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THE SYNTHESIS OF SOME DERIVATIVES BASED ON THE 4-BENZYL-1*H*-PYRAZOLE-3,5-DIAMINE CORE

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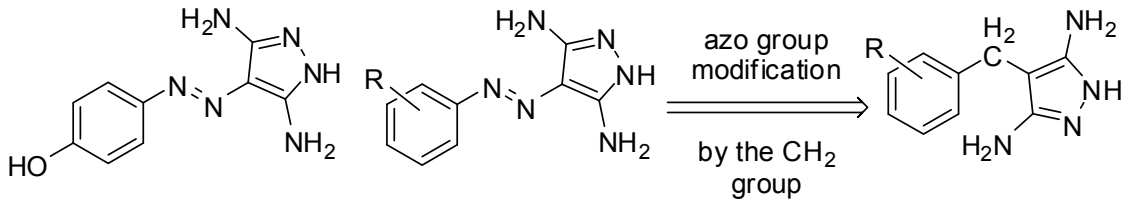
Abstract – The three-step synthesis of 4-benzyl-1*H*-pyrazole-3,5-diamines **2** from commercially available aldehydes **3** is given. The Knoevenagel condensation was utilized to assemble the initial carbon framework, resulting in the benzylidenemalononitriles **4** which were directly transformed by the reduction of the electron deficient C=C bond to benzylmalononitriles **5**. Subsequent cycloaddition of hydrazine with **5** afforded the desired pyrazoles **2**. Due to the high similarity with 4-arylazo-1*H*-pyrazole-3,5-diamines, the biological activities of the 4-benzyl-1*H*-pyrazole-3,5-diamines **2** were evaluated while focusing on the inhibition of cyclin-dependent kinases (CDKs), but no significant results were obtained.

In the last decade, protein kinases became an important class of drug targets for the pharmaceutical industry for the treatment of proliferative disorders such as cancer and inflammation.[1-4] The successful development of these molecular inhibitors resulted in the launch of several drug candidates that are currently on the market. Some examples of this class of drug that are commercially available include imatinib, dasatinib and nilotinib.[5] This selection, of course, only partially illustrates the vast amount of research completed in the area of protein kinase inhibitors.

A few years ago, we reported that the 4-arylazopyrazole-3,5-diamine derivatives **1** (Scheme 1) showed significant inhibitory activity against cyclin-dependent kinases (CDKs).[6] The most potent derivative, CAN508, displayed selectivity toward CDK9.[6] Inhibition of CDK9, which is the catalytic subunit of

positive transcription elongation factor b (P-TEFb),[7] can potentially play an important role in cardiac muscle-cell hyperthrophy,[8] HIV-1 Tat protein transactivation[9,10] or even cancers[11,12] and therefore is of potential pharmaceutical interest.

If the structures of low molecular weight protein kinase inhibitors are examined in more detail, some similar structural features can be traced.[13] The inhibitor CAN508 contains a rigid pyrazole ring and a benzene ring that are connected by an azo group moiety. Many compounds containing an azo group are considered to be non-toxic; some, such as tartrazine (E102), still are used as food additives.[14] However, in recent years, several reports suggest their possible genotoxicity and a possible role in cancer induction.[15] Therefore, we attempted to replace this pharmacologically unfavorable functionality by substituting a methylene group for the azo group, as in pyrazole **2**. Additionally, we were interested in how the methylene linker would change the inhibitory activity against CDKs. Therefore, we selected commercially available starting materials to provide benzene-substituted pyrazole products with functional groups more likely to interact with the active site of the protein kinase. The pyrazoles **2b**,[16] **2e**[17] and **2f**[18] have previously been reported but usually as intermediates in a multi-step synthesis, and the preparation of only a limited number of benzene-substituted derivatives has been described.



