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Reference:

Rogacki Maciej, Pitta Eleni, Balabon Olga, Huss Sophie, Lopez-Roman Eva Maria, Argyrou Argyrides, Blanco-Ruano Delia, Cacho Monica, Vande Velde Christophe, Augustyns Koen,- Identification and profiling of hydantoins : a novel class of potent antimycobacterial DprE1 inhibitors
Journal of medicinal chemistry - ISSN 0022-2623 - 61:24(2018), p. 11221-11249
Full text (Publisher's DOI): <https://doi.org/10.1021/ACS.JMEDCHEM.8B01356>
To cite this reference: <https://hdl.handle.net/10067/1559050151162165141>

Identification and profiling of hydantoins - a novel class of potent antimycobacterial DprE1 inhibitors

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Keywords: tuberculosis, mycobacterium, anti-mycobacterial, hydantoin, DprE1, drug discovery, inhibitor.

ABSTRACT

Tuberculosis is the leading cause of death worldwide from infectious diseases. With the development of drug-resistant strains of *Mycobacterium tuberculosis*, there is an acute need for new medicines with novel modes of action. Herein we are reporting the discovery and profiling of a novel hydantoin-based family of antimycobacterial inhibitors of the decaprenylphospho-beta-D-ribofuranose 2-oxidase (DprE1). In this study we have prepared a library of more than a 100 compounds and evaluated them for their biological and physicochemical properties. The series is characterized by high enzymatic and whole-cell activity, low cytotoxicity and a good overall physicochemical profile. Additionally we show that the series acts via reversible inhibition of the DprE1 enzyme. Overall, the novel compound family forms an attractive base for progression to further stages of optimization and may provide a promising drug candidate in the future.

INTRODUCTION

Tuberculosis (TB) is a disease caused primarily by the pathogen *Mycobacterium tuberculosis*. According to the World Health Organization, TB is the world's most deadly infectious disease with over 10 million new cases and 1.8 million TB-related deaths in 2015 alone.¹ The current first-choice therapy regimen still consists of drugs identified more than 60 years ago and requires administration of medicines for at least 6 months, even in the case of less severe infections. Long treatment regimens and pronounced side effects lead to poor patient compliance and have accelerated the emergence of drug-resistant strains of mycobacteria. With the WHO estimation of over 0.5 million multi-drug resistant cases developing in 2015¹ there is an acute need for new medicines with novel modes of action. Amongst the new potential bacterial targets that recently gained increased attention is the decaprenylphospho-beta-D-ribofuranose 2-oxidase (DprE1), a

periplasmic protein involved in the biosynthesis of the mycobacterial cell wall.² The enzyme is a flavoprotein catalyzing the first step in the redox epimerization of decaprenylphosphoryl-D-ribose. This epimerization process produces decaprenylphosphoryl-arabinose (DPA). DPA in turn is the biochemical donor of arabinofuranose residues during the biosynthesis of critical constituents of the mycobacterial cell wall. More specifically, biosynthesis of lipoarabinomannan and the peptidoglycan-arabinogalactone-mycolic acid complex depends on the availability of DPA and, hence, on DprE1 activity.³ DprE1 gained recognition during investigation of the benzothiazinone series (BTZ), a class of covalent suicide inhibitors.^{4,5} Several groups have later on contributed to the validation of DprE1 as an attractive drug target, generated insight in the binding mode and have reported multiple, structurally distinct compound series with either a covalent/irreversible or a non-covalent binding mode. A very exhaustive review covering all relevant DprE1-inhibitor literature to date was recently published in this journal.^{6,7} Noteworthy, the most advanced DprE1 inhibitors (benzothiazinones BTZ043, PBTZ-169/macozinone and the azaindole AZ7371) have recently entered the clinical development phase.^{8,9,10} **(Figure 1)** Sparked by the promising antimycobacterial profile of reported compounds, a target-based high-throughput screening campaign for inhibitors of DprE1 was performed by GlaxoSmithKline, leading to identification of a novel hydantoin-based hit scaffold. This report for the first time discloses the structure of the scaffold, which is unrelated to other known DprE1-ligands. In addition, research in the report aims at optimization of the hits, at generating insight in the Structure-Activity-Relationships (SARs) governing antimycobacterial potency, and at providing evidence that activity is effected through DprE1-inhibition. Finally, with the extensive biological and physicochemical profiling data we report, we wish to underscore the potential of the hydantoin scaffold for antimycobacterial drug discovery .

Figure 1. Selected reported DprE1 inhibitors.^{2, 8, 10-12}

RESULTS AND DISCUSSION

Chemistry and SAR. The hydantoin hit compound, **1** (Figure 2), appeared especially interesting due to high enzymatic inhibition (DprE1 $\text{pIC}_{50} = 7.0$) and good whole-cell activity ($\text{MIC} = 8.3 \mu\text{M}$). In addition to a promising activity profile, the compound was characterized by good solubility, acceptable lipophilicity and no detectable cytotoxicity (Table 1). Overall, **1** presented a good starting point for hit-to-lead optimization. Subsequent similarity-based clustering revealed 10 additional analogues showing detectable inhibition of the enzyme (see Table 1 and supporting information: Table S1). In order to organize further exploration of the series, the general structure common for all of the compounds was divided into 5 main parts as shown in Figure 2. During our initial assessment of **1** we focused on identifying the moieties and structural features of the scaffold that might represent potential liabilities for the series. Structure-activity relationship (SAR) data provided by this cluster was limited due to the small number of closely related structures. However, several compounds retained significant inhibitory activity ($\text{pIC}_{50} > 6$, Table 1), suggesting some possibility of modifications around rings A and B as well as the side-chain. All hit compounds preserved the hydantoin core as well as the acetyl linker indicating the potential importance of those fragments. At this stage the hit **1**, as well as all other analogues, were tested as racemates. Therefore, in order to better understand if one or both of the enantiomers contributed to the potency we prepared the racemic mixture of **1** via the Bucherer-Berg hydantoin cyclisation¹³ and

subsequent alkylation with the corresponding alpha-chloroketone **6** as shown in Scheme 1. Under these conditions alkylation occurred selectively on the *N*₃-nitrogen, providing the desired regioisomer as confirmed by crystal structure analysis (Figure 3). The enantiomers **7** and **8** were then separated by chiral HPLC. We were pleased to see that only the *R*-isomer contributed to both the enzymatic and whole-cell activity (Table 1). Although, with this information in hand, for procedural simplicity most compounds were prepared and tested as racemates.

Figure 2. Representative hits from the hydantoin cluster.

Scheme 1. Preparation of hit **1** and its *R* and *S* enantiomers **7** and **8**^a

^aReagents and conditions: (a) KCN, (NH₄)₂CO₃, EtOH, H₂O, 55°C, 18h; (b) K₂CO₃, acetone, rt, 18h; (c) separation via chiral HPLC.

Table 1. Biological and biochemical profile of hits **1-4** and enantiomers **7** and **8**.

Compound	1	2	3	4	7	8
DprE1 pIC ₅₀ ^[a]	7.0	6.8	6.4	4.4	7.2	4.2
MIC (μM) ^[b]	8.3	>80	40	>80	6.7	>125
HepG2 IC ₅₀ (μM) ^[c]	>100	49	88	n.d. ^[f]	>100	>100
Solubility (μM) ^[d]	202	n.d. ^[f]	n.d. ^[f]	n.d. ^[f]	355	344
ChromlogD (pH 7.4) ^[e]	4.54	n.d. ^[f]	n.d. ^[f]	n.d. ^[f]	4.51	4.51

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4; ^fn.d. = not determined.

Figure 3. Asymmetric unit of **1**. Anisotropic displacement ellipsoids drawn at the 50% level. Hydrogens are represented by spheres of arbitrary diameter. The asymmetric unit contains two molecules of opposite chirality, which are not related by crystallographic symmetry. They form a hydrogen bonded pair through the NH...O hydrogen bonds between the hydantoin moieties. The

2,4 fluorine substituted phenyl ring is disordered by 180° ring flipping. One molecule in the asu has a disorder 52:48 ratio, the other 63:37, reflecting the different environments that surround them.

We next moved to exploring the distinct elements of the hit scaffold. We were concerned that the acetyl linker could be a potential liability of the series. Aromatic ketones are generally considered to be reactive groups and can contribute to increased metabolic instability as well as possible non-specific covalent binding.¹⁴ Since the initial cluster did not provide information about the possible modifications of that part, we decided to explore the importance of the linker. For this purpose we envisaged modulating the linker length, as well as removal of the carbonyl group or introduction of known bioisosteric substitutions. Since hits **2** and **3** preserved good, albeit lower inhibitory activity, for some of the analogues we chose to include *para*-methoxyphenyl, and *para*-chlorophenyl substituted hydantoin counterparts in addition to the main *para*-cyanophenyl to probe the effect of alternative substitution and electron donation/withdrawal on the activity and physicochemical properties of the series.

Scheme 2. Synthetic approach to analogues with linker modifications^a

^aReagents and conditions: (a) K_2CO_3 , acetone or DMF, rt, 18-48h; (b) DAST (neat), rt, 1 week; (c) K_2CO_3 , DMF, 100°C , 2d; (d) EtOH, H_2SO_4 , 80°C , overnight (e) paraformaldehyde, $t\text{-BuOK}$, DMF, rt, overnight; (f) $\text{BH}_3\text{-THF}$, THF, rt, overnight; (g) (I) $n\text{-BuLi}$, Ts-Cl, THF, 0°C -rt, 1h20min, (II) $n\text{-BuLi}$, THF, 0°C -rt, 40min, (III) 65°C , 3.5h, (IV) $n\text{-BuLi}$, Ts-Cl, THF, 0°C -rt, 1h20min; (h) K_2CO_3 , DMF, 80°C - 100°C , 48h.

We prepared a number of analogues (**24-40**) with linker modifications via the alkylation of the previously prepared compound **5** or available hydantoins **22** and **23**, as shown in Scheme 2. The starting alkyl halides or pseudohalides were either commercially available (**6**, **9-12**) or prepared using standard literature conditions¹⁵ (**13-15**). Additionally, geminal difluorination of the ketone¹⁶ **16**, obtained by standard bromination of the 1-(2,4-difluorophenyl)ethan-1-one, led to **17**, which was further used to alkylate hydantoin **5** to provide the analogue **41**. The oxetane **42** was prepared following a 5-step synthesis. Briefly, esterification of **18**, followed by hydroxymethylation¹⁷ and

in-situ deprotection gave the acid **19**, which was reduced to the alcohol **20** with borane-THF complex.¹⁸ One-pot mono-tosylation, ether formation¹⁹ and subsequent second tosylation of the alcohol gave the oxetane **21**, which was used to alkylate the hydantoin **5** and provide the final product **42**.

Scheme 3. Preparation of analogues **43a-b** and **44**^a

^aReagents and conditions: (a) HONH₂HCl, MW, 100°C, 30min; (b) Red-Al, toluene, 120°C, 1h.

Alternatively, the ketone of the parent compounds **1** and **24** could be modified as shown on Scheme 3. Thus, microwave irradiation of a solution of **1** and hydroxylamine led to formation of two isomeric oximes²⁰ (**43a** and **b**). Lastly, Red-Al mediated reduction²¹ of **24** provided alcohol **44** as a diastereoisomeric mixture.

Throughout the synthesis of this series of analogues, we noticed that the alkylation of hydantoins consistently provided 3-substituted products (**24-42**) in moderate to good yields. However, inverse regioselectivity was observed in reactions of the acyl chloride **45** with hydantoins, leading to *N*₁-

substituted isomers **46** and **47** (Scheme 4). The desired 3-acylated analogue could be prepared by deprotonation of the hydantoin prior to the addition of the acyl chloride, however the imide **48** proved unstable in solution over time.

Scheme 4. Preparation of acylated hydantoins^a

^aReagents and conditions: (a) pyridine, rt, overnight; (b) NaH, DMF, rt, overnight.

Table 2. *In vitro* activity, cytotoxicity and physicochemical properties of the analogues with linker variation.

Nº	R ₁	linker	R ₂	DprE1 pIC ₅₀ ^[a]	<i>Mtb</i> MIC (µM) ^[b]	HepG2 IC ₅₀ (µM) ^[c]	Solubility (µM) ^[d]	Chrom logD ^[e]
1			CN	7.0	8.3	>100	202	4.54
24	F		OCH ₃	6.7	40	>100	358	4.78
25			Cl	6.7	>40	>100	17	5.54
26			CN	6.0	19.6	>100	132	4.26
27	H		OCH ₃	6.0	62	>100	260	4.53
28			Cl	4.2	>125	>100	7	5.28
29			CN	<4.0	125	>100	185	4.56
30	H		OCH ₃	<4.0	>125	>100	327	4.84
31			Cl	<4.0	>125	>100	19	5.68
32			CN	<4.0	>125	>100	≥404	4.50
33	H		OCH ₃	<4.0	>125	>100	≥381	4.72
34			Cl	<4.0	>125	63	188	5.59
35			CN	4.3	>125	>100	≥428	4.40
36	H		OCH ₃	4.4	>125	>100	≥409	4.60
37			Cl	<4.0	>125	>100	222	5.35
38	F		CN	5.40	80	>100	≥458	4.83
39	F		CN	4.60	>80	>100	314	4.33

40	F		CN	4.4	>125	>100	n.d. ^[f]	n.d. ^[f]
41	F		CN	5.0	>80	>100	≥261	5.1
42	F		CN	<4.0	>80	>100	295	4.21
43a	F		CN	4.0	>80	>100	≥480	3.38
43b				4.2	>80	>100	≥471	3.59
44	F		OCH ₃	4.2	>80	>100	287	3.96
46			OCH ₃	<4.0	>40	>100	≥484	3.43
47			Cl	<4.0	>125	>100	≥386	3.72

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4; ^fn.d. = not determined.

As shown in the Table 2, most modifications, such as modulation of linker length or removal of the keto group resulted in a complete loss of activity. Even methylation of the methylene group (**38**) led to 40-fold decrease of enzymatic potency. Surprisingly, even closely related analogues and bioisosteric transformations of the carbonyl moiety (**41-43**) did not retain activity, pointing to specific importance of that group. It is also worth noting that **44** could be expected to be one of the direct metabolites of the parent compound, therefore its lack of activity was particularly interesting.

Although all applied modifications led to significant loss of potency, it was encouraging to see the consistently low toxicity throughout the series of analogues. Except for a few more lipophilic representatives bearing chlorine substitutions (**25**, **28**, **31**), we also observed consistently good solubility.

Scheme 5. Preparation of analogues with modifications around ring A and the side-chain^a

^aReagents and conditions: (a) KCN, (NH₄)₂CO₃, EtOH, H₂O, 55°C, 18h; (b) K₂CO₃, acetone or DMF, rt, 18h.

Next, we moved to exploring substituents at the C₅-carbon of the hydantoin ring. Our attention was drawn by the nitrile in the position 4 of the “ring A”. Carbonitrile moieties have been generally reported and/or predicted to be able to react with cysteine^{22,23} and/or serine^{24,25} residues which could lead to potential off-target covalent binding. Comparison of **1**, **4**, **24** and **25** suggested the importance of the nitrile part, since substitution with a methoxy group (**24**) or a chlorine atom (**25**) led to a decrease in potency and lack of any group on the phenyl ring (**4**) provided a completely inactive compound. We therefore put our main focus on exploring the possible substitutions at that position. We also decided to introduce substituents at carbons 2 and 3 of ring A to probe the space available for further modifications. Additionally we were interested in exchanging the phenyl ring for a heterocyclic moiety and its impact on the activity as well as physicochemical properties of the series.

Most compounds were synthesized following the Bucherer-Berg hydantoin cyclisation and subsequent alkylation (Scheme 5), starting from ketones that were either commercially available or were prepared using standard literature procedures (see Supporting Information). Synthesis of hydantoin **139** was also attempted using the same approach, however the sulfonamide moiety appeared to be reactive during the alkylation step. We therefore decided to prepare **139** and **140** from the bromo-analogues **109** and **123** respectively, by palladium-catalyzed introduction of methanesulphonamide²⁶ as shown on Scheme 6. Anticipating similar reactivity problems, aniline **141** was obtained by acid catalyzed hydrolysis of the amide **119**. Finally, nitrile analogues **142** and **143** were prepared by microwave-promoted, palladium-catalyzed cyanation²⁷ of the corresponding bromopyridines **130** and **131**.

Scheme 6. Preparation of analogues with modifications around ring A and the side-chain^a

^aReagents and conditions: (a) CH₃SO₂NH₂, K₂CO₃, [Pd(allyl)Cl]₂, *t*-BuXPhos, 80°C, 4h; (b) EtOH, HCl, 65°C, 3d; (c) Zn(CN)₂, Pd₂(dba)₃, XantPhos, TMEDA, DMF, MW, 160°C, 5min.

Although initial substitutions at “ring A” led to less active compounds (**24**, **25**), we were pleased to observe that the nitrile group is not necessary and in fact can be exchanged for a variety of groups providing similarly or even more potent analogues, such as **114**, **116** or **139** (Table 3). It

was also interesting to note that the high activity is maintained in the case of slightly bulkier substituents, such as a tetrazole or a morpholine (**116**, **117**), which suggests that the enzymatic pocket could accommodate further expansion of the molecule in that direction. The obtained SAR data also showed that although some modifications are permitted at position 2 of the phenyl ring (**125**, **126**, **131**, **143**), substitutions at position 3 (**124**, **127**, **130**, **140**, **142**) generally gave a more pronounced loss of activity (Table 4), suggesting rather limited space around the aromatic ring. We were especially encouraged by the results obtained from the pyridine analogues **131** and **143**, which indicated that the phenyl ring A can be exchanged for a heterocyclic analogue, improving physicochemical properties of the compound. However the possibility of manipulation may prove to be limited, since substitution with a 5-membered heterocyclic analogue **132** led to a complete loss of both enzymatic as well as whole-cell activity.

Although some of the most potent compounds (**109**, **113** and **114**) were unfortunately linked to higher lipophilicity (and subsequently reduced solubility), good enzymatic and whole-cell potencies were also observed for the soluble and less lipophilic analogues such as the tetrazole **116** and the sulfonamide **139**.

It should be emphasized, that even for analogues with increased lipophilicity, no appreciable cytotoxicity was found throughout the series of modifications, indicating a promising safety profile of the investigated series.

Table 3. *In vitro* activity, cytotoxicity and physicochemical properties of the analogues with varying substituents at the 4-position of ring A.

№	R
		DprE1 pIC ₅₀ ^[a]	<i>Mtb</i> MIC (μM) ^[b]	HepG2 IC ₅₀ (μM) ^[c]	Solubility (μM) ^[d]	Chrom logD ^[e]
1	-CN	7	8.3	>100	202	4.54
24	-OCH ₃	6.7	40	>100	358	4.78
25	-Cl	6.7	>40	>100	17	5.54
109	-Br	7.1	20	>100	4	5.68
110	-CH ₃	6.1	80	>100	213	5.41
111	-OC ₂ H ₅	7.0	20	>100	167	5.43
112	-CF ₃	6.9	20	>100	43	6.01
113	-OCF ₃	7.3	35	>100	4	6.05
114	-OCHF ₂	7.4	10	>100	85	5.36
115	-(imidazol-1-yl)	6.5	20	>100	280	3.84
116	-(tetrazol-1-yl)	7.3	3.1	>100	379	3.78
117	-(N-morpholine)	7.1	10	>100	277	4.45
118		6.6	5.6	>100	359	3.94
119	-NHCOCH ₃	5.6	40	>100	≥489	3.28
120	-SO ₂ CH ₃	6.8	10	>100	≥461	3.64
139	-NHSO ₂ CH ₃	7.0	2.5	>100	≥487	3.57
141	-NH ₂	4.1	>80	>100	≥436	3.38

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4.

Interestingly, we observed that within the prepared series of compounds, higher enzymatic potency did not always translate into an improvement in MIC values, as seen in case of fluorinated methoxy (**113**, **114**) or bromine-bearing (**109**) analogues. It is worth noting that these molecules are characterized by higher lipophilicity as opposed to compounds showing an increase also in whole-cell potency (**116**, **139**), with ChromLogD values of 5.30-6.05 and 3.57-3.78 respectively. However, comparison of the pair **109** and **131** showed that lowering the ChromLogD value itself did not lead to an improvement in whole-cell potency in case of analogues with high enzymatic activity. This suggests that the overall lipophilicity of the molecule is not the driving factor behind the higher than expected MIC values. One could argue that the improved MIC potencies were a result of issues with borderline solubility of some compounds, such as **109** or **113**. However, as seen from the bromine-bearing analogues **109** and **131**, significant improvement of solubility did not translate into a substantial increase of whole-cell potency.

Table 4. *In vitro* activity, cytotoxicity and physicochemical properties of the analogues with modifications of the ring A.

№	R
		DprE1 pIC ₅₀ ^[a]	<i>Mtb</i> MIC (μM) ^[b]	HepG2 IC ₅₀ (μM) ^[c]	Solubility (μM) ^[d]	Chrom logD ^[e]
121		4.4	>80	>100	312	4.51
122		<4.0	>80	>100	285	4.96
123		4.5	80	>100	68	5.72

124		5.7	>80	>100	147	4.84
125		6.5	58	>100	104	5.05
126		6.7	30	>100	17	5.80
127		6.9	80	>100	106	5.50
128		<4.0	>80	>100	287	4.47
129		4.4	>125	>100	≥462	3.04
130		6.4	40	>100	369	4.67
131		7.3	15	>100	100	4.87
132		<4.0	>80	>100	417	4.67
140		4.2	>80	>100	≥369	3.67
142		6.0	20	>100	≥410	3.94
143		6.6	10	>100	≥474	3.97

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4.

While exploring the SAR around Ring A, we also decided to use the same synthetic approach (Scheme 5) to probe the possibility of manipulating the hydantoin side-chain (Table 5). However,

we found that only ethyl analogues (**133**, **134**) retained high activity as compared to their methyl-bearing counterparts (**109** and **1** respectively). Further extension of the chain (**135**) or introduction of a bulky substituent (**136**, **137**) resulted in significantly less potent compounds, suggesting very limited chemical space available around that part of the scaffold.

Table 5. *In vitro* activity, cytotoxicity and physicochemical properties of the analogues with modifications of the side-chain.

No			DprE1	<i>Mtb</i> MIC	HepG2 IC ₅₀	Solubility	Chrom
	R ₁	R ₂	pIC ₅₀	(μM) ^[a]	(μM) ^[b]	(μM) ^[d]	logD ^[e]
133	ethyl	-Br	6.9	10	56	31	6.17
134	ethyl	-CN	7.3	5	>100	154	4.99
135	propyl	-Br	6.2	80	51	7	6.64
136	isopropyl	-Br	4.7	>80	48	11	6.57
137	-CF ₃	-Br	5.1	>80	50	1	6.80
138			6.7	>80	>100	<1	5.71

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4.

Next, we turned our attention to the hydantoin core. Although attractive from the synthetic perspective and present in marketed drugs, this heterocycle has been linked to some undesired side-effects,²⁸ such as the congenital hydantoin syndrome²⁹ or inhibition of the hERG potassium channel.^{30,31} Cardiovascular risks are considered a major liability for further development of a lead

series, we were therefore interested in finding a suitable replacement for the core. Additionally, since the hydantoin core can serve as a platform for formation of multiple hydrogen bonds, we planned stepwise modifications to evaluate the role of the heterocycle in interacting with the DprE1 enzyme.

Some close analogues of hydantoin are succinimide, imidazolidin-2-one and γ -lactam rings. We considered these heterocycles to be attractive alternatives since they lack one or more hydrogen bond donors/acceptors while maintaining similar overall geometry of the original hit scaffold. Analogues built around these cores were prepared as shown on Scheme 7. The succinimide ring of **149** was constructed via the approach described by Tominaga et al.,³² starting with a reaction of 4-bromoacetonitrile and a commercially available nitroketene dithioacetal, followed by cyclisation to **145**. Subsequent acid-catalyzed hydrolysis of the oxime and further reduction with zinc provided the succinimide **147**. Alkylation of **147** with a halo ketone provided **148**, which was further methylated to yield the desired product **149**. The cyclic urea was obtained from hydantoin **24** by Red-Al mediated reduction, as reported by von Kieseritzky and Lindström.²¹ Under these conditions the carbonyl group in the linker was reduced to an alcohol, which was subsequently oxidized using the Swern reaction.³³ The resulting mixture was purified by chiral HPLC, providing pure enantiomers **151a** and **151b**. Finally, the lactam **155** was synthesized starting from 2-(4-bromophenyl)acetic acid. Briefly, the acid was protected with a methyl ester and alkylated first with methyl iodide and then with bromoacetonitrile. Resulting ester **153**, was cyclized to **154** under reductive conditions with sodium borohydride in the presence of cobalt chloride.³⁴ Alkylation of **154** yielded the final lactam **155**.

To further evaluate the role of N_1 -nitrogen, we decided to prepare analogues **156** and **157** by simple methylation or acylation of the hit **1**, as shown in Scheme 8.

Finally, we wanted to probe the possibility of a more aggressive scaffold hopping approach by replacing the original core with an aromatic heterocycle. For that purpose, we envisaged imidazole as a direct analogue of a hydantoin cycle. The aromatic analogues were obtained as shown in Scheme 9. The 4-substituted imidazoles **158** and **159** were constructed by cyclisation of the corresponding bromoketones with formamide.³⁵ Subsequent alkylation with chloroketone **6** provided analogues **160** and **161**. Additionally, we also prepared an analogue in which the core is replaced by a pyrazole. This compound was synthesised starting from a commercially available 4-bromopyrazole (Scheme 9). After initial trityl protection, the intermediate was coupled with a 4-cyanobenzeneboronic acid under Suzuki-Miyaura conditions.³⁶ Acid-mediated deprotection provided **162** which was further alkylated to obtain the final pyrazole analogue **163**.

