Volume 317, number 3, 211–215 FEBS 12096 February 1993

Transcriptional activation of adrenocortical steroidogenic genes by high potassium or low sodium intake

André Tremblay* and Jean-Guy LeHoux

Department of Biochemistry, Faculty of Medicine, University of Sherbrooke, Sherbrooke, Quebec, J1H 5N4, Canada

Received 29 December 1992

We have previously shown that the long-term alterations in the intake of sodium and potassium which stimulated aldosterone production in the rat adrenal significantly increased cytochrome $P450_{scc}$ ($P450_{scc}$) and $P450_{11\beta}$ ($P450_{11\beta}$) mRNA's and also the mRNA of their electron donor adrenodoxin. In the present study run-on analyses showed an accumulation of nascent RNA in isolated nuclei of zona glomerulosa cells in K^+-supplemented and Na^+-depleted rats for $P450_{scc}$ (5- and 6-fold), 3β-HSD (3.6- and 2.0-fold) and $P450_{11\beta}$ (6.0- and 6.1-fold), but not for $P450_{c21}$ (1.4- and 1.1-fold). In contrast, that of adrenodoxin decreased (0.6-fold) in high K^+ and remained near control (1.3-fold) in low Na^+ intake. Moderate variations in the rate of transcription of $P450_{scc}$, $P450_{c21}$, $P450_{11\beta}$ and adrenodoxin genes were observed in the zona fasciculata-reticularis cells of the treated rats. Our results thus demonstrated that positive modulators of aldosterone such as long-term K^+ supplementation and Na^+ restriction provoked an increase in transcription of the genes encoding key regulatory steroidogenic enzymes of aldosterone biosynthesis in the zona glomerulosa. The rates of transcription of the genes encoding 3β-HSD and $P450_{c21}$, two enzymes catalyzing intermediate steps in the aldosterone pathway, were moderately affected by such treatments. However, according to the known stimulation of adrenodoxin mRNA levels following these treatments, a decreased turnover of the adrenodoxin mRNA rather than initiation of transcription of its gene might be involved in the response to K^+ ions, and partially so in the response to Na^+ restriction. Finally, the effects of salt-modified intake were mainly restricted to the zona glomerulosa cells, which are solely responsible for aldosterone production.

Steroidogenesis; Aldosterone; Adrenal cortex; Run-on; Salt-modified diet

1. INTRODUCTION

The regulation of the expression of the adrenal cortex steroidogenic enzymes has been intensively investigated in relation to tissue specificity and cAMP-dependent mechanisms since the availability of their cDNAs [1–4]. Most of these studies were related to understanding the effects of corticotropin or cAMP mimicking agents on isolated adrenocortical cells in primary culture. The transcriptional regulation of the expression of the steroidogenic enzyme genes has also been studied by John et al. [5] who showed that the enhancement by corticotropin of mRNA levels of steroid hydroxylases in cultured bovine adrenal zona fasciculata-reticularis cells was due to an increased transcription rate of their genes.

In vivo studies showed that adrenal zona glomerulosa steroidogenesis is influenced by modulation of the intake of cations. Indeed, potassium supplementation and sodium restriction have been shown to increase the steroid hydroxylase activities of both the first and the final steps of aldosterone biosynthesis catalyzed by cytochrome ($P450_{scc}$) and cytochrome ($P450_{11\beta}$) respectively [6–9]. We have further demonstrated that in rats, the response to K^+ supplementation and to Na^+ restriction was also located at the messenger RNA level of steroidogenic enzymes since both regimens provoked specific increases in the levels of $P450_{scc}$ and adrenodoxin mRNA in the zona glomerulosa [10,11]. Time-dependent gradual increases have also been observed in levels of the mRNA of these two enzymes and also that of $P450_{11\beta}$ in the zona glomerulosa of either K^+-supplemented or Na^+-restricted rats [12]. These increases were paralleled by similar responses in the levels of the corresponding protein as measured by immunoblotting analysis [12]. Taken collectively, these results suggest that the maintenance of optimal steroidogenic capacity following varitions in cation intake may be a consequence of an increased synthesis of the above mentioned steroidogenic enzymes.