Scheme 1

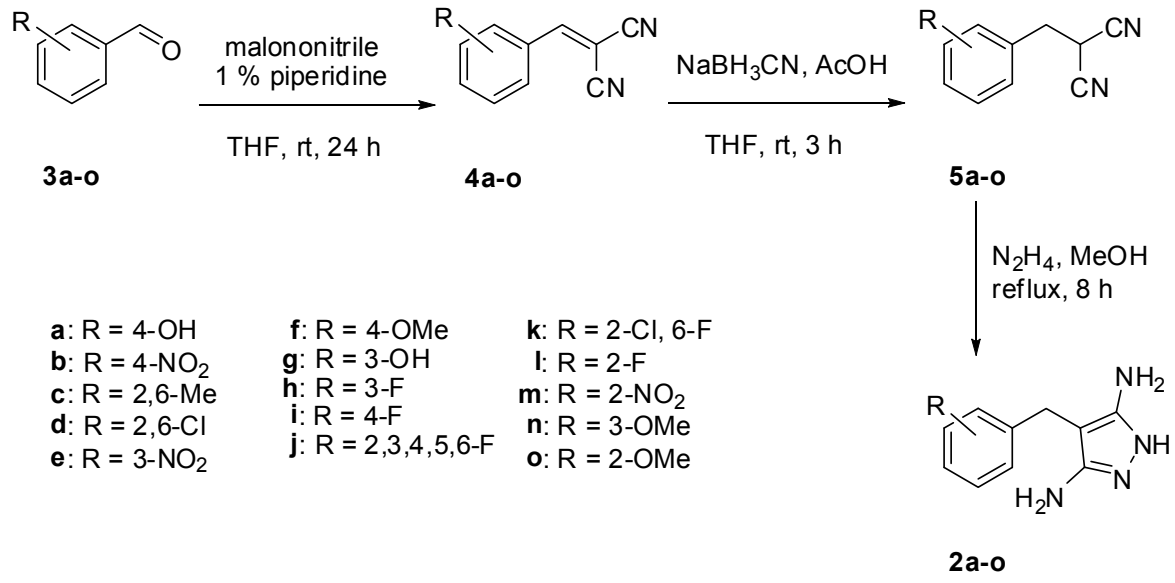
RESULTS AND DISCUSSION

The synthesis of the pyrazole derivative **2** was completed in three steps (Scheme 2). The carbon framework was assembled in the first step using a Knoevenagel condensation of an aldehyde and malononitrile. The activated C=C bond of the benzylidenemalononitrile derivative **4** was reduced to give benzylmalononitrile **5**. The alternative preparation of derivatives of **5** via the alkylation of malononitrile with a benzyl halide is not ideal for the following two reasons: the reaction usually results in the formation of bis-benzylated malononitriles, and more aldehydes are generally commercially available than benzyl halides.[19,20] Finally, the synthesis was completed by the cycloaddition of hydrazine and **5** to afford the desired pyrazoles **2**.

Our primary intention was to develop a practical method to utilize diverse substituted benzaldehyde derivatives **3** in a Knoevenagel condensation with malononitrile. To find suitable robust reaction

conditions, benzaldehydes **3a**, **3b** and **3c** were chosen for a model study. THF was used as the solvent because a wide range benzaldehyde derivatives are typically very soluble in it. Another reason to use this aprotic solvent was the possibility of performing the Knoevenagel condensation and the subsequent reductive step in a one-pot synthesis, eliminating the need to isolate the intermediate **4**. Although the Knoevenagel condensation was studied using various catalysts[21-23] and even performed in water alone without a catalyst,[24] we found the classical catalyst piperidine to be the most suitable in this case.

At first, we investigated the necessary concentration of piperidine at ambient temperature. The amount of piperidine was critical during the condensation of the most reactive aldehyde **3b** so that the product **4b** could be isolated in high purity. One mole percent of piperidine was found to be sufficient for complete condensation without any decrease in purity or yield. The reaction time was extended to 24 hours for the less reactive aldehydes **3a** and **3c**. In general, the optimized reaction conditions (THF, 1% piperidine, rt, 24 h) were found to be applicable for all selected aldehydes **3a-o,** and yields of the isolated benzylidenemalononitriles **4a-d** were higher than 80%. Except for the benzylidenemalononitriles **4a** and **4b,** which were isolated for model study purposes, only the benzylidenemalononitriles **4c** and **4d** were isolated and characterized because no data for these analogs was previously published. Benzylidenemalononitriles **4e-o** were directly reduced to benzylmalononitriles **5** without an isolation of the intermediates **4**.



Scheme 2

The second step, the reduction of a C=C bond, was optimized using purified benzylidenemalononitriles **4a** and **4b**. Although it is possible to find references describing the C=C bond reduction of some benzylidenemalononitriles,[25-28] we decided to use sodium cyanoborohydride as a new alternative reducing

agent for the electron deficient C=C bond in intermediates **4**. Some authors, when describing the reduction of heterocyclic systems with sodium cyanoborohydride, indicate that it might also be efficient in our system.[29-31] The first attempt to reduce **4a** or **4b** with sodium cyanoborohydride in THF at ambient temperature afforded only a mixture of compounds, but no desired benzylmalononitriles **5a** or **5b** were detected by HPLC/MS. When the same reaction was carried out in the presence of acetic acid, benzylmalononitriles **5a** and **5b** were the major products, and after work up, they were easily isolated by filtration. Next, we focused our effort on the preparation of benzylmalononitriles **5a** and **5b** directly from the corresponding aldehydes **3a** and **3b** without the need to isolate the intermediate benzylidenemalononitriles **4a** and **4b**. A simple one-pot procedure was developed as follows. After the first condensation step was completed, a THF solution containing sodium cyanoborohydride and acetic acid was added, and the resulting reaction mixture was continuously stirred for an additional three hours. The resulting benzylmalononitriles **5a-o** were then isolated by filtration or extraction and purified by recrystallization or chromatography. This one-pot synthetic strategy was applied for the preparation of all benzylmalononitriles **5a-o** with yields ranging between 37 and 70% over the two steps.

