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# Discovery of Novel Cyanodihydropyridines as Potent Mineralocorticoid Receptor Antagonists

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A new 1,4-dihydropyridine **5a**, containing a cyano group at the C3 position, was recently reported to possess excellent mineralocorticoid receptor (MR) antagonist in vitro potency and no calcium channel-blocker (CCB) activity. In the present study, we report the structure−activity relationships of this novel series of cyano ester dihydropyridines that resulted in $R_6$ substituted analogues with improved metabolic stability while maintaining excellent MR antagonist activity and selectivity against other nuclear receptors. Further structure optimization with the introduction of five-membered ring heterocycles at $R_6$ resulted in compounds with excellent MR antagonist potency and a suitable pharmacokinetic profile. In vivo studies of a promising tool compound in the Dahl salt-sensitive rat model of hypertension showed similar blood pressure (BP) reduction as the steroidal MR antagonist eplerenone, providing proof-of-concept (POC) for a nonsteroidal, orally efficacious MR antagonist.

## Introduction

Hypertension or high blood pressure (BP[a]) is the world's most common killer, with an estimated one in three adults in the U.S. being affected by this condition, putting them at a markedly increased risk of major cardiovascular and renal diseases and shortened life expectancy. Almost 70% of hypertensive patients fail to reach their BP targets, with many needing multiple agents to maintain their BP goal.[1] Despite the antihypertensive market being both mature and highly competitive, current drugs are poorly differentiated in terms of clinical efficacy. Major antihypertensive drug classes in the market comprise diuretics, calcium channel blockers (CCB), and drugs that target the renin−angiotensin−aldosterone system, including rennin inhibitors, angiotensin converting enzyme inhibitors (ACEi), and angiotensin receptor blockers (ARBs).[2] Aldosterone is a steroid hormone that mediates sodium reabsorption by binding to the mineralocorticoid receptor (MR, NR3C2), a member of the nuclear receptor superfamily of ligand-dependent transcription factors. Aldosterone binding initiates a series of events that lead to the interaction of MR with the regulatory regions of target genes. Abnormal activation of the MR by elevated levels of aldosterone and salt imbalance causes hypertension and other detrimental effects to the cardiovascular system such as heart failure and myocardial fibrosis.[3] Two approaches have been developed to treat this abnormal activation of the MR. One approach involves lowering elevated aldosterone levels using aldosterone synthase inhibitors.[4] Alternatively, MR antagonists, such as spironolactone **2** and more recently eplerenone **1** (Figure 1), bind to the receptor, blocking the activation of target gene transcription. This results in lowering of blood pressure in hypertensive patients and improves survival in heart failure patients.[5] Human clinical trials have also shown that administration of MR antagonists spironolactone and eplerenone resulted in a greater reduction in albuminuria compared to ACE inhibitors with similar hypotensive effects in hypertensive patients, demonstrating the therapeutic potential of MR antagonists for diabetic nephropathy.[6]

In general, steroidal MR antagonists present issues of complex chemical synthesis, undesirable physical properties, and poor selectivity against other steroid hormone receptors. The last results in unwanted side effects in the clinic. For example, poor selectivity against the androgen receptor is thought to be responsible for inducing gynocomastia in male patients treated with spironolactone.[7] Thus, there has been a keen interest in discovering novel classes of selective nonsteroidal MR antagonists in recent years.[8]

The 1,4-dihydropyridine (DHP) scaffold has been used to develop nonsteroidal MR antagonists. 4-Aryl-1,4-dihydropyridines have been claimed as inhibiting [$^3$H]aldosterone binding to MR at 10 $\mu$M. However, no data were disclosed regarding the functional activity of this series.[9] We previously reported that a number of approved 1,4-dihydropyridine CCBs were found to inhibit aldosterone-induced activation of a luciferase reporter driven by the MR ligand binding domain (LBD).[10] Further study of asymmetric diester DHPs, such as mebudipine **3**, showed that only the *R*-isomer was MR active while the other enantiomer was CCB active.[11] Replacement of the methyl ester in the mebudipine scaffold with a cyano group resulted in **5a** which possesses excellent MR in vitro potency and is devoid of

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[a] Abbreviations: BP, blood pressure; CCB, calcium channel blocker; ACEi, angiotensin converting enzyme inhibitor; ARBs, angiotensin receptor blockers; MR, mineralocorticoid receptor; DHP, 1,4-dihydropyridine; LBD, ligand binding domain; POC, proof of concept; NHR, nuclear hormone receptor; SS, salt sensitive; SAR, structure−activity relantionship; ER, estrogen receptor; PR, progesterone receptor; GR, glucocorticoid receptor; AR, androgen receptor; PK, pharmacokinetic; CL, clearance, HLM, human liver microsome; RLM, rat liver microsome; CYP, cytochrome P450.

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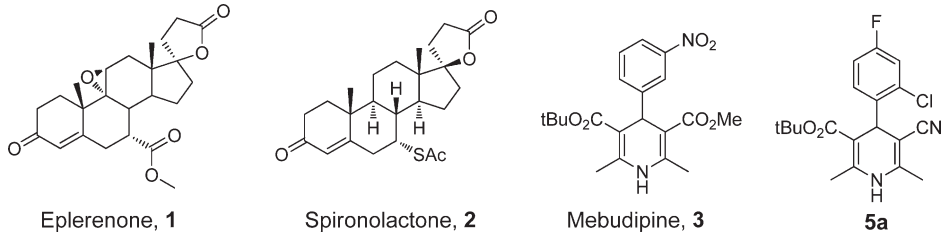
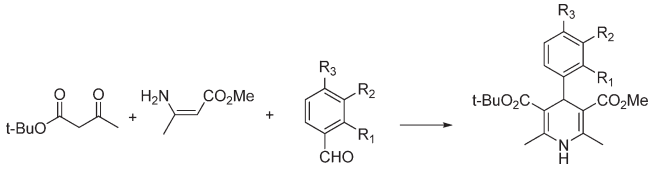



**Figure 1**

**Scheme 1.** Preparation of Diester DHPs **4a**−**t**[a]



[a] Reagents and conditions: piperidine (cat.), MeOH, 65 °C, overnight.

CCB activity. These findings increased our interest in the development of DHPs as selective MR antagonists. (*R*)-Mebudipine was an attractive starting point in terms of its potent MR in vitro activity, selectivity against the L-calcium channel, and being a druggable scaffold.[12] However, the presence of a nitro group and poor physicochemical properties, such as high lipophilicity and low water solubility, required significant modifications to transform mebudipine into a viable MR antagonist lead. Herein, we report the discovery and optimization of novel cyano DHPs that led to identification of potent and selective MR antagonists. Compound **6d** was advanced to in vivo studies using the Dahl salt-sensitive (SS) rat model of hypertension[13] where it provided proof of concept (POC) for a nonsteroidal, orally efficacious MR antagonist.

## Results and Discussion

**Chemistry.** A library of dihydropyridine diesters **4a**−**t** was prepared by a modified Hantzsch reaction as shown in Scheme 1.[14] Condensation of *tert*-butyl oxobutanoate with (*Z*)-methyl 3-aminobut-2-enoate and the corresponding benzaldehyde in the presence of piperidine afforded the desired dihydropyridines in acceptable yields.

