Available online at www.derpharmachemica.com

ISSN 0975-413X
CODEN (USA): PCHHAX

Der Pharma Chemica, 2016, 8(4):363-376
(http://derpharmachemica.com/archive.html)

A convenient route to 5-aminopyrazole, bispyrazole and pyrazolo[1,5-*a*]pyrimidines incorporating antipyrine or furan moiety as potent antimicrobial agents

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ABSTRACT

Development of new antimicrobial agents is a good solution to overcome drug-resistance problems. In this context, (E)-3-(anthracen-9-yl)-2-cyano-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)acrylamide ***(3)*** *was synthesized and allowed to react with hydrazines to afford 5-aminopyrazoles* ***4a,b****. Also, an efficient three-component, two-step synthesis of 5-aminopyrazoles* ***7*** *was reported, via reaction of 2-cyano-N-(furan-2-ylmethyl)acetamide* ***(2b)****, phenyl isothiocyanate and dimethylsulphate to produce the ketene N,S-acetal* ***6****, which reacted with hydrazine hydrate to furnish 5-aminopyrazole* ***7****. The latter was allowed to react with some electrophilic reagents and many 5-(benzylideneamino)pyrazole* ***8a,b****, bispyrazole* ***9*** *and pyrazolo[1,5-a]pyrimidines* ***12****,* ***17****,* ***20****,* ***22****,* ***25****,* ***26****,* ***28*** *and* ***29*** *were obtained. The synthesized compounds were evaluated for their in vitro antibacterial and antifungal activities. Among the synthesized compounds, bispyrazole* ***9*** *showed equipotent to ampicillin and gentamycin against both of S. epidermidis (MIC 0.49µ/mL), B. subitilis (MIC 0.24µ/mL), P. vulgaris (MIC 0.98µg/mL) and K. pneumonia (MIC 0.49µ/mL), and displayed equipotent to amphotericin B versus A. clavatus (MIC 0.98 µ/mL). Azomethine derivatives* ***8a,b*** *were equipotent to amphotericin B in inhibiting the growth of A. clavatus (MIC 0.98 µ/mL). Pyrazolo[1,5-a]pyrimidine* ***17*** *was equipotent to amphotericin B in inhibiting the growth of G. candidium (MIC 0.49 µ/mL). Structures of the new synthesized compounds were established by elemental analysis and spectral data.*

Keywords: aminopyrazole; azomethine; bispyrazoles; pyrazolo[1,5-*a*]pyrimidines; antibacterial; antifungal.

INTRODUCTION

Several pyrazole derivatives received great attention due to their biological and pharmacological activities; not only as potential inhibitors of HIV-1[1], pesticides [2], fungicides [3], antihypertensive agents [4], and anticancer activity [5], but they are also important and useful starting materials for the synthesis of other fused heterocyclic systems. The synthesis of pyrazolo[1,5-*a*]pyrimidine and their derivatives have attracted attention due to their interesting pharmacological properties[6-10]. Also, many biologically important derivatives of furan substituted at 2- and 5-positions are frequently observed in nature. These furan derivatives show broad-spectrum phytocidal, antibacterial and insecticidal activities [11,12] and exhibit pharmacological properties which include serving as antidepressant and anti-inflammatory agents [13,14]. Furthermore, Antipyrine derivatives are well known compounds used mainly as analgesic, antipyretic drugs, antitumoragent, antivirus, anticancer and radiosensitizing agent [15-17]. One of the best known antipyrine derivatives is 4-aminoantipyrine (4-amino-2,3-dimethyl-1-phenyl-3-pyrazolin-5-one), which presents a structural similarity with metamizole, a well-known and very effective analgesic, anti-inflammatory agent [18]. In view of the above facts and in continuation of our research Program [19-26] directed towards the development of a new, simple and less toxic antimicrobial agents. It seems of considerable interest to synthesize newly pyrazole and pyrazolo[1,5-*a*]pyrimidine derivatives bearing antipyrine or furan moiety. Additionally, my

target is also to study the antimicrobial activities of the synthesized compounds, hoping to add some synergistic biological significance to the target molecules.

MATERIALS AND METHODS

All chemicals and solvents were commercially available and used without purification. Melting points were determined by open glass capillary method on a Cintex melting point apparatus and are uncorrected. IR spectra were recorded on a Perkin Elmer spectrometer in KBr pellets. ^{1}H NMR and ^{13}C NMR spectra were obtained on a Varian Gemini 300 MHz spectrometer using TMS as internal standard; chemical shifts are reported as (ppm). Mass spectra were obtained on GCMS\QP 1000 Ex mass spectrometer at 70 ev. Elemental analyses were performed on a LECO-932 analyzer at the Department of Chemistry, Faculty of Science, Cairo University, Egypt. Microbiology screening was carried out in the Regional Center for Microbiology and Biotechnology (RCMB), Antimicrobial unit test organisms, Al-azhr University, Cairo, Egypt.

Chemistry

Preparation of acrylamides 3and 6

E-3-(anthracen-9-yl)-2-cyano-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)acrylamide (3)

A mixture of compound **2a** (0.01 mol), anthracene-9-carbaldehyde (0.01 mol) and piperidine (0.5ml) in ethanol (30 mL) was heated under reflux for 3 hrs; then it was poured into ice/water and acidified with 0.1 N HCl at pH 3-4. The resulting precipitate was filtered off, dried, and recrystallized from methanol as red solid. Yield 65%; mp 140-142ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 2.31, 3.15 (2s, 6H, $2CH_3$), 7.16-8.86 (m, 14H, Ar-H), 9.15(s, 1H, =CH), 10.15 (s, 1H, NH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3432 (br, NH), 3048 (arom. CH), 2931 (aliph. CH), 2220 (C≡N) and 1646 (C=O; amide); MS: m/z 458 (M^+, 28%), 56 (100%); Anal. Calcd for $C_{29}H_{22}N_4O_2$. Calcd: C, 75.97; H, 4.84; N, 12.22. Found: C, 75.99; H, 4.88; N, 12.16%.

2-cyano-N-(furan-2-ylmethyl)-3-(methylthio)-3-phenylamino)acrylamide (6)

To suspend finely powdered potassium hydroxide (0.01 mol) in dry dimethyl formamide (10 mL), phenyl isothiocyanate (0.01 mol) and 2-cyano-*N*-(furan-2-ylmethyl)acetamide(**2b**) (0.01 mol) were added gradually. The reaction was stirred at room temperature for 3 hrs, then treated with dimethylsulfate (0.01mol) and stirred at room temperature for an additional 6 hrs. Then it was poured into ice/water and acidified with 0.1 *N* HCl at pH 3-4.; the resulting precipitate was filtered off, dried and recrystallized from ethanol as white crystals. Yield 70%; m p 90-92 ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 2.41(s, 3H, SCH_3), 4.39 (d, 2H, CH_2-N, *J* = 6 Hz),6.15 (d, 1H, furan-H3, *J* = 3.3 Hz), 6.35 (t, 1H, furan-H4, *J* = 3.5 Hz), 7.02-7.61 (m, 6H, Ar-H + furan-H5), 8.26 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz), 10.52(s, 1H, NH); IR (KBr,*v*, cm^{-1}): 3337(NH), 2924 (aliph. CH), 2194(C≡N), 1621(C=O; amide); MS: m/z 313 (M^+, 8.08%), 81(100%); Anal. Calcd for $C_{16}H_{15}N_3O_2S$. Calcd: C, 61.32; H, 4.82; N, 13.41. Found: C, 61.28; H, 4.78; N, 13.44%.

Preparation of pyrazoles 4a,b and 7: General procedure:

A mixture of acrylamide **3**&/or **6** (0.01 mol) and hydrazines (namely; hydrazine hydrate and / or phenyl hydrazine) (0.01 mol) in ethanol (30 mL) was heated under reflux for 3 hrs. Then it was poured into ice/water and acidified with 0.1 N HCl at pH 3-4.; the resulting precipitate was filtered off, dried and recrystallized from the proper solvent.

5-Amino-3-(anthracen-9-yl)-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)-1H-pyrazole-4-carboxamide (4a)

Yield 55%; orange solid (MeOH); mp 268-270ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 2.30, 3.42 (2s, 6H, $2CH_3$), 7.01(s, 2H, NH_2; Cancelled with D_2O), 7.35-9.18 (m, 14H, Ar-H), 9.44, 10.07(2s, 2H, 2NH; Cancelled with D_2O); IR (KBr, *v*, cm^{-1}): 3427 (br, NH_2), 3047 (arom.CH) and 1625 (C=O; amide); MS: m/z 488 (M^+, 9.50%),81(100%); Anal. Calcd for $C_{29}H_{24}N_6O_2$. Calcd: C, 71.30; H, 4.95; N, 17.20. Found: C, 71.35; H, 4.92; N, 17.25%.