Scheme 7. Preparation of analogues with modified cores^a

^aReagents and conditions: (a) NaOH, DMSO, rt, 4h; (b) MeOH, reflux, 30min; (c) HCl, MeOH, reflux, 5d; (d) Zn, AcOH, reflux, 2.5h; (e) **16**, K₂CO₃, acetone, rt, overnight; (f) MeI, K₂CO₃, acetone, rt, 2d; (g) Red-Al, toluene, 120°C, 2h15min; (h) (I) oxalyl chloride, DMSO, DCM, -78°C, 15min, (II) TEA, rt, 2h; (i) H₂SO₄, MeOH, 80°C, overnight; (j) (I) diisopropylamine, *n*-BuLi, THF, -78°C, 20min, (II) MeI, 0°C, 30min; (k) (I) diisopropylamine, *n*-BuLi, THF, -78°C, 20min, (II) bromoacetonitrile, -78°C, 1.5h - , 0°C, 30min; (l) NaBH₄, CoCl₂·6H₂O, 0°C – rt, overnight; (m) **16**, NaH, THF, rt, overnight.

Scheme 8. Preparation of analogues with substituted N_1 -nitrogen.

^aReagents and conditions: (a) K_2CO_3 , acetone, rt, 2d; (b) $AcCl$, TEA, dioxane, rt, 3d.

Scheme 9. Preparation of analogues with aromatic cores.

^aReagents and conditions: (a) formamide, $170^\circ C$, 2-3h; (b); K_2CO_3 , DMF, $0^\circ C$ – rt, overnight; (c) Cs_2CO_3 , DMF, $80^\circ C$, 90min; (d) $TrCl$, DMF, $0^\circ C$, 1h; (e) (4-cyanophenyl)boronic acid, $Pd(PPh_3)_4$, dioxane, $85^\circ C$, overnight; (f) TFA, $70^\circ C$, 1h; (g) **6**, K_2CO_3 , DMF, MW, $60^\circ C$, 30min.

Table 6 summarizes the results obtained from the modifications of the core hydantoin. It was disappointing to observe that none of the compounds tested was able to retain potency comparable to the parent hydantoins. Almost complete loss of enzymatic activity of both ureas **151a** and **151b** suggests that the carbonyl moiety at position 4 may take part in interactions that are crucial for the

potency of the series. Modifications around position 1 were also quite noteworthy. Both placing substituents at the nitrogen (**156**, **157**) as well as exchanging it for a carbon (**149**) led to a significant decrease of enzymatic activity. This could suggest that the *N*₁-nitrogen atom is important for binding to the enzyme. Interestingly enough, although methylation of this nitrogen led to a complete loss of enzymatic activity (**156**), amide **157** retained whole cell potency comparable to the original hit (MIC = 10 and 8.3 μ M respectively). Since **157** appears to be chemically stable in solution, one can speculate that it could be subject to hydrolysis by the bacterial amidases and hence act like a prodrug.

Unsurprisingly, the aromatic analogues **160**, **161** and **163** also did not retain potency. Pronounced differences in the overall geometry of the final molecules as well as lack of important hydrogen bond donors/acceptors were the likely culprits for this loss of activity and demonstrated that in this case the imidazole and pyrazole heterocycles are not suitable candidates for a scaffold hopping approach.

Taken together, these results indicate that the core hydantoin cycle not only serves as a linking scaffold for proper spatial organization of the peripheral moieties, but seems to also be involved in interactions with the protein that are crucial for the potency of the series.

Table 6. *In vitro* activity, cytotoxicity and physicochemical properties of the analogues with modifications of core hydantoin.

№			DprE1 pIC ₅₀	<i>Mtb</i> MIC (μM) ^[a]	HepG2 IC ₅₀ (μM) ^[b]	Solubility (μM) ^[d]	Chrom logD ^[e]
	Core	R					
149		-Br	5.8	>80	>100	191	6.86
151a ^[f]		-OCH ₃	<4.0	>80	>100	88	4.72
151b ^[f]			4.5	>80	>100	177	4.72
155		-Br	5.1	>80	>100	89	6.74
156		-CN	4.9	>80	>100	167	5.22
157		-CN	4.5	10	>100	61	5.77
160		-CN	4.7	>125	>100	25	4.58
161		-OCH ₃	<4.0	>125	>100	352	4.68
163		-CN	<4.0	>125	>100	21	5.17

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4; ^fseparated enantiomers – absolute configuration was not determined.

Finally, we turned our attention to substitution around ring B. Structural features of that fragment were not identified as liabilities, therefore we were mainly interested in probing the chemical space available for manipulations and their contribution towards the SAR. Additionally we also envisaged introduction of more polar groups that could help decrease the lipophilicity of the molecule and improve the overall physicochemical profile of the series. For this exploration, in addition to the hydantoin counterparts bearing the hit 4-cyano group, we decided to also include the 4-OCHF₂ substituted analogues. Since **114** showed the highest enzymatic activity without an increase in the whole-cell potency, the difluoromethoxy analogues were prepared to further explore this apparent disconnection.

Compounds **180-198** were prepared following the standard alkylation approach shown on Scheme 10 from haloketones that were either commercially available or prepared using literature procedures (see Supporting Information). Additionally, the library was expanded with molecules **199-209**, that were available in GSK compound database (see Table 7).

Scheme 10. Preparation of analogues with modifications around ring B.

^aReagents and conditions: (a) K₂CO₃, acetone or DMF, rt, overnight.

As shown in Table 7, we observed that a number of modifications are permitted in that part of molecule. Although the original 2,4-difluorophenyl ring from hit **1** remained amongst the most active groups, single fluorine (**180, 182**) or methyl substituents (**204**) also provided analogues with good enzymatic potency. Although introduction of more polar groups such as methoxy (**184-187**),

nitro (**189**) or dioxole (**182**) usually led to less potent molecules, it was particularly interesting to see that the phenyl ring can also be exchanged for some other aromatic heterocycles, such as pyridines (**192**, **193**) or a thiophene (**206-207**), leaving some potential for optimization of the physicochemical properties without a significant loss of potency. It seemed however, that all aliphatic substitutions (**178-181**) resulted in a more pronounced loss of activity indicating that an aromatic moiety at ring B is important for the high potency of the compound. Taken together, these results suggest that the SAR around Ring B is not confined to limited chemical space and ring B can be modified during further optimization of the series.

Table 7. *In vitro* activity, cytotoxicity and physicochemical properties of the analogues with modifications of ring B.

No			DprE1 pIC ₅₀ ^[a]	<i>Mtb</i> MIC (μM) ^[b]	HepG2 IC ₅₀ (μM) ^[c]	Solubility (μM) ^[d]	Chrom logD ^[e]
	R ₁	R ₂					
1		-CN	7	8.3	>100	202	4.54
114		-OCHF ₂	7.4	10	>100	85	5.63
26		-CN	6.0	19.6	>100	132	4.26
180		-OCHF ₂	7.2	15	>100	98	5.08
181		-OCHF ₂	6.4	20	>100	144	5.13
182		-OCHF ₂	7.3	20	>100	141	5.23
183		-OCHF ₂	7.2	30	>100	180	4.97
184		-CN	5.7	>80	>100	270	4.25
185		-OCHF ₂	6.2	80	>100	104	4.92

186		-CN	5.9	80	>100	283	4.39
187		-CN	4.4	>80	>100	≥402	4.45
188		-CN	6.5	20	>100	149	5.3
189		-CN	5.6	>80	>100	29	4.25
190		-OCHF ₂	6.3	30	>100	≥328	3.52
191		-OCHF ₂	5.7	80	>100	≥503	3.54
192		-OCHF ₂	6.7	15	>100	≥497	4.35
193		-CN	7	5	>100	117	3.89
194		-OCHF ₂	6.8	20	>100	356	4.59
195		-CN	5.7	80	>100	≥470	3.42
196		-OCHF ₂	6.7	20	>100	≥409	4.45
197		-CN	5.7	>80	>100	≥481	5.07
198	Adamantyl	-CN	4.7	>80	>100	87	6.31
199 ^[f]		-OCHF ₂	5.3	80	>100	≥472	4.03
200 ^[f]		-OCHF ₂	4.6	>80	>100	≥467	3.37
201 ^[f]		-OCHF ₂	6.6	>80	>100	80	4.90
202 ^[f]		-OCHF ₂	6.4	80	>100	105	5.03

203 ^[f]		-OCHF ₂	5.4	>80	>100	183	5.42
204 ^[f]		-OCHF ₂	7.0	80	63	69	5.97
205 ^[f]		-OCHF ₂	5.0	>80	50	18	6.48
206 ^[f]		-OCHF ₂	7.0	80	>100	132	5.06
207 ^[f]		-OCHF ₂	7.2	20	>100	97	5.49
208 ^[f]		-OCHF ₂	4.3	>80	>100	≥456	3.91
209 ^[f]		-OCHF ₂	4.4	>80	>100	≥455	4.12

^aDprE1 pIC₅₀ is the negative logarithm of the IC₅₀-concentration expressed in M (molar) obtained in the DprE1-inhibition assay; ^bMIC against *Mycobacterium tuberculosis* (H37Rv), isoniazid was used as a reference with MIC= 1.8 μM; ^ccytotoxicity against HepG2 human caucasian hepatocyte carcinoma; ^dkinetic aqueous solubility (CLND); ^elipophilicity - chromlogD at pH = 7.4; ^fmolecules present in GSK compound library and not synthesized in the course of this study.

Metabolic stability. Several potent analogues were tested for *in vitro* metabolic stability. The results summarized in Table 8 illustrate that the compounds were generally stable with most representatives showing intrinsic clearance values below 3 ml/min/g in mouse microsomes and lower than 0.4 ml/min/g in human microsomes. It was particularly interesting to observe that essentially all potent compounds were characterized by an improved microsomal stability over the original hit **1**. We were therefore pleased to conclude that the lack of available SAR space around the acetophenone linker might not be so critical to the optimization of the series.

Table 8. Microsomal stability of selected potent analogues.

Mouse	Human
Nº	R	Cl _{int} (ml/min/g)	Cl _{int} (ml/min/g)
1	-CN	3.52	0.49
109	-Br	1.48	<0.40
114	-OCHF ₂	1.78	<0.40
116		2.44	<0.40
118		3.54	<0.40
120	-SO ₂ CH ₃	2.50	0.46
139		3.29	0.47

hERG inhibition. Since any attempted alteration of the core hydantoin provided loss of potency, we were concerned with potential cardiotoxicity linked to that heterocycle. Indeed, the hit compound **1** as well as the difluoromethoxy (**114**) and tetrazole (**116**) analogues showed measurable inhibition of the hERG potassium channel. However, as presented in the Table 9, we also observed considerable improvement for several analogues, with compounds such as **118** or **120** showing no appreciable inhibition. Interestingly, also substituting the phenyl ring B of the hit **1** for a pyridine ring provided a close analogue **193** with no hERG inhibition. Overall these results suggest that potential cardiotoxicity is not inherent to the series but rather is dependent on the substituents on rings A and B. Although activity against the hERG channel should be monitored

for future analogues, the hydantoin core itself should not be treated as a major concern for further development of this compound family.

Table 9. hERG inhibition of selected potent analogues.

№	R		hERG pIC ₅₀
1	-CN		4.6
4	-OCH ₃		<4.3
114	-OCHF ₂		5.3
116			5.2
118			<4.3
120	-SO ₂ CH ₃		<4.3
139			4.4
193			<4.3

Validation of the mode action via overexpression of DprE1. Although the existing enzymatic and whole cell data appeared to indicate that these compounds were in fact operating on DprE1, we were interested whether the whole-cell potency solely depends only on that interaction or whether there are also contributions from alternative modes of action. Therefore we selected several of the most potent analogues (**1**, **114**, **116** and **139**) for MIC determination against *M. tuberculosis* strain engineered to overexpress the DprE1 protein. As presented in Table 10, higher expression of the target enzyme conferred resistance to all tested compounds with a more than 8

fold shift in the MIC values relative to the wild type strain. These results are in agreement with data from the other known DprE1 inhibitors, both reversible (TCA1)¹¹ and irreversible (BTZ043).³⁷ Therefore, inhibition of DprE1 was confirmed as the only major mechanism responsible for antimycobacterial activity of the hydantoin series.

Table 10. MIC against *Mtb* overexpressing DprE1^a

No	R
		MIC _{WT} (μM) ^[b]	MIC _{OE} (μM) ^[c]	MIC _{OE} / MIC _{WT} ^[d]
1	-CN	8.3	125	15
114	-OCHF ₂	10	>80	>8
116		3.1	>80	>25
139		2.5	>80	>32
TCA1		0.5	100	200
BTZ043		<0.6	>16	>26

^aThe results were compared with MIC activity of the tested compounds against a control *Mycobacterium tuberculosis* strain transformed with an empty vector; ^bMIC against *Mycobacterium tuberculosis* (H37Rv); ^cMIC against *Mycobacterium tuberculosis* overexpressing DprE1; ^dRatio of MIC values against the DprE1-overexpressing strain and the wild type strain.

Hydantoins are reversible DprE1 inhibitors. Since hit **1** contains potentially reactive groups, such as the ketone in the linker part or the nitrile substituent on the ring A, we were concerned with possible formation of covalent interactions with the target protein. To assess the mechanism

of inhibition, the rates of reaction catalyzed by DprE1 were monitored in the presence of varying amounts of the inhibitor. Irreversible covalent inhibition leads to gradual depletion of the active enzyme and hence a decrease of reaction rate over time. As shown on Figure 4, almost perfectly linear increase of the product's concentration was maintained throughout a period of 60 minutes with no apparent time-dependent inhibition. Increasing concentration of the inhibitor did not lead to loss of this linearity, suggesting that **1** is not a suicide inhibitor and binds to the enzyme in a reversible manner.

Figure 4. DprE1-catalyzed reaction progress in presence of varying concentrations of **1**.

Intracellular activity. Since *Mycobacterium tuberculosis* is an intracellular pathogen and survives within macrophages, we were interested in the ability of the investigated series to inhibit the growth of bacilli in such environment. Selected potent analogues were therefore tested for their activity in an intracellular model of *M. tuberculosis* infection of human macrophages. As shown in Table 11 the obtained results were mostly consistent for the series with the intracellular activity generally following the MIC values. The active *R*-enantiomer **7** of the hit compound and the tetrazole **116** showed good potency ($IC_{50} = 0.40 \mu M$ and $0.13 \mu M$ respectively), while **114** exhibited slightly lower activity ($IC_{50} = 0.74 \mu M$). Interestingly, analogues **118** and **139**, although potent in the whole-cell antimycobacterial assay, and showing higher activity than the racemic

hit **1** (IC_{50} = 0.85 μ M, 0.44 μ M and 1 μ M respectively) did not provide increased intracellular activity over the enantiomer **7**. These results suggest, that the representatives of the investigated series exhibit good activity in a macrophage infection model. However the actual potency is not always directly translatable from the standard MIC values and additional factors might have an impact on the compound's potency in an intracellular setting.

Table 11. Activity of selected potent analogues in an intracellular macrophage infection model.

No	R	<i>Mtb</i> MIC (μ M) ^[a]	Intracellular IC_{50} (μ M) ^[b]
1	-CN	8.3	1.00
7	-CN (R-enantiomer)	6.7	0.40
114	-OCHF ₂	10	0.74
116		3.1	0.13
118		5.6	0.85
139		2.5	0.44

^aMIC against *Mycobacterium tuberculosis* (H37Rv); ^b IC_{50} against *Mycobacterium tuberculosis* (H37Rv) in infected Human THP-1 macrophages.

Hydantoins are selective against Actinobacteria. As a means of further characterizing the series, we were also interested in its spectrum of activity against other bacteria. For the hit **1** and compounds **118** and **139**, MIC values were determined against a panel of medically relevant strains of gram positive and gram negative bacteria. Microbial selectivity is an important factor in development of an antibiotic series. Antitubercular therapy requires long-term administration of

drugs and activity against other bacterial species could not only cause eradication of non-pathogenic bacteria, but also lead to development and spread of drug resistance. Since conserved DprE1 orthologues are found in the Actinobacteria and in particular in Mycobacteria,² we were not expecting any activity against other non-actinobacteria-related species. We were therefore pleased to observe that no appreciable inhibition of growth was found in any of the experiments, as shown in Table S2 in Supporting Information. These results indicate the hydantoin compound family are selective inhibitors of growth of Mycobacteria and closely related species, as well as further support the proposed mechanism of action.

CONCLUSIONS

In summary, we report here a new family of potent and selective antimycobacterial compounds identified through a target-based high-throughput screening against the decaprenylphospho-beta-D-ribofuranose 2-oxidase (DprE1). Our medicinal chemistry efforts presented in this work provided more than 100 analogues of the original hit and led to identification of potent representatives characterized by high *in vitro* enzymatic activity (pIC_{50} 7-7.4) and whole-cell MIC values in the low micromolar range. Although initial examination of the original hit identified some structural concerns with potential developability or safety issues, we were able to show that the most essential structural features do not constitute any major liability inherent to the series and provide a platform for simple modular synthetic accessibility of a variety of analogues. The new scaffold class is generally characterized by good metabolic stability, no appreciable cytotoxicity and possesses a physicochemical profile acceptable for further development. We were also able to show that the series acts via a reversible mechanism of inhibition of DprE1 and that no other underlying modes of action seem to strongly contribute to its antibacterial activity. Overall, the

novel family of compounds forms an attractive base for progression to lead-optimization stage and may provide a promising drug candidate in the future. Further structural optimization of the compound family is currently underway. Results of the latter work will be published as soon as possible, including *in vivo* evaluation data of new, maximally optimized molecules in a rodent model of respiratory TB.

EXPERIMENTAL SECTION

General Information. Laboratory reagent grade solvents were used unless mentioned otherwise. Reagents were purchased from Sigma-Aldrich, Acros Organics, Fluorochem, TCI, Apollo Scientific or Enamine and were used without further purification unless otherwise stated. Reactions were monitored by TLC on silica gel and/or by UPLC-MS. Silica gel TLC analysis was performed using Polygram® precoated silica gel TLC sheets SIL G/UV₂₅₄ with detection by UV light (254 nm).

Characterization of all compounds was done using ¹H NMR and ¹³C-NMR spectroscopy and mass spectrometry. ¹H NMR (400 MHz) and ¹³C-NMR (100 MHz) spectra were recorded on a Bruker Avance III Nanobay Ultrashield 400 or a Bruker DPX 400 spectrometer. The chemical shift (δ) values are expressed in parts per million (ppm) and coupling constants are in Hertz (Hz). The chemical shifts δ were given relative to the residual ¹H and ¹³C signals of the solvent peak as an internal standard: in ¹H NMR (400 MHz) δ 2.50 ppm (quin, C₂D₅HOS) for DMSO-*d*₆, δ 2.05 ppm (quin, C₃D₅HO) for Acetone-*d*₆, δ 3.31 ppm (pent, CD₂HOD) for Methanol-*d*₄; in ¹³C NMR (101 MHz) δ 39.51 ppm (sept) for DMSO-*d*₆, δ 29.84 ppm (sept), δ 206.26 ppm (s) for Acetone-*d*₆, δ 49.00 ppm (sept) for Methanol-*d*₄. Legend: s = singlet, d = doublet, t = triplet, q = quartet, quin = quintet, sept = septet, m = multiplet (denotes complex pattern), br = broad signal, dd = doublet of doublets, dt = doublet of triplets, td = triplet of doublets, etc.

UPLC-MS analysis was performed according to the following methods A, B and C. Method A involved the Waters Acquity UPLC system coupled to a Waters SQ detector. A Waters Acquity UPLC BEH C18 1.7 μm, 3 mm × 50 mm column was used. The concentration of the measured samples was 0.1 mg/ml and the flow was 0.8 mL/min. Solvent A consisted of aqueous ammonium acetate 25mM and 10% acetonitrile at pH 6.6 and Solvent B was acetonitrile. The method was as

follows: 0.0-0.2 min A:B 99.9:0.1, 0.2-1.0 min 10:90, 1.0-1.8 min 10:90, 1.9-2.0 min 99.9:0.1 at temperature 40°C. The UV detection was an averaged signal from wavelength of 210 nm to 400 nm. In methods B and C ESI mass spectra were obtained with an Esquire 3000plus ion trap mass spectrometer (Bruker Daltonics), using the direct infusion mode. A Waters acquity H-class UPLC system coupled to a waters TQD ESI mass spectrometer and a Waters TUV detector was used with a Waters acquity UPLC BEH C18 1.7µm 2.1'50 mm column. Solvent A consisted of water with 0.1% formic acid. Solvent B consisted of acetonitrile with 0.1% formic acid. Method B involved the following: flow 0.7 mL/min, 0.15 min isocratic elution (A:B = 95:5), then gradient elution during 1.85 min (A:B = from 95:5 to 0:100), followed by 0.25min of isocratic elution (A:B = 0:100), then 0.75 min of isocratic elution (A:B = 95:5). Method C involved the following: flow 0.4 mL/min, 0.15 min isocratic elution (A:B = 95:5), then gradient elution during 4.85 min (A:B = from 95:5 to 0:100), followed by 0.25min of isocratic elution (A:B = 0:100), then 0.75 min of isocratic elution (A:B = 95:5). In methods B and C the wavelength for UV detection was 254 nm unless stated otherwise.

For the High Resolution Mass Spectrometry (HRMS) measurements positive ion mass spectra were acquired using a QSTAR Elite System (AB Sciex Instruments) mass spectrometer, equipped with a turbospray source, over a mass range of 250–700.

Where necessary, flash purification was performed on a Biotage® ISOLERA One or Four flash systems equipped with an internal variable dual-wavelength diode array detector (200-400nm). For normal phase purifications Biotage SNAP (10-340g), Sylicicle SiliaSep (4-120g) or Götec-Labortechnik EasyVarioFlash (5-100g) cartridges were used (flow rates 10-100mL/min). Reversed phase purifications were performed with Biotage KP-C18 containing cartridges. Gradients used varied for each purification. However, typical gradients used for normal phase were

gradient of 0–100% ethyl acetate in *n*-heptane or cyclohexane, or 0-15% methanol in ethyl acetate. For reverse phase typically a gradient of 5% MeCN in water to 50% MeCN in water was used.

The preparative HPLC purification was conducted on the Agilent 1200 or Agilent 1100 instrument, employing either on an X-Bridge C₁₈ column (19 mm x 150 mm, i.d. 5 µm packing diameter or 30 mm x 150 mm, i.d. 5 µm packing diameter) or a SunFire C₁₈ column (19 mm x 150 mm or 30 mm x 250 mm) at 35°C. The solvents employed were: A = 10 mM ammonium bicarbonate in water; B = acetonitrile (“basic” method) or A = 0.1 M formic acid in water; B = 0.1 M formic acid in acetonitrile (“acidic” method) respectively. The purification was run as a gradient (A:B) typically from 40 to 100% over either 20 min or 25 min, with a flow rate of 17 mL/min (19 mm column) or 35 mL/min (30 mm column). The UV detection wavelengths were 210 nm and 254 nm.

Microwave radiation-assisted reactions were performed in a Biotage Initiator instrument. The initial absorption was set as ‘high’ and 2 min of pre-stirring was applied before heating commenced.

The isolated yields are reported. The purity of all final compounds, tested on *in vitro* and *in vivo* assays, was 95% or higher (unless stated otherwise), verified by UPLC-MS and ¹H NMR data. All products were obtained as amorphous solids, and melting points were not measured.

The following section reports the synthetic procedures and analytical data for all final compounds and some representative intermediates reported in this publication. Complementary data for the rest of the intermediate compounds can be found in the Supporting Information. Synthetic procedures that were used in the preparation of several products are summarized here as “General methods”.

General method A: hydantoin core synthesis. A modified Bucherer-Berg protocol¹³ was employed. The appropriate ketone (1.5.0-4.0 mmol, 1.0 eq.), ammonium carbonate $(\text{NH}_4)_2\text{CO}_3$ (9.0 eq.) and potassium cyanide KCN (1.3 eq.) were dissolved in a mixture of ethanol and water (1:1) (reaction molarity ~ 0.25 - 0.4 mol/l). The reaction mixture was heated at 55°C for 18-48 hr or irradiated in microwave oven at 70°C for 3-9 hours. After the reaction was completed, the reaction mixture was cooled down to room temperature and neutralized with 6M hydrochloric acid to pH $\sim$ 7-8. In case of precipitate formation, the product was collected by filtration, washed with water and dried. Otherwise, the solvent was removed under reduced pressure and the residue was diluted with water and extracted with ethyl acetate; the combined organic phase was washed with brine, dried over Na_2SO_4 , filtered and evaporated to dryness. When necessary the product was additionally dried in the vacuum oven (40°C , 0-10 mbar). Typically no additional purification was performed.