Cyano ester DHPs **5a**−**v** were prepared by a similar procedure as described above from 3-aminobutenenitrile as shown in Scheme 2. Chiral separation using supercritical fluid chromatography (SFC) on an asymmetric resin with a mobile phase containing a mixture of ethanol/$CO_2$ afforded the corresponding enantiomers. Chloromethyl DHP **6**[11] was used as an advanced intermediate to obtain $R_6$ substituted analogues **7a**−**d** via nucleophilic displacement of the chloro group with the desired heterocycle in the presence of potassium carbonate in NMP, as shown in Scheme 2.

4,4-Disubstituted DHP **5c** was prepared in three steps from pyridine **8** by intramolecular addition of sulfonyl carbanion to generate intermediate **9**, as shown in Scheme 3.[15]

DHPs **5d** and **5s** were further derivatized as shown in Scheme 4. The sodium salt of **5d** was alkylated with bromoethane and 1-bromo-2-methoxyethane to give **5e** and **5f**, respectively. The sodium salt of **5s** in turn was alkylated with iodomethane to give **5t**.

## Results

MR antagonist potency was evaluated in a cellular assay based on aldosterone-induced activation of a luciferase reporter driven by MR ligand binding domain fused to a heterologous DNA binding domain from yeast transcription factor Gal4.[16] Initial structure−activity relationship (SAR) studies consisted of a library focused on 4-aryl substituents to identify preferred groups for the replacement of the nitro group in **3**. Selected examples are shown in Table 1. Small nonpolar substituents, e.g., F, Cl, and $CF_3$, were well tolerated at the $R_1$ (**4a**−**c**) and $R_3$ positions (**4m**, **4o**, **4r**). Disubstitution with Cl at $R_1$ and F at $R_3$ was found to be optimal (**4s**).

The same general SAR for 4-aryl substituents in the diester series was observed in the cyano *tert*-butyl ester series where disubstitution with halogens (e.g., **5a** and **5d**) produced the most potent analogues, as shown in Table 2. Substitution at the nitrogen in the DHP ring (**5e**, **5f**, **5t**) was poorly tolerated, suggesting a potential H-bond interaction of the DHP NH group with the receptor. Replacement of the *tert*-butyl ester by methyl ester in **5g** did not result in significant changes in potency.

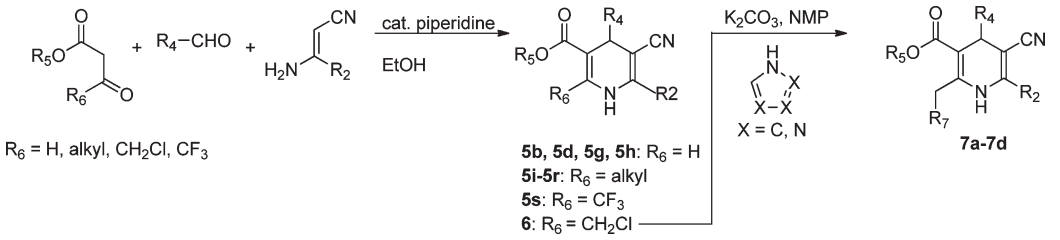
Substitution of the methyl group at the $R_6$ position by phenyl or benzyl groups was detrimental to potency (**5n**, **5o**, **5p**). However, substitution with smaller $C_2$−$C_4$ alkyl groups yielded more potent compounds (**5i**−**l**), with the gains in potency reaching a plateau for propyl and butyl. The $CF_3$ group was also well-tolerated at this position (**5s**). An initial assessment on the MR selectivity for a select group of potent analogues against other steroidal nuclear hormone receptors was determined in a similar Gal4 cellular assay format (Table 3). In general, cyano DHPs exhibit excellent selectivity against the estrogen receptor (ER) (data not shown). They exhibit good selectivity against the glucocorticoid receptor (GR), while selectivity against the progesterone receptor (PR) and the androgen receptor (AR) tends to be more compound dependent. Compound **5s** was found to be especially selective across all other nuclear hormone receptors (NHRs) tested.

Despite the significant gains in potency for this series with respect to the corresponding diester DHPs, the analogues had high lipophilicity and thus suffered from poor solubility and in vitro metabolic stability as shown in Table 3.

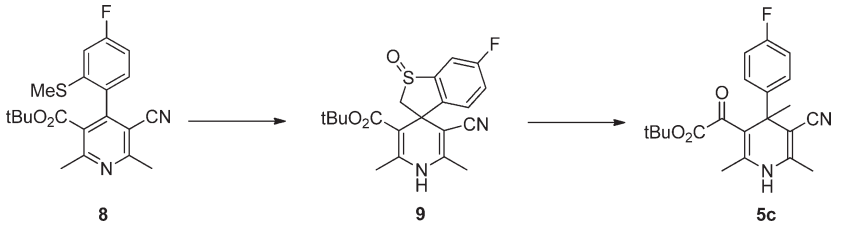
Replacement of alkyl groups at $R_6$ by a methylene linker tethering five-membered ring heterocycles resulted in analogues that maintained excellent MR potency and showed improved selectivity against the other NHRs, as shown in Table 4. Although imidazole **7a** and triazole **7d** showed improved rat microsomal stability (RLM), they were potent CYP inhibitors. In contrast, tetrazole **7d** showed improved in vitro metabolic stability and solubility without CYP inhibition liability. On the basis of its MR potency and favorable in vitro pharmacokinetic profile, compound **7d** was advanced for rat pharmacokinetic (PK) studies. Its moderate clearance and good half-life, summarized in Table 5, made **7d** a suitable candidate for in vivo efficacy studies.

The effect of **7d** on blood pressure (BP) and kidney injury was tested in male Dahl SS rats fed with 4% NaCl. Rats were

**Scheme 2.** General Synthesis of Cyano Ester DHPs

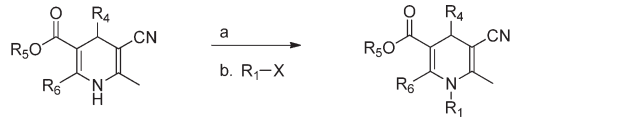



**Scheme 3.** Synthesis of DHP **5c**[a]




[a] (a) 30% $H_2O_2$, hexfluoro-2-propanol, 2 h; (b) LDA, THF, −78 °C; (c) Raney Ni, EtOH, 90 C, 30 min.

**Scheme 4.** Synthesis of DHPs **5e**−**f** and **5t**[a]




**5d**: $R_4$ = 2,4-diFPh, $R_5$ = tBu, $R_6$ = Me
**5s**: $R_4$ = 2-Cl, 4-FPh, $R_5$ = Me, $R_6$ = $CF_3$

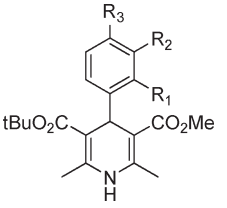
**5e**: $R_1$ = Me, $R_4$ = 2,4-diFPh, $R_5$ = tBu, $R_6$ = Me
**5f**: $R_1$ = $CH_2CH_2OMe$, $R_4$ = 2,4-diFPh, $R_5$ = tBu, $R_6$ = Me
**5t**: $R_1$ = Me, $R_4$ = 2-Cl, 4-FPh, $R_5$ = Me, $R_6$ = $CF_3$

[a] (a) NaH, DMA, room temp, 4 h; (b) **10**, room temp, overnight.

dosed orally via gavage with **7d** in vehicle at 10 and 60 (mg/kg)/d, twice a day. For comparison, **1** was dosed in chow at 100 mpk/d because of its very short half-life in rats. Treatment was initiated at the beginning of salt feeding, and BP was monitored using telemetry units. The results are shown in Figure 3. After 21 days of salt feeding, animals treated with vehicle showed a dramatic increase (40−50 mmHg) in BP (mean 24 h SBP), typical of Dahl SS rats. As has been demonstrated previously, BP increase was much lower in the group fed with **1**, which represented a significant BP reduction compared to the vehicle group. At 60 (mg/kg)/d b.i.d. **7d** caused BP reduction approaching the effect by **1**, demonstrating POC for this class of nonsteroidal MR antagonist. The PK of **7d**, its short half-life and low bioavailability, needs to be further optimized to achieve the maximum BP lowering effect of eplerenone. Dahl SS rats also develop kidney damage after high salt feeding as evidenced by increased urinary albumin. Both eplerenone and 60 (mg/kg)/d **7d** achieved comparable reduction of albumin levels, although statistical significance was not achieved because of small sample size (data not shown). Together, these results demonstrated that the DHP class MR antagonist is capable of reducing BP and renal injury in vivo.