5-Amino-3-(anthracen-9-yl)-N-(1,5-dimethyl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)-1-phenyl-1H-pyrazole-4-carboxamide (4b)

Yield 53%; orange solid (AcOH); mp 199-200 ºC; ^{1}HNMR (300 MHz, DMSO-d_6): δ =2.31, 3.15 (2s, 6H, $2CH_3$),6.70 (s, 2H, NH_2; exchangeable with D_2O), 7.35-9.18 (m, 19H, Ar-H), 10.66 (s, H, NH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3430, 3300 (br, NH_2/NH), 3047 (arom.CH) and 1625 (C=O; amide); MS: m/z564 (M^+, 25.4%), 458(100%); Anal. Calcd for $C_{35}H_{28}N_6O_2$. Calcd: C, 74.45; H, 5.00; N, 14.88. Found: C, 74.39; H, 4.96; N, 14.92%.

5-Amino-N-(furan-2-ylmethyl)-3-(phenylamino)-1H-pyrazole-4-carboxamide (7)

Yield 75%; white solid (MeOH); mp 98-100 ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 4.41 (d, 2H, CH_2-N, *J* = 6 Hz), 5.84 (s, 2H, NH_2; exchangeable with D_2O), 6.18 (d, 1H, furan-H3, *J* = 3 Hz), 6.36 (t, 1H, furan-H4, *J* = 3 Hz), 6.72-

7.32 (m, 6H, Ar-H + furan-H5), 7.53, 11.06 (2s, 2H, 2NH; exchangeable with D_2O), 8.83 (t, 1H, O=C-NH-CH_2; *J* = 6 Hz, exchangeable with D_2O); ^{13}C NMR (300 MHz, DMSO-d_6) δ = 35.09(CH_2), 106.40, 164.45, 164.47(pyrazole-C), 110.36, 115.76, 141.72, 142.91 (furan-C), 119.50, 121.90, 128.76 (Ph-C), 152.84 (C=O); IR (KBr,*v*, cm^{-1}): 3351, 3233 (NH_2/NH) and 1620(C=O; amide); MS: m/z 297(M^+, 28.52%), 81(100%); Anal. Calcd for $C_{15}H_{15}N_5O_2$. Calcd: C, 60.60; H, 5.09; N, 23.56. Found: C, 60.63; H, 5.05; N, 23.51%.

Preparation of azomethines 8a,b and 9: General procedure:
A mixture of compound **7** (0.01mol), aromatic aldehyde (namely; *o*-chlorobenzaldehyde, *p*-chlorobenzaldehyde, terephthalaldehyde) (0.01 mol) and piperidine (0.5ml) in ethanol (30 mL) was heated under reflux for 1 h; the solid product which produced by heating was collected and recrystallized from the proper solvent.

5-(2-Chlorobenzylideneamino)-N-(furan-2-ylmethyl)-3-(phenylamino)-1H-pyrazole-4-carboxamide (8a)
Yield 55%; orange solid (Dioxane); mp 200-202 ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 4.41 (d, 2H, CH_2-N, *J* = 6 Hz), 6.19 (d, 1H, furan-H3, *J* = 3 Hz), 6.36 (t, 1H, furan-H4, *J* = 3 Hz), 6.44-8.09 (m, 10H, Ar-H + furan-H5), 8.64 (s, 1H, N=CH), 8.91 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz), 8.97, 12.84 (2s, 2H, 2NH); IR (KBr,*v*, cm^{-1}): 3113 (NH), 3039 (arom.CH) and 1645 (C=O; amide); MS: m/z 419.5 [M^+ (27.15%), M^{+1} (10.53%)], 73(100%); Anal. Calcd for $C_{22}H_{18}ClN_5O_2$. Calcd: C, 62.93; H, 4.32; N, 16.68. Found: C, 62.89; H, 4.35; N, 16.72%.

5-(4-Chlorobenzylideneamino)-N-(furan-2-ylmethyl)-3-(phenylamino)-1H-pyrazole-4-carboxamide (8b)
Yield 60%; yellow solid (AcOH); mp 158-160 ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 4.55 (d, 2H, CH_2-N, *J* = 3 Hz), 6.36 (d, 1H, furan-H3, *J* = 3 Hz), 6.44 (t, 1H, furan-H4, *J* = 3 Hz), 6.85-8.05 (m, 10H, Ar-H + furan-H5), 8.14 (s, 1H, N=CH), 8.93 (t, 1H, O=C-NH-CH_2, *J* = 3 Hz), 9.22, 13.00 (2s, 2H, 2NH); IR (KBr, *v*, cm^{-1}): 3105 (NH), 3119 (arom.CH) and 1642 (C=O; amide); MS: m/z 419.5 [M^+ (25.20%), M^{+1} (10.20%)], 81(100%); Anal. Calcd for $C_{22}H_{18}ClN_5O_2$. Calcd: C, 62.93; H, 4.32; N, 16.68. Found: C, 62.97; H, 4.28; N, 16.62%.

5,5'-(1,4-Phenylenebis(methan-1-yl-1-ylidene))bis(azan-1-yl-1-ylidene)-bis(N-(furan-2-ylmethyl)-3-(phenylamino)-1H-pyrazole-4-carboxamide) (9)
Yield 60%; red solid (Dioxane); mp 289-290 ºC; IR (KBr, *v*, cm^{-1}): 3352, 3267 (NH), 3063 (arom.CH) and 1634 (C=O; amide); MS: m/z 692 (M^+, 12.5%), 64 (100%); Elemental analysis for $C_{38}H_{32}N_{10}O_4$. Calcd: C, 65.89; H, 4.66; N, 20.22. Found: C, 65.85; H, 4.66; N, 20.17%.

Preparation of 12a* and *12b*: *General procedure:
A mixture of **7**(0.01 mol), α-cinnamonitrile **10** (0.01 mol) and piperidine (0.5mL) in ethanol (30 mL) was heated under reflux for 3 hrs; the solid product which produced by heating was filtered off and recrystallized from the proper solvent to give **12**.

7-Amino-5-(2-chlorophenyl)-6-cyano-N-(furan-2-ylmethyl)-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (12a)
Yield 60%; white solid (Dioxane); mp 300-302 ºC; ^{1}HNMR (300 MHz, DMSO-d_6) δ = 4.54 (d, 2H, CH_2-N, *J* = 6 Hz), 6.25 (d, 1H, furan-H3, *J* = 3 Hz), 6.35 (t, 1H, furan-H4, *J* = 3 Hz), 6.96-7.88 (m, 10H, Ar-H + furan-H5), 8.10 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz), 9.15 (s, 2H, NH_2), 9.54 (s, 1H, NH); IR (KBr, *v*, cm^{-1}): 3416, 3334, 3162 (NH_2/NH), 2216 (C≡N), and 1655 (C=O; amide); MS: m/z 483 (M^+, 20%), 387 (100%); Anal. Calcd for $C_{25}H_{18}ClN_7O_2$. Calcd: C, 62.05; H, 3.75; N, 20.26. Found: C, 62.10; H, 3.72; N, 20.21%.

7-Amino-6-cyano-N-(furan-2-ylmethyl)-5-(4-methoxyphenyl)-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (12b)
Yield 62%; white solid (Dioxane); mp 298-300 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 3.86 (s, 3H, OCH_3), 4.58 (d, 2H, CH_2-N, *J* = 6 Hz), 6.33 (d, 1H, furan-H3, *J* = 3 Hz), 6.43 (t, 1H, furan-H4, *J* = 3 Hz), 6.97-7.86 (m, 10H, Ar-H + furan-H5), 8.32 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz), 8.96 (s, 2H, NH_2), 9.52 (s, 1H, NH); ^{13}C NMR (300 MHz, DMSO-d_6): δ = 35.24 (CH_2), 53.14 (OCH_3), 116.35, 137.50, 139.80,142.50 (furan-C), 127.60, 128.65, 130.37, 132.15, 133.40 (Ar-C), 88.65 (pyrimidine-C5), 149.56 (pyrimidine-C2), 160.59 (pyrimidine-C6), 161.35 (pyrimidine-C4), 106.90 (pyrazole-C3), 158.01 (pyrazole-C2), 152.80 (CN), 163.57 (C=O); IR (KBr, *v*, cm^{-1}): 3326-3270 (NH_2/NH), 2200 (C≡N), and 1660 (C=O; amide); MS: m/z 479(M^+, 18.2%), 386 (100%); Anal. Calcd for $C_{26}H_{21}N_7O_3$. Calcd: C, 65.13; H, 4.41; N, 20.45. Found: C, 65.19; H, 4.36; N, 20.40 %.

7-Amino-5,6-dicyano-N-(furan-2-ylmethyl)-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (17)
A mixture of compound **7** (0.01mol), tetracyanoethylene (**15**) (0.01 mol) and piperidine (0.5ml) in dioxane (30 mL) was heated under reflux for 3 hrs; the solid product which produced by heating was filtered off, dried and recrystallized from acetic acid as brown solid. Yield 45%; mp 285-287 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 4.61 (d, 2H, CH_2-N, *J* = 6 Hz), 6.31 (d, 1H, furan-H3, *J* = 3 Hz), 6.41 (t, 1H, furan-H4, *J* = 3 Hz), 6.97-7.86 (m, 6H, Ar-

H + furan-H5), 8.01 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz), 9.41 (br, 2H, NH_2), 9.61 (s, 1H, NH); IR (KBr, *v*, cm^{-1}): 3422, 3358 (NH_2/NH), 2958 (aliph.CH), 2229(C≡N), 1650 (C=O; amide); MS: m/z 398 (M^+, 55.20%), 96 (100%); Anal. Calcd for $C_{20}H_{14}N_8O_2$. Calcd: C, 60.30; H, 3.54; N, 28.13. Found: C, 60.26; H, 3.49; N, 28.18%.

