



Aldosterone as a target in congestive heart failure

Sanjay Rajagopalan*, Bertram Pitt

University of Michigan School of Medicine, Ann Arbor, MI, USA

The significance of aldosterone in systolic heart failure

Aldosterone is a major prognostic determinant in heart failure [1–3]. Aldosterone has been traditionally viewed as a hormonal mediator downstream of angiotensin II, that plays an important role in the pathophysiology in heart failure by way of its effects on sodium retention and potassium loss. Emerging data, however, suggests that aldosterone synthesis from the adrenal gland, the heart, and the vasculature, independent of angiotensin II, may exert pleiotropic effects on these organs [4]. In this regard, angiotensin II reduction strategies, such as through the usage of an ACE inhibitor, an angiotensin receptor blocking agent (ARB), or their combination are insufficient to block aldosterone production; this phenomenon is termed “aldosterone escape” and has been recognized for more than 2 decades [5,6]. Recent results from the Randomized Evaluation of Strategies for Left Ventricular Dysfunction Study (RESOLVD) trial in patients with heart failure caused by systolic left ventricular dysfunction confirmed this effect. In this trial, angiotensin converting enzyme inhibitor (ACEI), ARB, or the combination (enalapril, 10 mg twice a day and candesartan, 8 mg every day) was insufficient to suppress aldosterone levels [7]. Furthermore in this trial, β -blocking agents, despite their known effects on plasma renin activity, failed to suppress aldosterone levels. Thus, standard therapy for patients with heart failure including an ACE inhibitor, an angiotensin receptor blocking agent, their combination, or a β -adrenergic receptor blocking agent do not suppress aldosterone production in patients with heart failure. This is consistent with observations that stimuli other than angiotensin II, such as potassium,

* Corresponding author. Division of Cardiology, L3119, Women’s Hospital, 1500 East Medical Center Drive, Ann Arbor, MI 48109-0273.

E-mail address: srajagop@umich.edu (S. Rajagopalan).

corticotropin, nitric oxide, endothelin, oxygen-free radicals, and adrenomedullin have all been shown to modulate aldosterone production [8–11].

Insights from the Randomized Aldactone Evaluation Study

The Randomized Aldactone Evaluation Study (RALES) trial is proof of principle that aldosterone is of pathophysiologic importance in patients with heart failure that is caused by systolic left ventricular dysfunction [12]. In this study more than 1600 patients with evidence of systolic left ventricular dysfunction and a history of New York Heart Association (NYHA) class III/IV heart failure within the past 6 months who were treated with standard therapy including an ACE inhibitor, loop diuretic, \pm digoxin, and a β -adrenergic receptor blocking agent, were randomized to the aldosterone receptor antagonist spironolactone 25 mg daily or placebo. Patients with a serum creatinine that was greater than or equal to 2.5 mg dL or a serum potassium that was greater than or equal to 5.0 meq/L were excluded. Serum potassium was closely monitored; investigators decreased the dose of study medication to one tablet every other day if there was evidence of hyperkalemia (potassium \geq 5.5 meq/L). If after 8 weeks there was no evidence of hyperkalemia, however, the study medication could be increased to 50 mg daily. Most of the patients who were randomized to spironolactone remained on a dose of 25 mg daily, whereas approximately one sixth decreased their dose to 25 mg every other day (12.5 mg daily) and another one sixth increased their dose to 50 mg daily. The mean dose of spironolactone was 26 mg daily. With this dosing strategy there was no significant effect of spironolactone on blood pressure (subhemodynamic dosage), heart rate, or serum creatinine. There was, however, a small, but significant, effect of spironolactone on serum potassium with a mean serum potassium of 4.34 meq/L in the group that received placebo, compared with 4.57 meq/L in the group that was given spironolactone. RALES showed a 30% reduction in mortality that was the result of a decrease in progressive heart failure-related death as well as sudden cardiac death. The Kaplan Meier event curves were still diverging at the time the study was discontinued at a mean follow-up of approximately 2 years. Subgroup analysis suggested a uniform effect of spironolactone without regard to age, gender, cause of heart failure, use of concomitant potassium supplements, creatinine level, and serum potassium level. There was also a significant improvement in NYHA class, as well as a 35% reduction in hospitalization for heart failure. The only notable side effect in the RALES trial was an excess of gynecomastia and breast pain in males. This is a recognized side effect of spironolactone because it binds to androgen and progestosterone receptor. There was no increase in the incidence of severe hyperkalemia (potassium \geq 6.0 meq/L). Only one placebo patient in the RALES trial died with evidence of severe hyperkalemia (serum potassium \geq 6.0 meq/L).

The pathophysiologic basis for targeting aldosterone in the vasculature and myocardium

Brilla et al [13] were among the first to demonstrate that aldosterone may have distinct effects on the vasculature and myocardium. Since then several groups have confirmed their findings and extended their observations to a multitude of effects, some of which are classic mineralocorticoid receptor (MR) mediated, whereas some are more direct, rapid effects as illustrated in Fig. 1.

Effects on the endothelium

Aldosterone-synthesizing enzymes and MR have been demonstrated in endothelial cells and smooth muscle cells; this suggested that these cells are capable of producing aldosterone and responding to it. Aldosterone induces rapid, nongenomic effects, including a rapid increase in intracellular calcium in porcine aortic endothelial cells that reaches a plateau within 2 to 3 minutes and which cannot be blocked by classic MR antagonists [14]. Although angiotensin II and thrombin induce large fluxes in $[Ca^{2+}]_i$, sensitization of cells with aldosterone reduces the concentration of angiotensin II to effect the same change in $[Ca^{2+}]_i$, by nearly 1000-fold [15]. Thus aldosterone could potentiate the effects of other vasoconstrictors. There are emerging data that aldosterone may exert important effects on the