Several attempts to synthesize pyrazoles **2** from benzylmalononitriles **5a** and **5b** in a one-pot procedure were carried out. Because the cycloaddition reaction with hydrazine hydrate in boiling THF failed, it was obvious that the preceding isolation of benzylmalononitrile **5** is necessary. The cyclization proceeds in EtOH but was troublesome due to the formation of greasy products that were difficult to isolate when using the reported procedure for benzene-unsubstituted derivatives.[32] Despite our difficulties using EtOH in the final step, MeOH was found to be a suitable solvent. Utilization of MeOH enabled to simplify the isolation of cycloaddition reaction products. The pyrazoles **2a-o**, prepared from corresponding benzylmalononitriles **5a-o** with hydrazine hydrate in boiling MeOH, were obtained in yields between 29 and 88%. Isolation of the pyrazoles **2a-l** was simple. After evaporation of MeOH, the residue was triturated with DCM or recrystallized from a mixture of MeOH and ethyl acetate. The attempts to isolate pure pyrazoles **2m-o** as a free base or a hydrochloride salt failed, although they were identified as part of the reaction mixture by HPLC-MS.

In the ^{13}C NMR spectra of the isolated compounds, we observed an unusual overlap of carbon signals for pyrazole **2d**. The HSQC NMR spectrum revealed that carbon signals representing the meta and para position were overlapped, so the ^{13}C NMR spectrum showed only 6 singlets instead of the expected 7 singlets. In addition to this result, another complication can occur when measuring the ^{13}C NMR spectrum of pyrazole **2**. The pyrazole carbon signal around 149 ppm, common to all the pyrazoles we isolated, was difficult to observe in some derivatives. The signal is broadened probably due to tautomerism of the pyrazole ring; hence, the number of scans should be increased sufficiently.

Previous biochemical assays showed the ability of 4-arylazo-1*H*-pyrazole-3,5-diamines **1** to inhibit the

enzymatic activity of cyclin-dependent kinases (CDKs), which are critical regulators of the cell cycle and some other cellular processes.[6] Since the prepared pyrazoles **2a-l** contain structural moieties similar to derivatives **1** we decided to test them for CDK inhibition and antiproliferative activity. However, none of the prepared compounds inhibited the kinase CDK2/cyclin E, probably due to differences in conformation between **1** and **2**. Arylazo derivatives **1** are able to set almost planar conformation which was proven by co-crystallization of CAN508 with CDK2 or with CDK9.[6,33] In contrast, our benzyl analogues probably adopt a less planar conformation due to the sp^3 configuration of the carbon in the methylene group. Such molecules cannot enter the active site of CDKs and thus will not inhibit their catalytic activity.

Despite this result that 4-benzyl-1*H*-pyrazole-3,5-diamines **2a-l** are not CDK inhibitors, we tested their anticancer cytotoxicity under *in vitro* conditions in two cancer cell lines, K-562 (myeloid leukemia) and MCF-7 (breast adenocarcinoma). The rationale for performing the cytotoxicity assay was based on a hypothesis that the cytotoxicity of 4-arylazo-1*H*-pyrazole-3,5-diamines **1** found in our previous work can be mediated by interaction(s) with other cellular targets besides CDKs.[6] However, the assays showed that all prepared compounds were not cytotoxic against the two cell lines. These results confirm that cytotoxicity of 4-arylazo-1*H*-pyrazole-3,5-diamines **1** is at least partially a consequence of CDK inhibition in cells.

A novel alternative one-pot procedure to afford benzylmalononitriles **5** from commercially available benzaldehydes **3** was developed. Sodium cyanoborohydride turned out to be an efficient reagent to reduce the electron deficient C=C bond in the benzylidenemalononitriles **4**. The choice of solvent is critical for the final step. The cycloaddition of hydrazine with the benzylmalononitriles **5** to give the desired benzylpyrazoles **2** works best in MeOH. All three steps were evaluated for various derivatives. None of the prepared pyrazoles **2a-l** showed antiproliferative activity against the cell lines K562 or MCF7.

EXPERIMENTAL

All starting materials used are commercially available. Melting points were determined on a Boetius stage. The LC/MS analyses were carried out on an UHPLC-MS system consisting of an Accela UHPLC chromatograph with a photodiode array detector and a TSQ Quantum Access triple quadrupole mass spectrometer (both Thermo Scientific, CA, USA), using a Nucleodur Gravity C18 column (Macherey-Nagel, 1.8 µm, 2.1 x 50 mm, Germany) at 30 °C and a flow rate of 800 µL/min. The APCI source operated at a discharge current of 5 µA, vaporizer temperature of 400 °C and capillary temperature of 200 °C. The ^{1}H and ^{13}C NMR spectra were measured in DMSO-d_6 at 20 °C on a Bruker Avance 300 FT NMR spectrometer. Elemental analyses were performed using an EA 1108 Elemental Analyzer (Fison Instruments).

Typical procedure for the synthesis of 2-benzylidenemalononitriles (4)

The corresponding aldehyde **3** (5 mmol) and piperidine (0.05 mmol) were added to a solution of malononitrile (5 mmol) in THF (5 mL). The reaction mixture was stirred for 24 h at room temperature and then was diluted with water (25 mL). The precipitate was collected by filtration, washed with water and recrystallized from EtOH or a mixture of EtOH and water.