Chiral resolution of **7d** by supercritical fluid chromatography (SFC) afforded only one MR-active enantiomer, assigned as (*R*)-**7d** based on our previous studies.[11] Intermediate (*R*)-**6** was also obtained by SFC, as previously described, and used in the preparation of (*R*)-**7d** to confirm its configuration assignment.

**Table 1.** Initial Screening Results for Diester DHPs[a]



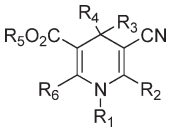

| compd | $R_1$ | $R_2$ | $R_3$ | yield (%) | MR $IC_{50}$ (nM) |
|---|---|---|---|---|---|
| **1** | | | | | 122 |
| **3** | H | $NO_2$ | H | | 126 |
| **4a** | Cl | H | H | 22 | 621 |
| **4b** | F | H | H | 11 | 2020 |
| **4c** | $CF_3$ | H | H | 8 | 224 |
| **4d** | Me | H | H | 19 | 208 |
| **4e** | OMe | H | H | 17 | 553 |
| **4f** | H | Me | H | 41 | 2790 |
| **4g** | H | CN | H | 26 | 3200 |
| **4h** | H | OMe | H | 27 | 888 |
| **4i** | H | Cl | H | 30 | 518 |
| **4j** | H | $CF_3$ | H | 19 | 702 |
| **4k** | H | OH | H | 24 | 2830 |
| **4l** | H | F | H | 38 | 1390 |
| **4m** | H | H | $CF_3$ | 17 | 309 |
| **4n** | H | H | OH | 35 | 2570 |
| **4o** | H | H | F | 25 | 288 |
| **4p** | H | H | Me | 24 | 12010 |
| **4q** | H | H | OMe | 19 | 2620 |
| **4r** | H | H | Cl | 24 | 319 |
| **4s** | Cl | H | F | 44 | 78 |
| **4t** | H | F | Cl | 28 | 574 |

[a] $IC_{50}$ values were obtained through curve-fitting of dose response ($n \geq 3$/concentration, 6−10 concentrations) using the four-parameter logistic model. Standard error of the $IC_{50}$ was generally less than 30%.

(*R*)-**7d** showed a similar in vitro metabolic profile and similar solubility to the racemic compound (see Table 4).

Induced-fit docking[17] studies suggest that (*R*)-**7d** can adopt the previously proposed DHP MR binding pose[10,11] (Figure 2). In this pose, the 4-aryl group occupies the A-ring pocket of the

**Table 2.** Cyano Ester DHP SAR[a]




| compd | $R_1$ | $R_2$ | $R_3$ | $R_4$ | $R_5$ | $R_6$ | MR $IC_{50}$ (nM) |
|---|---|---|---|---|---|---|---|
| **5a** | H | Me | H | 2-chloro-4-fluorophenyl | $^t$Bu | Me | 59 |
| **5b** | H | Me | H | 4-fluorophenyl | $^t$Bu | Me | 329 |
| **5c** | H | Me | Me | 4-fluorophenyl | $^t$Bu | Me | 661 |
| **5d** | H | Me | H | 2,4-difluorophenyl | $^t$Bu | Me | 201 |
| **5e** | Et | Me | H | 2,4-difluorophenyl | $^t$Bu | Me | 1620 |
| **5f** | $(CH_2)_2OMe$ | Me | H | 2,4-difluorophenyl | $^t$Bu | Me | 3740 |
| **5g** | H | Me | H | 2-chloro-4-fluorophenyl | Me | Me | 92 |
| **5h** | H | Me | H | 2-chlorophenyl | Me | Me | 594 |
| **5i** | H | Me | H | 2-chlorophenyl | Me | Et | 232 |
| **5j** | H | Me | H | 2-chlorophenyl | Me | Pr | 64 |
| **5k** | H | Me | H | 2-chlorophenyl | Me | $^i$Pr | 208 |
| **5l** | H | Me | H | 2-chlorophenyl | Me | Bu | 58 |
| **5m** | H | Me | H | 2-chlorophenyl | Me | $CH_2OCH_3$ | 655 |
| **5n** | H | Me | H | 2-chlorophenyl | Et | Ph | 923 |
| **5o** | H | Me | H | 2-chlorophenyl | Et | 4-ClPh | 1090 |
| **5p** | H | Me | H | 2-chlorophenyl | Me | Bn | 852 |
| **5q** | H | Me | H | phenyl | Me | Pr | 7430 |
| **5r** | H | Me | H | 2-chloro-4-fluorophenyl | Me | Pr | 16 |
| **5s** | H | Me | H | 2-chloro-4-fluorophenyl | Me | $CF_3$ | 52 |
| **5t** | Me | Me | H | 2-chloro-4-fluorophenyl | Me | $CF_3$ | 275 |

[a] $IC_{50}$ values were obtained through curve-fitting of dose response ($n \geq 3$/concentration, 6−10 concentrations) using the four-parameter logistic model. Standard error of the $IC_{50}$ was generally less than 30%.

**Table 3.** NHR Selectivity and in Vitro PK Properties of Select 2,6-Dimethyl Cyano Ester DHPs[a]

| compd | MR $IC_{50}$ (nM) | GR $IC_{50}$ ($\mu$M) | AR $IC_{50}$ ($\mu$M) | PR $IC_{50}$ ($\mu$M) | HLM % rem | RLM % rem | CYP 3A4 % inh | CYP 2C9 % inh | CYP 2D 6% inh | solubility ($\mu$M) |
|---|---|---|---|---|---|---|---|---|---|---|
| **5g** | 92 | 5.1 | 0.3 | 0.9 | 37 | 14 | 40 | 44 | 0 | 10 |
| **5j** | 64 | 1.6 | 0.3 | 0.2 | 36 | 13 | 71 | 83 | 19 | < 5 |
| **5l** | 58 | 1.6 | 0.9 | 0.3 | 11 | 3 | | | | < 5 |
| **5r** | 16 | 0.7 | 0.1 | > 10.0 | 4 | 4 | | | | 5 |
| **5s** | 52 | 3.7 | 0.8 | 7.8 | 0 | 6 | 30 | 72 | 6.8 | |

[a] $IC_{50}$ values were obtained through curve-fitting of dose response ($n \geq 3$/concentration, 6−10 concentrations) using the four-parameter logistic model. Standard error of the $IC_{50}$ was generally less than 30%.

receptor, making a face−edge (tee) interaction with Phe 829, while the DHP ring plane is approximately orthogonal to the typical steroidal plane and the DHP NH donates a hydrogen bond to Asn 770. In this orientation, the ester is accommodated in the hydrophobic α-face pocket, and the 2-methyl and 3-cyano groups protrude toward the $\beta$ face, either directly impinging on Leu 960 from helix 12 or doing so indirectly by perturbing Trp 806. Although no explicit hydrogen bonds are formed between the cyano nitrogen and the hydroxyl of Ser 810 in this type of pose, they are typically about 3.5 Å apart and an interaction cannot be ruled out. Addition of the pendent tetrazole is readily accommodated, occupying a position similar to that of the steroid C20 side chain, with the potential for accepting hydrogen bonds from Asn 770 or Thr 945 depending on the particular pose. However, the DHP fails to overlap the entire steroidal D-ring and much of the steroidal C-ring volume (Figure 2b). Incomplete occupation/stabilization of the receptor binding pocket[18] and steric clashes induced by DHP binding are both possible mechanisms for antagonism.

