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Synthesis of Some New Fused Pyridines and Prediction their Biological Activity via PASS INET.

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Abstract: A new series of fused heterocyclic compounds containing pyridine moieties were prepared via the reaction of 2-amino-6-(methylthio)-4-phenylpyridine-3,5-dicarbonitrile with some halo reagents, active methylenes, acetic anhydride, phenylisothiocyanate, hydrazine hydrate or thioacetamide. Accompanied with Predictions of the activity spectra of some selected compounds using PASS INET.at Pa > 70%, showing high probability of Atherosclerosis treatment, Antineoplastic, DNA intercalator, Protein kinase inhibitor and Signal transduction pathways inhibitor.

Keywords: phase transfer catalysis; pyridine; pyrrole; pyrimidine; pyrrazole; fused pyridine; PASS INET.

1 Introduction

The considerable biological and medicinal activities of polyfunctionally substitutedand condensed pyridines[1-7] have stimulated considerable recent research aimedat developing syntheses of these compounds. Pyrroles, pyrimidines and pyrazoles are well known examples of hetero-organic compounds associated with diverse biological and pharmacological properties. Pyrrole derivatives were reported as having important synthetic and biological activities[8,9] such as COX-1/COX-2 inhibitors[10] and cytotoxic activity against a variety of marine and human tumour models[11] .Pyrimidines are reported to have a broad spectrum of biological activities. Some are endowed with antitumor[12], antiviral[13], anti-inflammatory[14], antipyretic[15], antimicrobial[16], and antifungal properties[17]. Pyrazole derivatives are synthetic targets of utmost importance in the pharmaceutical industry, since the pyrazole ring has been known as an important frame-work in a large number of drugs[18-21].

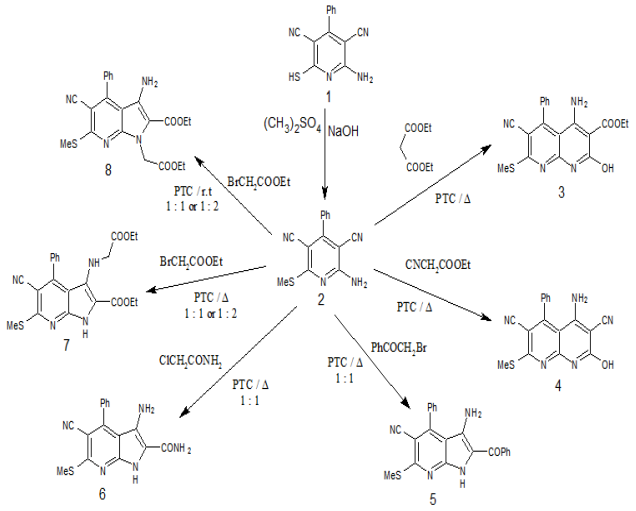
Considering the above very interesting pharmacological properties, we have now designed and synthesized some novel functionalized pyridines and their fused polycyclic ring systems, with important heterocycles such as pyrrole, pyrimidines and pyrazole with the hope to possess better biological activity.

2 Results and Discussions

The starting compound 2-amino-6-(methylthio)-4-phenylpyridine3,5-dicarbonitrile **2** was prepared by the reaction of 6-amino-4-phenyl-2-thioxo-1,2-dihydropyridine-3,5-dicarbonitrile(**1**)[15] with dimethyl sulphate in sodium hydroxide solution.

Under phase transfer catalysis conditions (PTC) using dioxane as the organic phase, potassium carbonate as the solid phase and tetrabutylammonium bromide (TBAB) as a catalyst, compound **2** was allowed to react with diethyl malonate, ethyl cyanoacetate, phenacyl bromide or 2-chloroacetamide to give ethyl 4-amino-6-cyano-2-hydroxy-7-(methylthio)-5-phenyl-1,8-naphthyridine-3-carboxylate 3,4-amino-2-hydroxy-7-(methylthio)-5-phenyl-1,8-naphth-yridine-3,6-dicarbonitrile 4, 3-amino-2-benzoyl-6-(methylthio)-4-phenyl-1H-pyrrolo[2,3-b]-pyridine-5-carbonitrile **5** or 3-amino-5-cyano-6-(methylthio)-4-phenyl-1H-pyrrolo[2,3-b]pyr-idine-2-carboxamide **6**, respectively. Under similar PTC conditions, the reaction of compound **2** with ethyl bromoacetate in (1 : 1 or 1 : 2) molar ratio at room temperature afforded ethyl 2-(3-amino-5-cyano-2-ethyloxycarbonyl-6-methylsulfanyl-4-phenyl-1H-pyrrolo-[2,3-b]- pyridin-1-yl)acetate 7, while, on heating gave ethyl 2-(5-cyano-2-ethyloxycarbonyl-6-methyl-sulfanyl-4-phenyl-1H-pyrrolo[2,3-b]pyridin-3-ylamino) acetate (**8**), respectively (Scheme 1).