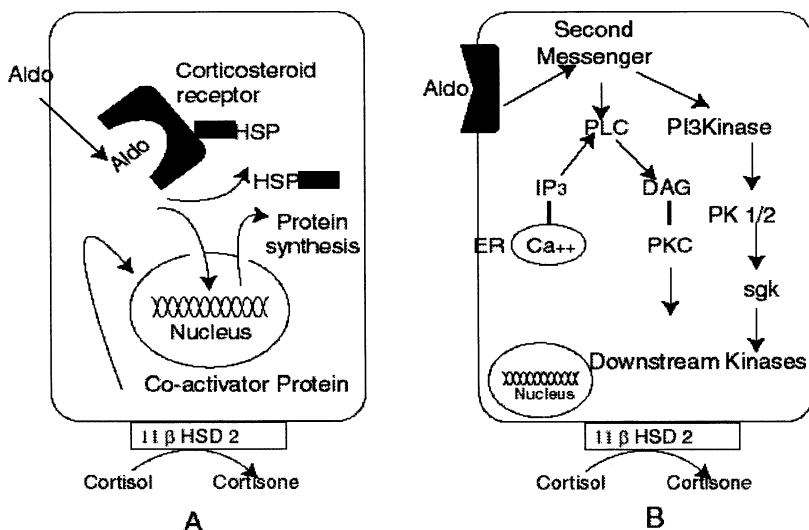


Fig. 1. (A) Classic Type I mineralocorticoid receptor mediated effects of aldosterone. (B) Nongenomic effects of aldosterone in vascular cells. *Abbreviations:* Aldo, aldosterone; DAG, diacylglycerol; ER, endoplasmic reticulum; HSD, hydroxysteroid dehydrogenase; HSP, heat shock protein; IP₃, inositol 1, 4-5 biphosphate; PKC, protein kinase C; PK1/2, protein kinase 1/2; PLC, phospholipase C; sgk, serum and glucocorticoid induced kinase.

nitric oxide pathway in the endothelium. Although it has been known for some time that conditions associated with high levels of circulating aldosterone, such as in primary hyperaldosteronism and heart failure (secondary hyperhyperaldosteronism), are associated with impairment of vasodilatation to acetylcholine [16], more direct evidence was lacking. Farquharson et al [17] demonstrated that patients with advanced heart failure who were already on ACE inhibitor therapy markedly improved their endothelial function (Fig. 2) after 1 month of aldosterone blockade with spironolactone at 50 mg per day. The mechanisms by which aldosterone might influence endothelial function are speculative at this point, but there are some interesting links to suggest that this might involve excess free radical generation. Deoxycorticosterone acetate (DOCA), a precursor of aldosterone has been used for a long time in experimental hypertension. Somers et al [18] treated rats with DOCA for approximately 3 weeks. Systolic blood pressure and aortic superoxide productions were increased in the group that was given DOCA-salt compared with controls. The responses to the endothelium dependent agonist, acetylcholine, were impaired in DOCA-salt hypertension. Treatment with the superoxide scavenger heparin-binding superoxide dismutase had no effect on blood pressure, but significantly improved relaxation to acetylcholine and A23187. As expected in a low renin model, treatment with losartan did not correct the hypertension

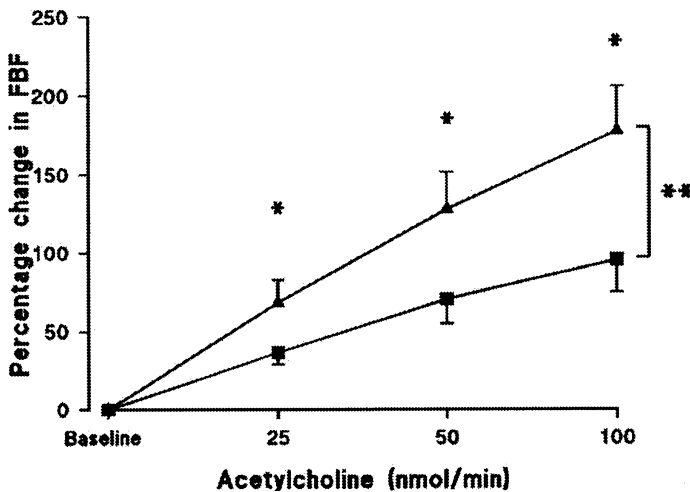


Fig. 2. Forearm blood flow responses to acetylcholine after placebo (■) or spironolactone (▲) therapy. Values are mean \pm SEM. * $P < 0.05$, ** $P < 0.001$ for differences between treatments. Abbreviation: FBF, forearm blood flow. (From Farquharson CJ, Struthers AD. Spironolactone increases nitric oxide bioactivity, improves endothelial vasodilator dysfunction, and suppresses vascular angiotensin I/angiotensin II conversion in patients with chronic heart failure. *Circulation* 2000;101:594–7; with permission.)

or the impaired vascular relaxations. When the animals were treated with the DOCA alone, without the salt, there were no deleterious effects on the vasculature. These findings are in concordance with the notion that salt potentiates aldosterone effect. The authors recently demonstrated that the selective aldosterone receptor antagonist, eplerenone, significantly improved endothelial function in the lipid fed rabbit model and was associated with a decrease in oxygen free radical formation secondary to inhibition of NADH/NADPH oxidase activity. Thus, aldosterone blockade may potentially alter nitric oxide bioavailability in the vasculature in conditions that are associated with upregulation of the renin angiotensin aldosterone axis [19].

Effects on vascular smooth muscle cells

Funder et al [20] initially described vascular type I aldosterone binding sites in rat mesenteric arteries; they found that these were physiologic mineralocorticoid receptors. Since then numerous other investigators demonstrated the presence of a complete aldosterone-synthesizing apparatus in the vascular smooth muscle cells [21]. As in endothelial cells, aldosterone may induce rapid, nongenomic effects in vascular smooth muscle cells, such as increases intracellular calcium, diacylglycerol and inositol-1, 4-5 phosphate at subnanomolar concentrations. The physiologic correlate of these changes may translate into the rapid increase in peripheral vascular resistance and blood pressure that is observed 5 minutes after injecting aldosterone intravenously [22]. Through MR, aldosterone may also potentiate the hypertrophic effects of mitogens, such as angiotensin II [23].

