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Synthesis of Spiro-fused Pyrazolidoylisoxazolines

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Abstract

Routes to structurally unique spiro-fused pyrazolidoylisoxazolines are reported. These methods start with mono-substituted hydrazines or hydrazides and utilize the nitrile oxide 1,3-dipolar cycloaddition reaction to generate the targeted spiro-fused bis-heterocycles. Molecular shape space diversity analyses were performed on these pyrazolidoylisoxazolines showing that manipulation of the appended R groups significantly changes the molecular shape.

Introduction

The spiro-isoxazoline motif is an important structural feature found in a number of biologically active natural products. Examples include 11-deoxyfistularin-3 and 11-oxoaerotherionin (isolated from the Caribbean sponges *Aplysina fistularis* and *Aplysina lacunosa*, respectively; **Figure 1a**), which exhibit activity against human breast (cell line MCF-7)¹ and human colon (cell line HCT 116) cancers,² respectively. Furthermore, three spirocyclic-isoxazolines, isolated from the marine sponge *Suberea clavata*, (aerophobin 1, purealdin L, and aplysinamisine II; **Figure 1b**) have been found to inhibit serine protease factors IXa and FXIa, and thus show potential as antithrombotic agents.³ The pyrazolidine moiety is also a feature in many synthetically designed bioactive compounds showing anti-hyperglycemic activity via inhibition of the serine peptidase, dipeptidyl peptidase IV (DPP-IV).⁴

Herein, synthetic routes to the unique scaffold comprised of spiro-fused pyrazolidine and isoxazoline components are presented (**1**; **Figure 1c**). Connecting these two heterocycles spirocyclically produces a uniquely functionalized molecular architecture that could find utility as a scaffold for the construction of probes targeting various biological receptors.

Results and Discussion

Retrosynthetically, the spiro-5-pyrazolidoylisoxazoline scaffold **1** can be envisioned as arising from *N'*-alkylacylhydrazide, 3-chloro-2-(chloromethyl)prop-1-ene, and nitrile oxide building-blocks as shown in **Figure 1c**. As an initial investigation of this strategy, hydrazide **2** was treated with 3-bromobenzyl bromide to afford **3** in a 68% yield (requires slow addition of the alkylating agent to minimize over-alkylation; **Scheme 1**). The resulting *N'*-alkylacylhydrazide **3** was then subjected to alkylation with 3-chloro-2-(chloromethyl)prop-1-ene in K₂CO₃/NaI/acetone to form pyrazolidine **4**. Having

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Supporting Information. Copies of ¹H-NMR and ¹³C-NMR spectra for all new compounds can be found in the supporting information file. This material is available free of charge via the Internet at <http://pubs.acs.org>.

constructed the requisite 4-methylenepyrazolidine dipolarophile, treatment of **4** with 4-chloro-*N*-hydroxybenzimidoyl chloride (**5**; prepared in two steps from 4-chlorobenzaldehyde⁵) in the presence of triethylamine affected the key 1,3-dipolar cycloaddition and provided the spirocycle **6** in 70% yield.

With Lipinski's *Rule of 5* in mind,⁶ we next set out to prepare lower molecular weight analogs of **1** (the molecular weight of **6** is 545.3 g/mol) where R¹ or R² would be non-aryl groups. Attempts to introduce the R² diversity via direct alkylation of arylhydrazide **7e** (**Scheme 2**) with low molecular weight alkylating agents (i.e., methyl iodide) resulted in very low yields (i.e. ~5%) of the desired product due to over alkylation. Reductive amination, a logical alternative, with acetaldehyde gave ~10% yield of the desired *N*-ethylacylhydrazide; use of formaldehyde in this type of reductive amination has also been reported to be ineffective.^{7,8} Interestingly, hydrazone formation from **7a/d/e/g** with acetone followed by reduction with sodium cyanoborohydride afforded the *N*-isopropylacylhydrazides **8a/d/e/g** in respectable yields (59-86%) with no chromatographic purification required (**Scheme 2**).⁷ This favorable result could be due to the purported stability of the di-substituted acylhydrazone intermediate relative to the mono- and unsubstituted hydrazones.⁷ Indeed, aldehyde/acylhydrazide condensation and subsequent reduction results in cyclic trimerization of the intermediate hydrazone to give a 1,3,5-triazinane as reported by Fox *et al.*⁸ Given these reductive amination results, spiro-fused analogs where R² was fixed as isopropyl were prepared using *Method B* as delineated in **Scheme 2**. While the conditions for pyrazolidine ring formation employed in *Method A* (**Scheme 1**) were unsuccessful (resulted in oligomer formation), we found that treatment of **8a/d/e/g** with sodium hydride in THF afforded **9a/d/e/g** in moderate yields (56-86%). Subsequent 1,3-dipolar cycloaddition via the nitrile oxide derived from **10** delivered spirocycles **11a-h**.

Four spiro-fused pyrazolidoylisoxazolines with the R¹ diversity element of **1** fixed as a methyl substituent were prepared next using *Method C* (**Scheme 3**). Arylhydrazine **12a/c** was treated with acetic anhydride to afford *N*-arylacetohydrazide **13a/c** in high yields (>95%) with no purification necessary.⁹ Employing the same conditions used in *Method B* (**Scheme 2**), these acetohydrazides were then converted to pyrazolidines **14a/c** and subsequent 1,3-dipolar cycloaddition delivered spirocycles **15a-d**.

Finally, acylation of methyl hydrazine with benzoic anhydride was investigated as an entry point in an attempt to prepare spiro-fused heterocycle **1** adorned with a methyl group at the R² position. Unfortunately, this effort resulted in formation of the undesired regioisomer **16** instead of the required hydrazide **17** (**Scheme 4**). Hydrazinolysis of methyl benzoate with methyl hydrazine was also unsuccessful in providing *N*-methylbenzohydrazide **17** (**Scheme 4**). In contrast, hydrazinolysis of methyl pyrazine-2-carboxylate (**18**; **Scheme 4**) delivered *N*'-methylpyrazine-2-carbohydrazide (**19**) in excellent yield (90%).¹⁰ The divergent regioselectivities observed with these two methods can be rationalized on the basis of the mechanistic differences described by Condon *et al.*:¹¹ acylation using an anhydride is governed by the greater nucleophilicity of the substituted nitrogen whereas acylation using a simple ester is governed by steric congestion around the neutral, tetrahedral intermediate.¹¹ With hydrazide **19** in hand, the methods employed in **Scheme 2** and **3** delivered the targeted pyrazolidine and the sequence-ending 1,3-dipolar cycloaddition delivered final product **20** in 55% yield.