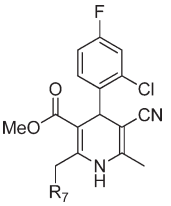
## Conclusions

We have designed novel nonsteroidal MR antagonists by modifying marketed 1,4-dihydropyridine CCBs, such as nifedipine and mebudipine. Replacement of one ester group by cyano, rendering the MR antagonists devoid of CCB activity, followed by optimization of the dihydropyridine core substituents resulted in the identification of lead compounds **5j**, **5g**, **5r**, and **5s**, which showed comparable in vitro MR antagonist potency to steroid **1** and good selectivity against other NHRs. However, in vitro PK data indicated these compounds did not possess adequate metabolic stability to be studied in vivo. Appending five-membered heterocycles at the $R_6$ position resulted in further improvements in the physicochemical properties of the analogues while maintaining excellent potency and selectivity for MR. Compound **7d** was advanced to in vivo studies using the Dahl SS rat model of hypertension where it provided POC for a nonsteroidal, orally active MR antagonist.

## Experimental Section

All materials were obtained from commercial sources and used as purchased. Chromatography solvents were HPLC grade and were used without further purification. Thin layer chromatography (TLC) analysis was performed using Merck silica gel 60 F-254 thin layer plates. LC−MS analyses were performed on Mariner TOF from Perseptive Biosystems. The scan range was $m/z$

**Table 4.** Cyano DHPs Containing Five-Membered Ring Heterocycles[a]




| Example | R7 | MR IC50 (nM) | GR IC50 (uM) | AR IC50 (uM) | PR IC50 (uM) | HLM %rem | RLM %rem | 3A4 IC50 (uM) | 2C9 IC50 (uM) | 2D6 IC50 (uM) | Sol. (uM) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **7a** |  | 9 | 2.2 | 3.9 | >10.0 | 8 | 64 | 0.2 | 1 | >10 | 5 |
| **7b** |  | 54 | 3.3 | 0.8 | 5.5 | 2 | 31 | 1.1 | 0.6 | >10 | <5 |
| **7c** |  | 123 | 2.9 | 4.6 | 7.0 | 9 | 34 | 0.8 | 0.1 | >10 | <5 |
| **7d** |  | 64 | 1.3 | 2.7 | >10.0 | 3 | 60 | 1.2 | 1.9 | >10 | 31 |
| ***(R)*-7d** |  | 52 | 1.0 | 0.3 | 7.2 | -- | * | >10 | >10 | >10 | 32 |

[a] $IC_{50}$ values were obtained through curve-fitting of dose response ($n \geq 3$/concentration, 6−10 concentrations) using the four-parameter logistic model. Standard error of the $IC_{50}$ was generally less than 30%. (∗) Determined as CL intrinsic 8.71 mL/(kg·min).

**Table 5.** Rat Pharmacokinetic Data for **7d**[a]

| compd | dose (mpk) | CL ((mL/min)/kg) | $T_{1/2}$ (h) | $V_{dss}$ (L·kg) |
|---|---|---|---|---|
| **7d** | 5 | 14.1 | 4.84 | 0.9 |

[a] Male Sprague−Dawley rats ($n = 2$) dosed intravenously (iv) at 5 mg/kg. Compound **7d** was formulated for iv dosing in 10% ethanol/40% PEG400/50% phosphate buffered saline, pH 7.4. Plasma samples were analyzed by LC/MS/MS.

100−1000. The sample was introduced by flow inject from an Agilent 1100 with 100 μL/min MeOH (10 mM ammonium acetate) into the electrospray source. Preparative reverse phase HPLC was performed on a Gilson 215 liquid handler equipped with a Dynamax Microsorb C18 (300 Å) column (41.4 mm × 25 cm, 8 μm) and Gilson 156 variable length UV detector. The purity of tested compounds was >95% as determined by combustion analysis or by HPLC conducted on an Agilent 1100 system using a reverse phase C8 column with diode array detector. NMR spectra were recorded on a Bruker 400 spectrometer. The signal of the deuterated solvent was used as internal reference. Chemical shifts ($\delta$) are given in ppm and are referenced to residual not fully deuterated solvent signal. Coupling constants ($J$) are given in Hz.

**General Procedure for the Preparation of Diester DHPs 4a−t.** To a solution of methyl 3-aminocrotonate (4.8 mmol) in MeOH (5 mL) in a 4 dram vial was added *tert*-butyl acetoacetate (1 equiv), the relevant aldehyde (1 equiv), and piperidine (1 drop). The reaction mixture was stirred overnight at 65 °C. The solvent was evaporated under a stream of nitrogen and the residue was dissolved in DMSO and purified by reverse phase HPLC (ACN/water with 0.1%TFA, 20−75% ACN) to yield the desired diester DHP.

**General Procedure for the Preparation of Cyano DHPs.** To a solution of the appropriate aldehyde (4 mmol) in THF (10 mL) was added the acetoacetate (4 mmol) followed by 3 drops of piperidine. The mixture was stirred at reflux under nitrogen atmosphere for 4 h. The solvent was concentrated in vacuo, and the residue was dissolved in ethanol (10 mL). 3-Aminocrotononitrile (4 mmol) was added to the solution, and the reaction mixture was stirred at reflux under nitrogen overnight. Solvent was evaporated in vacuo and the resulting crude product was purified by reverse phase HPLC (ACN/water with 0.1%TFA, 35−95% ACN) to give the desired cyano DHP.

***tert*-Butyl 5-Cyano-4-(4-fluorophenyl)-2,6-dimethyl-1,4-dihydropyridine-3-carboxylate 5b.** This compound was prepared by the general method with 4-fluorobenzaldehyde and *tert*-butyl acetoacetate as starting materials. Yield 37%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 9.06 (s, 1 H), 7.11−7.21 (m, 4 H), 4.41 (s, 1 H), 2.71 (s, 1 H), 2.56 (s, 1 H), 2.23 (s, 3 H), 1.99 (s, 3 H), 1.46 (s, 1 H), 1.22 (s, 11 H); ES-MS $m/z$ 329 (M + H).