Effects on adventitial/vascular matrix

Fibroblasts that reside within the interstitial space of the adventitia, as well as media, are well-known to have Type I and Type II mineralocorticoid receptors and are responsive to the effects of aldosterone [24]. Experiments by Zhou et al [25] demonstrated that aldosterone increased the synthesis of collagen I at the mRNA and protein level. Recent evidence from animal experiments and in patients with heart failure suggested that chronic aldosterone receptor blockade using spironolactone markedly reduced procollagen type III amino-terminal peptide (PIIINP); this suggested that aldosterone is an important determinant of collagen turnover [26,27]. In the human study (a substudy of the larger RALES trial), baseline PIIINP of greater than 3.85 g/L was an important determinant of subsequent death and of death and hospitalization [27]. Remodeling of the vessel wall occurs in diverse conditions, including atherosclerosis, postangioplasty restenosis, transplant arteriosclerosis, and hypertensive vasculopathy. Because fibrillar collagen (Type I and III) is a major structural component of arteries,

a disproportionate increase in its content is expected to raise tissue stiffness and reduce compliance. Conversely, a reduction in collagen content may have opposite effects. Benetos et al [28] examined whether targeting the MR would attenuate aortic fibrosis in spontaneously hypertensive rats (SHR) [28]. The latter model is associated with an upregulated renin angiotensin aldosterone system and is characterized by extensive vascular remodeling. The effect of a 4-month treatment of spironolactone was compared with the ACE inhibitor quinapril in SHR. Control SHR and Wistar-Kyoto rats received placebo. At the end of treatment, quinapril completely prevented the development of hypertension, whereas spironolactone produced only a slight reduction in blood pressure. Aortic hypertrophy was significantly attenuated by ACE inhibition, but not by MR blockade. In contrast, collagen accumulation in the aorta was completely prevented by quinapril and spironolactone. Spironolactone reduced collagen density below that of controls. Van Belle et al [29] examined the effects of aldosterone receptor blockade in experimental vessel wall remodeling that follows restenotic injury. Neointimal thickening in hyperlipidemic pigs in the study was significantly increased after balloon injury at the level of the aorta and iliac arteries in animals that concomitantly received aldosterone infusions. In contrast, aldosterone receptor blockade with spironolactone inhibited neointimal proliferation in the aorta and iliac arteries. These results support the role of aldosterone in vascular remodeling following balloon angioplasty.

The effects on myocardium

Expression of terminal enzymes of corticosterone and aldosterone synthesis (11 β -hydroxylase and aldosterone synthase) and production of both steroids were demonstrated within rat heart; this confirmed the potential for steroid metabolism in cardiac tissue [30]. MR and 11 β -hydroxysteroid dehydrogenase, which confers mineralocorticoid selectivity to aldosterone target tissues, were detected in heart; this raised the possibility that systemically or locally-synthesized mineralocorticoids influence ventricular remodeling [31,32]. In vivo, aldosterone infusion in the presence of high salt resulted in a disproportionate increase in atrial and perivascular fibrosis of the pulmonary artery, as well as in the aorta. Increased fibrosis in the low pressure atria supports the hypothesis that the fibrotic effects of aldosterone is likely unrelated to hemodynamic effects [33]. In these experiments, however, fibrosis appeared only if aldosterone and sodium intake were increased simultaneously. The role of sodium in the pathogenesis of fibrosis is still unresolved [34] but there is some evidence that salt content may mediate its effects through the aldosterone pathway [35]. Recent experiments showed that aldosterone directly regulates ACE mRNA expression in neonatal cardiomyocytes. This effect was observed with physiologic doses of aldosterone and was dose- and time-dependent.

The magnitude of effect observed (23-fold increase) was profound and could be blocked by spironolactone [36].

Potential mechanisms for the beneficial effects of spironolactone on mortality and morbidity in patients with heart failure

The diuretic effects of spironolactone are insufficient to explain the significant 30% reduction in mortality and 35% reduction in hospitalizations for heart failure in the RALES study. The patients in the RALES trial were already on a loop diuretic and physicians were free to vary the dosage of the loop diuretic over the course of the trial. There was also no evidence of a difference in the clinical occurrence of edema or change in weight between patients randomized to spironolactone or placebo; this suggested that the diuretic effects of spironolactone were not a major factor in the RALES trial [12].

Effects of aldosterone on vascular and myocardial fibrosis

One explanation for the beneficial effect of spironolactone on mortality and morbidity in patients with heart failure relates to the effects of aldosterone on myocardial and vascular fibrosis, which are powerful determinants of mortality [37–39]. Aldosterone is an important determinant of large vessel wall compliance in heart failure and in hypertension [40,41]. It can therefore be postulated that aldosterone receptor antagonists play an important role in conditions such as congestive heart failure, that are associated with decreased large vessel compliance. In concordance with the insights that were gained in experimental animal models whereby aldosterone exerted important effects on collagen turnover in the myocardium, in a substudy of the RALES trial, Zannad et al [27] demonstrated that spironolactone significantly decreased collagen formation as evidenced by a significant decrease in procollagen 1 and 3. A decrease in collagen formation may be expected to result in an improvement in diastolic myocardial function, and, therefore, the trend toward progressive heart failure and pulmonary congestion. A reduction in myocardial fibrosis may also have important implications for the homogeneity of ventricular conduction and sudden cardiac death. For example, spironolactone was shown to improve heart rate variability [26] and QT dispersion [42]. Heart rate variability and baroreceptor function, reflecting vagal tone, are important predictors of sudden cardiac death. The RALES trial confirmed an important effect of spironolactone on sudden cardiac death. Therefore, the effect of aldosterone receptor blocking agents on myocardial fibrosis has important implications for patients with heart failure that is caused by systolic left ventricular dysfunction and may theoretically provide benefit in diastolic heart failure and reduce mortality secondary to sudden cardiac death in patients with both of these conditions.

