Brand S C, Whittan R. 1984. The effect of furosemide on sodium movements in human red blood cells.
J physiol. 384 : 301-306.

Brock T A, Smith J B, Overbeck H W. 1982. Relationship of vascular sodium potassium pump activity to intracellular sodium in hypertensive rats.
Hypertension. 4 (suppl II) : 43-48.

Brooks B A, Lant A F. 1978. The use of the human erythrocyte as a model for studying the action of diuretics on sodium and chloride transport.
Clin Sci Molec Med. 54 : 679-683.

Brugnara C, Corrocher R, Foroni L, Steinmayr M, Bonfanti F, De Sandre G. 1983. Lithium-sodium countertransport in erythrocytes of normal and hypertensive subjects.
Hypertension. 5 (4) : 529-534.

Brunner H R, Laragh J H, Baer L, Newton M A, Goodwin F T, Krakoff L R, Bard R H, Buhler F R. 1972. Essential hypertension : renin and aldosterone, heart attack and stroke.
N Engl J Med. 441-449.

Brunner H R, Sealy D E, Laragh J H. 1973. Renin as a risk factor in essential hypertension : more evidence.
Am J Med. 55 : 295-301.

Bryant J C. 1975. Earles balanced salt solution : Preparation of the saline. Tissue culture association manual.
1 : 4 : 185-187.

Bulpitt C J. 1981. Sodium excess or potassium lack as a cause of hypertension : A discussion paper.
Journal of the Royal Soc Med. 74 : 896.

Burg M B. 1976. Renal chloride transport and diuretics.
Circulation. 53 (4) : 587-588.

Canessa M, Adragna N, Solomon H S, Connolly T M, Tosteson D C. 1980. Increased sodium-lithium countertransport in red cells of patients with essential hypertension.
N Engl J Med. 302 : 772-776.

Canessa M, Spalvins A, Adragna N, Falkner B. 1984. Red cell sodium countertransport and cotransport in normotensive and hypertensive Blacks.
Hypertension. 6 : 344-351.

Chaudhuri B R. 1978. Down with high blood pressure.
J Indian M A. 10 (7) : 166-167.

Cohen S L, Jhetam D, DaSilva J, Milne F J, Van der Walt A, 1982. Sodium and potassium status, plasma renin and aldosterone profiles in normotensive and hypertensive Johannesburg Blacks.
S Afr Med J. 62 : 941-944.

Cole A C E. 1959. Some observations on heart disease in Dar-es-Salaam.
E. Afr. Med. J. 36 : 538.

Cole H C 1983. Erythrocyte membrane sodium transport in patients with treated and untreated essential hypertension.
Circulation. 68 (1) : 17-22.

Creditor M C, Loschky V K. 1967. Plasma renin activity in hypertension.
Am J Med. 43 : 371-382.

Creditor M C, Loschky V K. 1968. Incidence of suppressed renin activity and of normokalaemic primary aldosteronism in hypertensive negro patients.
Circulation. 37 : 1027-1031.

Cruickshank J K, Beevers D G. 1982. Epidemiology of hypertension : blood pressure in blacks and Whites.
Clin Sci. 62 : 1-6.

Cruz-Coke R, Etcheverry R, Nagel R. 1964. Influence of migration on blood pressure of Easter Islanders.
Lancet, i : 697-699.

Dagher G, Garay R P. 1980. A Na^+, k^+ Co-transport assay for essential hypertension.
Can J Biochem. (58) : 1069-1074.

Dahl L K, Love R A. 1957. Etiological role of sodium chloride in essential hypertension in humans.
JAMA. 164, 4 : 397-400.

Dahl L K, Heine M, Tassinari L. 1962. Effects of chronic salt ingestion. Evidence that genetic factors play an important role in the susceptibility to experimental hypertension.
J Exp Med. 115 : 1173-90.

Dahl L K, Knudsen K D, Heine M, Leitl G. 1967. Effects of chronic excess salt ingestion : Genetic influence on the development of salt and hypertension in parabiotic rats : evidence for a humeral factor.
J Exp Med. 126 : 687-699.

Dahl L K, Leite G, Heine M. 1972. Influence of dietary potassium and sodium/potassium molar ratios on the development of salt hypertension.
J Exp Med. 136 : 318-330.

D'Amico G. 1958. Red cell sodium and potassium in congestive heart failure, essential hypertension and myocardial infarction.
Am J Med Sci. 236 : 156-161.

Davidson J S, Opie L H, Kedding B. 1982. Sodium-potassium cotransport activity as genetic marker in essential hypertension.
Br Med J. 284 : 539-542.

Department of Statistics. Population of South Africa 1904-1970. Report No. 020512 Pretoria. Government Printer.

Dixon N J (ed). 1982 BMDP Statistical Software. Los Angeles : Regents of University of California, Dept of Biomathematics.

Dunn M J. 1970. The effects of transport inhibitors on sodium outflux and influx in red blood cells : evidence for exchange diffusion.
J Clin Investig. (49) : 1804-1814.

Edmonson R P S, Thomas R D, Hilton P J, Patrick J, Jones N F. 1975. Abnormal leucocyte composition and sodium transport in essential hypertension.
Lancet. 1 : 1003-1005.