General method B: hydantoin core alkylation. A mixture of the appropriate hydantoin (0.2-3.7 mmol, 1 eq.) and potassium carbonate K_2CO_3 (1.1-2.0 eq.) was dissolved in DMF or acetone. After 10-15 min, the corresponding alkyl halide was added in a slight excess (1.1-1.5 eq.) (reaction molarity ~ 0.08 - 0.2 mol/l). The reaction mixture was stirred at room temperature for 20-72 hr. After the reaction completion, the solvent was removed under reduced pressure and the residue was diluted with saturated ammonium chloride solution or water and extracted with ethyl acetate. The combined organic phase was typically washed with 1M NaOH, brine, dried over Na_2SO_4 , filtered and evaporated under vacuum. The residue was purified by normal phase flash chromatography on silicagel (gradient c-Hex/Hep:EtOAc = 100:0 to 10:90) or reversed-phase flash chromatography (gradient water:ACN = 90:10 to 50:50). The final product was typically lyophilized.

COMPOUND SYNTHESSES

4-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile

(1). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (250 mg, 1.16 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (244 mg, 1.28 mmol) following general method B. White solid, yield 75% (321 mg, 0.869 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.20 (s, 1H), 7.99 (dt, *J* = 6.82, 8.46 Hz, 1H), 7.93 (d, *J* = 8.59 Hz, 2H), 7.75 (d, *J* = 8.34 Hz, 2H), 7.50 (ddd, *J* = 2.40, 9.28, 11.56 Hz, 1H), 7.28 (dt, *J* = 2.27, 8.34 Hz, 1H), 4.81 (d, *J* = 2.27 Hz, 2H), 1.78 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.0 (d, ³*J*_{CF} = 5.12 Hz), 174.3, 165.7 (dd, ¹*J*_{CF} = 255.41 Hz, ³*J*_{CF} = 13.17 Hz), 162.4 (dd, ¹*J*_{CF} = 257.60 Hz, ³*J*_{CF} = 13.17), 154.9, 144.6, 132.7 (dd, ³*J*_{CF} = 5.85, ³*J*_{CF} = 10.24 Hz), 132.6, 126.7, 119.4 (dd, ²*J*_{CF} = 13.17 Hz, ⁴*J*_{CF} = 3.66 Hz), 118.5, 112.8 (dd, ²*J*_{CF} = 21.22 Hz, ⁴*J*_{CF} = 2.93 Hz), 111.0, 105.4 (t, ²*J*_{CF} = 26.30 Hz), 63.2, 47.3 (d, ⁴*J*_{CF} = 10.98 Hz), 24.9. UPLC-MS (A) (ESI) *t*_R = 1.13 min, *m/z* 368 [M-H]⁻ (>95%), HRMS (ESI) *m/z* calcd for C₁₉H₁₃F₂N₃O₃ [M-H]⁻: 368.0852; found: 368.0849.

4-(4-Methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (5). The title compound was prepared from 4-acetylbenzonitrile (1 g, 6.89 mmol) following general method A. White solid, yield 72% (1.06 g, 4.94 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 10.91 (br. s., 1H), 8.73 (s, 1H), 7.88 (d, *J* = 8.34 Hz, 2H), 7.69 (d, *J* = 8.59 Hz, 2H), 1.67 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 176.1, 156.1, 145.2, 132.5, 126.5, 118.5, 110.8, 63.9, 25.1. UPLC-MS (A) *t*_R = 0.87 min; *m/z* 214 [M-H]⁻ (>95%).

(R)- and (S)-4-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (7 and 8). The title compounds were obtained by chiral separation of **1** via preparative HPLC (Chiralpak IC 20x250 cm, Heptane:Ethanol 80:20, 25 min, 18 mL/min) and

were obtained as white solids. Chiral separation >95%. HPLC (Chiralpak IC 0.46x25 cm, Heptane:Ethanol 80:20, 20 min, 1 mL/min) **7**: t_R = 16.05min; **8**: t_R = 11.47min.

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (24). The title compound was prepared from 5-(4-methoxyphenyl)-5-methyl-2,4-imidazolidinedione **22** (100 mg, 0.454 mmol) and **6** (95 mg, 0.50 mmol) following general method B. White solid; yield 25% (45 mg, 0.11 mmol) ^1H NMR (400 MHz, DMSO- d_6) δ 9.00 (s, 1H), 8.00 (td, J = 8.6, 6.8 Hz, 1H), 7.50 (ddd, J = 11.6, 9.3, 2.4 Hz, 1H), 7.41 - 7.46 (m, 2H), 7.28 (td, J = 8.5, 2.3 Hz, 1H), 6.90 - 7.03 (m, 2H), 4.79 (d, J = 2.5 Hz, 2H), 3.76 (s, 3H), 1.72 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $^3J_{CF}$ = 5.1 Hz), 175.4, 165.7 (dd, $^1J_{CF}$ = 255.4 Hz, $^3J_{CF}$ = 13.2 Hz), 162.4 (dd, $^1J_{CF}$ = 256.9 Hz, $^3J_{CF}$ = 13.9 Hz), 159.0, 155.0, 132.6 (dd, $^3J_{CF}$ = 11.0 Hz, $^3J_{CF}$ = 4.4 Hz), 131.3, 126.9, 119.5 (dd, $^2J_{CF}$ = 13.2 Hz, $^4J_{CF}$ = 3.7 Hz), 113.8, 112.8 (dd, $^2J_{CF}$ = 22.0 Hz, $^4J_{CF}$ = 3.7 Hz), 105.4 (t, $^2J_{CF}$ = 27.1 Hz), 62.9 (C5), 55.2, 47.1 (d, $^4J_{CF}$ = 10.3Hz), 24.8. UPLC-MS (A) t_R = 1.16 min, m/z 375 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{19}\text{H}_{17}\text{F}_2\text{N}_2\text{O}_4$ $[\text{M}+\text{H}]^+$: 375.1151; found: 375.1139.

5-(4-Chlorophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (25). The title compound was prepared from 5-(4-chlorophenyl)-5-methylimidazolidine-2,4-dione **23** (25 mg, 0.11 mmol) and **6** (23.33 mg, 0.12 mmol) following general method B. White solid; yield 45% (21 mg, 0.05 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.12 (s, 1H), 7.99 (dt, J = 6.82, 8.46 Hz, 1H), 7.47 - 7.58 (m, 5H), 7.28 (dt, J = 2.40, 8.40 Hz, 1H), 4.80 (d, J = 2.53 Hz, 2H), 1.74 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $^3J_{CF}$ = 4.39 Hz), 174.8, 165.7 (dd, $^1J_{CF}$ = 255.41, $^3J_{CF}$ = 13.17 Hz), 162.4 (dd, $^1J_{CF}$ = 257.60, $^3J_{CF}$ = 13.17 Hz), 155.0, 138.3, 132.9, 132.6 (dd, $^3J_{CF}$ = 10.98, $^3J_{CF}$ = 4.39 Hz), 128.5, 127.6, 119.5 (dd, $^2J_{CF}$ = 13.17, $^4J_{CF}$ = 3.66, Hz),

112.8 (dd, $^2J_{CF} = 21.96$, $^4J_{CF} = 2.93$ Hz), 105.4 (t, $^2J_{CF} = 27.10$ Hz), 62.9, 47.2 (d, $^4J_{CF} = 10.98$ Hz), 24.9. UPLC-MS (A) $t_R = 0.87$ min, m/z 379 $[M+H]^+$ (>95%).

4-(1-(2-(4-Fluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile

(26). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (50 mg, 0.23 mmol) and 2-chloro-4'-fluoroacetophenone **9** (44.1 mg, 0.26 mmol) following general method B. White solid; yield 74% (60 mg, 0.17 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.21 (s, 1H), 8.12 (dd, $J = 5.56$, 8.84 Hz, 2H), 7.94 (d, $J = 8.59$ Hz, 2H), 7.77 (d, $J = 8.59$ Hz, 2H), 7.41 (t, $J = 8.72$ Hz, 2H), 4.99 (s, 2H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 190.8, 174.5, 165.6 (d, $^1J_{CF} = 253.22$ Hz), 155.1, 144.6, 132.6, 131.3 (d, $^3J_{CF} = 9.51$ Hz), 130.7 (d, $^4J_{CF} = 2.93$ Hz), 126.7, 118.5, 116.1 (d, $^2J_{CF} = 21.22$ Hz), 111.0, 63.3, 44.6, 25.0. UPLC-MS (A) $t_R = 1.12$ min, m/z 350 $[M+H]^+$ (>95%). HRMS (ESI) m/z calcd for $C_{19}H_{14}FN_3O_3$ $[M+H]^+$: 374.0911; found: 374.0908.

3-(2-(4-Fluorophenyl)-2-oxoethyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione

(27). The title compound was prepared from 5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione **22** (100 mg, 0.454 mmol) and 2-chloro-4'-fluoroacetophenone **9** (78 mg, 0.45 mmol) following general method B. White solid; yield 69% (112 mg, 0.314 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.99 (s, 1H), 8.07 - 8.19 (m, 2H), 7.35 - 7.51 (m, 4H), 6.94 - 7.03 (m, 2H), 4.96 (s, 2H), 3.76 (s, 3H), 1.73 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 191.0, 175.6, 165.5 (d, $^1J_{CF} = 251.0$ Hz), 159.0, 155.2, 131.3, 131.2 (d, $^3J_{CF} = 10.3$ Hz), 130.8 (d, $^4J_{CF} = 2.2$ Hz), 126.9, 116.1 (d, $^3J_{CF} = 22.7$ Hz), 113.8, 62.9, 55.2, 44.4, 24.8. UPLC-MS (A) $t_R = 1.14$ min, m/z 357 $[M+H]^+$ (>95%).

5-(4-Chlorophenyl)-3-(2-(4-fluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione

(28). The title compound was prepared from 5-(4-chlorophenyl)-5-methylimidazolidine-2,4-dione

23 (25 mg, 0.111 mmol) and 2-chloro-4'-fluoroacetophenone **9** (19.21 mg, 0.111 mmol) following general method B. White solid; yield 80% (32 mg, 0.089 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.11 (s, 1H), 8.13 (dd, $J = 5.56, 8.84$ Hz, 2H), 7.49 - 7.60 (m, 4H), 7.41 (t, $J = 8.84$ Hz, 2H), 4.98 (s, 2H), 1.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 190.9, 175.0, 165.5 (d, $^1J_{CF} = 253.95$ Hz), 155.1, 138.4, 132.9, 131.3 (d, $^3J_{CF} = 10.25$ Hz), 130.7 (d, $^4J_{CF} = 2.93$ Hz), 128.5, 127.6, 116.1 (d, $^2J_{CF} = 21.22$ Hz), 62.9, 44.5, 25.0. UPLC-MS (A) $t_R = 1.20$ min, m/z 359 $[\text{M-H}]^-$ (>95%).

4-(1-(4-fluorophenethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (29). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (50 mg, 0.232 mmol) and 1-(2-bromoethyl)-4-fluorobenzene **10** (51.9 mg, 0.256 mmol) following general method B. White solid; yield 84% (66 mg, 0.196 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 8.95 (br. s., 6H), 7.85 (d, $J = 8.34$ Hz, 13H), 7.55 (d, $J = 8.59$ Hz, 13H), 7.07 (dd, $J = 5.68, 8.46$ Hz, 13H), 6.96 (t, $J = 8.84$ Hz, 12H), 3.55 - 3.66 (m, 12H), 2.82 (t, $J = 6.82$ Hz, 12H), 1.58 (s, 19H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 174.2, 160.9 (d, $^1J_{CF} = 241.51$ Hz), 155.2, 144.7, 133.9 (d, $^4J_{CF} = 2.93$ Hz), 132.4, 130.6 (d, $^3J_{CF} = 8.05$ Hz), 126.5, 118.5, 114.8 (d, $^2J_{CF} = 20.49$ Hz), 110.8, 62.6, 39.0, 31.9, 24.9. UPLC-MS (A) $t_R = 1.15$ min, m/z 336 $[\text{M-H}]^-$ (>95%).

3-(4-Fluorophenethyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (30). The title compound was prepared from 5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione **22** (200 mg, 0.91 mmol) and 1-(2-bromoethyl)-4-fluorobenzene **10** (184 mg, 0.91 mmol) following general method B. White solid; yield 78% (242 mg, 0.707 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 8.76 (s, 1H), 7.18 - 7.27 (m, 2H), 7.06 - 7.16 (m, 2H), 6.96 - 7.06 (m, 2H), 6.85 - 6.95 (m, 2H), 3.74 (s, 3H), 3.52 - 3.66 (m, 2H), 2.83 (t, $J = 6.8$ Hz, 2H), 1.53 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 175.3, 160.9 (d, $^1J_{CF} = 242.2$ Hz), 158.8, 155.4, 134.0 (d, $^4J_{CF} = 2.9$ Hz), 131.4, 130.6

(d, $^3J_{CF}$ = 8.1 Hz), 126.6, 114.9 (d, $^2J_{CF}$ = 21.2 Hz), 113.7, 62.1, 55.1, 38.8, 32.0, 24.7. UPLC-MS (A) t_R = 1.17 min, m/z 341 $[M-H]^-$ (>95%).

5-(4-Chlorophenyl)-3-(4-fluorophenethyl)-5-methylimidazolidine-2,4-dione (31). The title compound was prepared from 5-(4-chlorophenyl)-5-methylimidazolidine-2,4-dione **23** (50 mg, 0.223 mmol) and 1-(2-bromoethyl)-4-fluorobenzene **10** (0.031 ml, 0.223 mmol) following general method B. White solid; yield 95% (73 mg, 0.211 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.86 (s, 1H), 7.40 - 7.48 (m, 2H), 7.32 - 7.39 (m, 2H), 7.06 - 7.17 (m, 2H), 6.95 - 7.02 (m, 2H), 3.60 (dt, J = 2.78, 6.82 Hz, 2H), 2.83 (t, J = 6.95 Hz, 2H), 1.55 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ ^{13}C NMR (101 MHz, DMSO- d_6) δ 174.7, 160.9 (d, $^1J_{CF}$ = 242.24 Hz), 155.3, 138.5, 134.0 (d, $^4J_{CF}$ = 2.93 Hz), 132.7, 130.6 (d, $^3J_{CF}$ = 8.05 Hz), 128.3, 127.3, 114.9 (d, $^2J_{CF}$ = 21.22 Hz), 62.2, 32.0, 24.8. UPLC-MS (A) t_R = 1.24 min, m/z 345 $[M-H]^-$ (>95%).

4-(1-(4-Fluorobenzyl)-4-methyl-2,5-dioximidazolidin-4-yl)benzonitrile (32). The title compound was prepared from 4-(4-methyl-2,5-dioximidazolidin-4-yl)benzonitrile **5** (50 mg, 0.232 mmol) and 1-(bromomethyl)-4-fluorobenzene **11** (0.029 ml, 0.232 mmol) following general method B. White solid; yield 75% (56 mg, 0.174 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.17 (s, 1H), 7.84 - 7.93 (m, 2H), 7.63 - 7.73 (m, 2H), 7.21 - 7.30 (m, 2H), 7.08 - 7.19 (m, 2H), 4.53 (s, 2H), 1.71 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 174.3, 161.4 (d, $^1J_{CF}$ = 239.3 Hz), 155.2, 144.6, 132.7 (d, $^4J_{CF}$ = 2.9 Hz), 132.6, 129.4 (d, $^3J_{CF}$ = 8.0 Hz), 126.5, 118.4, 115.4 (d, $^2J_{CF}$ = 22.0 Hz), 111.0, 62.9, 40.7, 25.1. UPLC-MS (A) t_R = 1.14 min, m/z 322 $[M-H]^-$ (>95%).

3-(4-Fluorobenzyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (33). The title compound was prepared from 5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione **22** (100 mg, 0.45 mmol) and 1-(bromomethyl)-4-fluorobenzene **11** (86 mg, 0.45 mmol) following general method B. White solid; yield 79% (118 mg, 0.359 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.97 (s,

1H), 7.31 - 7.41 (m, 2H), 7.20 - 7.30 (m, 2H), 7.10 - 7.19 (m, 2H), 6.88 - 6.98 (m, 2H), 4.52 (s, 2H), 3.74 (s, 3H), 1.65 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 175.3, 161.4 (d, ¹J_{CF} = 243.0 Hz), 158.9, 155.3, 132.9 (d, ⁴J_{CF} = 2.9 Hz), 131.3, 129.4 (d, ³J_{CF} = 8.8 Hz), 126.6, 115.3 (d, ²J_{CF} = 21.2 Hz), 113.9, 62.5, 55.1, 40.5, 25.0. UPLC-MS (A) t_R = 1.16 min, m/z 327 [M-H]⁻ (>95%).

5-(4-Chlorophenyl)-3-(4-fluorobenzyl)-5-methylimidazolidine-2,4-dione (34). The title compound was prepared from 5-(4-chlorophenyl)-5-methylimidazolidine-2,4-dione **23** (50 mg, 0.223 mmol) and 1-(bromomethyl)-4-fluorobenzene **11** (0.027 ml, 0.223 mmol) following general method B. White solid; yield 70% (52 mg, 0.156 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.08 (s, 1H), 7.44 - 7.51 (m, 4H), 7.21 - 7.27 (m, 2H), 7.11 - 7.18 (m, 2H), 4.53 (s, 2H), 1.68 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 174.7, 161.4 (d, ¹J_{CF} = 242.97 Hz), 155.2, 138.4, 132.8, 132.8 (d, ⁴J_{CF} = 2.93 Hz), 129.3 (d, ³J_{CF} = 8.05 Hz), 128.5, 127.3, 115.4 (d, ²J_{CF} = 21.22 Hz), 62.6, 40.6, 25.1. UPLC-MS (A) t_R = 1.23 min, m/z 331 [M-H]⁻ (>95%).

4-(1-(3-(4-Fluorophenyl)-3-oxopropyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (35). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (100 mg, 0.465 mmol) and 3-chloro-1-(4-fluorophenyl)propan-1-one **12** (95 mg, 0.511 mmol) following general method B. Off-white solid; yield 10% (20.4 mg, 0.047 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.04 (s, 1H), 7.95-8.02 (m, 2H), 7.85-7.90 (m, 2H), 7.64-7.70 (m, 2H), 7.26-7.36 (m, 2H), 3.72 (t, J = 7.07 Hz, 2H), 3.27-3.36* (m, 2H), 1.68 (s, 3H). *Multiplet overlaps with the water peak. ¹³C NMR (101 MHz, DMSO-d₆) δ 196.5, 174.2, 165.1 (d, ¹J_{CF} = 251.7 Hz), 155.2, 144.7, 132.9 (d, ⁴J_{CF} = 2.9 Hz), 132.4, 130.9 (d, ³J_{CF} = 9.5 Hz), 126.6, 118.5, 115.7 (d, ²J_{CF} = 22.0 Hz), 110.9, 62.7, 35.8, 34.1, 25.0. UPLC-MS (A) t_R = 1.13 min, m/z 366 [M+H]⁺ (>90%).

3-(3-(4-Fluorophenyl)-3-oxopropyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (36). The title compound was prepared from 5-(4-methoxyphenyl)-5-methylimidazolidine-

2,4-dione **22** (100 mg, 0.45 mmol), and 3-chloro-1-(4-fluorophenyl)propan-1-one **12** (85 mg, 0.45 mmol) following general method B. White solid; 25% (42 mg, 0.11 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 8.84 (s, 1H), 7.94 - 8.04 (m, 2H), 7.28 - 7.40 (m, 4H), 6.89 - 6.96 (m, 2H), 3.74 (s, 3H), 3.71 (t, J = 7.1 Hz, 2H), 3.27-3.36* (m, 2H), 1.62 (s, 3H). *Multiplet overlaps with the water peak. ^{13}C NMR (101 MHz, DMSO- d_6) δ 196.5, 175.3, 165.1 (d, $^1J_{CF}$ = 252.3 Hz), 158.8, 155.3, 132.9 (d, $^4J_{CF}$ = 2.9 Hz), 131.4, 131.0 (d, $^3J_{CF}$ = 9.7 Hz), 126.7, 115.7 (d, $^2J_{CF}$ = 22.2 Hz), 113.8, 62.2, 55.1, 36.0, 33.8, 24.8. UPLC-MS (A) t_R = 1.14 min, m/z 371 $[\text{M}+\text{H}]^+$ (>95%).

5-(4-Chlorophenyl)-3-(3-(4-fluorophenyl)-3-oxopropyl)-5-methylimidazolidine-2,4-dione (37). The title compound was prepared from 5-(4-chlorophenyl)-5-methylimidazolidine-2,4-dione **23** (25 mg, 0.111 mmol), and 3-chloro-1-(4-fluorophenyl)propan-1-one **12** (22.84 mg, 0.122 mmol) following general method B. White solid; 45.6 % yield (19 mg, 0.051 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 8.94 (s, 1H), 7.99 (dd, J = 5.68, 8.46 Hz, 2H), 7.43 - 7.50 (m, 4H), 7.32 (t, J = 8.72 Hz, 2H), 3.71 (t, J = 6.95 Hz, 2H), 3.26 - 3.36* (m, 2H), 1.64 (s, 3H). *Multiplet overlaps with the water peak. ^{13}C NMR (101 MHz, DMSO- d_6) δ 196.5, 174.7, 165.1 (d, $^1J_{CF}$ = 252.48 Hz), 155.3, 138.5, 132.9 (d, $^4J_{CF}$ = 2.93 Hz), 132.7, 131.0 (d, $^3J_{CF}$ = 9.51 Hz), 128.4, 127.4, 115.7 (d, $^2J_{CF}$ = 22.69 Hz), 62.3, 35.9, 34.0, 25.0. UPLC-MS (A) t_R = 1.21 min, m/z 375 $[\text{M}+\text{H}]^+$ (>95%).

4-(1-(1-(2,4-Difluorophenyl)-1-oxopropan-2-yl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (38). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (50 mg, 0.232 mmol), and 2-bromo-1-(2,4-difluorophenyl)propan-1-one **13** (63.6 mg, 0.256 mmol) following general method B. White solid; 74.8 % yield (321 mg, 0.869 mmol). Note: the product is a diastereoisomeric mixture and was not further separated. The ratio of isomers is approx. A:B = 1:1.35 as judged by NMR. Based on the relative integration intensity

the peaks can be assigned as such: ^1H NMR (400 MHz, DMSO- d_6) δ isomer A: 9.18 (s, 1H), 7.76 (d, $J = 8.59$ Hz, 2H), 7.43 - 7.48 (m, 1H)*, 7.41 (d, $J = 8.59$ Hz, 2H), 7.05 - 7.13 (m, 1H), 7.01 (dt, $J = 2.27, 8.46$ Hz, 1H)*, 5.17 (q, $J = 6.57$ Hz, 1H), 1.57 (s, 3H), 1.40 (d, $J = 6.82$ Hz, 3H)*; isomer B: 9.10 (s, 1H), 7.84 (d, $J = 8.59$ Hz, 2H), 7.52 - 7.63 (m, 1H), 7.48 (d, $J = 8.34$ Hz, 2H)*, 7.17 - 7.29 (m, 1H), 7.05 - 7.13 (m, 1H)*, 5.24 (q, $J = 6.74$ Hz, 1H), 1.52 (s, 3H), 1.41 (d, $J = 6.82$ Hz, 3H)*. The mentioned relative integration intensities are calculated for one isomer. * Indicated peaks overlap with peaks of the other isomer at the corresponding chemical shift. UPLC-MS (A) $t_R = 1.40$ min, m/z 382 $[\text{M-H}]^-$ (>95%).

4-(1-(5,7-Difluoro-1-oxo-2,3-dihydro-1H-inden-2-yl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (39). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (50 mg, 0.232 mmol), and 2-bromo-5,7-difluoro-2,3-dihydro-1H-inden-1-one **14** (63.1 mg, 0.256 mmol) following general method B. White solid; 31% (27 mg, 0.071 mmol). Note: the product is a diastereoisomeric mixture and therefore produces a complex NMR image. ^1H NMR (400 MHz, DMSO- d_6) δ 9.29 (s, 1H), 7.90 - 7.97 (m, 2H), 7.68 - 7.75 (m, 2H), 7.30 - 7.38 (m, 2H), 4.99 (ddd, $J = 3.41, 5.05, 8.46$ Hz, 1H), 3.53 (dd, $J = 8.84, 17.43$ Hz, 1H), 3.25 (dd, $J = 5.18, 17.56$ Hz, 1H), 1.75 (s, 3H). UPLC-MS (A) $t_R = 1.19$ min, m/z 380 $[\text{M-H}]^-$ (>95%).

4-(1-(2-(2,4-Difluorophenoxy)ethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (40). To a stirred solution of 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) in DMF (2 ml), sodium hydride (14.31 mg, 0.358 mmol) was added and the mixture was stirred for 15 min. Subsequently a solution of 2-(2,4-difluorophenoxy)ethyl methanesulfonate **15** (98 mg, 0.390 mmol) in 1 ml DMF was added dropwise. The resulting mixture was stirred for 72 h at room temperature. The reaction mixture was diluted with ethyl acetate and washed with saturated ammonium chloride and brine, dried over Na_2SO_4 , filtered and evaporated to dryness.

The residue was purified by preparative HPLC (Sunfire 19, ACN:Water 40-100%, 0.1% formic acid). Off-white solid; yield 3.5% (5.0 mg, 0.011 mmol). ¹H NMR (400 MHz, Methanol-d₄) δ 7.69-7.78 (m, 4H), 7.03 (dt, *J* = 5.31, 9.22 Hz, 1H), 6.92 (ddd, *J* = 2.91, 8.53, 11.31 Hz, 1H), 6.76-6.84 (m, 1H), 4.22 - 4.27 (m, 2H), 3.89 (t, *J* = 5.43 Hz, 2H), 1.79 (s, 3H). *NH signal was not detected. UPLC-MS (A) *t_R* = 1.51 min, *m/z* 370 [M-H]⁻ (>85%).