A molecular shape space diversity analysis (**Figure 2**), which categorizes a compound's shape as ratios of rod-, disc-, or sphere-like character,¹² was performed on the collection of compounds reported above as well as on virtual analogs thereof. This analysis revealed that manipulation of the various R groups on the spiro-fused scaffold significantly alters the

shape ratios in the resulting triangle plots. For instance, analysis of a virtual library showed that, if R¹ is a methyl substituent, molecular shapes are localized into the rod-like region (**Figure 2a**; calculations included multiple low-energy conformers for each analog). A methyl group at R² gives results much like having large substituents at all three diversity points, but slightly shifted to the rod-like region (**Figure 2b**). Placing a methyl at R³ shifts the shape distribution toward the center of the plot (“goblet-like” shape, **Figure 2c**). In contrast, when all substituents are large (substituted phenyl groups), molecular shapes are widely distributed between the rod- and disc-like regions and cover a significant amount of shape space (**Figure 2d**). These results suggest that shape biasing can be achieved by manipulating the substituents about the spiro-fused pyrazolidoylisoxazoline core. Of course, shape control is an important factor in the rational design of a biological ligand.

In conclusion, synthetic entry into the novel spiro-fused pyrazolidoylisoxazolines has been achieved.¹⁵ These spirocycles (and virtual analogs thereof) were examined computationally to characterize their overall molecular shapes. The compounds reported here are currently being evaluated by collaborators at Dow AgroSciences for pesticidal activity and have been submitted to the NIH molecular libraries small molecule repository (MLSMR).

Experimental Section

I. General Procedures

Materials and Methods—All reagents and solvents were purchased from commercial vendors and used without further purification. Concentration of reaction mixtures refers to rotary evaporation under reduced pressure carried out at 40°C. Thin layer chromatography (TLC) was performed on silica-coated glass plates (silica gel 60 F₂₅₄, 0.5 mm thickness) and visualized at 254 nm. Silica gel (60 Å pores, particle size ranging from 32-63 μm) was used for all flash chromatography purification. NMR spectral data was obtained at ambient temperature unless otherwise specified. ¹H NMR spectra were recorded at 300, 400, or 600 MHz in CDCl₃. ¹³C NMR spectra were recorded at 75, 100, or 150 MHz in CDCl₃. δ values are reported and shown in parts per million (ppm) and referenced against CDCl₃ (7.26 ppm for ¹H and 77.16 ppm for ¹³C) and *J* coupling values are listed in Hz. Infrared spectral data was obtained on an FT-IR spectrometer with major peaks listed. LC/MS data was obtained for mass verification and purity analysis; the specifications of the instrument are as follows: electrospray (+) ionization, mass range of 150-1500 Da, 20 V cone voltage, MS C₁₈ column (2.1 mm × 50 mm × 3.5 μm), mobile phase consisting of water and acetonitrile with a 0.1% TFA buffer, and a flow rate of 0.2 ml/min.

II. Preparation of 2-substituted hydrazides (procedures and characterization data)

N'-(3-bromobenzyl)-4-chlorobenzohydrazide (**3**)—

To a solution of 4-chlorobenzohydrazide (1.50 g, 8.79 mmol) in *N,N*-dimethylformamide (15 mL) in a round bottom flask equipped with a stir bar was added *N,N*-diisopropylethylamine (0.919 mL, 5.28 mmol). 3-Bromobenzyl bromide (439 mg, 1.76 mmol), dissolved in *N,N*-dimethylformamide (7 mL), was added to the solution via syringe pump over 12 hours (0.6 mL/hr) at room temperature. The reaction mixture was allowed to stir until TLC indicated the disappearance of 3-bromobenzyl bromide. The resulting mixture was then diluted with saturated aqueous ammonium chloride (40 mL) and extracted with

95:5 ethyl acetate: hexanes (3x40 mL). The combined organic extracts were washed with brine (100 mL), dried over sodium sulfate, filtered, and concentrated. The crude material was purified by flash chromatography (gradient mobile phase: 2:3 EtOAc:hexanes → 1:1 EtOAc:hexanes) yielding **3**, a white solid (406 mg, 68% yield): mp 149 °C; IR (neat) $\nu_{\max}$ 3303, 3290, 3052, 2913, 2453, 2398, 1625, 1593, 1411, 1384, 1089, 1009, 869, 844 cm^{-1} ; ^1H NMR (300 MHz, CDCl_3) δ 7.63 (d, J = 8.4 Hz, 2H), 7.58 (s, 1H), 7.52 (s, 1H), 7.45 – 7.39 (m, 3H), 7.31 (d, J = 7.6 Hz, 1H), 7.22 (t, J = 7.7 Hz, 1H), 5.14 (s, 1H), 4.06 (s, 2H); ^{13}C NMR (75 MHz, CDCl_3) δ 166.8, 140.0, 138.5, 132.1, 131.1, 131.0, 130.3, 129.2, 128.4, 127.7, 122.8, 55.4; HRMS (ESI) Calcd for $\text{C}_{14}\text{H}_{12}\text{BrClN}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 338.9895; Found,

General procedure for the preparation of 2-isopropyl aryl hydrazides (8a/d/e/g): 4-bromo-*N'*-isopropylbenzohydrazide (8g)—

Prepared according to literature procedures found in Benstead, D. J.; Hulme, A. N.; McNab, H.; Wight, P. *Synlett*. **2005**, 10, 1571-74 yielding a white solid (79 mg, 66% yield): mp 141-142 °C; IR (neat) $\nu_{\max}$ 3306, 3242, 2967, 1638, 1543, 1496, 1379, 1313, 1073, 1012, 894, 842 cm^{-1} ; ^1H NMR (400 MHz, CDCl_3) δ 7.89 (s, 1H), 7.63 (d, J = 8.5 Hz, 2H), 7.57 (d, J = 8.5 Hz, 2H), 4.86 (s, 1H), 3.21 (sept, J = 6.3 Hz, 1H), 1.09 (d, J = 6.3 Hz, 6H); ^{13}C NMR (100 MHz, CDCl_3) δ 166.7, 132.1, 131.8, 128.6, 126.7, 51.5, 21.0; HRMS (ESI) Calcd for $\text{C}_{10}\text{H}_{13}\text{BrN}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 257.0284; Found, 257.0287.

***N'*-isopropylfuran-2-carbohydrazide (8e)—**

Synthesized according to the general procedure for the preparation of 2-isopropyl aryl hydrazides (**8a/d/e/g**), which yielded a white solid (2.248 g, 84% yield): mp 100-102 °C; IR (neat) $\nu_{\max}$ 3325, 3233, 2977, 1647, 1567, 1489, 1306, 940, 884 cm^{-1} ; ^1H NMR (400 MHz, CDCl_3) δ 8.12 (s, 1H), 7.41 (t, J = 0.8 Hz, 1H), 7.10 (d, J = 3.5 Hz, 1H), 6.47 (ddd, J = 3.5, 1.7 & 0.8 Hz, 1H), 4.74 (s, 1H), 3.19 (sept, J = 6.3 Hz, 1H), 1.07 (d, J = 6.3 Hz, 6H); ^{13}C NMR (100 MHz, CDCl_3) δ 158.3, 146.9, 144.2, 114.7, 112.1, 51.5, 20.8; HRMS (ESI) Calcd for $\text{C}_8\text{H}_{12}\text{N}_2\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 169.0972; Found, 169.0969.