2-(4-Hydroxybenzylidene)malononitrile (4a)

Prepared from 4-hydroxybenzaldehyde. 0.80 g (94%), mp 188–189 °C (EtOH/H_2O). MS (-APCI, *m/z*): 169.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 6.96 (d, J = 8.6 Hz, 2 H), 7.88 (d, J = 8.6 Hz, 2 H), 8.27 (s, 1 H), 11.05 (bs, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 75.1, 114.2, 115.1, 116.6, 122.8, 133.9, 160.5, 163.9. Anal. Calcd for $C_{10}H_6N_2O$ (170.17): C, 70.58; H, 3.55; N, 16.46. Found: C, 70.42; H, 3.61; N, 16.26.

2-(4-Nitrobenzylidene)malononitrile (4b)

Prepared from 4-nitrobenzaldehyde. 0.96 g (96%), mp 159-160 °C (EtOH). MS (+APCI, m/z): 198.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 8.13 (dd, J = 8.8, 1.8 Hz, 2 H), 8.42 (dd, J = 8.8, 1.8 Hz, 2 H), 8.72 (s, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 85.9, 112.5, 113.6, 124.4, 131.5, 136.7, 149.7, 159.3. Anal. Calcd for $C_{10}H_5N_3O_2$ (199.17); C, 60.31; H, 2.53; N, 21.10. Found: C, 60.45; H, 2.60; N, 21.18.

2-(2,6-Dimethylbenzylidene)malononitrile (4c)

Prepared from 2,6-dimethylbenzaldehyde. 0.89 g (98%), mp 78-80 °C (EtOH). MS (-APCI, *m/z*): 181.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 2.27 (s, 6 H), 7.18 (d, J = 7.5 Hz, 2 H), 7.31 (t, J = 7.5 Hz, 1 H), 8.85 (s, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 19.6, 90.4, 112.1, 112.9, 128.0, 130.4, 131.1, 135.8, 165.7. Anal. Calcd for $C_{12}H_{10}N_2$ (182.22); C, 79.10; H, 5.53, N, 15.37. Found: C, 79.22; H, 5.39; N, 15.56.

2-(2,6-Dichlorbenzylidene)malononitrile (4d)

Prepared from 2,6-dichlorbenzaldehyde. 0.89 g (80%), mp 73-74 °C (EtOH). MS (-APCI, *m/z*): 221.9, 223.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 7.57-7.73 (m, 3H), 8.85 (s, 1H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 93.1, 111.1, 112.1, 129.0, 129.2, 132.7, 133.5, 159.6. Anal. Calcd for $C_{10}H_4Cl_2N_2$ (223.06); C, 53.85; H, 1.81; N, 12.56. Found: C, 53.96; H, 1.99; N, 12.32.

Typical procedure for the synthesis of benzylmalononitriles (5)

The corresponding aldehyde **3** (5 mmol) and piperidine (0.05 mmol) were added to a solution of malononitrile (5 mmol) in THF (5 mL). The reaction mixture was stirred for 24 hours at room temperature, then a solution of acetic acid (0.6 mL) and sodium cyanoborohydride (5 mmol) in THF (15 mL) was added. After three hours of stirring at ambient temperature, THF was evaporated under reduced pressure and water (30 mL) was added to the residue. The precipitated solid was collected by filtration

and recrystallized from MeOH or a mixture of MeOH and water (method A). If a precipitate was not formed, the crude product was extracted with DCM (20 mL) and purified by the column chromatography (method B).

2-(4-Hydroxybenzyl)malononitrile (5a)

Prepared from 4-hydroxybenzaldehyde (method A). 0.54 g (63%), mp 173-176 °C (MeOH/H_2O). MS (-APCI, *m/z*): 171.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.19 (d, J = 6.7 Hz, 2 H), 4.99 (t, J = 6.8 Hz, 1 H), 6.75 (d, J = 8.4 Hz, 2 H), 7.16 (d, J = 8.6 Hz, 2 H), 9.49 (s, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.6, 34.1, 114.2, 115.4, 124.8, 130.5, 157.1. Anal. Calcd for $C_{10}H_8N_2O$ (172.18); C, 69.76; H, 4.68; N, 16.27. Found: C, 69.45; H, 4.47; N, 16.01.

2-(4-Nitrobenzyl)malononitrile (5b)

Prepared from 4-nitrobenzaldehyde (method A). 0.51 g (51%), mp 153-154 °C (MeOH). MS (-APCI, *m/*z): 199.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.54 (d, J = 7.1 Hz, 2 H), 5.22 (t, J = 7.0 Hz, 1 H), 7.67 (d, J = 8.6 Hz, 2 H), 8.27 (d, J = 8.7 Hz, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 23.8, 33.9, 113.8, 123.8, 130.9, 142.5, 147.3. Anal. Calcd for $C_{10}H_7N_3O_2$ (201.18); C, 59.70; H, 3.51; N, 20.89. Found: C, 59.96; H, 3.23; N, 20.57.