4-(1-(2-(2,4-Difluorophenyl)-2,2-difluoroethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (41). 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (80 mg, 0.372 mmol) and potassium carbonate (77 mg, 0.558 mmol) were suspended in dry DMF (Volume: 3 ml) and stirred at room temperature for 5 minutes. 1-(2-Bromo-1,1-difluoroethyl)-2,4-difluorobenzene **17** (143 mg, 0.558 mmol) was added and the reaction was left stirring at 100 °C for 2 days. The mixture was cooled down to room temperature and concentrated *in vacuo*. The residue was added to water and the precipitate was filtered, washed with water, dried and purified via flash column chromatography on silicagel (EtOAc:Heptane 5-50%). The product was dissolved in a mixture of acetonitrile/water and lyophilized. White solid; 65% (94.5 mg, 0.241 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.16 (s, 1H), 7.89 (d, *J* = 8.53 Hz, 2H), 7.57 (d, *J* = 8.53 Hz, 2H), 7.26 - 7.46 (m, 2H), 7.03 (dt, *J* = 2.01, 8.53 Hz, 1H), 4.15 (t, *J* = 13.43 Hz, 2H), 1.63 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 173.9, 163.9 (dd, ¹*J*_{CF} = 251.64 Hz, ³*J*_{CF} = 12.47 Hz), 159.8 (dd, ¹*J*_{CF} = 253.84 Hz, ³*J*_{CF} = 13.21 Hz), 154.2, 144.4, 132.6, 128.8, 126.4, 118.5, 117.5 (ddt, ⁴*J*_{CF} = 3.70 Hz, ²*J*_{CF} = 11.70 Hz, ²*J*_{CF} = 27.10 Hz), 118.4 (dt, ⁴*J*_{CF} = 2.20 Hz, ¹*J*_{CF} = 245.80 Hz), 111.6 (dd, ⁴*J*_{CF} = 3.67, ²*J*_{CF} = 22.01 Hz), 111.1, 105.2 (t, ²*J*_{CF} = 26.40 Hz), 62.9, 42.9 (t, ²*J*_{CF} = 33.70 Hz), 24.7. UPLC-MS (B) *t_R* = 1.73 min, *m/z* 390 [M-H]⁻ (>95%).

4-(1-((3-(2,4-Difluorophenyl)oxetan-3-yl)methyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (42). 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (25 mg, 0.071

mmol), (3-(2,4-difluorophenyl)oxetan-3-yl)methyl 4-methylbenzenesulfonate **21** (30.4 mg, 0.141 mmol) and potassium carbonate (10.73 mg, 0.078 mmol) were suspended in dry DMF (500 μ l) and stirred at 80 °C for 24 h and then at 100 °C for another 24 h. The mixture was cooled down to room temperature and concentrated *in vacuo*. The residue was suspended in a mixture of EtOAc and water. The water phase was extracted with ethyl acetate (3x) and the organic phases were combined, washed 2x with brine, dried with Na₂SO₄ and evaporated. The residue was purified via preparative HPLC (Sunfire 19, ACN:Water 40-100%, 0.1% formic acid) and lyophilized. White solid; 22% (6.3 mg, 0.016 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.00 (s, 1H), 7.85 (d, *J* = 8.34 Hz, 2H), 7.51 (d, *J* = 8.34 Hz, 2H), 6.85 - 7.04 (m, 2H), 6.77 (dt, *J* = 2.27, 8.46 Hz, 1H), 4.69 - 4.89 (m, 4H), 3.95 - 4.12 (m, 2H), 1.53 (s, 3H). UPLC-MS (A) *t_R* = 1.09 min, *m/z* 396 [M-H]⁻ (>95%).

(E/Z)-4-(1-(2-(2,4-difluorophenyl)-2-(hydroxyimino)ethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (43a, 43b). A solution of 4-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **1** (50 mg, 0.135 mmol) and hydroxylamine hydrochloride (47.0 mg, 0.677 mmol) in a mixture of pyridine (500 μ L) and ethanol (500 μ L) was irradiated in a microwave at 100 °C for 30 minutes, cooled down to rt, poured into saturated sodium bicarbonate (5ml) and extracted with DCM (3x5ml). Combined organic layers were washed with brine (10 ml), dried with Na₂SO₄ and evaporated. The residue was purified via preparative HPLC (Sunfire 19, ACN:Water 40-100%, 0.1% formic acid) to provide two isomeric products. **43a**, white solid; 27% (14 mg, 0.036 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 11.84 (s, 1H), 9.00 (s, 1H), 7.77 (d, *J* = 8.59 Hz, 2H), 7.40 (d, *J* = 8.59 Hz, 2H), 7.09 - 7.15 (m, 1H), 6.95 - 7.01 (m, 1H), 6.84 - 6.89 (m, 1H), 4.52 - 4.58 (m, 2H), 1.48 (s, 3H). UPLC-MS (A) *t_R* = 1.37 min, *m/z* 383 [M-H]⁻ (>95%). **43b**, white solid; 13% (7 mg, 0.018 mmol). ¹H NMR

(400 MHz, DMSO- d_6) δ 11.28 (s, 1H), 9.02 (s, 1H), 7.84 (d, J = 8.59 Hz, 2H), 7.54 (d, J = 8.34 Hz, 2H), 7.11 - 7.26 (m, 2H), 6.97 - 7.05 (m, 1H), 4.30 - 4.45 (m, 2H), 1.60 (s, 3H). UPLC-MS (A) t_R = 1.36 min, m/z 383 [M-H] $^-$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-hydroxyethyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (44). **24** (100 mg, 0.267 mmol) was dissolved in anhydrous toluene (50 mL) and sodium bis(2-methoxyethoxy)aluminum hydride (Red-Al) solution in toluene (0.16 mL, 0.53 mmol) was added to the mixture under nitrogen atmosphere. The reaction mixture was heated at 120 °C for 1 h. Subsequently, the reaction mixture was cooled down and quenched by careful addition of aqueous NaOH (2 M, 2 mL) followed by addition of ethyl acetate (20 mL) and water (70 mL). The resulting mixture was separated and the organic layer was washed with water (2x 50 mL) and evaporated. The crude product was purified by flash column chromatography on silicagel (EtOAc:cyclohexane 0-50%). White solid; 50% (50 mg, 0.13 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.76 (d, J = 5.6 Hz, 1H), 7.40 - 7.52 (m, 1H), 7.21 - 7.33 (m, 2H), 6.95 - 7.17 (m, 2H), 6.84 - 6.94 (m, 2H), 5.71 - 5.78 (m, 1H), 5.09 (q, J = 6.5 Hz, 1H), 3.75 (d, J = 0.8 Hz, 3H), 3.45 - 3.63 (m, 2H), 1.55 (d, J = 14.1 Hz, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) (mixture of diastereoisomers) δ ppm 175.3, 175.2, 160.1 - 162.9 (m), 158.8, 158.8 (s), 157.9 - 160.7 (m), 155.4, 155.4, 131.5, 131.4, 129.2 - 129.7 (m), 126.7, 126.6, 125.4 - 125.9 (m), 113.7, 113.6, 111.1 - 111.6 (m), 103.1 (m), 62.8, 62.7, 62.2, 62.2, 55.1, 43.9, 43.8, 24.8. UPLC-MS (A) t_R = 1.18-1.19 min, m/z 375 [M-H] $^-$ (>95%).

1-(2,4-Difluorobenzoyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (46). 5-(4-methoxyphenyl)-5-methyl-2,4-imidazolidinedione **22** (100 mg, 0.454 mmol) was dissolved in anhydrous pyridine (1 mL) and 2,4-difluorobenzoyl chloride **45** (80 mg, 0.45 mmol) was added dropwise to the mixture under nitrogen atmosphere. The reaction mixture was left stirring at room

temperature overnight. The solvent was removed under reduced pressure and the obtained residue was dissolved in ethyl acetate (10 mL). The solution was filtered through a 2-cm thick pad of silicagel which was subsequently washed with ethyl acetate (5 mL x 2). The combined filtrates were concentrated by rotary evaporation to provide the crude product which was purified by flash column chromatography on silicagel (EtOAc:Cyclohexane 20%). Subsequently, the organic phase was washed with saturated aqueous sodium bicarbonate (3x 40 mL) and water (40 mL), dried with MgSO₄ and evaporated. Off-white solid; 63% (103 mg, 0.286 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 12.04 (br. s., 1H), 7.59 (td, *J* = 8.3, 6.4 Hz, 1H), 7.30 - 7.40 (m, 3H), 7.17 (td, *J* = 8.5, 2.3 Hz, 1H), 6.93 - 7.02 (m, 2H), 3.76 (s, 3H), 2.05 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 174.8, 163.5 (dd, ¹*J*_{CF} = 250.3 Hz, ³*J*_{CF} = 12.4 Hz), 161.7, 159.0, 159.1 (dd, ¹*J*_{CF} = 250.3 Hz, ³*J*_{CF} = 13.2 Hz), 153.7, 130.9 (dd, ³*J*_{CF} = 10.2 Hz, ³*J*_{CF} = 4.4 Hz), 129.0, 126.7, 121.3 (dd, ²*J*_{CF} = 15.7 Hz, ⁴*J*_{CF} = 4.0 Hz), 114.0, 111.7 (dd, ²*J*_{CF} = 22.0 Hz, ⁴*J*_{CF} = 3.7 Hz), 104.0 (t, ²*J*_{CF} = 26.3 Hz), 68.1, 55.2, 20.3. UPLC-MS (A) *t*_R = 1.06 min, *m/z* 361 [M+H]⁺ (>95%).

5-(4-Chlorophenyl)-1-(2,4-difluorobenzoyl)-5-methylimidazolidine-2,4-dione (47). 5-(4-chlorophenyl)-5-methyl-2,4-imidazolidinedione **22** (50 mg, 0.223 mmol) was dissolved in dry pyridine (2 ml) under nitrogen. 2,4-Difluorobenzoyl chloride **45** (0.027 ml, 0.223 mmol) was added and the reaction was left stirring at room temperature overnight. The reaction mixture was concentrated *in vacuo*, redissolved in ethyl acetate, washed 3x with aqueous ammonium chloride, and 3x with aqueous sodium bicarbonate, dried with sodium sulfate and evaporated. Flash column chromatography on silicagel (EtOAc:Cyclohexane 0-30%) provided crude fractions containing 5-(4-chlorophenyl)-1-(2,4-difluorobenzoyl)-5-methylimidazolidine-2,4-dione and 5-(4-chlorophenyl)-1,3-bis(2,4-difluorobenzoyl)-5-methylimidazolidine-2,4-dione. 40 mg of the crude mixture was dissolved in a mixture of methanol (2 ml) and water (2 ml). Potassium carbonate

(10.95 mg, 0.079 mmol) was added and the solution was stirred at room temperature for 10 minutes. The reaction mixture was evaporated *in vacuo* and the residue was dissolved in a mixture of ethyl acetate and water. Water phase was extracted with ethyl acetate. Organic phases were combined and extracted with aqueous sodium bicarbonate and water, dried with sodium sulfate, evaporated with silicagel and purified via flash column chromatography on silicagel (EtOAc:Cyclohexane 0-30%). White solid, 26% (21 mg, 0.058 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 12.14 (br. s., 1H), 7.63 (dt, *J* = 6.57, 8.34 Hz, 1H), 7.44 - 7.55 (m, 4H), 7.38 (ddd, *J* = 2.40, 9.41, 10.67 Hz, 1H), 7.19 (dt, *J* = 2.15, 8.53 Hz, 1H), 2.08 (s, 3H). UPLC-MS (A) *t_R* = 1.08 min, *m/z* 363 [M-H]⁻ (>95%).

3-(2,4-Difluorobenzoyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (48). 5-(4-methoxyphenyl)-5-methyl-2,4-imidazolidinedione **22** (100 mg, 0.454 mmol) and sodium hydride (60% dispersion in mineral oil) (22mg, 0.92 mmol) were dissolved in anhydrous DMF (1 mL) and the mixture was stirred at room temperature for 15 min. Subsequently, 2,4-difluorobenzoyl chloride **45** (0.056 μL, 0.45 mmol) was added to the mixture under nitrogen atmosphere and stirred at room temperature overnight. The reaction mixture was poured into water (40 mL) and the target compound was extracted with ethyl acetate (3x 50 mL). The combined organic layers were evaporated. Attempts to purify the product via silica gel column chromatography or HPLC provided only a partially hydrolyzed product. The product hydrolyzed further in solution. ¹H NMR (400 MHz, DMSO-d₆) δ 9.47 (s, 1H), 7.84 (td, *J* = 8.5, 6.4 Hz, 1H), 7.40 - 7.49 (m, 3H), 7.27 (td, *J* = 8.5, 2.0 Hz, 1H), 6.97 - 7.04 (m, 2H), 3.74 - 3.80 (m, 3H), 1.77 (s, 3H). UPLC-MS (A) *t_R* = 1.08 min, *m/z* 361 [M+H]⁺ (90%).

5-(4-Bromophenyl)-5-methylimidazolidine-2,4-dione (79). The title compound was prepared from 1-(4-bromophenyl)ethan-1-one **49** (2.3 g, 11.56 mmol) following general method A. White

solid, yield 96% (2.97 g, 11.06 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 10.82 (br. s., 1H), 8.64 (s, 1H), 7.60 (d, $J = 8.59$ Hz, 2H), 7.43 (d, $J = 8.59$ Hz, 2H), 1.64 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 176.5, 156.1, 139.4, 131.4, 127.6, 121.2, 63.6, 25.0. UPLC-MS (A) $t_{\text{R}} = 1.11$ min; m/z 267 $[\text{M-H}]^-$ (>95%).

5-(4-(Difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione (84). The title compound was prepared from 1-(4-(difluoromethoxy)phenyl)ethanone **54** (500 mg, 2.69 mmol) following general method A. White solid, yield 95% (651 mg, 2.54 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 10.79 (br. s., 1H), 8.62 (s, 1H), 7.48 - 7.54 (m, 2H), 7.03 - 7.43 (m, 3H), 1.64 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 176.8, 156.1, 150.5 (t, $^3J_{\text{CF}} = 3.3$ Hz), 136.8, 127.1, 118.7, 116.3 (t, $^1J_{\text{CF}} = 258.3$ Hz), 63.5, 25.0. UPLC-MS (A) $t_{\text{R}} = 1.07$ min; m/z 255 $[\text{M-H}]^-$ (>95%).

5-(4-Bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (109). The title compound was prepared from 5-(4-bromophenyl)-5-methylimidazolidine-2,4-dione **79** (800 mg, 2.97 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (623 mg, 3.27 mmol) following general method B. The product was purified via crystallization from cold DCM. White solid, yield 64% (804 mg, 1.900 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.11 (s, 1H), 7.96 - 8.03 (m, 1H), 7.64 (d, $J = 8.59$ Hz, 2H), 7.46 - 7.54 (m, 3H), 7.25 - 7.31 (m, 1H), 4.80 (d, $J = 2.53$ Hz, 2H), 1.74 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J = 5.12$ Hz), 174.8, 165.7 (dd, $J = 12.44, 255.41$ Hz), 162.4 (dd, $J = 13.17, 257.61$ Hz), 155.0, 138.8, 132.6 (dd, $J = 4.39, 10.98$ Hz), 131.4, 127.9, 121.5, 119.5 (dd, $J = 3.66, 13.17$ Hz), 112.8 (dd, $J = 2.93, 21.96$ Hz), 105.4 (m, $J = 27.10, 27.10$ Hz), 62.9, 47.2 (d, $J = 10.98$ Hz), 24.8. UPLC-MS (A) $t_{\text{R}} = 4.04$ min; m/z 421 $[\text{M-H}]^-$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(p-tolyl)imidazolidine-2,4-dione (110). The title compound was prepared from 5-methyl-5-(p-tolyl)imidazolidine-2,4-dione **80** (105 mg,

0.514 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (147 mg, 0.771 mmol) following general method B. White solid, yield 74% (144 mg, 0.382 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.02 (s, 1H), 8.00 (dt, $J=6.57, 8.59$ Hz, 1H), 7.50 (ddd, $J=2.40, 9.28, 11.56$ Hz, 1H), 7.41 (d, $J=8.34$ Hz, 2H), 7.20-7.32 (m, 3H), 4.79 (d, $J=2.78$ Hz, 2H), 2.31 (s, 3H), 1.73 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J=5.1$ Hz), 175.3, 165.7 (dd, $J=255.4, 12.4$ Hz), 162.4 (dd, $J=257.6, 13.2$ Hz), 155.1, 137.3, 136.4, 132.6 (dd, $J=11.0, 4.4$ Hz), 129.0, 125.5, 119.5 (dd, $J=13.2, 3.7$ Hz), 112.8 (dd, $J=22.0, 3.2$ Hz), 105.4 (t, $J=26.8$ Hz), 63.1, 47.1 (d, $J=10.2$ Hz), 24.7, 20.6. UPLC-MS (A) $t_R = 1.26\text{min}$; m/z 359 $[\text{M}+\text{H}]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-(4-ethoxyphenyl)-5-methylimidazolidine-2,4-dione (111). The title compound was prepared from 5-(4-ethoxyphenyl)-5-methylimidazolidine-2,4-dione **81** (105 mg, 0.514 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (88 mg, 0.38 mmol) following general method B. White solid, yield 22% (32 mg, 0.082 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 8.99 (s, 1H), 8.00 (td, $J = 8.6, 6.8$ Hz, 1H), 7.50 (ddd, $J = 11.6, 9.2, 2.4$ Hz, 1H), 7.39 - 7.46 (m, 2H), 7.29 (td, $J = 8.5, 2.3$ Hz, 1H), 6.92 - 7.00 (m, 2H), 4.79 (d, $J = 2.5$ Hz, 2H), 4.03 (q, $J = 7.1$ Hz, 2H), 1.72 (s, 3H), 1.33 (t, $J = 6.9$ Hz, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $^3J_{CF} = 5.1$ Hz), 175.5, 165.7 (dd, $^1J_{CF} = 255.4$ Hz, $^3J_{CF} = 13.2$ Hz), 162.4 (dd, $^1J_{CF} = 259.8$ Hz, $^3J_{CF} = 13.2$ Hz), 158.2, 155.0, 132.6 (dd, $^3J_{CF} = 11.7$ Hz, $^3J_{CF} = 3.7$ Hz), 131.1, 126.9, 119.5 (dd, $^2J_{CF} = 13.2$ Hz, $^4J_{CF} = 3.7$ Hz), 114.3, 112.8 (dd, $^2J_{CF} = 22.0$ Hz, $^4J_{CF} = 2.9$ Hz), 105.4 (t, $^2J_{CF} = 26.3$ Hz), 63.1, 62.9, 47.1 (d, $^4J_{CF} = 10.2$ Hz), 24.8, 14.6. UPLC-MS (A) $t_R = 1.27\text{min}$; m/z 389 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{20}\text{H}_{19}\text{F}_2\text{N}_2\text{O}_4$ $[\text{M}+\text{H}]^+$: 389.1307; found: 389.1303.

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(4-(trifluoromethyl)phenyl)imidazolidine-2,4-dione (112). The title compound was prepared from

5-methyl-5-(4-(trifluoromethyl)phenyl)imidazolidine-2,4-dione **82** (100 mg, 0.387 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (111 mg, 0.581 mmol) following general method B. White solid, yield 61% (97 mg, 0.24 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.21 (s, 1H), 7.99 (td, $J=8.6, 6.6$ Hz, 1H), 7.80 (q, $J=8.7$ Hz, 4H), 7.49 (ddd, $J=11.7, 9.3, 2.5$ Hz, 1H), 7.28 (td, $J=8.5, 2.3$ Hz, 1H), 4.81 (d, $J=2.8$ Hz, 2H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, $^3J_{\text{CF}} = 5.1$ Hz), 174.5, 165.7 (dd, $^1J_{\text{CF}} = 255.4$ Hz, $^3J_{\text{CF}} = 13.2$ Hz), 162.4 (dd, $^1J_{\text{CF}} = 257.6$ Hz, $^3J_{\text{CF}} = 13.2$ Hz), 155.0, 143.9, 132.6 (dd, $^3J_{\text{CF}} = 10.6$ Hz, $^3J_{\text{CF}} = 4.0$ Hz), 128.7 (q, $^2J_{\text{CF}} = 31.8$ Hz), 126.6, 125.5 (q, $^3J_{\text{CF}} = 3.6$ Hz), 124.1 (q, $^1J_{\text{CF}} = 272.2$ Hz), 119.5 (dd, $^2J_{\text{CF}} = 13.2$ Hz, $^4J_{\text{CF}} = 3.7$ Hz), 112.8 (dd, $^2J_{\text{CF}} = 22.0$ Hz, $^4J_{\text{CF}} = 3.7$ Hz), 105.4 (t, $^2J_{\text{CF}} = 27.0$ Hz), 63.2, 47.2 (d, $^4J_{\text{CF}} = 11.0$ Hz), 25.0. UPLC-MS (A) $t_{\text{R}} = 1.26\text{min}$; m/z 412 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{19}\text{H}_{14}\text{F}_5\text{N}_2\text{O}_3$ $[\text{M}+\text{H}]^+$: 413.0919; found: 413.0911.

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(4-(trifluoromethoxy)phenyl)imidazolidine-2,4-dione (113). The title compound was prepared from 5-methyl-5-(4-(trifluoromethoxy)phenyl)imidazolidine-2,4-dione **83** (80 mg, 0.29 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (83 mg, 0.44 mmol) following general method B. Off-white solid, yield 59% (74 mg, 0.17 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.14 (s, 1H), 7.94 - 8.05 (m, 1H), 7.63 - 7.71 (m, 2H), 7.40 - 7.53 (m, 3H), 7.28 (td, $J = 8.5, 2.5$ Hz, 1H), 4.81 (d, $J = 2.8$ Hz, 2H), 1.77 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $^3J_{\text{CF}} = 5.1$ Hz), 174.8, 165.7 (dd, $^1J_{\text{CF}} = 255.4$ Hz, $^3J_{\text{CF}} = 12.4$ Hz), 162.4 (dd, $^1J_{\text{CF}} = 257.6$ Hz, $^3J_{\text{CF}} = 13.2$ Hz), 155.0, 148.0, 138.7, 132.6 (dd, $^3J_{\text{CF}} = 11.3$ Hz, $^3J_{\text{CF}} = 4.0$ Hz), 127.8, 121.1, 120.0 (q, $^1J_{\text{CF}} = 256.1$ Hz), 119.5 (dd, $^2J_{\text{CF}} = 13.2$ Hz, $^4J_{\text{CF}} = 3.7$ Hz), 112.8 (dd, $^2J_{\text{CF}} = 22.0$ Hz, $^4J_{\text{CF}} = 3.7$ Hz), 105.4 (t, $^2J_{\text{CF}} = 26.4$ Hz), 62.9, 47.2 (d, $^4J_{\text{CF}} = 10.2$ Hz), 25.0. UPLC-MS (A) $t_{\text{R}} = 1.32\text{min}$; m/z 429 $[\text{M}+\text{H}]^+$ (>95%).

5-(4-(Difluoromethoxy)phenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (114). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (100 mg, 0.390 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (112 mg, 0.585 mmol) following general method B. Off-white solid, yield 59% (95 mg, 0.23 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.09 (s, 1H), 8.00 (td, *J* = 8.6, 6.8 Hz, 1H), 7.56 - 7.62 (m, 2H), 7.50 (ddd, *J* = 11.6, 9.3, 2.4 Hz, 1H), 7.06 - 7.45 (m, 4H), 4.80 (d, *J* = 2.8 Hz, 2H), 1.75 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.1, 175.0, 165.7 (dd, ¹*J*_{CF} = 255.4 Hz, ³*J*_{CF} = 12.4 Hz), 162.4 (dd, ¹*J*_{CF} = 257.6 Hz, ³*J*_{CF} = 13.2 Hz), 155.0, 150.7 (t, ³*J*_{CF} = 3.2 Hz), 136.2, 132.6 (dd, ³*J*_{CF} = 10.6 Hz, ³*J*_{CF} = 4.0 Hz), 127.4, 119.5 (dd, ²*J*_{CF} = 13.2 Hz, ⁴*J*_{CF} = 3.7 Hz), 118.7, 116.3 (t, ¹*J*_{CF} = 257.6 Hz), 112.8 (dd, ²*J*_{CF} = 22.0 Hz, ⁴*J*_{CF} = 3.2 Hz), 105.4 (t, ²*J*_{CF} = 27.1 Hz), 62.9, 47.2 (d, ⁴*J*_{CF} = 10.2 Hz), 24.9. UPLC-MS (A) *t*_R = 1.28min; *m/z* 411 [M+H]⁺ (>95%). HRMS (ESI) *m/z* calcd for C₁₉H₁₅F₄N₂O₄ [M+H]⁺: 411.0962; found: 411.0950.

