RSC Advances

REVIEW

View Article Online
View Journal | View Issue

Cite this: *RSC Adv.*, 2026, **16**, 25459

The utility of pyrimidine derivatives in the construction of azolo[*d*]pyrimidine: part II-multi-nitrogen azole systems with potential biological applications

Ahmed H. Shamroukh * and Aymn E. Rashad

In this review (part II), we continue our compilation of studies highlighting the use of the pyrimidine ring as a key core precursor for the construction of azolo[*d*]pyrimidine scaffolds containing more than one nitrogen atom in the azole moiety. Fusion of the azole ring owing to the presence of multiple nitrogen atoms onto the pyrimidine nucleus affords structurally diverse regioisomeric systems, including pyrazolo[3,4-*d*]pyrimidines, pyrazolo[4,3-*d*]pyrimidines, imidazolo[4,5-*d*]pyrimidines (purines) and [1,2,3]triazolo[4,5-*d*]pyrimidines (8-azapurines), depending on the position of the nitrogen atom within the fused heterocycle. This review provides a comprehensive overview of recent advances during the last decade (2015–2026) in the synthesis and biological applications of fused azolo[*d*]pyrimidine scaffolds (Part II). In addition, the results presented in this review highlight the importance of various azolo[*d*]pyrimidine derivatives as promising scaffolds for the development of new therapeutic agents.

Received 29th March 2026
Accepted 27th April 2026

DOI: 10.1039/d6ra02611g

rsc.li/rsc-advances

Introduction

Building upon our previous study (Part I), which explored pyrimidine nucleus as a key building block for the synthesis of azolo[*d*]pyrimidine frameworks bearing mono-nitrogen azole rings,[1] this work extends that approach to the construction of azolo[*d*]pyrimidine scaffolds incorporating azole moieties containing more than one nitrogen atom, utilizing pyrimidine derivatives as versatile precursors. No doubt that the pyrimidine ring represents a fundamental heterocyclic scaffold in organic and medicinal chemistry owing to its presence in numerous biologically essential molecules, including nucleic acid bases, vitamins, and pharmaceuticals.[2–18] The electron-deficient aromatic character of pyrimidine, together with its versatile substitution patterns, renders it a valuable core precursor for the construction of fused-pyrimidine scaffolds.[19] In fact, the diversity of pyrimidine derivatives is further expanded through the fusion of the pyrimidine ring with other heterocyclic moieties leading to the formation of more bioactive molecules compared to the individual separated moieties. Among them, azolo[*d*]pyrimidines constitute an important class of bicyclic heterocyclic frameworks formed through the annulation of azole rings onto the pyrimidine nucleus (Fig. 1). The fusion of nitrogen-rich five-membered heterocycles, such as pyrazole, imidazole, and triazole, onto pyrimidine scaffolds has resulted in structurally diverse compounds exhibiting a broad spectrum of biological activities.[20–40]

Specifically, fusion of a pyrazole ring onto the pyrimidine nucleus affords structurally diverse regioisomers, including pyrazolo[3,4-*d*]pyrimidines and pyrazolo[4,3-*d*]pyrimidines, which have been extensively studied due to their structural resemblance to purine analogues and their notable bioactivities, particularly in kinase inhibition and anticancer drug discovery.[41–44]

Several FDA-approved drugs contain the pyrazolo-pyrimidine nucleus, reflecting its pharmaceutical importance (Fig. 2). For example, *allopurinol*, a pyrazolo[3,4-*d*]pyrimidine, was the first xanthine oxidase inhibitor approved by the U.S. Food and Drug Administration for the treatment of chronic gout and hyperuricemia.[45] Additionally, *ibrutinib*, a covalent Bruton's tyrosine kinase (BTK) inhibitor bearing a pyrazolo[3,4-*d*]pyrimidine core, has been approved for the treatment of several B-cell malignancies.[46] One of the most significant pharmacological applications of pyrazolo[4,3-*d*]pyrimidine derivatives is exemplified by *sildenafil* (Viagra®), a selective phosphodiesterase type 5 (PDE5) inhibitor widely used as an oral therapeutic agent for the treatment of male erectile dysfunction.[47] Consequently, a series of sildenafil analogues (R = Me and Et; R_1 = Me, Et, and –CH_2CH_2OH) was synthesized, and their inhibitory activities were evaluated. The results demonstrated enhanced inhibitory potency along with improved selectivity toward PDE5.[48]

Similarly, fusion of an imidazole ring with pyrimidine yields purine frameworks, key heterocycles that constitute the core of nucleosides, nucleotides, and numerous therapeutic agents

Photochemistry Department, Chemical Industries Research Institute, National Research Centre, 33 El Buhouth Street, P.O. Box 12622, Cairo, Egypt. E-mail: ahshamroukh@yahoo.com; Tel: +2021124477409

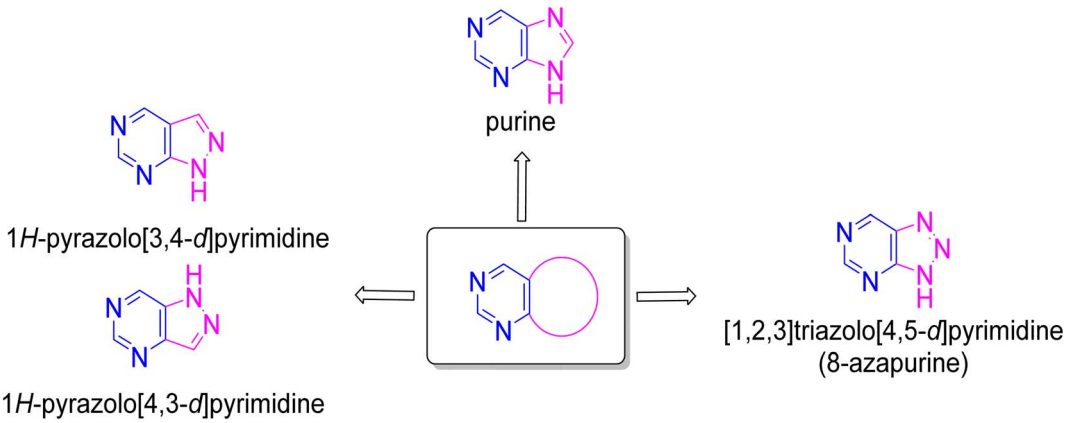


Fig. 1 Azolo[*d*]pyrimidines generated by combining pyrimidines at the *d* position with different heterocyclic rings such as pyrazole, imidazole, and triazole.

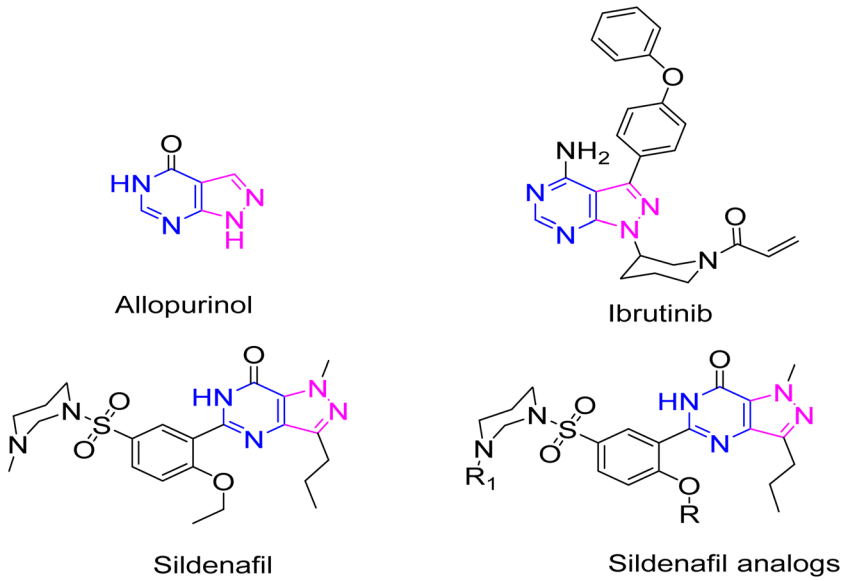


Fig. 2 FDA approved-drug containing a pyrazolopyrimidine moiety.

