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AGE RELATED CHANGES IN CARDIAC AND AORTIC PHOSPHODIESTERASE ACTIVITIES IN NORMOTENSIVE AND HYPERTENSIVE RATS

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Abstract—The activities of cAMP* and cGMP phosphodiesterase were studied in the aorta (freed of adventitia layer) and in the heart (ventricles) of normotensive and mineralocorticoid hypertensive rats of 8 or 16 weeks of age. The enzyme activities were determined at low (1 μM) and high (100 μM) substrate concentrations. The changes in activity were compared to the changes in organ weight, protein and DNA content. The increase in organ weight that occurred with both age and hypertensive treatment corresponded mostly to a marked elevation in protein content in the aorta, but not in the heart, where the DNA content increased without any significant variation in protein content. In both tissues, cGMP phosphodiesterase activity measured at low substrate concentration was sensitive to endogenous Ca^{2+}-dependent activation and markedly increased with age. This increase was proportionally larger than the variations in DNA content of the tissues, but lower than those of total protein in the aorta. It could not be ascribed to an increase in the activator content of the tissues, which was in excess. By contrast, cGMP phosphodiesterase activity measured at high substrate concentration and cAMP phosphodiesterase activity, measured at either substrate concentration, were not sensitive to the Ca^{2+}-dependent activation and did not undergo large changes with age except for a significant decrease in cAMP phosphodiesterase activity at high substrate concentration per mg heart cytosol protein. No relationship could be found between the elevation of blood pressure, due to age or to the influence of the mineralocorticoid treatment, and phosphodiesterase activities, which varied in a similar manner in control and hypertensive rats. The results are consistent with the view that a cGMP phosphodiesterase, which is sensitive to Ca^{2+}-dependent endogenous activation, increases in aorta and heart cells with the age of the rat.

The roles of cAMP and cGMP in heart and in arterial smooth muscle function [1–4] and their possible involvement in the pathogenesis of hypertension [5–15] have been investigated in several laboratories. Alterations of cyclic nucleotide metabolism have been reported in hypertensive patients and in rats [10, 12], but phosphodiesterase activities were found to be decreased [8], unchanged [9, 11, 13], or increased [5–7, 15] in cardiovascular tissues of mature hypertensive rats.

We recently reported [16] that the Ca^{2+}-dependent phosphodiesterase activator content increases with age in the aorta during maturation of the rat, and is less in mineralocorticoid–hypertensive rats than in control rats. We wondered whether cAMP and cGMP phosphodiesterase activities in the aorta and the heart ventricles of control rats also vary with age and are also altered during the onset of mineralocorticoid-hypertension. Unlike previous studies, we related the phosphodiesterase activities to the protein and also to the DNA content of the tissues and to the whole organ, in order to take into account the possible hypertrophy of the tissues, which seems closely related to hypertension [17–23].

* Abbreviations used: cAMP, 3′,5′-adenosine monophosphate; cGMP, 3′,5′-guanosine monophosphate; EGTA, ethylene glycol bis amino-2-ethylether *N,N*′tetraacetate; DOCA, desoxycorticosterone acetate.

MATERIALS AND METHODS

Chemicals

[^{3}H]cAMP (30–50 Ci/mmole), [^{3}H]cGMP (5–10 Ci/mmole), and [^{14}H]guanosine (>400 mCi/mmole) were obtained from New England Nuclear Corp. [^{14}C]Adenosine (500–600 mCi/mmole) was furnished by Radiochemical Centre Amersham. Snake venom (*Ophiaphagus hannah*), bovine serum albumin (fraction V powder) and cyclic nucleotides were purchased from Sigma Chemical Co. QAE-Sephadex A25 was delivered by Pharmacia Sweden. Desoxycorticosterone acetate pellets (25 mg) were prepared by the Laboratoire de Pharmacie Galénique of the Faculty of Strasbourg. Other chemicals were analytical grade. [^{3}H]Cyclic nucleotides were purified by thin layer chromatography [24] and stored at −20° in 50% ethanol. Water from milli Q_2 system (Millipore, 18 Mohm/cm at 25°) was used throughout.

Mineralocorticoid hypertension

Six-week-old male Sprague–Dawley rats (Iffa-Credo, St Germain sur l'Arbresle, France) were randomly distributed into two groups: rats in one group were sham-operated and drank tap water *ad lib.* (control), and those in the other group had one 25 mg dexosycorticosterone acetate (DOCA) pellet implanted subcutaneously and drank 0.9% NaCl solution *ad lib.* (DOCA–NaCl). Each group was randomly divided into subgroups in which the rats were

killed at various ages and times after the onset of the mineralocorticoid treatment.