5-(4-(1H-imidazol-1-yl)phenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (115). The title compound was prepared from 5-(4-(1H-imidazol-1-yl)phenyl)-5-methylimidazolidine-2,4-dione **85** (100 mg, 0.390 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (112 mg, 0.585 mmol) following general method B. Off-white solid, yield 57% (95.6 mg, 0.221 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.15 (s, 1H), 8.29 (s, 1H), 7.95-8.07 (m, 1H), 7.77 (t, *J* = 1.26 Hz, 1H), 7.70-7.75 (m, 2H), 7.65-7.70 (m, 2H), 7.51 (ddd, *J* = 2.40, 9.22, 11.62 Hz, 1H), 7.29 (dt, *J* = 2.40, 8.40 Hz, 1H), 7.12 (s, 1H), 4.82 (d, *J* = 2.53 Hz, 2H), 1.79 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.1 (d, *J* = 5.1 Hz), 175.0, 165.7 (dd, *J* = 255.4, 13.2 Hz), 162.4 (dd, *J* = 257.6, 13.2 Hz), 155.0, 137.8, 136.6, 135.6, 132.6 (dd, *J* = 11.0, 3.6 Hz),

129.9, 127.1, 120.3, 119.5 (dd, $J = 13.2, 3.7$ Hz), 118.0, 112.8 (dd, $J = 22.0, 3.2$ Hz), 105.4 (t, $J = 26.7$ Hz), 63.0, 47.2 (d, $J = 11.0$ Hz), 24.9. UPLC-MS (A) $t_R = 1.15$ min; m/z 411 $[M+H]^+$ (>95%).

5-(4-(1H-tetrazol-1-yl)phenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (116). The title compound was prepared from 5-(4-(1H-tetrazol-1-yl)phenyl)-5-methylimidazolidine-2,4-dione **86** (100 mg, 0.387 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (81 mg, 0.426 mmol) following general method B. White solid, yield 68% (109 mg, 0.264 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 10.12 (s, 1H), 9.21 (s, 1H), 7.96 - 8.04 (m, 3H), 7.82 (d, $J = 8.59$ Hz, 2H), 7.46 - 7.54 (m, 1H), 7.28 (dt, $J = 2.27, 8.46$ Hz, 1H), 4.83 (d, $J = 2.53$ Hz, 2H), 1.82 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J = 5.12$ Hz), 174.7, 165.7 (dd, $J = 13.17, 256.14$ Hz), 162.4 (dd, $J = 13.20, 258.30$ Hz), 155.0, 142.4, 140.8, 133.4, 132.6 (dd, $J = 4.39, 11.71$ Hz), 127.4, 121.3, 119.5 (dd, $J = 2.93, 13.17$ Hz), 112.8 (dd, $J = 2.93, 21.96$ Hz), 105.4 (t, $J = 27.80$ Hz), 63.1, 47.2 (d, $J = 10.98$ Hz), 24.9. UPLC-MS (A) $t_R = 1.09$ min; m/z 411 $[M-H]^-$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(4-morpholinophenyl)imidazolidine-2,4-dione (117). The title compound was prepared from 5-methyl-5-(4-morpholinophenyl)imidazolidine-2,4-dione **87** (100 mg, 0.363 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (104 mg, 0.545 mmol) following general method B. Off-white solid, yield 62% (102 mg, 0.226 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.95 (s, 1H), 8.00 (dt, $J = 6.57, 8.59$ Hz, 1H), 7.50 (ddd, $J = 2.40, 9.22, 11.62$ Hz, 1H), 7.36 (d, $J = 8.84$ Hz, 2H), 7.29 (dt, $J = 2.40, 8.40$ Hz, 1H), 6.97 (d, $J = 9.09$ Hz, 2H), 4.72-4.85 (m, $J = 2.53$ Hz, 2H), 3.66-3.79 (m, 4H), 3.05-3.15 (m, 4H), 1.71 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $J = 4.4$ Hz), 175.5, 165.7 (dd, $J = 255.9, 13.0$ Hz), 162.4 (dd, $J = 257.7, 13.2$ Hz), 155.1, 150.7, 132.6 (dd, $J = 11.1, 3.7$ Hz), 129.5, 126.3, 119.6 (dd, $J = 13.5, 3.3$ Hz), 114.8, 112.8 (dd, $J = 21.6, 2.8$ Hz), 105.4

(t, $J = 26.9$ Hz), 66.0, 62.8, 48.2, 47.1 (d, $J = 11.0$ Hz), 24.6. UPLC-MS (A) $t_R = 1.14$ min; m/z 430 $[M+H]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(4-(2-oxooxazolidin-3-yl)phenyl)imidazolidine-2,4-dione (118). The title compound was prepared from 5-methyl-5-(4-(2-oxooxazolidin-3-yl)phenyl)imidazolidine-2,4-dione **88** (80 mg, 0.29 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (138 mg, 0.727 mmol) following general method B. Off-white solid, yield 58% (72 mg, 0.17 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.06 (s, 1H), 8.00 (td, $J = 8.6, 6.8$ Hz, 1H), 7.46 - 7.66 (m, 5H), 7.28 (td, $J = 8.4, 2.4$ Hz, 1H), 4.80 (d, $J = 2.5$ Hz, 2H), 4.45 (t, $J = 8.0$ Hz, 2H), 4.03 - 4.11 (m, 2H), 1.75 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $^3J_{CF} = 5.1$ Hz), 175.2, 165.7 (dd, $^1J_{CF} = 256.1$ Hz, $^3J_{CF} = 13.2$ Hz), 162.4 (dd, $^1J_{CF} = 257.6$ Hz, $^3J_{CF} = 13.9$ Hz), 155.1, 154.9, 138.3, 134.2, 132.6 (dd, $^3J_{CF} = 11.7$, $^3J_{CF} = 3.7$ Hz), 126.2, 119.5 (dd, $^2J_{CF} = 13.2$ Hz, $^4J_{CF} = 3.7$ Hz), 117.9, 112.8 (dd, $^2J_{CF} = 22.0$ Hz, $^4J_{CF} = 2.9$ Hz), 105.4 (t, $^2J_{CF} = 27.1$ Hz), 62.9, 61.5, 47.1 (d, $^4J_{CF} = 10.2$ Hz), 44.7, 24.7. UPLC-MS (A) $t_R = 1.19$ min; m/z 430 $[M+H]^+$ (>95%). HRMS (ESI) m/z calcd for $C_{21}H_{17}F_2N_3O_5Na$ $[M+Na]^+$: 452.1028; found: 452.1030.

N-(4-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)phenyl)acetamide (119). The title compound was prepared from N-(4-(4-methyl-2,5-dioxoimidazolidin-4-yl)phenyl)acetamide **89** (200 mg, 0.809 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (231 mg, 1.21 mmol) following general method B. White solid, yield 40% (131 mg, 0.326 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 10.01 (s, 1H), 9.00 (s, 1H), 8.00 (td, $J = 8.6, 6.8$ Hz, 1H), 7.60 (d, $J = 8.6$ Hz, 2H), 7.50 (ddd, $J = 11.6, 9.2, 2.4$ Hz, 1H), 7.44 (d, $J = 8.8$ Hz, 2H), 7.28 (td, $J = 8.5, 2.3$ Hz, 1H), 4.79 (d, $J = 2.5$ Hz, 2H), 3.32 (s, 1H), 2.04 (s, 3H), 1.73 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $^3J_{CF} = 4.4$ Hz), 175.3, 168.3, 165.7 (dd,

$^1J_{CF} = 255.4$ Hz, $^3J_{CF} = 12.4$ Hz), 162.4 (dd, $^1J_{CF} = 258.3$ Hz, $^3J_{CF} = 13.9$ Hz), 155.1, 139.1, 133.6, 132.6 (dd, $^3J_{CF} = 11.0$ Hz, $^3J_{CF} = 4.4$ Hz), 126.0, 119.5 (dd, $^2J_{CF} = 13.9$ Hz, $^4J_{CF} = 3.7$ Hz), 118.9, 112.8 (dd, $^2J_{CF} = 22.0$ Hz, $^4J_{CF} = 2.9$ Hz), 105.4 (t, $^2J_{CF} = 27.1$ Hz), 63.0, 47.1 (d, $^4J_{CF} = 10.2$ Hz), 24.6, 24.0. UPLC-MS (A) $t_R = 3.31$ min; m/z 402 $[M+H]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(4-(methylsulfonyl)phenyl)imidazolidine-2,4-dione (120). The title compound was prepared from 5-methyl-5-(4-(methylsulfonyl)phenyl)imidazolidine-2,4-dione **90** (100 mg, 0.373 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (78 mg, 0.410 mmol) following general method B. White solid, yield 60% (95 mg, 0.225 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.23 (s, 1H), 7.96 - 8.03 (m, 3H), 7.83 (d, $J = 8.34$ Hz, 2H), 7.47 - 7.54 (m, 1H), 7.28 (dt, $J = 2.40, 8.40$ Hz, 1H), 4.82 (d, $J = 2.53$ Hz, 2H), 3.24 (s, 3H), 1.80 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, $J = 4.39$ Hz), 174.4, 165.7 (dd, $J = 13.17, 255.41$ Hz), 162.4 (dd, $J = 13.90, 258.34$ Hz), 154.9, 144.8, 140.6, 132.6 (dd, $J = 3.66, 10.98$ Hz), 127.3, 126.7, 119.4 (dd, $J = 3.29, 12.81$ Hz), 112.8 (dd, $J = 2.30, 22.10$ Hz), 105.4 (t, $J = 26.30$ Hz), 63.2, 47.2 (d, $J = 10.98$ Hz), 43.4, 25.0. UPLC-MS (A) $t_R = 1.09$ min; m/z 423 $[M+H]^+$ (>95%).

3-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (121). The title compound was prepared from 3-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **91** (100 mg, 0.465 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (133 mg, 0.697 mmol) following general method B. White solid, yield 75% (134.7 mg, 0.346 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.16 (s, 1H), 7.95-8.05 (m, 2H), 7.85-7.93 (m, 2H), 7.64-7.72 (m, 1H), 7.50 (ddd, $J = 2.40, 9.22, 11.62$ Hz, 1H), 7.28 (dt, $J = 2.40, 8.40$ Hz, 1H), 4.74-4.89 (m, 2H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J = 4.4$ Hz), 174.5, 165.7 (dd, $J = 255.7, 12.8$ Hz), 162.4 (dd, $J = 257.1, 12.8$ Hz), 154.9, 140.9, 132.6 (dd, $J = 11.3, 4.0$ Hz), 132.0,

130.6, 129.9, 129.4, 119.4 (dd, $J = 13.1, 3.7$ Hz), 118.5, 112.8 (dd, $J = 22.0, 2.9$ Hz), 111.6, 105.4 (t, $J = 26.7$ Hz), 62.9, 47.3 (d, $J = 11.0$ Hz), 24.8. UPLC-MS (A) $t_R = 1.26$ min; m/z 370 $[M+H]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-(3-methoxyphenyl)-5-methylimidazolidine-2,4-dione (122). The title compound was prepared from 5-(3-methoxyphenyl)-5-methylimidazolidine-2,4-dione **92** (100 mg, 0.454 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (130 mg, 0.682 mmol) following general method B. White solid, yield 26% (43 mg, 0.12 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.06 (s, 1H), 8.00 (td, $J = 8.6, 6.8$ Hz, 1H), 7.50 (ddd, $J = 11.6, 9.2, 2.4$ Hz, 1H), 7.34 (t, $J = 8.0$ Hz, 1H), 7.28 (td, $J = 8.4, 2.4$ Hz, 1H), 7.06 - 7.15 (m, 2H), 6.94 (dd, $J = 8.2, 1.9$ Hz, 1H), 4.80 (d, $J = 2.8$ Hz, 2H), 3.79 (s, 3H), 1.74 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $J = 5.1$ Hz), 175.0, 165.7 (dd, $J = 255.4$ Hz, 13.2 Hz), 162.4 (dd, $J = 257.6$ Hz, 13.2 Hz), 159.3, 155.0, 140.9, 132.6 (dd, $J = 10.6$ Hz, 3.3 Hz), 129.6, 119.5 (dd, $J = 13.2$ Hz, 3.7 Hz), 117.7, 113.2, 112.8 (dd, $J = 22.0$ Hz, 2.9 Hz), 111.7, 105.4 (t, $J = 26.3$ Hz), 63.2, 55.1, 47.1 (d, $J = 11.0$ Hz), 24.9. UPLC-MS (A) $t_R = 1.18$ min; m/z 375 $[M+H]^+$ (>95%).

5-(3-Bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (123). The title compound was prepared from 5-(3-bromophenyl)-5-methylimidazolidine-2,4-dione **93** (400 mg, 1.486 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (425 mg, 2.230 mmol) following general method B. White solid, yield 53% (352 mg, 0.790 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.02-9.08 (m, 1H), 7.86-7.97 (m, 1H), 7.60-7.66 (m, 1H), 7.46-7.52 (m, 2H), 7.38-7.45 (m, 1H), 7.30-7.36 (m, 1H), 7.15-7.23 (m, 1H), 4.68-4.76 (m, 2H), 1.66 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J = 5.1$ Hz), 174.6, 165.7 (dd, $J = 255.4, 13.2$ Hz), 162.4 (dd, $J = 257.6, 13.2$ Hz), 154.9, 142.0, 132.6 (dd, $J = 11.0, 3.7$ Hz), 131.0, 130.8, 128.4,

124.8, 121.9, 119.5 (dd, $J = 13.2, 3.7$ Hz), 112.8 (dd, $J = 21.9, 3.1$ Hz), 105.4 (t, $J = 26.7$ Hz), 62.9, 47.2 (d, $J = 11.0$ Hz), 25.0. UPLC-MS (A) $t_R = 1.24$ min; m/z 421 $[M-H]^-$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-(3-fluoro-4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (124). The title compound was prepared from 5-(3-fluoro-4-methoxyphenyl)-5-methylimidazolidine-2,4-dione **94** (98 mg, 0.411 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (118 mg, 0.617 mmol) following general method B. White solid, yield 38% (65.3 mg, 0.158 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.05 (s, 1H), 8.00 (dt, $J = 6.69, 8.53$ Hz, 1H), 7.50 (ddd, $J = 2.27, 9.16, 11.56$ Hz, 1H), 7.18-7.38 (m, 4H), 4.80 (d, $J = 2.53$ Hz, 2H), 3.85 (s, 3H), 1.72 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $J = 5.1$ Hz), 175.0, 165.7 (dd, $J = 255.4, 13.2$ Hz), 162.4 (dd, $J = 257.6, 13.2$ Hz), 154.9, 151.1 (d, $J = 243.7$ Hz), 146.8 (d, $J = 10.6$ Hz), 132.6 (dd, $J = 11.0, 3.7$ Hz), 132.1 (d, $J = 5.9$ Hz), 121.9 (d, $J = 2.9$ Hz), 119.5 (dd, $J = 13.2, 3.3$ Hz), 113.8 (d, $J = 2.2$ Hz), 113.5 (d, $J = 19.8$ Hz), 112.8 (dd, $J = 21.8, 3.2$ Hz), 105.4 (t, $J = 26.8$ Hz), 62.6 (d, $J = 1.5$ Hz), 56.1, 47.2 (d, $J = 11.0$ Hz), 24.7. UPLC-MS (A) $t_R = 1.16$ min; m/z 393 $[M+H]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-(2-fluoro-4-methoxyphenyl)-5-methylimidazolidine-2,4-dione (125). The title compound was prepared from 5-(2-fluoro-4-methoxyphenyl)-5-methylimidazolidine-2,4-dione **95** (97 mg, 0.407 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (116 mg, 0.611 mmol) following general method B. White solid, yield 73% (137 mg, 0.297 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.81 (s, 1H), 8.03 (dt, $J = 6.82, 8.59$ Hz, 1H), 7.42-7.57 (m, 2H), 7.30 (dt, $J = 2.40, 8.40$ Hz, 1H), 6.80-6.91 (m, 2H), 4.84 (d, $J = 2.53$ Hz, 2H), 3.79 (s, 3H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J = 5.1$ Hz), 175.3, 165.6 (dd, $J = 255.3, 12.6$ Hz), 162.4 (dd, $J = 257.2, 13.2$ Hz), 160.9 (d, $J = 11.7$ Hz), 161.0 (d, $J = 248.8$ Hz), 155.0 (s), 132.6 (dd, $J = 11.0, 3.7$ Hz), 129.0 (d, $J = 5.1$ Hz), 119.6

(dd, $J = 13.2, 3.7$ Hz), 117.5 (d, $J = 11.7$ Hz), 112.8 (dd, $J = 22.0, 2.9$ Hz), 109.9 (d, $J = 2.2$ Hz), 105.4 (t, $J = 26.6$ Hz), 102.4 (d, $J = 25.6$ Hz), 60.5, 55.8, 47.0 (d, $J = 10.2$ Hz), 23.6. UPLC-MS (A) $t_R = 1.16$ min; m/z 393 $[M+H]^+$ (>95%).

5-(4-Bromo-2-fluorophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (126). The title compound was prepared from 5-(4-bromo-2-fluorophenyl)-5-methylimidazolidine-2,4-dione **96** (200 mg, 0.697 mmol) and 2-bromo-1-(2,4-difluorophenyl)ethan-1-one **16** (246 mg, 1.045 mmol) following general method B. White solid, yield 31% (95 mg, 0.215 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.91 (s, 1H), 8.03 (dt, $J = 6.82, 8.59$ Hz, 1H), 7.64 (dd, $J = 1.64, 11.24$ Hz, 1H), 7.46-7.58 (m, 3H), 7.30 (dt, $J = 2.40, 8.40$ Hz, 1H), 4.86 (d, $J = 2.53$ Hz, 2H), 1.82 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, $J = 4.4$ Hz), 174.6, 165.7 (dd, $J = 255.4, 13.2$ Hz), 162.4 (dd, $J = 257.6, 13.2$ Hz), 160.2 (d, $J = 254.0$ Hz), 155.0, 132.7 (dd, $J = 11.0, 4.4$ Hz), 130.2 (d, $J = 3.7$ Hz), 127.6 (d, $J = 3.7$ Hz), 125.3 (d, $J = 11.7$ Hz), 122.5 (d, $J = 9.5$ Hz), 119.7 (d, $J = 25.6$ Hz), 119.6 (dd, $J = 13.0, 3.6$ Hz), 112.8 (dd, $J = 22.0, 2.9$ Hz), 105.4 (t, $J = 26.8$ Hz), 60.7, 47.1 (d, $J = 11.0$ Hz), 23.4. UPLC-MS (A) $t_R = 1.23$ min; m/z 439 $[M-H]^-$ (>95%).

5-(4-(Difluoromethoxy)-3-methoxyphenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (127). The title compound was prepared from 5-(4-(difluoromethoxy)-3-methoxyphenyl)-5-methylimidazolidine-2,4-dione **97** (70 mg, 0.25 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (70 mg, 0.37 mmol) following general method B. White solid, yield 49% (53 mg, 0.12 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.11 (s, 1H), 8.00 (td, $J = 8.6, 6.8$ Hz, 1H), 7.50 (ddd, $J = 11.6, 9.3, 2.4$ Hz, 1H), 6.87 - 7.34 (m, 5H), 4.81 (d, $J = 2.5$ Hz, 2H), 3.88 (s, 3H), 1.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $J = 4.4$ Hz), 174.9, 165.7 (dd, $J = 255.4$ Hz, 12.4 Hz), 162.4 (dd, $J = 258.3$ Hz, 13.9 Hz), 155.0, 150.5, 139.2,

137.9, 132.6 (dd, $J = 11.0$ Hz, 4.4 Hz), 120.9, 119.5 (dd, $J = 13.2$ Hz, 3.7 Hz), 117.9, 116.6 (t, $J = 259.1$ Hz), 112.8 (dd, $J = 22.0$ Hz, 2.9 Hz), 110.8, 105.4 (t, $J = 27.1$ Hz), 63.1, 55.9, 47.2 (d, $J = 11.0$ Hz), 24.9. UPLC-MS (A) $t_R = 1.26$ min; m/z 441 $[M+H]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(3,4,5-trimethoxyphenyl)imidazolidine-2,4-dione (128). The title compound was prepared from 5-methyl-5-(3,4,5-trimethoxyphenyl)imidazolidine-2,4-dione **98** (100 mg, 0.357 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (74.8 mg, 0.392 mmol) following general method B. White solid, yield 75% (116 mg, 0.267 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.05 (s, 1H), 8.01 (s, 1H), 7.46 - 7.54 (m, 1H), 7.25 - 7.32 (m, 1H), 6.85 (s, 2H), 4.81 (d, $J = 2.27$ Hz, 2H), 3.82 (s, 6H), 3.67 (s, 3H), 1.74 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $J = 4.39$ Hz), 175.1, 165.7 (dd, $J = 12.44$, 255.41 Hz), 162.4 (dd, $J = 13.90$, 258.34 Hz), 155.0, 152.7, 137.3, 135.0, 132.6 (dd, $J = 3.66$, 10.98 Hz), 119.5 (dd, $J = 3.66$, 13.17 Hz), 112.8 (dd, $J = 2.93$, 21.22 Hz), 105.4 (t, $J = 26.30$ Hz), 103.3, 63.3, 60.0, 56.0, 47.2 (d, $J = 10.25$ Hz), 24.9. UPLC-MS (A) $t_R = 1.15$ min; m/z 435 $[M+H]^+$ (>95%).

3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-methyl-5-(pyridin-4-yl)imidazolidine-2,4-dione (129). The title compound was prepared from 5-methyl-5-(pyridin-4-yl)imidazolidine-2,4-dione **99** (100 mg, 0.523 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (100 mg, 0.523 mmol) following general method B. White solid, yield 39% (71 mg, 0.20 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.19 (s, 1H), 8.58 - 8.71 (m, 2H), 8.00 (td, $J = 8.6$, 6.8 Hz, 1H), 7.53 - 7.57 (m, 2H), 7.43 - 7.53 (m, 1H), 7.28 (td, $J = 8.5$, 2.5 Hz, 1H), 4.81 (d, $J = 2.8$ Hz, 2H), 1.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, $J = 4.4$ Hz), 174.1, 165.7 (dd, $J = 253.2$ Hz, 13.2 Hz), 162.4 (dd, $J = 257.6$ Hz, 13.2 Hz), 155.0, 150.0, 147.9, 132.6 (dd, $J = 11.7$ Hz, 4.4 Hz), 120.6,

119.4 (dd, $J = 13.2$ Hz, 3.7 Hz), 112.8 (dd, $J = 22.0$ Hz, 2.9 Hz), 105.4 (t, $J = 26.3$ Hz), 62.8, 47.3 (d, $J = 11.0$ Hz), 24.5. UPLC-MS (A) $t_R = 1.04$ min; m/z 346 $[M+H]^+$ (>95%).

5-(6-Bromopyridin-3-yl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (130). The title compound was prepared from 5-(6-bromopyridin-3-yl)-5-methylimidazolidine-2,4-dione **100** (200 mg, 0.741 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (155 mg, 0.815 mmol) following general method B. Off-white solid, yield 78% (244 mg, 0.575 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.19 (s, 1H), 8.56 (d, $J = 2.53$ Hz, 1H), 8.00 (dt, $J = 6.82, 8.46$ Hz, 1H), 7.90 (dd, $J = 2.65, 8.46$ Hz, 1H), 7.75 (d, $J = 8.59$ Hz, 1H), 7.46 - 7.54 (m, 1H), 7.28 (dt, $J = 2.40, 8.40$ Hz, 1H), 4.82 (d, $J = 2.53$ Hz, 2H), 1.78 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, $J = 5.12$ Hz), 174.3, 165.7 (dd, $J = 13.17, 255.41$ Hz), 162.5 (dd, $J = 13.17, 258.34$ Hz), 154.9, 147.9, 141.2, 137.1, 135.0, 132.7 (dd, $J = 4.02, 11.34$ Hz), 128.0, 119.4 (dd, $J = 3.66, 13.17$ Hz), 112.8 (dd, $J = 2.20, 22.69$ Hz), 105.4 (t, $J = 27.10$ Hz), 61.7, 47.3 (d, $J = 10.25$ Hz), 24.7. UPLC-MS (A) $t_R = 1.14$ min; m/z 424 $[M+H]^+$ (>95%).

5-(5-Bromopyridin-2-yl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (131). The title compound was prepared from 5-(5-bromopyridin-2-yl)-5-methylimidazolidine-2,4-dione **101** (150 mg, 0.555 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (116 mg, 0.611 mmol) following general method B. Off-white solid, yield 96% (226 mg, 0.533 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.04 (s, 1H), 8.76 (d, $J = 2.27$ Hz, 1H), 8.17 (dd, $J = 2.40, 8.46$ Hz, 1H), 8.01 (dt, $J = 6.95, 8.53$ Hz, 1H), 7.47 - 7.61 (m, 2H), 7.29 (dt, $J = 2.27, 8.34$ Hz, 1H), 4.83 (d, $J = 2.53$ Hz, 2H), 1.80 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.1 (d, $J = 4.39$ Hz), 174.1, 166.1 (dd, $J = 13.17, 255.41$ Hz), 162.4 (dd, $J = 13.17, 258.34$ Hz), 156.2, 155.3, 149.8, 140.0, 132.7 (dd, $J = 4.03, 11.34$ Hz), 122.5, 120.1, 119.5 (dd, J

= 3.66, 13.17 Hz), 112.8 (dd, J = 2.93, 21.22 Hz), 105.5 (t, J = 27.80 Hz), 64.9, 47.2 (d, J = 10.25 Hz), 22.5. UPLC-MS (A) t_R = 1.16min; m/z 424 $[M+H]^+$ (>95%).