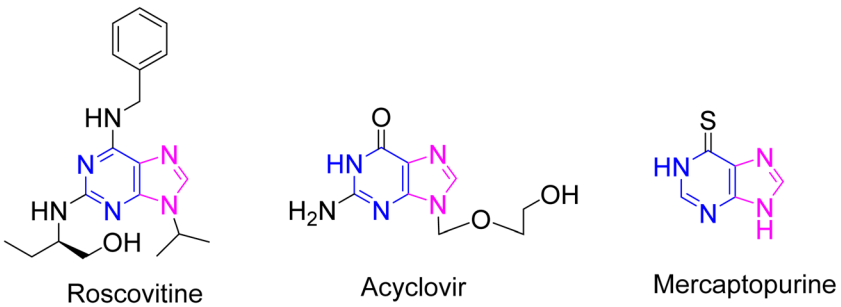


Fig. 3 Therapeutic agents containing a purine moiety.

(Fig. 3), such as *roscovitine*, a cyclin-dependent kinase (CDK) inhibitor with anticancer potential,[49] *acyclovir* (an antiviral nucleoside analogue),[50] and *mercaptopurine* (an antileukemic agent).[51]

Furthermore, the incorporation of a triazole ring into the pyrimidine nucleus affords [1,2,3]triazolo[4,5-*d*]pyrimidines (8-azapurines), a class of pharmacologically active heterocycles with diverse therapeutic potential.[34–41] Notable examples include *zaprinast*, a [1,2,3]triazolo[4,5-*d*]pyrimidine derivative

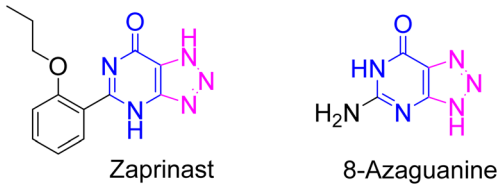


Fig. 4 Therapeutic agents containing a [1,2,3]triazolo[4,5-*d*]pyrimidine moiety.

investigated as a phosphodiesterase inhibitor that contributed to the development of subsequent PDE-targeting drugs,[52] and *8-azaguanine*, an early purine analog exhibiting antineoplastic activity (Fig. 4).[53] Beyond anticancer applications, triazolopyrimidine frameworks have been broadly reviewed for their wide spectrum of biological activities, including antimicrobial,[54] antiviral,[35,55,56] antiplatelet,[57] antimalarial,[58] antiepileptic,[59] and anti-Alzheimer's activities,[60] emphasizing their versatility as therapeutic scaffolds.

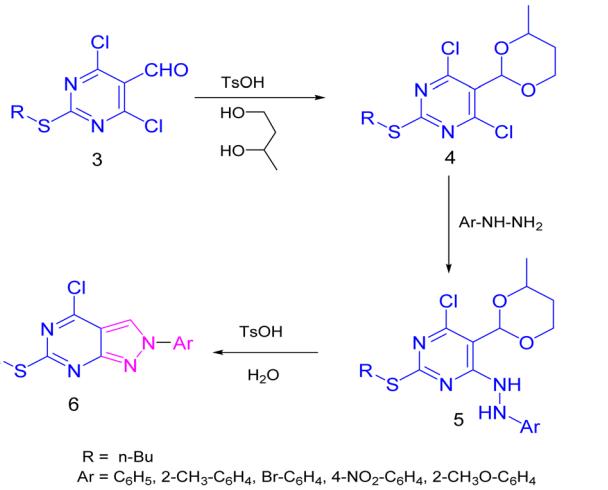
The present review summarizes synthetic strategies leading to these frameworks and emphasizes their importance in heterocyclic and medicinal chemistry during the last decade (2015–2026).

Pyrazolo[*d*]pyrimidines

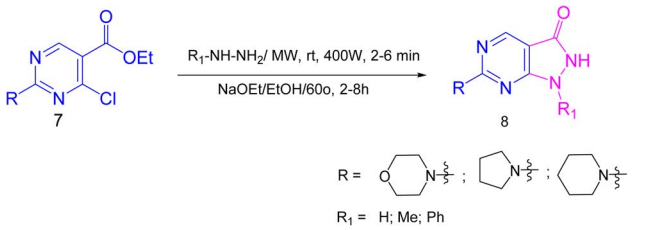
Pyrazolo[*d*]pyrimidines are bicyclic heterocycles formed by the fusion of a pyrazole ring with a pyrimidine nucleus, resulting in a rigid, planar framework. Depending on the mode of annulation, two regioisomeric systems can be obtained: pyrazolo[3,4-*d*]pyrimidine and pyrazolo[4,3-*d*]pyrimidine. Numerous synthetic methodologies have been developed for the preparation of pyrazolo[4,3-*d*]pyrimidines; however, both classical and contemporary approaches predominantly rely on pre-substituted pyrazole derivatives as key starting materials.

In contrast, strategies that construct the pyrazolo[4,3-*d*]pyrimidine scaffold directly from pyrimidine substrates remain relatively limited.[61–63] This limitation is largely attributed to the inherent challenges associated with the regioselective activation of the electron-deficient pyrimidine ring, as well as the controlled formation of the N–N bond during pyrazole annulation.

Accordingly, this section focuses exclusively on synthetic approaches that utilize the pyrimidine ring as the principal core precursor for the construction of pyrazolo[3,4-*d*]pyrimidine frameworks.



Scheme 2 Preparation of pyrazolo[3,4-*d*]pyrimidine from protected 4,6-dichloropyrimidine-5-carbaldehydes.

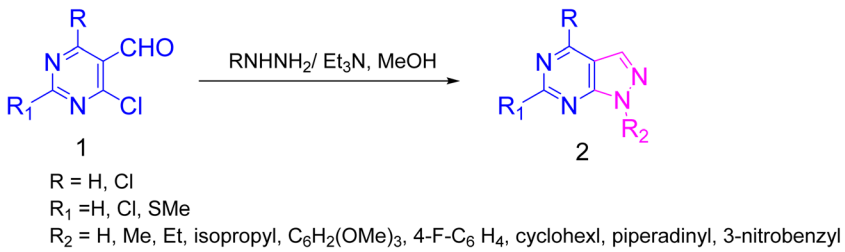


Scheme 3 Preparation of pyrazolo[3,4-*d*]pyrimidine from ethyl 4-chloropyrimidine-5-carboxylates.

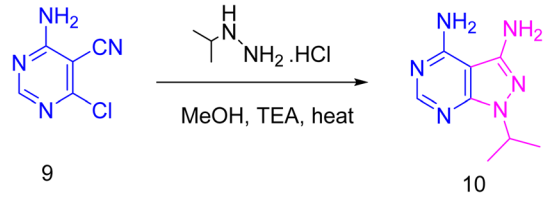
Synthesis of pyrazolo[3,4-*d*]pyrimidine derivatives

6-Chloropyrimidine-5-carbaldehyde (**1**) has been widely employed in the literature for the preparation of 1*H*-pyrazolo[3,4-*d*]pyrimidine derivatives **2**. Thus, condensation of **1** with various hydrazine derivatives in the presence of triethylamine afforded the corresponding pyrazolo[3,4-*d*]pyrimidine derivatives **2** bearing different substituents (Scheme 1).[64–70]

According to Ofitserova *et al.*, protection of the aldehyde group in 4,6-dichloropyrimidine-5-carbaldehydes alters the



Scheme 1 Preparation of pyrazolo[3,4-*d*]pyrimidines from 6-chloropyrimidine-5-carbaldehyde.



