

1 Steroidal and non-steroidal mineralocorticoid receptor antagonists in cardiorenal medicine

2 Supplementary material

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4 Supplementary Table S1: Molecular insights from preclinical MR models

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Model	Phenotype	Pathophysiology	Reference
Complete knock-out of the MR	Death within 10 days, pseudohypoaldosteronism	Increased renin, aldosterone and potassium; decreased sodium and ENaC in the kidney (24%) and colon (16%), but no change in mRNA abundance of ENaC	Berger 1998 ¹
Kidney-specific MR knock-out			
Principal cell, renal tubule	Salt wasting, survival to adulthood. Sodium/water loss on low-sodium diet	Increased aldosterone; decreased ENaC in CCD, impaired trafficking	Ronzaud 2007 ²
Renal nephron	Severe pseudohypoaldosteronism-1. Death on sodium-deficient diet	Increased aldosterone and potassium; decreased sodium and ENaC-a; increased GR; decreased sodium chloride co-transporter downstream change to increased potassium	Canonica 2016; ³ Terker 2016 ⁴
Cardiomyocyte-specific MR knock-out			
Infarct-induced cardiac remodelling		Increased infarct healing and capillary density; decreased pulmonary oedema, progressive adverse cardiac remodelling, contractile dysfunction,	Fraccarollo 2011 ⁵

		and Nox2 upregulation; reduced ECM deposition; increased stress-induced activation and subsequent suppression of nuclear factor- κ B and decreased apoptosis	
DOCA–salt-induced cardiac inflammation and fibrosis		Similar recruitment of macrophages decreased cardiac inflammation and fibrosis at baseline, at 8 days and 8 weeks; increased decorin (at baseline and 8 weeks), mRNA and protein. BP response	Rickard 2012 ⁶
Uninephrectomy–salt \pm DOCA, ischaemia reperfusion heart		MR excess. Increased peak contraction during ischaemia, regardless of genotype. Recovery of LV pressure and rates of contraction increased in KO. Incidence of arrhythmic activity during early reperfusion significantly higher in WT than in KO hearts	Bienvendu 2015 ⁷
Cell-specific MR knock-out			
Vascular smooth muscle-specific MR knock-out	Decreased BP with age, without defects in renal sodium handling or vascular structure	Decreased vascular myogenic tone, agonist-dependent contraction, activity of L-type calcium channels; reduced ang II-induced vascular oxidative stress	McCurley 2012 ⁸
Endothelial cell-specific MR knock-out	DOCA–salt-induced cardiac fibrosis	Endothelial function better in response to aldosterone in conduit artery (aorta) but not resistance artery (mesenteric); cardiac inflammation less at 8 days, and fibrosis and inflammation less at 8 weeks. BP unchanged with WT or EC-MRKO	Rickard 2014 ⁹
MR deletion in podocytes	No different from WT mice; eplerenone was protective		Huang 2014 ¹⁰
MR deletion in myeloid cells	Albuminuria reduced on day 1; reduced tubular damage and fibrosis	Decreased cystatin C, crescents, macrophages and neutrophils; pro-inflammatory cytokines	Huang 2014 ¹⁰

Myeloid/macrophage-specific knock out of MR, with extra stimuli			
DOCA–salt uninephrectomy	Decreased function of macrophages; similar recruitment of macrophages; reduced cardiac fibrosis and hypertension		Rickard 2009 ¹¹
Ang II/L-NAME	Macrophages from mice lacking MR in myeloid cells exhibited a transcription profile of alternative activation (in contrast to classical activation of M1 macrophages)	Cardiac hypertrophy, fibrosis and vascular damage. BP increased in KO, especially in PP and complete loss of PP circadian variation	Usher 2010 ¹²
Stroke–ischaemia reperfusion	Infarct volume reduced by 65%; decreased recruitment of macrophages; suppressed M1 macrophages and partially restored M2 macrophages		Frieler 2011 ¹³
Anti-Thy1 GN	Decreased fibrosis and inflammation, and proteinuria at 1 week compared with eplerenone, GFR protection		Huang 2014 ¹⁰
Salt ± DOCA. Cardiac fibrosis at 8 weeks	Activation of the c-Jun NH2-terminal kinase pathway in macrophages after a tissue injury and inflammatory stimuli in the DOCA–salt model is MR-dependent and regulates the transcription of downstream pro-fibrotic factors		Shen 2017 ¹⁴
Myeloid cell-restricted MR deletion by Cre-Lox	Compared with WT controls, MRKO mice had: improved cardiac function and remodelling after		Fraccarollo 2019 ¹⁵

recombination	MI; enhanced neutrophil efferocytosis; reduced superoxide production; improved post-ischaeamic wound healing		
MR overactivation			
Human MR overexpression in MR-sensitive tissues	Renal and cardiac abnormalities including enlarged kidneys (associated with tubular dilation) and mild dilatated cardiomyopathy	Increased renal and cardiac SGK expression and increased cardiac ANP expression. Dilated cardiomyopathy not associated with induction of cardiac fibrosis	Le Menuet 2001 ¹⁶
Conditional cardiomyocyte-specific overexpression of the human MR	Embryonic and/or early postnatal lethality. High early mortality in surviving mice	Normal cardiac structure but functional heart defect predisposing to fatal arrhythmia. No increase in cardiac fibrosis or inflammation.	Ouvrard-Pascaud 2005 ¹⁷
Rac1 knockout mice	Podocytes undergo conversion to a motile phenotype	Podocyte injury, proteinuria, FSGS, cardiac hypertrophy, cardiomyopathy, heart failure, arrhythmia	Nagase 2013 ¹⁸

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2 ang II, angiotensin II; ANP, atrial natriuretic peptide; BP, blood pressure; CCD, cortical-collecting duct; DOCA, deoxycorticosterone acetate; EC-
3 MRKO, endothelial cell mineralocorticoid receptor knock-out; ECM, extracellular matrix; ENaC, endothelial sodium channel; FSGS, focal
4 segmental glomerulosclerosis; GBM, glomerular basement membrane; GN, glomerulonephritis; GR, glucocorticoid receptor; KO, knock-out; LV,
5 left ventricular; MI, myocardial infarction; MR, mineralocorticoid receptor; MRKO, mineralocorticoid receptor knock-out; PP, pulse pressure;
6 SGK, serum and glucocorticoid-inducible kinase; WT, wild-type

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