2-(2,6-Dimethylbenzyl)malononitrile (5c)

Prepared from 2,6-dimethylbenzaldehyde (method B). 0.45 g (49%), mp 71-73 °C. MS (-APCI, *m/z*): 183.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 2.37 (s, 6 H), 3.44 (d, J = 8.2 Hz, 2 H), 4.99 (t, J = 8.2 Hz, 1 H), 7.04-7.15 (m, 3 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 19.6, 21.8, 28.9, 114.2, 127.8, 128.5, 131.3, 137.4. Anal. Calcd for $C_{12}H_{12}N_2$ (184.24); C, 78.23; H, 6.57; N, 15.21. Found: C, 78.27; H, 6.61; N, 15.43.

2-(2,6-Dichlorobenzyl)malononitrile (5d)

Prepared from 2,6-dichlorobenzaldehyde (method A). 0.79 g (70%), mp 84-86 °C (MeOH/H_2O). MS (-APCI, *m/*z): 222.8. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.68 (d, J = 7.9 Hz, 2 H), 5.17 (t, J = 8.0 Hz, 1 H), 7.43 (t, J = 7.7 Hz, 1 H), 7.57 (d, J = 7.7 Hz, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 21.2, 30.6, 113.4, 129.0, 130.1, 131.1, 135.4. Anal. Calcd for $C_{10}H_6Cl_2N_2$ (225.07); C, 53.36; H, 2.69; N, 12.45. Found: C, 53.42; H, 2.78; N, 12.14.

2-(3-Nitrobenzyl)malononitrile (5e)

Prepared from 3-nitrobenzaldehyde (method A). 0.54 g (54%), mp 126-128 °C (MeOH). MS (-APCI, *m/*z): 199.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.56 (d, J = 7.3 Hz, 2 H), 5.22 (t, J = 7.2 Hz, 1 H), 7.72 (t, J = 8.1 Hz, 1 H), 7.86 (d, J = 7.7 Hz, 1 H), 8.22 (dd, J = 8.2, 1.3 Hz, 1 H), 8.34 (s, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.0, 33.7, 113.8, 122.9, 124.2, 130.2, 136.3, 136.9, 147.8. Anal. Calcd for $C_{10}H_7N_3O_2$ (201.18); C, 59.70; H, 3.51; N, 20.89. Found: C, 59.76; H, 3.38; N, 20.65.

2-(4-Methoxybenzyl)malononitrile (5f)

Prepared from 4-methoxybenzaldehyde (method B). 0.34g (40%), mp 80-81 °C. MS (-APCI, *m/*z): 185.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.26 (d, J = 6.7 Hz, 2 H), 3.75 (s, 3 H), 5.03 (t, J = 6.8 Hz, 1 H), 6.95 (d, J = 8.6 Hz, 2 H), 7.29 (d, J = 8.6 Hz, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.5, 33.9, 55.0, 114.0, 114.1, 126.5, 130.5, 158.9. Anal. Calcd for $C_{11}H_{10}N_2O$ (186.21); C, 70.95; H, 5.41; N, 15.04. Found: C, 70.88; H, 5.63; N, 14.99.

2-(3-Hydroxybenzyl)malononitrile (5g)

Prepared from 3-hydroxybenzaldehyde (method B). 0.52 g (60%), mp 77-78 °C. MS (-APCI, *m/z*): 170.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.23 (d, J = 6.7 Hz, 2 H), 5.05 (t, J = 6.8 Hz, 1 H), 6.63 - 6.86 (m, 3 H), 7.05-7.26 (m, 1 H), 9.54 (s, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.3, 34.7, 114.2, 115.0, 116.2, 119.9, 129.7, 136.0, 157.5. Anal. Calcd for $C_{10}H_8N_2O$ (172.18); C, 69.76; H, 4.68; N, 16.27. Found: C, 69.89; H, 4.47; N, 16.11.

2-(3-Fluorobenzyl)malononitrile (5h)

Prepared from 3-fluorobenzaldehyde (method B). 0.42 g (48%), mp < 65 °C. MS (-APCI, *m/*z): 172.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.38 (d, J = 6.9 Hz, 2 H), 5.14 (t, J = 7.1 Hz, 1 H), 7.09 - 7.31 (m, 3 H), 7.39-7.50 (m, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.0, 34.0, 113.9, 114.6, 114.9, 116.0, 116.3, 125.50, 125.53, 130.6, 130.7, 137.4, 137.5, 160.4, 163.6. Anal. Calcd for $C_{10}H_7FN_2$ (174.17); C, 68.96; H, 4.05; N, 16.08. Found: C, 68.81; H, 4.22; N, 16.35.

2-(4-Fluorobenzyl)malononitrile (5i)

Prepared from 4-fluorobenzaldehyde (method A). 0.55 g (64%), mp 107-110 °C (MeOH/H_2O). MS (-APCI, *m/*z): 172.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.34 (d, J = 6.8 Hz, 2H), 5.08 (t, J = 6.9 Hz, 1 H), 7.17–7.28 (m, 2 H), 7.37–7.48 (m, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.4, 33.7, 114.0, 115.3, 115.6, 130.91, 130.93, 131.4, 131.5, 160.2, 163.5. Anal. Calcd for $C_{10}H_7FN_2$ (174.17); C, 68.96; H, 4.05; N, 16.08. Found: 68.72; H, 4.17; N, 15.84.