[Caution: this reaction liberate HCN gas, which is very toxic]

6-Cyano-5-(cyanomethyl)-N-(furan-2-ylmethyl)-7-imino-2-(phenylamino)-6,7-dihydropyrazolo[1,5-a]pyrimidine-3-carboxamide (20)

A mixture of **7** (0.01 mol), 1,1,3-tricyano-2-aminopropene (**18**) (0.01 mol) and piperidine (0.5mL) in ethanol (30 mL) was heated under reflux for 3 hrs; the solid product which was produced by heating was filtered off, dried and recrystallized from dioxane as white solid. Yield 45%; mp 220-222 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 2.20 (s, 2H, CH_2), 4.42 (d, 2H, CH_2-N, *J* = 6 Hz), 6.21 (d, 1H, furan-H3, *J* = 3 Hz), 6.37 (t, 1H, furan-H4, *J* = 3 Hz), 6.77-7.54 (m, 6H, Ar-H + furan-H5), 7.61 (s, 1H, pyrimidine-H), 8.55 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz; exchangeable with D_2O), 10.16, 12.49 (2s, 2H, NH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3302 (NH), 2936 (aliph.CH), 2213(C≡N) and 1676 (C=O; amide); MS: m/z 412 (M^+, 10.50 %), 81 (100%); Anal. Calcd for $C_{21}H_{16}N_8O_2$. Calcd: C, 61.16; H, 3.91; N, 27.17. Found: C, 61.11; H, 3.88; N, 27.12%.

Ethyl 7-amino-3-((furan-2-ylmethyl)carbamoyl)-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-6-carboxylate (22)

A mixture of **7** (0.01 mol), ethyl 2-cyano-3-ethoxyacrylate (**21**) (0.01 mol) and piperidine (0.5mL) in ethanol (30 mL) was heated under reflux for 3 hrs; Then it was poured into ice/water and acidified with 0.1 N HCl at pH 3-4.; the resulting precipitate was filtered off, dried and recrystallized from AcOH as white solid. Yield 42%; mp 189-190 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 1.36 (t, 3H, CH_3), 4.35 (q, 2H, CH_2), 4.59 (d, 2H, CH_2-N, *J* = 6 Hz), 6.33 (d, 1H, furan-H3, *J* = 3 Hz), 6.40 (t, 1H, furan-H4, *J* = 3 Hz), 6.74 -7.83 (m, 6H, Ar-H + furan-H5), 8.27 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz; exchangeable with D_2O), 8.69 (s, 1H, pyrimidine-H), 9.57 (s, 2H, NH_2; exchangeable with D_2O), 11.06 (s, H, NH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3439, 3313 (NH_2/NH), 2926 (aliph.CH), 1691(C=O; ester) and 1650 (C=O; amide); MS: m/z 420 (M^+, 39.40 %), 278 (100%); Anal. Calcd for $C_{21}H_{20}N_6O_4$. Calcd: C, 59.99; H, 4.79; N, 19.99. Found: C, 59.92; H, 4.83; N, 19.95%.

N-(furan-2-ylmethyl)-5-(4-methoxyphenyl)-7-phenyl-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide(25)

A mixture of **7** (0.01 mol), α,β-unsaturated carbonyl derivative **24** (0.01 mol) and piperidine (0.5mL) in ethanol (30 mL) was heated under reflux for 3 hrs; the solid product which produced by heating was filtered off, dried and recrystallized from dioxane as white solid. Yield 40%; mp 190-192 ºC; IR (KBr, *v*, cm^{-1}): 3109 (NH), 2951 (aliph.CH) and 1655(C=O; amide); MS: m/z 515 (M^+, 12.24%), 517 (M^{+2}, 100%); Anal. Calcd for $C_{31}H_{25}N_5O_3$. Calcd: C, 72.22; H, 4.89; N, 13.58. Found: C, 72.17; H, 4.84; N, 13.54%.

Preparation of compounds* 26a**,**b**, **28 and 29: *General procedure:

A mixture of compound **7** (0.01mol) and 1,3-dicarbonyl compound (namely; acetylacetone, benzoylacetone, ethyl acetoacetate and ethyl 2-methyl-3-oxobutanoate) (0.01 mol) in glacial acetic acid (10 mL) was heated under reflux for 3hrs, the solid product which produced by heating was collected and recrystallized from the proper solvent to give **26a**,**b**, **28** and **29** respectively.

N-(furan-2-ylmethyl)-5,7-dimethyl-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (26a)

Yield 60%; white solid (Dioxane); mp 200 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 2.48, 2.68 (2s, 6H, $2CH_3$), 4.59 (d, 2H, CH_2-N, *J* = 6 Hz), 6.31 (d, 1H, furan-H3, *J* = 3 Hz), 6.39 (t, 1H, furan-H4, *J* = 3 Hz), 6.92-7.70 (m, 6H, Ar-H + furan-H5), 7.58 (s, 1H, pyrimidine-H), 8.23 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz; exchangeable with D_2O), 9.50 (s, H, NH; exchangeable with D_2O); ^{13}C NMR (300 MHz, DMSO-d_6): δ = 17.11, 24.63 ($2CH_3$), 35.46 (CH_2), 117.53, 121.32, 124.50.129.53 (Ar-C), 109.47, 110.98, 140.70,142.76 (furan-C), 86.51, 146.88 (pyrazole-C),107.27, 156.52, 161.42, 164.38 (pyrimidine-C), 152.78 (C=O); IR (KBr, *v*, cm^{-1}): 3307 (NH), 2917 (aliph.CH) and 1641(C=O; amide); MS: m/z 361(M^+, 39.42%), 265 (100%); Anal. Calcd for $C_{20}H_{19}N_5O_2$. Calcd: C, 66.47; H, 5.30; N, 19.38. Found:C, 66.42; H, 5.25; N, 19.42%.

N-(furan-2-ylmethyl)-5-methyl-7-phenyl-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (26b)

Yield 65%; white solid (Dioxane); mp 189-190 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 2.49 (s, 3H, CH_3), 4.63 (d, 2H, CH_2-N, *J* = 6 Hz), 6.34 (d, 1H, furan-H3, *J* = 3 Hz), 6.43 (t, 1H, furan-H4, *J* = 3 Hz), 6.90-8.21 (m, 12H, Ar-H + furan-H5+ pyrimidine-H5), 8.38 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz; exchangeable with D_2O), 9.52 (s, H, NH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3101 (NH), 2920 (aliph.CH) and 1643 (C=O; amide). MS: m/z 423 (M^+, 10.40 %), 327(100%); Anal. Calcd for $C_{25}H_{21}N_5O_2$. Calcd: C, 70.91; H, 5.00; N, 16.54. Found: C, 70.85; H, 4.98; N, 16.60%.

N-(furan-2-ylmethyl)-7-hydroxy-5-methyl-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (28)
Yield 63%; white solid (Dioxane); mp 298-300 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 2.35 (s, 3H, CH_3), 4.52 (d, 2H, CH_2-N, *J* = 6 Hz), 5.76 (s, 1H, pyrimidine-H), 6.32 (d, 1H, furan-H3, *J* = 3 Hz), 6.40 (t, 1H, furan-H4, *J* = 3 Hz), 6.87-7.64 (m, 6H, Ar-H + furan-H5), 8.21 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz; exchangeable with D_2O), 9.33(s, H, NH; exchangeable with D_2O), 11.57 (s, H, OH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3379 (NH), 2925 (aliph.CH) and 1658(C=O; amide); MS: m/z 363 (M^+, 35.30 %), 81(100%); Anal. Calcd for $C_{19}H_{17}N_5O_3$. Calcd: C, 62.80; H, 4.72; N, 19.27. Found: C, 62.76; H, 4.69; N, 19.23%.

N-(furan-2-ylmethyl)-7-hydroxy-5,6-dimethyl-2-(phenylamino)pyrazolo[1,5-a]pyrimidine-3-carboxamide (29)
Yield 63%; white solid (Dioxane); mp 290-292 ºC; ^{1}H NMR (300 MHz, DMSO-d_6) δ =1.98, 2.37 (2s, 6H, $2CH_3$), 4.51 (d, 2H, CH_2-N, *J* = 6 Hz), 6.31 (d, 1H, furan-H3, *J* = 3 Hz), 6.40 (t, 1H, furan-H4, *J* = 3 Hz), 6.87-7.64 (m, 6H, Ar-H + furan-H5), 8.18 (t, 1H, O=C-NH-CH_2, *J* = 6 Hz; exchangeable with D_2O), 9.43(s, H, NH; exchangeable with D_2O), 11.30 (s, H, OH; exchangeable with D_2O); IR (KBr, *v*, cm^{-1}): 3370, (NH), 2841 (aliph.CH) and 1655(C=O; amide); MS: m/z 377 (M^+, 54.21%), 64 (100%); Anal. Calcd for $C_{20}H_{19}N_5O_3$. Calcd: C, 63.65; H, 5.07; N, 18.56. Found: C, 63.60; H, 5.02; N, 18.60%.