4-(*tert*-butyl)-*N'*-isopropylbenzohydrazide (8a)—

Synthesized according to the general procedure for the preparation of 2-isopropyl aryl hydrazides (**8a/d/e/g**), which yielded a light pink solid (2.079 g, 85% yield): mp 103-104 °C; IR (neat) ν_{max} 3267, 3238, 2961, 2927, 2875, 1648, 1536, 1473, 1310, 849 cm^{-1} ; ^1H NMR (300 MHz, CDCl_3) δ 8.14 (s, 1H), 7.71 (d, $J = 8.4$ Hz, 2H), 7.43 (d, $J = 8.4$ Hz, 2H), 4.92 (s, 1H), 3.22 (sept, $J = 6.3$ Hz, 1H), 1.32 (s, 9H), 1.08 (d, $J = 6.3$ Hz, 6H); ^{13}C NMR (75 MHz, CDCl_3) δ 167.4, 155.3, 130.1, 126.9, 125.6, 51.4, 35.0, 31.2, 20.9; HRMS (ESI) Calcd for $\text{C}_{14}\text{H}_{22}\text{N}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 235.1805; Found, 235.1807.

N'-isopropylnicotinohydrazide (8d)—

Synthesized according to the general procedure for the preparation of 2-isopropyl aryl hydrazides (**8a/d/e/g**), which yielded a white crystalline material (939 mg, 59%): mp 108-110 °C; IR (neat) ν_{max} 3269, 3234, 2969, 2870, 1621, 1588, 1544, 1471, 1414, 1347, 1320 cm^{-1} ; ^1H NMR (400 MHz, CDCl_3) δ 8.97 (dd, $J = 2.2$ & 0.8 Hz, 1H), 8.78 (s, 1H), 8.68 (dd, $J = 4.9$ & 1.7 Hz, 1H), 8.10 (dt, $J = 7.9$ & 1.9 Hz, 1H), 7.36 (ddd, $J = 7.9$, 4.9 & 0.8 Hz, 1H), 4.93 (s, 1H), 3.20 (sept, $J = 6.3$ Hz, 1H), 1.07 (d, $J = 6.3$ Hz, 6H); ^{13}C NMR (100 MHz, CDCl_3) δ 165.6, 152.5, 148.0, 135.3, 129.0, 123.7, 51.5, 20.9; HRMS (ESI) Calcd for $\text{C}_9\text{H}_{13}\text{N}_3\text{O}$ ($\text{M}+\text{H}$) $^+$, 180.1132; Found, 180.1125.

General procedure for the preparation of 2-aryl acetohydrazides (13a/c):N'-(4-chlorophenyl)acetohydrazide (13a)—

Prepared according to literature procedures found in Hearn M. J.; Grinwade, J. E. *Org. Prep. Proced. Int.* **1980**, 12(3-4), 249-51. Characterization data is in agreement with published values.

***N'*-(3-chlorophenyl)acetohydrazide (13b)—**

Prepared according to the general procedure for the preparation of 2-aryl acetohydrazides (13a/c). Characterization data is in agreement with published values.

Procedure for the preparation of *N'*-methylpyrazine-2-carbohydrazide (19)—

Prepared according to literature procedures found in Gobis, K.; Foks, H.; Zwolska, Z.; Augustynowicz-Kopeć, E. *Phosphorous, Sulfur, and Silicon*. **2006**, 181, 965-75. Characterization data is in agreement with published values.

III. Preparation of pyrazolidine intermediates (procedures and characterization data)**(2-(3-bromobenzyl)-4-methylenepyrazolidin-1-yl)(4-chlorophenyl)methanone (4)—**

To a solution of 3-chloro-2-(chloromethyl)prop-1-ene (630 μ L, 5.98 mmol) in acetone (15 mL) in a round bottom flask with sidearm equipped with a stir bar and reflux condenser were added potassium carbonate (826 mg, 5.98 mmol) and sodium iodide (90 mg, 0.598 mmol). The resulting mixture was brought to reflux and *N'*-(3-bromobenzyl)-4-chlorobenzohydrazide (**3**; 406 mg, 1.20 mmol), dissolved in acetone (7 mL), was added via syringe pump over 12 h. The reaction mixture was subsequently concentrated, diluted with a saturated aqueous solution of ammonium chloride (20 mL), and extracted with ethyl acetate (3x15 mL). The organic extracts were combined and washed with brine (30 mL), dried over sodium sulfate, filtered, concentrated, and purified by flash chromatography (gradient mobile phase: 1:4 EtOAc:Hexanes $\rightarrow$ 2:3 EtOAc:Hexanes) to afford pyrazolidine **4**, a white solid (84 mg, 18% yield): mp 128-129 $^{\circ}$ C; IR (neat) ν_{max} 2919, 2902, 2850, 1609, 1451,

1432, 1417, 1090, 898, 839 cm^{-1} ; ^1H NMR (300 MHz, CDCl_3) δ 7.52 (s, 1H), 7.40 (t, J = 7.8 Hz, 3H), 7.33 (d, J = 5.6 Hz, 1H), 7.27 (t, J = 7.3 Hz, 2H), 7.17 (t, J = 7.8 Hz, 1H), 5.27 (s, 1H), 5.21 (s, 1H), 4.25 (s, 2H), 4.18 (s, 2H), 3.79 (s, 2H); ^{13}C NMR (150 MHz, CDCl_3) δ 167.8, 143.5, 138.2, 136.5, 133.6, 132.8, 131.1, 130.1, 130.0, 128.0, 127.9, 122.7, 108.4, 60.8, 58.8, 47.8; HRMS (ESI) Calcd for $\text{C}_{18}\text{H}_{16}\text{BrClN}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 391.0208; Found, 391.0212.