9 10

Scheme 4 Preparation of pyrazolo[3,4-*d*]pyrimidine from 6-chloropyrimidine-5-carbonitrile.

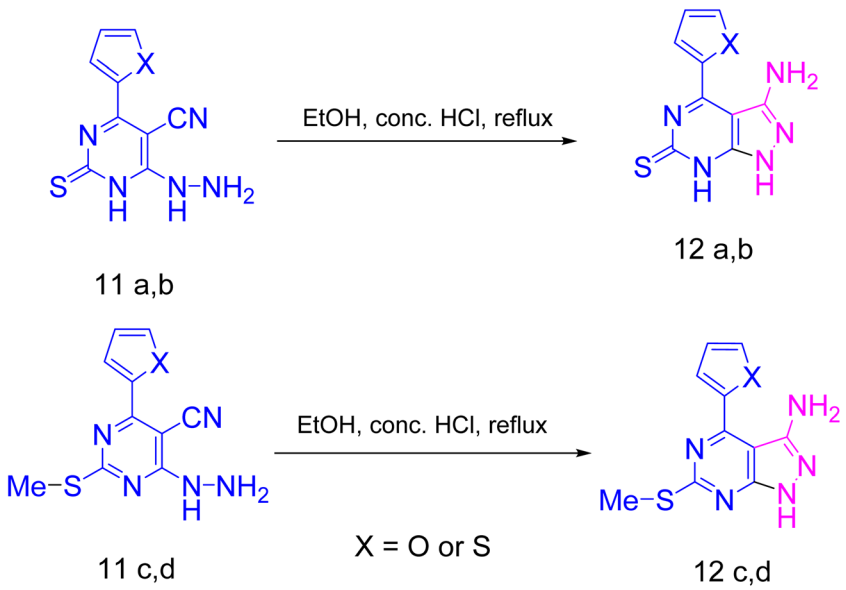
selectivity of their reaction with arylhydrazines. This protection is presumably attributed to steric hindrance and a change in reaction sequence between the reacting centers. Thus, treatment of 4,6-dichloropyrimidine-5-carbaldehyde derivatives **3** with 1,3-butylene glycol in the presence of *p*-toluenesulfonic acid monohydrate afforded 4,6-dichloro-5-(4-methyl-1,3-dioxan-2-yl)pyrimidines **4**. The latter compounds were treated with hydrazine derivatives to give the corresponding 4-chloro-5-(4-methyl-1,3-dioxan-2-yl)-6-(2-arylhydrazinyl)pyrimidines **5**, which were cyclized using *p*-toluenesulfonic acid, leading to the formation of the desired 2-arylpyrazolo[3,4-*d*]pyrimidines **6** (Scheme 2).[71]

On the other hand, Ebrahimpour *et al.* reported that the microwave-assisted transformation of ethyl 4-chloropyrimidine-5-carboxylates **7** provides direct access to the corresponding pyrazolo[3,4-*d*]pyrimidines **8** through reaction with hydrazine, methylhydrazine, or phenylhydrazine. In contrast, the classical synthesis of compounds **8** was achieved by heating the reactants in ethanol in the presence of sodium ethoxide as a base (Scheme 3).[72]

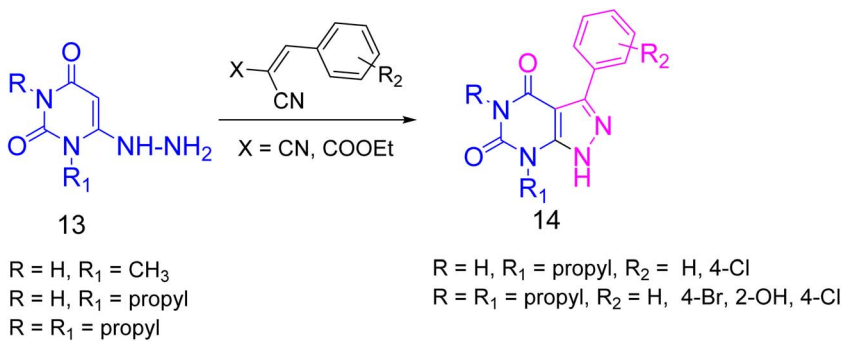
Wang *et al.* reported that the treatment of 4-amino-6-chloropyrimidine-5-carbonitrile **9** with isopropylhydrazine hydrochloride afforded the corresponding pyrazolo[3,4-*d*]pyrimidine derivatives **10** (Scheme 4).[73]

Ragab *et al.* described the acid-mediated intramolecular cyclization of 6-hydrazinylpyrimidine-5-carbonitrile derivatives **11a–d**, where treatment with HCl promoted ring closure to afford the corresponding 1*H*-pyrazolo[3,4-*d*]pyrimidin-3-amine derivatives **12a–d** (Scheme 5).[74]

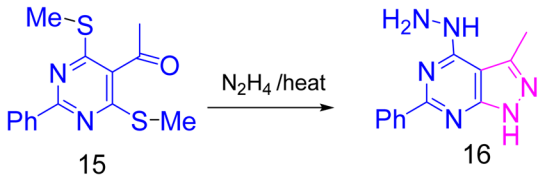
El-Kalyoubi *et al.* described the Michael-type addition of 6-hydrazinyluracils **13a–c** to benzylidene malononitrile or



Scheme 5 Preparation of pyrazolo[3,4-*d*]pyrimidine from 6-hydrazinylpyrimidine-5-carbonitriles.



Scheme 6 Preparation of pyrazolo[3,4-*d*]pyrimidine from 6-hydrazinyluracils.



Scheme 7 Preparation of pyrazolo[3,4-*d*]pyrimidine from a methylsulfanylpyrimidine derivative.

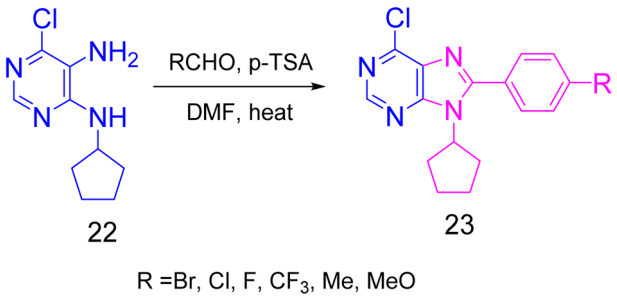
benzylidene ethyl cyanoacetate. Refluxing the reaction mixture in DMF in the presence of triethylamine promoted cyclization, followed by the elimination of malononitrile or ethyl cyanoacetate to furnish the corresponding 3-substituted-7-propyl- and/or 5,7-dipropyl-1*H*-pyrazolo[3,4-*d*]pyrimidine-4,6(5*H*,7*H*)-diones **14a–f** (Scheme 6).[75]

In another study, Komkov *et al.* reported that treatment of 5-acetyl-4,6-di(methylsulfanyl)-2-phenylpyrimidine (**15**) with hydrazine hydrate promoted cyclization to afford 4-hydrazino-3-methyl-6-phenyl-1*H*-pyrazolo[3,4-*d*]pyrimidine (**16**) (Scheme 7).[76]

El-Mekabaty reported that the treatment of enaminones **17** with hydrazine hydrate or phenyl hydrazine in refluxing DMF containing triethylamine furnished the corresponding pyrazolo[3,4-*d*]pyrimidinones **19** through the non-isolable intermediates **18** (Scheme 8).[77]