The present study was thus performed to more precisely define the effects of modified cation intakes on the in vivo transcriptional regulation of the expression of the genes of steroidogenic enzymes. Our results demonstrated that positive modulators of aldosterone such as long-term K^+ supplementation and Na^+ restriction provoked a de novo initiation of the transcription of genes encoding steroidogenic enzymes and that such re-

Correspondence address: J.-G. LeHoux, Department of Biochemistry, Faculty of Medicine, University of Sherbrooke, Sherbrooke, Quebec, J1H 5N4, Canada. Fax: (1) (819) 564 5340.

**Present address:* Endocrine Laboratories, Royal Victoria Hospital, and the Department of Medicine, McGill University, Montreal, Canada.

sponses were restricted mainly to the zona glomerulosa, which is reponsible for aldosterone production.

2. MATERIALS AND METHODS

2.1. *Animals and treatments*

Male Long Evans rats, weighing 250–300 g, were obtained from a local breeding colony. The animals were maintained on a sodium-deficient diet (< 0.01 mEq Na^+/g; ICN Biochemicals, Cleveland, OH) with demineralized water as drinking fluid or a high potassium intake (2% KCl, 5% dextrose in tap water) and Purina rat chow. Control rats were fed Purina chow and tap water. After one week on the various treatments, rats were sacrificed by decapitation and blood was collected for hormone analysis. The adrenal glands were removed, freed of fat and separated into the zona glomerulosa and zona fasciculata-reticularis (with medulla).

2.2. *Elongation of nascent RNA chains in isolated nuclei*

The in vitro nuclear transcription assay was performed according to the procedures described previously [13–15] with some modifications. Nuclei were purified from both zona glomerulosa and zona fasciculata-reticularis for run-on experiments. Tissues were homogenized in 15 vols. of 50 mM ice-cold Tris, pH 7.5 containing 25 mM KCl, and 5 mM $MgCl_2$ (TKM) where sucrose was added to a final concentration of 0.25 M. The homogenate was filtered through cheese cloth and layered onto a 4 ml cushion of TKM containing 1 M sucrose and a 8 ml cushion of TKM containing 1.9 M sucrose. The mixture was centrifuged at 27,000 × g for 1 hour at 0°C in a SW 28 rotor. The nuclear pellet was suspended in 20 mM HEPES, pH 7.6, containing 75 mM NaCl, 0.5mM EDTA, 0.85 mM DTT, 0.125 mM PMSF and 50% glycerol and rapidly frozen in liquid nitrogen.

The in vitro nuclear transcription assay was performed by modification of the previously described procedure [14,15]. Nuclei ($1–5 \times 10^7$) were suspended in 4 × reaction buffer (80 mM HEPES, pH 7.8, 200 mM NaCl, 600 mM KCl, 20 mM $MgCl_2$, 16 mM $MnCl_2$, 1.6 mM EDTA and 0.4 mM PMSF) and in vitro elongation of nascent RNA chains was carried out at 26°C for 45 min in a 250 μl cocktail containing 1 × reaction buffer, 5 mM DTT, 2 mM each of GTP, ATP and CTP, 10 mM creatine phosphate, 20 U/ml RNAsin (Promega, Montreal, Quebec), 29% glycerol, 40 μg/ml phosphokinase and 250 μCi of [α-^{32}P]UTP (3000 Ci/mmol; Amersham, Oakville, Ontario). The reaction was stopped by a 20 min incubation at 25°C with 80 U of DNase I, and yeast tRNA was added as carrier. One volume of 20 mM HEPES, pH 7.8 containing 20 mM EDTA and 1% SDS was then added with 240 μg/ml of proteinase K (Boehringer Mannheim, Laval, Quebec) and the mixture was incubated at 42°C for 30 min. RNA was extracted with phenol/chloroform and ethanol-precipitated.