Eichelberger L. 1943. The distribution of water and electrolytes between blood and skeletal muscle in experimental hypertension.
J Exp Med. 77 : 205-213

Ellory J C, Dunham P B, Logue P J, Stewart G W. 1982. Anion dependent cation transport in erythrocytes.
Phil Trans R Soc. B. 299 : 483-495.

Etkin L N, Mahoney J R, Forsthoefel M W, Echman J R, McSwingan J D, Gillum R F, Eaton J W. 1982. Racial differences in hypertension - associated red cell sodium permeability. Nature. 297 : 588-589.

Freis E D. 1983. How diuretics lower blood pressure. Am Heart J. 106 (2) : 185-187.

Frisancho A R, Leonard W R, Bollettino L A. 1984. Blood pressure in Blacks and Whites and its relationship to dietary sodium and potassium intake. J Chron Dis. 37 (7) : 515-519.

Ganguly A, Weinberger M H 1979. Low renin hypertension. A current review of definition and controversies. Am Heart J. 98 : 5 : 642-648.

Garay R, Nazaret C, Dagher G, Bertrand G, Meyer P. 1981. A genetic approach to the Geography of hypertension : examination of Na^+ - K^+ cotransport in Ivory Coast Africans. Clin Exp Hyper. 3 : 861-870.

Genest J, Nowaczynski W, Boucher R, Kuchell O, Mannel J, Ortega-Rojo M. 1975. Aldosterone and renin in essential hypertension. Can Med Assoc J. 113 : 421-31.

Ghosh B N, Bausal R D, Bhardway U D, Joshi S C. 1983. A blood pressure survey in Simla Hills. J Indian M A. 80 (3-4) : 47-52.

Gillum R F. 1979. Pathophysiology of hypertension in Blacks and Whites. Hypertension. i (5) : 468-475.

Goodman and Gillman. 1980. The pharmacological basis of therapeutics. MacMillan Publishing Co. 901.

Grim C E, Luft F C, Miller J Z, Meneely G R, Battarbee H D, Hames C G, Dahl L K. 1980. Racial differences in blood pressure in Evans County Georgia. Relationship to sodium and potassium intake and plasma renin activity.
J Chron Dis. 33 : 87-94.

Gulati C, Channick B J, Adlin U, Biddle L M, Marks A D. 1975. Low renin hypertension.
Arch Intern Med. 135 : 260-263.

Gyton A C. 1986. Textbook of medical physiology.
W B Saunders Company. 944-948. 254-255. 912-914.

Hamlyn J M, Ringel R, Schoeffer J. 1982. A circulating inhibitor of Na + K -ATP associated with essential hypertension.
Nature. 300 : 650-652.

Heyman A, Karp H R, Heyden S, Bartel A, Cassel J C, Tyroler A, Hames C G. 1971. Cerebro-vascular disease in the biracial population of Evans County, Georgia.
Arch Intern Med. 128 : 949-955.

Hicks C S, Matters R F. 1933. The standard metabolism of the Australian Aborigines.
Aust J Exp Biol Med Sci. 11 : 177-183.

Hilton P J, Patrick J. 1973. Sodium and potassium flux rates in normal human leucocytes in an artificial extracellular fluid.
Clin Sci. 44 : 439-445.

Holland O B, Fairchild C. 1982. Renin classification for diuretic and beta-blocker treatment of Black hypertensive patients.
J Chron Dis. 35 : 179-182.

Holland O B, Gomez-Sanchez C, Fairchild C, Kaplan N.M. 1979. Role of renin classification for diuretic treatment of Black hypertensive patients.
Arch Intern Med. 139 : 1365-1370.

Hout S J, Pamnani M B, Clough D L. 1983. Sodium-potassium pump activity in reduced renal mass hypertension.
Hypertension. 5 (Suppl I) : 94-100.

Hsiao Z K, Wang S Y, Hong Z G, Liu K, Cheng T Y, Stamler J, Tao S C. 1986. Timed overnight sodium and potassium excretion and blood pressure in steel workers in North China.
J Hypertens. 4 : 345-350.

Iimura O, Kijima T, Kikuchi K. 1981. Studies on the hypotensive effect of high potassium intake in patients with essential hypertension.
Clin Sci. 61 (7) : 77s-80s.

Isaacson C, Kincaid-Smith P. 1961. Study of the kidney in the Bantu with hypertension.
Brit Heart J. 24 : 372-374.

Isaacson C. 1977. The changing pattern of heart disease in South African Blacks.
S Afr Med J. 52 : 793-798.

Iwai J, Dahl L K, Knudsen K D. 1973. Genetic influence on the renin angiotensin system.
Circ Res. 32 : 678-684.

Johnson B C, Remington R D. 1961. A sampling study of blood pressure levels in White and Negro residents of Nassau, Bahamas.
J Chron Dis. 13 : 39.

Kaminer B, Lutz W P M. 1960. Blood pressure in Bushmen of the Kalahari dessert.
Circulation. 22 : 289.

Kaplan N M, Kem D C, Holland O B, Kramer N J, Higgins J T, Gomez-Sanchez C. 1976. The intravenous furosemide test : a simple way to evaluate renin responsiveness.
Ann Intern Med. 84 : 639-645.

Karlberg B E, Kagedal B, Tegler L, Tolagen K. 1976. Renin concentrations and effects of propranolol and spironolactone in patients with hypertension.
Br med J. 1 : 251-254.