Antimicrobial evaluation

Antimicrobial screening
The disks of Whatman filter paper were prepared with standard size (6.0 mm diameter) and kept into 1.0 Oz screw capped wide mouthed containers for sterilization. These bottles were kept into hot air oven at a temperature of 150 ºC. Then, the standard sterilized filter paper disks impregnated with a solution of the test compound in DMF (100 µL, 5 mg/mL) were placed on nutrient agar plate seeded with the appropriate test organism in triplicates. Standard concentrations of 10^6 CFU/mL (Colony Forming Units/mL) and 10^4 CFU/mL were used for antibacterial and antifungal assay, respectively. Pyrex glass Petri dishes (9 cm in diameter) were used and two disks of filter paper were inoculated in each plate. The utilized test organisms were *S. aureus*, *S. epidermidis* and *B. subitilis* as examples of Gram-positive bacteria and *P. aeruginosa*, *P. vulgaris* and *K. pneumonia* as examples of Gram-negative bacteria. They were also evaluated for their *in vitro* antifungal potential against *A. fumigatus*, *A. clavatus* and *G. candidium* fungal strain. Ampicillin and gentamycin were used as standard antibacterial agents; while amphotericin B was used as standard antifungal agent. DMF alone was used as control at the same above-mentioned concentration and due to this, there was no visible change in bacterial growth. The plates were incubated at 37 ºC for 24 hrs for bacteria and for 48 hrs at 25 ºC for fungi. The mean zone of inhibition measured in mm ± standard deviation beyond well diameter (6 mm) produced on a range of environmental and clinically pathogenic microorganisms. Compounds that showed significant growth inhibition zones using the twofold serial dilution technique, were further evaluated for their minimal inhibitory concentrations (MICs).

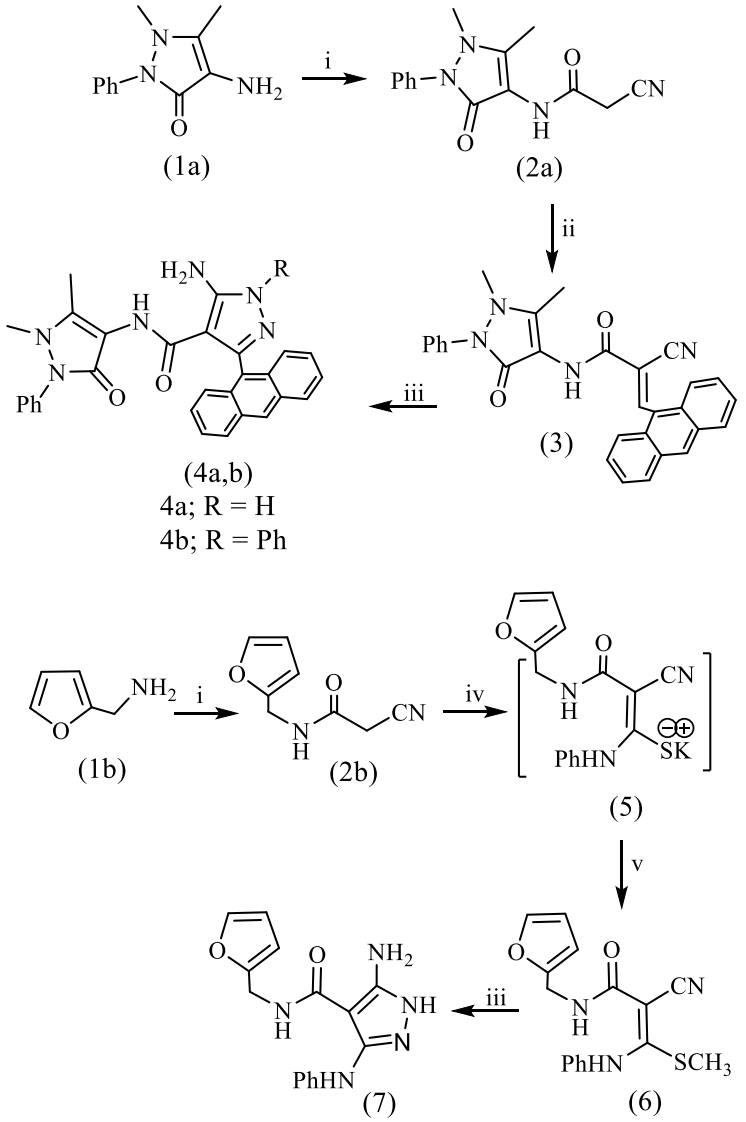
Minimal inhibitory concentration (MIC) measurement
The micro dilution susceptibility test in Müller-Hinton Broth (Oxoid) and Subouraud Liquid Medium (Oxoid) was used for the determination of antibacterial and antifungal activity, respectively. Stock solutions of the tested compounds, ampicillin, gentamycin, amphotericin B and sulfisoxazole were prepared in DMF at concentrations 1000 µg/mL. Each stock solution was diluted with standard method broth (Difco) to prepare serial twofold dilutions in the range of 500-0.007 µg/ mL. 10 mL of the broth containing about 10^6 CFU/mL of test bacteria or 10^4 CFU/mL of the test fungus was added to each well of 96-well microtiter plate. The sealed microplates were incubated at 37 ºC for 24 hrs for antibacterial activity and at 25 ºC for 48 hrs for antifungal activity in a humid chamber. At the end of the incubation period, the minimal inhibitory concentrations (MIC) values were recorded as the lowest concentrations of the substance that had no visible turbidity. Control experiments with DMF and inoculated media were run parallel to the test compounds under the same conditions.

RESULTS AND DISCUSSION

Chemistry

The starting materials **2a**,**b** were achieved in a good yield (90%) by the solvent free reaction of 4-aminoantipyrine and /or furan-2-ylmethanamine (**1a**,**b)** with ethyl cyanoacetate [27]. *Knoevenagel* condensation of compound **2a** with anthracene-9-carbaldehyde in refluxing ethanol containing a catalytic amount of piperidine produced a single stereoisomer, identified as (*E*)-3-(anthracen-9-yl)-2-cyano-*N*-(1,5-dimeth-yl-3-oxo-2-phenyl-2,3-dihydro-1H-pyrazol-4-yl)acrylamide (**3**). The *E*-configuration of compound **3** was assigned based on its ^{1}H NMR spectrum which displayed a downfield singlet signal at δ 9.15 ppm due to the olefinic (CH) proton that agrees with the chemical shift of the olefinic proton of similar structure, (*E*)-*N*-(pyridin-2-yl)-2-cyano-3-phenylprop-2-enamide, confirmed by X-ray analysis [28]. Treatment of acrylamide **3** with hydrazine hydrate and/ or phenylhydrazine in

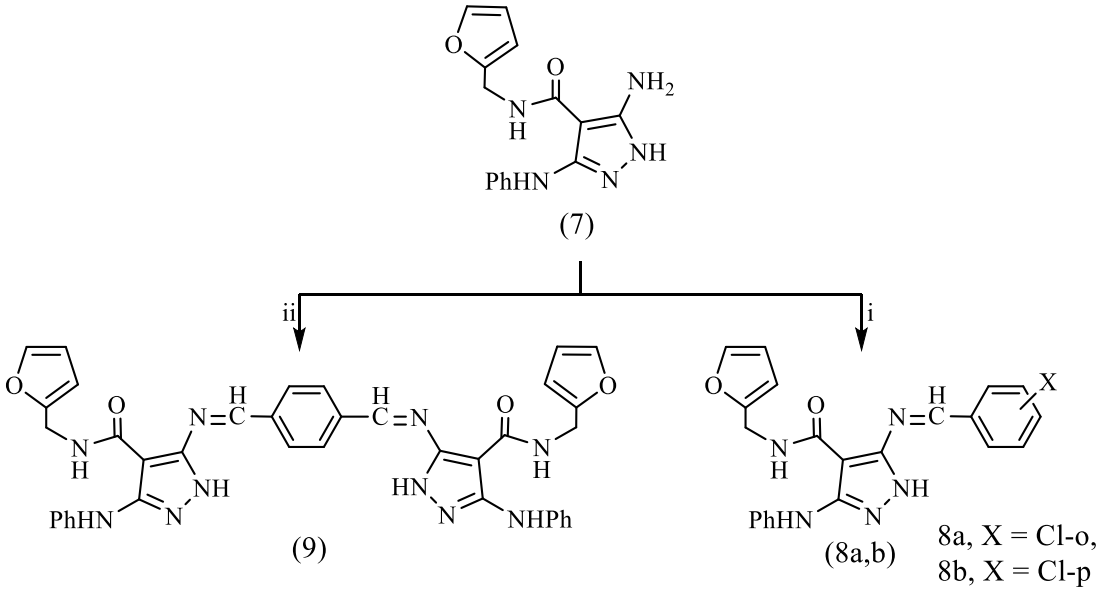
ethanol under reflux, furnished in each case a single product identified as 5-aminopyrazole derivatives **4a,b** (scheme 1). ^{1}H NMR spectrum of **4a** in DMSO-d_6 displayed singlet signals at δ 2.30, 3.42 ppm characteristic of two methyl protons and three D_2O exchangeable signals at 7.01, 9.44 and 10.07 ppm corresponding to NH_2 and 2NH protons, respectively. Mass spectrum of compound **4b** revealed a molecular ion peak at m/z = 564 (M^+, 25.4%) which is characteristic of the molecular formula $C_{35}H_{28}N_6O_2$. An efficient three-component, two-step "catch and release" synthesis of 5-aminopyrazoles was reported. Thus, treatment of 2-cyano-*N*-(furan-2-ylmeth-yl)acetamide (**2b**) with phenyl isothiocyanate in DMF in the presence of potassium hydroxide at room temperature gave the non-isolated adduct **5** which was then treated with dimethylsulfate to afford the 2-cyano-*N*-(furan-2-ylmethyl)-3-(methylthio)-3-(phenylamino)acrylamide (**6**). Reaction of ketene *N*, S-acetal **6** with hydrazine hydrate in refluxing ethanol furnished 5-aminopyrazole derivative **7** (scheme 1). The isolated product **7** was confirmed by analytical and spectral data. ^{1}H NMR (DMSO-d_6) spectrum showed doublet and singlet signals at δ 4.41, 5.84 ppm characteristic for $\underline{CH_2}$-NHand NH_2 protons in addition to, two singlet signals at δ 7.53, 11.06 ppm for 2 NH protons with triplet signals at δ 8.83 ppm characteristic of the amide group proton.^{13}C NMR (DMSO-d_6) of compound **7** revealed signals at δ 35.09 (CH_2), 106.40, 164.45, 164.47 (pyrazole carbons) in addition to, signal at 152.84 ppm (C=O).