General Procedure for the preparation of 4-methylenepyrazolidines (9a/d/e/g & 14a/c): (4-bromophenyl)(2-isopropyl-4-methylenepyrazolidin-1-yl)methanone (9g)—

4-Bromo-*N'*-isopropylbenzohydrazide (**8g**; 375 mg, 1.46 mmol) and 3-chloro-2-(chloromethyl)prop-1-ene (0.768 mL, 7.29 mmol) were combined in a round bottom flask equipped with a stir bar and flushed with nitrogen. Dry THF (10 mL) was then added to the flask and the contents were cooled in an ice bath. Sodium hydride (350 mg, 60% dispersion in mineral oil, 8.75 mmol) was added slowly and the reaction mixture was allowed to warm to room temperature with stirring under nitrogen overnight. Water (10 mL) was cautiously added and the mixture was concentrated, diluted with a saturated aqueous solution of NH_4Cl (10 mL), and extracted with ethyl acetate (3x10 mL). The organic extracts were combined and washed with brine (20 mL), dried over sodium sulfate, filtered, concentrated, and purified by flash chromatography (1:1 EtOAc/hexane) to afford pyrazolidine **9g** as a yellow oil (388 mg, 86% yield): IR (neat) ν_{max} 2969, 2926, 2867, 1619, 1587, 1434, 1414, 1382, 1608, 880, 836 cm^{-1} ; ^1H NMR (300 MHz, CDCl_3) δ 7.68 (d, J = 8.3 Hz, 2H), 7.43 (d, J = 8.3 Hz, 2H), 5.13 (d, J = 17.9 Hz, 2H), 4.59 (d, J = 14.6 Hz, 1H), 3.93 (d, J = 14.6 Hz, 1H), 3.61 (d, J = 15.4 Hz, 2H), 2.72 (m, 1H), 0.80 (d, J = 32.0 Hz, 6H); ^{13}C NMR (75 MHz, CDCl_3) δ 167.5, 144.7, 134.1, 131.0, 130.6, 124.5, 106.8, 56.5, 55.2, 48.5, 20.7, 20.2; HRMS (ESI) Calcd for $\text{C}_{14}\text{H}_{17}\text{BrN}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 309.0597; Found, 309.0598.

Furan-2-yl(2-isopropyl-4-methylenepyrazolidin-1-yl)methanone (9e)—

Prepared according to the general procedure for the preparation of 4-methylenepyrazolidines (**9a/d/e/g** & **14a/c**), which yielded a yellow oil (828 mg, 64% yield): IR (neat) $\nu_{\max}$ 3111, 2986, 2973, 2860, 1627, 1563, 1476, 1451, 1424, 1366, 1017, 908 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.39 (s, 1H), 7.36 (d, $J = 0.7$ Hz, 1H), 6.28 (dd, $J = 3.4$ & 1.7 Hz, 1H), 5.00 (app pent, $J = 2.1$ Hz, 1H), 4.94 (app pent, $J = 2.1$ Hz, 1H), 4.54 (d, $J = 13.1$ Hz, 1H), 3.70 (d, $J = 13.1$ Hz, 1H), 3.40 (d, $J = 7.0$ Hz, 2H), 2.67 (sept, $J = 6.3$ Hz, 1H), 0.88 (d, $J = 6.3$ Hz, 6H); ^{13}C NMR (150 MHz, CDCl_3) δ 159.1, 146.2, 144.4, 144.3, 117.9, 110.9, 106.3, 56.2, 54.2, 47.7, 20.5, 19.6; HRMS (ESI) Calcd for $\text{C}_{12}\text{H}_{16}\text{N}_2\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 221.1285; Found, 221.1281.

4-(*tert*-butyl)phenyl)(2-isopropyl-4-methylenepyrazolidin-1-yl)methanone (9a**)**

(Prepared according to the general procedure for the preparation of 4-methylenepyrazolidines (**9a/d/e/g** & **14a/c**), which yielded a yellow oil (966 mg, 62% yield): IR (neat) $\nu_{\max}$ 2966, 2904, 2866, 1626, 1433, 1416, 1382, 895 cm^{-1} ; NMR spectra obtained at -30 $^{\circ}\text{C}$ displaying a mixture of rotamers (most abundant listed) ^1H NMR (600 MHz, CDCl_3) δ 7.78 (d, $J = 8.2$ Hz, 2H), 7.34 (d, $J = 8.2$ Hz, 2H), 5.21 (s, 1H), 5.15 (s, 1H), 4.66 (d, $J = 15.9$ Hz, 1H), 3.98 (d, $J = 15.9$ Hz, 1H), 3.73 (d, $J = 14.7$ Hz, 1H), 3.62 (d, $J = 14.7$ Hz, 1H), 2.74 (sept, $J = 6.1$ Hz, 1H), 1.29 (s, 9H), 0.89 (d, $J = 6.1$ Hz, 3H), 0.78 (d, $J = 6.3$ Hz, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 168.5, 153.2, 144.9, 132.0, 129.0, 124.4, 106.9, 56.6, 55.1, 48.4, 34.8, 31.2, 20.9, 20.2; HRMS (ESI) Calcd for $\text{C}_{18}\text{H}_{26}\text{N}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 287.2118; Found, 287.2114.

(2-isopropyl-4-methylenepyrazolidin-1-yl)(pyridin-3-yl)methanone (9d**)**—

Prepared according to the general procedure for the preparation of 4-methylenepyrazolidines (**9a/d/e/g** & **14a/c**), which yielded a yellow oil (688 mg, 56% yield): IR (neat) $\nu_{\max}$ 2974, 2877, 1625, 1589, 1451, 1410, 888 cm^{-1} ; ^1H NMR (400 MHz, CDCl_3) δ 8.68 (s, 1H), 8.21 (s, 1H), 7.75 (d, $J = 5.9$ Hz, 1H), 6.94 – 6.91 (m, 1H), 4.84 (s, 1H), 4.79 (s, 1H), 4.30 (d, $J = 15.3$ Hz, 1H), 3.59 (d, $J = 15.3$ Hz, 1H), 3.31 (d, $J = 23.8$ Hz, 2H), 2.38 (s, 1H), 0.45 (d, $J = 47.5$ Hz, 6H); ^{13}C NMR (100 MHz, CDCl_3) δ 165.7, 149.9, 149.5, 143.9, 136.1, 130.6, 121.8, 106.3, 56.0, 54.5, 20.1, 19.5; HRMS (ESI) Calcd for $\text{C}_{13}\text{H}_{17}\text{N}_3\text{O}$ ($\text{M}+\text{H}$) $^+$, 232.1445; Found, 232.1443.

1-(2-(3-chlorophenyl)-4-methylenepyrazolidin-1-yl)ethanone (14c**)—**

Prepared according to the general procedure for the preparation of 4-methylenepyrazolidines (**9a/d/e/g** & **14a/c**), which yielded a yellow oil (340 mg, 27% yield): IR (neat) $\nu_{\max}$ 1659, 1590, 1476, 1449, 1397, 897, 853 cm^{-1} ; ^1H NMR (300 MHz, CDCl_3) δ 7.19 (t, $J = 8.1$ Hz, 1H), 6.97 – 6.91 (m, 2H), 6.80 (ddd, $J = 8.3, 2.3$ & 0.8 Hz, 1H), 5.08 (dt, $J = 4.2$ & 2.1 Hz, 1H), 5.03 (dt, $J = 4.2$ & 2.2 Hz, 1H), 4.73 (s, 1H), 4.08 – 4.05 (m, 2H), 3.79 (s, 1H), 2.07 (s, 3H); ^{13}C NMR (75 MHz, CDCl_3) δ 173.3, 151.8, 143.3, 135.3, 130.5, 122.6, 115.8, 113.8, 107.1, 60.2, 46.9, 20.7; HRMS (ESI) Calcd for $\text{C}_{12}\text{H}_{13}\text{ClN}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 237.0789; Found, 237.0789.