The role of aldosterone in ventricular remodeling

Silvestre et al [43] evaluated the effects of MR blockade with spironolactone or angiotensin receptor blockade with losartan or the combination on a rat post-infarction model. Treatment with losartan but not spironolactone myocardial infarction (MI) corrected raised aldosterone synthase (CYP11B2) mRNA (the terminal enzyme of aldosterone synthesis) aldosterone levels and cardiac angiotensin II levels; these findings suggest an angiotensin type 1 (AT-1) receptor mediated mechanism of aldosterone synthesis. The MI-induced collagen deposition in noninfarcted myocardium was prevented 1.6-fold with MR blockade and by 2.5-fold with losartan. Losartan and spironolactone reduced cardiac norepinephrine levels. These findings suggest that in pathophysiologic conditions such as infarction, there is an activation of the aldosterone cascade with a consequent preferential increase in collagen density that can be reversed by MR blockade.

Aldosterone as an inflammatory mediator

In the SHR Rocha et al [44] demonstrated that exogenous aldosterone in the presence of an ACE inhibitor led to a loss of the protective effect of the latter and accentuated end-organ damage in the heart and the kidney through proinflammatory effects. Conversely, the use of spironolactone in this animal model in subhemodynamic doses decreased proteinuria, histologic evidence of inflammatory injury, and improved survival in comparison with control animals [45]. A recent study by Oelkers [46] suggested that the deleterious effects of aldosterone on the kidney may be related to aldosterone's role as a potent hydroxyl radical generator. Stier showed that the hydroxyl radical inhibitor dimethylthiourea can prevent the renal damage that is associated with aldosterone at subhemodynamic doses [47]. The finding that aldosterone is a potent oxygen free radical generator has important implications for the therapy of heart failure as well as for other major cardiovascular diseases. Fiebeler and his colleagues [48] demonstrated that in an angiotensin excess transgenic model, spironolactone reduced the activation of proinflammatory NF- κ B and AP-1 signaling pathways that may lead to activation of adhesion molecules, endothelial damage, and fibrosis. Harada et al [36] demonstrated a profound effect of aldosterone on ACE expression. The effects of aldosterone were completely abolished by spironolactone which suggested an MR effect. This supported the existence of a positive feedback loop in the myocardium, in conditions such as heart failure, where aldosterone generates more angiotensin II and continues a vicious cycle. Angiotensin II, in turn, serves as a prototypical proinflammatory mediator and may further potentiate aldosterone injury. Taken together, these findings support the idea that aldosterone results in proinflammatory effects and antagonism of MR may exert powerful

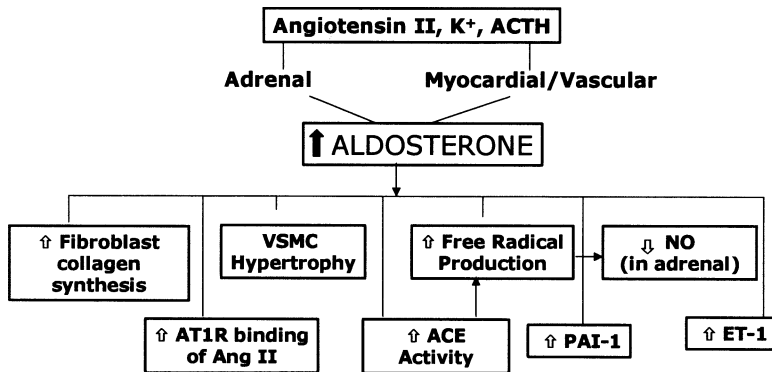


Fig. 3. Mechanisms of aldosterone-mediated potentiation of heart failure.

antiinflammatory effects. Collectively, the effects of aldosterone receptor antagonists have important implications for the therapy of heart failure and other cardiovascular disease (Fig. 3).

Effects of aldosterone on PAI-1: implications for remodeling

Aldosterone contributes to the regulation of plasminogen activator inhibitor-1 (PAI-1) expression in cultured human endothelial cells and smooth muscle cells. Using PAI-1 promoter reporter constructs, Brown et al [49] demonstrated dose-dependent increases in luciferase activity by aldosterone when used with angiotensin II (100 nmoles/L). Neither dexamethasone nor aldosterone alone increased PAI-1 expression. This effect was abolished by mutation in the region of a putative glucocorticoid-responsive element. The time course of the effect of aldosterone on angiotensin II-induced PAI-1 expression was consistent with an MR-dependent mechanism. In humans, PAI-1 levels correlate strongly with aldosterone levels [50]. Conversely, angiotensin II/aldosterone reducing strategies decrease plasminogen activator inhibitor levels, and, therefore, may improve fibrinolysis and prevent thrombosis [51]. A decrease in plasminogen activator inhibitor levels has important implications for thrombosis and the development of fibrosis because the plasmin system plays an important upstream role in the regulation of a variety of serine proteases, such as matrix metalloproteinases (MMPs) [52].

When should an aldosterone receptor antagonist be used in patients with systolic heart failure?

In patients with mild to moderate heart failure, ACE inhibitors and β -adrenergic receptor blocking agents have been shown to be effective in reducing morbidity and mortality [53–56]. There are no data from

well-designed, prospective, randomized trials about the effectiveness of aldosterone receptor antagonists in these patients. Based on the RALES study, spironolactone, at a daily dosage of 25 mg, with monitoring of serum potassium, is indicated in patients with severe or progressive heart failure caused by systolic left ventricular dysfunction [12]. In a patient with severe heart failure and evidence of volume overload, an ACE inhibitor, loop diuretic \pm digoxin, and spironolactone is the preferred therapy. After the patient is euvolemic, an attempt should be made to add a β -adrenergic receptor blocking agent. There is evidence that β -adrenergic receptor blocking agents are effective in reducing mortality in patients with severe heart failure who are euvolemic but not in those with evidence of volume overload or edema [56]. Although only 10% to 11% of patients in the RALES trial were on a β -adrenergic receptor blocking agent, the reduction in mortality was not significantly different from those without a β -adrenergic receptor blocking agent. In fact, the point estimate for a reduction in mortality in those on an ACE inhibitor, a β -adrenergic blocking agent, and spironolactone was greater than those on an ACE inhibitor and spironolactone alone [57]. The interaction of an aldosterone receptor antagonist and a β -adrenergic blocking agent should be further elucidated by the results of the ongoing Eplerone Neurohormonal Efficacy and Survival Study (EPHESUS) trial [58].