The IR spectra of compounds **3** and **4** showed new absorption bands at 3475 – 3396 cm^{-1} corresponding to OH groups and at 1734 cm^{-1} corresponding to C=Oester, in the case of compound **3**. The ^{1}H-NMR spectra of compounds **3** and **4** revealed new singlet signals at 4.20–4.00 ppm corresponding to OH groups and in the case of compound **3**, new quartet signal at 4.00–3.80 ppm corresponding to



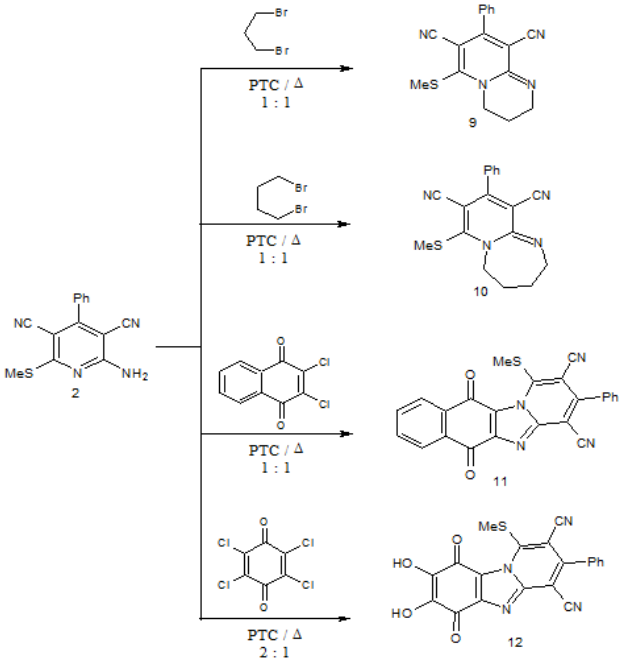
Scheme 1

CH_2ester group and a triplet signal at 1.20–1.00 ppm corresponding to CH_3ester.

The IR spectra of compounds **5** and **6**showed new absorption bands at 3454–3213 cm^{-1} corresponding to NH, NH_2 groups and at 1691–1680 cm^{-1} corresponding to C=O groups. The ^{1}H-NMR spectra of compounds **5** and **6** revealed multiplet signals at 7.50–6.70 corresponding to (10 aromatic protons + NH_2 + NH) groups, , and new singlet signal at 6.20 ppm corresponding to $CONH_2$ group, in the case of compound **5**.

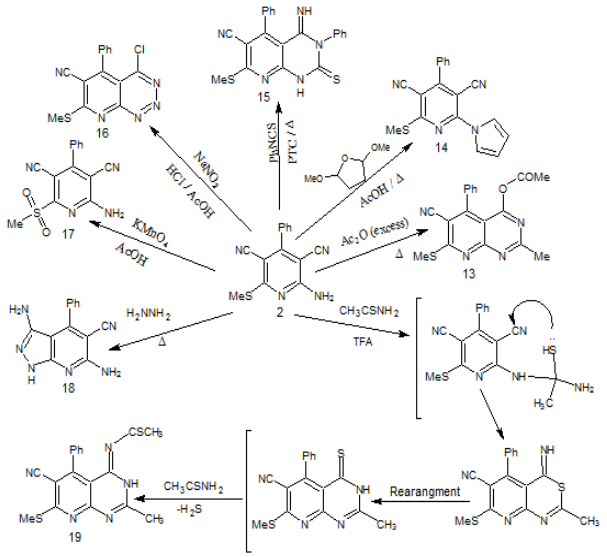
The IR spectra of compounds **7** and **8**showed new absorption bands at 1756 – 1750 cm^{-1} corresponding to C=Oester groups. The mass spectrum of **7** showed the following fragmentation pattern m/e (rel. intensity %): 438 (M^+, 58.51), 352 (M^++2 –CH_3COOEt, 51.07) and 279 (M^+ –$EtOOCCH_2COOEt$, 100).While, the mass spectrum of **8** showed the following fragmentation pattern m/e (rel. intensity %): 438 (M^+, 100), 365 (M^++1 –HCOOEt, 53.21) and 319 (M^+–MeSCOOEt, 34.36).

Under similar reaction (PTC) conditions compound **2** was allowed to undergo cycloalkylation by heating with some halo reagents namely; 1,3-dibromopropane, 1,4-dibromobutane, 2,3-dichloro-1,4-naphthoquinone or 2,3,5,6-tetrachloro-1,4-benzoquinone to give 6-(methylthio)-8-phenyl-3,4-dihydro-2H-pyrido[1,2-a]pyri-midine-7,9-dicarbonitrile **9**, 7-(methylthio)-9-phenyl-2,3,4,5-tetrahydro-pyrido[1,2-a][1,3]diazepine-8,10-dicarbonitrile **10**, 4-methylsulfanyl-6,11-dioxo-2-phenyl-6,11-dihydronaphtho[2',3':4,5]imidazo[1,2-a]pyridine-1,3-dicarbonitrile **11** or 7,8-dihydroxy-1-(methylthio)-6,9-dioxo-3-phenyl-6,9-dihydropyrido[1,2-a]benzimid-azole-2,4-dicarbonitrile **12**, respectively(Scheme 2). The IR spectra of compounds **9-12** showed the absence of absorption bands corresponding to NH_2 group while exhibit absorption bands at 2970–2947 cm^{-1}which revealed the presence of protons attached to SP^3 carbons, in the case of compounds **9**, **10** and at 1673–1624 cm^{-1} corresponding to C=O, in the case of compounds **11**, **12**. Also, the IR spectrum of compound **12** exhibit new absorption bands at 3736, 3444 cm^{-1} corresponding to OH groups. The ^{1}H-NMR spectra of compounds **9-12** showed the absence of the signals corresponding to NH_2 group while revealed alicyclic protons signals at 4.40–1.60 ppm, in the case of compounds **9**, **10** and aromatic protons signals at 7.80–7.00 ppm, in the case of compound **11**. The ^{1}H-NMR spectrum of compound **12** showed new singlet signal at 3.80 ppm corresponding to 2 OH groups.