5-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)thiophene-2-carbonitrile (132). The title compound was prepared from 5-(4-methyl-2,5-dioxoimidazolidin-4-yl)thiophene-2-carbonitrile **102** (100 mg, 0.452 mmol) and 2-bromo-1-(2,4-difluorophenyl)ethan-1-one **16** (159 mg, 0.678 mmol) following general method B. Off-white solid, yield 44% (74.3 mg, 0.198 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.49 (br. s., 1H), 8.01 (dt, J = 7.03, 8.53 Hz, 1H), 7.96 (d, J = 4.02 Hz, 1H), 7.52 (ddd, J = 2.26, 9.29, 11.55 Hz, 1H), 7.38 (d, J = 3.77 Hz, 1H), 7.30 (dt, J = 2.26, 8.41 Hz, 1H), 4.84 (d, J = 2.51 Hz, 2H), 1.83 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 188.9 (d, J = 5.1 Hz), 173.1, 165.8 (dd, J = 255.4, 13.2 Hz), 162.5 (dd, J = 257.6, 13.2 Hz), 154.8, 152.0, 139.7, 132.7 (dd, J = 11.8, 3.7 Hz), 126.4, 119.4 (dd, J = 13.2, 3.7 Hz), 114.0, 112.8 (dd, J = 22.0, 3.0 Hz), 108.1, 105.5 (t, J = 26.8 Hz), 62.1, 47.4 (d, J = 11.0 Hz), 25.4. UPLC-MS (B) t_R = 1.60min; m/z 374 $[M-H]^-$ (>95%).

5-(4-Bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-ethylimidazolidine-2,4-dione (133). The title compound was prepared from 5-(4-bromophenyl)-5-ethylimidazolidine-2,4-dione **103** (200 mg, 0.706 mmol) and 2-bromo-1-(2,4-difluorophenyl)ethan-1-one **16** (249 mg, 1.060 mmol) following general method B. Off-white solid, yield 61% (188 mg, 0.430 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.16 (s, 1H), 7.98 (dt, J = 6.69, 8.65 Hz, 1H), 7.60-7.67 (m, 2H), 7.45-7.54 (m, 3H), 7.27 (dt, J = 2.40, 8.40 Hz, 1H), 4.79 (d, J = 2.53 Hz, 2H), 2.09-2.21 (m, 1H), 1.92-2.04 (m, 1H), 0.90 (t, J = 7.33 Hz, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 188.9 (d, J = 4.4 Hz), 174.0, 165.7 (dd, J = 255.6, 13.2 Hz), 162.4 (dd, J = 257.6, 13.2 Hz), 155.3, 138.1, 132.6 (dd, J = 11.0, 3.7 Hz), 131.4, 127.8, 121.4, 119.5 (dd, J = 13.2, 3.7 Hz), 112.8 (dd, J = 21.8, 3.0 Hz), 105.4

(t, $J = 26.6$ Hz), 67.1, 47.2 (d, $J = 11.0$ Hz), 31.2, 7.9. UPLC-MS (A) $t_R = 1.26$ min; m/z 435 $[M-H]^-$ (>95%).

4-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-ethyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (134). The title compound was prepared from 4-(4-ethyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **104** (100 mg, 0.436 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (91 mg, 0.480 mmol) following general method B. White solid, yield 88% (147 mg, 0.383 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.27 (s, 1H), 7.98 (dt, $J = 6.95, 8.53$ Hz, 1H), 7.92 (d, $J = 8.34$ Hz, 2H), 7.75 (d, $J = 8.59$ Hz, 2H), 7.49 (ddd, $J = 2.27, 9.28, 11.43$ Hz, 1H), 7.27 (dt, $J = 2.27, 8.46$ Hz, 1H), 4.80 (d, $J = 2.53$ Hz, 2H), 2.13 - 2.25 (m, 1H), 1.95 - 2.07 (m, 1H), 0.91 (t, $J = 7.33$ Hz, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 188.9 (d, $J = 5.12$ Hz), 173.6, 165.7 (dd, $J = 13.17, 256.14$ Hz), 162.4 (dd, $J = 13.17, 256.88$ Hz), 155.2, 143.9, 132.7 (m)*, 132.5*, 126.7, 119.5 (dd, $J = 3.66, 13.17$ Hz), 118.5, 112.8 (dd, $J = 1.46, 21.22$ Hz), 111.0, 105.4 (t, $J = 27.10$ Hz), 67.4, 47.3 (d, $J = 10.25$ Hz), 31.3, 7.9. *Peaks overlap. UPLC-MS (A) $t_R = 1.16$ min; m/z 382 $[M-H]^-$ (>95%).

5-(4-Bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-propylimidazolidine-2,4-dione (135). The title compound was prepared from 5-(4-bromophenyl)-5-propylimidazolidine-2,4-dione **105** (200 mg, 0.673 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (141 mg, 0.740 mmol) following general method B. White solid, yield 82% (248.8 mg, 0.551 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.18 (s, 1H), 7.92 - 8.05 (m, 1H), 7.63 (d, $J = 8.59$ Hz, 2H), 7.43 - 7.55 (m, 3H), 7.27 (dt, $J = 2.40, 8.40$ Hz, 1H), 4.78 (d, $J = 2.53$ Hz, 2H), 1.86 - 2.15 (m, 2H), 1.16 - 1.39 (m, 2H), 0.90 (t, $J = 7.33$ Hz, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, $J = 5.12$ Hz), 174.1, 165.7 (dd, $J = 11.71, 253.95$ Hz), 162.4 (dd, $J = 13.17, 257.61$ Hz), 155.2, 138.2, 132.6 (dd, $J = 3.66, 10.98$ Hz), 131.4, 127.8, 121.4, 119.5 (d, $J = 13.17$ Hz), 112.8 (d, $J = 24.88$ Hz),

105.4 (t, $J = 24.90$ Hz), 66.6, 47.1 (d, $J = 9.51$ Hz), 26.3, 16.6, 13.7. UPLC-MS (A) $t_R = 1.32$ min; m/z 449 $[M-H]^-$ (>95%).

5-(4-Bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-isopropylimidazolidine-2,4-dione (136). The title compound was prepared from 5-(4-bromophenyl)-5-isopropylimidazolidine-2,4-dione **106** (40 mg, 0.135 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (28.2 mg, 0.148 mmol) following general method B. White solid, yield 62% (37.7 mg, 0.084 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 9.23 (s, 1H), 7.94 - 8.01 (m, 1H), 7.63 (d, $J = 8.59$ Hz, 2H), 7.44 - 7.53 (m, 3H), 7.26 (dt, $J = 2.40, 8.40$ Hz, 1H), 4.76 (d, $J = 2.27$ Hz, 2H), 2.50 - 2.57 (m, 1H)*, 0.99 (d, $J = 6.57$ Hz, 3H), 0.68 (d, $J = 7.07$ Hz, 3H). *Overlaps with solvent peak. ^{13}C NMR (101 MHz, DMSO- d_6) δ 188.9 (d, $J = 5.12$ Hz), 174.0, 165.7 (dd, $J = 13.17, 254.68$ Hz), 162.3 (dd, $J = 13.90, 257.61$ Hz), 155.6, 137.8, 132.6 (dd, $J = 4.39, 11.71$ Hz), 131.4, 127.8, 121.3, 119.5 (dd, $J = 3.66, 13.17$ Hz), 112.7 (m, $J = 3.66, 21.96$ Hz), 105.4 (t, $J = 27.80$ Hz), 70.5, 47.1 (d, $J = 10.98$ Hz), 35.2, 16.7, 16.3. UPLC-MS (A) $t_R = 1.31$ min; m/z 449 $[M-H]^-$ (>95%).

5-(4-Bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-(trifluoromethyl)imidazolidine-2,4-dione (137). The title compound was prepared from 5-(4-bromophenyl)-5-(trifluoromethyl)imidazolidine-2,4-dione **107** (158 mg, 0.489 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (103 mg, 0.538 mmol) following general method B. White solid, yield 63% (148 mg, 0.310 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 10.49 (s, 1H), 8.00 (dt, $J = 6.82, 8.46$ Hz, 1H), 7.71 - 7.82 (m, 4H), 7.46 - 7.55 (m, 1H), 7.28 (dt, $J = 2.27, 8.46$ Hz, 1H), 4.92 (d, $J = 2.27$ Hz, 2H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 188.1 (d, $J = 5.12$ Hz), 166.4, 165.9 (dd, $J = 13.17, 256.14$ Hz), 162.5 (dd, $J = 13.17, 257.61$ Hz), 154.7, 132.7 (dd, $J = 4.03, 11.34$ Hz), 132.0, 129.2, 128.8, 123.9, 119.3 (dd, $J = 3.29, 12.81$ Hz), 122.3, 112.8 (dd, $J =$

2.93, 21.96 Hz), 105.4 (t, $J = 27.00$ Hz), 66.7 (q, $J = 30.00$ Hz), 47.7 (d, $J = 10.98$ Hz). UPLC-MS (A) $t_R = 1.32$ min; m/z 475 $[M-H]^-$ (>95%).

5'-Bromo-1-(2-(2,4-difluorophenyl)-2-oxoethyl)-2',3'-dihydrospiro[imidazolidine-4,1'-indene]-2,5-dione (138). The title compound was prepared from 5'-bromo-2',3'-dihydrospiro[imidazolidine-4,1'-indene]-2,5-dione **108** (300 mg, 1.067 mmol) and 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (224 mg, 1.174 mmol) following general method B. White solid, yield 56% (258 mg, 0.593 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.96 (s, 1H), 7.98 - 8.11 (m, 1H), 7.59 (s, 1H), 7.46 - 7.57 (m, 2H), 7.20 - 7.35 (m, 2H), 4.85 (d, $J = 2.27$ Hz, 2H), 3.06 (t, $J = 7.20$ Hz, 2H), 2.54 - 2.66 (m, 1H), 2.28 (td, $J = 7.77, 13.52$ Hz, 1H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.2 (d, $J = 5.12$ Hz), 174.9, 165.8 (dd, $J = 12.44, 255.41$ Hz), 162.5 (dd, $J = 13.17, 258.34$ Hz), 155.2, 146.8, 140.5, 132.7 (dd, $J = 5.12, 11.71$ Hz), 130.1, 128.1, 125.2, 122.4, 119.5 (dd, $J = 3.66, 13.17$ Hz), 112.8 (dd, $J = 3.66, 22.69$ Hz), 105.5 (t, $J = 26.30$ Hz), 70.5, 47.2 (d, $J = 10.25$ Hz), 36.2, 29.6. UPLC-MS (A) $t_R = 1.23$ min; m/z 433 $[M-H]^-$ (>95%).

N-(4-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)phenyl)methanesulfonamide (139). 5-(4-bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione **109** (300 mg, 0.709 mmol), methanesulfonamide (81 mg, 0.851 mmol) potassium carbonate (196 mg, 1.418 mmol), allylpalladium (II) chloride dimer ($[Pd(allyl)Cl]_2$) (2.59 mg, 7.09 μ mol) and 2-di-*tert*-butylphosphino-2',4',6'-triisopropylbiphenyl (t-BuXPhos) (12.04 mg, 0.028 mmol) were placed in a vial. The vial was sealed with a septum and was evacuated with vacuum and backfilled with nitrogen 3 times. Subsequently dry 2-methyltetrahydrofuran (6 mL) was added and the vial was evacuated with vacuum and backfilled with nitrogen 3 times. The vial was sealed under nitrogen flow and the reaction mixture was stirred for 10 minutes at room temperature, followed by heating at 80 °C for 4h. Reaction was cooled

down to room temperature, diluted with ethyl acetate (8 ml) and washed with 1M HCl (8ml). The water phase was extracted 3x with ethyl acetate (15ml). Combined organic phases were filtered through a plug of silica and eluted with ethyl acetate and evaporated. Flash column chromatography on silicagel (EtOAc:Cyclohexane 20-60%) provided the crude product which was further purified by preparative HPLC (Sunfire 19, ACN:Water 40-100%, 0.1% formic acid). The obtained fractions were concentrated *in vacuo* and extracted twice with ethyl acetate. Combined organic fractions were washed with water (3x), dried with sodium sulfate, evaporated and lyophilized. White solid, yield 63% (194 mg, 0.444 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.85 (br. s., 1H), 9.03 (s, 1H), 7.93 - 8.16 (m, 1H), 7.42 - 7.59 (m, 3H), 7.10 - 7.36 (m, 3H), 4.80 (br. s., 2H), 3.01 (s, 3H), 1.73 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.2 (d, *J* = 4.39 Hz), 175.2, 165.7 (dd, *J* = 12.44, 255.41 Hz), 162.4 (dd, *J* = 13.17, 257.61 Hz), 155.1, 138.2, 134.5, 132.7 (dd, *J* = 3.66, 10.98 Hz), 126.7, 119.6*, 119.5*, 112.8 (dd, *J* = 2.93, 21.96 Hz), 105.4 (t, *J* = 26.30 Hz), 62.9, 47.1 (d, *J* = 10.25 Hz), 39.5 (overlaps with solvent peak), 24.7. *Peaks overlap. UPLC-MS (A) t_R = 1.17min; *m/z* 438 [M+H]⁺ (>95%).

N-(3-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioximidazolidin-4-yl)phenyl)methanesulfonamide (140). The title compound was prepared following a procedure analogous to **139**, starting from 5-(3-bromophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione **109** (150 mg, 0.354 mmol). White solid, yield 56% (91 mg, 0.198 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.86 (s, 1H), 9.10 (s, 1H), 8.00 (dt, *J* = 6.69, 8.53 Hz, 1H), 7.51 (ddd, *J* = 2.40, 9.28, 11.56 Hz, 1H), 7.35-7.44 (m, 2H), 7.25-7.32 (m, 2H), 7.20 (dd, *J* = 1.26, 7.83 Hz, 1H), 4.80 (d, *J* = 2.53 Hz, 2H), 3.02 (s, 3H), 1.73 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.0 (d, *J* = 4.4 Hz), 174.9, 165.7 (dd, *J* = 255.4, 13.0 Hz), 162.4 (dd, *J* = 257.6, 13.2 Hz), 155.0, 140.6, 138.6, 132.6 (dd, *J* = 11.3, 3.8 Hz), 129.5, 121.0, 119.5 (dd, *J* = 13.2, 3.6

Hz), 119.1, 116.8, 112.8 (dd, $J = 21.9, 3.1$ Hz), 105.4 (t, $J = 26.6$ Hz), 63.1, 47.1 (d, $J = 11.0$ Hz), 39.3 (overlaps with solvent peak), 24.9. UPLC-MS (A) $t_R = 1.06$ min; m/z 438 $[M+H]^+$ (>95%).

5-(4-Aminophenyl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (141). N-(4-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)phenyl)acetamide **119** (51 mg, 0.13 mmol) was dissolved in ethanol (4 mL) and hydrochloric acid (HCl) (1M, 2 mL) was added and the mixture was left stirring at 65°C for 3 days. The reaction mixture was concentrated *in vacuo* and the residue was diluted with water (30 mL), neutralized using sodium carbonate (1M) and extracted with ethyl acetate (2x 30 mL). The combined organic layers were dried over sodium sulfate and evaporated to give the title compound. White solid; yield 94% (43 mg, 0.12 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 8.83 (s, 1H), 8.00 (td, $J = 8.6, 6.8$ Hz, 1H), 7.51 (ddd, $J = 11.6, 9.3, 2.4$ Hz, 1H), 7.29 (td, $J = 8.5, 2.3$ Hz, 1H), 7.06 - 7.19 (m, 2H), 6.51 - 6.62 (m, 2H), 5.17 (s, 2H), 4.77 (d, $J = 2.3$ Hz, 2H), 1.66 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.3 (d, $^3J_{CF} = 5.1$ Hz), 175.9, 165.6 (dd, $^1J_{CF} = 255.4$ Hz, $^3J_{CF} = 13.2$ Hz), 162.4 (dd, $^1J_{CF} = 256.9$ Hz, $^3J_{CF} = 13.2$ Hz), 155.1, 148.5, 132.6 (dd, $^3J_{CF} = 10.6$ Hz, $^3J_{CF} = 4.0$ Hz), 126.3, 125.9, 119.6 (dd, $^2J_{CF} = 13.9$ Hz, $^4J_{CF} = 3.7$ Hz), 113.5, 112.8 (dd, $^2J_{CF} = 22.0$ Hz, $^4J_{CF} = 2.9$ Hz), 105.4 (t, $^2J_{CF} = 26.3$ Hz), 62.9, 47.0 (d, $^4J_{CF} = 10.2$ Hz), 24.4. UPLC-MS (A) $t_R = 1.04$ min; m/z 360 $[M+H]^+$ (>95%). HRMS (ESI) m/z calcd for $C_{18}H_{16}F_2N_3O_3$ $[M+H]^+$: 360.1154; found: 360.1156.

5-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)picolinonitrile (142). Under nitrogen, 5-(6-bromopyridin-3-yl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione **130** (25 mg, 0.059 mmol), zinc cyanide (4.15 mg, 0.035 mmol), 4,5-bis(diphenylphosphino)-9,9-dimethylxanthene (XantPhos) (0.682 mg, 1.179 μ mol) and tris(dibenzylideneacetone)dipalladium(0) ($Pd_2(dba)_3$) (0.540 mg, 0.589 μ mol) were placed in a microwave vial. Air was evacuated with vacuum and the vial was backfilled with

nitrogen 3 times. Anhydrous DMF (200 μ L) and tetramethylethylenediamine (TMEDA) (1.370 mg, 0.012 mmol) were added and nitrogen was bubbled through the mixture for 1 minute. The vial was sealed and the reaction was irradiated in a microwave at 160 $^{\circ}$ C for 5 minutes. Reaction mixture was filtered through a PTFE syringe filter and purified via preparative HPLC (Sunfire 19, ACN:Water 40-100, 0.1% formic acid). Obtained fractions were concentrated *in vacuo* and extracted with ethyl acetate (3x). Combined organic layer was washed with saturated sodium bicarbonate and brine, dried with sodium sulfate, evaporated and lyophilized. White solid; yield 73% (16 mg, 0.043 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.27 (br. s., 1H), 8.94 (d, J = 1.77 Hz, 1H), 8.18 - 8.22 (m, 1H), 8.11 - 8.17 (m, 1H), 7.96 - 8.03 (m, 1H), 7.46 - 7.54 (m, 1H), 7.28 (dt, J = 2.27, 8.34 Hz, 1H), 4.83 (d, J = 2.53 Hz, 2H), 1.83 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 188.9 (d, J = 4.39 Hz), 173.9, 165.8 (dd, J = 13.17, 255.41 Hz), 162.5 (dd, J = 13.17, 256.88 Hz), 154.8, 148.7, 139.1, 135.3, 132.7 (dd, J = 4.39, 10.98 Hz), 132.3, 128.9, 119.4 (dd, J = 3.29, 12.81 Hz), 117.2, 112.8 (dd, J = 2.93, 21.22 Hz), 105.4 (t, J = 27.80 Hz), 62.1, 47.4 (d, J = 10.25 Hz), 24.7. UPLC-MS (A) t_R = 1.15min; m/z 371 $[\text{M}+\text{H}]^+$ (>95%).

6-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)nicotinonitrile (143).

The title compound was prepared following a procedure analogous to **142**, starting from 5-(5-bromopyridin-2-yl)-3-(2-(2,4-difluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione **131** (30 mg, 0.071 mmol). White solid; yield 73% (19 mg, 0.051 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.13 (s, 1H), 9.09 (d, J = 1.26 Hz, 1H), 8.44 (dd, J = 2.27, 8.34 Hz, 1H), 7.98 - 8.05 (m, 1H), 7.83 (d, J = 8.34 Hz, 1H), 7.48 - 7.55 (m, 1H), 7.29 (dt, J = 2.27, 8.46 Hz, 1H), 4.84 (d, J = 2.27 Hz, 2H), 1.83 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 189.0 (d, J = 5.12 Hz), 173.7, 165.7 (dd, J = 12.44, 255.41 Hz), 162.5 (dd, J = 13.90, 258.34 Hz), 161.1, 155.3, 152.1, 141.3, 132.7 (dd, J

= 2.93, 10.98 Hz), 120.9, 119.5 (dd, J = 2.93, 12.44 Hz), 116.7, 112.8 (dd, J = 2.93, 22.30 Hz), 108.8, 105.4 (t, J = 26.30 Hz), 65.3, 47.3 (d, J = 10.25 Hz), 22.4. UPLC-MS (A) t_R = 1.27min; m/z 369 $[M-H]^-$ (>95%).

3-(4-Bromophenyl)pyrrolidine-2,5-dione (147). Crude 3-(4-bromophenyl)-4-(methylthio)-1H-pyrrole-2,5-dione **146** (650 mg, 2.180 mmol) was dissolved in glacial acetic acid (42 mL). Powdered zinc (713 mg, 10.90mmol) was added and the reaction was refluxed for 1.5h. Additional portion of powdered zinc (603 mg, 9.22 mmol) was added and the reaction was refluxed for another 1 hour. Reaction mixture was cooled to room temperature, water was added and the mixture was extracted with DCM (4x). Combined organic fractions were washed with saturated sodium bicarbonate (3x - until the emission of gas was not observed anymore), and brine, dried with sodium sulfate and evaporated. The product was purified via flash column chromatography on silicagel (EtOAc:Cyclohexane 10-60%). White solid, yield 75% (415.3 mg, 1.633 mmol). 1H NMR (400 MHz, DMSO- d_6) δ 11.34 (br. s., 1H), 7.54 (d, J = 8.34 Hz, 2H), 7.28 (d, J = 8.34 Hz, 2H), 4.15 (dd, J = 5.56, 9.35 Hz, 1H), 3.10 (dd, J = 9.47, 18.06 Hz, 1H), 2.73 (dd, J = 5.56, 17.94 Hz, 1H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 179.1, 177.5, 137.4, 131.5, 130.2, 120.4, 46.3, 37.6. UPLC-MS (A) t_R = 1.04min; m/z 252 $[M-H]^-$ (>95%).

3-(4-Bromophenyl)-1-(2-(2,4-difluorophenyl)-2-oxoethyl)pyrrolidine-2,5-dione (148). The title compound was prepared from 3-(4-bromophenyl)pyrrolidine-2,5-dione **147** (100 mg, 0.394 mmol) and 2-bromo-1-(2,4-difluorophenyl)ethan-1-one **16** (102 mg, 0.433 mmol) following general method B. White solid, yield 41% (66 mg, 0.162 mmol). 1H NMR (400 MHz, CHLOROFORM- d) δ 8.04 (dt, J = 6.65, 8.47 Hz, 1H), 7.49 - 7.58 (m, 2H), 7.25 (d, J = 8.28 Hz, 2H), 6.92 - 7.05 (m, 2H), 4.91 (d, J = 4.02 Hz, 2H), 4.13 (dd, J = 4.77, 9.54 Hz, 1H), 3.35 (dd, J = 9.79, 18.57 Hz, 1H), 2.89 (dd, J = 5.02, 18.57 Hz, 1H). ^{13}C NMR (101 MHz, CHLOROFORM- d)

δ 186.9 (d, $J = 5.87$ Hz), 176.8, 175.2, 166.7 (dd, $J = 12.47, 259.71$ Hz), 163.4 (dd, $J = 13.21, 258.24$ Hz), 136.3, 133.1 (dd, $J = 4.40, 11.01$ Hz), 132.3, 129.4, 122.0, 119.0 (dd, $J = 3.67, 13.94$ Hz), 112.9 (dd, $J = 2.93, 22.01$ Hz), 104.9 (t, $J = 28.60$ Hz), 48.5 (d, $J = 14.67$ Hz), 45.6, 37.4. UPLC-MS (B) $t_R = 1.96$ min; m/z 406 $[M-H]^-$ (>95%).

3-(4-Bromophenyl)-1-(2-(2,4-difluorophenyl)-2-oxoethyl)-3-methylpyrrolidine-2,5-dione (149). 3-(4-Bromophenyl)-1-(2-(2,4-difluorophenyl)-2-oxoethyl)pyrrolidine-2,5-dione **148** (55 mg, 0.135 mmol) and potassium carbonate (20.48 mg, 0.148 mmol) were suspended in dry Acetone (1 mL). Iodomethane (0.042 mL, 0.674 mmol) was added and the reaction was left stirring at room temperature for 2 days. Reaction mixture was evaporated, and dissolved in a mixture of ethyl acetate and water. Aqueous phase was washed with ethyl acetate. Combined organic fractions were washed with brine, dried with sodium sulfate and evaporated. Column chromatography on silicagel provided the product which was further lyophilized. White solid, yield 28% (16 mg, 0.038 mmol). 1H NMR (400 MHz, CHLOROFORM- d) δ 7.98 - 8.09 (m, $J = 6.50, 8.40, 8.40$ Hz, 1H), 7.50 - 7.59 (m, 2H), 7.32 - 7.41 (m, 2H), 7.00 - 7.08 (m, 1H), 6.96 (ddd, $J = 2.38, 8.60, 11.11$ Hz, 1H), 4.91 (d, $J = 4.02$ Hz, 2H), 3.16 (d, $J = 18.32$ Hz, 1H), 3.00 (d, $J = 18.57$ Hz, 1H), 1.81 (s, 3H). ^{13}C NMR (101 MHz, CHLOROFORM- d) δ 187.0 (d, $J = 5.87$ Hz), 180.2, 174.7, 166.7 (dd, $J = 13.21, 258.24$ Hz), 163.4 (dd, $J = 13.21, 258.97$ Hz), 140.7, 133.1 (dd, $J = 4.40, 11.00$ Hz), 132.0, 127.7, 121.7, 119.1 (dd, $J = 3.67, 13.94$ Hz), 112.9 (dd, $J = 2.93, 22.01$ Hz), 104.9 (dd, $J = 25.68, 27.14$ Hz), 48.4 (d, $J = 13.94$ Hz), 47.9, 45.4, 25.5. UPLC-MS (B) $t_R = 2.04$ min; m/z 422 $[M+H]^+$ (>95%).