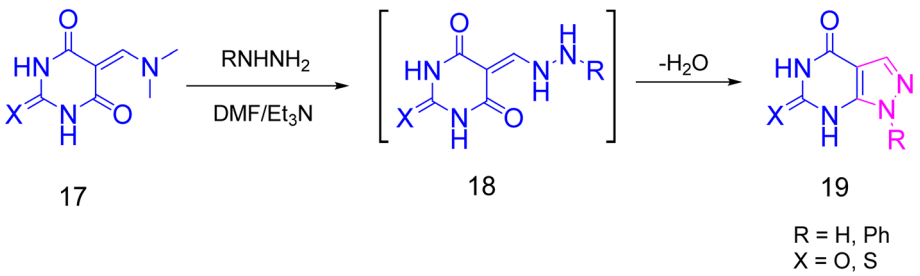
Synthesis of imidazolo[4,5-*d*] pyrimidines (purines)

Various synthetic strategies have been developed for the construction of the imidazolo[4,5-*d*]pyrimidine fused heterocyclic system. The most common approaches involve the

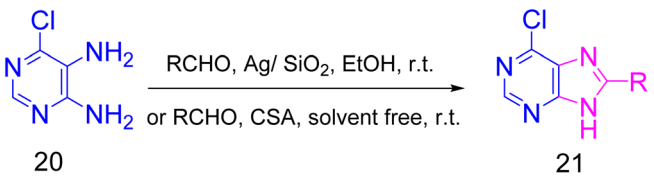


Scheme 10 Preparation of purine derivative *via* the reaction of 6-chloro-*N*-cyclopentylpyrimidine-4,5-diamine with substituted benzaldehyde.

cyclization of appropriate substituted pyrimidine derivatives, particularly 4,5-diaminopyrimidines with suitable reagents such as aldehydes, ketones, carboxylic acids, carboxylic anhydrides, amides, *ortho*-esters, cyanoacetimidate, malononitrile, isothiocyanates, carbon disulfide or primary alcohols. These reactions typically lead to the formation of an imidazole ring fused to the pyrimidine nucleus, producing the desired framework. Consequently, the synthesis of substituted purine derivatives has attracted considerable attention, and a variety of strategies have been developed using different catalysts, such as acetic anhydride, ethanesulfonic acid, NaH, $FeCl_3$–SiO_2, polyphosphoric acid (PPA), $Pd(OAc)_2$–CuI, Cs_2CO_3, H_2S/base, H_2S/S_8, $Pd_2(dba)_3$, cellulose sulfuric acid, and pyridine, as well as microwave-assisted conditions. However, many of these methods suffer from several drawbacks, including the use of toxic organic solvents, tedious work-up procedures, prolonged reaction times, and the requirement for expensive reagents or catalysts.

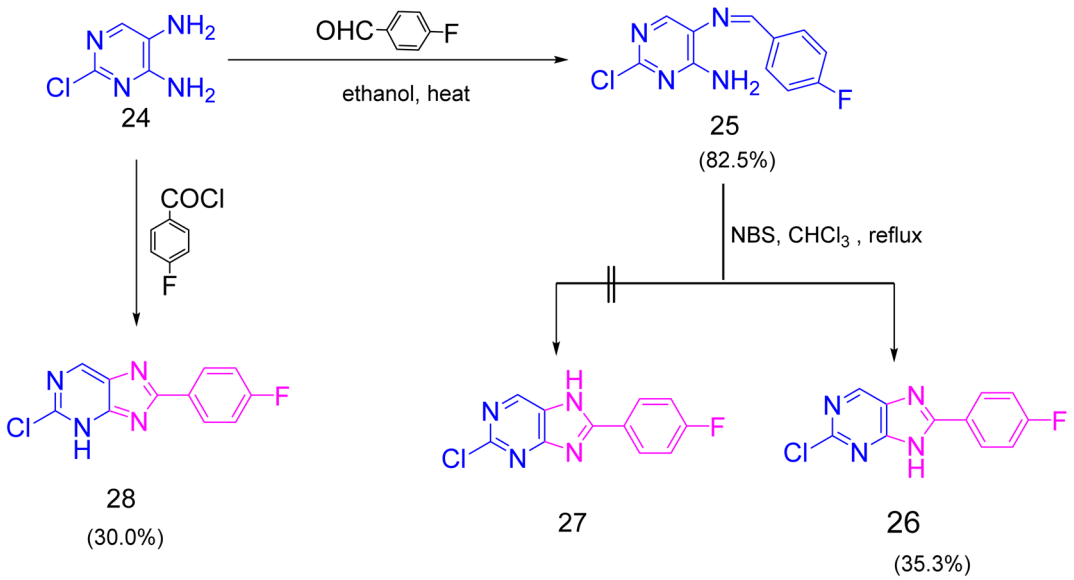


Scheme 8 Preparation of pyrazolo[3,4-*d*]pyrimidine from enaminones.

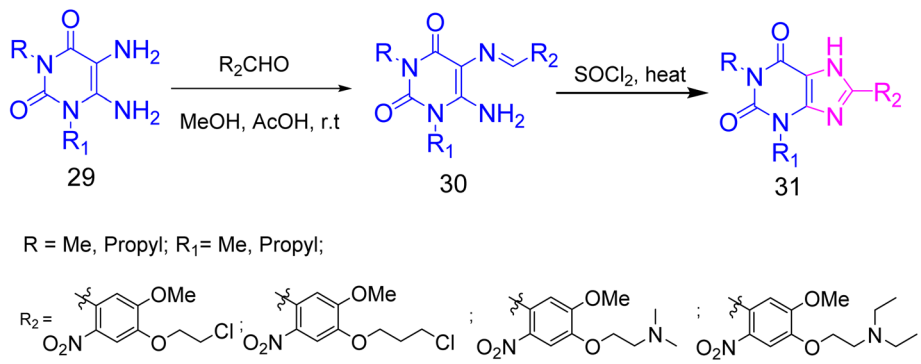


R =H, C_6H_5, 2-Br-C_6H_5, 4-Br-C_6H_5, 4-MeO$C_6H_5CH_2$, 4-HO-C_6H_5, 3,4-HO-C_6H_3, 2-CF_3-C_6H_5, 2,4-Cl_2-C_6H_3, $C_6H_5CH_2$, 4-MeC_6H_5, 4-MeO$C_6H_5CH_2$, 4-NO_2-$C_6H_5CH_2$, 4-Br-$C_6H_5CH_2$, 2-Cl-$C_6H_5CH_2$, 2,4-Cl_2-C_6H_3

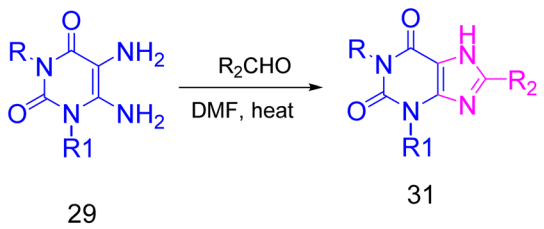
Scheme 9 Preparation of purine derivative *via* the reaction of 6-chloro-pyrimidine-4,5-diamine with substituted benzaldehyde.