Kcan B H. 1944. The blood pressure of the Cuna Indians.
Am J Trop Med. 24 : 341-3.

Kcan B H, Hammill J F. 1949. Anthropology of arterial tension.
Arch Int Med. 83 : 355.

Kempner W. 1948. Treatment of hypertensive vascular disease with rice diet.
Am J Med. 4 : 545-577.

Khaw K T, Thom S. 1982. Randomised double blind cross-over trial of potassium on blood pressure in normal subjects.
Lancet. ii : 1127-1129.

Khokhar A M, Slater J D H, Jowett T P, Payne N N. 1976. Suppression of the renin-aldosterone system in mild essential hypertension.
Clin Sci mol med. 50 : 269-276.

Khosla T, Lowe C R. 1967. Indices of obesity derived from body weight and height.
Brit J Prev Soc Med. 21 : 122-128

Kisch E S, Dluhy R G, Williams G H. 1976. Enhanced aldosterone response to angiotensin II in human hypertension.
Circ Res. 38 : 502-505.

Kloppenberg P W C, Drayer J M, Smals A G H, Benraad T J. 1982. Aldosterone, sodium and potassium in essential hypertension. Clin and Exper hyper. Theory and practise. A4 (9,10) : 1881-1893.

Kloppers P J, Meiring J H, Van der Walt I D, de Wet C S. 1981. Hypertension in the Bushman.
S Afr Med J. 26 Dec : 982.

Kuller L, Seitser R. 1967. Cerebrovascular disease mortality in Maryland.
Am J Epidemiol. 2 : 442-450.

Laragh J H, Baer L, Brunner H R, Buhler F R, Sealy J E, Vaughan E D. 1972. Renin, Angiotensin and aldosterone system in pathogenesis and management of hypertensive vascular disease.
Am J Med. 52 : 633-652.

Laragh J H, Sealy J E, Sommers S C. 1966. Patterns of adrenal secretion and urinary excretion of aldosterone and plasma renin activity in normal and hypertensive subjects.
Circ Res. (suppl) 1 : 158.

Lijnen P, M'buyamba-Kabangu J R, Fagard P, Staessen J, Lissens W, Moerman E, Amery A. 1985. Racial differences in renin, aldosterone, prostaglandin and kallikrein of normal male subjects.
Ann Soc Belge Med Trop. 65 : 293-301.

Lilley J J, Hsu L, Stone R A. 1976. Racial disparity of plasma volume in hypertensive man.
Ann Intern Med. 84. (6) : 707-708.

Liu K, Dyer A R, Cooper R S, Stamler R, Stamler J. 1979. Can overnight urine replace 24 hour urine collections to assess salt intake?
Hypertension. 1 : 529-536.

Ljungman S, Aurell M, Hartford M, Witstrand J, Berglund G. 1982. Blood pressure in relation to the Renin Angiotensin Aldosterone system.
Acta Med Scand. 211 : 351-360.

Losse H, Zidek W, Zumkley H, Wessels F, Vetter H. 1981. Intracellular sodium as a genetic marker of essential hypertension.
Clin exp hypertens. (A) 3 (4) : 627-640.

Lowenstein F W. 1961. Blood pressure in relation to age and sex in the tropics and subtropics.
Lancet. i : 389-392.

Luft F C, Grim C E, Higgins J T, Weinberger H M. 1977. Differences in response to sodium administration in normotensive White and black subjects.
J Lab Clin Med. 3 : 555-561.

Luft F C, Rankin L I, Bloch R Weyman A E, Willis L R, Murray R H Grim C E, Weinberger M H. 1979. Cardiovascular and humoral responses to extremes of sodium intake in normal Black and White men.
Circulation. 60 : 697-706.

Maddocks I. 1967. Blood pressure in Melanesians.
Med J Aust. 1 : 1123-1126.

MacGregor G A, Smith S J, Markandu N D, Banks R A, Sagnella G A. 1982. Moderate potassium supplementation in essential hypertension. Lancet. 8298 : 567-570.

Matlou S M, Isles C G, Higgs A, Milne F J, Murray G D, Schultz E, Starke I F (1986). Potassium supplementation in Blacks with mild to moderate essential hypertension.
J Hypertens. 1986. 4 : 61-64.

Mathur K, Wahi P N, Gahlant D. 1963. Blood pressure studies in the adult population of Agra, India.
The American J Cardiol. Jan. 61-65.

Marks S L, Maxwell M H. 1979. Tigerstedt and the discovery of rennin. An historical note. Hypertension. 4 : 384-388.

M'Buyamba-Kabangu J R, Fagard R, Lijnen P, Mbuy Wa, Mbuy R, Staessen J, Amery A. 1986. Blood pressure and urinary cations in urban Bantu of Zaire.
Am J Epidemiol. 124 : 957-968.

McDonough J R, Garrison G E, Haines C G. 1964. Blood pressure and hypertensive disease among Negro and Whites.
Ann Intern Med. 61 : 208-228.

Meade T W, Imeson J D, Gordon D, Peart W S. 1983. The epidemiology of plasma renin.
Clin Sci. 64 : 273-280.

Miall W E. 1959. Follow up study of arterial pressure in the population of a Welsh Mining Valley.
Br Med J. ii : 1204-1210.