The systolic blood pressure of 20 rats randomly chosen from each experimental group was measured, using a plethysmographic tail-cuff technique (PNS Racia, Bordeaux, France).

Preparation of tissue samples

The rats were killed by decapitation. The whole aorta (from the left coronary artery to the iliac artery) and the heart were rapidly removed and cleaned of blood. The aorta was freed from adventitia and the ventricles freed from auricles. Pools of five aorta or ventricles were homogenized with a glass–glass Potter homogenizer in 4 v/w of buffer containing 40 mM Tris–HCl (pH 8.0), 0.33 M sucrose, 1 μM EDTA. The homogenates were centrifuged at 105,000 *g* for 1 hr. The supernatants (soluble fractions) and the pellets (particulate fractions) were then dialysed against 20 vol of buffer (40 mM Tris–HCl (pH 8.0), 5 mM $MgSO_4$, 3.75 mM 2-mercaptoethanol) for 48 hr with 3 changes. The soluble and particulate preparations were distributed in small volumes and stored at −80°.

Cyclic nucleotide phosphodiesterase assay

Cyclic nucleotide phosphodiesterase was assayed using a modified Thompson and Appleman [25] assay: QAE-Sephadex A25 [26] was used instead of Biorad AG 1 × 2 slurry to separate the products from the residual substrate, and the recovery of adenosine or guanosine was estimated using [^{14}C]adenosine or guanosine as tracer. The anion exchange resin QAE-Sephadex A25 was chosen to improve the recovery of the nucleotides without decreasing the binding of the residual cyclic nucleotides to the resin. Unless otherwise stated, the experimental conditions were those described by Thompson and Appleman [25]. The reaction mixture contained approximately 30,000 c.p.m. of purified [^{3}H]-labelled cyclic nucleotide, a total concentration of 1 or 100 μM cyclic nucleotide, and an appropriate amount of the enzyme, and were incubated for various times. The reaction was stopped by immersing the test tubes in boiling water for 1 min. Then 120 mg of snake venom and 10,000 c.p.m. of [^{14}C]adenosine or guanosine were added to each tube. After 10 min of incubation at 37°, 1 ml of QAE-Sephadex A25 slurry, prepared according according to Schultz [26] was added. The mixture was stirred occasionally, centrifuged, and then 0.5 ml of the supernatant was counted in the scintillation fluid. The results were corrected for the yield of adenosine or guanosine recovered in each sample, and expressed as nmoles of cyclic nucleotides hydrolysed per minute.

Blanks were run under identical conditions, with buffer replacing the enzyme, since in preliminary experiments we verified that blanks run with boiled enzyme were identical. Phosphodiesterase concentrations used in the assay were adjusted to give linear reaction rates under our conditions. No more than 15 per cent of the substrate was hydrolysed during the enzyme assay. We also verified that separation of the final products by thin layer chromatography [24] gives the same percentage of hydrolysis; this shows that deaminases and other enzymes metabolizing the products of the reaction [27] did not interfere in the experimental conditions.

Other methods

The amount of protein in each fraction was determined by the method of Lowry *et al.* [28] using bovine serum albumin as standard.

DNA contents were measured in particulate fractions according to Hill and Whatley [29, 30].

Calcium dependent activator was prepared according to Cheung [31] using rat brain.

Expression of the data

Phosphodiesterase activities found in particulate and soluble fractions were related to the protein content of the corresponding fraction, to the whole organ, or its DNA content. Unless otherwise stated, Student's *t*-test was used for the statistical analysis of the data.

RESULTS

Changes in blood pressure, protein and DNA content

As expected, blood pressure increased with age and was higher in DOCA–NaCl treated rats than in control rats (Table 1). The data contained in Table 2 show that the wet weight of aortas and heart ventricles increased significantly with age in both control and DOCA–NaCl rats. After 2 weeks of mineralocorticoid treatment the wet weight of the organs of treated rats was significantly elevated over that of controls. This increase was more pronounced in heart ventricles than in aortas. After 10 weeks of treatment, there was no significant difference in organ weight between control and DOCA–NaCl aortas and heart ventricles.