How should an aldosterone receptor antagonist (spironolactone) be used in heart failure?

The authors advocate starting spironolactone at 25 mg a day. Serum potassium should be monitored at the end of 1 week of treatment, and, periodically thereafter, for the first 2 to 3 months. If serum potassium levels increase to 5.5 meq/L or greater, the authors advise reducing the dose of spironolactone to 25 mg every other day. The authors do not advise discontinuing spironolactone in patients whose serum potassium is between 5.5 meq/L and 6.0 meq/L, however, unless there were concomitant electrocardiographic changes that suggest hyperkalemia. It would, however, be prudent to discontinue spironolactone in any patient in whom serum potassium levels increase to 6.0 meq/L or higher. Before discontinuing spironolactone for patients with hyperkalemia, care should be taken to ensure that the blood sample was not hemolyzed or elevated secondary to other medications that are known to increase serum potassium levels, such as supplements and NSAIDs. Since the publication of the RALES trial there have been suggestions that when spironolactone is used in clinical practice there is an increased incidence of hyperkalemia [59]. Although severe hyperkalemia is a potential risk of using spironolactone in conjunction with an ACE inhibitor in patients with heart failure, this risk should be minimal if the agent is avoided in individuals with a serum creatinine level that is greater than or equal to 2.5 mg/dL or a potassium level that is greater than

or equal to 5.0 meq/L, as was done in the RALES trial. Some of the cases of hyperkalemia in clinical practice occurred when spironolactone was instituted at a dose greater than 25 mg or when serum potassium levels were not closely monitored [59].

Selective aldosterone receptor antagonists: an emerging treatment paradigm

A variety of compounds with antimineralocorticoid (partial or complete) can be generated by modifying the nature of substitution in the steroid nucleus [60]. One such compound with pure antagonistic properties is eplerenone. The structure of eplerenone differs from spironolactone by having the lactone group in the C-17 position replaced with a carbomethoxyl group (Fig. 4). In doing so, a molecule with 10- to 20-fold less affinity for MR, that is largely devoid of antiprogestational and antiandrogenic effects, and with minimal agonistic properties is generated. The half-life of eplerenone is approximately 3.5 hours in human beings; it lacks significant metabolites. The ongoing EPHESUS trial is the first large trial to test the effect of a selective aldosterone receptor antagonist strategy in post-infarct patients. In this study, 6200 patients with evidence of systolic left ventricular dysfunction postmyocardial infarction and evidence of heart failure, 3 to 14

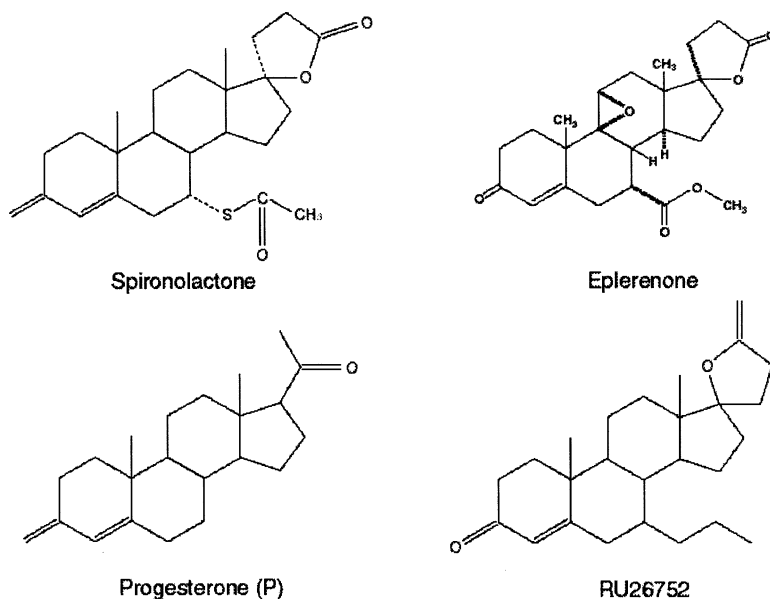
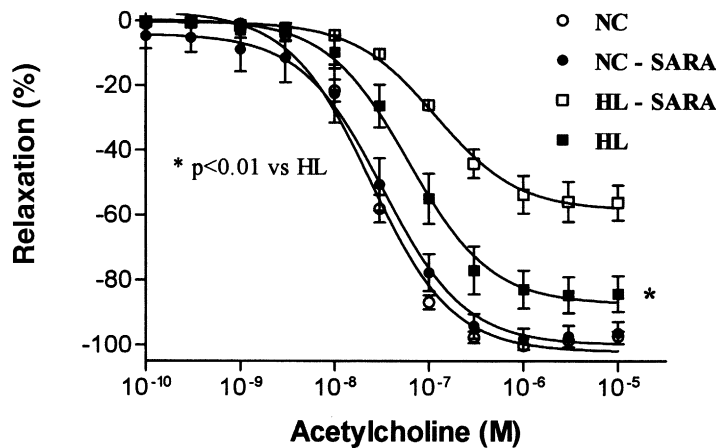
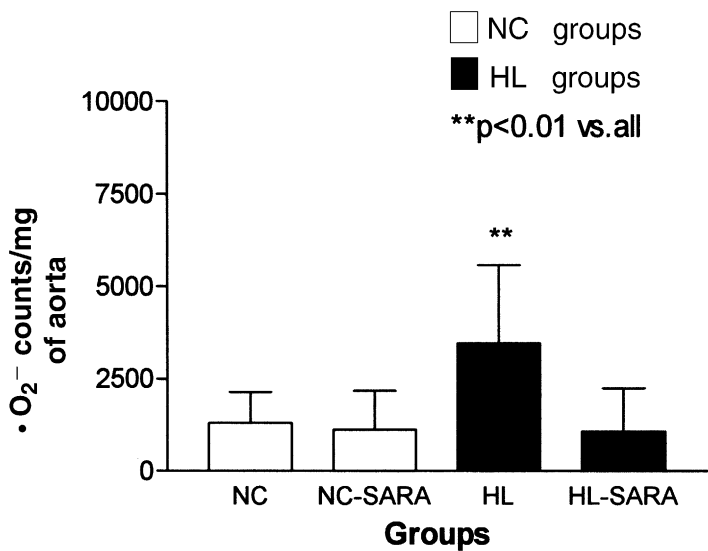


