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PSEUDOHYPOALDOSTERONISM AND MINERALOCORTICOID RECEPTOR ABNORMALITIES

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Summary—Pseudohypoaldosteronism is a rare inherited disease characterized by renal salt loss, hyperkalemia and metabolic acidosis despite highly elevated aldosterone values. We previously reported absent or reduced numbers of mineralocorticoid receptors in mononuclear leukocytes and defective effector mechanism as shown by no response *in vitro* to the incubation of aldosterone in terms of intracellular electrolyte content. We have studied the inheritance of this disorder in ten families and found two different kinds of inheritance: autosomal recessive—often in interrelated families—and autosomal dominant in unrelated families. Parallel studies in the families with the autosomal dominant form of inheritance demonstrated in addition that the effector mechanism of aldosterone is impaired *in vitro* both in the affected patients and in the carrier relatives characterized by a low number of mineralocorticoid receptors.

INTRODUCTION

Pseudohypoaldosteronism is characterized by sodium wasting in the neonatal period associated with hyponatremia, hyperkalemia, metabolic acidosis and extremely high aldosterone values [1, 2]. We reported in a previous paper that in patients with pseudohypoaldosteronism the number of mineralocorticoid receptors in mononuclear leukocytes are absent or extremely low, thus suggesting a genetic defect of the mineralocorticoid receptors [3]. It is interesting to note that the clinical symptoms of this disorder ameliorate spontaneously after a period of sodium supplementation and that in general in older patients therapy can be discontinued [4]. However, serum aldosterone levels remain elevated and mineralocorticoid receptors low. It seems that in these patients factors other than aldosterone are in someway able to maintain electrolyte balance [5].

In the present paper we review the inheritance of pseudohypoaldosteronism regarding not only aldosterone receptors but also aldosterone effector mechanisms [6].

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EXPERIMENTAL

Ten families with a total of eleven patients were studied. In all cases plasma aldosterone and the number of mineralocorticoid receptors was evaluated in mononuclear leukocytes. In one family the electrolyte content in mononuclear leukocytes were measured and related to receptor number.

Plasma aldosterone was measured by routine RIA. The number of mineralocorticoid receptors was determined as described previously [7]. Peripheral mononuclear leukocytes were separated from heparinized blood by Percoll gradient centrifugation and the cells washed several times in order to free them of endogeneous steroids. An aliquot of cells was then incubated with increasing concentrations of [^{3}H]aldosterone and an excess of RU-26988 to exclude binding to glucocorticoid receptors. In parallel incubation was performed by adding an excess of radioinert aldosterone to determine unspecific binding. After incubations cells were washed, the radioactivity counted and the number of receptors calculated according to the Scatchard analysis [8].

The electrolyte content of cells was determined after incubating washed cells in medium alone or with the addition of aldosterone and by comparing these data with those obtained immediately after separation. Intracellular electrolytes were

Table 1. A review of the findings of plasma aldosterone and mineralocorticoid receptor determinations in the 10 families studied

	Plasma aldosterone			Mineralocorticoid receptors		
Family	Patients	Father	Mother	Patients	Father	Mother
Families with an autosomal recessive mode of inheritance						
1	⇑⇑	N	N	0/⇓	N	N
2	⇑	N	N	⇓	N	N
3	⇑	N	N	⇓	N	N
4	⇑	N	N	0	⇓	⇓
Families with an autosomal dominant mode of inheritance						
5	⇑	N	⇑	⇓	N	⇓
6	⇑	N	⇑	0	N	⇓
7	⇑	N	⇑	⇓	N	⇓
8	⇑	N	N	⇓	N	⇓
9	⇑	N	⇑	⇓	N	⇓
10	⇑	N	⇑	0	N	⇓

N: normal level; (⇑) increased; and (⇓) decreased.

measured with flame photometry and results expressed as mmol/kg wet cells [9].

RESULTS

Table 1 presents the results in the 10 families studied. The inheritance of pseudohypoaldosteronism was interpreted to be recessive in 4 families (3 of them being related) and dominant in the others (none of these parents were interrelated). In the families with a dominant form of inheritance, we found carriers of the defect with low mineralocorticoid receptors and high plasma aldosterone values indistinguishable from affected and symptomatic patients. These individuals were entirely asymptomatic. The electrolyte content of mononuclear leukocytes of the symptomatic patients as well as the asymptomatic carriers did not respond to the incubation with aldosterone as shown as an example in 1 family in Fig. 1.