In the aorta, the changes in protein content paralleled the changes in wet weight. However, the

Table 1. Elevation with age of blood pressure in control and DOCA–NaCl rats

Age (weeks)	Time after onset of the treatment (weeks)	Blood pressure mmHg	
		Control	DOCA–NaCl
8	2	121 ± 1.3	149 ± 1.5^{+++}
16	10	153 ± 2.3$^{●●●}$	192 ± 4.6^{+++} $^{●●●}$

Values are the means of 20 individual determinations done in duplicate ± S.E.M. $^{+++}$ $P < 0.001$. Student's *t*-test for comparison between DOCA–NaCl and control rats of the same age. $^{●●●}$ $P < 0.001$ for comparison between 8- and 16-week-old rats of the same experimental group.

Table 2. Changes in weight, protein and DNA content of aorta and heart ventricles in control and mineralocorticoid hypertensive rats

Organ	Age (weeks)	mg wet weight Control	mg wet weight DOCA–NaCl	mg protein . organ^{-1} Control	mg protein . organ^{-1} DOCA–NaCl	μg DNA organ^{-1} Control	μg DNA organ^{-1} DOCA–NaCl
Aorta	8	73 ± 5	88 ± 3^{+}	1.61 ± 0.14	2.36 ± 0.19^{++}	55 ± 8	64 ± 3
	16	109 ± 3$^{●●●}$	120 ± 5$^{●●●}$	3.31 ± 0.13$^{●●●}$	3.37 ± 0.18$^{●●}$	60 ± 3	70 ± 5
Heart ventricles	8	727 ± 22	816 ± 18^{++}	88.8 ± 5.6	98.3 ± 3.5	577 ± 69	708 ± 89
	16	994 ± 9$^{●●●}$	1089 ± 55$^{●●●}$	82.0 ± 5.4	86.8 ± 5.0	666 ± 31	832 ± 58^{+}

Results are the means ± S.E.M. of 8 experiments in duplicate.
+ $P < 0.05$, ++ $P < 0.01$, Student's t-test for comparison between treated and control rats of the same age.
●● $P < 0.01$, ●●● $P < 0.001$ versus 8-week-old rats receiving the same treatment.

DNA content of the aorta varied much less than its protein content. A two-way layout variance analysis was applied to the values of the DNA content: in the aorta, it shows that there was no significant interference between age and treatment, no significant influence of age, but a significant influence of treatment on DNA contents ($P < 0.01$).

By contrast with what was found in the aorta, Table 2 also shows that the protein content of the ventricles did not parallel their wet weight, since it did not vary significantly with age or under the influence of the mineralocorticoid treatment. However, the DNA content increased slightly with age and treatment. The two-way layout variance analysis of heart DNA content shows an interaction between age and treatment ($P < 0.05$), a significant effect of age ($P < 0.05$), and a more significant influence of treatment ($P < 0.01$) on DNA change.

A great difference in the variation of protein content could thus be seen between aorta and heart ventricles. By contrast, wet weight and DNA content changed more or less in the same way in both organs.

Changes in cyclic nucleotide phosphodiesterase activities in whole organs

In these experiments, cyclic nucleotides phosphodiesterases were determined at 1 μM and 100 μM substrate concentrations. The lower concentration (1 μM) is within the limits estimated physiological concentrations of cyclic nucleotides in tissues. It also corresponds to the low apparent K_m value of phosphodiesterase which was characterized by the existence of two apparent K_ms as reported by several authors [5, 25] and as determined in our previous experiments [32]. The 100 μM substrate concentration, ten fold greater than the high apparent K_m, gives the enzyme activity of the high K_m phosphodiesterase.

Figure 1 demonstrates cyclic nucleotide phosphodiesterase activities in soluble and particulate fractions of rat aorta.

No significant variation of cAMP hydrolysis could be found at 1 μM substrate concentration. At 100 μM, cyclic AMP phosphodiesterase activity was