***tert*-Butyl 5-Cyano-4-(4-fluoro-2-(methylthio)phenyl)-2,6-dimethylnicotinate 8.** 4-Fluoro-2-methylthiobenzaldehyde and *tert*-butyl acetoacetate were reacted according the general procedure for the preparation of cyano DHPs to give the *tert*-butyl 5-cyano-4-(4-fluoro-2-(methylthio)phenyl)-2,6-dimethyl-1,4-dihydropyridine-3-carboxylate intermediate. Yield 72%. To a solution of *tert*-butyl 5-cyano-4-(4-fluoro-2-(methylthio)phenyl)-2,6-dimethyl-1,4-dihydropyridine-3-carboxylate (400 mg, 1.1 mmol) in toluene (5 mL) in a capped vial was added chloranil (330 mg, 1.3 mmol). The reaction mixture was heated at 120 °C for 18 h and then cooled to room temperature. A precipitate developed and was filtered off. The solid was purified on silica gel

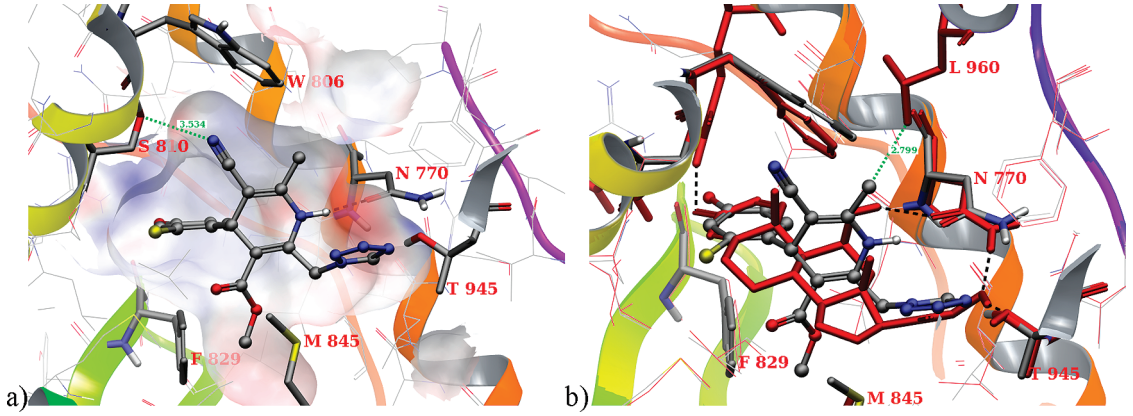



**Figure 2.** (a) Representative induced-fit docking pose for tetrazole (*R*)-**7d**. (b) Overlay of induced-fit docking pose (gray) with the native corticosterone-MR crystal structure 2A3I (red). Steroid hydrogen bonding in the native steroid structure is shown. Movement of W806 induced by DHP is shown, as is close contact with L960 by the 2-methyl group.

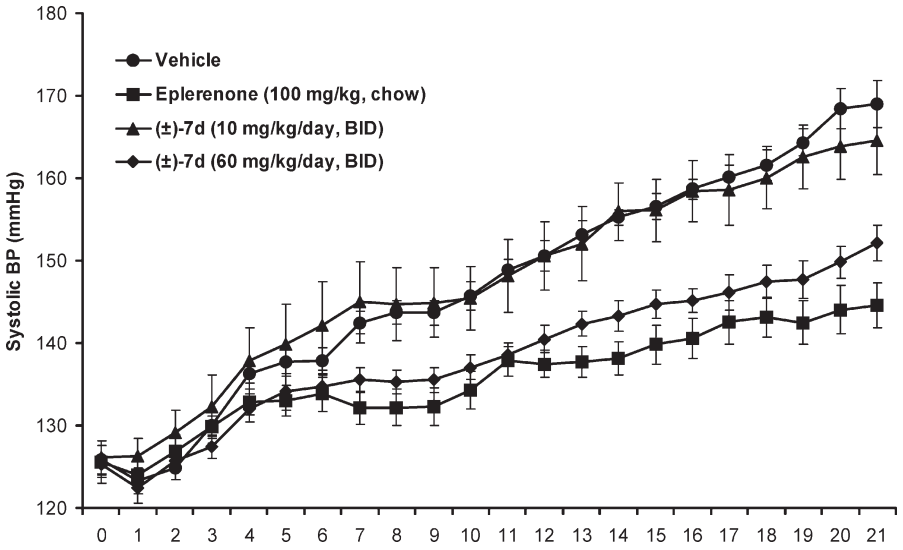



**Figure 3.** Blood pressure lowering effect of **7d** in Dahl SS rats. Radiotelemetrized arterial systolic blood pressure (SBP) was measured with the DATAQUEST A.R.T., version 3.0, Gold software. The values represent the average of all data points collected from each animal every 15 min for a 10 s interval over a 24 h period (6:00 a.m. to 6:00 a.m. the following day). SBP data were collected continuously over the course of the entire study (days 1−21). Blood was drawn at day 5 and day 16 for PK analysis. $N = 6$ for the 100 mpk group, and $N = 9$ for all other groups.

(dichloromethane 100%) to give pure **8**. Yield 75%; HRMS M + H calcd for $C_{20}H_{22}FN_2O_2S$, 373.1381; obsd, 373.1358.

***tert*-Butyl 5-Cyano-2,4-dimethyl-4-spiro-3′-(5′-fluoro-2′,3′-dihydro-1′-benzothiophene 1′-oxide)-1,4-dihydropyridine-3-carboxylate 9. Step 1.** To a stirred solution of pyridine **8** (147 mg) in hexafluoro-2-propanol (2 mL) was added 30% hydrogen peroxide (0.6 mmol), and the mixture was stirred at room temperature. After 2 h the reaction was quenched with a saturated solution of potassium metabisulfite and the mixture was concentrated under a stream of nitrogen. The residue was filtered through a short column of silica gel, eluting with methanol to give crude *tert*-butyl 5-cyano-4-(4-fluoro-2-(methylsulfinyl)phenyl)-2,6-dimethylnicotinate (85%). ES-MS *m*/*z* 389 (M + H).

**Step 2.** A solution of LDA (0.5 mmol) prepared from diisopropylamine and *n*-BuLi in THF was added to a solution of *tert*-butyl 5-cyano-4-(4-fluoro-2-(methylsulfinyl)phenyl)-2,6-dimethylnicotinate (130 mg) in THF (5 mL) cooled at −78 °C. Reaction was quenched with saturated $NH_4Cl$ and the resulting solid was washed with water to give **9** (50%). ES-MS *m*/*z* 387 (M + H).

***tert*-Butyl 5-Cyano-4-(4-fluorophenyl)-2,4,6-trimethyl-1,4-dihydropyridine-3-carboxylate 5c.** To a solution of intermediate **9** (25 mg) in ethanol (3 mL) was added Raney Ni (slurry, 200 μL), and the mixture was heated at 90 °C for 30 min. The mixture was cooled and filtered. The solid was rinsed with hot ethanol, and the combined ethanol solution was concentrated under a $N_2$ stream. The residue was chromatographed on silica gel (hexanes/EtOAc) to yield **5c** (60%). $^1$H NMR (400 MHz, chloroform-*d*) δ ppm 7.36 (dd, $J = 8.86, 5.37$ Hz, 2 H), 7.25 (s, 1 H), 6.99 (t, $J = 8.73$ Hz, 2 H), 2.15 (s, 3 H), 2.04 (s, 3 H), 1.83 (s, 4 H), 1.54 (s, 2 H), 1.09 (s, 10 H), 1.05 (s, 1 H); ES-MS *m*/*z* 343 (M + H).