Scheme 1. Synthetic route to 5-aminopyrazoles

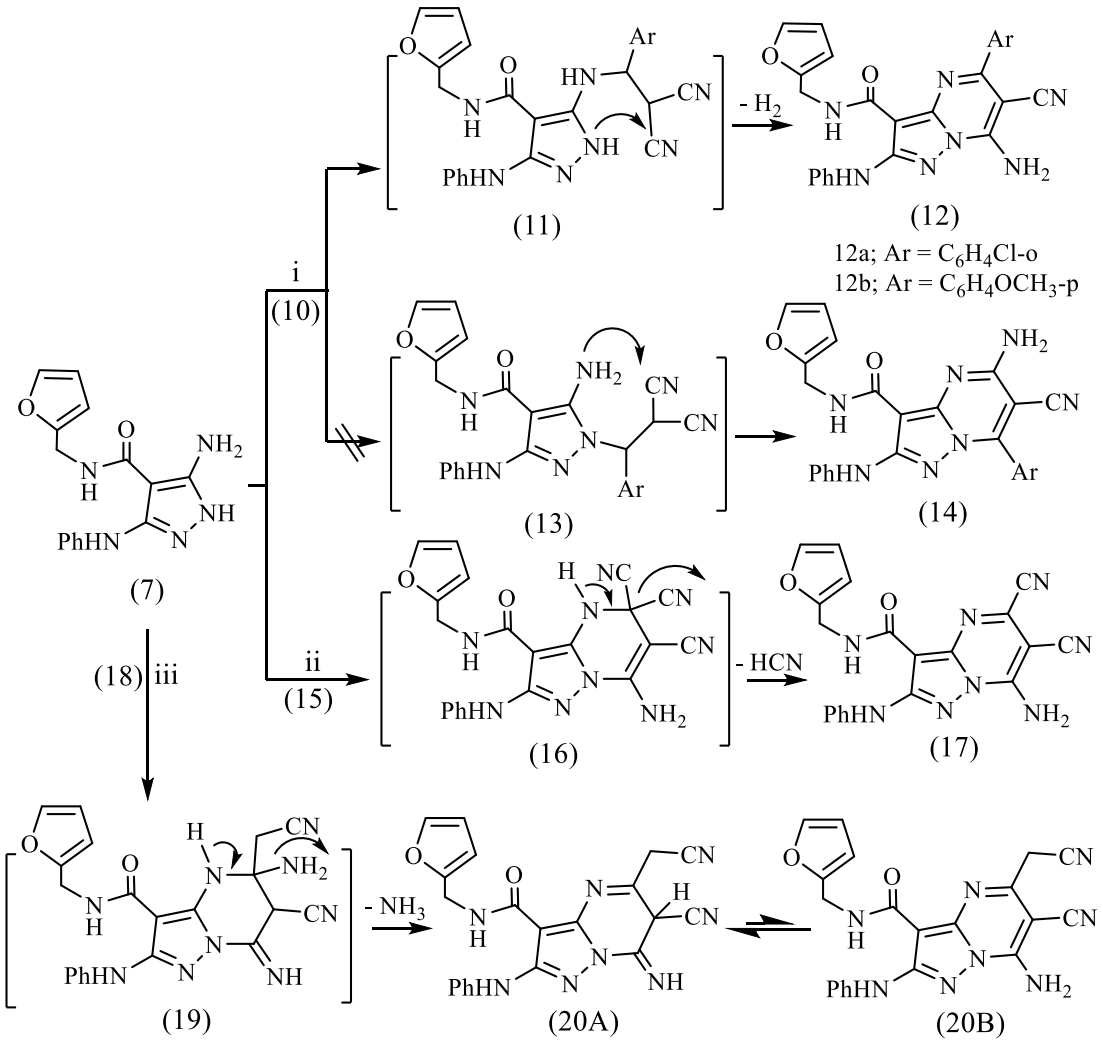
To investigate the structure reactivity relationship with respect to antimicrobial properties, the reactivity of 5-amino-*N*-(furan-2-ylmethyl)-3-(phenylamino)-1H-pyrazole-4-carboxamide (**7**) toward some electrophilic reagents was investigated. Schiff base derivatives were reported to possess significant biological activities and new series

have been tested for their antitumor, antimicrobial, and antiviral activities [29, 30]. In the light of these facts, 5-(benzylideneamino)pyrazole derivatives **8a**,**b** were obtained by condensation of 5-aminopyrazole **7** with aromatic aldehydes in ethanol in the presence of piperidine under reflux.^{1}H NMR (DMSO-d_6) spectra of **8a,b** showed singlet downfield CH=N signals at δ 8.14-8.64 ppm. Also, bis(azomethine)pyrazole derivative **9** was achieved by condensation of compound **7** with terephthalaldehyde (2 : 1 molar ratio) in refluxing ethanol in the presence of piperidine (scheme 2). A molecular ion peak at *m/z* = 692 (12.5%) was observed in the mass spectrum of compound **9** with a base peak at *m/z*= 64, which is compatible with its molecular formula of $C_{38}H_{32}N_{10}O_4$.



Scheme 2. Synthetic route to azomethine derivatives

5-Aminopyrazoles are versatile reagents and have been extensively used as synthetic starting materials for the synthesis of several polysubstituted fused pyrazoles of potential biological activity [31-34]. Thus, the reactivity of 5-aminopyrazole **7** towards some activated nitriles was investigated as an alternative route to obtain pyrazolo[1,5-*a*]pyrimidine derivatives. Reaction of 5-aminopyrazole **7** with α-cinnamonitriles**10a**,**b** in refluxing ethanol in the presence of a catalytic amount of piperidine yielded a single product for which structure **12** or **14** seemed possible. Structure of **12** appears more likely than **14** on the basis of single crystal X-ray structure analysis [35] and HMBC-^{15}N[36].^{1}H NMR spectrum of **12b** in DMSO-d_6 revealed singlet signals at δ 3.86, 8.96 ppm characteristic of OCH_3 and amino group protons. ^{13}C NMR spectrum of **12b** in DMSO-d_6 revealed signals at δ = 53.14 (OCH_3), 88.65, 149.56, 160.59, 161.35 (pyrimidine-C), 106.90, 158.01 (pyrazole-C), 152.80 (CN), 163.57 (C=O) ppm. The formation of **12** is assumed to proceed via an initial *Michael* addition of the oxocyclic NH_2 in **7** to the activated double bond in **10** to yield the non-isolable *Michael* adduct **11**, followed by intramolecular cyclization and aromatization by loss of hydrogen molecule. Similarly, pyrazolo[l,5-*a*]pyrimidine derivative **17** was obtained via the reaction of compound **7** with tetracyanoethylene (**15**) in refluxing dioxane in the presence of a catalytic amount of piperidine. Infrared spectrum of **17** showed the characteristic absorption bands at 3422, 3358, 2229, assignable for NH_2 and C≡N groups. The mass spectrum of compound **17** revealed a molecular ion peak at m/z = 398 (55.2%) which is characteristic of the molecular formula $C_{20}H_{14}N_8O_2$ (Chart 1). Formation of pyrazolopyrimidine**17** takes place as depicted in scheme 3, via the formation of *Michael* adduct **16**, followed by intramolecular cyclization with HCN elimination. Interaction of 5-aminopyrazole **7** with 1,1,3-tricyano-2-aminopropene (**18**) in ethanol in the presence of piperidine afforded two possible isomeric products **20A** and **20B.** Structure **20A** appears more likely than **20B** on the basis of spectral data. The infrared spectrum of the isolated product afforded intense absorption bands at 3302 (NH) and 2213 cm^{-1} (C≡N).^{1}H NMR spectrum of **20A** showed three D_2O exchangeable signals at δ 8.55, 10.16, 12.49 attributed to 3NH protons. In addition to, singlet signal at δ 7.61 ppm characteristic of pyrimidine-H proton. The reaction proceeds through the formation of *Michael* adduct **19** as intermediate, followed by the ammonia molecule elimination (scheme 3).