Scheme 2

Cycloacetylation of compound **2** using acetic anhydride afforded 6-cyano-2-methyl-7-(methylthio)-5-phenylpyrido-[2,3-d]pyrimidin-4-yl acetate **13**.Treatment of compound 2 with 2,5-dimethoxytetrahydrofuran yielded 2-(methylthio)-4-phenyl-6-(1H-pyrrol-1-yl)pyridine-3,5-dicarbonitrile **14**. Under PTC reaction conditions compound **2** was allowed to react with phenylisothiocyanate to give 4-imino-7-(methylthio)-3,5-diphenyl-2-thioxo-1,2,3,4-tetrahydropyri-do[2,3-d]pyramid-ine-6-carbonitrile **15**. Diazotization of compound 2with sodium nitrite and HCl/AcOH mixture gave 4-chloro-7-(methylthio)-5-phenylpyrido[2,3-d][1,2,3]-triazine-6-carbonitrile **16**. Oxidation of compound **2** with potassium permanganate in acetic acid afforded 2-amino-6-(methylsulfonyl)-4-phenylpyridine-3,5-dicarbonitrile **17**. The sulfur-free compound 3,6-diamino-4-phenyl-1H-pyrazolo[3,4-b]pyridine-5-carbonitrile **18** was obtained by heating compound **2** with excess hydrazine hydrate. Also compound **2** was treated with thioacetamide in trifluoro-acetic acid to give 2-methyl-7-methylsulfanyl-5-phenyl-4-(1-thioxoethylimino)-3,4-dihydropyrido-[2,3-d]pyrimidin-6-yl cyanide **19**. The reaction mechanism for the formation of product 19 was suggested as shown in scheme 3.



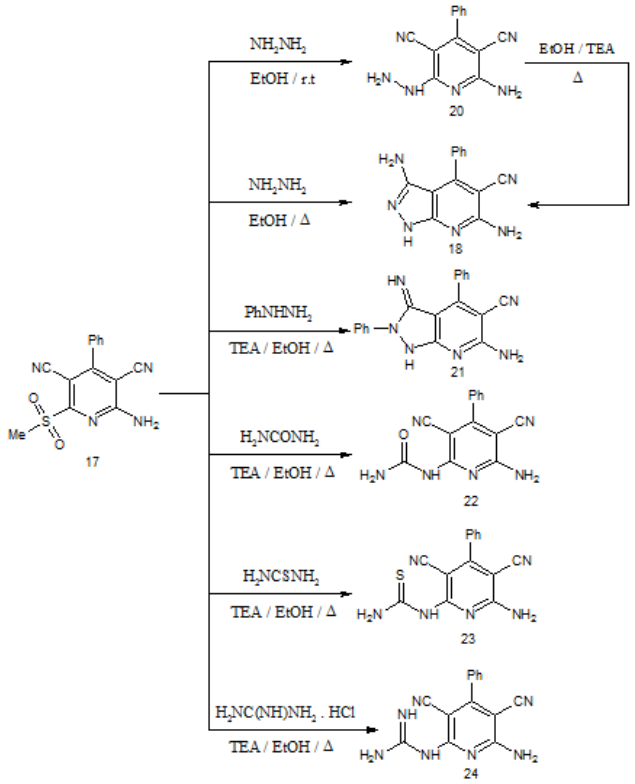
Scheme 3

The IR and ^{1}H-NMR spectra of compounds**13-16** showed the absence of absorption bands corresponding to NH_2 group. ^{1}H-NMR spectrum of compound **13** revealed new singlet signals at 2.80 ppm and at 2.30 ppm corresponding to $2CH_3$ groups. The IR spectrum of compound **17** showed new absorption bands at 1325, 1148 cm^{-1} corresponding to O=S=O group. The IR spectrum of compound **18** showed the absence of absorption bands corresponding to protons attached to SP^3 carbons while exhibit new absorption bands at 3449, 3208, 3160 cm^{-1} corresponding to NH_2, NH groups. The ^{1}H-NMR spectrum of compound **18** showed the absence of the signal corresponding to SCH_3 group while revealed new singlet signals at 11.80 ppm and at 4.30 ppm corresponding to NH and NH_2 groups, respectively. Moreover, the mass spectrum of compound **18** gave *m/z* 250 [M^+] (I_{rel}100%) which corresponds to the molecular weight of the molecular formula $C_{13}H_{10}N_6$ of the assigned structure.

2-Amino-6-hydrazino-4-phenylpyridine-3,5-dicarbo-nitrile **20** was yielded by treating compound **17** with hydrazine hydrate at room temperature, which in turn underwent intermolecular cyclization into compound **18**. Also compound **18** was synthesized directly in one step by heating compound **17** with hydrazine hydrate in ethanol (Scheme 4).

In ethanol and TEA as a catalyst, compound **17** was heated with phenyl hydrazine, urea, thiourea or guanidine hydrochloride giving 6-amino-3-imino-2,4-diphenyl-2,3-dihydro-1H-pyrazolo[3,4-b]pyridi-ne-5-carbonitrile **21**, 1-(6-amino-3,5-dicyano-4-phenyl-pyridin-2-yl)urea **22**, 1-(6-amino-3,5-dicyano-4-phenylpyridin-2-yl)thiourea **23** or 1-(6-amino-3,5-dicyano-4-phenylpyridin-2-yl)guanidine **24**, respectively (Scheme 4).