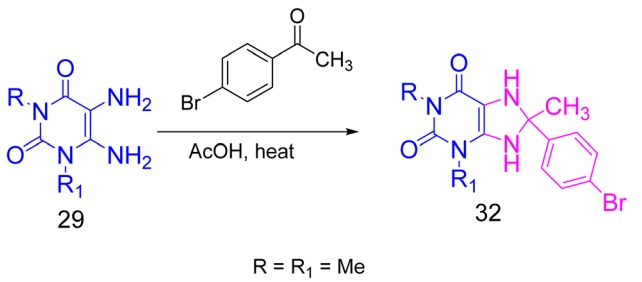
Scheme 11 Preparation of 9*H*-purines *via* the reaction of a Schiff base with NBS, and 3*H*-purine *via* the reaction of 4,5-diaminopyrimidine with 4-fluorobenzoyl chloride.



Scheme 12 Preparation of xanthine derivatives from 5,6-diaminouracils *via* reaction with substituted aldehydes followed by thionyl chloride.



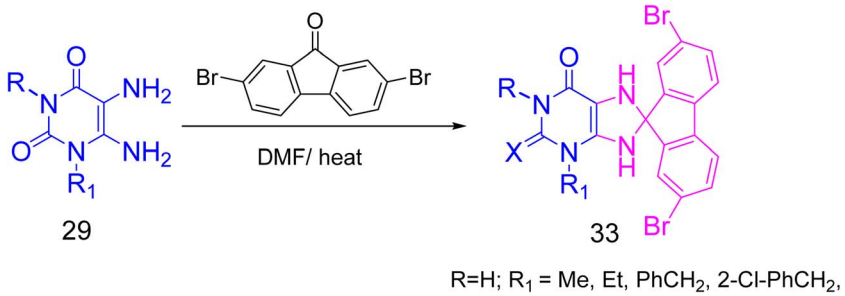
Scheme 13 Preparation of 8-aryl xanthine derivatives from 5,6-diaminouracil with aromatic aldehydes.



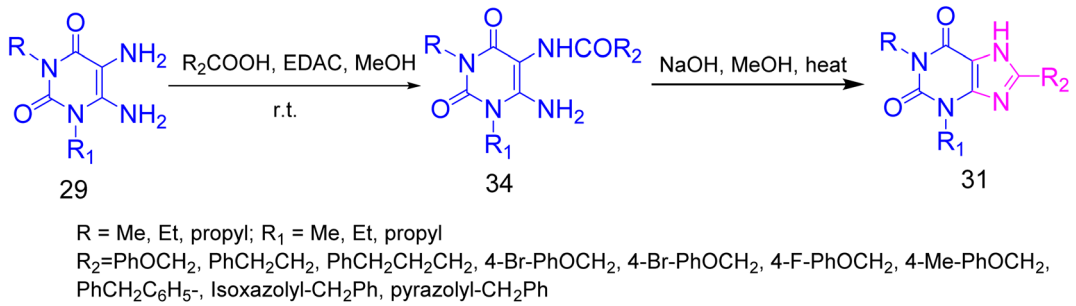
Scheme 14 Synthesis of 8-(4-bromophenyl)-1*H*-purine-2,6-dione from 1,3-dimethyl-5,6-diaminouracil and *p*-bromoacetophenone.

In recent years, heterogeneous catalysts have gained increasing importance in heterocyclic chemistry due to their significant advantages, such as simple work-up procedures, environmental compatibility, low cost, low toxicity, and ease of separation from the reaction mixtures.

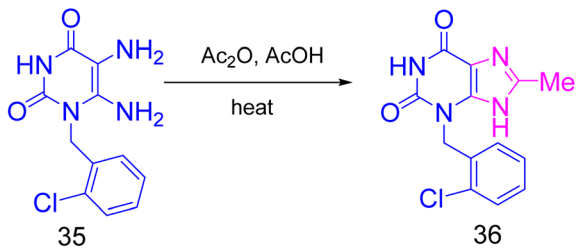
In this concept, Maddila *et al.* developed a convenient, simple, and efficient catalytic procedure for the synthesis of 6-chloro-8-substituted-9*H*-purine derivatives **21**. This method involves the one-pot condensation of 6-chloropyrimidine-4,5-diamine (**20**) with various aldehydes using silver supported on silica (Ag/SiO_2) or cellulose sulfuric acid as a heterogeneous



Scheme 15 Preparation of 2,7-dibromo-dihydrospiro[fluorene-9,8′-purine] derivatives from 5,6-diaminouracil with 2,7-dibromo-9*H*-fluoren-9-one.



Scheme 16 Synthesis of purine derivatives from 5,6-diaminouracil using carboxylic acids.



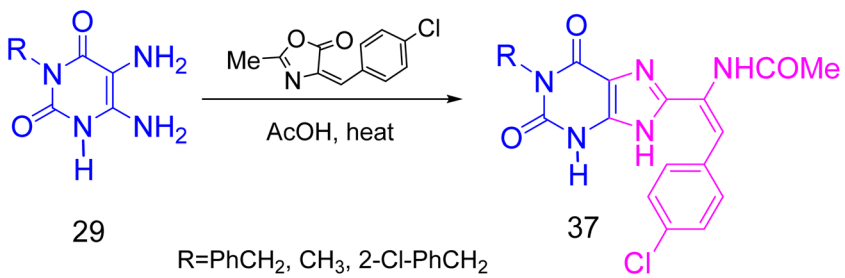
Scheme 17 Synthesis of 3-(2-chlorobenzyl)-8-methyl-3,9-dihydro-1*H*-purine-2,6-dione from a 5,6-diaminouracil derivative with acetic anhydride.

catalyst. The target compounds were obtained in excellent yields within short reaction times under environmentally friendly conditions (Scheme 9).[78,79]

Similarly, Polat *et al.* reported the use of another catalyst for the preparation of 6-chloro-*N*-cyclopentylpyrimidine-4,5-diamine **23** *via* the one-pot condensation of 6-chloro-N^4-cyclopentylpyrimidine-4,5-diamine (**22**) with substituted benzaldehydes in the presence of *p*-toluenesulfonic acid (*p*-TSA) as a catalyst (Scheme 10).[80]

Doganc *et al.* described a two-step synthesis of 2-chloro-8-(4-fluorophenyl)-9*H*-purine **26.** In the first step, Schiff base **25** was formed by the reaction of 2-chloropyrimidine-4,5-diamine (**24**) with 4-fluorobenzaldehyde under reflux conditions. In the second step, the resulting Schiff base **25** was treated with *N*-bromosuccinimide (NBS) in chloroform under reflux, yielding the *N*9 isomer **26** and not the *N*7 isomer **27.** Interestingly, they also reported that the reaction of 4,5-diaminopyrimidine **24** with 4-fluorobenzoyl chloride produced 6-chloro-8-(4-fluorophenyl)-3*H*-purine (**28**) as the sole product (Scheme 11).[81]

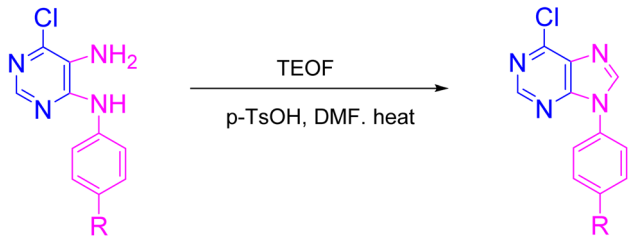
Similarly, other reports have described the use of thionyl chloride as a cyclizing agent in the two-step synthesis of 8-substituted-xanthine **31** *via* treatment of 5,6-diaminouracils **29**



Scheme 18 Synthesis of *N*-substituted-8-purine derivatives *via* the cyclo-condensation of 5,6-diaminouracils with an oxazolone derivative.