Milne FJ, Gear JSS, Laidley L, Ritchie M, Schultz E 1980. Spot urinary electrolyte concentrations and 24 hour excretion.
Lancet. i : 1135.

Modlin M, Isaacson L C, Jackson W P U. 1963. Normal Bantu urine - a preliminary report.
S Afr Med J. 37 : 121-122.

Morse W R, Beh Y T. 1937. Blood pressure amongst aboriginal ethnic groups of Szechwan province, West China.
Lancet. i : 966-967.

Munro-Faure A D, Hill D M, Anderson J. 1971. Ethnic differences in human blood cell sodium concentration.
Nature. 231 : 457-458.

Nechay B R. 1977. Biochemical basis of diuretic action.
J Clin Pharmacol, October : 626-641.

Neter J, Wasserman W. 1974. Applied linear statistical models. Richard D. Irwin Inc. Homewood. Illinois. p. 730.

Nielsen J R, Johansen T, Jest P, Petersen K E, Klitgaard N A. 1986. Cell membrane disturbance and the pathogenesis of essential hypertension: Several hypothesis.
Scand J Clin Lab Invest. 46 : Supplement 180 : 7-10.

Nowaczynski W, Kuchel O, Genest J. 1975. Aldosterone, deoxycorticosterone, 18 hydroxydeoxy corticosterone and progesterone in benign essential hypertension. In: Epidemiology and control of hypertension (ed. O Paul) p 265.

Overlack A, Muller H M, Kolloch R, Ollig A, Moch B, Kleinmann R, Krück F, Stumpe K. 1983. Long term antihypertensive effect of oral potassium in essential hypertension.
Journal of Hypertension. 1 (52) : 165-167.

Padmavati S, Gupta S. 1959. Blood pressure studies in rural and urban groups in Delhi.
Circulation. Vol XIX. March. 395-405.

Parfrey P S, Markandu N D, Roulston J E, Jones B E, Jones J C, MacGregor G A. 1981. Relation between arterial pressure, dietary sodium intake and renin system in essential hypertension.
Br Med J. 283 : 94-97.

Pietinen P I, Findley T W, Clausen J D, Finnerty F A, Altschul A M. 1976. Studies in community nutrition, estimation of sodium output.
Prev Med. 5 : 400-407.

Prior I A M, Grimely E. 1969. Sodium intake and blood pressure in Pacific populations.
Israel J Med Sci. 5 : 608-11.

Poston L, Sewell R B, Wilkinson S P, Richardson P J, Williams R, Clarkson E M, Mac Gregor G A, de Wardener H E. 1982. Evidence for a circulating sodium transport inhibitor in essential hypertension.
Br Med J. 282 : 847-849.

Ray A A. (ed) 1982. SAS User's Guide. Statistics. Cary, North Carolina. SAS Institute.

Richards A M, Nicholls, M G, Espiner E A, Ikram H, Maslowski A H, Hamilton E J, Wells E J. 1984. Blood pressure response to moderate sodium restriction and to potassium supplementation in mild essential hypertension.
Lancet. i : 757-761.

Ringel R E, Hamlyn J M, Schaefer J, Hamilton B P, Kowarski A, Blaustein M P, Berman M A. 1984. Red cell cotransport activity and sodium content in Black men.
Hypertension. 6 (5) : 724-730.

Sagar S, Anand I S, Sharma R, Rai R, Jani P K, Chakravati R N. 1982. Plasma renin activity in essential hypertensives in North India.
Indian J Med Res. 75 : 101-106.

Sasaki N. 1962. High blood pressure and the salt intake of the Japanese.
Japanese Heart Journal. 3 : 313-324.

Sasaki N. 1979. The salt factor in apoplexy and hypertension. Epidemiological studies in Japan. In: Perspectives in cardiovascular Research. Edited by Katz AM. Vol. 4 : 467-474.

Saunders G M, Barcroft H. 1942. Blood pressure studies on Negro and White men living in Virgin Islands of United States.
Amer Heart J. 23 : 410.

Schneckcloth R E, Stuart K L, Corcoran A C, Moore F E. 1962. Arterial pressure and hypertensive disease in a West Indian Negro population. Report of a survey in St. Kitts, West Indies.
Amer Heart J. 63 : 607.

Seedat Y K, Seedat M A, Nkomo M N. 1978. The prevalence of hypertension in the urban Zulu.
S Afr Med J. 53 : 923-927.

Seedat Y K, Seedat M A, Reddy K. 1978. The prevalence of Hypertension in the Indian population of Durban.
S Afr Med J. 54 : 10-15.

Seedat Y K, Seedat M A, Hackland D B T. 1982. Biosocial factors and hypertension in urban and rural Zulus.
S Afr Med J. 61 : 999-1002.

Seedat Y K, Seedat M A. 1982. An inter-racial study of the prevalence of hypertension in an urban South African population.
Trans R Soc Trop Med Hyge. 76 (1) : 62-71.

Seftel H C, Johnson S, Muller E A. 1980. Distribution and biosocial correlations of blood pressure levels in Johannesburg Blacks.
S Afr Med J. 57 : 313-320.

Sever P S, Peart W S, Meade T W, Davis I B, Gordon D. 1979. Ethnic differences in blood pressure with observations on noradrenaline and renin.
Clin Exp Hypertens. 1 (6) : 733-744.