The IR spectrum of compound **20** showed the absence of absorption bands corresponding to SO_2 group while exhibit new absorption bands at 3470-3230 cm^{-1} corresponding to NH_2, NH groups. The ^{1}H-NMR spectrum of compound **20** showed the absence of the signal corresponding to CH_3 group. The IR spectra of compounds **21-24** showed the absence of absorption bands corresponding to SO_2 group while exhibit new absorption bands at 3466 – 3180 cm^{-1} corresponding to NH_2, NH groups. The ^{1}H-NMR spectra of compounds **21-24** showed the absence of the signal corresponding to CH_3 group.



Scheme 4

3 Biological activity predicted by PASS

The biological activity spectra of new compounds **2-24** were obtained by PASS software. The predictions were carried out based on analysis of training set containing about 46,000 drugs and biologically active compounds. This set consider as reference compounds for known chemical compounds as well as different biological activities. It estimates the probability of the molecule to be active (Pa) and inactive (Pi) for each type of activity from the biological activity spectrum. Interpretation of prediction results is based on consideration of Pa values [22,24].

1. Pa > 0.7: the chance of finding activity experimentally is high; in many cases the compound may be a close analogue of known pharmaceutical agents.

2. 0.5 < Pa < 0.7: the chance of finding activity

experimentally is less; the compound is not so similar to known pharmaceutical agents.

3. Pa < 0.5: the chance of finding activity experimentally is even less; the compound has only a low similarityto the compounds from the training set.

Percent activity (Pa) and inactivity (Pi) of new compounds, which have Pa more than 0.700, represented in table 1.

According to these data the most frequently predicted types of biological activities are Atherosclerosis treatment, Antineoplastic and DNA intercalator. we got a very important targets for compound **17** that can be confirmed with experiments especially the target of Antiarthritic which has high probability at (Pa=0,940) and Atherosclerosis treatment at (Pa= 0.918). Whereas, compound 18 is expected to exhibit good Protein kinase inhibitor (Pa=0,944) and Signal transduction pathways inhibitor (Pa=0,943).

Table 1: Biological activity predicted by PASS

Compound No.	Activities	Pa	Pi
2	Adenosine A2b receptor agonist Atherosclerosis treatment Adenosine A1 receptor agonist Antidiabetic Antihypertensive	0,896 0,896 0,713 0,706 0,705	0,000 0,003 0,002 0,006 0,005
4	Heart failure treatment	0,714	0,004
8	Atherosclerosis treatment Heart failure treatment	0,765 0,733	0,004 0,004
11	DNA intercalator	0,730	0,003
12	DNA intercalator	0,712	0,003
13	Heart failure treatment Atherosclerosis treatment Antihypertensive	0,784 0,743 0,710	0,004 0,004 0,005
14	Atherosclerosis treatment Centromere associated protein inhibitor Heart failure treatment	0,828 0,817 0,803	0,004 0,004 0,004
17	Antiarthritic Atherosclerosis treatment Adenosine A2b receptor agonist	0,940 0,800 0,705	0,004 0,004 0,000
18	Protein kinase inhibitor Signal transduction pathways inhibitor Tyrosine kinase inhibitor Histidine kinase inhibitor Antineoplastic	0,944 0,943 0,722 0,723 0,704	0,004 0,004 0,005 0,007 0,025
20	Beta-Lysine 5,6-aminomutase inhibitor CDK9/cyclin T1 inhibitor Glucose oxidase inhibitor Antineoplastic (brain cancer)	0,867 0,845 0,798 0,718	0,000 0,001 0,012 0,003
22	Alopecia treatment	0,702	0,006
23	Antineoplastic (melanoma)	0,733	0,004
24	CDP-glycerol glycerophosphotransferase inhibitor Alopecia treatment	0,868 0,737	0,016 0,005

4 Conclusion

This study illustrates that 2-amino-6-(methylthio)-4-phenylpyridine3,5-dicarbonitrile(**2**) is a convenient starting material for the synthesis of new series of fused pyridine derivatives. Due to the availability of the starting materials, the simplicity of the procedures, and the comparatively reasonable yields of the products, this synthetic approach might be valuable for the synthesis of such ring systems. Accompanied with predictions of the activity spectra of some selected compounds using PASS INET.at Pa > 70%, showing high probability of Atherosclerosis treatment, Antineoplastic, DNA intercalator, Protein kinase inhibitor and Signal transduction pathways inhibitor.

5 Experimental

All melting points are uncorrected and were recorded on Melt-Temp II melting point apparatus. IR spectra were measured as KBr pellets on a Shimadzu DR-8001 spectrometer. ^{1}H-NMR spectra were recorded in deuterated chloroform or dimethyl sulfoxide at 60 MHz on a Varian EM 360L and also at 200 MHz on a Varian Gemini NMR spectrometer using tetramethylsilane as an internal reference. Mass spectra were performed on a Shimadzu GCMS-QP 1000 mass spectrometer at 70 ev. The elemental analyses were carried out on a Perkin-Elmer 240C. All compounds were checked for their purity on TLC plates.