Fig. 4. Structures of mineralocorticoid receptor antagonists, spironolactone, eplerenone, and RU26752 (progestogen with MR antagonist properties). The structure of progesterone is provided for comparison.



days postmyocardial infarction are being randomized to placebo or the specific aldosterone receptor antagonist, eplerenone. Patients in this trial may receive standard therapy, including reperfusion, revascularization, an ACE inhibitor, β -adrenergic receptor blocking agent, aspirin, and a statin. The primary endpoints all cause mortality and the combination of cardiovascular mortality and cardiovascular hospitalizations. The results of this trial are expected by early 2003. A small, randomized study with the aldosterone receptor antagonist, potassium canrenoate, in patients with anterior myocardial infarction demonstrated a reduction in collagen synthesis and progressive left ventricular dilation. These findings are encouraging, especially because more than 40% of patients were on concomitant β -adrenergic blocking agents and all were on an ACE-inhibitor; this is reflective of contemporary practice and suggests an incremental benefit with aldosterone receptor antagonists [61].

Progestogens as selective aldosterone receptor antagonist (SARA)

Progesterone can also act as an MR antagonist (Fig. 5) and dissociates rapidly from the receptor after binding that results in deactivation [62]. Synthetic derivatives of progesterone with high affinity binding to MR can be generated and may serve the dual purpose of a progestogen as well as aldosterone receptor antagonists. Drospirenone is a synthetic progesterone derivative that binds to the mineralocorticoid receptor. Thus in addition to contraception, it also has a weak diuretic effect and prevents weight gain during the menstrual cycle. In addition, it is purported to have favorable effects on mood and is being promoted as a medication to counteract premenstrual dysphoria [46,63].

Summary

Based upon the results of the RALES trial and accumulating evidence about the role of aldosterone and aldosterone receptor antagonism in various disease states, the authors anticipate that aldosterone receptor antagonists will become standard therapy, along with ACE inhibitors and

◀ Fig. 5. The effect of SARA on O_2^- production (*upper graph*). (*Lower graph*) Dose response curves to acetylcholine and superoxide production in diet-induced hyperlipidemic rabbits who were treated with eplerenone or placebo. Values represent responses to graded dosages of drug expressed as a percentage of preconstricted tension in response to phenylephrine. All values represent means \pm SEM. *Abbreviations:* NC, normal chow; NC-SARA, normal chow and selective aldosterone receptor antagonist; HL, hyperlipidemic chow; HL-SARA, hyperlipidemic chow with selective aldosterone receptor antagonist; P, progesterone. * $P < 0.01$ vs. HL-SARA and NC groups. ** $P < 0.01$ vs. HL. P is not significant for HL-SARA vs. NC and NC-SARA by ANOVA. (From Rajagopalan S, Duquaine D, King S, et al. Mineralocorticoid receptor antagonism in experimental atherosclerosis. *Circulation* 2002;105:2212–6; with permission.)

β -adrenergic receptor blocking agents, in patients with heart failure that is caused by systolic left ventricular dysfunction. Furthermore, the prospect of the use of these agents in other disease states that have implicated an activated rennin-angiotensin-aldosterone cascade, such as diastolic dysfunction, aging, and atherosclerosis, remains to be tested. Until further data from well-designed, prospective, randomized trials are available, the use of aldosterone receptor antagonists should be restricted to patients with severe or progressive heart failure caused by systolic left ventricular dysfunction in whom serum creatinine level is ≤ 2.0 mg/dL and serum potassium levels are <5.0 meq/L at baseline.

References

- [1] Dzau VJ, Colucci WS, Hollenberg NK, et al. Relation of the renin-angiotensin-aldosterone system to clinical state in congestive heart failure. *Circulation* 1981;63:645–51.
- [2] Schrier RW, Abraham WT. Hormones and hemodynamics in heart failure. *N Engl J Med* 1999;341:577–85.
- [3] Vantrimpont P, Rouleau JL, Ciampi A, et al. Two-year time course and significance of neurohumoral activation in the Survival and Ventricular Enlargement (SAVE) Study [Comments]. *Eur Heart J* 1998;19:1552–63.
- [4] Rajagopalan S, Pitt B. Aldosterone antagonists in the treatment of hypertension and target organ damage. *Curr Hypertension Repts* 2001;3:240–8.
- [5] Pitt B. Escape of aldosterone production in patients with left ventricular dysfunction treated with an angiotensin converting enzyme inhibitor: implications for therapy. *Cardiovasc Drugs Ther* 1995;9:145–9.
- [6] Staessen J, Lijnen P, Fagard R, et al. Rise in plasma concentration of aldosterone during long-term angiotensin II suppression. *J Endocrinol* 1981;91:457–65.
- [7] McKelvie RS, Yusuf S, Pericak D, et al. Comparison of candesartan, enalapril, and their combination in congestive heart failure: randomized evaluation of strategies for left ventricular dysfunction (RESOLVD) pilot study. The RESOLVD Pilot Study Investigators [Comments]. *Circulation* 1999;100:1056–64.
- [8] Andreis PG, Tortorella C, Malendowicz LK, et al. Endothelins stimulate aldosterone secretion from dispersed rat adrenal zona glomerulosa cells, acting through ETB receptors coupled with the phospholipase C-dependent signaling pathway. *Peptides* 2001;22:117–22.
- [9] Hinson JP, Kapas S. The role of endothelial cell products in the regulation of adrenocortical function: actions of endothelin, nitric oxide, adrenomedullin and PAMP. *Horm Metab Res* 1998;30:334–40.
- [10] Natarajan R, Lanting L, Bai W, et al. The role of nitric oxide in the regulation of aldosterone synthesis by adrenal glomerulosa cells. *J Steroid Biochem Mol Biol* 1997;61:47–53.
- [11] Quinn S, Williams G. Regulation of aldosterone secretion. *Annu Rev Physiol* 1988;50:409–26.
- [12] Pitt B, Zannad F, Remme W, et al. The effect of spironolactone on morbidity and mortality in patients with severe heart failure. *N Engl J Med* 1999;341:709–17.
- [13] Brilla CG, Pick R, Tan LB, et al. Remodeling of the rat right and left ventricles in experimental hypertension. *Circ Res* 1990;67:1355–64.
- [14] Wehling M, Ulsenheimer A, Schneider M, et al. Rapid effects of aldosterone on free intracellular calcium in vascular smooth muscle and endothelial cells: subcellular localization.