The newly synthesized ^{32}P-labeled transcripts were quantified by hybridization to an excess of plasmid bound to nitrocellulose, the plasmid DNA was first linearized, heat- and alkali-denatured and spotted (3 μg/dot) onto nitrocellulose.

Prehybridization was carried out for 48–72 h at 42°C in a solution containing 33% deionized formamide, 0.5 M NaCl, 50 mM HEPES, pH 7.8, 0.4% SDS, 5 × Denhardt's, 2 mM EDTA, 200 μg/ml tRNA, and 500 μg/ml salmon sperm DNA. The filters were hybridized for 3 days at 42°C in a fresh buffer containing an equal number of counts ($> 5 \times 10^5$ cpm/ml) of newly synthesized ^{32}P-labeled RNA. The filters were washed in 0.1 × SSC, 0.1% SDS at 60°C and autoradiographed, the bands were then quantified by densitometry. Duplicate filters were hybridized with the same ^{32}P-labeled RNA hybridization solutions that were used in the experiments to ensure the absence of any residual radioactivity.

2.3. *Probes and plasmids*

The plasmids used for the run-on assays included: pBSCC containing a full-length cDNA insert specific for bovine $P450_{scc}$ [16]; pBAdx4, which contains a full-length bovine adrenodoxin cDNA insert [17]; pCMV5H3β-HSD, with a full-length insert for human placental 3β-hydroxysteroid dehydrogenase [18]; pBC21–1, containing a partial-length insert coding for bovine $P450_{c21}$ [19]; and p11β-OHase-1.2, which contains a 1.2-kb cDNA insert cloned from a mouse Y1 cDNA library that contains $P450_{11\beta}$ sequences from the 5'-untranslated region to +1153 [20]. This cDNA recognizes 11β-hydroxylase and aldosterone synthase mRNAs [21]. For use as probes for Northern blotting analyses, each of these plasmids was cleaved with an appropriate restriction endonuclease to generate the specific insert which was then isolated by electrophoresis. The probes were labelled with [α-^{32}P]dCTP by random priming using a kit from Amersham (Oakville, Ontario) and the specific activities were always greater than 10^8 cpm/μg DNA.

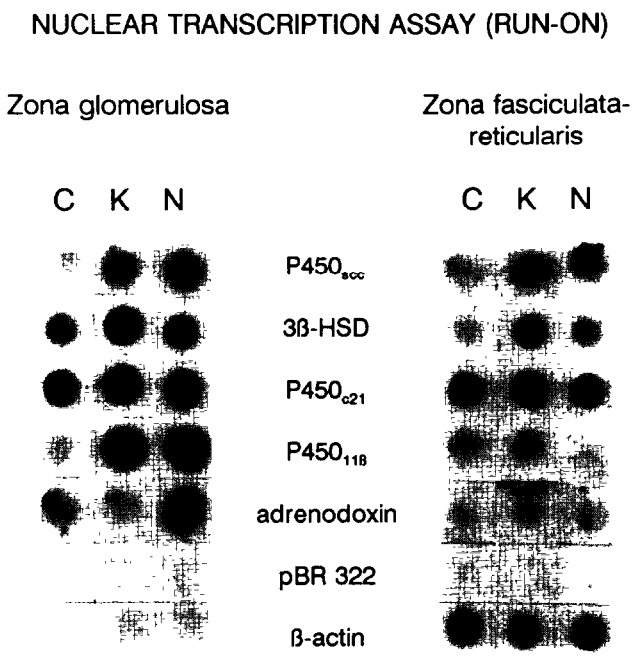

Fig. 1. Nuclear transcription assay of steroidogenic genes. Nuclei were isolated from both the zona glomerulosa and the zona fasciculata-reticularis of control (C), K^+-supplemented (K), and Na^+-depleted (N) rats. In vitro elongation of nascent RNA chains was carried out in the presence of [α-^{32}P]UTP and newly synthetized RNA transcripts were quantified by hybridization to specific cDNAs as indicated. The plasmid pBR322 was used as a measure of non-specific hybridization and the β-actin cDNA served to correct for amounts of [^{32}P]RNA hybridized in each sample.