2-amino-6-(methylthio)-4-phenylpyridine-3,5-dicarbonitrile (**2**):
To a solution of compound **1** (0.005 mol, 1.26 g) in sodium hydroxide (0.5 g in 20 ml H_2O), dimethyl sulphate (0.005 mol, 0.47 ml) was added drop wise with stirring. The stirring was continued for 2 h. The solid product was filtered off, crystallized from dioxaneand dried in the air ,1.14 g(86%), mp305^{o}C; IR: 3323, 3217 (NH_2); 3069 ($CH_{arom.}$); 2986 (SP3 C-H.); 2219 (2CN). cm^{-1}; ^{1}H-NMR: 7.60(s, 2H, NH_2); 7.30 – 7.00 (m, 5H, arom.); 2.50 (s, 3H, CH_3).; Anal. Calcd. for $C_{14}H_{10}N_4S$:C, 63.14 , H, 3.78; N, 21.04; S, 12.04. Found: C, 63.10; H, 3.64; N, 21.13; S, 11.92.

Synthesis of compounds 3-12:
General procedure:
An equimolar mixture of compound **2** (0.001 mol, 0.266 g) and the appropriate halo compound, diethyl malonate, ethyl cyanoacetate, phenacyl bromide or 2-chloroacetamide in dioxane (20 ml) was treated with anhydrous potassium carbonate (3 g) and a catalytic amount of tetrabutylammonium bromide. The reaction mixture was stirred for a period of time, filtered off and the solvent was evaporated in vacuo. The resulting solid was crystallized from ethanol.

Ethyl 4-amino-6-cyano-2-hydroxy-7-(methylthio)-5-phenyl-1,8-naphthyridine-3-carboxylate (3):

Yield: 40%, mp271^{o}C; IR: 3475 (OH); 3324, 3215 (NH_2);

3056 ($CH_{arom.}$); 2954 (SP^3 C-H.); 2211 (CN); 1734 (C=O).cm^{-1}; ^{1}H nmr: 7.7 – 7.0 (m, 5H, arom.); 6.4 (s, 2H, NH_2); 4.2 (s, 1H, OH); 4.0 – 3.8 (q, 2H, CH_2 ester); 2.5 (s, 3H, CH_3S); 1.2–1.0 (t, 3H, CH_3 ester).; Anal. Calcd. for $C_{19}H_{16}N_4O_3S$: C, 59.99; H, 4.24; N, 14.73; S, 8.43. Found: C, 60.06; H, 4.35; N, 14.65; S, 8.32.

4-amino-2-hydroxy-7-(methylthio)-5-phenyl-1,8-naphthyridine-3,6-dicarbonitrile (4):

Yield: 64%, mp265ºC; IR: 3396 (OH); 3324, 3215 (NH_2); 3050 ($CH_{arom.}$); 2959 (SP^3 C-H); 2198 (2CN).cm^{-1}; ^{1}H-NMR : 7.50–7.00 (m, 5H, arom.); 6.30 (s, 2H, NH_2); 4.00 (s, 1H, OH); 2.50 (s, 3H, CH_3).; Anal. Calcd. for $C_17H_{11}N_5OS$: C, 61.25; H, 3.33; N, 21.01; S, 9.62. Found: C, 61.38; H, 3.21; N, 21.10; S, 9.55.

3-amino-2-benzoyl-6-(methylthio)-4-phenyl-1H-pyrrolo[2,3-b]pyridine-5-carbonitrile (5):

Yield: 70%, mp132ºC; IR: 3418, 3360, 3240 (NH_2, NH); 3060 ($CH_{arom.}$); 2926 (SP^3 C-H.); 2214 (CN); 1680 (C=O). cm^{-1}; ^{1}H-NMR: 7.50–6.70(m, 13H, arom. + NH_2 + NH); 2.40 (s, 3H, CH_3).; Anal. Calcd. for $C_{22}H_{16}N_4OS$: C, 68.73; H, 4.19; N, 14.57; S, 8.34. Found: C, 68.66; H, 4.11; N, 14.66; S, 8.23.

3-amino-5-cyano-6-(methylthio)-4-phenyl-1H-pyrrolo[2,3-b]-pyridine-2-carboxamide (6):

Yield: 60%, mp230ºC; IR: 3454, 3323, 3213 ($2NH_2$, NH); 3060 (CHarom.); 2931 (SP^3 C-H); 2209 (CN); 1691 (C=O).cm^{-1}; ^{1}H-NMR: 9.90 (s, 1H, NH); 7.50 – 7.10 (m, 7H, arom. + NH_2); 6.20 (s, 2H, $CONH_2$); 2.50 (s, 3H, CH^3).; Anal. Calcd. for $C_{16}H_{13}N_5OS$: C, 59.43; H, 4.05; N, 21.66; S, 9.92. Found: C, 59.34; H, 3.97; N, 21.62; S, 9.82.

ethyl 2-(3-amino-5-cyano-2-ethyloxycarbonyl-6-methylsulfanyl-4-phenyl-1H-pyrrolo[2,3-b]pyridin-1-yl)acetate (7):

Yield: 62%, mp146ºC; IR: 3464, 3338 (NH_2); 3047 (CHarom.); 2989, 2933 (SP^3 C-H.); 2213 (CN); 1750 (C=Oester).cm-1; ^{1}H-NMR: 7.40 – 6.90 (m, 5H, arom.); 4.90 (s, 2H, N–CH_2); 4.50 (s, 2H, NH_2); 4.10 – 3.70 (q, 4H, $2CH_2$ ester); 2.40 (s, 3H, SCH_3); 1.40 – 1.00 (t, 6H, $2CH_3$ ester).; Anal. Calcd. for $C_{22}H_{22}N_4O_4S$: C, 60.26; H, 5.06; N, 12.78; S, 7.31. Found: C, 60.18; H, 5.11; N, 12.86; S, 7.39.