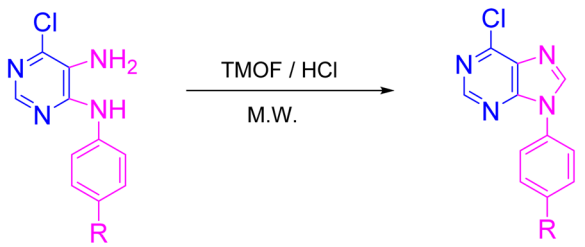
R = H, R_1 = Me, $(CH_2)_3$ OMe, $(CH_2)_3$, OCH_2Ph

Scheme 19 Synthesis of 3-substituted-1*H*-purine-2,6(3*H*,7*H*)-dione *via* the reaction of 5,6-diaminopyrimidine with triethyl orthoformate (TEOF).



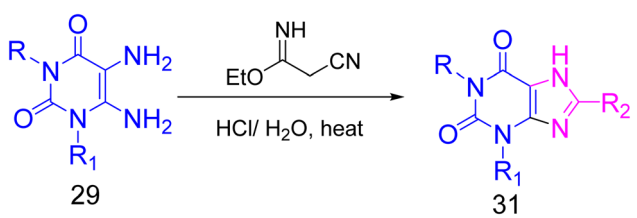
a: R = Cl
b: R = CF_3

Scheme 20 Synthesis of 3-substituted-1*H*-purine-2,6(3*H*,7*H*)-dione *via* the reaction of 5,6-diaminopyrimidine with triethyl orthoformate (TEOF).



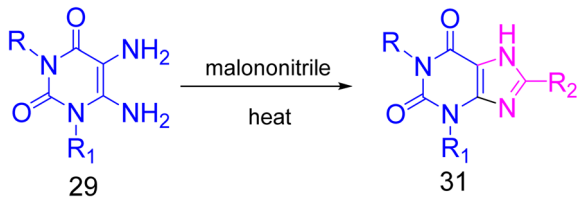
R = 3-NO_2

Scheme 21 Synthesis of 3-substituted-1*H*-purine-2,6(3*H*,7*H*)-dione *via* the reaction of 5,6-diaminopyrimidine with trimethyl orthoformate (TMOF).



R = R_1 = Me, R_2 = CH_2CN

Scheme 22 Synthesis of 1,3-dimethyl-2,6-dioxo-1*H*-purine derivatives from diaminopyrimidine.



R = H, R_1 = 2-Cl-$C_6H_4CH_2$, R_2 = CH_2CN

Scheme 23 Synthesis of purine derivatives from 5,6-diaminouracil with malononitrile.

with various substituted aldehydes in a MeOH : AcOH (4 : 1) mixture at room temperature to afford the corresponding Schiff bases **30.** Subsequent oxidative cyclization of these benzylidene derivatives using thionyl chloride furnished the desired 8-substituted-xanthine **31** in good yields (Scheme 12).[82–84]

However, El-Kalyoubi *et al.* reported that 8-aryl xanthine derivatives **31** could be obtained directly *via* the condensation of 5,6-diaminouracils **29** with various aromatic aldehydes in DMF under reflux (Scheme 13).[85]

Furthermore, El-Kalyoubi *et al.* reported a simple procedure for the synthesis of the substituted purine derivative **32** in high yield by heating 1,3-dimethyl-5,6-diaminouracil **29** with *p*-bromoacetophenone in DMF under a light torch flame, producing 8-(4-bromophenyl)-1,3,8-trimethyl-3,8-dihydro-1*H*-purine-2,6-dione (**32**) (Scheme 14).[86]

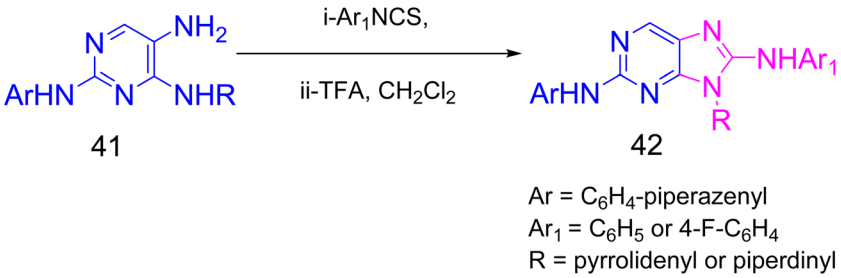
In another report, El-Kalyoubi *et al.* heated diaminouracils **29** in DMF with 2,7-dibromo-9*H*-fluoren-9-one, resulted in an intramolecular Aza-Michael addition, furnishing 2,7-dibromo-dihydrospiro[fluorene-9,8′-purine] derivatives **33** in moderate yields (Scheme 15).[87]

The synthesis of substituted purine derivatives was reported by several authors *via* the reaction of 5,6-diaminouracil derivatives **29** with appropriate carboxylic acids, such as 4-phenylbutanoic acid, phenylpropanoic acid, phenoxyacetic acid, phenyl carboxylic acid, isoxazole carboxylic acid, pyrazole-3-carboxylic acids, and pyrazole-4-carboxylic in the presence of the coupling/dehydrating reagent *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (EDAC) at room temperature. The resulting intermediates, 1,3-dialkyl-6-amino-5-carboxamidouracils **34**, were subsequently treated with aqueous NaOH to afford the corresponding 3,7-dihydro-1*H*-purine-2,6-dione derivatives **31** (Scheme 16).[88–91]

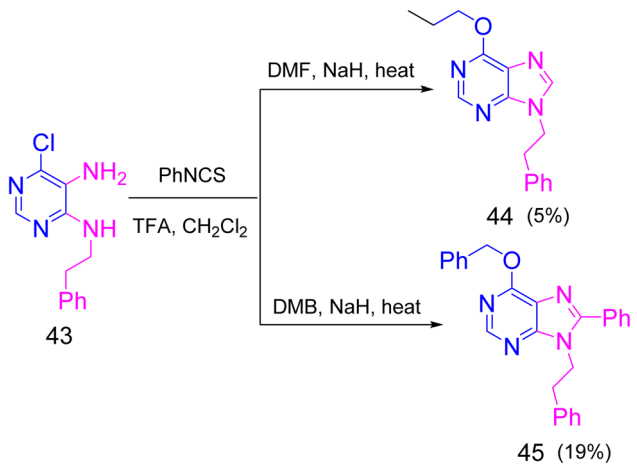
Furthermore, 3-(2-chlorobenzyl)-8-methyl-3,9-dihydro-1*H*-purine-2,6-dione (**36**) was efficiently synthesized in higher yield and shorter reaction time, without the use of toxic reagents, by refluxing 5,6-diamino-1-(2-chlorobenzyl)pyrimidine-2,4(1*H*,3*H*)-dione (**35**) with acetic anhydride in acetic acid (Scheme 17).[92]

Moreover, El-Kalyoubi *et al.* reported the synthesis of *N*-substituted purine derivatives **37** through cyclocondensation of suitable 5,6-diaminouracil derivatives **29** with an azlactone (oxazolone) derivative in acetic acid (Scheme 18).[93]

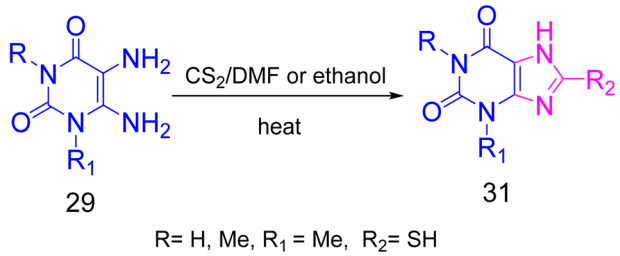
Pretze *et al.* reported that the treatment of 5,6-diaminopyrimidine-2,4(1*H*,3*H*)-diones **29** with triethyl



Scheme 24 Synthesis of 9-heterocyclyl-substituted 9*H*-purine derivatives.