2.4. *Analytical methods*

Plasma aldosterone was measured by the Coat-A-Count RIA procedure (Diagnostic Products, Los Angeles, CA), plasma corticosterone was measured by RIA using a rabbit antiserum (Sigma, St-Louis, MO) and plasma renin activity was determined with the Gamma Coat RIA kit (Baxter, Montreal, Quebec).

3. RESULTS

3.1. *Effects of both diets on plasma hormone levels and renin activity*

Table I shows the effects of high K^+ or low Na^+ intake for one week on plasma aldosterone and corticosterone levels and on plasma renin activity. It can be seen that both diets provoked significant increases in plasma aldosterone and corticosterone. Plasma renin activity was significantly increased by the low Na^+ intake, and sig-

nificantly decreased by the high K^+ intake. These results indicate that low Na^+ and high K^+ intakes properly altered the kidney and adrenal functions [12,21]. Our previous observations showed a significant induction of steroid hydroxylases and adrenodoxin mRNA by those alterations in the intake of sodium and potassium which stimulated aldosterone production [10–12]. The present investigations were initiated to determine whether such increased mRNA levels are due to an increased rate of transcription or if they simply reflect the metabolic fate of the mRNA transcripts.

3.2. *Estimation of the rate of transcription of steroidogenic enzymes by run-on assays*

Measurement of de novo transcription is achieved by the quantitation in isolated nuclei of nascent RNA chains which have been elongated in an in vitro transcription assay, a procedure which should primarily assess the rate at which initiation of transcription was occurring in vivo at the time that the animals were sacrificed.

As shown in Fig. 1 (left panel) and in Table II, an accumulation of nascent RNA in isolated nuclei from the zona glomerulosa in K^+ supplementation and Na^+ restriction was evident for $P450_{scc}$ (5- and 6-fold), 3β-HSD (3.6- and 2-fold) and $P450_{11\beta}$ (6.0- and 6.1-fold). $P450_{c21}$ transcription rates, in contrast, were only slightly affected by these variations in intake (1.4- and 1.2-fold) while that of adrenodoxin decreased (0.6-fold) with K^+ supplementation and remained to near basal level (1.3-fold) with Na^+ restriction.

Similar experiments were performed on nuclei isolated from the zona fasciculata reticularis of control and treated animals (Fig. 1, right panel). The results showed only slight differences in the transcription rates of $P450_{scc}$, $P450_{c21}$, $P450_{11\beta}$ and adrenodoxin with either K^+ supplementation or Na^+ restriction. On the other hand, these two regimens elicited increases in 3β-HSD initial transcripts by 4.5- in K^+-loaded and 1.7-fold in Na^+-restricted rats (Table I). It thus appears that such alterations in both K^+ and Na^+ intakes act at the level of the initiation of transcription of steroid hydroxylase genes solely in the zona glomerulosa and that their primary targets are $P450_{scc}$ and $P450_{11\beta}$. In contrast, the variations in intake of Na^+ and K^+ seemed to be without positive effect upon the de novo transcription of $P450_{c21}$ and adrenodoxin, whereas the rate of transcription of 3β-HSD was found to be elevated throughout the adrenal cortex.

Table I

Effects of high K^+ and low Na^+ diet on plasma aldosterone and corticosterone concentration and renin activity

Diet	Aldosterone (ng/dl)	Corticosterone (μg/dl)	Renin activity (ng A_I/ml/h)
Normal	15.7 ± 1.3 (9)	5.6 ± 1.1 (8)	8.5 ± 0.5 (11)
High K^+	50.2 ± 7.8* (8)	12.9 ± 1.3* (7)	4.8 ± 1.1* (8)
Low Na^+	107.7 ± 10.2* (9)	10.6 ± 0.6* (7)	16.6 ± 1.8* (7)

Aldosterone and corticosterone concentration and angiotensin I (A_I) production measured for renin activity were determined by RIA in plasma isolated from truncular blood. All values are expressed as mean ± S.E.M. of the number of experiments indicated in parentheses. Statistical significance of difference was calculated by Student's *t*-test where $*P < 0.01$ vs. untreated animals.