2-(2,3,4,5,6-Pentafluorobenzyl)malononitrile (5j)

Prepared from 2,3,4,5,6-pentafluorobenzaldehyde (method A). 0.73 g (60%), mp 95-98 °C (MeOH/H_2O). MS (-APCI, *m/*z): 244.8. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.58 (d, J = 6.2 Hz, 2 H), 5.19 (t, J = 6.5 Hz, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 22.5, 22.7, 108.4 (t), 113.3, 135.8 (m), 138.3 (m), 139.3 (m), 141.8 (m), 143.9 (m), 146.3 (m). Anal. Calcd for $C_{10}H_3F_5N_2$ (246.14); C, 48.80; H, 1.23; N, 11.38. Found: C, 48.95; H, 1.36; N, 11.13.

2-(2-Chloro-6-fluorobenzyl)malononitrile (5k)

Prepared from 2-chloro-6-fluorobenzaldehyde (method A). 0.75 g (72%), mp 119-120 °C (MeOH). MS (-APCI, *m/z*): 206.9, 208.9. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.55 (dd, J = 7.3, 1.4 Hz, 2 H), 5.15 (t, J = 7.4 Hz, 1 H), 7.24-7.38 (m, 1 H), 7.39 - 7.53 (m, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 21.9, 26.37, 26.4, 113.5, 114.7, 115.0, 120.1, 120.4, 125.8, 125.9, 131.2, 131.3, 134.8, 134.9, 159.5, 162.8.

Anal. Calcd for $C_{10}H_6ClFN_2$ (208.62); C, 57.57; H, 2.90; N, 13.43. Found: C, 57.45; H, 3.01; N, 13.22.

2-(2-Fluorobenzyl)malononitrile (5l)

Prepared from 2-fluorobenzaldehyde (method B). 0.60 g (70%), mp 68-70 °C. MS (-APCI, *m/z*): 173.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.41 (d, J = 6.7 Hz, 2 H), 5.14 (t, J = 6.9 Hz, 1 H), 7.16 - 7.34 (m, 2 H), 7.36-7.55 (m, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 23.4, 28.02, 28.05, 113.8, 115.5, 115.7, 121.4, 121.6, 124.7, 124.8, 130.4, 130.5, 131.80, 131.84, 159.0, 162.2. Anal. Calcd for $C_{10}H_7FN_2$ (174.17); C, 68.96; H, 4.05; N, 16.08. Found: C, 68.84; H, 4.23; N, 16.20.

2-(2-Nitrobenzyl)malononitrile (5m)

Prepared from 2-nitrobenzaldehyde (method B). 0.97 g (97%), mp 85-86 °C. MS (-APCI, *m/z*): 200.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.73 (d, J = 7.5 Hz, 2 H), 5.23 (t, J = 7.4 Hz, 1 H), 7.57 - 7.74 (m, 2 H), 7.77-7.91 (m, 1 H), 8.16 (d, J = 8.2 Hz, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 23.3, 32.1, 113.8, 125.5, 129.5, 129.9, 133.2, 134.3, 148.5. Anal. Calcd for $C_{10}H_7N_3O_2$ (201.18); C, 59.70; H, 3.51; N, 20.89. Found: C, 59.57; H, 3.34; N, 20.61.

2-(3-Methoxybenzyl)malononitrile (5n)

Prepared from 3-methoxybenzaldehyde (method B). 0.35 g (37%), mp < 65 °C. MS (-APCI, *m/z*): 185.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.31 (d, J = 6.9 Hz, 2 H), 3.76 (s, 3 H), 5.11 (t, J = 7.2 Hz, 1 H), 6.85-7.01 (m, 3 H), 7.30 (t, J = 7.9 Hz, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.1, 34.5, 55.0, 113.2, 114.0, 115.0, 121.4, 129.7, 136.1, 159.3. Anal. Calcd for $C_{11}H_{10}N_2O$ (186.21); C, 70.95; H, 5.41; N, 15.04. Found: C, 70.69; H, 5.28; N, 14.86.

2-(2-Methoxybenzyl)malononitrile (5o)

Prepared from 2-methoxybenzaldehyde (method B). 0.53 g (57%), mp < 65 °C. MS (-APCI, *m/z*): 185.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.30 (d, J = 6.9 Hz, 2 H), 3.81 (s, 3 H), 5.02 (t, J = 7.2 Hz, 1 H), 6.96 (td, J = 7.4, 0.9 Hz, 1 H), 7.05 (d, J = 8.0 Hz, 1 H), 7.24-7.41 (m, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 22.7, 29.9, 55.4, 111.0, 114.1, 120.4, 122.2, 129.6, 130.8, 157.3. Anal. Calcd for $C_{11}H_{10}N_2O$ (186.21); C, 70.95; H, 5.41; N, 15.04. Found: C, 70.76; H, 5.21; N, 15.28.

Typical procedure for the synthesis of 4-aryl(methyl)-1*H*-pyrazole-3,5-diamines (2)

Hydrazine monohydrate (2.75 mmol) was added to a solution of the benzylmalononitrile **5** (2.5 mmol) in MeOH (17 mL). The reaction mixture was heated at 65°C for 8 hours. After the reaction was complete (TLC), the solvent was removed under reduced pressure, and the residue was suspended in DCM (20 mL). The precipitate was collected by filtration and recrystallized from a mixture of MeOH and ethyl acetate.