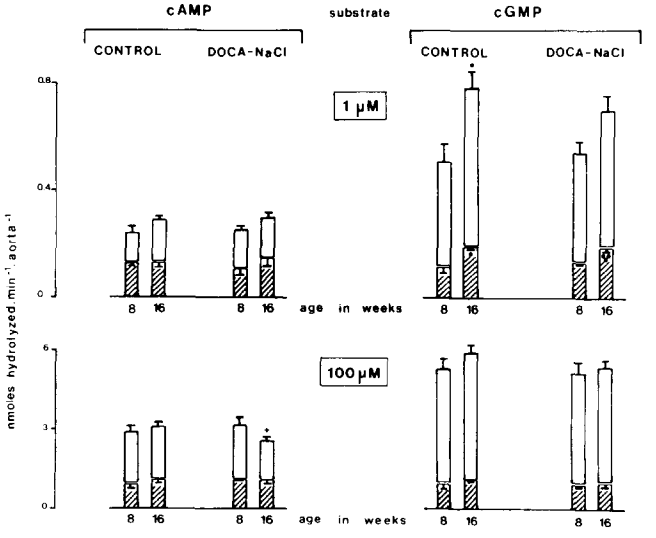

Fig. 1. Changes with age of cyclic nucleotide phosphodiesterase activity in particulate ▨ and soluble □ fractions of the aorta of control and mineralocorticoid hypertensive rats. Results are the means ± S.E.M. of 8 experiments performed in duplicate. ● $P < 0.05$, ●●● $P < 0.001$. Student's t-test between 8- and 16-week-old rats receiving the same treatment; + $P < 0.05$. Student's t-test for comparison between treated and control rats of the same age.

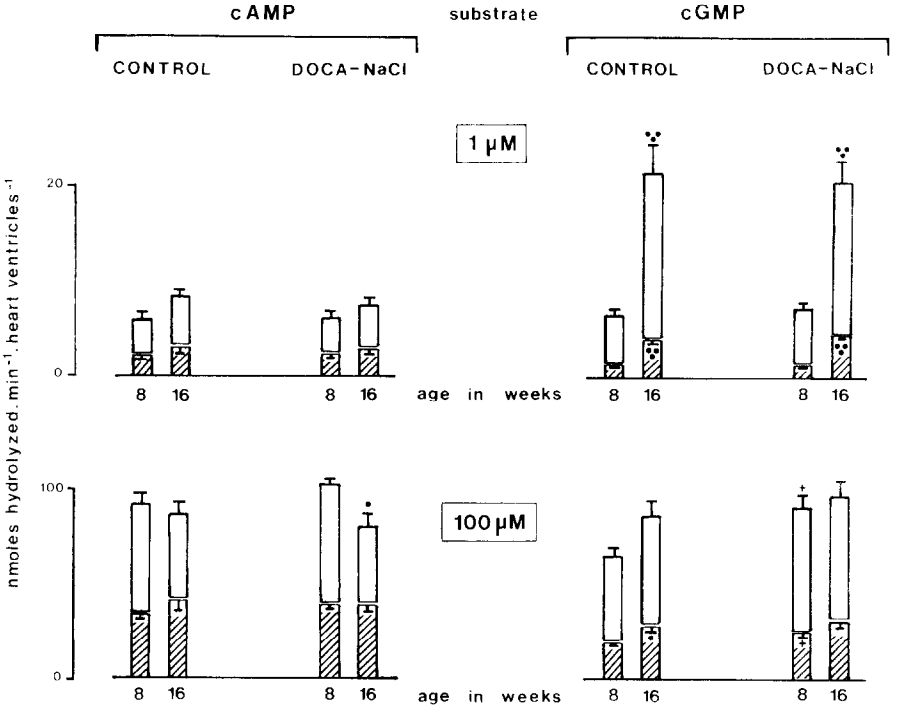

Fig. 2. Changes with age of cyclic nucleotide phosphodiesterase activity in particulate ▨ and soluble □ fractions of the ventricles of control and mineralocorticoid hypertensive rats. Results are the means ± S.E.M. of 8 experiments performed in duplicate. ● $P < 0.05$, ●●● $P < 0.001$. Student's *t*-test between 8- and 16-week-old rats receiving the same treatment; + $P < 0.05$, for comparison between treated and control rats of the same age.

significantly lower in soluble fractions of 16-week-old treated rats than in the controls of the same age. Under the same conditions, cyclic GMP phosphodiesterase activities, determined at 1 μM, increased markedly with age in both particulate and soluble fractions of control and treated aortas. In these conditions, the hydrolysis of cyclic GMP in the soluble fractions was identical in the aortas of 8-week-old mineralocorticoid rats as in those of controls, but was less in the aortas of treated rats than in those of controls 16 weeks of age. Thus the increase with age of cytosolic phosphodiesterase activity was less in mineralocorticoid treated rats than in the controls. In the particulate fractions cyclic GMP phosphodiesterase activities increase with age in both DOCA–NaCl and control rats. At 100 μM cGMP in aorta phosphodiesterase activity did not vary with age or with treatment.