Sever P S, Peart W S, Gordon D, Beighton P. 1980. Blood pressure and its correlates in urban and tribal Africa.
Lancet. ii : 60-64.

Shaper A G, Leonard P J, Jones K W, Jones M. 1969. Environmental effects on body build, blood pressure and blood chemistry of nomadic warriors serving in the army in Kenya.
E Afr Med J. 46 : 282.

Simon G Conklin D J. 1986. In vivo erythrocyte sodium concentration in human hypertension is reduced not increased.
J Hypertens. 4 : 71-75.

Simpson F O, Waal-Manning H J, Bolli P, Phelan E L, Spears G F S. 1978. Relationship of blood pressure to sodium excretion in a population survey.
Clin Sci Mol Med. 55 : 373s - 375s.

Smith J B, Ash K O, Hunt S C, Hentschel W M, Sprowell W, Dadone M M, Williams R R. 1984. The three red cell sodium transport systems in hypertensive and normotensive Utah adults.
Hypertension. 6 : 159-166.

Smith S J, Markandu N D, Sagnella G A, Poston L, Hilton P J, MacGregor G A. 1983. Does potassium lower blood pressure by increasing sodium excretion? A metabolic study in patients with mild to moderate essential hypertension.
J Hypertens. 1 (Suppl 2) : 27-30.

Staessen J, Bulpitt C, Fagard R, Joossens J U, Lignen P, Amery A. 1981. Salt and blood pressure in Belgium.
J Epidemol Community Health. 35 : 256-261.

Tedde R, Sechi L A, Marigliano A, Campus S. 1983. Sodium concentration in erythrocytes in arterial hypertension.
J Hypertens. 1 (Suppl 2) : 140-141.

Thomas W A. 1927. Health of a carnivorous race : a study of the Eskimo.
JAMA. 88 : 1559-1560.

Thulin T, Karlberg B E, Scherstein B. 1978. Plasma renin activity, aldosterone and sodium excretion in women with high and low casual blood pressure levels.
Acta Med Scand. 203 : 405-410.

Tobian L, Binion J. 1952. Tissue cations and water in arterial hypertension.
Circulation. 5 : 754-758.

Tolagen K. 1978. Aldosterone in primary hypertension, relationship to plasma renin activity and urinary electrolytes and a comparison with normotensive subjects.
Scand J Clin Lab Invest. 38 : 487-493.

Trowell H C. 1980. From normotension to hypertension in Kenyans and Ugandans. 1928-1978.
E Afr Med J. 57 (3) : 167-173.

Truswell A S, Kennelly B M, Hansen J D L, Lee R B. 1972. Blood pressure of !kung bushmen in Northern Botswana.
Am Heart J. 84 (1) : 5-12.

Turner P P. 1962. The pattern of disease as seen by medical admissions to the Coast Province General Hospital in 1960.
E Afr Med J. 39 : 121.

Touyz R M. 1984. Magnesium, calcium, sodium and potassium status in normotensive and hypertensive Johannesburg residents.
M Sc Thesis. University of Witwatersrand.

Tuck M L, Gross C, Maxwell M H, Brickman A S, Krasnoshtein G, Mayes D. 1984. Erythrocyte Na^+, K^+ cotransport and Na^+, K^+ pump in black and caucasian hypertensive patients.
Hypertension. 6 : 536-544.

Tuck M L, Williams G H, Cain J P, Sullivan J M, Dluhy R B. 1973. Relation of age, diastolic pressure and known duration of hypertension to presence of low renin essential hypertension.
Am J Cardiol. 32 : 637-642.

Valiallah A A, Gear J S S. 1983. The prevalence of hypertension in the Indian population of Roodepoort (Transvaal).
S Afr Med J. 63 : 964-965.

Verma B L, Srivastava R N. 1983. Hypertension in Indian rural adults.
Int J Epidemiol. 12 (3) : 374-375.

Voors A W, Dalferes E R, Frank G C, Aristimuno G G, Berenson G S. 1983. Relation between ingested potassium and sodium balance in young Blacks and Whites.
Am J Clin Nutr. 37 : 583-594.

Wainwright J. 1969. Cardiovascular disease in the Asiatic (Indian) population of Durban.
S Afr Med J. 43 : 136-138.

Walker W G, Whelton P K, Saito H, Russell R P, Herman J. 1979. Relation between blood pressure and renin, renin substratae, angiotensin II, aldosterone and urinary sodium and potassium in 574 ambulatory subjects.
Hypertension. 1 (3) : 287-291.

Wallach L, Nyarai I, Dawson K G. 1975. Stimulated renin : A screening test for hypertension.
Ann Intern Med. 82 : 27-34.

Walter U, Distler A. 1982. Abnormal sodium efflux in erythrocytes of patients with essential hypertension.
Hypertension. 4 : 205-210.
Walter U, Distler A. 1980. Effects of oubain and furosemide on ATPase - activity and sodium transport in erythrocytes of normotensive and of patients with essential hypertension. In: Intracellular electrolytes and arterial hypertension. Edited by: H Zumkley and H Loose. Georg Theime Verlag. Stuttgart, New York : 170-182.

Watson R L, Langford H G. 1970. Usefulness of overnight urines in populations.
Am J Clin Nutr. 23 : 290-304.