Table II

Relative transcription rates of adrenocortical steroidogenic genes in K^+ supplementation and Na^+ restriction

	Zona glomerulosa		Zona fasciculata reticularis	
	High K^+	Low Na^+	High K^+	Low Na^+
$P450_{acc}$	5.0, 4.9	5.7, 6.2	1.4	1.7, 1.8
3β-HSD	3.6	2.0	4.5	1.7
$P450_{c21}$	1.3, 1.4	1.1, 1.2	1.2	0.7, 0.6
$P450_{11\beta}$	5.9, 6.0	6.0, 6.1	1.3, 1.9	0.6, 0.5
Adrenodoxin	0.6, 0.5	1.2, 1.4	0.9, 0.8	0.9, 0.8
β-Actin	1.0, 1.0	1.3, 1.3	1.0, 1.0	1.0, 1.0

Nuclei were isolated from both adrenocortical zones of rats maintained 7 days on the various diets and in vivo initiated RNA chains were elongated in vitro in the presence of [^{32}P]UTP. Autoradiographic signals obtained by hybridization with specific cDNAs were then quantitated by densitometric analysis and expressed as the values of two independent experiments where the control (complete diet) is set arbitrarily at 1.0. Transcription rates for 3β-HSD was determined on a single experiment.

4. DISCUSSION

It has been well established that aldosterone biosynthesis is modulated by alterations in the intake of potassium and sodium. In this study, we present evidence that the transcriptional activation of genes encoding key steroidogenic enzymes is involved in the induction of aldosterone production following either supplementation of K^+ or restriction of the intake of Na^+. The transcription rates of the genes encoding $P450_{scc}$ and $P450_{11\beta}$, the two enzymes catalyzing the first and the final steps in aldosterone pathway, were the most highly induced, with values ranging between 5- and 6-fold in the zona glomerulosa of treated rats, followed by their electron donor adrenodoxin, as measured by run-on assays. The comparison of these responses with the corresponding mRNA levels in the zona glomerulosa of K^+-supplemented and Na^+-depleted rats, as observed in our previous reports [11,12], strongly suggests that the increases in $P450_{scc}$ and $P450_{11\beta}$ mRNA result from an increased initiation of the transcription of their genes. A similar response was also found when the content of nuclear precursor RNA was measured by Northern

blotting analysis (unpublished results). In the case of $P450_{11\beta}$, the magnitude of change in transcription was probably underestimated because the mouse $P450_{11\beta}$ cDNA probe we used recognized both forms of $P450_{11\beta}$ (11β-hydroxylase and aldosterone synthase) mRNAs. Indeed, by Northern blotting analyses with specific oligonucleotide probes that discriminate between the two forms of rat $P450_{11\beta}$, we found that only aldosterone synthase mRNA was increased by high K^+ or low Na^+ diet given to rats for one week [21]. Unfortunately it was not possible to use these oligonucleotide probes for run-on assays. Consequently, these results indicate that the sustained response to long-term variations in salt intake might not involve the stabilization of mRNA encoding either $P450_{scc}$ or $P450_{11\beta}$. It has been reported by Boggaram et al. [22] that such stabilization was occurring for $P450_{scc}$ mRNA when bovine adrenocortical cells were stimulated in vitro by corticotropin, although the transcriptional regulation of the expression of the $P450_{scc}$ gene was also contributing to the response [5]. The corticotropin effect on the other steroid hydroxylase enzymes was shown to be mediated at the transcriptional level of their gene expression, without any evidence for change in mRNA half-life [5,22].