Ethyl 2-(5-cyano-2-ethyloxycarbonyl-6-methyl-sulfanyl-4-phenyl-1H-pyrrolo[2,3-b]pyridin-3-ylamino)- acetate (8):

Yield: 85%, mp170 ºC; IR: 3463, 3329 (2NH); 3050 (CHarom.); 2988, 2933 (SP^3 C-H); 2216 (CN); 1756 ($C=O_{ester}$).cm-1; ^{1}H-NMR: 7.50–7.20 (m, 6H, arom. + NH); 5.10 (s, 2H, N–CH_2); 4.60 (s, 1H, NH); 4.30 – 3.90 (q, 4H, $2CH_2$ ester); 2.50 (s, 3H, SCH_3); 1.50 – 1.10 (t, 6H, $2CH_3$ ester).; Anal. Calcd. for $C_{22}H_{22}N_4O_4S$: C, 60.26; H, 5.06; N, 12.78; S, 7.31. Found: C, 60.21; H, 5.01; N, 12.71; S, 7.25.

6-(methylthio)-8-phenyl-3,4-dihydro-2H-pyrido-[1,2-a]pyrimidine-7,9-dicarbonitrile (9):

Yield: 76%, mp217ºC; IR: 3050 (CH-arom.); 2947 (SP^3 C-H); 2211 (2CN).cm^{-1}; ^{1}H-NMR: 7.40-7.00 (m, 5H, arom.); 4.40– 4.00 (t, 4H, $2CH_2$–N); 2.50 (s, 3H, CH_3); 2.50 – 2.20 (m, 2H, CH_2).; Anal. Calcd. for $C_{17}H_{14}N_4S$: C, 66.64; H, 4.61; N, 18.29; S, 10.47. Found: C, 66.51; H, 4.49; N, 18.21; S, 10.55.

7-(methylthio)-9-phenyl-2,3,4,5-tetrahydropyrido-[1,2-a][1,3]-diazepine-8,10-dicarbonitrile (10):

Yield: 96%, mp182 ºC; IR: 3056 (CHarom.); 2970 (SP^3 C-H.); 2205 (2CN).cm^{-1}; ^{1}H-NMR: 7.50-7.00 (m, 5H, arom.); 3.90 – 3.40 (t, 4H, $2CH_2$-N); 2.50 (s, 3H, CH_3); 2.20–1.60 (m, 4H, $2CH_2$).; Anal. Calcd. for $C_{18}H_{16}N_4S$: C, 67.47; H, 5.03; N, 17.49; S, 10.01. Found: C, 67.39; H, 5.12; N, 17.33; S, 10.08.

4-methylsulfanyl-6,11-dioxo-2-phenyl-6,11-dihydrona-phtho-[2',3':4,5]imidazo[1,2-a]pyridine-1,3-dicarbonitrile (11):

Yield: 77%, mp310ºC; IR: 3059 (CHarom.); 2928 (SP^3 C-H.); 2204 (2CN); 1673 (2C=O).cm-1; ^{1}H-NMR: 7.80 -7.20 (m, 9H, arom.); 2.40 (s, 3H, CH_3).; Anal. Calcd .for $C_{24}H_{12}N_4O_2S$: C, 68.56; H, 2.88; N, 13.33; S, 7.63. Found: C, 68.45; H, 2.95; N, 13.24; S, 7.52.

7,8-dihydroxy-1-(methylthio)-6,9-dioxo-3-phenyl-6,9-dihydropyrido[1,2-a]benzimidazole-2,4-dicarbonitrile (12):

Yield: 73 %, mp280ºC; IR: 3736, 3444 (2OH); 3062 (CHarom.); 2932 (SP^3 C-H); 2212 (2CN); 1644 (2C=O).cm^{-1}; ^{1}H-NMR: 7.50–7.00 (m, 5H, arom.); 3.80 (s, 2H, 2OH); 2.50 (s, 3H, CH_3).; Anal. Calcd. for $C_{20}H_{10}N_4O_4S$: C, 59.70; H, 2.50; N, 13.92; S, 7.97. Found: C, 59.62; H, 2.63; N, 14.01; S, 7.83.

6-cyano-2-methyl-7-(methylthio)-5-phenylpyrido-[2,3-d]pyrimidin-4-yl acetate (13):

A solution of compound**2** (0.001 mol, 0.266 g) in acetic anhydride (15 ml) was heated under reflux for 4 h and then allowed to cool to room temperature. The reaction mixture was poured onto ice cold water. The obtained solid product was filtered, washed with water and crystallized from ethanol, 0.29 g(83%), mp175ºC; IR: 3016 (CHarom.); 2934 (SP^3 C-H.); 2223 (CN); 1733 ($C=O_{ester}$). cm^{-1}; ^{1}H-NMR: 7.40–7.10(m, 5H, arom.); 2.80 (s, 3H, CH_3–C=N); 2.50 (s, 3H, CH_3CO); 2.30 (s, 3H, CH_3S).; Anal. Calcd. for $C_{18}H_{14}N_4O_2S$:C, 61.70, H, 4.03; N, 15.99; S, 9.15. Found: C, 61.60; H, 4.00; N, 16.07; S, 9.09.