***tert*-Butyl 5-Cyano-4-(2,4-difluorophenyl)-2,6-dimethyl-1,4-dihydropyridine-3-carboxylate 5d.** This compound was prepared by the general method with 2,4-difluorobenzaldehyde and *tert*-butyl acetoacetate as starting materials. Yield 72%; $^1$H NMR (400 MHz, DMSO-$d_6$) δ ppm 9.08 (s, 1 H), 7.20 (td, $J = 8.59$, 6.71 Hz, 1 H), 7.15 (td, $J = 9.87$, 2.55 Hz, 1 H), 7.03 (td, $J = 8.53$, 2.55 Hz, 1 H), 4.73 (s, 1 H), 2.21 (s, 3 H), 1.94 (s, 3 H), 1.16 (s, 9 H); ES-MS *m*/*z* 347 (M + H).

***tert*-Butyl 5-Cyano-4-(2,4-difluorophenyl)-1-ethyl-2,6-dimethyl-1,4-dihydropyridine-3-carboxylate 5e.** To a solution of **5d** (16.6 g) in DMA (46 mL) was added sodium hydride (60%, 1.2 equiv), and the solution was stirred at room temperature for 4 h. An aliquot of the solution (0.58 mL) was placed in a vial under nitrogen, and bromoethane (2.5 equiv) was added. The reaction mixture was stirred at room temperature overnight. The solution was

purified by reverse phase HPLC (ACN/water with 0.1%TFA, 25−95% ACN) to give pure **5e** (84%). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 7.19 (dt, $J$ = 8.59, 4.30 Hz, 2 H), 4.76 (s, 1 H), 3.72 (s, 1 H), 3.66 (s, 1 H), 2.42 (s, 3 H), 2.19 (s, 3 H), 1.23 (s, 9 H), 1.15 (t, $J$ = 7.12 Hz, 3 H); ES-MS $m/z$ 375 (M + H).

***tert*-Butyl 5-Cyano-4-(2,4-difluorophenyl)-1-(2-methoxyethyl)-2,6-dimethyl-1,4-dihydropyridine-3-carboxylate 5f. 5f** was prepared as example **5e** using 1-bromo-2-methoxyethane (2.5 equiv) in place of bromoethane. Yield 31%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 7.26 (td, $J$ = 8.59, 6.71 Hz, 1 H), 7.19 (td, $J$ = 9.87, 2.55 Hz, 1 H), 7.00−7.12 (m, 1 H), 4.76 (s, 1 H), 3.87−4.09 (m, 1 H), 3.72−3.87 (m, 1 H), 2.40 (s, 3 H), 2.15 (s, 3 H), 1.24 (s, 9 H), signal at 3.45 blocked for water peak. HRMS M + H calcd for $C_{22}H_{27}F2N_2O_2$, 406.2016; obsd 406.2049.

**Methyl 4-(2-Chloro-4-fluorophenyl)-5-cyano-1,4-dihydro-2,6-dimethylpyridine-3-carboxylate 5g.** This compound was prepared by the general method with 2-chloro-4-fluorobenzaldehyde and methyl acetoacetate as starting materials. Yield 56%; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ ppm 1.98 (s, 3 H), 2.28 (s, 3 H), 3.42 (s, 3 H), 5.02 (s, 1 H), 7.19 (m, 1 H), 7.31 (m, 2 H), 9.26 (s, 1 H); ES-MS $m/z$ 321 (M + H).

**Methyl 4-(2-Chlorophenyl)-5-cyano-1,4-dihydro-2,6-dimethylpyridine-3-carboxylate 5h.** This compound was prepared by the general method with 2-chlorobenzaldehyde and methyl acetoacetate as starting materials. Yield 60%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 1.98 (s, 3 H), 2.28 (s, 3 H), 3.41 (s, 3 H), 5.05 (s, 1 H), 7.20 (m, 1 H), 7.26 (dd, $J$ = 7.79, 2.15 Hz, 1 H), 7.31 (m, 1 H), 7.36 (dd, $J$ = 7.79, 1.34 Hz, 1 H), 9.22 (s, 1 H). HRMS M + H calcd for $C_{16}H_{16}ClN_2O_2$, 303.0895; obsd 303.0902.

**Methyl 4-(2-Chlorophenyl)-5-cyano-2-ethyl-1,4-dihydro-6-methylpyridine-3-carboxylate 5i.** This compound was prepared by the general method with 2-chlorobenzaldehyde and methyl propionylacetate. Yield 57%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 1.14 (t, $J$ = 7.38 Hz, 3 H), 1.98 (s, 3 H), 2.62 (m, 1 H), 2.72 (m, 1 H), 3.41 (s, 3 H), 5.05 (s, 1 H), 7.21 (m, $J$ = 7.79, 1.88 Hz, 1 H), 7.24 (dd, $J$ = 7.79, 1.88 Hz, 1 H), 7.30 (m, 1 H), 7.36 (dd, $J$ = 8.06, 1.34 Hz, 1 H), 9.21 (s, 1 H). HRMS M + H calcd for $C_{17}H_{18}ClN_2O_2$, 317.1051; obsd 317.1010. Anal. ($C_{17}H_{18}ClN_2O_2$) C, H, N.

**Methyl 4-(2-Chlorophenyl)-5-cyano-1,4-dihydro-6-methyl-2-propylpyridine-3-carboxylate 5j.** This compound was prepared by the general method with 2-chlorobenzaldehyde and methyl butyrylacetate. Yield 50%; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ ppm 0.94 (t, $J$ = 7.35 Hz, 3 H), 1.57 (m, 2 H), 1.98 (s, 3 H), 2.57 (m, 1 H), 2.72 (m, 1 H), 3.41 (s, 3 H), 5.05 (s, 1 H), 7.27 (m, 4 H), 9.19 (s, 1 H); ES-MS $m/z$ 331 (M + H).

**Methyl 4-(2-Chlorophenyl)-5-cyano-1,4-dihydro-2-isopropyl-6-methylpyridine-3-carboxylate 5k.** This coumpound was prepared by the general method with 2-chloro-4-fluorobenzaldehyde and methyl acetoacetate. Yield 52%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 1.13 (d, $J$ = 6.98 Hz, 3 H), 1.20 (d, $J$ = 6.98 Hz, 3 H), 2.04 (s, 3 H), 3.41 (s, 3 H), 4.08 (m, 1 H), 5.04 (s, 1 H), 7.21 (m, 2 H), 7.32 (m, 1 H), 7.36 (m, 1 H), 8.60 (s, 1 H). HRMS M + H calcd for $C_{18}H_{20}ClN_2O_2$, 331.1208; obsd, 331.1192.

**Methyl 2-Butyl-4-(2-chlorophenyl)-5-cyano-1,4-dihydro-6-methylpyridine-3-carboxylate 5l.** This compound was prepared by the general procedure with 2-chlorobenzaldehyde and 3-oxoheptanoic acid methyl ester. Yield 40%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.91 (t, $J$ = 7.25 Hz, 3 H), 1.36 (m, 2 H), 1.53 (m, 2 H), 1.98 (s, 3 H), 2.58 (m, 1 H), 2.75 (m, 1 H), 3.41 (s, 3 H), 5.05 (s, 1 H), 7.22 (m, 2 H), 7.30 (m, 1 H), 7.36 (m, 1 H), 9.19 (s, 1 H). HRMS M + H calcd for $C_{19}H_{22}ClN_2O_2$, 345.1364; obsd, 345.1339. Anal. ($C_{19}H_{22}ClN_2O_2$) C, H, N.