Scheme 25 Synthesis of 6,9-disubstituted purines from diaminopyrimidine.



Scheme 26 Synthesis of 8-mercapto-purines from diaminopyrimidine.

orthoformate (TEOF) in the presence of *p*-toluenesulfonic acid (*p*-TsOH) afforded the corresponding 3-substituted-1*H*-purine-2,6(3*H*,7*H*)-dione derivatives **38** (Scheme 19).[94]

In the same manner, Kucukdumlu *et al.* designed and synthesized novel purine analogs, by the multistep reactions, through condensation of 4,6-dichloro-5-nitropyrimidines **39a** and **39b** with triethyl orthoformate in the presence of *p*-toluenesulfonic acid, affording 9-substituted purine derivatives **40a** and **40b**, respectively (Scheme 20).[95]

Also, Orduña *et al.* illustrated the synthesis of new substituted purine derivatives using low-power microwave irradiation. The treatment of 5,6-diaminopyrimidine derivative **39c** with trimethyl orthoformate (TMOF), in the presence of HCl, afforded the corresponding 9-substituted purine derivative **40c** in good yield (Scheme 21).[96]

Verma *et al.* described a new strategy for the synthesis of 1,3-dimethyl-2,6-dioxo-1*H*-purine derivatives **31** containing an acetonitrile group. The reaction was carried out by treating 1,3-dimethyl-5,6-diaminouracil **29** with ethyl 2-cyanoacetimidate in the presence of hydrochloric acid under reflux conditions (Scheme 22).[97]

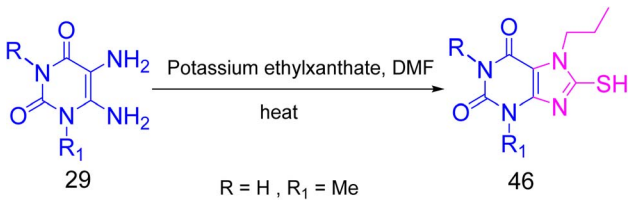
Moreover, El-Kalyoubi *et al.* developed a novel method for access to numerous functionalized purine scaffolds **31** in less time and improvement in yield by refluxing of 5,6-diaminouracil **29** with malononitrile under solvent-free condition (Scheme 23).[92]

Lei *et al.* treated compound **41** with phenyl isothiocyanate or 4-fluorophenyl isothiocyanate, followed by trifluoroacetic acid in dichloromethane, to generate 9-heterocyclyl-substituted 9*H*-purine derivatives **42** (Scheme 24).[98]

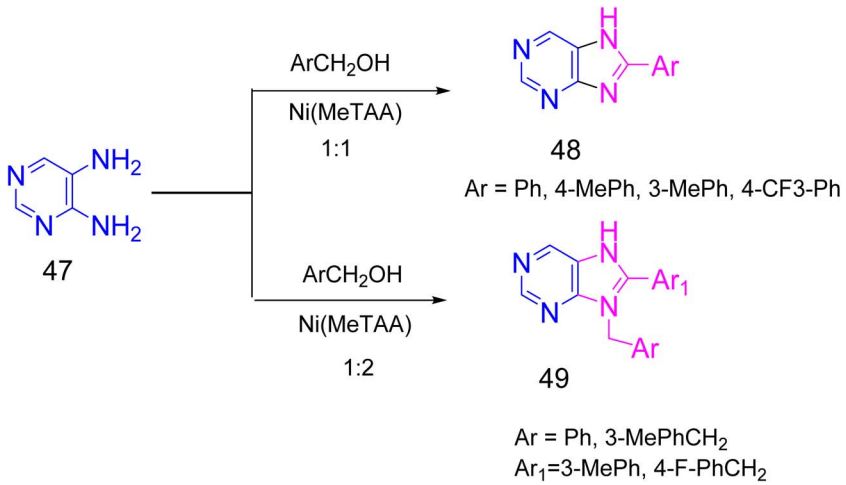
Lorente-Macías *et al.* explored that the 6-alkoxy purine derivatives were prepared *via* the reaction compound **43** with phenyl isothiocyanate. It was noticed that using dimethylformamide (DMF) facilitated the synthesis of 6,9-disubstituted purines **44** with monosubstituted C8; however, the use of dimethylbenzamide (DMB) resulted in the synthesis of trisubstituted purines **45** with fragments at both C6 and C8 (Scheme 25).[99]

Hisham *et al.* reported a direct cyclization of 5,6-diamino-1,3-dimethyluracil **29** using carbon disulfide in DMF or in an ethanolic solution of KOH under reflux, affording 8-mercapto-1,3-dimethyl-1,2,3,7-tetrahydro-6*H*-purin-6-one **31** (Scheme 26).[100]

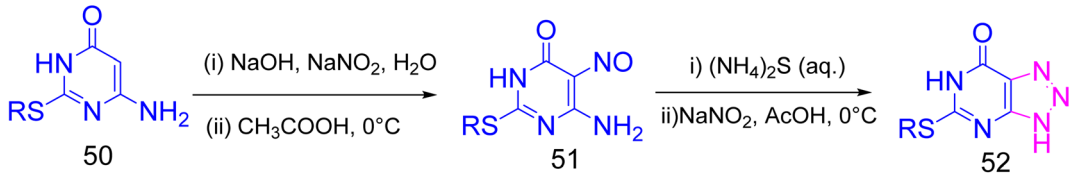
Also, compound **46** was obtained by heating substituted diamino-pyrimidine-2,4-one derivative **29** with potassium ethylxanthate in DMF (Scheme 27).[101]



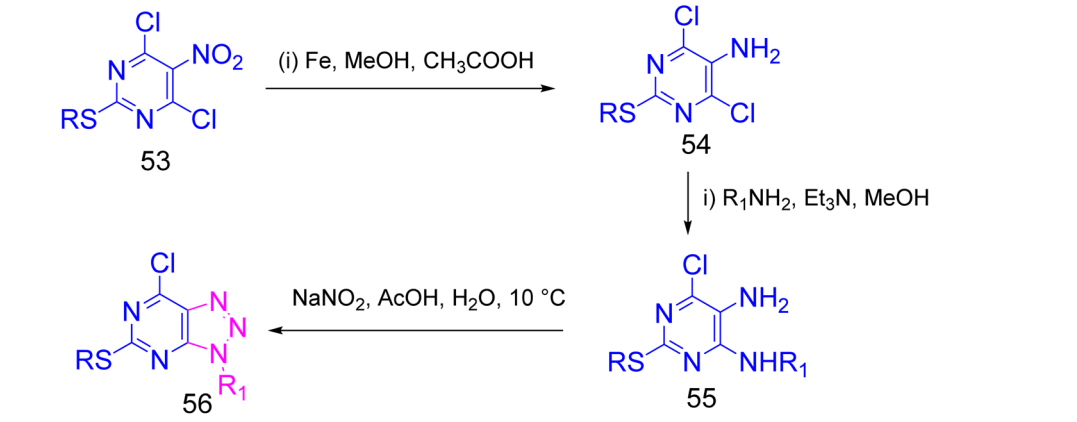
Scheme 27 Synthesis of 8-mercapto-purines from diaminopyrimidine.



Scheme 28 Synthesis of 6-substituted purines from diaminopyrimidine using primary alcohol.



Scheme 29 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidine from 6-amino-2-alkylsulfanylpyrimidine.