Measurement of the initiation of the rate of transcription of the adrenodoxin gene indicated that while low Na^+ intake induced a slight increase (1.3-fold) in the zona glomerulosa, a high K^+ intake appeared to down-regulate the expression of the adrenodoxin gene by approximately 40%. This finding suggests that, according to the known stimulation of adrenodoxin mRNA levels [11,12], a stabilization of adrenodoxin mRNA rather than de novo transcription of its gene might be involved in the response to K^+ ions and, in part, in the response to Na^+ restriction. However, we cannot rule out a positive transcient increase in the adrenodoxin gene transcription during the treatment period which might also lead to a sustained increase in mRNA levels and, in such case, could not have been detected in our assay. Nevertheless, long-term variations in cation intakes seem to trigger various transcriptional events that differentially regulate the expression of the two mitochondrial P450 genes and that of their electron donor. The two regimens also enhanced the transcription of the 3β-HSD gene but to a lesser extent than that of $P450_{scc}$ and $P450_{11\beta}$. The coordinate control that seems to exist between the 3β-HSD and the $P450_{scc}$ and $P450_{11\beta}$ transcriptional activity in response to variations in the intake of K^+ and Na^+ may be related to a common mechanism in the zona glomerulosa that regulates both genes. Responsiveness to cAMP as well as to protein kinase C activators has been reported for these genes [2,4,23]. These transcriptional events seem also to occur in the zona fasciculata-reticularis for the expression of $P450_{scc}$, 3β-HSD and $P450_{11\beta}$ but the responses obtained were reduced as compared with the zona glomerulosa. It is well known, however, that fasciculata-reticularis cells are non-responsive to factors that modulate aldosterone secretion such as angiotensin-II and alterations in K^+ and Na^+ intake since no active renin-angiotensin system has been detected in these cells [24–26]. Furthermore, the levels of mRNA of the steroidogenic enzymes were found to be unchanged in the zona fasciculata-reticularis of K^+-supplemented and Na^+-restricted rats [11,12]. Thus, the inability of the zona fasciculata-reticularis to completely abolish the transcription of $P450_{scc}$, 3β-HSD and $P450_{11\beta}$ genes is suggestive of other mecanism(s) which may modulate the turnover of the mRNA encoding these enzymes, thus leading to a suppression of the steroidogenic response in the zona fasciculata-reticularis.

The rate of transcription of the gene encoding $P450_{c21}$ (present study), and also its mRNA level [12] was not affected by variations in cation intake. These results, along with the rather poor response of 3β-HSD expression to the regimens, compared with those of $P450_{scc}$ and $P450_{11\beta}$, clearly indicate that the initial rate-limiting step of steroidogenesis and the final steps leading to aldosterone are the two principal sites of regulation in the mediation of effects of K^+ and Na^+ in the zona glomerulosa. A number of studies have also documented that dietary sodium restriction and high potassium intake act through these two key steps by stimulating conversion of cholesterol to pregnenolone [7,8] and corticosterone and deoxycorticosterone to aldosterone [6–9]. We extended these findings by demonstrating that the stimulation of aldosterone production requires a de novo transcription of the $P450_{scc}$ and $P450_{11\beta}$ genes.

Little is known concerning the intracellular signalling pathway induced by variations in potassium and sodium intake. The demonstration that the zona glomerulosa contains all of the components of the renin-angiotensin system [25–28] led to a model postulating that a local generation of angiotensin II regulates mineralocorticoid production by mediating the effects of alterations in intake of K^+ and Na^+. In support of this model, inhibitors of angiotensin converting enzyme were shown to suppress the response of aldosterone to such regimens [31–33] by decreasing the level of expression of steroidogenic enzymes [21,29]. However, cAMP may also be involved in the response of the adrenal to potassium ion as suggested by Kojima et al. [30]. Hence, K^+ ions might follow a different intracellular route than the one involved in the Na^+ restriction effect. Moreover, recent studies have provided evidence for interactions between the cAMP and inositol phosphate transduction systems in glomerulosa cell responses to major stimuli [31–33]. The responsiveness to cAMP of a number of steroid hydroxylase genes has been shown to be mediated through cAMP-responsive sequences, such as in $P450_{scc}$ [34–37] and $P450_{11\beta}$ [38,39] genes. However, AP-1 sites and other putative regulatory elements have also been found in the promoter regions of genes encoding the two cytochromes [36,39,40] as well as adrenodoxin