Weder A B, Torretti B A, Julius S. 1984. Racial differences in erythrocyte cation transport.
Hypertension. 6 (1) : 115-123.

Wessels F, Zumkley H. 1980. Sodium metabolism of RBC in hypertensive patients. In: Zumkley H, Losse E. Ed. Intracellular electrolytes and arterial hypertension.
Georg Theime Verlag. Stuttgart. New York : 59-68.

Williams A W. 1944. Heart disease of the native population of Uganda. Part IV. Hypertensive heart disease.
E Afr Med J. 21 : 328-368.

Williams A W. 1941-1942. The blood pressure of Africans.
E Afr Med J. 18 : 109.

Williams G H. 1982. The renin-angiotensin system and hypertension.
Clin Exp Pharmcol Physiol. 7 : 31s - 40s.

WHO. 1962 Arterial hypertensive ischaemic heart disease : preventative aspects. World Health Organisation Technical Report No. 231. Geneva: WHO.

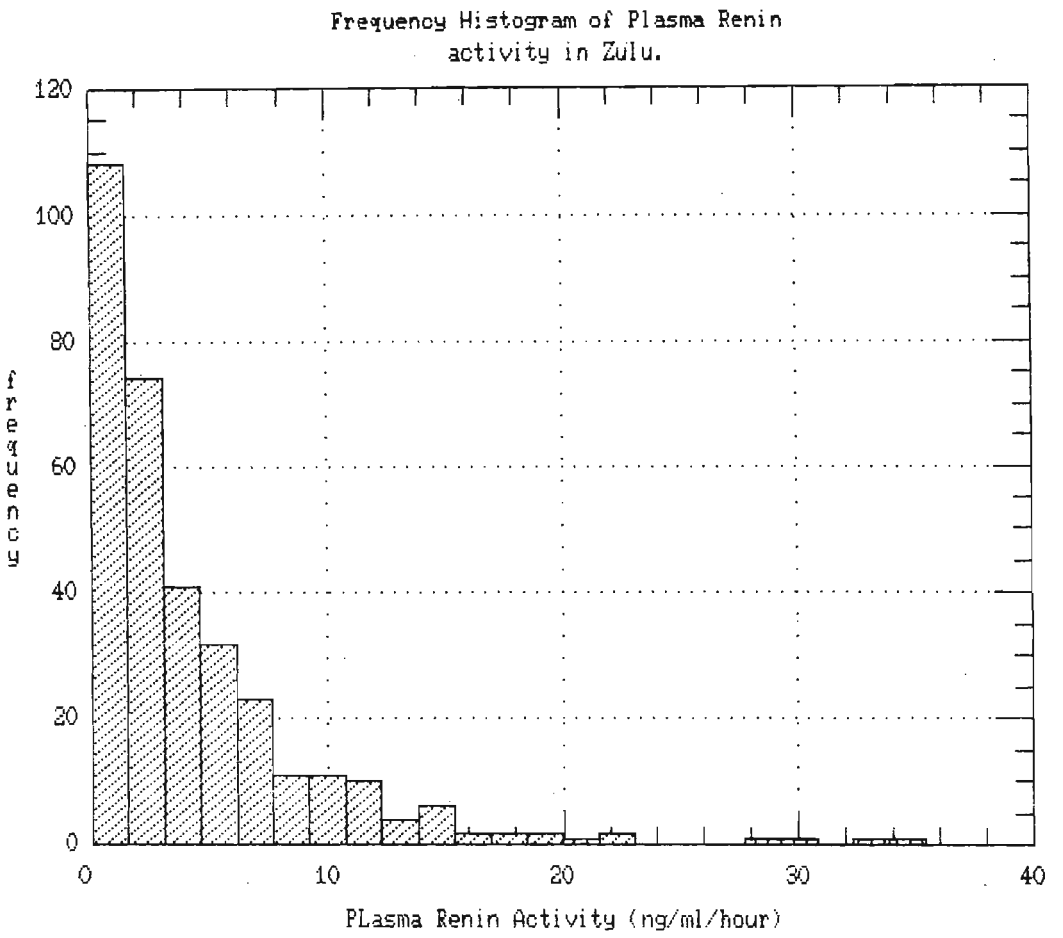
Zidek W, Knorr M, Losse H, Baumgart P, Vetter H. 1983. Sodium and calcium in lymphocytes and erythrocytes in essential hypertension.
J Hypertens. 1 (25) : 3s - 5s.

Zidek W, Losse H, Schmidt W, Fehske K J, Vetter H. 1984. Intracellular electrolytes during anti-hypertensive treatment with a loop diuretic.
J Hypertens. 2 : 393-395.