**Methyl 4-(2-Chlorophenyl)-5-cyano-1,4-dihydro-2-(2-methoxymethyl)-6-methylpyridine-3-carboxylate 5m.** This compound was prepared by the general procedure with 2-chlorobenzaldehyde and methyl 4-methoxyacetoacetate. Yield 30%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.03 (s, 3 H), 3.34 (s, 3 H), 3.43 (s, 3 H), 4.55 (s, 2 H), 5.08 (s, 1 H), 7.29 (m, 4 H), 9.07 (s, 1 H). HRMS M + H calcd for $C_{17}H_{18}ClN_2O_3$, 333.1000; obsd, 333.1005.

**Ethyl 4-(2-Chlorophenyl)-5-cyano-2-methyl-6-phenyl-1,4-dihydropyridine-3-carboxylate 5n.** This compound was prepared by the general procedure with 2-chlorobenzaldehyde and ethyl benzoylacetate. Yield 82%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.66 (t, $J$ = 7.03 Hz, 3 H), 2.01 (s, 3 H), 3.61 (qd, $J$ = 7.03, 3.12 Hz, 2 H), 5.18 (s, 1H), 7.27 (dt, $J$ = 7.52, 3.86 Hz, 2 H), 7.36 (m, 2 H), 7.44 (m, 4 H), 7.50 (dd, $J$ = 7.81, 1.56 Hz, 1 H), 9.39 (s 1 H); ES-MS $m/z$ 379 (M + H).

**Ethyl 4-(2-Chlorophenyl)-6-(4-chlorophenyl)-5-cyano-2-ethyl-1,4-dihydropyridine-3-carboxylate 5o.** This compound was prepared by the general procedure with 2-chlorobenzaldehyde and ethyl 4-chlorobenzoylacetate. Yield 80%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.70 (t, $J$ = 7.03 Hz, 3 H), 2.00 (s, 3 H), 3.63 (dd, $J$ = 7.23, 2.15 Hz, 2 H), 5.18 (s, 1 H), 7.27 (dd, $J$ = 7.82, 1.95 Hz, 1 H), 7.41 (d, $J$ = 7.82 Hz, 1 H), 7.37−7.42 (m, 3 H), 7.50 (d, $J$ = 8.60 Hz, 2 H), 7.48−7.51 (m, 1 H); ES-MS $m/z$ 413 (M + H).

**Methyl 2-Benzyl-4-(2-chlorophenyl)-5-cyano-1,4-dihydro-6-methylpyridine-3-carboxylate 5p.** This compound was prepared by the general method with 2-chlorobenzaldehyde and methyl 3-oxo-4-phenylbutanoate. Yield 50%; $^1$H NMR (300 MHz, DMSO-$d_6$) $\delta$ ppm 1.99 (s, 3 H), 3.40 (s, 3 H), 4.01 (d, $J$ = 14.10 Hz, 1 H), 4.16 (m, 1 H), 5.13 (s, 1 H), 7.27 (m, 9 H), 9.37 (s, 1 H); ES-MS $m/z$ 379 (M + H).

**Methyl 5-Cyano-1,4-dihydro-6-methyl-4-phenyl-2-propylpyridine-3-carboxylate 5q.** This compound was prepared by the general method with benzaldehyde and methyl butyrylacetate. Yield 54%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.91 (m, 3 H), 1.70 (m, 2 H), 2.73 (m, 2 H), 2.73 (s, 3 H), 3.52 (s, 3 H), 4.44 (s, 1 H), 7.37 (m, 2 H), 7.52 (m, 3 H), 9.16 (m, 1 H); ES-MS $m/z$ 297 (M + H).

**Methyl 4-(2-Chloro-4-fluorophenyl)-5-cyano-1,4-dihydro-6-methyl-2-propylpyridine-3-carboxylate 5r.** This compound was prepared by the general method with 2-chloro-4-fluorobenzaldehyde and methyl butyrylacetate. Yield 70%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 0.94 (t, $J$ = 7.38 Hz, 3 H), 1.56 (m, 2 H), 1.98 (s, 3 H), 2.56 (m, 1 H), 2.71 (m, 1 H), 3.42 (s, 3 H), 5.03 (s, 1 H), 7.20 (m, 1 H), 7.27 (m, 1 H), 7.34 (dd, $J$ = 9.00, 2.55 Hz, 1 H), 9.22 (s, 1 H); ES-MS $m/z$ 349 (M + H).

**Methyl 4-(2-Chloro-4-fluorophenyl)-5-cyano-6-methyl-2-(trifluoromethyl)-1,4-dihydropyridine-3-carboxylate 5s.** To a solution of 2-chlorobenzaldehyde (0.215 g, 1.53 mmol) in THF (10 mL) was added methyl trifluoroacetoacetate (0.236 g, 1.39 mmol) followed by piperidine (3 drops), and the reaction mixture was stirred at reflux under nitrogen. After 4 h the solution was concentrated in vacuo to yield a solid. This crude product was dissolved in EtOH (10 mL), and 3-aminocrotononitrile (0.114 g, 1.39 mmol) was added. The reaction mixture was stirred at reflux under nitrogen overnight. The solvent was evaporated and the resulting product was purified by reverse phase HPLC (ACN/water with 0.1%TFA, 35−95% ACN) to give a solid. Solid was dissolved in acetonitrile (1 mL) and 4 N HCl in dioxane (1 mL), and the mixture was stirred at 55 °C for 18 h. The solvent was removed and the crude product was purified by reverse phase HPLC to give the title compound. Yield 72%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.08 (s, 3 H), 3.50 (s, 3 H), 5.12 (s, 1 H), 7.28 (m, 2 H), 7.41 (m, 2 H), 9.68 (s, 1 H); HRMS M + H calcd for $C_{16}H_{11}F_4ClN_2O_2NH_4$, 392.0783; obsd, 392.0820.

**Methyl 4-(2-Chloro-4-fluorophenyl)-5-cyano-1,6-dimethyl-2-(trifluoromethyl)-1,4-dihydropyridine-3-carboxylate 5t.** This compound was prepared by the same procedure described for **5e** from **5s** and iodomethane. Yield 40%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 7.48 (dd, $J$ = 8.86, 2.69 Hz, 1 H), 7.42 (dd, $J$ = 8.59, 6.18 Hz, 1 H), 7.29 (td, $J$ = 8.46, 2.69 Hz, 1 H), 4.98 (s, 1 H), 3.61 (s, 3 H), 3.22 (m, 3 H), 2.28 (s, 3 H); ES-MS $m/z$ 389 (M + H).

**Methyl 2-((1*H*-Imidazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate 7a.** To a solution of **6** (2.0 mmol) in NMP (6 mL) were added imidazole (2.0 mmol) and $K_2CO3$ (4.4 mmol). The mixture was stirred at

room temperature overnight. The mixture was poured into water and extracted with EtOAc. The combined organic extract was dried over sodium sulfate and the solvent evaporated in vacuo to give the crude product which was purified by reverse phase HPLC. Yield 64%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.01 (s, 3 H), 3.49 (s, 3 H), 5.08 (s, 1 H), 5.14 (d, $J$ = 2.95 Hz, 2 H), 6.91 (s, 1 H), 7.20 (m, 3 H), 7.37 (dd, $J$ = 9.00, 2.55 Hz, 1 H), 7.69 (s, 1 H), 9.64 (s, 1 H). HRMS M + H calcd for $C_{19}H_{17}ClFN_4O_2$, 387.1019; obsd, 387.1058. Anal. ($C_{19}H_{17}ClFN_4O_2 \cdot 0.3H_2O$) C, H, N.