2-(methylthio)-4-phenyl-6-(1H-pyrrol-1-yl)pyridine-3,5-dicarbonitrile (14):

An equimolar ratio of compound **2** (0.001 mol, 0.266 g) and 2,5-dimethoxytetrahydrofuran (0.001 mol, 0.13 ml) in glacial acetic acid (10 ml), was heated under reflux for 4 h

and left to cool. The precipitated crystals was collected by filtration and crystallized from ethanol, 0.20 g(64%), mp198ºC; IR: 3070 (CHarom.); 2930 (SP^3 C-H); 2223 (2CN). cm^{-1}; ^{1}H-NMR: 7.60–7.40 (d, 2H, =CH-N); 7.30 – 7.10 (m, 5H, arom.); 6.30–6.10 (t, 2H, 2 =CH-C); 2.60 (s, 3H, CH_3).; Anal. Calcd. for $C_{18}H_{12}N_4S$:C, 68.33, H, 3.82; N, 17.71; S, 10.13. Found: C, 68.21; H, 3.74; N, 17.61; S, 10.22.

4-imino-7-(methylthio)-3,5-diphenyl-2-thioxo-1,2,3,4-tetrahydropyrido[2,3-d]pyrimidine-6-carbonitrile (15):

An equimolarmixture of compound **2** (0.001 mol, 0.266 g) and phenylisothiocyanate (0.001 mol, 0.12 ml) in dioxane(20 ml) was treated with anhydrous potassium carbonate (3 g) and a catalytic amount of tetrabutylammonium bromide. The reaction mixture was stirred for 4 h at 60-70 °C The precipitated crystals was collected by filtration and crystallized from ethanol, 0.18 g(45 %), mp160ºC; IR: 3439 (NH); 3330 (NH); 3062 (CHarom.); 2935(SP^3 C-H); 2213(CN); 1533 (C=S). cm^{-1}; ^{1}H-NMR: 7.30 – 6.90 (m, 10H, arom.); 6.20 (s, 1H, NH); 4.00 (s, 1H, NH); 2.50 (s, 3H, CH_3).; Anal. Calcd. for $C_{21}H_{15}N_5S_2$:C, 62.82, H, 3.77; N, 17.44; S, 15.97. Found: C, 62.95; H, 3.86; N, 17.37; S, 16.00.

4-chloro-7-(methylthio)-5-phenylpyrido[2,3-d]-[1,2,3]-triazine-6-carbonitrile (16):

To a chilled solution of compound**2**(0.001 mol, 0.266 g) in a mixture of acetic acid (10 ml) and concentrated hydrochloric acid (7 ml), a sodium nitrite solution 10% (1 ml) was added with stirring during 5 min. The stirring was continued at 5ºC for 3 h. The formed precipitate was collected by filtration and crystallized from ethanol, 0.24 g (78%), mp192ºC; IR: 3054 (CHarom.); 2929(SP^3 C-H); 2211 (CN). cm^{-1}; ^{1}H-NMR: 7.40–7.10 (m, 5H, arom.); 2.50 (s, 3H, CH_3).; Anal. Calcd. for $C_{14}H_8ClN_5S$:C, 53.59, H, 2.57; N, 22.32; S, 10.22. Found: C, 53.51; H, 2.45; N, 22.43; S, 10.29.

2-amino-6-(methylsulfonyl)-4-phenylpyridine-3,5-dicarbonitrile (17):

To a solution of compound **2**(0.001 mol, 0.266 g) in glacial acetic acid (30 ml), potassium permanganate solution (0.001 mol, 0.158 g in 1 ml H_2O) was added during 5 min. The stirring was continued at r.t for 3 h. The reaction mixture was filtrated. The filtrate poured onto water and the resulting solid was crystallized from ethanol, 0.17 g (58%), mp240ºC; IR: 3331, 3223 (NH_2); 3014 (CHarom.); 2924 (SP^3 C-H); 2224 (2CN); 1325, 1148 (SO_2). cm-1; ^{1}H-NMR: 7.90 (s, 2H, NH_2); 7.40–7.10 (m, 5H, arom.); 3.20 (s, 3H, CH_3).; Anal. Calcd. for $C_{14}H_{10}N_4O_2S$:C, 56.37, H, 3.38; N, 18.78; S, 10.75. Found: C, 56.50; H, 3.21; N, 18.66; S, 10.62.

3,6-diamino-4-phenyl-1H-pyrazolo[3,4-b]pyridine-5-carbonitrile (18):

Method A:
A mixture of compound **2**(0.001 mol, 0.266 g) and hydrazine hydrate (5 ml) was heated under reflux for about 9 h until the odor of CH3SH ceased and then allowed to cool to room temperature. The mixture triturated with ethanol (10 ml). The solid product was filtered and crystallized from ethanol.

Method B:
A mixture of compound **17**(0.001 mol, 0.298 g) and hydrazine hydrate (0.1 ml) in ethanol (20 ml) was refluxed for 2 h. The product that formed by cooling was collected by filtration and crystallized from ethanol.

Method C:
To a solution of compound **20** (0.001 mol, 0.25 g) in ethanol (20 ml), TEA (2 drops) was added. The reaction mixture was refluxed for 2 h. The product that formed after cooling was collected by filtration and crystallized from ethanol.