1-(2-(4-chlorophenyl)-4-methylenepyrazolidin-1-yl)ethanone (14a**)—**

Prepared according to the general procedure for the preparation of 4-methylenepyrazolidines (**9a/d/e/g** & **14a/c**), which yielded a yellow oil (343 mg, 27% yield): IR (neat) $\nu_{\max}$ 1663, 1486, 1401, 826 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.22 (s, 2H), 6.86 (s, 2H), 5.08 (s, 1H), 5.03 (s, 1H), 4.73 (s, 1H), 4.04 (s, 2H), 3.79 (s, 1H), 2.07 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 173.1, 149.2, 143.3, 129.4, 127.7, 117.0, 107.1, 60.6, 46.9, 20.7; HRMS (ESI) Calcd for $\text{C}_{12}\text{H}_{13}\text{ClN}_2\text{O}$ ($\text{M}+\text{H}$) $^+$, 237.0789; Found, 237.0790.

(2-methyl-4-methylenepyrazolidin-1-yl)(pyrazin-2-yl)methanone—

Prepared according to the general procedure for the preparation of 4-methylenepyrazolidines (**9a/d/e/g** & **14a/c**), which yielded a yellow oil (81 mg, 5% yield): IR (neat) $\nu_{\max}$ 2959, 2925, 2865, 2794, 1608, 1405, 1319, 1114, 1013 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 9.08 (d, J = 1.5 Hz, 1H), 8.49 (dd, J = 2.5 & 1.5 Hz, 1H), 8.45 (d, J = 2.5 Hz, 1H), 5.05 (s, 1H),

5.00 (s, 2H), 4.95 (s, 1H), 3.86 (s, 2H), 2.89 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 147.3, 144.1, 143.8, 143.4, 143.3, 141.2, 112.1, 73.4, 62.6, 48.9; HRMS (ESI) Calcd for $\text{C}_{10}\text{H}_{12}\text{N}_4\text{O}$ ($\text{M}+\text{H}$) $^+$, 205.1084; Found, 205.1078.

IV. General procedure for the preparation of Spiro-fused Pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**)

(4-bromophenyl)(8-isopropyl-3-(2-methoxyphenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)methanone (**11g**)—

To a dichloromethane (5 mL) solution of pyrazolidine **9g** (109 mg, 0.35 mmol) under N_2 was added triethylamine (122 μL , 0.88 mmol) and the resulting solution was cooled in an ice bath. Chlorooxime **10** ($\text{R}^3 = o\text{-OMe-C}_6\text{H}_4$; 131 mg, 0.71 mmol), dissolved in dichloromethane (5 mL), was added to the solution via syringe pump over 12 h. The resulting mixture was allowed to warm to room temperature with stirring over 5 h. Water (10 mL) was added and the mixture subsequently extracted with dichloromethane (3x10 mL). The combined organic extracts were washed with saturated ammonium chloride (20 mL) and brine (20 mL), dried over sodium sulfate, filtered, concentrated by rotary evaporation, and purified by flash chromatography (EtOAc/hexane 2:3) to afford spiro-fused pyrazolidoylisoxazoline **11g** as a white solid (101 mg, 62% yield): mp 60–65 $^\circ\text{C}$; IR (neat) ν_{max} 2971, 2934, 1626, 1599, 1490, 1461, 1433, 1384, 1357, 1252, 1012, 911 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.76–7.64 (m, 3H), 7.48 (d, $J = 7.5$ Hz, 2H), 7.38 (t, $J = 7.0$ Hz, 1H), 6.97 (t, $J = 6.5$ Hz, 1H), 6.93 (d, $J = 7.9$ Hz, 1H), 4.43 (s, 1H), 3.91 (d, $J = 17.1$ Hz, 1H), 3.84 (s, 3H), 3.76 (s, 1H), 3.69 (d, $J = 12.6$ Hz, 1H), 3.58 (d, $J = 17.7$ Hz, 1H), 3.37 (s, 1H), 3.15 (d, $J = 11.9$ Hz, 1H), 0.88 (d, $J = 142.8$ Hz, 6H); ^{13}C NMR (600 MHz, CDCl_3) δ 157.5, 155.8, 134.1, 131.8, 131.0, 130.7, 129.2, 124.8, 121.0, 118.0, 111.5, 93.5, 63.1, 57.4, 55.5, 54.2, 46.5, 21.3, 20.6; HRMS (ESI) Calcd for $\text{C}_{22}\text{H}_{24}\text{BrN}_3\text{O}_3$ ($\text{M}+\text{H}$) $^+$, 458.1074; Found, 458.1069.

(8-(3-Bromobenzyl)-3-(4-chlorophenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)(4-chlorophenyl)methanone (**6**)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoaxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (77 mg, 70% yield): mp 78-85 °C; IR (neat) $\nu_{\max}$ 2446, 2414, 2394, 2355, 1644, 1595, 1092, 914, 828 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.53 (d, J = 8.6 Hz, 2H), 7.50 – 7.46 (m, 3H), 7.38 (dd, J = 8.0 & 0.7 Hz, 1H), 7.35 (d, J = 8.6 Hz, 2H), 7.30 (d, J = 8.6 Hz, 2H), 7.24 (d, J = 8.0 Hz, 1H), 7.14 (t, J = 7.8 Hz, 1H), 4.31 (d, J = 13.1 Hz, 1H), 4.24 (d, J = 13.1 Hz, 1H), 3.84 (d, J = 11.2 Hz, 1H), 3.60 (d, J = 11.2 Hz, 1H), 3.53 (d, J = 14.7 Hz, 1H), 3.50 (d, J = 14.7 Hz, 1H), 3.42 (d, J = 17.1 Hz, 1H), 3.37 (d, J = 17.1 Hz, 1H); ^{13}C NMR (150 MHz, CDCl_3) δ 166.1, 156.3, 139.4, 138.1, 136.6, 132.4, 131.9, 131.1, 130.2, 129.2, 129.0, 128.5, 128.1, 128.1, 127.6, 122.7, 89.7, 61.2, 59.0, 45.9, 41.0; HRMS (ESI) Calcd for $\text{C}_{25}\text{H}_{20}\text{BrCl}_2\text{N}_3\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 544.0189; Found, 544.0179.