- tion of calcium elevations by single cell imaging. *Biochem Biophys Res Commun* 1994;204:475–81.
- [15] Wehling M, Neylon CB, Fullerton M, et al. Nongenomic effects of aldosterone on intracellular Ca^{2+} in vascular smooth muscle cells. *Circ Res* 1995;76:973–9.
- [16] Taddei S, Virdis A, Mattei P, et al. Vasodilation to acetylcholine in primary and secondary forms of human hypertension. *Hypertension* 1993;21:929–33.
- [17] Farquharson CA, Struthers AD. Spironolactone increases nitric oxide bioactivity, improves endothelial vasodilator dysfunction, and suppresses vascular angiotensin II/angiotensin II conversion in patients with chronic heart failure. *Circulation* 2000;101:594–7.
- [18] Somers MJ, Mavromatis K, Galis ZS, et al. Vascular superoxide production and vasomotor function in hypertension induced by deoxycorticosterone acetate-salt. *Circulation* 2000;101:1722–8.
- [19] Rajagopalan S, Duquaine D, King S, et al. Mineralocorticoid receptor antagonism in experimental atherosclerosis. *Circulation* 2002;105:2212–6.
- [20] Funder JW, Pearce PT, Smith R, et al. Vascular type I aldosterone binding sites are physiological mineralocorticoid receptors. *Endocrinology* 1989;125:2224–6.
- [21] Hatakeyama H, Miyamori I, Fujita T, et al. Vascular aldosterone. Biosynthesis and a link to angiotensin II-induced hypertrophy of vascular smooth muscle cells. *J Biol Chem* 1994;269:24316–20.
- [22] Klein KHW. Klinisch-experimentelle untersuchungen uber den ein-flub von aldosteron auf hamodynamik und gerinnung. *Z Kreisl Forsch* 1963;52:40–53 [in German].
- [23] Xiao F, Puddefoot JR, Vinson GP. Aldosterone mediates angiotensin II-stimulated rat vascular smooth muscle cell proliferation. *J Endocrinol* 2000;165:533–6.
- [24] Lombes M, Oblin ME, Gasc JM, et al. Immunohistochemical and biochemical evidence for a cardiovascular mineralocorticoid receptor. *Circ Res* 1992;71:503–10.
- [25] Zhou G, Kandala JC, Tyagi SC, et al. Effects of angiotensin II and aldosterone on collagen gene expression and protein turnover in cardiac fibroblasts. *Mol Cell Biochem* 1996;154:171–8.
- [26] MacFadyen RJ, Barr CS, Struthers AD. Aldosterone blockade reduces vascular collagen turnover, improves heart rate variability and reduces early morning rise in heart rate in heart failure patients. *Cardiovasc Res* 1997;35:30–4.
- [27] Zannad F, Alla F, Dousset B, et al. Limitation of excessive extracellular matrix turnover may contribute to survival benefit of spironolactone therapy in patients with congestive heart failure: insights from the Randomized Aldactone Evaluation Study (RALES). *Circulation* 2000;102:2700–6.
- [28] Benetos A, Lacolley P, Safar ME. Prevention of aortic fibrosis by spironolactone in spontaneously hypertensive rats. *Arteriosclerosis. Thrombosis Vascular Biol* 1997;17:1152–6.
- [29] Van Belle E, Bauters C, Wernert N, et al. Neointimal thickening after balloon denudation is enhanced by aldosterone and inhibited by spironolactone, and aldosterone antagonist. *Cardiovasc Res* 1995;29:27–32.
- [30] Silvestre JS, Robert V, Heymes C, et al. Myocardial production of aldosterone and corticosterone in the rat. Physiological regulation. *J Biol Chem* 1998;273:4883–91.
- [31] Funder JW. Glucocorticoid and mineralocorticoid receptors: biology and clinical relevance. *Annu Rev Med* 1997;48:231–40.
- [32] Slight SH, Ganjam VK, Gomez-Sanchez CE, et al. High affinity NAD(+)-dependent 11 beta-hydroxysteroid dehydrogenase in the human heart. *J Mol Cell Cardiol* 1996;28:781–7.
- [33] Sun Y, Ratajska A, Zhou G, et al. Angiotensin-converting enzyme and myocardial fibrosis in the rat receiving angiotensin II or aldosterone. *J Lab Clin Med* 1993;122:395–403.
- [34] Funder JW. Aldosterone, salt and cardiac fibrosis. *Clin Exp Hypertens* 1997;19:885–99.