4-[(3,5-Diamino-1*H*-pyrazol-4-yl)methyl]phenol (2a)

Prepared from 2-(4-hydroxybenzyl)malononitrile **5a**. 0.31 g (60%), mp 203-205 °C. MS (+APCI, *m/z*): 205.8. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.37 (s, 2 H), 4.21 (NH), 6.61 (d, J = 8.2 Hz, 2 H), 7.00 (d,

J = 8.2 Hz, 2 H), 9.51 (OH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 26.1, 87.4, 114.7, 129.0, 132.4, 149.2, 155.0. Anal. Calcd for $C_{10}H_{12}N_4O$ (204.23); C, 58.81; H, 5.92; N, 27.43. Found: C, 58.56; H, 5.77; N, 27.09.

4-(4-Nitrobenzyl)-1*H*-pyrazole-3,5-diamine (2b)

Prepared from 2-(4-nitrobenzyl)malononitrilu **5b**. 0.28 g (47%), mp 155-156 °C. MS (+APCI, *m/z*): 234.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.65 (s, 2 H), 4.43 (bs, NH), 7.48 (d, *J* = 8.7 Hz, 2 H), 8.11 (d, *J* = 8.7 Hz, 2 H), 10.05 (bs, NH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 26.9, 85.4, 123.2, 129.3, 145.5, 149.5, 151.3. Anal. Calcd for $C_{10}H_{11}N_5O$ (233.23); C, 51.50; H, 4.75; N, 30.03. Found: C, 51.64; H, 4.33; N, 29.89.

4-(2,6-Dimethylbenzyl)-1*H*-pyrazole-3,5-diamine (2c)

Prepared from 2-(2,6-dimethylbenzyl)malononitrile **5c**. 0.15 g (29%), mp 170-173 °C. MS (+APCI, *m/z*): 217.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 2.25 (s, 6 H), 3.48 (s, 2 H), 3.82 (bs, NH), 6.98–7.03 (m, 3 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 19.8, 22.7, 83.5, 125.8, 128.0, 136.47, 136.52, 148.8. Anal. Calcd for $C_{12}H_{16}N_4$ (216.28); C, 66.64; H, 7.46; N, 25.90. Found: C, 66.47; H, 7.35; N, 26.12.

4-(2,6-Dichlorobenzyl)-1*H*-pyrazole-3,5-diamine (2d)

Prepared from 2-(2,6-dichlorobenzyl)malononitrile **5d**. 0.38 g (59%), mp 182-190 °C. MS (+APCI, *m/*z): 257.1; 259.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.74 (s, 2 H), 4.26 (bs, NH), 7.25 (t, *J* = 8.4 Hz, 1 H), 7.44 (d, *J* = 8.1 Hz, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 24.2, 83.4, 128.6, 134.9, 135.8, 149.0. Anal. Calcd for $C_{10}H_{10}Cl_2N_4$ (257.12); C, 46.71; H, 3.92; N, 21.79. Found: C, 46.56; H, 3.73.; N, 22.04.

4-(3-Nitrobenzyl)-1*H*-pyrazole-3,5-diamine (2e)

Prepared from 2-(3-nitrobenzyl)malononitrile **5e**. 0.42 g (72%), mp 147-149 °C. MS (+APCI, *m/*z): 234.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.67 (s, 2 H), 4.71 (bs, NH), 7.52 (t, *J* = 7.7 Hz, 1 H), 7.71 (d, *J* = 7.7 Hz, 1 H), 8.00 (d, *J* = 7.7 Hz, 1 H), 8.09 (s, 1 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 26.5, 85.8, 120.5, 122.7, 129.4, 135.3, 145.3, 147.7, 149.7. Anal. Calcd for $C_{10}H_{11}N_5O$ (233.23); C, 51.50; H, 4.75; N, 30.03. Found: C, 51.73; H, 4.86; N, 29.85.

4-(4-Methoxybenzyl)-1*H*-pyrazole-3,5-diamine (2f)

Prepared from 2-(4-methoxybenzyl)malononitrile **5f**. 0.48 g (88%), mp 143-145 °C. MS (+APCI, m/z): 219.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.43 (s, 2 H); 3.69 (s, 3 H); 4.22 (bs, NH); 6.79 (d, *J* = 8.6 Hz, 2 H); 7.13 (d, *J* = 8.6 Hz, 2 H), 9.98 (bs, NH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 26.1, 54.9, 87.2, 113.3, 129.0, 134.3, 149.1, 157.1. Anal. Calcd for $C_{11}H_{14}N_4O$ (218.26); C, 60.53; H, 6.47; N, 25.67. Found: C, 60.61; H, 6.58; N, 25.34.