1-(2-(2,4-Difluorophenyl)-2-hydroxyethyl)-4-(4-methoxyphenyl)-4-methylimidazolidin-2-one (150). 3-(2-(2,4-Difluorophenyl)-2-oxoethyl)-5-(4-methoxyphenyl)-5-methylimidazolidine-2,4-dione **24** (100 mg, 0.267 mmol) was dissolved in anhydrous toluene (50 mL) and sodium bis(2-

methoxyethoxy)aluminum hydride solution (Red-Al) (0.16 mL, 0.53 mmol) was added to the solution under nitrogen atmosphere. The reaction mixture was heated at 120 °C for 2h 15 min. The reaction mixture was cooled down and quenched by careful addition of aqueous NaOH (2N, 2 mL) followed by addition of ethyl acetate (20 mL) and water (70 mL). The resulting biphasic system was separated and the organic layer was further washed with water (2x 50 mL). The organic layer was evaporated under reduced pressure to provide the crude product which was purified via flash column chromatography on silicagel (EtOAc:Cyclohexane 0-100%). White solid; yield 48% (46 mg, 0.13 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 7.38 - 7.56 (m, 1H), 7.24 (dd, *J* = 8.8, 1.5 Hz, 2H), 6.95 - 7.17 (m, 3H), 6.84 - 6.91 (m, 2H), 5.59 (dd, *J* = 4.8, 2.5 Hz, 1H), 4.93 (q, *J* = 5.5 Hz, 1H), 3.74 (d, *J* = 4.3 Hz, 3H), 3.47 (dd, *J* = 46.7, 8.6 Hz, 1H), 3.15 - 3.38 (m, 3H), 1.40 (d, *J* = 2.5 Hz, 3H). ¹³C NMR (101 MHz, CDCl₃) (mixture of diastereoisomers) δ 162.5, 160.9 - 163.6 (m), 158.95, 158.93, 159.2 (dd, *J* = 247.4 Hz, 11.7 Hz), 137.1, 136.9, 128.7 - 129.2 (m), 125.9, 125.8, 124.8 - 125.3 (m), 114.0, 113.9, 111.2 (dd, *J* = 20.5 Hz, 3.7 Hz), 103.32 (t, *J* = 25.6 Hz), 103.26 (t, *J* = 25.6 Hz), 67.71, 67.66, 62.3, 62.0, 57.8, 57.7, 55.3, 51.1, 50.8, 28.0. UPLC-MS (A) *t*_R = 1.20min; *m/z* 363 [M+H]⁺ (>95%).

1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-4-(4-methoxyphenyl)-4-methylimidazolidin-2-one (151a, 151b). A solution of oxalyl chloride (1 drop – approx. 6 µL, approx. 0.07 mmol) in DCM (0.14 mL) was cooled to -78 °C. Dimethyl sulfoxide (1 drop – approx. 9.9 µL, approx. 0.14 mmol) was added and the mixture was allowed to stir for 5 min. 3-(2-(2,4-Difluorophenyl)-2-hydroxyethyl)-5-(4-methoxyphenyl)-5-methylimidazolidin-4-one **150** (23 mg, 0.063 mmol) dissolved in DCM (1 mL) was added slowly and the reaction was stirred for 15 mins before addition of triethylamine (TEA, 0.04 mL, 0.3 mmol). The reaction mixture was left to warm to room temperature and stirred for 2 h. Then, the reaction mixture was diluted with water (50 mL)

and organics were extracted with ethyl acetate (50 mL x 3). The combined organic layers were washed with brine (50 mL), dried over magnesium sulfate and evaporated. Purification by preparative HPLC (Chiralpak IC 20x250 cm, Heptane:Methanol-Ethanol 80:20, 45 min, 18 mL/min) to obtain two enantiomers (**151a** and **151b**). **151a**: colourless solid; yield 35 % (8 mg, 0.02 mmol); ^1H NMR (400 MHz, CDCl_3) δ 8.01 (td, $J = 8.5, 6.6\text{Hz}$, 1H), 7.38 - 7.46 (m, 2H), 6.95 - 7.04 (m, 1H), 6.85 - 6.94 (m, 3H), 4.87 (s, 1H), 4.64 (dd, $J = 18.7, 4.3\text{Hz}$, 1H), 4.49 (dd, $J = 18.9, 4.0\text{Hz}$, 1H), 3.82 (s, 3H), 3.61 (dd, $J = 12.6, 8.3\text{Hz}$, 2H), 1.77 (s, 3H). ^{13}C NMR (101 MHz, CDCl_3) δ ppm 191.6 (d, $^3J_{\text{CF}} = 6.6\text{Hz}$), 166.3 (dd, $^1J_{\text{CF}} = 258.3\text{Hz}$, $^3J_{\text{CF}} = 12.4\text{Hz}$), 163.1 (dd, $^1J_{\text{CF}} = 256.9\text{Hz}$, $^3J_{\text{CF}} = 12.4\text{Hz}$), 160.8, 158.9, 137.6, 132.8 (dd, $^3J_{\text{CF}} = 10.2\text{Hz}$, $^3J_{\text{CF}} = 5.1\text{Hz}$), 126.3, 119.9 (dd, $^2J_{\text{CF}} = 15.4\text{Hz}$, $^4J_{\text{CF}} = 3.7\text{Hz}$), 113.9, 112.6 (dd, $^2J_{\text{CF}} = 22.0\text{Hz}$, $^4J_{\text{CF}} = 2.9\text{Hz}$), 104.8 (t, $^2J_{\text{CF}} = 27.1\text{Hz}$), 60.6, 57.0, 55.3, 53.2 (d, $^4J_{\text{CF}} = 12.4\text{Hz}$), 28.1. UPLC-MS (A) $t_{\text{R}} = 1.19\text{min}$; m/z 361 $[\text{M}+\text{H}]^+$ (>95%). **151b**: colourless solid; yield 32 % (7 mg, 0.02 mmol); ^1H NMR (400 MHz, CDCl_3) δ ppm 8.01 (td, $J = 8.5, 6.6\text{Hz}$, 1H), 7.38 - 7.45 (m, 2H), 6.96 - 7.03 (m, 1H), 6.86 - 6.94 (m, 3H), 4.87 (s, 1H), 4.64 (dd, $J = 18.9, 4.0\text{Hz}$, 1H), 4.49 (dd, $J = 18.7, 4.0\text{Hz}$, 1H), 3.82 (s, 3H), 3.61 (dd, $J = 12.4, 8.1\text{Hz}$, 2H), 1.77 (s, 3H). ^{13}C NMR (101 MHz, CDCl_3) δ ppm 191.6 (d, $^3J_{\text{CF}} = 5.9\text{Hz}$), 166.3 (dd, $^1J_{\text{CF}} = 258.3\text{Hz}$, $^3J_{\text{CF}} = 12.4\text{Hz}$), 163.1 (dd, $^1J_{\text{CF}} = 257.6\text{Hz}$, $^3J_{\text{CF}} = 12.4\text{Hz}$), 160.8, 158.9, 137.6, 132.8 (dd, $^3J_{\text{CF}} = 10.6\text{Hz}$, $^3J_{\text{CF}} = 4.8\text{Hz}$), 126.3, 119.9 (dd, $^2J_{\text{CF}} = 14.6\text{Hz}$, $^4J_{\text{CF}} = 4.4\text{Hz}$), 113.9, 112.6 (dd, $^2J_{\text{CF}} = 22.0\text{Hz}$, $^4J_{\text{CF}} = 2.9\text{Hz}$), 104.8 (t, $^2J_{\text{CF}} = 26.8\text{Hz}$), 60.6, 57.0, 55.3, 53.2 (d, $^4J_{\text{CF}} = 12.4\text{Hz}$), 28.1. UPLC-MS (A) $t_{\text{R}} = 1.19\text{min}$; m/z 361 $[\text{M}+\text{H}]^+$ (>95%).

3-(4-Bromophenyl)-3-methylpyrrolidin-2-one (154). Methyl 2-(4-bromophenyl)-3-cyano-2-methylpropanoate **153** (1.373 g, 4.86 mmol) and cobalt (II) chloride hexahydrate ($\text{CoCl}_2 \times 6\text{H}_2\text{O}$) (2.315 g, 9.73 mmol) were dissolved in dry methanol (50 mL) under nitrogen atmosphere and cooled to 0°C . Sodium borohydride (1.840 g, 48.6 mmol) was added in portions while maintaining

the temperature of the mixture below 5°C. The reaction mixture was allowed to reach room temperature and was left stirring at room temperature overnight. Subsequently the reaction was quenched with 1M HCl and neutralised with saturated sodium bicarbonate. The mixture was extracted with ethyl acetate (3x). Combined organic phases were washed with brine, dried with sodium sulfate, evaporated and purified via flash column chromatography on silicagel (EtOAc:Cyclohexane 0-60%). White solid, yield 57% (708 mg, 2.79 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 7.83 (br. s., 1H), 7.47 - 7.55 (m, 2H), 7.29 - 7.41 (m, 2H), 3.18 - 3.28 (m, 1H), 3.04 - 3.14 (m, 1H), 2.35 (ddd, *J* = 5.05, 7.45, 12.76 Hz, 1H), 2.14 (ddd, *J* = 6.57, 7.71, 12.76 Hz, 1H), 1.37 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 178.7, 143.7, 131.0, 128.5, 119.5, 46.8, 38.0, 36.7, 24.3. UPLC-MS (A) *t*_R = 1.07min; *m/z* 254 [M+H]⁺ (>95%).

3-(4-Bromophenyl)-1-(2-(2,4-difluorophenyl)-2-oxoethyl)-3-methylpyrrolidin-2-one (155).

3-(4-bromophenyl)-3-methylpyrrolidin-2-one **154** (100 mg, 0.394 mmol) and sodium hydride (60% in mineral oil) (14.95 mg, 0.374 mmol) were suspended in dry THF (Volume: 3 ml) and stirred at room temperature for 10 minutes. 2-Bromo-1-(2,4-difluorophenyl)ethanone **16** (102 mg, 0.433 mmol) was added and the reaction was left stirring at room temperature overnight. The reaction was concentrated *in vacuo* and resuspended in ethyl acetate and water. The aqueous phase was washed with ethyl acetate and the combined organic phases were washed with brine (3x), dried with sodium sulfate, evaporated and purified via flash column chromatography on silicagel (EtOAc:Heptane 0-40%). White solid, yield 4.9% (7.8 mg, 0.019 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 7.95 - 8.06 (m, 1H), 7.46 - 7.62 (m, 3H), 7.41 (d, *J* = 8.53 Hz, 2H), 7.29 (dt, *J* = 2.26, 8.41 Hz, 1H), 4.59 - 4.80 (m, 2H), 3.39 - 3.50 (m, 2H), 2.28 - 2.39 (m, 1H), 2.14 - 2.26 (m, 1H), 1.47 (s, 3H). UPLC-MS (B) *t*_R = 1.94min; *m/z* 408 [M+H]⁺ (>95%).

4-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-3,4-dimethyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (156). To a suspension of 4-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **1** (25 mg, 0.068 mmol) and potassium carbonate (10.29 mg, 0.074 mmol) in acetone (1 mL), methyl iodide (10.57 mg, 0.074 mmol) was added and the reaction mixture was left stirring overnight. Additional portions of methyl iodide (10.57 mg, 0.074 mmol) and potassium carbonate (10.29 mg, 0.074 mmol) were added and the reaction was left stirring overnight. The reaction mixture was evaporated and suspended in a mixture withyl acetate and water. Organic layer was washed with brine, dried with sodium sulfate, evaporated and purified via flash column chromatography on silicagel (EtOAc:Cyclohexane 10-50%). White solid, yield 52% (13.6 mg, 0.035 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 7.99 - 8.05 (m, 1H), 7.97 (d, *J* = 8.59 Hz, 2H), 7.66 (d, *J* = 8.34 Hz, 2H), 7.48 - 7.55 (m, 1H), 7.25 - 7.32 (m, 1H), 4.82 - 4.93 (m, 2H), 2.75 (s, 3H), 1.87 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.5 (d, *J* = 5.12 Hz), 174.0, 166.3 (m, *J* = 13.17, 256.14 Hz), 163.0 (dd, *J* = 13.17, 257.61 Hz), 155.1, 142.3, 133.4, 133.1 (dd, *J* = 4.03, 11.34 Hz), 128.0, 119.9 (dd, *J* = 3.66, 13.17 Hz), 118.8, 113.3 (dd, *J* = 2.93, 22.69 Hz), 112.1, 106.0 (t, *J* = 27.10 Hz), 67.1, 48.2 (d, *J* = 10.98 Hz), 25.7, 19.9. UPLC-MS (A) t_R = 1.22min; *m/z* 384 [M+H]⁺ (>95%).

4-(3-Acetyl-1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (157). To a mixture of 4-(1-(2-(2,4-difluorophenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **1** (50 mg, 0.135 mmol) and triethylamine (0.038 ml, 0.271 mmol) in dioxane (Volume: 2 ml), acetyl chloride (0.019 ml, 0.271 mmol) was added and the reaction was left stirring at room temperature overnight. Additional portions of triethylamine (0.038 ml, 0.271 mmol) and acetyl chloride (0.019 ml, 0.271 mmol) were added and the reaction was left stirring at room temperature overnight. Additional portions of triethylamine (0.038 ml,

0.271 mmol) and acetyl chloride (0.019 ml, 0.271 mmol) were added and the reaction was left stirring at room temperature for another night until no further progress was observed. Reaction mixture was concentrated *in vacuo* and resuspended in ethyl acetate and water. Aqueous phase was washed with ethyl acetate and the combined organic phases were washed with brine (3x), dried with sodium sulfate, evaporated and purified via flash column chromatography on reversed phase (Acetonitrile:Water 40-75%). White solid, yield 25% (13.9 mg, 0.034 mmol). ¹H NMR (400 MHz, CDCl₃) δ 8.07 (dt, *J* = 6.65, 8.47 Hz, 1H), 7.57 - 7.80 (m, 4H), 6.89 - 7.13 (m, 2H), 4.83 - 5.04 (m, 2H), 2.60 (s, 3H), 2.20 (s, 3H). UPLC-MS (B) *t_R* = 1.79min; *m/z* 410 [M-H]⁻ (>95%).

4-(1H-imidazol-4-yl)benzonitrile (158). 4-(2-Bromoacetyl)benzonitrile (1g, 4.46 mmol) was heated in formamide (10 ml, 252 mmol) at 170 °C for 1h20min. The reaction mixture was cooled to room temperature, and diluted with saturated aqueous sodium bicarbonate. The precipitate was filtered and washed with water to form batch 1. The filtrate was extracted with ethyl acetate (4x). Combined organic fractions were extracted with brine, dried with sodium sulfate and evaporated to form batch 2. Flash column chromatography on reversed phase of batch 1 (Methanol:Water 0-100%) yielded the pure product as well as fractions containing the product contaminated with formamide. These fractions were combined with batch 2 and lyophilized. The residue was further purified with preparative HPLC (XBridge30, ACN:Water 20-100%, 5min, 10mM ammonium bicarbonate). Obtained fractions were evaporated, dissolved in ethyl acetate, washed with water, dried with sodium sulfate, evaporated and combined with purified fractions of batch 1. Off-white solid, yield 52% (390 mg, 2.305 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 12.38 (br. s., 1H), 7.96 (d, *J* = 7.58 Hz, 2H), 7.86 (br. s., 1H), 7.70 - 7.82 (m, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 139.5, 138.4, 136.7, 132.5, 124.7, 119.3, 115.3, 107.9. UPLC-MS (A) *t_R* = 0.93min; *m/z* 170 [M+H]⁺ (>95%).

4-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-1H-imidazol-4-yl)benzonitrile (160). 4-(1H-imidazol-4-yl)benzonitrile **158** (200 mg, 1.182 mmol) and potassium carbonate (180 mg, 1.300 mmol) were stirred in a water/ice bath for 15 minutes. Subsequently 2-chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (248 mg, 1.30 mmol) was added and the reaction was left stirring for 3h in a water/ice bath. Reaction was left stirring at room temperature overnight. Reaction mixture was filtered and evaporated and the residue was partitioned between DCM and water. The aqueous phase was extracted with 2xDCM and the combined organic phases were extracted 2x with water, dried with sodium sulfate, evaporated and purified by preparative HPLC (Xbridge19, ACN:Water 20-100, 10mM ammonium bicarbonate). Collected fractions were evaporated, dissolved in DCM, washed 2x with water, dried with sodium sulfate and evaporated. Yellowish solid, yield 9% (36 mg, 0.111 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 8.06 (dt, *J* = 6.95, 8.53 Hz, 1H), 7.93 (d, *J* = 8.34 Hz, 2H), 7.80 (d, *J* = 8.84 Hz, 3H), 7.73 (s, 1H), 7.52 - 7.59 (m, 1H), 7.33 (dt, *J* = 2.40, 8.40 Hz, 1H), 5.64 (d, *J* = 3.03 Hz, 2H). UPLC-MS (A) *t_R* = 1.13min; *m/z* 324 [M+H]⁺ (>95%).

1-(2,4-Difluorophenyl)-2-(4-(4-methoxyphenyl)-1H-imidazol-1-yl)ethanone (161).

4-(4-Methoxyphenyl)-1H-imidazole **159** (100 mg, 0.574 mmol) and cesium carbonate (224 mg, 0.687 mmol) were suspended in DMF (2.5 mL). 2-Chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (219 mg, 1.15 mmol) was added to the reaction mixture and it was heated at 80 °C for 90 min. The reaction mixture was cooled down, poured into water (50 mL) and extracted with ethyl acetate (50 mL x 3). The combined organic layers were evaporated under reduced pressure. The residue was purified via flash column chromatography on silicagel (Ethyl acetate:Cyclohexane 20-80%). Off-white solid; yield 41% (77 mg, 0.24 mmol); ¹H NMR (400 MHz, DMSO-d₆) δ 8.06 (dd, *J* = 8.6, 1.8 Hz, 1H), 7.62 - 7.72 (m, 2H), 7.60 (d, *J* = 1.0 Hz, 1H), 7.54 (ddd, *J* = 11.6, 9.3, 2.4 Hz, 1H),

7.42 (d, $J = 1.0$ Hz, 1H), 7.32 (td, $J = 8.4, 2.4$ Hz, 1H), 6.88 - 6.98 (m, 2H), 5.58 (d, $J = 3.0$ Hz, 2H), 3.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 190.3 (d, $^3J_{\text{CF}} = 5.1$ Hz), 165.6 (dd, $^1J_{\text{CF}} = 254.7$ Hz, $^3J_{\text{CF}} = 12.4$ Hz), 162.4 (dd, $^1J_{\text{CF}} = 258.3$ Hz, $^3J_{\text{CF}} = 13.2$ Hz), 157.8, 140.0, 138.6, 132.6 (dd, $^3J_{\text{CF}} = 11.0$ Hz, $^3J_{\text{CF}} = 3.7$ Hz), 127.4, 125.4, 119.9 (dd, $^2J_{\text{CF}} = 13.9$ Hz, $^4J_{\text{CF}} = 3.7$ Hz), 115.8, 113.9, 112.7 (dd, $^2J_{\text{CF}} = 22.0$ Hz, $^4J_{\text{CF}} = 3.7$ Hz), 105.4 (t, $^2J_{\text{CF}} = 26.8$ Hz), 55.6 (d, $^4J_{\text{CF}} = 11.0$ Hz), 55.0. UPLC-MS (A) $t_{\text{R}} = 1.14\text{min}$; m/z 329 $[\text{M}+\text{H}]^+$ (>95%).

4-(1H-pyrazol-4-yl)benzonitrile (162). 4-Bromopyrazole (1.5 g, 10.21 mmol) was dissolved in anhydrous DMF (15 mL) under nitrogen and cooled down to 0 °C. Potassium *tert*-butoxide (1.374 g, 12.25 mmol) was added, following by the addition of triphenylmethyl chloride (3.13 g, 11.23 mmol). The reaction was allowed to warm up to room temperature and was left stirring for 1h. Reaction mixture was carefully diluted with water and neutralized with saturated aqueous ammonium chloride and extracted with ethyl acetate (3x). Combined organic fractions were washed with brine, dried with sodium sulfate and evaporated. Flash column chromatography on silicagel (Cyclohexane:EtOAc 0-5%) provided 4-bromo-1-trityl-1H-pyrazole. White solid, yield 36% (1.410 g, 3.62 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 7.76 (s, 1H), 7.51 (s, 1H), 7.30 - 7.41 (m, 9H), 7.00 - 7.09 (m, 6H).

Obtained 4-bromo-1-trityl-1H-pyrazole (400 mg, 1.028 mmol) and (4-cyanophenyl)boronic acid (166 mg, 1.130 mmol) were dissolved in a degassed mixture of 1,4-dioxane (12 mL) and water (5.00 mL) under nitrogen in an oven-dried vial. Nitrogen was bubbled through the mixture for 5 minutes. Tetrakis(triphenylphosphine)palladium (0) (119 mg, 0.103 mmol) was added followed by a degassed aqueous solution of potassium carbonate (0.411 mL, 2.055 mmol). The final mixture was flushed with nitrogen and the vial was sealed. The reaction mixture was heated at 85 °C overnight. Subsequently the reaction mixture was cooled down to room temperature,

trifluoroacetic acid (5ml, 65.3 mmol) was added and the reaction was heated at 70 °C for 1 h. The reaction mixture was neutralized with 1M NaOH, concentrated *in vacuo* and partitioned between DCM and water. The aqueous phase was washed 4x with DCM. The combined organic phases were dried with sodium sulfate, evaporated and purified via flash column chromatography on silicagel (Acetone:DCM 0-100%). Off-white solid, yield 51% (118.3 mg, 0.699 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 13.15 (br. s., 1H), 8.40 (br. s., 1H), 8.08 (br. s., 1H), 7.70 - 7.87 (m, 4H). UPLC-MS (A) t_R = 0.97min; m/z 168 [M-H]⁻ (>95%).

4-(1-(2-(2,4-Difluorophenyl)-2-oxoethyl)-1H-pyrazol-4-yl)benzonitrile (163). A mixture of 2-Chloro-1-(2,4-difluorophenyl)ethan-1-one **6** (74.3 mg, 0.390 mmol), 4-(1H-pyrazol-4-yl)benzonitrile **162** (60 mg, 0.355 mmol) and potassium carbonate (53.9 mg, 0.390 mmol) in DMF (1 ml) was irradiated for 30 minutes in a microwave at 60 °C. The reaction mixture was cooled down to room temperature and poured into water. Reaction mixture was extracted with ethyl acetate (4x), dried with sodium sulfate and evaporated. The residue was purified via preparative HPLC (XBridge 30, ACN:Water 40-100%, 10mM ammonium bicarbonate). The collected fractions were washed with water, dried with sodium sulfate, evaporated and lyophilised. White solid, yield 7% (8 mg, 0.025 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 8.33 (s, 1H), 8.11 (s, 1H), 8.00 - 8.07 (m, 1H), 7.78 - 7.84 (m, 4H), 7.49 - 7.56 (m, 1H), 7.31 (dt, *J* = 2.40, 8.40 Hz, 1H), 5.73 (d, *J* = 2.78 Hz, 2H). ¹³C NMR (101 MHz, DMSO-d₆) δ 190.1 (d, *J* = 5.12 Hz), 165.6 (dd, *J* = 12.44, 255.41 Hz), 161.9 (dd, *J* = 13.17, 257.61 Hz), 137.3, 132.9, 132.6 (dd, *J* = 4.39, 10.98 Hz), 130.1, 125.5, 120.7, 119.9 (dd, *J* = 3.66, 13.17 Hz), 119.1, 112.7 (dd, *J* = 2.93, 21.96 Hz), 108.1, 105.4 (t, *J* = 27.10 Hz), 60.7 (d, *J* = 10.25 Hz). UPLC-MS (A) t_R = 1.20min; m/z 322 [M-H]⁻ (>95%).

5-(4-(Difluoromethoxy)phenyl)-3-(2-(4-fluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (180). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-chloro-1-(4-fluorophenyl)ethanone **9** (51 mg, 0.29 mmol) following general method B. White solid; yield 72% (55 mg, 0.14 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.10 (s, 1H), 8.13 (m, *J* = 8.8, 5.6 Hz, 2H), 7.60 (d, *J* = 8.8 Hz, 2H), 7.06 - 7.47 (m, 5H), 4.98 (s, 2H), 1.76 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 190.9, 175.2, 165.5 (d, ¹*J*_{CF} = 253.2 Hz), 155.1, 150.7 (t, ³*J*_{CF} = 2.9 Hz), 136.3, 131.3 (d, ³*J*_{CF} = 9.5 Hz), 130.7 (d, ⁴*J*_{CF} = 2.9 Hz), 127.4, 118.7, 116.1 (d, ²*J*_{CF} = 22.0 Hz), 116.3 (t, ¹*J*_{CF} = 257.6 Hz), 62.9, 44.5, 25.0. UPLC-MS (A) *t*_R = 1.21min; *m/z* 391 [M-H]⁻ (>95%). HRMS (ESI) *m/z* calcd for C₁₉H₁₆F₃N₂O₄ [M+H]⁺: 393.1057; found: 393.1052.