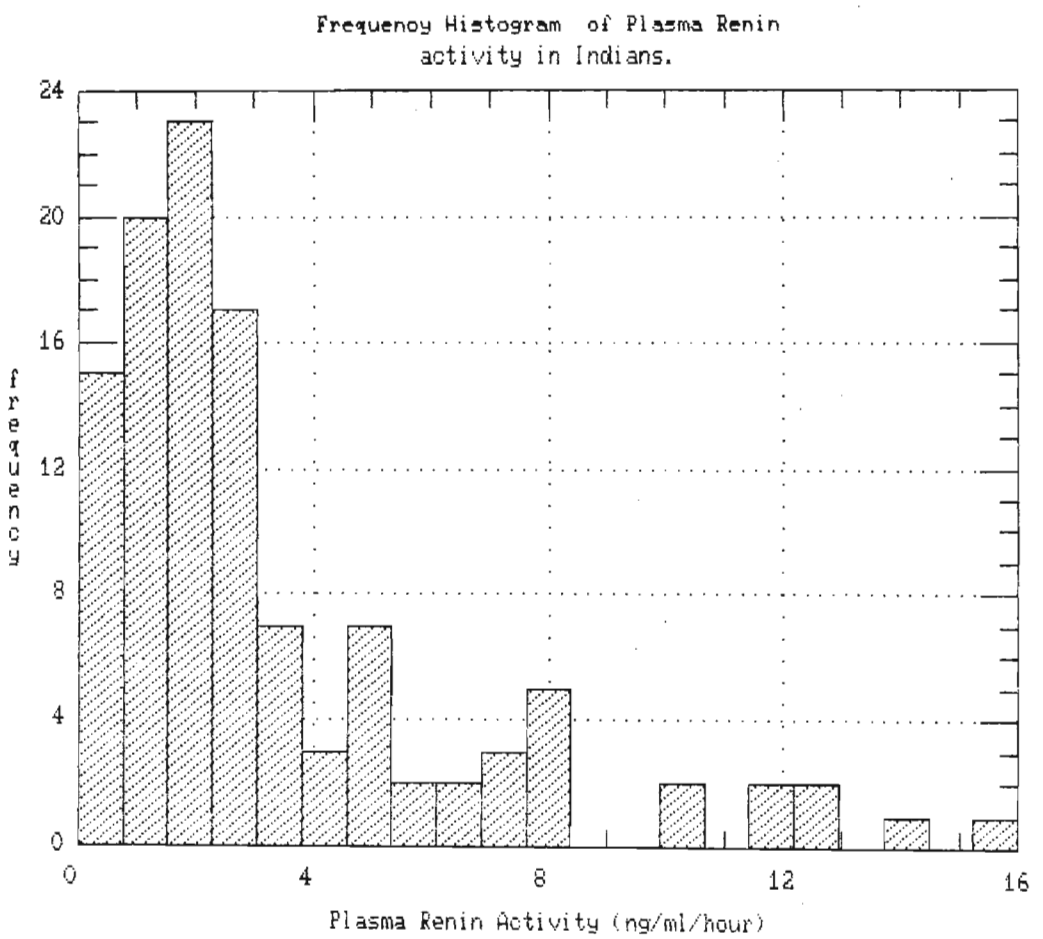
Zumkley H, Wessels F. 1980. Diagnostic aspects of increased sodium concentration in red blood cells. In: Zumkley H, Losse H Ed. Intracellular electrolytes and arterial hypertension. Georg Thieme Verlag. Stuttgart. New York : 104-110.

Zoccali C, Cumming A M M, Hutcheson M J, Barnett P, Semple P F. 1985. Effects of potassium on sodium balance, renin, noradrenaline and arterial pressure.
J Hypertens. 3 : 67-72.