(4-(Tert-butyl)phenyl)(3-(2-fluorophenyl)-8-isopropyl-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)methanone (11a)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoaxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (83 mg, 56% yield): mp 69-71 °C; IR (neat) $\nu_{\max}$ 2967, 2903, 2870, 1622, 1452, 1425, 1384, 1362, 1227, 922 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.84 (t, J = 7.0 Hz, 1H), 7.75 (s, 2H), 7.43 – 7.38 (m, 1H), 7.36 (d, J = 8.4 Hz, 2H), 7.18 (t, J = 7.6 Hz, 1H), 7.11 (dd, J = 11.0 & 8.6 Hz, 1H), 4.43 (s, 1H), 3.95 (s, 1H), 3.70 (d, J = 12.9 Hz, 2H), 3.56 (d, J = 16.8 Hz, 1H), 3.38 (s, 1H), 3.20 (d, J = 10.1 Hz, 1H), 1.32 (s, 9H), 0.98 (d, J = 95.1 Hz, 6H); ^{13}C NMR (150 MHz, CDCl_3) δ 169.3, 161.3, 159.6, 153.8, 152.9 (d, J_{CF} = 2.6 Hz), 132.4, 132.1 (d, J_{CF} = 8.7 Hz), 129.0 (m, 1C), 124.7 (m, 1C), 117.40 (d, J_{CF} = 11.5 Hz), 116.6 (d, J_{CF} = 22.2 Hz), 93.9, 62.7, 58.0, 54.5, 45.6, 34.9, 31.3, 20.9, 20.5; HRMS (ESI) Calcd for $\text{C}_{25}\text{H}_{30}\text{FN}_3\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 424.2395; Found, 424.2392.

(4-(Tert-butyl)phenyl)(8-isopropyl-3-(2-(trifluoromethyl)phenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)methanone (11b)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (104 mg, 63% yield): mp 68 °C; IR (neat) ν_{max} 2969, 2906, 2876, 1624, 1427, 1314, 1169, 1117, 1075, 1035 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.75 (d, J = 7.7 Hz, 3H), 7.61 (t, J = 7.5 Hz, 1H), 7.56 (t, J = 7.5 Hz, 1H), 7.53 (d, J = 7.3 Hz, 1H), 7.37 (d, J = 8.4 Hz, 2H), 4.46 (s, 1H), 3.99 (s, 1H), 3.74 (d, J = 12.9 Hz, 1H), 3.53 (s, 1H), 3.46-3.38 (m, 2H), 3.18 (d, J = 11.2 Hz, 1H), 1.32 (s, 9H), 1.06 (s, 3H), 0.88 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 169.3, 156.1, 153.8, 132.2, 130.7, 130.1, 129.1, 128.8 (q, J_{CF} = 31.3 Hz), 128.6, 126.8 (q, J_{CF} = 5.2 Hz), 124.6, 123.9 (q, J_{CF} = 273.8 Hz) 94.4, 62.5, 57.3, 54.6, 47.3, 34.9, 31.3, 20.9, 20.50; HRMS (ESI) Calcd for $\text{C}_{26}\text{H}_{30}\text{F}_3\text{N}_3\text{O}_2$ ($\text{M}+\text{H}$) $^+$ 474.2363; Found, 474.2352.

(4-(Tert-butyl)phenyl)(3-(2,4-dichlorophenyl)-8-isopropyl-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)methanone (11c)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (93 mg, 56% yield): mp 85 °C; IR (neat) ν_{max} 2966, 2951, 2903, 2869, 1621, 1586, 1424, 1383, 1363, 1106, 917 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.75 (s, 2H), 7.60 (d, J = 8.4 Hz, 1H), 7.44 (d, J = 1.9 Hz, 1H), 7.36 (d, J = 8.4 Hz, 2H), 7.29 (dd, J = 8.4 & 1.9 Hz, 1H), 4.41 (s, 1H), 3.94 (s, 1H), 3.71 (d, J = 12.9 Hz, 2H), 3.60 (d, J = 17.0 Hz, 1H), 3.36 (s, 1H), 3.20 (d, J = 9.4 Hz, 1H), 1.32 (s, 9H), 0.98 (d, J = 88.4 Hz, 6H); ^{13}C NMR (150 MHz, CDCl_3) δ 169.3, 155.3, 153.9, 136.8, 133.7, 132.4, 131.3, 130.6, 129.1, 127.6, 127.3, 124.6, 94.6, 62.4, 57.6, 54.6, 45.9, 34.9, 31.3, 20.9, 20.5; HRMS (ESI) Calcd for $\text{C}_{25}\text{H}_{29}\text{Cl}_2\text{N}_3\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 474.1710; Found, 474.1706.

(8-Isopropyl-3-(2-methoxyphenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)(pyridin-3-yl)methanone (11d)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (205 mg, 64% yield): mp 58-65 °C; IR (neat) ν_{max} 2971, 2929, 1635, 1589, 1462, 1434, 1408, 1248, 1028, 909, 897 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 9.02 (s, 1H), 8.59 (s, 1H), 8.07 (d, J = 6.4 Hz, 1H), 7.71 (d, J = 7.1 Hz, 1H), 7.36 (td, J = 8.4 & 1.6 Hz, 1H), 7.28 (dd, J = 7.1 & 5.1 Hz, 1H), 6.95 (t, J = 7.5 Hz, 1H), 6.91 (d, J = 8.4 Hz, 1H), 4.43 (d, J = 11.0 Hz, 1H), 3.91 – 3.86 (m, 1H), 3.82 (s, 3H), 3.77 (d, J = 18.7 Hz, 1H), 3.69 (d, J = 13.0 Hz, 1H), 3.57 (d, J = 18.1 Hz, 1H), 3.35 (s, 1H), 3.14 (d, J = 13.0 Hz, 1H), 0.84 (d, J = 145.4 Hz, 6H); ^{13}C NMR (150 MHz, CDCl_3) δ 166.7, 157.5, 155.8, 150.8, 150.1, 136.9, 131.7, 131.2, 129.2, 122.6, 120.9, 117.9, 111.4, 93.5, 63.1, 57.1, 55.5, 54.2, 46.4, 21.2, 20.6; HRMS (ESI) Calcd for $\text{C}_{21}\text{H}_{24}\text{N}_4\text{O}_3$ ($\text{M}+\text{H}$) $^+$, 381.1921; Found, 381.1929.