[41,42], but a functional activity of these sites has not yet been associated with these regions. Thus, several DNA elements might be involved in the regulation of steroidogenic enzyme gene expression but it remains to be determined which of these, or perhaps other regions are required to mediate the effects of K^+ and Na^+ on steroidogenesis.

Acknowledgements: André Tremblay was the recipient of a studentship from the Canadian Heart Foundation, and from the Fonds de la Recherche en Santé du Québec. This work was supported by MRC Grant MT-4653. We thank the assistance of M. des Chênes, A. Lefebvre, and L. Ducharme, and advice of Dr D. Shapcott. The generous gifts of cDNAs by Drs M.R. Warterman, J.I. Mason, and K.L. Parker are also gratefully acknowledged

REFERENCES

[1] Waterman, M.R. and Simpson, E.R. (1989) Recent Prog. Horm. Res. 45, 533–566.
[2] Simpson, E.R. and Waterman, M.R. (1988) Annu. Rev. Physiol. 50, 427–440.
[3] Lachance, Y., Luu-The, V., Labrie, C., Simard, J., Dumont, M., de Launoit, Y., Guerin, S., Leblanc, G. and Labrie, F. (1990) J. Biol. Chem. 265, 20649–20675.
[4] Naville, D., Rainey, W.E. and Mason, J.I. (1991) Mol. Cell. Endocrinol. 75, 257–263.
[5] John, M.E. John, M.C., Boggaram, V., Simpson, E.R. and Waterman, M.R. (1986) Proc. Natl. Acad. Sci. USA 83, 4715–4719.
[6] Lehoux, J.G., Sandor, T., Henderson, I.W. and Jones, I.C. (1974) Can. J. Biochem. 52, 1–6.
[7] Aguilera, G. and Catt, K.J. (1979) Endocrinology 104, 1046–1052.
[8] Kramer, R.E., Gallant, S., and Brownie, A.C. (1979) J. Biol. Chem. 254, 3953–3958.
[9] Muller, J. (1980) J. Steroid Biochem. 13, 253–257.
[10] Tremblay, A. and Lehoux, J.G. (1989) 71st. Annual Meeting of the Endocrine Society. June 21–24, 1989, Seattle, WA. Abstract 1459.
[11] Tremblay, A. and Lehoux, J.G. (1989) J. Steroid Biochem. 34, 385–390.
[12] Tremblay, A., Waterman, M.R., Parker, K.L. and Lehoux, J.G. (1991) J. Biol. Chem. 266, 2245–2251.
[13] Lamers, W.H., Hanson, R.W. and Meisner, H.M. (1982) Proc. Natl. Acad. Sci. USA 79, 5137–5141.
[14] Schibler, U., Hagenbüchle, O., Wellaver, P.K. and Pittet, A.C. (1983) Cell 33, 501–508.
[15] Varguese, S. Deaven, L.L., Yu-Chu, H., Gill, R.K., Iacopino, A. and Christakos, S. (1989) Mol. Endocrinol. 3, 495–502.
[16] Zuber, M.X., Mason, J.I., Simpson, E.R. and Waterman, M.R. (1988) Proc. Natl. Acad. Sci. USA 85, 699–703.
[17] Okamura, T., John, M.E., Zuber, M.X., Simpson, E.R. and Waterman, M.R. (1985) Proc. Natl. Acad. Sci. USA 82, 5705–5709.