Scheme 3. Synthetic route to pyrazolo[1,5-*a*]pyrimidine

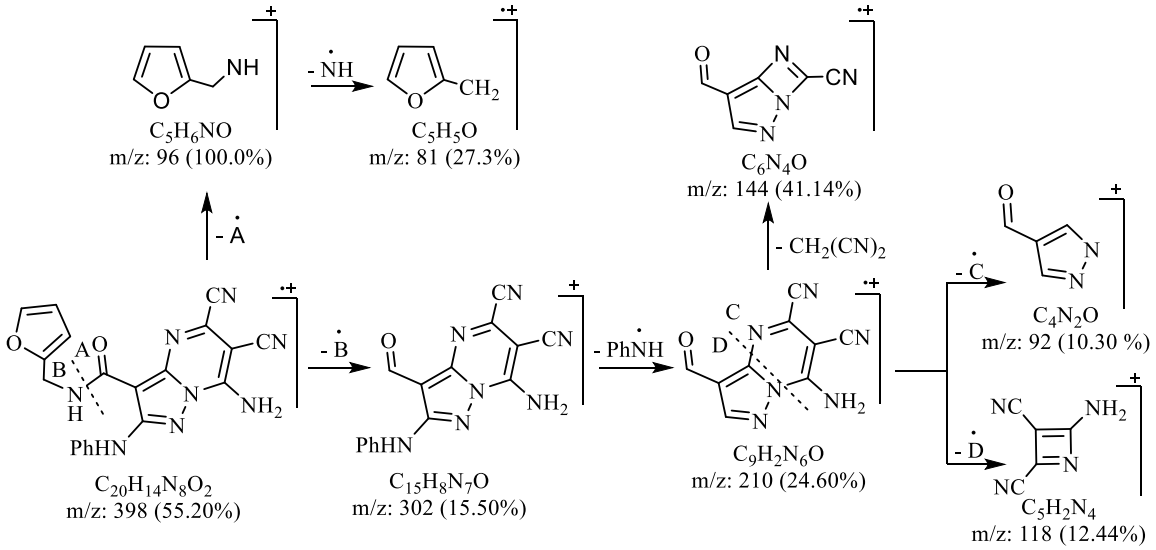
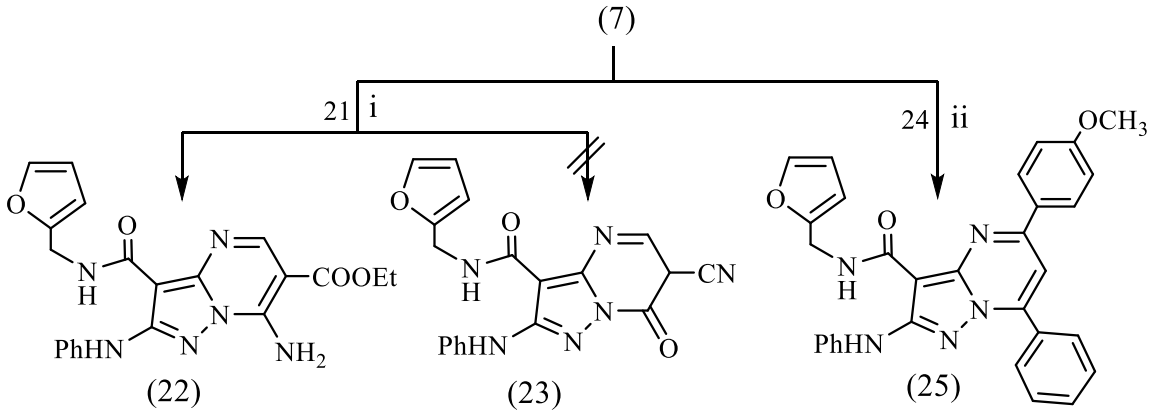


Chart 1: Fragmentation pattern of compound 17

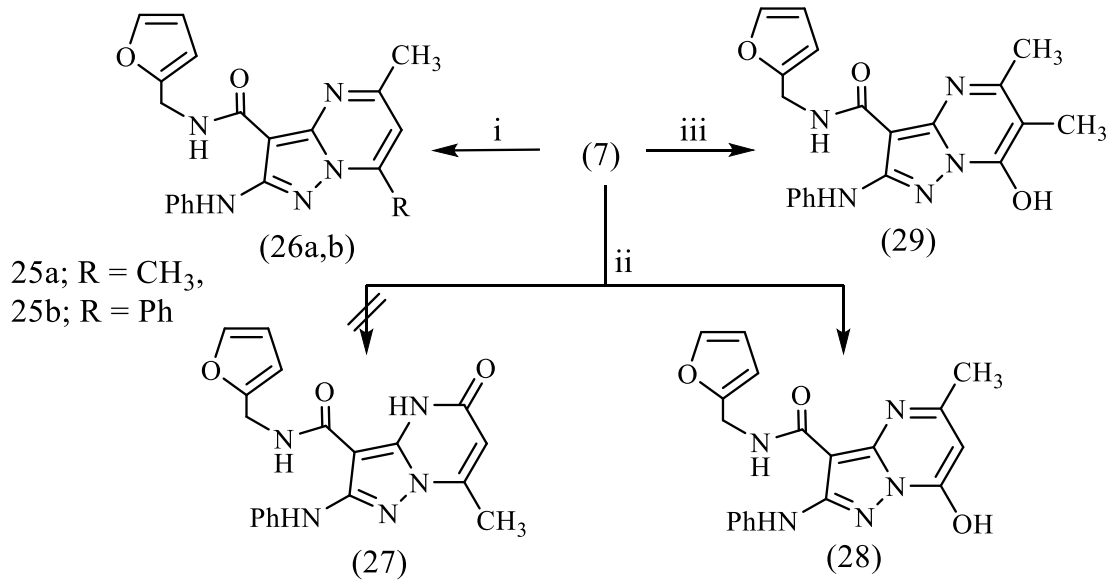
Reaction of 5-aminopyrazole **7** with ethyl 2-cyano-3-ethoxyacrylate (**21**) in ethanolic piperidine solution, afforded pyrazolo[1,5-*a*]pyrimidine derivative **22** rather than **23** (Scheme 4). ^{1}HNMR spectrum of **22** revealed triplet and quartet signals at 1.36, 4.35 assignable for ester moiety with two singlet signals at, 8.69, 9.57ppmdue to CH-pyrimidine and amino protons. Furthermore, the reaction of 5-aminopyrazole **7** with α,β-unsaturated carbonyl derivative **24** in ethanolic piperidine solution under reflux, afforded pyrazolo[1,5-*a*]pyrimidine derivative **25**. Its mass spectrum revealed a molecular ion peak at m/z = 515 (M^+, 12.24%) and a base peak was observed in the spectrum at *m/z* = 517 (M^{+2}), which is compatible with its molecular formula of $C_{31}H_{25}N_5O_3$.



i = EtOCH=C(CN)CO_2Et, EtOH/Pip.,
ii =*p*-CH_3O-C_6H_4CH=CHCOC_6H_5, EtOH/Pip.

Scheme 4. Synthetic route to pyrazolo[1,5-*a*]pyrimidine

The general literature procedure for the synthesis of pyrazolo[1,5-*a*]pyrimidines involves cyclocondensation of aminopyrazoles with reagents having 1,3-electrophilic centers such as *β*-diketones [37-40]. Thus, cyclocondensation of 5-aminopyrazole **7** with acetylacetone and / or benzoylacetone in boiling glacial acetic acid gave the pyrazolo[l,5-*a*]pyrimidine derivatives **26a** and **26b** - evidence for assigned structures being provided by analytical and spectral data. For example, ^{13}C NMR (DMSO-d_6) of compound **26a** revealed signals at δ 17.11, 24.63 (2CH_3), 107.27, 156.52, 161.42, 164.38 (pyrimidine carbons) in addition to, singlet signal at 152.78 ppm (C=O). The mass spectrum of compound **26a** revealed a molecular ion peak at m/z = 361 (M+, 39.42%), and a base peak was observed in the spectrum at m/z = 265, which is compatible with its molecular formula of $C_{20}H_{19}N_5O_2$ (Chart 2). Similarly, treatment of 5-aminopyrazole **7** with ethyl acetoacetate in boiling glacial acetic afforded two possible isomeric products **27** and **28**. Structure **27** was excluded on the basis of spectral data and analogy with previous work [41-44]. ^{1}H NMR spectrum of **28** in DMSO-d_6 displayed singlet signals at δ 2.35, 5.76 ppm characteristic of methyl protons and pyrimidine-H, in addition to, three D_2O exchangeable signals at δ 8.21, 9.33 and 11.57 ppm attributable to 2NH and OH protons. The mass spectrum of compound **28** revealed a molecular ion peak at *m/z* = 363 (M+, 35.3%), and a base peak was observed in the spectrum at *m/z* = 81, which is compatible with its molecular formula of $C_{19}H_{17}N_5O_3$. Finally, pyrazolo[l,5-*a*]pyrimidine derivative **29** could be achieved in excellent yield via cyclocond-ensation of compound **7** with ethyl 2-methyl-3-oxo-butanoate in glacial acetic acid under reflux condition (Scheme 5).The chemical structure of **29** was supported on the basis of elemental analysis and spectral data. Its ^{1}H NMR spectrum in DMSO-d_6 displayed singlet signals at δ 1.98 and 2.37 ppm due to 2 CH_3 protons in addition to three D_2O exchangeable signals at δ 8.18, 9.43 and 11.30 assignable to 2NH and OH protons.