(3-(2-Fluorophenyl)-8-isopropyl-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl) (furan-2-yl)methanone (11e)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a yellow solid (179 mg, 68% yield): mp 53-60 °C; IR (neat) ν_{max} 2975, 2932, 1631, 1473, 1453, 1418, 1083, 1050, 1017, 917 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.78 (t, J = 7.3 Hz, 1H), 7.54 (s, 1H), 7.52 (m, 1H), 7.35 (tdd, J = 7.2, 5.2 & 1.6 Hz, 1H), 7.13 (t, J = 7.6 Hz, 1H), 7.06 (ddd, J = 11.2, 8.5 & 0.9 Hz, 1H), 6.42 (dd, J = 3.4 & 1.7 Hz, 1H), 4.51 (d, J = 12.2 Hz, 1H), 3.79 (d, J = 11.5 Hz, 1H), 3.66 (t, J = 13.3 Hz, 2H), 3.45 (d, J = 17.9 Hz, 1H), 3.39 (pent, J = 6.0 Hz, 1H), 2.99 (d, J = 13.3 Hz, 1H), 1.17 (d, J = 6.0 Hz, 3H), 1.03 (d, J = 6.0 Hz, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 160.3 (d, J_{CF} = 252.8 Hz), 159.9, 152.9, 146.4, 145.0, 132.1, 128.8,

124.6, 118.6, 117.1, 116.5 (d, $J_{\text{CF}} = 22.2$ Hz), 111.2, 94.0, 63.1, 57.0, 53.7, 45.5, 21.3, 20.2; HRMS (ESI) Calcd for $\text{C}_{19}\text{H}_{20}\text{FN}_3\text{O}_3$ ($\text{M}+\text{H}$)⁺, 358.1562; Found, 358.1563.

Furan-2-yl(8-isopropyl-3-(2-(trifluoromethyl)phenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)methanone (11f)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoaxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a yellow solid (114 mg, 66% yield): mp 130–137 °C; IR (neat) ν_{max} 2985, 2968, 2934, 2927, 1627, 1418, 1312, 1167, 1115, 1076 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.72 (d, $J = 7.7$ Hz, 1H), 7.60–7.56 (m, 2H), 7.53 (t, $J = 7.5$ Hz, 2H), 7.50 (d, $J = 7.5$ Hz, 1H), 6.44 (dd, $J = 3.4$ & 1.7 Hz, 1H), 4.54 (d, $J = 12.6$ Hz, 1H), 3.85 (d, $J = 12.6$ Hz, 1H), 3.73 (d, $J = 13.1$ Hz, 1H), 3.52 (d, $J = 17.5$ Hz, 1H), 3.42 (pent, $J = 6.0$ Hz, 1H), 3.35 (d, $J = 17.5$ Hz, 1H), 3.00 (d, $J = 13.1$ Hz, 1H), 1.20 (d, $J = 6.0$ Hz, 3H), 1.05 (d, $J = 6.0$ Hz, 3H); ^{13}C NMR (151 MHz, CDCl_3) δ 160.0, 156.1, 146.5, 145.1, 132.2, 130.6, 130.1, 128.7 (q, $J_{\text{CF}} = 31.6$ Hz), 128.4, 126.8 (q, $J_{\text{CF}} = 5.1$ Hz), 123.9 (q, $J_{\text{CF}} = 273.0$ Hz), 118.6, 111.2, 94.5, 62.9, 56.5, 53.8, 47.3, 21.3, 20.2; HRMS (ESI) Calcd for $\text{C}_{20}\text{H}_{20}\text{F}_3\text{N}_3\text{O}_3$ ($\text{M}+\text{H}$)⁺, 408.1530; Found, 408.1528.

(4-Bromophenyl)(8-isopropyl-3-(thiophen-2-yl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)methanone (11h)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoaxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a brown solid (51 mg, 76% yield): mp 71–80 °C; IR (neat) ν_{max} 2973, 2930, 1621, 1588, 1424, 1383, 1012, 911 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.70 (d, $J = 8.3$ Hz, 2H), 7.49 (d, $J = 8.3$ Hz, 2H), 7.41 (d, $J = 5.1$ Hz, 1H), 7.20 (d, $J = 3.5$ Hz, 1H), 7.07 (t, $J = 4.6$ Hz, 1H), 4.36 (s, 1H), 3.94 (s, 1H), 3.70 (d, $J = 11.0$ Hz, 1H), 3.60 (d, $J = 16.8$ Hz, 1H), 3.46 (d, $J = 16.8$ Hz, 1H), 3.33 (s, 1H), 3.19 (d, $J = 11.0$ Hz, 1H), 1.00 (s, 3H), 0.86 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ

168.2, 152.0, 134.3, 131.7, 131.0, 131.0, 128.8, 128.7, 127.6, 125.0, 93.9, 63.0, 57.5, 54.7, 44.7, 21.0, 20.6; HRMS (ESI) Calcd for $C_{19}H_{20}BrN_3O_2S$ (M+H)⁺, 434.0533; Found, 434.0532.

1-(8-(4-chlorophenyl)-3-(2-fluorophenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)ethanone (15a)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (62 mg, 29% yield): mp 146 °C; IR (neat) ν_{max} 1660, 1488, 1450, 1405, 919, 828, 818 cm^{-1} ; 1H NMR (600 MHz, $CDCl_3$) δ 7.78 (t, J = 7.2 Hz, 1H), 7.41 – 7.36 (m, 1H), 7.26 (d, J = 8.2 Hz, 2H), 7.15 (t, J = 7.6 Hz, 1H), 7.09 (dd, J = 8.8 & 8.9 Hz, 1H), 6.91 (d, J = 8.2 Hz, 2H), 4.36 (s, 1H), 4.02 (d, J = 12.2 Hz, 1H), 3.82 (s, 1H), 3.60 (s, 1H), 3.47 (s, 2H), 2.09 (s, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 173.6, 160.4 (d, J_{CF} = 252.6 Hz), 152.9, 149.1, 132.2 (d, J_{CF} = 8.8 Hz), 129.5, 128.9 (d, J_{CF} = 2.9 Hz), 127.7, 124.7 (d, J_{CF} = 3.4 Hz), 117.2 (d, J_{CF} = 11.0 Hz), 116.7, 116.5 (d, J_{CF} = 21.8 Hz), 93.4, 66.0, 54.9, 44.0, 20.6; HRMS (ESI) Calcd for $C_{19}H_{17}ClFN_3O_2$ (M+H)⁺, 374.1066; Found, 374.1069.

1-(8-(4-Chlorophenyl)-3-(2-(trifluoromethyl)phenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)ethanone (15b)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (124 mg, 65% yield): mp 157-158 °C; IR (neat) $\nu_{\max}$ 1659, 1490, 1407, 1314, 1177, 1110, 1035, 828 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.65 (d, $J = 7.5$ Hz, 1H), 7.49 (dt, $J = 23.2$ & 7.5 Hz, 2H), 7.40 (d, $J = 7.5$ Hz, 1H), 7.18 (d, $J = 8.8$ Hz, 2H), 6.84 (d, $J = 8.8$ Hz, 2H), 4.29 (s, 1H), 3.97 (d, $J = 12.0$ Hz, 1H), 3.77 (s, 1H), 3.40 (s, 1H), 3.27 (s, 2H), 2.01 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 173.5, 156.1, 149.0, 132.2, 130.6, 130.1, 129.5, 128.7 (q, $J_{\text{CF}} = 31.9$ Hz), 128.3, 127.7, 126.8 (q, $J_{\text{CF}} = 5.0$ Hz), 123.9 (q, $J_{\text{CF}} = 273.3$ Hz), 116.6, 93.7, 65.6, 54.4, 46.0, 20.5; HRMS (ESI) Calcd for $\text{C}_{20}\text{H}_{17}\text{ClF}_3\text{N}_3\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 424.1034; Found, 424.1034.