Figure 2 represents changes of cyclic nucleotide phosphodiesterase activity in rat heart ventricles which paralleled those reported above for rat aorta. No variation of cyclic AMP hydrolysis could be seen at 1 μM in rat ventricles, while there was a marked increase with age in cyclic GMP hydrolysis in both control and DOCA–NaCl rats. The increase with age was less in mineralocorticoid rats. At 100 μM,

Table 3. Phosphodiesterase activities in the soluble fraction of rat aorta

Substrate		1 μM cAMP	100 μM cAMP	1 μM cGMP	100 μM cGMP
Treatment	Age (weeks)	(pmoles . mg protein $^{-1}$. min $^{-1}$)			
Control	8	231 ± 26	3486 ± 376	721 ± 100	8134 ± 231
	16	147 ± 4●●	1847 ± 223●●●	646 ± 61	4444 ± 376●●●
DOCA–NaCl	8	177 ± 30+	2345 ± 322++	558 ± 149	5767 ± 738++
	16	165 ± 20	1509 ± 155●●●	596 ± 35	3866 ± 252●●
		(nmoles . mg DNA $^{-1}$. min $^{-1}$)			
Control	8	2.1 ± 0.1	31.8 ± 2.5	6.6 ± 0.7	74.0 ± 3.4
	16	2.8 ± 0.2●	34.7 ± 2.3	12.3 ± 1.3●●	85.1 ± 6.7
DOCA–NaCl	8	1.7 ± 0.2	22.7 ± 2.0+	5.5 ± 1.1	56.4 ± 6.1+
	16	2.9 ± 0.3●	26.3 ± 1.7+	10.4 ± 0.7●	67.2 ± 3.0

In each experimental group the results are the means of 4 experiments in duplicate ± S.E.M. + $P < 0.05$, ++ $P < 0.001$, +++ $P < 0.001$. Student's *t*-test for comparison of DOCA–NaCl rats and controls of the same age. Comparison of 16-week-old rats versus 8-week-old rats receiving the same treatment: ● $P < 0.05$, ●● $P < 0.01$, ●●● $P < 0.001$.

cyclic GMP phosphodiesterase activity increased in soluble and particulate fractions of 8-week-old-DOCA–NaCl rats as compared to controls of the same age.

Changes in phosphodiesterase activities related to protein or DNA content

In Tables 3 and 4 the enzyme activities found in soluble fractions were related to the protein content of the preparation or to the DNA content of the tissue.

In the aorta (Table 3) when related to protein contents, both cAMP and cGMP hydrolysing activities decreased with age and the hypertensive treatment. These changes were large and highly significant at high substrate concentrations, but were much less and generally not significant at low substrate concentrations. There was no significant change in aorta phosphodiesterase activities when related to DNA content, except for a moderate but significant increase with age in cAMP hydrolysis at 1 μM in control and treated aortas and a decrease with treatment in cAMP hydrolysis at 100 μM in 16-week-old-hypertensive rats.

Contrary to what was observed in the aorta, the phosphodiesterase activities found in the soluble fraction of heart varied identically whatever the mode of expression of the data (Table 4). The hydrolysis of cAMP did not vary significantly at 1 μM but decreased with age at 100 μM. By contrast, the hydrolysis of cGMP increased with age at low substrate concentration, but did not vary at high substrate concentration. There was no significant influence of the hypertensive treatment.

Influence of Ca^{2+}-dependent activation

We wondered whether the increase with age of phosphodiesterase activity at low substrate concentration that we report here could be ascribed to the increase in activator content that we reported previously [16]. In order to investigate this question, we studied firstly the influence of the Ca^{2+} chelator EGTA, and secondly the effect of Ca^{2+} and activator in excess on soluble and particulate phosphodiesterase activities of the aortas and the hearts of control and hypertensive rats of different ages.