**Methyl 2-((1*H*-Pyrazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate 7b.** This compound was prepared by the same procedure described for **6a** with **6** and pyrazole. Yield 48%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.00 (s, 3 H), 3.45 (s, 3 H), 5.07 (s, 1 H), 5.21 (d, $J$ = 13.96 Hz, 1 H), 5.46 (d, $J$ = 13.96 Hz, 1 H), 6.25 (m, 1 H), 7.14 (m, 1 H), 7.33 (m, 2 H), 7.51 (d, $J$ = 1.88 Hz, 1 H), 7.72 (d, $J$ = 2.42 Hz, 1 H), 9.67 (s, 1 H). HRMS M + H calcd for $C_{19}H_{17}ClFN_4O_2$, 387.1019; obsd, 387.1052. Anal. ($C_{19}H_{15}ClFN_4O_2 \cdot 0.8HCl$) C, H, N.

**Methyl 2-((1*H*-1,2,4-Triazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate 7c.** This compound was prepared by the same procedure described for **7a** with **6** and 1,2,4-triazole. Yield 58%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.01 (s, 3 H), 3.44 (s, 3 H), 5.07 (s, 1 H), 5.23 (d, $J$ = 14.23 Hz, 1 H), 5.52 (d, $J$ = 14.23 Hz, 1 H), 7.17 (m, 1 H), 7.35 (m, 2 H), 8.01 (s, 1 H), 8.50 (s, 1 H), 9.77 (s, 1 H). Anal. ($C_{18}H_{15}ClFN_5O_2 \cdot 0.55HCl$) C, H, N.

**Methyl 2-((1*H*-Tetrazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate 7d.** To a solution of **6** (1.8 mmol) in NMP (3 mL) was added a solution of tetrazole in ACN (3% w/w, 6.5 mL) and $K_2CO_3$ (4.35 mmol). The mixture was stirred at room temperature overnight. The mixture was poured into water and extracted with EtOAc. The combined organic extract was dried over sodium sulfate and the solvent evaporated in vacuo to give crude product which was purified by reverse phase HPLC. Yield 60%; $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ ppm 2.02 (s, 3 H), 3.44 (s, 3 H), 5.10 (s, 1 H), 5.52 (d, $J$ = 14.23 Hz, 1 H), 5.65 (m, 1 H), 7.26 (m, 3 H), 9.40 (s, 1 H), 9.88 (s, 1 H). HRMS M + H calcd for $C_{17}H_{15}ClFN_6O_2$, 389.0924; obsd, 389.0924. Anal. ($C_{17}H_{15}ClFN_6O_2$) C, H, N.

**(*R*)-Methyl 2-((1*H*-Tetrazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate (*R*)-7d.** Racemic methyl 2-((1*H*-tetrazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate **7d** was loaded on a Berger SFC MultiGram II SFC system equipped with a Chiralpak AS-H (Chiral Technologies, 30 mm × 250 mm) and eluted with 20% MeOH/$CO_2$, 70 mL/min. The two peaks were collected to yield the two enantiomers. Peak 1: (*R*)-methyl 2-((1*H*-tetrazol-1-yl)methyl)-4-(2-chloro-4-fluorophenyl)-5-cyano-6-methyl-1,4-dihydropyridine-3-carboxylate (*R*)-**7d**; ES-MS $m/z$ 389 (M + H); >99% ee determined by analytical chiral chromatography performed on the equivalent Chiralpak AS-H column (Chiral Technologies, 4.6 mm × 100 mm, 3 mL/min).

**Biological Assays. Cell-Based Gal4 Response Element-Controlled Luciferase Reporter Assays. MR Luciferase Reporter Antagonist Assay.** HUH7 human hepatocyte cells were maintained in RPMI 1640 plus 10% FBS and transfected with Gal4-MRLBD construct and a luciferase reporter under Gal4 control. After transfection, compounds were added in RPMI 1640 media plus 10% heat-inactivated and charcoal dextran stripped FBS (Hyclone) with and without 1 nM aldosterone. Cells were harvested 20 h later for reporter activity as described.[16] Selectivity against other steroid receptors was assayed in the same manner using Gal4-driven luciferase as reporter.

**Dahl Salt Sensitive Rat Model of Hypertension and Nephropathy.**[13] All animals were outfitted with radiotelemetry units (Data Sciences Inc., St. Paul, MN) for conscious, unrestricted SBP measurements. After recovery from surgery, baseline SBP was measured and all animals were then randomized to various treatment groups and compounds were continued for 21 days. The vehicle group received 0.5% methylcellulose/0.1% Tween 80. All compounds given to the treatment groups were dissolved in 0.5% methylcellulose/0.1% Tween 80. For compound treated groups, animals were dosed with the compounds daily, via gavage. For eplerenone treated groups, eplerenone was incorporated into the 4% NaCl rodent chow at various concentrations (Research Diets, Inc., New Brunswick, NJ). Radiotelemetrized arterial SBP was measured with the DATAQUEST A.R.T., version 3.0, Gold software (Data Sciences International, St. Paul, MN).

Twenty-four hours prior to the termination of the study, animals were placed in metabolism caging and urine was collected at 24 h. Animals were not fasted for the 24 h period. After 21 days of treatment, animals were exsanguinated using a 20 gauge needle inserted into the abdominal aorta. Blood samples were immediately transferred into Vacutainer collection tubes (Becton-Dickinson and Co., Franklin Lakes, NJ) and placed on wet ice. Blood was centrifuged for 15 min at 3000 rpm, 4 °C, and plasma was collected and frozen at −80 °C until further analysis. Plasma and urine chemistries (e.g., albumin, creatine, and electrolytes) were analyzed with the Hitachi 912 automated diagnostic clinical chemistry analyzer (Roche Diagnostics Corp., Indianapolis, IN) according to standard procedures. The St. Louis Pfizer Institutional Animal Care and Use Committee reviewed and approved the animal use in these studies. The animal care and use program is fully accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International.

**Modeling Studies.** Molecular modeling was conducted using the Schrodinger Suite 2009 (Schrodinger Suite 2009 Induced Fit Docking protocol, Glide version 5.5, Prime version 2.1, Jaguar version 7.6; Schrodinger LLC, New York, NY). The wild type MR docking structure was prepared from the 1.95 Å MR structure with bound corticosterone (PDB code 2A3I)[19] by removal of waters, truncation of helix 12 (AF-2) to the C-terminus (residues 958−984), and capping of Pro 957 as the *N*-methylacetamide. Dihydropyridine (*R*)-**7d** was geometry-optimized using Jaguar (HF/6-31G**//HF/6-31G**) to ensure appropriate dihydropyridine ring geometry. The induced fit docking protocol[17] was used to dock (*R*)-**7d** into the truncated receptor model, using the default induced fit docking protocol parameters with the exception of truncating the side chain of Met 845 and Trp 806 during initial Glide docking, requirement of a hydrogen bond to Asn 770, refinement of side chains within 6.0 Å of ligand poses, and the use of XP scoring for the redock step. Pictures were generated using Maestro 9.0.111 (Schrodinger LLC).

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**Supporting Information Available:** Elemental analysis data for compounds **5i**, **5l**, and **7a**−**d**. This material is available free of charge via the Internet at http://pubs.acs.org.

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