Scheme 5. Synthetic route to pyrazolo[1,5-*a*]pyrimidine

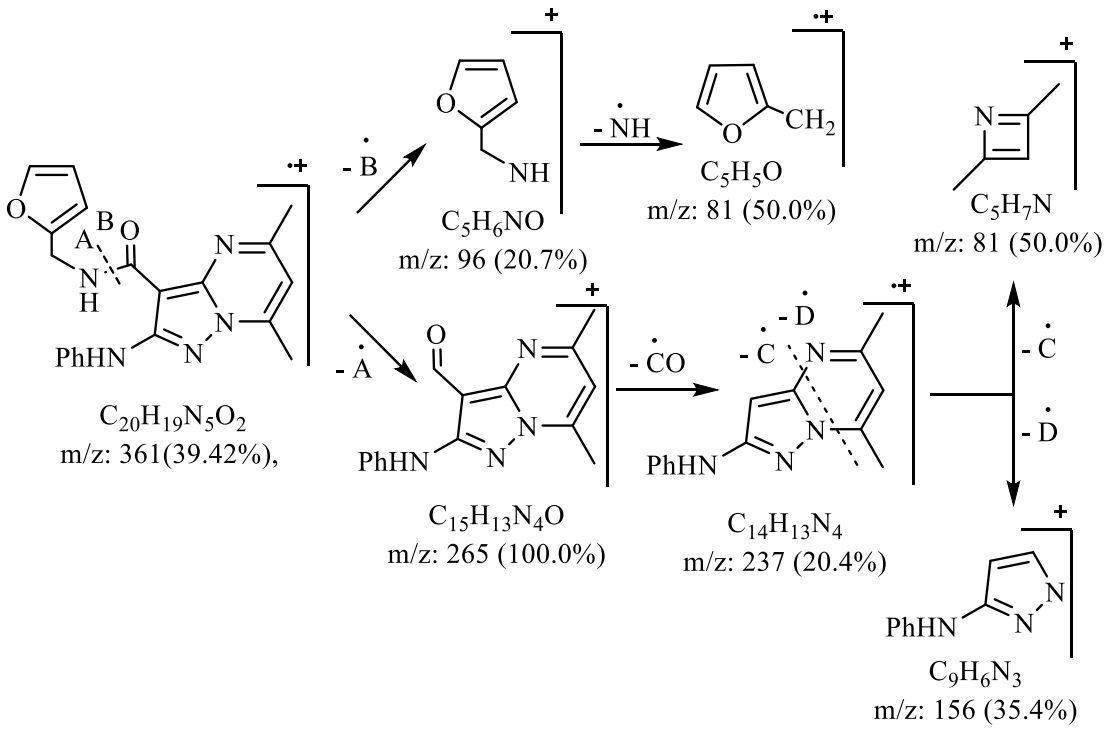


Chart 2: Fragmentation pattern of compound 26a

Antimicrobial evaluation

The antimicrobial screening and minimal inhibitory concentrations of the tested compounds were carried out at the Regional Center for Mycology and Biotechnology, Al-Azhar University, Cairo, Egypt. Eighteen of the newly synthesized target compounds were evaluated for their *in vitro* antibacterial activity against *Staphylococcus aureus* (RCMB 010027), *Staphylococcus epidermidis* (RCMB 010024) and *Bacillis subitilis* (RCMB 010063) as examples of Gram-positive bacteria and *Pseudomonas aeruginosa* (RCMB 010043), *Proteous vulgaris* (RCMB 010085) and *Klebsiella pneumonia* (RCMB 010093) as examples of Gram-negative bacteria. They were also evaluated for their

in vitro antifungal potential against *Aspergillus fumigatus, Aspergillus clavatus* and *Geotricum candidium* fungal strain.

Agar-diffusion method [45] was used for the determination of the preliminary antibacterial and antifungal activity. Ampicillin, Gentamycin and Amphotericin B were used as reference drugs. The results were recorded for each tested compound as the average diameter of inhibition zones (IZ) of bacterial or fungal growth around the disks in mm. The minimum inhibitory concentration (MIC) measurement was determined for compounds and showed significant growth inhibition zones using twofold serial dilution method [46].The MIC (µg/mL) and inhibition zone diameters values are recorded in Table 1.

Compounds no.	Gram-positive bacteria			Gram-negative bacteria			Fungi		
	S. aureus	*S. epidermidis*	*B. subitilis*	*P. aeruginosa*	*P. vulgaris*	*K. pneumonia*	*A. fumigatus*	*A. clavatus*	*G. candidium*
3	18.3	20.1	20.1	*NA	18.3	19.6	16.3	18.2	19.3
4a	17.3	19.2	19.9	NA	17.4	18.2	15.6	17.4	19.4
4b	20.6(3.9)	21.8(0.98)	22.3(0.98)	NA	21.3(1.95)	22.4(0.98)	21.4(1.95)	23.6(0.98)	24.2(0.49)
6	17.3	19.2	19.8	NA	15.3	17.2	16.3	18.2	19.3
7	15.2	17.2	18.1	NA	16.8	18.1	14.6	14.8	16.2
8a	17.6(3.9)	19.2(0.98)	20.3(0.24)	NA	16.4(3.9)	18.1(0.98)	19.3(1.95)	19.8(0.98)	20.4(0.98)
8b	20.8(3.9)	22.4(0.98)	22.9(0.98)	NA	20.4(3.9)	21.3(1.95)	21.2(1.95)	23.2(0.98)	24.2(0.98)
9	22.3(0.98)	25.4(0.49)	26.4(0.24)	NA	22.1(0.98)	26.3(0.49)	20.2(3.9)	22.3(0.98)	23.4(0.98)
12a	NA	NA	NA	NA	NA	NA	NA	NA	NA
12b	14.3	14.7	16.3	NA	18.2	19.4	NA	NA	NA
17	20.6(1.95)	21.4(2.94)	22.9(0.98)	NA	21.7(1.95)	22.1(1.95)	19.9(3.9)	21.9(1.95)	20.8(0.49)
20	15.2	16.3	18.2	NA	15.2	16.3	14.3	16.2	17.3
22	19.6	20.3	21.4	NA	18.6	19.2	17.3	18.8	19.2
25	14.9	15.8	19.1	NA	16.5	17.1	15.4	17.2	17.5
26a	15.2	18.3	20.4	NA	18.3	20.2	14.3	16.1	16.9
26b	17.6	19.2	20.3	NA	16.4	18.1	19.3	19.8	20.4
28	20.3	21.2	22.3	NA	21.4	21.9	15.6	18.2	19.4
29	16.8	19.1	20.2	NA	17.3	19.1	15.3	17.2	18.1
Ampicillin	28.9(0.24)	25.4(0.49)	36.6(0.24)	*NT	NT	NT	NT	NT	NT
Gentamycin	NT	NT	NT	(3.9)19.9	25.4(0.98)	26.3(0.49)	NT	NT	NT
Amphotericin B	NT	NT	NT	NT	NT	NT	23.7(0.98)	21.9(0.98)	25.4(0.49)

**NA: no activity, *NT: not tested*

Table1: Antimicrobial inhibition zone in mm and minimal inhibitory concentrations (MIC, µg/ml, between brackets) of some new synthesized compounds

Antibacterial activity

Regarding the Antibacterial activities of acrylamides, acrylamide **3** was more potent than **6** against both Gram-positive and Gram-negative bacteria (fig.1).The pyrazoles **4b** and **4a** bearing antipyrine moiety displayed better antibacterial activity than pyrazole **7** bearing furan moiety when compared with ampicillin and gentamycin. Pyrazole **4b** showed relatively good growth inhibitory profiles versus both of *S. epidermidis* (MIC 0.98µg/mL), *P. vulgaris* (MIC 1.95µg/mL) and *K. pneumonia* (MIC 0.98µg/mL), which was about 50% of the activity of ampicillin and gentamycin. When pyrazole **7** converted into Bis(azomethine)pyrazole derivative **9**, it showed equal activity to ampicillin versus both of *S. epidermidis* (MIC 0.49µ/mL) and *B. subitilis* (MIC 0.24µ/mL), it also showed equipotent to gentamycin versus both of *P. vulgaris* (MIC 0.98µ/mL) and *K. pneumonia* (MIC 0.49µ/mL). Azomethine derivatives **8a** displayed equipotent to ampicillin against *B. subitilis* (MIC 0.24µ/mL), but showed relatively good growth inhibitory profiles against both of *S. epidermidis* (MIC 0.98µg/mL) and *K. pneumonia* (MIC 0.98µg/mL), which was about 50% of the activity of ampicillin and gentamycin. Also, Azomethine **8b** has more activities than 5-amino-pyrazole **7** when compared with ampicillin and gentamycin. Regarding the activity of pyrazolo[1,5-*a*]pyrimidines, the results revealed that, pyrazolo[1,5-*a*]pyrimidine **17** contain two cyano groups recorded higher activity than all other pyrazolo[1,5-*a*]pyrimidine derivatives when compared with ampicillin and gentamycin. Compounds **20**, **22**, **25**, **26a**, **28** and **29** showed relatively moderate growth inhibitory profiles versus both Gram-positive and Gram-negative bacteria. While, pyrazolo[1,5-*a*]pyrimidines **12b** and **26b** revealed weak activities when compared with ampicillin and gentamycin. Unfortunately, pyrazolo[1,5-*a*]pyrimidine **12a** showed completely inactive toward Gram-positive and Gram-negative bacteria (fig.2). On the other hand, all the synthesized compounds showed were completely inactive toward *P. aeruginosa* (RCMB 010043) compared to gentamycin.