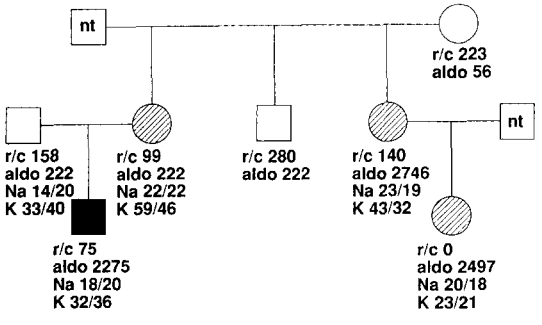

Fig. 1. Family pedigree with one affected patient (■) and three asymptomatic members (◎): r/c, receptors/cell (normal range in children, 100–400; normal range in adults, 150–400); aldo, aldosterone (normal range, 27–695 pmol/l); nt, not tested; and Na and K represent sodium and potassium content before/after incubation with aldosterone given as mmol/kg wet cells.

DISCUSSION

In all patients with pseudohypoaldosteronism mineralocorticoid receptors and effector mechanisms are deficient. At least two different interpretations of these results are feasible: 1, there is a genetic absence or a marked deficiency of mineralocorticoid receptors itself in target tissues; or 2, the reduction or absence of mineralocorticoid receptors is not the primary defect but is the consequence of an abnormality at the level of the plasma membrane which impaires the action of aldosterone, which in turn stimulates the aldosterone synthesis via the renin–angiotensin-system. Consequently the high aldosterone levels down-regulate mineralocorticoid receptors.

From our data we believe that a genetic defect causing the absence or the deficiency of mineralocorticoid receptors is more likely: 1, in cases of primary or secondary hyperaldosteronism mineralocorticoid receptors are down-regulated [10, 11] but never to the level seen in pseudohypoaldosteronism [3]; 2, in patients with pseudohypoaldosteronism who recover from the clinical symptoms aldosterone levels remain high and receptors low even without any electrolyte abnormalities or signs of volume depletion; and 3, in the asymptomatic carriers of pseudohypoaldosteronism the biochemical picture and the receptor status is consistent with pseudohypoaldosteronism but these individuals never show any signs of sodium wasting [6].

As already mentioned, two kinds of inheritance have been observed [6] i.e. a recessive form of inheritance with the most severe expression of clinical symptoms in the interrelated families; and a dominant form of inheritance, with asymptomatic carriers, which seems to be more frequent in our families and is also more frequently reported in the literature. The results in our families with the dominant form of inheritance are compatible with a X-chromosomal dominant form of inheritance, since in our cases only mothers are the asymptomatic carriers with biochemical abnormalities. However there are families reported in which there is a father-to-son transmission thus excluding a X-chromosomal location of the gene [13]. In addition recent studies have located the gene encoding for the mineralocorticoid receptors to the chromosome 4 thus further supporting an autosomal dominant form of inheritance [14].

Intracellular electrolyte content is normal in mononuclear leukocytes of patients with pseudo-

hypoaldosteronism who were in sodium balance. In contrast to normal controls, however, *in vitro* incubation of these cells with aldosterone is unable to prevent the loss of intracellular sodium and potassium. Since this phenomenon can be observed within 30 min it is unlikely to be a receptor related effect but is more likely due to secondary consequences of the mineralocorticoid receptor defect.

In vitro incubation of mononuclear leukocytes from healthy controls with aldosterone is able to prevent electrolyte loss, but unable to increase intracellular electrolyte content. Thus, we might speculate that similarly *in vivo* aldosterone is not the major determinator of intracellular electrolyte concentrations.

A possible mechanism seems to be that aldosterone is able to increase the number of sodium channels in the plasma membranes and that all the subsequent steps are not directly related to the receptor but to changes in electrolyte concentrations and potential differences. Furthermore one has to relate the measurement of intracellular electrolyte content not only to aldosterone alone but also to other mechanisms by whom the action of aldosterone can be impaired. Thus we could already show, that the atrial natriuretic factor can *in vitro* impair the action of aldosterone [12]. Further factors seem to be possible and have to be included in the consideration.

In conclusion, we postulate that a defect in the mineralocorticoid receptor is the cause of pseudohypoaldosteronism. The impaired response of mononuclear leukocytes *in vitro* of patients with pseudohypoaldosteronism regarding intracellular electrolyte content is a secondary phenomenon not primarily related to the receptor defect.

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