3-[(3,5-Diamino-1*H*-pyrazol-4-yl)methyl]phenol (2g)

Prepared from 2-(3-hydroxybenzyl)malononitrile **5g**. 0.34 g (68%), mp 217-219 °C. MS (+APCI, *m/*z): 205.2. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.44 (s, 2 H), 4.33 (bs, NH), 6.52 (dd, *J* = 7.8, 1.7 Hz, 1 H),

6.59 - 6.70 (m, 2 H), 7.01 (t, *J* = 7.8 Hz, 1 H), 9.56 (bs, OH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 27.0, 86.7, 112.4, 115.2, 119.0, 128.8, 143.8, 149.5, 157.3. Anal. Calcd for $C_{10}H_{12}N_4O$ (204.23); C, 58.81; H, 5.92; N, 27.43. Found: C, 58.87; H, 5.84; N, 27.36.

4-(3-Fluorobenzyl)-1*H*-pyrazole-3,5-diamine (2h)

Prepared from 2-(3-fluorobenzyl)malononitrile **5h**. 0.23 g (44%), mp 123-124 °C. MS (+APCI, *m/*z): 207.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.52 (s, 2 H), 4.37 (bs, NH), 6.92 (td, *J* = 8.6, 2.3 Hz, 1 H), 7.01 (d, *J* = 10.6 Hz, 1 H), 7.08 (d, *J* = 7.6 Hz, 1 H), 7.19-7.30 (m, 1 H), 10.01 (bs, NH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 26.6, 86.3, 111.8, 112.1, 114.6, 114.9, 124.28, 124.31, 129.6, 129.7, 145.8, 145.9, 149.5 (bs), 160.6, 163.8. Anal. Calcd for $C_{10}H_{11}FN_4$ (206.22); C, 58.24; H, 5.38; N, 27.17. Found: C, 58.16; H, 5.22; N, 27.36.

4-(4-Fluorobenzyl)-1*H*-pyrazole-3,5-diamine (2i)

Prepared from 2-(4-fluorobenzyl)malononitrile **5i**. 0.26 g (52%), mp 150-152 °C. MS (+APCI, *m/*z): 207.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.49 (s, 2 H), 4.36 (bs, NH), 7.04 (t, *J* = 8.9 Hz, 2 H), 7.16–7.33 (m, 2 H), 9.96 (bs, NH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 26.1, 86.8, 114.4, 114.6, 129.8, 129.9, 138.70, 138.73, 149.4, 158.8, 162.0. Anal. Calcd for $C_{10}H_{11}FN_4$ (206.22); C, 58.24; H, 5.38; N, 27.17. Found: C, 58.25; H, 5.57; N, 27.30.

4-(2,3,4,5,6-Pentafluorobenzyl)-1*H*-pyrazole-3,5-diamine (2j)

Prepared from 2-(2,3,4,5,6-pentafluorobenzyl)malononitrile **5j**. 0.23 g (33%), mp 200-202 °C. MS (+APCI, *m/*z): 279.1. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.60 (s, 2 H), 4.24 (bs, NH), 10.10 (bs, NH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 14.6, 83.7, 115.1, (t), 135.6 (m), 137.4 (m), 138.0 (m), 139.9 (m), 143.3 (m), 145.7 (m), 149.2 (bs). Anal. Calcd for $C_{10}H_7F_5N_4$ (278.18); C, 43.18; H, 2.54; N, 20.14. Found: C, 43.25; H, 2.61; N, 20.06.

4-(2-Chloro-6-fluorobenzyl)-1*H*-pyrazole-3,5-diamine (2k)

Prepared from 2-(2-chloro-6-fluorobenzyl)malononitrile **5k**. 0.39 g (66%), mp 161-168 °C. MS (+APCI, *m/z*): 241.0, 243.0. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.61 (d, *J* = 2.0 Hz, 2 H), 4.42 (bs, NH), 7.08-7.23 (m, 1 H), 7.24 - 7.39 (m, 2 H). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 18.88, 18.92, 84.2, 114.2, 114.5, 125.4, 125.5, 126.4, 126.6, 128.3, 128.4, 133.95, 134.04, 149.0, 159.4, 162.6. Anal. Calcd for $C_{10}H_{10}ClFN_4$ (240.67); C, 49.91; H, 4.19; N, 23.28. Found: C, 49.85; H, 3.96; N, 23.39.

4-(2-Fluorobenzyl)-1*H*-pyrazole-3,5-diamine (2l)

Prepared from 2-(2-fluorobenzyl)malononitrile **5l**. 0.34 g (67%), mp 150-151 °C. MS (+APCI, *m/z*): 207.2. ^{1}H NMR (300 MHz, DMSO-d_6) δ ppm 3.55 (s, 2 H), 4.33 (bs, NH), 7.00-7.25 (m, 4 H), 9.98 (bs, NH). ^{13}C NMR (75 MHz, DMSO-d_6) δ ppm 19.77, 19.83, 84.2, 114.5, 114.8, 123.98, 124.02, 127.2, 127.3, 128.5, 128.7, 130.2, 130.3, 149.6, 158.9, 162.0. Anal. Calcd for $C_{10}H_{11}FN_4$ (206.22); C, 58.24; H, 5.38; N, 27.17. Found: C, 58.18; H, 5.33; N, 26.90.

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