- [35] Takeda Y, Yoneda T, Demura M, et al. Sodium-induced cardiac aldosterone synthesis causes cardiac hypertrophy. *Endocrinology* 2000;141:1901–4.
- [36] Harada E, Yoshimura M, Yasue H, et al. Aldosterone induces angiotensin-converting-enzyme gene expression in cultured neonatal rat cardiocytes. *Circulation* 2001;104:137–9.
- [37] Safar ME. Epidemiological findings imply that goals for drug treatment of hypertension need to be revised. *Circulation* 2001;103:1188–90.
- [38] Safar ME. Mechanical factors predicting cardiovascular risk and drug treatment of hypertension. *J Hypertens* 2002;20:349–52.
- [39] Safar ME. Systolic blood pressure, pulse pressure and arterial stiffness as cardiovascular risk factors. *Curr Opin Nephrol Hypertens* 2001;10:257–61.
- [40] Blacher J, Amah G, Girerd X, et al. Association between increased plasma levels of aldosterone and decreased systemic arterial compliance in subjects with essential hypertension. *Am J Hypertens* 1997;10:1326–34.
- [41] Duprez DA, De Buyzere ML, Rietzschel ER, et al. Inverse relationship between aldosterone and large artery compliance in chronically treated heart failure patients [Comments]. *Eur Heart J* 1998;19:1371–6.
- [42] Yee KM, Pringle SD, Struthers AD. Circadian variation in the effects of aldosterone blockade on heart rate variability and QT dispersion in congestive heart failure. *J Am Coll Cardiol* 2001;37:1800–7.
- [43] Silvestre J-S, Heymes C, Oubenaissa A, et al. Activation of cardiac aldosterone production in rat myocardial infarction: effect of angiotensin ii receptor blockade and role in cardiac fibrosis. *Circulation* 1999;99:2694–701.
- [44] Rocha R, Chander PN, Zuckerman A, et al. Role of aldosterone in renal vascular injury in stroke-prone hypertensive rats. *Hypertension* 1999;33:232–7.
- [45] Rocha R, Chander PN, Khanna K, et al. Mineralocorticoid blockade reduces vascular injury in stroke-prone hypertensive rats. *Hypertension* 1998;31:451–8.
- [46] Oelkers W. Drospirenone—a new progestogen with antimineralocorticoid activity, resembling natural progesterone. *Eur J Contracept Reprod Health Care* 2000;5:17–24.
- [47] Stier C Jr. Anti-oxidants reduce aldosterone-induced renal vascular injury in stroke-prone spontaneously hypertensive rats [abstract]. 18th scientific meeting of the International Society of Hypertension, Chicago, IL.
- [48] Fiebeler A, Schmidt F, Muller DN, et al. Mineralocorticoid receptor affects AP-1 and nuclear factor- κ B activation in angiotensin II-induced cardiac injury. *Hypertension* 2001;37:787–93.
- [49] Brown NJ, Kim KS, Chen YQ, et al. Synergistic effect of adrenal steroids and angiotensin II on plasminogen activator inhibitor-1 production. *J Clin Endocrinol Metab* 2000;85:336–44.
- [50] Brown NJ, Agirbasli MA, Williams GH, et al. Effect of activation and inhibition of the renin-angiotensin system on plasma PAI-1. *Hypertension* 1998;32:965–71.
- [51] Brown NJ, Abbas A, Byrne D, et al. Comparative effects of estrogen and angiotensin-converting enzyme inhibition on plasminogen activator inhibitor-1 in healthy postmenopausal women. *Circulation* 2002;105:304–9.
- [52] Luttun A, Dewerchin M, Collen D, et al. The role of proteinases in angiogenesis, heart development, restenosis, atherosclerosis, myocardial ischemia, and stroke: insights from genetic studies. *Current Atherosclerosis Reports* 2000;2:407–16.
- [53] Anonymous. Effect of enalapril on survival in patients with reduced left ventricular ejection fractions and congestive heart failure. The SOLVD Investigators. *N Engl J Med* 1991;325:293–302.
- [54] Anonymous. Effect of metoprolol CR/XL in chronic heart failure: Metoprolol CR/XL Randomised Intervention Trial in Congestive Heart Failure (MERIT-HF). *Lancet* 1999;353:2001–7.
- [55] Anonymous. Effects of enalapril on mortality in severe congestive heart failure. Results of the Cooperative North Scandinavian Enalapril Survival Study (CONSENSUS). The CONSENSUS Trial Study Group. *N Engl J Med* 1987;316:1429–35.

- [56] Packer M, Bristow MR, Cohn JN, et al. The effect of carvedilol on morbidity and mortality in patients with chronic heart failure. U.S. Carvedilol Heart Failure Study Group. *N Engl J Med* 1996;334:1349–55.
- [57] Pitt B, Zannad F, Remme WJ, et al. The effect of spironolactone on morbidity and mortality in patients with severe heart failure. Randomized Aldactone Evaluation Study Investigators. *N Engl J Med* 1999;341:709–17.
- [58] Pitt B, Williams G, Remme W, et al. The EPHESUS Trial: eplerenone in patients with heart failure due to systolic dysfunction complicating acute myocardial infarction. *Cardio-vasc Drugs Ther* 2001;15:79–87.
- [59] Schepkens H, Vanholder R, Billioud JM, et al. Life-threatening hyperkalemia during combined therapy with angiotensin-converting enzyme inhibitors and spironolactone: an analysis of 25 cases. *Am J Med* 2001;110:438–41.
- [60] Delyani JA. Mineralocorticoid receptor antagonists: the evolution of utility and pharmacology. *Kidney Int* 2000;57:1408–11.
- [61] Modena MG, Aveta P, Menozzi A, et al. Aldosterone inhibition limits collagen synthesis and progressive left ventricular enlargement after anterior myocardial infarction. *Am Heart J* 2001;141:41–6.
- [62] Couette B, Lombes M, Baulieu EE, et al. Aldosterone antagonists destabilize the mineralocorticosteroid receptor. *Biochem J* 1992;282:697–702.
- [63] Mansour D. Yasmin—a new oral contraceptive, a new progestogen: the reasons why. *Eur J Contracept Reprod Health Care* 2000;5:9–16.