The data illustrated in Fig. 3 show no influence of EGTA or of the activator preparation on the hydrolysis of cAMP by the aorta soluble fraction at 1 μM substrate, whatever the age of the rat or the treatment. By contrast, the hydrolysis of cGMP was markedly inhibited in the presence of EGTA, but it was not modified by the addition of an excess of Ca^{2+} and activator. Thus, cGMP phosphodiesterase

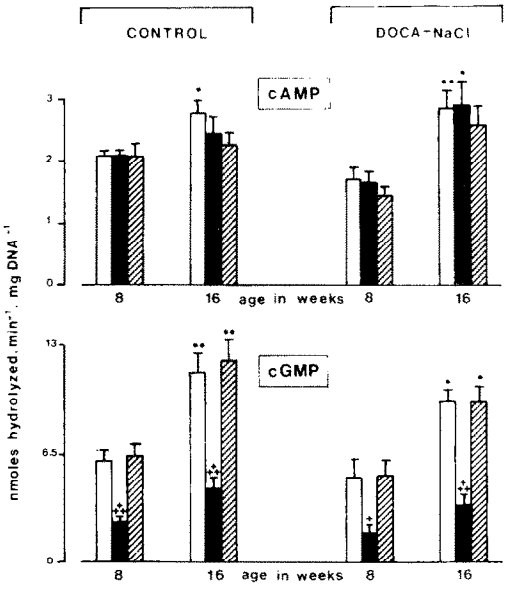

Fig. 3. Influence of Ca^{2+}-dependent activation on cyclic nucleotide phosphodiesterase activity at 1 μM substrate concentration in the aorta from control and mineralocorticoid hypertensive rats. □ 100.000 *g* supernatant + buffer; ■ *idem.* + EGTA 10 μM; ▨ *idem.* + $CaCl_2$ 20 μM + activator 97 μg in each test tube. Results are means of 4 experiments in duplicate ± S.E.M. • $P < 0.05$, •• $P < 0.01$. Student's *t*-test between 8- and 16-week-old rats receiving the same treatment; +$P < 0.05$, ++$P < 0.01$, +++$P < 0.001$. Student's *t*-test for paired values for comparison between □ and ■.

Table 4. Phosphodiesterase activity in the soluble fraction of heart ventricle

Substrate		1 μM cAMP	100 μM cAMP	1 μM cGMP	100 μM cGMP
Treatment	Age (weeks)	(pmoles . mg protein^{-1} . min^{-1})			
Control	8	189 ± 17	2574 ± 70	228 ± 7	2184 ± 210
	16	155 ± 7	1335 ± 119•••	511 ± 56•••	1699 ± 159
DOCA–NaCl	8	154 ± 18	2402 ± 97	211 ± 15	2588 ± 280
	16	138 ± 12	1253 ± 146•••	460 ± 52•••	1934 ± 157
		(nmoles . mg DNA^{-1} . min^{-1})			
Control	8	8.2 ± 1.3	108.6 ± 12.1	9.6 ± 1.1	95.2 ± 16.5
	16	8.2 ± 1.0	72.3 ± 11.9	28.1 ± 6.2••	92.5 ± 15.8
DOCA–NaCl	8	6.7 ± 1.4	94.8 ± 7.6	8.9 ± 1.5	111.5 ± 21.5
	16	5.4 ± 0.7	53.6 ± 7.6••	19.9 ± 3.2••	82.4 ± 9.8

In each experimental group the results are the mean of 8 experiments in duplicate ± S.E.M., •• $P < 0.01$, ••• $P < 0.001$. Student's *t*-test for comparison of 8- and 16-week-old rats receiving the same treatment.

activity was sensitive to Ca^{2+}-dependent activation and was already maximally activated by Ca^{2+} and activator present in the preparation. The age related moderate increase in cAMP phosphodiesterase activity and the more marked increase in cGMP phosphodiesterase activity that we observed at low substrate concentration with respect to DNA occurred in the presence of EGTA as well as in the presence of an excess Ca^{2+} and activator, in the aortas of both control and treated rats. In the case of cGMP, the increase in hydrolysis with age was approximately twofold whether in the absence or in presence of EGTA. The decrease in enzyme activity produced by EGTA (or, vice versa, its degree of activation by the endogenous activator) was practically identical (two- to three-fold) whatever the age of the rat.

The results obtained on the particulate fraction of the aorta and on the particulate and soluble fractions of the heart ventricles (data not shown) were similar to those shown above. In all cases cGMP but not cAMP hydrolysis was sensitive to EGTA and was maximally activated by Ca^{2+} and the endogenous activator contained in the preparations, so that the limiting factor was phosphodiesterase; in all cases cGMP phosphodiesterase display an identical Ca^{2+}-dependency, whatever the age of donors.