Yield: 65%, mp282ºC; IR: 3449, 3338, 3208, 3160 ($2NH_2$, NH); 3055 (CHarom.); 2205 (CN).cm^{-1}; ^{1}H-NMR: 11.80 (s, 1H, NH); 7.80–7.20 (m, 5H, arom.); 6.80 (s, 2H, NH_2); 4.30 (s, 2H, NH_2).; Anal. Calcd. for $C_{13}H_{10}N_6$: C, 62.39; H, 4.03; N, 33.58. Found: C, 62.28; H, 3.92; N, 33.66.

2-methyl-7-methylsulfanyl-5-phenyl-4-(1-thioxo-ethylimino)-3,4-dihydropyrido[2,3-d]pyrimidin-6-yl cyanide (19):

A mixture of compound **2** (0.001 mol, 0.266 g) and thioacetamide (0.001 mol, 0.075 g) in trifluoroacetic acid (5 ml) was refluxed for 24 h. The mixture was left to cool, was poured onto ice cold water. The obtained solid product was filtered, washed with water and crystallized from dioxane, 0.24 g (65%), mp289ºC; IR: 3322 (NH); 3063 (CHarom.); 2930 (SP^3 C-H); 2216 (CN); 1540 (C=S). cm^{-1}; ^{1}H-NMR: 7.40–7.10 (m, 6H, arom. + NH); 2.60 (s, 3H, CH_3); 2.50 (s, 6H, $2CH_3$).; Anal. Calcd. for $C_{18}H_{15}N_5S_2$:C, 59.15, H, 4.14; N, 19.16; S, 17.55. Found: C, 59.22; H, 4.11; N, 19.09; S, 17.46.

2-amino-6-hydrazino-4-phenylpyridine-3,5-dicarbo-nitrile (20):

A mixture of compound1**7** (0.001 mol, 0.298 g) and hydrazine hydrate (0.1 ml) in ethanol (20 ml) was stirred for 2 h at r.t. The product that formed was collected by filtration and crystallized from ethanol, 0.12 g (48%),mp225ºC; IR: 3470, 3332, 3230 ($2NH_2$, NH); 3050 (CHarom.); 2203 (2CN). cm^{-1}; ^{1}H-NMR: 7.60 – 7.00 (m, 5H, arom.); 6.90 – 6.30 (br, 1H, NH); 4.00 – 3.20 (br, 4H, $2NH_2$).; Anal. Calcd. for $C_{13}H_{10}N_6$:C, 62.39, H, 4.03; N, 33.58. Found: C, 62.48; H, 4.10; N, 33.27.

Synthesis of compounds 21–24:

General procedure:
To a mixture of compound **17**(0.001 mol, 0.298 g) and an equiomolar ratio of phenyl hydrazine, urea,

thiourea or guanidine hydrochloride in ethanol (20 ml), TEA (0.15 ml) was added. The reaction mixture was heated under reflux for 4 h, allowed to cool and concentrated. The product that formed was collected by filtration and crystallized from ethanol.

6-amino-3-imino-2,4-diphenyl-2,3-dihydro-1H-pyrazolo[3,4-b]-pyridine-5-carbonitrile (21):

Yield: 45%, mp220ºC; IR: 3466, 3319, 3206, 3180 (NH_2, 2NH); 3070 (CHarom.); 2219 (CN).cm^{-1}; ^{1}H-NMR: 7.80 – 7.00 (m, 12H, arom. + NH_2); 6.20 (s, 1H, NH); 4.80 (s, 1H, NH).; Anal. Calcd. for $C_{19}H_{14}N_6$: C, 69.92; H, 4.32; N, 25.75. Found: C, 7.03; H, 4.21; N, 25.83.

1-(6-amino-3,5-dicyano-4-phenylpyridin-2-yl)urea(22):

Yield: 42%, mp185ºC; IR: 3460, 3323, 3206 ($2NH_2$, NH); 3053 (CHarom.); 2207 (2CN); 1642 (C=O).cm-1; ^{1}H-NMR: 7.40 – 6.90 (m, 6H, arom. + NH); 4.20 (s, 4H, $2NH_2$).; Anal. Calcd. for $C_{14}H_{10}N_6O$: C, 60.43; H, 3.62; N, 30.20. Found: C, 60.35; H, 3.56; N, 30.27.

1-(6-amino-3,5-dicyano-4-phenylpyridin-2-yl)thiourea (23):

Yield: 40%, mp295ºC; IR: 3454, 3322, 3211 ($2NH_2$, NH); 3060 (CHarom.); 2216 (2CN); 1557 (C=S).cm-1; ^{1}H-NMR: 7.40– 6.90 (m, 6H, arom. + NH); 3.80 (s, 4H, $2NH_2$).; Anal. Calcd. for $C_{14}H_{10}N_6S$: C, 57.13; H, 3.42; N, 28.55; S, 10.89. Found: C, 57.21; H, 3.32; N, 28.43; S, 10.75.

1-(6-amino-3,5-dicyano-4-phenylpyridin-2-yl)guanidine (24):

Yield: 75%, mp222ºC; IR: 3457, 3323, 3218 ($2NH_2$, 2NH); 3055 (CHarom.); 2215 (2CN).cm^{-1}; ^{1}H-NMR: 7.40 – 6.90 (m, 6H, arom. + NH); 3.50 (s, 5H, $2NH_2$ + NH).; Anal. Calcd. for $C_{14}H_{11}N_7$: C, 60.64; H, 4.00; N, 35.36. Found: C, 60.56; H, 3.95; N, 35.28.

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