[18] Lorence, M.C., Murry, B.A., Trant, J.M. and Mason, J.I. (1990) Endocrinology 126, 2493–2498.
[19] John, M.E., Okamura, T., Dee, A., Adler, B., John, M.C., White, P.C., Simpson, E.R. and Waterman, M.R. (1986) Biochemistry 25, 2846–2853.
[20] Wong, M., Rice, D.A., Parker, K.L. and Schimmer, B.P. (1989) J. Biol. Chem. 264, 12867–12871.
[21] Tremblay, A., Parker, K.L. and Lehoux, J.G. (1992) Endocrinology 130, 3152–3158.
[22] Boggaram, V., Zuber, M.X. and Waterman, M.R. (1984) Arch. Biochem. Biophys. 231, 518–523.
[23] Rainey, W.E., Naville, D. and Mason, J.I. (1991) Endocrine Res. 17, 281–296.
[24] Braley, L.M. and Williams, G.H. (1977) Am. J. Physiol. 233, E402-E406.
[25] Doi, Y., Atarashi, K., Franco-Saenz, R. and Mulrow, P.J. (1984) Hypertension 6, I124-I129.
[26] Dzau, V.J., Ellison, K.E., Brody, T., Ingelfinger, J. and Pratt, R.E. (1987) Endocrinology 120, 2334–2338.
[27] Nakamaru, M., Misono, K.S., Naruse, M., Workman, R.J. and Inagami, T. (1985) Endocrinology 117, 1772–1778.
[28] Oda, H., Lotshaw, D.P., Franco-Saenz, R. and Mulrow, P.J. (1991) Proc. Soc. Exp. Biol. Med. 196, 175–177.
[29] Tremblay, A. and Lehoux, J.G. (1992) J. Steroid Biochem. 41, 799–808.
[30] Kojima, I., Kojima, K. and Rasmussen, H. (1985) Biochem. J. 228. 69–76.
[31] Brami, B., Vilgrain, I. and Chambaz, E.M. (1987) Mol. Cell Endocrinol. 50, 131–137.
[32] Baukal, A.J., Hunyady, L., Balla, T., Ely, J.A. and Call, K.J. (1990) Mol. Endocrinol. 4, 1712–1719.
[33] Lehoux, J.G., Grondin, F., Pacuraru, J.P. and Yachaoui, Y. (1991) Mol. Cell Endocrinol. 78, 97–106.
[34] Oonk, R.B., Parker, K.L., Gibson, J.L. and Richards, J.O. (1990) J. Biol. Chem. 265, 22392–22401.
[35] Inoue, H., Igashi, Y., Morohashi, K.I. and Fujii-Kuriyama, Y. (1988) Eur. J. Biochem. 171, 435–440.
[36] Rice, D.A., Kirkman, M.S., Aitken, L.D., Mouw, A.R., Schimmer, B.P. and Parker, K.L. (1990) J. Biol. Chem. 265, 11713–11720.
[37] Ahlgren, R., Simpson, E.R., Waterman, M.R. and Lund, J. (1990) J. Biol. Chem. 265, 3313–3319.
[38] Kirita, S., Hashimoto, T., Kitajima, M., Honda, S., Morohashi, K. and Omura, T. (1990) J. Biochem. 108, 1030–1041.
[39] Mouw, A.R., Rice, D.A., Meade, J.C., Chua, S.C., White, P.C., Schimmer, B.P. and Parker, K.L. (1989) J. Biol. Chem. 264, 1305–1309.
[40] Honda, S.I., Morohashi, K.I. and Omura, T. (1990) J. Biochem. 108, 1042–1049.
[41] Chang, C.Y., Wu DA Lai, C.C., Miller, W.L. and Chung, B.C. (1988) DNA 7, 609–615.
[42] Sagara, Y., Sawae, H., Kimura, A., Nanako, Y.S., Morohashi, K.I., Miyoshi, K.I. and Horiuchi, T. (1990) J. Biochem. 107, 77–83.