1-(8-(3-chlorophenyl)-3-(2,4-dichlorophenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)ethanone (15c)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (128 mg, 72% yield): mp 67-77 °C; IR (neat) ν_{max} 1668, 1590, 1476, 1428, 1384, 1339 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.54 (d, J = 8.2 Hz, 1H), 7.41 (s, 1H), 7.26 – 7.24 (m, 1H), 7.20 (t, J = 8.2 Hz, 1H), 6.96 (d, J = 6.7 Hz, 2H), 6.83 (d, J = 5.8 Hz, 1H), 4.38 (s, 1H), 4.05 (d, J = 11.5 Hz, 1H), 3.78 (s, 1H), 3.64 (s, 1H), 3.50 (d, J = 32.7 Hz, 2H), 2.08 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 173.8, 155.4, 151.7, 136.9, 135.5, 133.6, 131.3, 130.6, 127.7, 127.0, 122.6, 115.5, 113.4, 94.1, 65.4, 54.5, 44.4, 20.6; HRMS (ESI) Calcd for $\text{C}_{19}\text{H}_{16}\text{Cl}_2\text{N}_3\text{O}_2$ ($\text{M} + \text{H}^+$), 424.0381; Found, 424.0383.

4-(7-Acetyl-8-(3-chlorophenyl)-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-3-yl)benzonitrile (15d)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylisoxazolines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (61 mg, 41% yield): mp 89-95 °C; IR (neat) ν_{max} 3066, 2955, 2925, 2227, 1667, 1591, 1476, 1430, 1390,

1350, 915, 838 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 7.68 (d, J = 2.1 Hz, 4H), 7.23 (t, J = 7.9 Hz, 1H), 7.01 – 6.95 (m, 2H), 6.85 (d, J = 7.0 Hz, 1H), 4.41 (s, 1H), 4.07 (d, J = 12.2 Hz, 1H), 3.81 (s, 1H), 3.56 (s, 1H), 3.49 (s, 1H), 3.39 (s, 1H), 2.10 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 173.8, 155.0, 151.6, 135.6, 133.3, 132.7, 130.7, 127.2, 122.8, 118.1, 115.6, 114.2, 113.4, 94.2, 65.9, 54.9, 41.9, 20.6; HRMS (ESI) Calcd for $\text{C}_{20}\text{H}_{17}\text{ClN}_4\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 381.1113; Found, 381.1116.

(3-(2-Fluorophenyl)-8-methyl-1-oxa-2,7,8-triazaspiro[4.4]non-2-en-7-yl)(pyrazin-2-yl)methanone (20)—

Prepared according to the general procedure for the preparation of spiro-fused pyrazolidoylloxazolidines (**6**, **11a-h**, **15a-d**, & **20**), which yielded a white solid (75 mg, 55% yield); mp 56–65 °C; IR (neat) ν_{max} 2959, 2928, 1640, 1451, 1433, 1386, 1017, 917 cm^{-1} ; ^1H NMR (600 MHz, CDCl_3) δ 8.85 (s, 1H), 8.62 (d, J = 5.5 Hz, 2H), 7.84 (t, J = 7.1 Hz, 1H), 7.41 (q, J = 5.6 Hz, 1H), 7.19 (t, J = 7.4 Hz, 1H), 7.12 (t, J = 8.5 Hz, 1H), 4.17 (d, J = 12.9 Hz, 1H), 3.75 (d, J = 17.4 Hz, 1H), 3.56 (d, J = 17.4 Hz, 1H), 3.32 (d, J = 13.0 Hz, 1H), 3.13 (d, J = 13.0 Hz, 1H), 2.99 (s, 1H), 2.85 (s, 3H); ^{13}C NMR (150 MHz, CDCl_3) δ 166.6, 161.3, 159.6, 152.9, 149.8, 145.4, 143.7 (d, J_{CF} = 41.4 Hz), 132.3 (d, J_{CF} = 8.7 Hz), 128.9, 124.8 (d, J_{CF} = 3.2 Hz), 117.1 (d, J_{CF} = 11.5 Hz), 116.6 (d, J_{CF} = 22.5 Hz), 93.8, 67.4, 55.8, 45.8, 45.1; HRMS (ESI) Calcd for $\text{C}_{17}\text{H}_{16}\text{FN}_5\text{O}_2$ ($\text{M}+\text{H}$) $^+$, 342.1361; Found, 342.1361.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

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15. During review of this manuscript, the question was asked if the methods reported here for the synthesis of spiro-fused pyrazolidoylisoxazoline **1** could be adapted to a one-pot cascade format. Regrettably, 4-methylenepyrazolidine formation requires the use of excess (we employed 5 equiv; see **3** → **4** in the Experimental Section) 3-chloro-2-(chloromethyl)prop-1-ene to minimize the production of unwanted bis-hydrazide **21**. Preliminary studies also established that 3-chloro-2-(chloromethyl)prop-1-ene reacts with nitrile oxides to give isoxazoline **22**, which proved unreactive in the alkylation of *N'*-alkylacylhydrazides. Together, these two observations preclude the feasibility of developing an effective one-pot cascade route to **1**.

Figure 1.

(a) 11-deoxyfistularin-3 & 11-oxoaerothionin (active against human breast and colon carcinomas) (b) Aerophobin 1, Purealdin L, & Aplysinamisine II (serine protease inhibitors) (c) Retrosynthetic analysis of spiro-fused pyrazolidoylisoaxazolines **1**.

Scheme 1.

Method A route to spiro-fused pyrazolidoylisoxazoline 6.

Scheme 2.*Method B* route to spiro-fused pyrazolidoylisoxazolines **11a-h**.

Scheme 3.

Method C route to spiro-fused pyrazolidoylisoxazolines **15a-d**.

Scheme 4.

Method D route to spiro-fused pyrazolidoylisoxazoline **20**.

Figure 2.

Molecular shape space diversity for: (a) **Virtual library 1** (R^1 =Me, R^2 and R^3 = substituted phenyl groups); (b) **Virtual library 2** (R^2 =Me, R^1 and R^3 = substituted phenyl groups); (c) **Virtual library 3** (R^3 = Me, R^1 and R^2 = substituted phenyl groups); (d) **Virtual library 4** (R^1 , R^2 , and R^3 = substituted phenyl groups). Up to 200 conformations within 10 kcal/mol, as estimated by MMFF94,¹³ are plotted in blue while global minima for each member of a library is represented by red. Virtual library constituents and computational methods¹⁴ are defined in the Supporting Information file.