DISCUSSION

The choice of a reference to normalize the data is critical in the type of study reported here. The growth of the tissues with age and their hypertrophy related to increased blood pressure may be ascribed to three distinct phenomena: increase in the number of cells, reflected by an increase in DNA; increase in cell volume; increase in extracellular material. In accordance with other reports where rats of the same age were used [21–23], we found that the increase in aorta weight was paralleled by a corresponding increase in protein but not in DNA content. Age and hypertension both are known to cause increases in extracellular protein, primarily elastin and collagen [18, 20–23]. Thus, relating phosphodiesterase activities to aorta protein suggests that the enzyme activities decreased in this organ with age and hypertension (Table 3). By contrast the increase in heart weight with age and hypertension was not followed by a significant increase in protein content but corresponded to a moderate increase in DNA content. It may be misleading, however, to relate the data to DNA only, since the elevation of blood pressure may cause an elevation in cell volume [33]. This is why, in this study, we compared the changes in phosphodiesterase activities in the organs with the changes in their weight, protein and DNA content. The need for more than one reference may explain the conflicting conclusions which were previously reported regarding phosphodiesterase activities in cardiovascular tissues of hypertension rats [5–13, 15].

In spite of the weight increase of the organs with age and treatment, the changes of the corresponding cAMP phosphodiesterase activities measured at both substrate concentrations and of cGMP phosphodiesterase activities measured at high substrate concentration were either slight or non-significant in both aortas and hearts. Nevertheless there was a highly significant decrease with age of cAMP phosphodiesterase activities at high substrate concentration when activities were related to heart cytosolic protein (Table 4). However, since enzyme activities related to protein content take into account the synthesis of extracellular proteins, one might have expected that activities related to DNA could have led to quite different results. But due to the variability of individual data, differences in DNA contents with age and treatment were either non-significant or at the limit of significance. Thus it seems that the cAMP phosphodiesterase activity cellular contents remain constant with age and treatment.

In contrast, we found a significant increase with age in cGMP phosphodiesterase activity at low substrate concentration, in both control and hypertensive rats. This increase was greater in the ventricles than in the aorta. In both organs, however, it was proportionally larger than the increase in DNA content, so that the data related to DNA demonstrated significant variations per cell (Tables 3 and 4). At least in the heart, this increase in enzyme activity was not merely the result of a possible increase in cell volume, since it was also found per mg cytosolic protein while the other enzyme activities related to the same reference did not vary significantly or decreased with age.

The experiments in which EGTA or Ca^{2+} and endogenous activator in excess (Fig. 3) were added to the assay medium show that cGMP phosphodiesterase activity (but apparently not the other enzyme activities) was sensitive to Ca^{2+}-dependent activation. They also demonstrate that, under our conditions, the enzyme was maximally activated, probably because Ca^{2+} ions released during the homogenization procedure activated the endogenous protein regulator which was in excess in the cells studied as in other cells (see review 34). In the presence of EGTA, the "basal" activity also increased with age in the same proportion (approximately two-fold) as in the absence of EGTA. These findings show that the change with age in cGMP hydrolysis reported here cannot be ascribed to the increase in endogenous activator content reported previously [32], but is due to the enzyme itself. Such an increase in phosphodiesterase activity may result from a modification in kinetic properties or from an increase in concentration of a low K_m activable cGMP phosphodiesterase. A phosphodiesterase form which uses cGMP as preferred substrate in the micromolar concentration range and which is activated by the Ca^{2+}-dependent protein regulator has been found in many tissues including heart and arteries [11, 34–36]. Our observation that the degree of maximal activation in presence of 1 μM cGMP was not different (two- to three-fold) in the tissues of 8- and 16-week-old rats is consistent with the view that the age-related increase in cGMP hydrolysis resulted from an increase in activable cGMP phosphodiesterase.

In view of previous reports on possible involvement of cyclic nucleotides in hypertension [5–15], we investigated cyclic nucleotide phosphodiesterase activities during the onset of DOCA-salt elicited

hypertension. The differences between the data found in the tissues from control and hypertensive rats were slight, if any, during the onset phase of hypertension (after 2 weeks of treatment) as well as during the sustained or "metacorticoid" phase (10 weeks after implantation of DOCA pellets). Thus, the increase with age in cGMP phosphodiesterase activity that occurs in heart and aorta does not seem linked to the elevation of blood pressure, since the enzyme activity did not increase in hypertensive rats. The possible physiological significance of the changes in cyclic nucleotide phosphodiesterase activities reported here deserves further investigation.

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