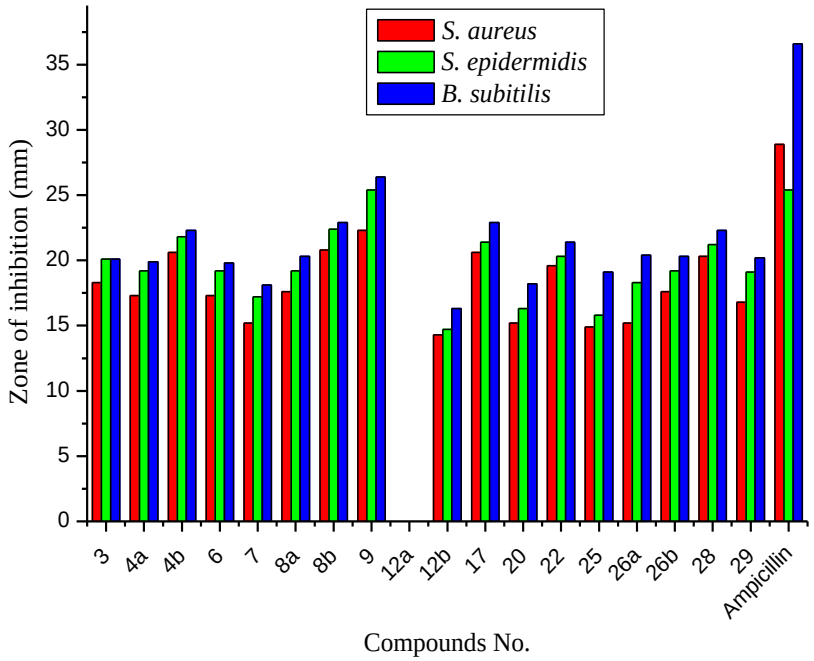


Fig.1: Preliminary antibacterial activity against Gram-positive bacteria

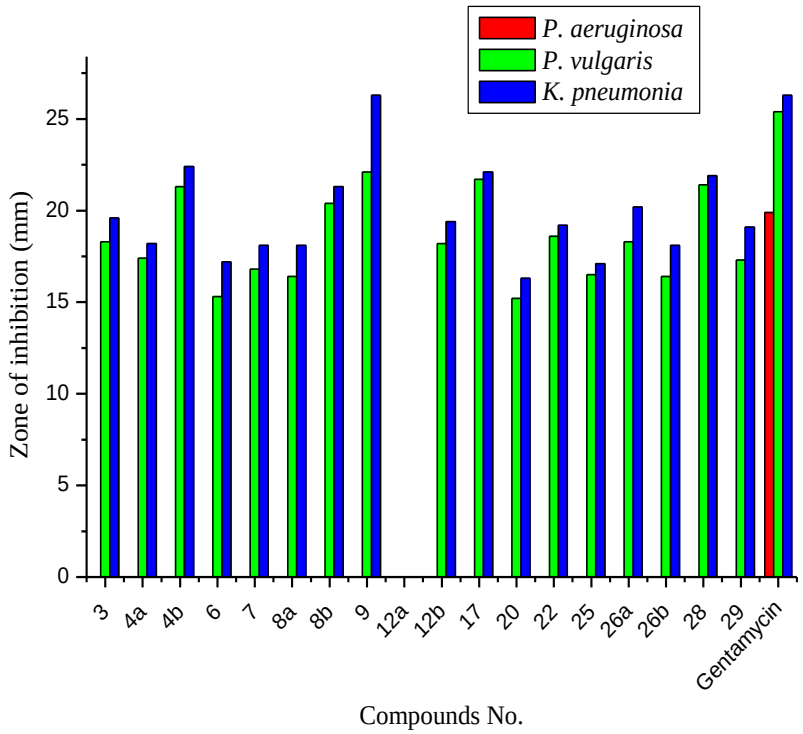


Fig.2: Preliminary antibacterial activity against Gram-negative bacteria

Antifungal activity

Both acrylamides **3** and **6** showed the same moderate activity when compared to amphotericine B in inhibiting the growth of *A .fumigatus, A. clavatus* and *G. candidium* fungal strain. Pyrazole **4b** displayed *invitro* antifungal activity equipotent to amphotericin B against both of *A. clavatus* (MIC 0.98 µ/mL), and *G. candidum* (MIC 0.49µ/mL), but revealed 50% of the activity of *A. fumigatus* (MIC 1.95µ/mL) when compared with amphotericin B.

Bis(azomethine)pyrazole **9** displayed equipotent to amphotericin B versus *A. clavatus* (MIC 0.98 µ/mL) and showed 50% of the activity of amphotericin B in inhibiting the growth of *G. candidium* (MIC 0.98 µ/mL). Azomethine derivatives **8a** and **8b** displayed 50% of the activity of amphotericin B versus the inhibiting of *A. fumigatus* (MIC 1.95µ/mL) and *G.candidium* (MIC 0.98 µ/mL), but they were equipotent to amphotericin B against *A. clavatus* (MIC 0.98 µ/mL). Pyrazolo[1,5-*a*]pyrimidine **17** was equipotent to amphotericin B in inhibiting the growth of *G. candidium* (MIC 0.49 µ/mL).Other pyrazolo[1,5-*a*]pyrimidine derivatives **20**. **22**, **25**, **26a**,**b**, **28** and **29** displayed the moderate activity comparable to amphotericin B. Unfortunately, all the synthesized compounds **12a** and **12b** showed were completely inactive toward *A. fumigatus*, *A. clavatus* and *G. candidium* fungal strain (fig. 3).

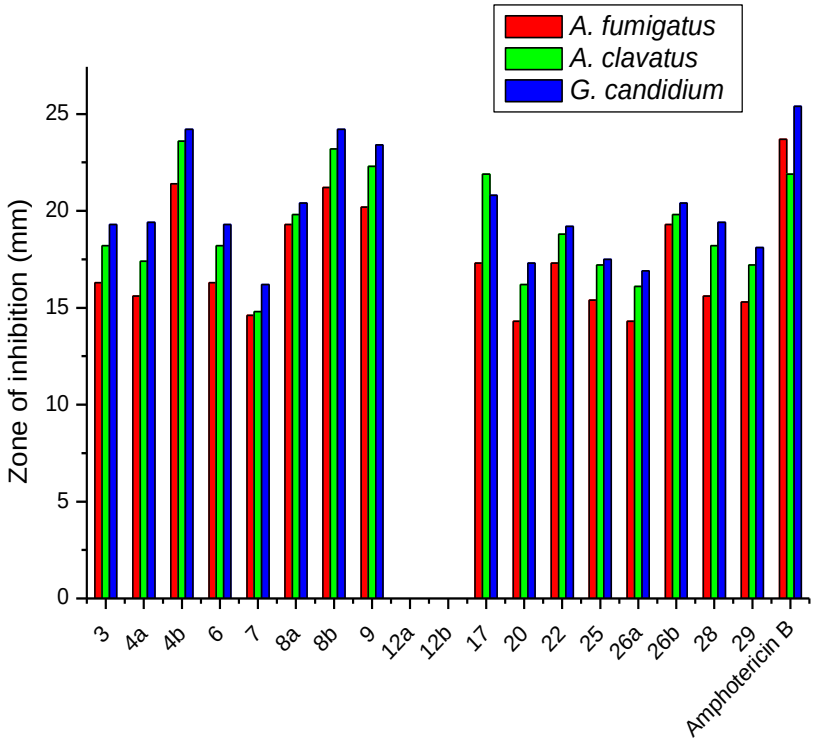


Fig. 3:Preliminary antifungal activity of the synthesized compounds

CONCUSION

The objective of this study was to synthesize and investigate the antimicrobial activities of some new 5-aminopyrazole and pyrazolo[1,5-*a*]pyrimidine derivatives. My aim has been verified by the synthesis of 5-aminopyrazole, 5-(benzylideneamino)pyrazole, bispyrazole and pyrazolo[1,5-*a*]pyrimidine derivatives bearing antipyrine or furan moiety. The obtained results clearly reveal that 5-aminopyrazoles **4b** and **4a** bearing antpyrine moiety display antimicrobial activity better than pyrazole **7** bearing furan moiety. Conversation of pyrazole**7** into 5-(benzylideneamino)pyrazole **8a,b** and bispyrazole derivative **9** increase the potential activity. Some pyrazolo[1,5-*a*]pyrimidine derivatives record good potential activity.

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