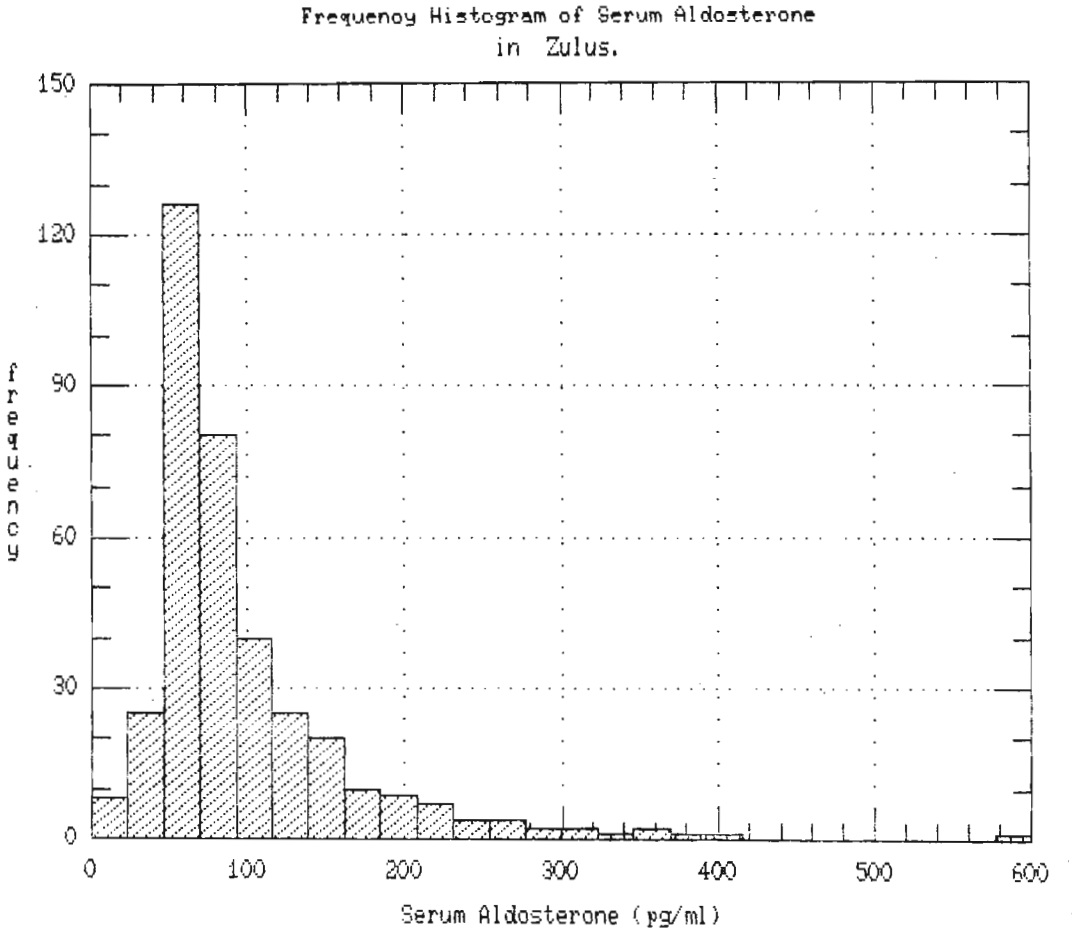
ADDENDUM 1


Frequency Histogram of Plasma Renin activity in Zulu.
120
100
80
60
40
20
0
frequency
0
10
20
30
40
PLasma Renin Activity (ng/ml/hour)

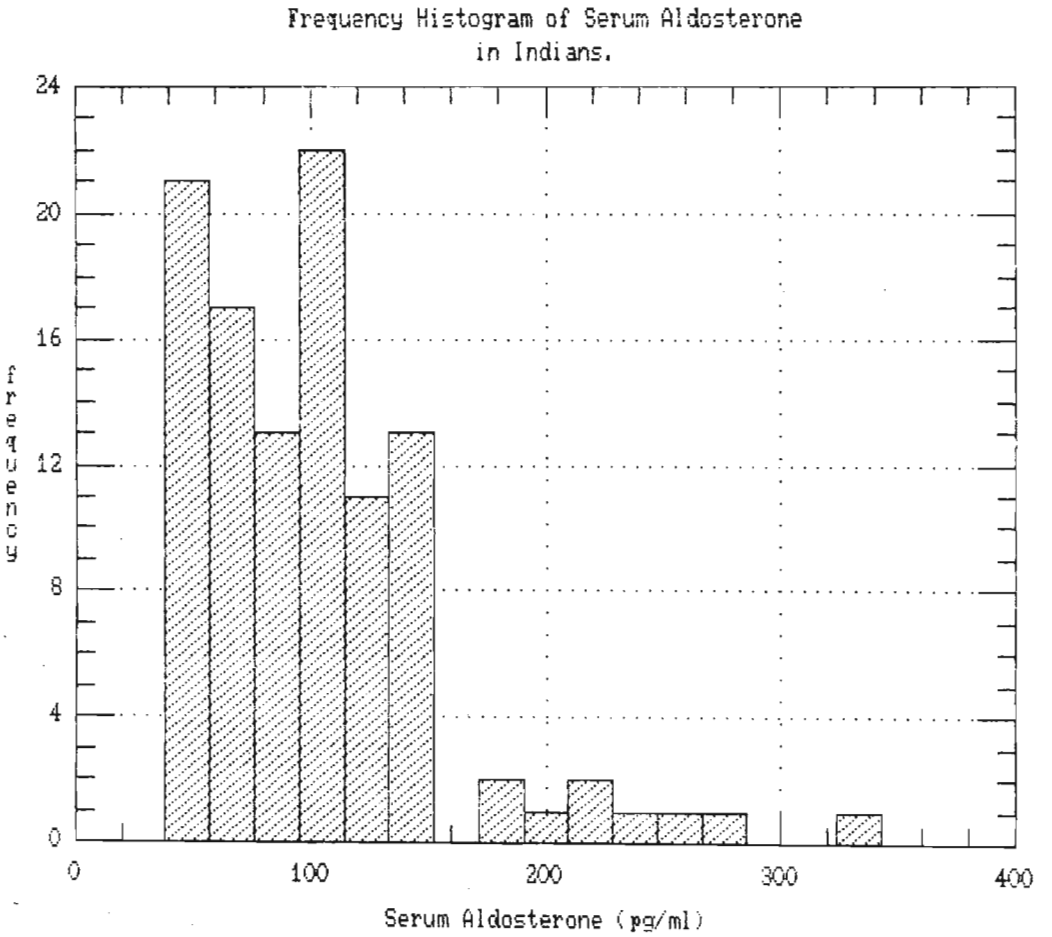
ADDENDUM 2


Frequency Histogram of Plasma Renin activity in Indians.
24
20
16
12
8
4
0
frequency
0
4
8
12
16
Plasma Renin Activity (ng/ml/hour)

ADDENDUM 3


Frequenoy Histogram of Serum Aldosterone
in Zulus.
150
120
90
60
30
0
frequency
0
100
200
300
400
500
600
Serum Aldosterone (pg/ml)

ADDENDUM 4


Frequency Histogram of Serum Aldosterone
in Indians.
24
20
16
12
8
4
0
frequency
0
100
200
300
400
Serum Aldosterone (pg/ml)