5-(4-(Difluoromethoxy)phenyl)-3-(2-(3-fluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (181). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(3-fluorophenyl)ethan-1-one **164** (64 mg, 0.29 mmol) following general method B. White solid; yield 81% (62 mg, 0.16 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.11 (s, 1 H), 7.90 (dt, *J* = 7.6, 1.0 Hz, 1H), 7.85 (dt, *J* = 9.5, 1.9 Hz, 1H), 7.55 - 7.69 (m, 4H), 7.05 - 7.50 (m, 3H), 5.00 (s, 2H), 1.76 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 191.5, 175.1, 162.1 (d, ¹*J*_{CF} = 245.9 Hz), 155.1, 150.7 (t, ³*J*_{CF} = 3.3 Hz), 136.2, 136.0 (d, ³*J*_{CF} = 6.6 Hz), 131.2 (d, ³*J*_{CF} = 8.1 Hz), 127.4, 124.4 (d, ⁴*J*_{CF} = 2.9 Hz), 121.2 (d, ²*J*_{CF} = 22.7 Hz), 118.7, 114.8 (d, ²*J*_{CF} = 22.7 Hz), 116.3 (t, ¹*J*_{CF} = 258.3 Hz), 62.9, 44.7, 24.9. UPLC-MS (A) *t*_R = 4.05min; *m/z* 391 [M-H]⁻ (>95%). HRMS (ESI) *m/z* calcd for C₁₉H₁₆F₃N₂O₄ [M+H]⁺: 393.1057; found: 393.1070.

5-(4-(Difluoromethoxy)phenyl)-3-(2-(2-fluorophenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (182). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-

methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(2-fluorophenyl)ethan-1-one **165** (64 mg, 0.29 mmol) following general method B. Orange solid; yield 73% (56 mg, 0.14 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.09 (s, 1H), 7.90 (td, J = 7.6, 1.8 Hz, 1H), 7.71 - 7.80 (m, 1H), 7.56 - 7.62 (m, 2H), 7.06 - 7.48 (m, 5H), 4.81 (d, J = 2.5 Hz, 2H), 1.75 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 190.3 (d, J_{CF} = 4.4 Hz), 175.0, 161.5 (d, J_{CF} = 254.7 Hz), 155.0, 150.7 (t, J_{CF} = 2.9 Hz), 136.3 (d, J_{CF} = 9.5 Hz), 136.2, 130.3 (d, J_{CF} = 2.9 Hz), 127.4, 125.1 (d, J_{CF} = 3.7 Hz), 122.4 (d, J_{CF} = 13.2 Hz), 118.7, 117.0 (d, J_{CF} = 22.7 Hz), 116.3 (t, J_{CF} = 257.6 Hz), 62.9, 47.3 (d, J_{CF} = 11.0 Hz), 24.9. UPLC-MS (A) t_{R} = 1.27 min; m/z 393 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{19}\text{H}_{16}\text{F}_3\text{N}_2\text{O}_4$ $[\text{M}+\text{H}]^+$: 393.1057; found: 393.1072.

4-(Difluoromethoxy)phenyl)-5-methyl-3-(2-oxo-2-phenylethyl)imidazolidine-2,4-dione (183). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-chloro-1-phenylethan-1-one **166** (45 mg, 0.29 mmol) following general method B. White solid; yield 61% (45 mg, 0.12 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.09 (s, 1H), 8.00 - 8.08 (m, 2H), 7.69 - 7.75 (m, 1H), 7.54 - 7.64 (m, 4H), 7.06 - 7.47 (m, 3H), 4.98 (s, 2H), 1.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 192.2, 175.2, 155.2, 150.6 (t, $^3J_{\text{CF}}$ = 2.9 Hz), 136.3, 134.2, 134.0, 129.0, 128.1, 127.4, 118.7, 116.3 (t, $^1J_{\text{CF}}$ = 259.1 Hz), 62.9, 44.6, 25.0. UPLC-MS (A) t_{R} = 1.21 min; m/z 375 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{19}\text{H}_{17}\text{F}_2\text{N}_2\text{O}_4$ $[\text{M}+\text{H}]^+$: 375.1151; found: 375.1147.

4-(1-(2-(4-Methoxyphenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (184). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) and 2-bromo-1-(4-methoxyphenyl)ethan-1-one **167** (82 mg, 0.358 mmol) following general method B. White solid, yield 60% (71 mg, 0.195 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.20 (s, 1H), 8.01 (d, J = 8.78 Hz, 2H), 7.94 (d, J = 8.28 Hz, 2H), 7.77

(d, $J = 8.53$ Hz, 2H), 7.09 (d, $J = 9.04$ Hz, 2H), 4.92 (s, 2H), 3.86 (s, 3H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 190.2, 174.6, 163.9, 155.2, 144.7, 132.6, 130.6, 126.8, 126.8, 118.5, 114.2, 111.0, 63.3, 55.7, 44.4, 25.0. UPLC-MS (B) $t_R = 1.57$ min; m/z 364 $[\text{M}+\text{H}]^+$ (>95%).

5-(4-(Difluoromethoxy)phenyl)-3-(2-(4-methoxyphenyl)-2-oxoethyl)-5-methylimidazolidine-2,4-dione (185). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(4-methoxyphenyl)ethan-1-one **167** (67 mg, 0.29 mmol) following general method B. White solid; yield 61% (52 mg, 0.13 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.06 (s, 1H), 7.94 - 8.09 (m, 2H), 7.56 - 7.64 (m, 2H), 7.03 - 7.47 (m, 5H), 4.90 (s, 2H), 3.86 (s, 3H), 1.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 190.3, 175.3, 163.8, 155.3, 150.6 (t, $^3J_{CF} = 2.9$ Hz), 136.3, 130.5, 127.4, 126.9, 118.7, 114.2, 116.3 (t, $^1J_{CF} = 257.6$ Hz), 62.8, 55.6, 44.2, 24.9. UPLC-MS (A) $t_R = 1.28$ min; m/z 405 $[\text{M}+\text{H}]^+$ (>95%).

4-(1-(2-(3-Methoxyphenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (186). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) and 2-bromo-1-(3-methoxyphenyl)ethan-1-one **168** (82 mg, 0.358 mmol) following general method B. White solid, yield 70% (83 mg, 0.228 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.23 (br. s., 1H), 7.94 (d, $J = 8.28$ Hz, 2H), 7.76 (d, $J = 8.53$ Hz, 2H), 7.64 (d, $J = 7.78$ Hz, 1H), 7.44 - 7.56 (m, 2H), 7.28 (dd, $J = 2.01, 8.28$ Hz, 1H), 4.99 (s, 2H), 3.82 (s, 3H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 192.0, 174.6, 159.5, 155.1, 144.6, 135.2, 132.6, 130.2, 126.8, 120.6, 120.5, 118.5, 112.5, 111.1, 63.3, 55.4, 44.9, 25.0. UPLC-MS (B) $t_R = 1.59$ min; m/z 364 $[\text{M}+\text{H}]^+$ (>95%).

4-(1-(2-(2-Methoxyphenyl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (187). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-

yl)benzonitrile **5** (70 mg, 0.325 mmol) and 2-bromo-1-(2-methoxyphenyl)ethan-1-one **169** (82 mg, 0.358 mmol) following general method B. White solid, yield 75% (88 mg, 0.242 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.17 (s, 1H), 7.94 (d, *J* = 8.53 Hz, 2H), 7.69 - 7.79 (m, 3H), 7.60 - 7.68 (m, 1H), 7.25 (d, *J* = 8.28 Hz, 1H), 7.08 (t, *J* = 7.40 Hz, 1H), 4.74 (s, 2H), 3.94 (s, 3H), 1.77 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 192.3, 174.5, 159.6, 155.2, 144.7, 135.6, 132.6, 130.2, 126.8, 123.7, 120.8, 118.6, 112.8, 111.0, 63.2, 56.0, 48.5, 24.9. UPLC-MS (B) t_R = 1.59min; *m/z* 364 [M+H]⁺ (>95%).

4-(4-Methyl-2,5-dioxo-1-(2-oxo-2-(3-(trifluoromethyl)phenyl)ethyl)imidazolidin-4-yl)benzonitrile (188). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) and 2-bromo-1-(3-(trifluoromethyl)phenyl)ethan-1-one **170** (96 mg, 0.358 mmol) following general method B. White solid, yield 28% (37 mg, 0.091 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.25 (br. s., 1H), 8.25 - 8.36 (m, 2H), 8.09 (d, *J* = 7.78 Hz, 1H), 7.94 (d, *J* = 8.53 Hz, 2H), 7.83 (t, *J* = 7.78 Hz, 1H), 7.76 (d, *J* = 8.53 Hz, 2H), 5.11 (s, 2H), 1.79 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 191.8, 174.5, 155.0, 144.6, 134.7, 132.6, 132.3, 130.6 (q, *J* = 3.70 Hz), 130.4, 129.7 (q, *J* = 32.30 Hz), 126.7, 124.7 (q, *J* = 3.67 Hz), 123.7 (q, *J* = 272.90 Hz), 118.5, 111.1, 63.3, 44.9, 25.0. UPLC-MS (B) t_R = 1.72min; *m/z* 400 [M-H]⁻ (>95%).

4-(4-Methyl-1-(2-(3-nitrophenyl)-2-oxoethyl)-2,5-dioxoimidazolidin-4-yl)benzonitrile (189). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) and 2-bromo-1-(3-nitrophenyl)ethan-1-one **171** (96 mg, 0.358 mmol) following general method B. White solid, yield 45% (55 mg, 0.145 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.26 (s, 1H), 8.70 (t, *J* = 1.76 Hz, 1H), 8.53 (dd, *J* = 1.51, 8.28 Hz, 1H), 8.46 (d, *J* = 8.03 Hz, 1H), 7.94 (d, *J* = 8.53 Hz, 2H), 7.88 (t, *J* = 8.03 Hz, 1H), 7.76 (d, *J* = 8.53

Hz, 2H), 5.13 (s, 2H), 1.79 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 191.4, 174.5, 155.0, 148.1, 144.6, 135.0, 134.5, 132.6, 130.8, 128.4, 126.7, 122.7, 118.5, 111.1, 63.4, 45.0, 25.0. UPLC-MS (B) t_R = 1.72min; m/z 400 $[\text{M-H}]^-$ (>95%).

5-(4-(Difluoromethoxy)phenyl)-5-methyl-3-(2-oxo-2-(pyridin-4-yl)ethyl)imidazolidine-2,4-dione (190). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (80 mg, 0.31 mmol) and 2-bromo-1-(pyridin-4-yl)ethan-1-one hydrobromide **172** (132 mg, 0.468 mmol) following general method B. White solid, yield 10% (12 mg, 0.032 mmol). ^1H NMR (400 MHz, CDCl_3) δ 8.86 (d, J = 5.1 Hz, 2H), 7.68 - 7.76 (m, 2H), 7.54 - 7.63 (m, 2H), 7.13 - 7.22 (m, 2H), 6.30 - 6.76 (m, 2H), 4.85 - 4.97 (m, 2H), 1.94 (s, 3H). ^{13}C NMR (101 MHz, CDCl_3) δ 190.6, 174.8, 155.7, 151.3 (t, $^3J_{CF}$ = 2.9 Hz), 151.2, 140.0, 135.2, 127.2, 120.8, 119.9, 115.6 (t, $^1J_{CF}$ = 261.3 Hz), 63.9, 44.8, 25.3. UPLC-MS (A) t_R = 1.27min; m/z 376 $[\text{M+H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{18}\text{H}_{16}\text{F}_2\text{N}_3\text{O}_4$ $[\text{M+H}]^+$: 376.1103; found: 376.1118.

5-(4-(Difluoromethoxy)phenyl)-5-methyl-3-(2-oxo-2-(pyridin-3-yl)ethyl)imidazolidine-2,4-dione (191). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(pyridin-3-yl)ethan-1-one **173** (59 mg, 0.29 mmol) following general method B. White solid, yield 34% (25 mg, 0.067 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.21 (d, J = 1.5 Hz, 1H), 9.12 (s, 1H), 8.86 (dd, J = 4.8, 1.8 Hz, 1H), 8.37 (dt, J = 8.0, 1.9 Hz, 1H), 7.60 (m, J = 8.8 Hz, 3H), 7.06 - 7.46 (m, 3H), 5.06 (s, 2H), 1.76 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 192.1, 175.1, 155.1, 154.3, 150.7 (t, $^3J_{CF}$ = 2.9 Hz), 149.3, 136.2, 135.7, 129.5, 127.4, 124.0, 118.8, 116.3 (t, $^1J_{CF}$ = 257.6 Hz), 62.9, 44.7, 24.9. UPLC-MS (A) t_R = 1.09min; m/z 376 $[\text{M+H}]^+$ (>95%).

5-(4-(Difluoromethoxy)phenyl)-5-methyl-3-(2-oxo-2-(pyridin-2-yl)ethyl)imidazolidine-2,4-dione (192). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(pyridin-2-yl)ethan-1-one **174** (59 mg, 0.29 mmol) following general method B. White solid, yield 28% (21 mg, 0.055 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.11 (s, 1H), 8.75 - 8.81 (m, 1H), 8.07 (td, *J* = 7.6, 1.5 Hz, 1H), 7.95 - 8.00 (m, 1H), 7.75 (ddd, *J* = 7.3, 4.8, 1.3 Hz, 1H), 7.57 - 7.65 (m, 2H), 7.04 - 7.49 (m, 3H), 5.04 (s, 2H), 1.76 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 193.3, 175.2, 155.2, 150.9, 150.7 (t, ³*J*_{CF} = 2.9 Hz), 149.5, 138.0, 136.3, 128.8, 127.5, 121.9, 118.8, 116.3 (t, ¹*J*_{CF} = 257.6 Hz), 62.9, 44.2, 25.0. UPLC-MS (A) *t*_R = 1.19min; *m/z* 376 [M+H]⁺ (>95%). HRMS (ESI) *m/z* calcd for C₁₈H₁₆F₂N₃O₄ [M+H]⁺: 376.1103; found: 376.1100.

4-(1-(2-(3,5-Difluoropyridin-2-yl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (193). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (100 mg, 0.465 mmol) and 2-chloro-1-(3,5-difluoropyridin-2-yl)ethan-1-one **175** (98 mg, 0.511 mmol) following general method B. White solid, yield 78% (134 mg, 0.362 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.23 (s, 1H), 8.70 (d, *J* = 2.26 Hz, 1H), 8.18 (ddd, *J* = 2.26, 9.10, 10.98 Hz, 1H), 7.94 (d, *J* = 8.53 Hz, 2H), 7.76 (d, *J* = 8.53 Hz, 2H), 4.98 (s, 2H), 1.78 (s, 3H). ¹³C NMR (101 MHz, DMSO-d₆) δ 189.7 (d, *J* = 4.40 Hz), 174.5, 161.3 (dd, *J* = 6.60, 267.05 Hz), 158.5 (dd, *J* = 8.07, 278.05 Hz), 155.1, 144.6, 136.1 (t, *J* = 4.40 Hz), 134.4 (dd, *J* = 4.40, 24.21 Hz), 132.6, 126.8, 118.5, 114.6 (t, *J* = 22.00 Hz), 111.1, 63.3, 45.0, 25.0. UPLC-MS (B) *t*_R = 1.47min; *m/z* 369 [M-H]⁻ (>95%).

5-(4-(Difluoromethoxy)phenyl)-5-methyl-3-(2-oxo-2-(thiophen-2-yl)ethyl)imidazolidine-2,4-dione (194). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(thiophen-2-yl)ethan-1-one

176 (66 mg, 0.32 mmol) following general method B. White solid, yield 64% (52 mg, 0.14 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.09 (s, 1 H), 8.17 (dd, J = 3.8, 1.0 Hz, 1H), 8.13 (dd, J = 4.9, 0.9 Hz, 1H), 7.56 - 7.63 (m, 2H), 7.32 (dd, J = 4.8, 3.8 Hz, 1H), 7.06 - 7.46 (m, 3H), 4.92 (s, 2H), 1.75 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 185.3, 175.2, 155.1, 150.7 (t, $^3J_{\text{CF}}$ = 3.7 Hz), 140.1, 136.2, 136.1, 134.5, 129.1, 127.4, 118.7, 116.3 (t, $^1J_{\text{CF}}$ = 257.6 Hz), 62.9, 44.2, 24.8. UPLC-MS (A) t_{R} = 1.17min; m/z 381 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{17}\text{H}_{15}\text{F}_2\text{N}_2\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 381.0715; found: 381.0724.

4-(4-Methyl-2,5-dioxo-1-(2-oxo-2-(thiazol-2-yl)ethyl)imidazolidin-4-yl)benzonitrile (195). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (80 mg, 0.372 mmol) and 2-bromo-1-(thiazol-2-yl)ethan-1-one **177** (84 mg, 0.409 mmol) following general method B. White solid, yield 4.9% (6.2 mg, 0.018 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.26 (s, 1H), 8.35 (d, J = 3.01 Hz, 1H), 8.23 (d, J = 3.01 Hz, 1H), 7.94 (d, J = 8.53 Hz, 2H), 7.76 (d, J = 8.53 Hz, 2H), 5.00 (s, 2H), 1.78 (s, 3H). UPLC-MS (B) t_{R} = 1.43min; m/z 339 $[\text{M}-\text{H}]^-$ (>95%).

5-(4-(Difluoromethoxy)phenyl)-5-methyl-3-(2-oxo-2-(thiazol-2-yl)ethyl)imidazolidine-2,4-dione (196). The title compound was prepared from 5-(4-(difluoromethoxy)phenyl)-5-methylimidazolidine-2,4-dione **84** (50 mg, 0.20 mmol) and 2-bromo-1-(thiazol-2-yl)ethan-1-one **177** (60 mg, 0.29 mmol) following general method B. White solid, yield 20% (15 mg, 0.038 mmol). ^1H NMR (400 MHz, DMSO- d_6) δ 9.13 (s, 1H), 8.34 (d, J = 3.0 Hz, 1H), 8.23 (d, J = 3.0 Hz, 1H), 7.54 - 7.64 (m, 2H), 7.01 - 7.47 (m, 3H), 5.00 (s, 2H), 1.75 (s, 3H). ^{13}C NMR (101 MHz, DMSO- d_6) δ 186.0, 175.0, 163.3, 154.9, 150.7 (t, $^3J_{\text{CF}}$ = 3.3 Hz), 145.6, 136.2, 129.0, 127.4, 118.8, 116.3 (t, $^1J_{\text{CF}}$ = 257.6 Hz), 63.0, 44.1, 24.8. UPLC-MS (A) t_{R} = 1.16min; m/z 382 $[\text{M}+\text{H}]^+$ (>95%). HRMS (ESI) m/z calcd for $\text{C}_{16}\text{H}_{14}\text{F}_2\text{N}_3\text{O}_4\text{S}$ $[\text{M}+\text{H}]^+$: 382.0668; found: 382.0669.

4-(1-(2-Cyclohexyl-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile (197).

The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) and 2-bromo-1-cyclohexylethan-1-one **178** (73.4 mg, 0.358 mmol) following general method B. White solid, yield 89% (98 mg, 0.289 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.13 (s, 1H), 7.92 (d, *J* = 8.53 Hz, 2H), 7.72 (d, *J* = 8.53 Hz, 2H), 4.36 (s, 2H), 2.52 - 2.62 (m, 1H), 1.47 - 1.96 (m, 8H), 1.03 - 1.35 (m, 5H). ¹³C NMR (101 MHz, DMSO-d₆) δ 205.8, 174.3, 155.0, 144.7, 132.6, 126.7, 118.5, 111.0, 63.1, 46.9, 45.3, 27.7, 27.6, 25.3, 24.9. UPLC-MS (B) *t_R* = 1.66min; *m/z* 338 [M-H]⁻ (>95%).

4-(1-(2-(Adamantan-1-yl)-2-oxoethyl)-4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile

(198). The title compound was prepared from 4-(4-methyl-2,5-dioxoimidazolidin-4-yl)benzonitrile **5** (70 mg, 0.325 mmol) and 1-(adamantan-1-yl)-2-bromoethan-1-one **179** (92 mg, 0.358 mmol) following general method B. White solid, yield 68% (86 mg, 0.220 mmol). ¹H NMR (400 MHz, DMSO-d₆) δ 9.14 (s, 1H), 7.93 (d, *J* = 8.53 Hz, 2H), 7.73 (d, *J* = 8.53 Hz, 2H), 4.40 (s, 2H), 1.94 - 2.04 (m, 3H), 1.77 - 1.85 (m, 6H), 1.74 (s, 3H), 1.64 - 1.72 (m, 6H). ¹³C NMR (101 MHz, DMSO-d₆) δ 206.9, 174.4, 155.1, 144.7, 132.6, 126.7, 118.5, 111.0, 63.1, 44.8, 42.7, 37.1, 35.8, 27.2, 25.0. UPLC-MS (B) *t_R* = 1.92min; *m/z* 390 [M-H]⁻ (>95%).

Strain and growth conditions. *M. tuberculosis* H37Rv (ATC25618) wild-type was grown in Middlebrook 7H9-ADC broth (Difco) supplemented with 0.05% Tween 80 and on 7H10-OADC or 7H11-OADC agar (Difco) at 37 °C. Isoniazid and hygromycin were purchased from Sigma-Aldrich. When required, hygromycin (50 µg/ml) was added to the culture medium.

MIC determination. MIC determination assay was performed using a Resazurin reduction assay with fluorescent readout as described previously.³⁸ Isoniazid was used as a positive control (MIC= 1.8 µM) and Rifampicin was used as a no-growth control.

Intracellular IC₅₀ determination. The assays were performed as described previously³⁸ using Human THP-1 macrophages differentiated with PMA.

Microsomal fraction stability assays were performed as described previously.³⁸ The human biological samples were sourced ethically and their research use was in accord with the terms of the informed consents.

DprE1 enzymatic inhibition, time-dependent DprE1 inhibition and DprE1 overexpressing strain. Expression and purification of Mt-DprE1 and cloning of Mt-DprE1 were performed as described by Batt *et al.*³⁹ Enzymatic data were generated using a modified version of the assay described in that report. The new protocol is in the process of being submitted for publication.

HepG2 cytotoxicity assay; artificial membrane permeability (AMP), kinetic aqueous solubility (CLND) and hydrophobicity (chromlogD_{pH7.4}). These assays were performed as described previously.^{38,40}

hERG inhibition. Inhibition of the hERG potassium channel was determined using in vitro IonWorks patch-clamp electrophysiology as described in literature.⁴¹

Crystal structure. Crystals were grown from a mixture of ethyl acetate/heptane by slow evaporation. A suitable crystal was mounted on an Agilent SuperNova diffractometer and irradiated during ω -scans with Cu K α (1.54184Å) from a microsource, monochromated by mirror optics. Frames were collected with an Atlas CCD detector. The structure was solved with SHELXT and refined with SHELXL-2017/1⁴² via the shelxle GUI.⁴³ Graphics were prepared with ORTEP3 for Windows.⁴⁴ C₁₉H₁₃F₂N₃O₃, MM 369.32, monoclinic P2₁/n, a=7.1647(2)Å, b=18.2987(6)Å, c=25.1405(6)Å, β =95.199(2)°, data collection temperature 100.0(1)K, Z=8, R[F_o>4 σ (F_o)]=0.0678 (4684 unique reflections), wR₂=0.2048 (6647 unique reflections), GOF=1.026. The CIF was

deposited at the Cambridge Crystallographic Data Centre and can be obtained under deposition number CCDC 1828259.

Vibrational Circular Dichroism. VCD analysis and assignment was performed according to an analogous protocol published previously.⁴⁵

ASSOCIATED CONTENT

Supporting Information: Additional experimental data for intermediate compounds (synthetic protocols and analytical details); protocols used in antimicrobial evaluation experiments; chemical structures and antimicrobial activities of additional screening hits; antimicrobial activities of selected analogues against a panel of clinically relevant bacteria.

Molecular Formula Strings are available for all reported final products.

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ACKNOWLEDGMENTS

This research was funded by a Marie Curie Intra-European Grant within the 7th European Community Framework Programme (FP7-PEOPLE-2012-ITN) under the European Industrial Doctorates Scheme of the Marie Curie Initial Training Networks actions with grant agreement No 316773 and it is part of the OpenMedChem project.

The authors thank Kristof Van Hecke (UGent) for diffractometer time, and the Hercules Foundation (project AUGÉ/11/029 "3D-SPACE: 3D Structural Platform Aiming for Chemical Excellence") for funding of the diffractometer.

The authors gratefully acknowledge many scientists from GSK DDW, involved in the research.

ABBREVIATIONS USED

ACN, acetonitrile; asu, asymmetric unit; Cl_{int} , hepatic intrinsic clearance; CLND, chemiluminescent nitrogen detection; DAST, diethylaminosulfur trifluoride; dba, dibenzylideneacetone; DprE1, decaprenylphospho-beta-D-ribofuranose 2-oxidase, EtOAc, ethyl acetate; HepG2, hepatocellular carcinoma, human; *Mtb*, *Mycobacterium tuberculosis*; MeOH, methanol; MIC, minimum inhibitory concentration; MW, microwave; SAR, structure–activity relationship; TB, tuberculosis; *t*-BuXPhos, 2-Di-tert-butylphosphino-2',4',6'-triisopropylbiphenyl; TEA, trimethylamine; UPLC-MS, ultra-performance liquid chromatography-mass spectrometry; WHO, World Health Organization; XantPhos, 4,5-Bis(diphenylphosphino)-9,9-dimethylxanthene.

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MmpL3. *PLoS One* **2013**, 8 (4), e60933.

Graphical Abstract