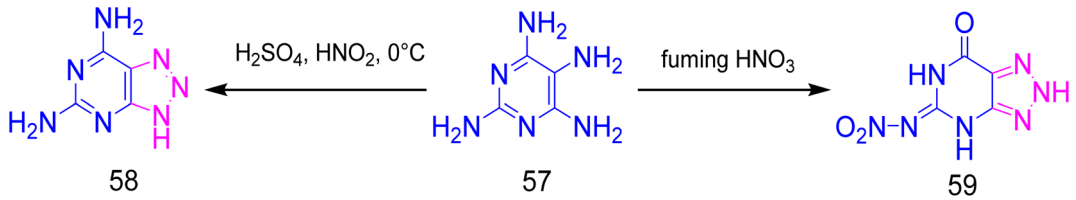


Scheme 30 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidines from nitrosopyrimidine derivatives.

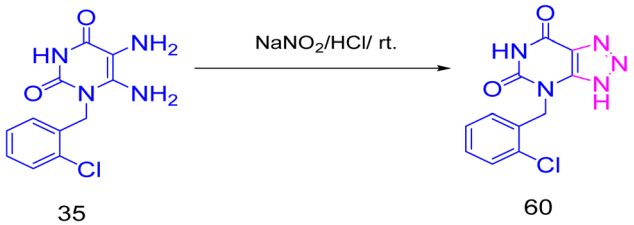
Chakraborty *et al.* explored the synthesis of a variety of 8-substituted purines **48** and 8,9-disubstituted purines **49** *via* the dehydrogenative coupling reaction of different substituted benzyl alcohol derivatives with 4,5-diaminopyrimidine **47** using a nickel catalyst [Ni(MeTAA)] for a prolonged reaction time and under an argon atmosphere. The ratio between the reactants significantly affected the type and yield of the obtained products (Scheme 28).[102]

Synthesis of [1,2,3]triazolo[4,5-*d*] pyrimidines (azapurine derivatives)

Some reports have described the design and synthesis of this scaffold *via* a multistep transformation starting from 6-amino-2-alkylsulfanylpyrimidine analogues **50**. Initially, nitrosation of compounds **50** in the presence of acetic acid affords the



Scheme 31 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidine from tetraaminopyrimidine.



Scheme 32 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidine from the 4,5-diaminopyrimidine derivative.

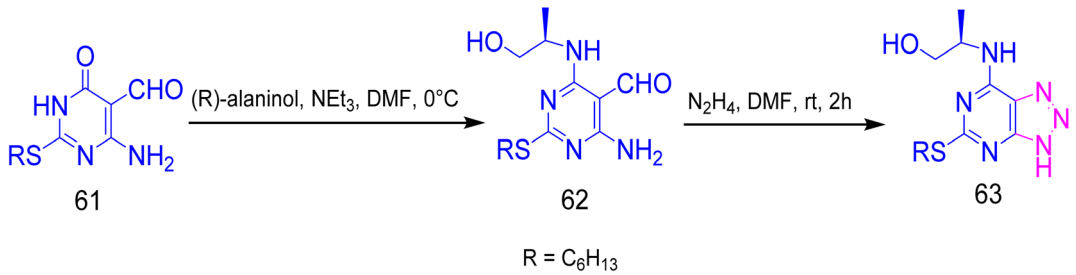
corresponding 5-nitrosopyrimidine derivatives **51.** Subsequently, a Zinin reduction of the nitroso group using ammonium sulfide provides the corresponding 5,6-diaminopyrimidine intermediates. Due to their high susceptibility to oxidation, these intermediates are not isolated but are immediately subjected to diazotization in aqueous acetic acid, leading to intramolecular cyclization and affording the desired 1,2,3-triazolo[4,5-*d*]pyrimidine derivatives **52** (Scheme 29).[103,104]

Alternatively, many reports stated that the reduction of nitrosopyrimidine derivatives **53** using iron powder in methanol/acetic acid afforded the corresponding aminopyrimidine derivatives **54.** Subsequently, heating of compounds **54** with the appropriate amine produces the intermediates **55.** Without further purification, upon treatment of **55** with sodium nitrite in the presence of acetic acid, it undergoes intramolecular cyclization to furnish the corresponding [1,2,3]triazolo[4,5-*d*]pyrimidine derivatives **56** (Scheme 30).[105–109]

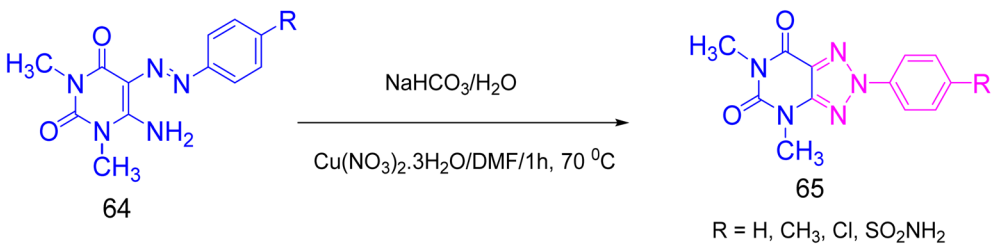
Moreover, diazotization of tetraaminopyrimidine (**57**) using sodium nitrite in sulfuric acid, followed by intramolecular cyclization, leads to the formation of the fused [1,2,3]triazolo[4,5-*d*]pyrimidine ring system **58.** In addition, nitration of compound **57** using nitric acid as the nitrating agent and upon quenching the reaction mixture with ice-water, the fused heterocyclic compound *N*-(7-oxo-6,7-dihydro-2*H*-[1,2,3]triazolo[4,5-*d*]pyrimidin-5-yl)nitramide (**59**) was isolated in good yield (Scheme 31).[110]

Similarly, triazolo[4,5-*d*]pyrimidine **60** was synthesized in good yield *via* cyclocondensation of the 4,5-diaminopyrimidine precursor **35** with *in situ* generated nitrous acid (HNO_3) at ambient temperature (Scheme 32).[92]

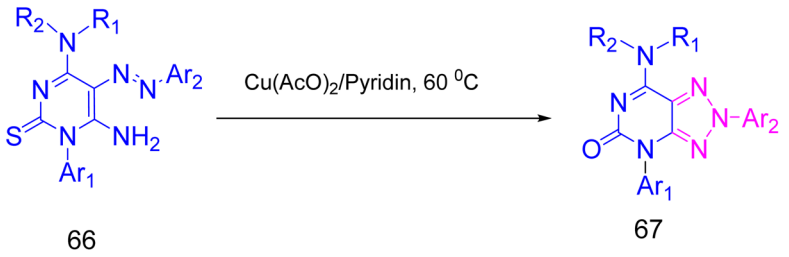
Recently, another important approach utilizes pyrimidine-5-carboxaldehyde derivatives as key precursors for the construction of the [1,2,3]triazolo[4,5-*d*]pyrimidine framework. For instance, in 2026, Omanakuttan *et al.* reported the synthesis of triazolo[4,5-*d*]pyrimidine analogues *via* nucleophilic aromatic



Scheme 33 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidine from 4,6-dichloropyrimidine-5-carboxaldehyde.

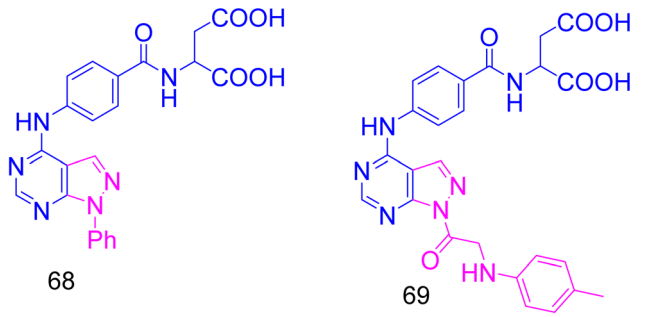


Scheme 34 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidine from (arylazo)-6-aminouracil derivatives in the presence of copper ions.

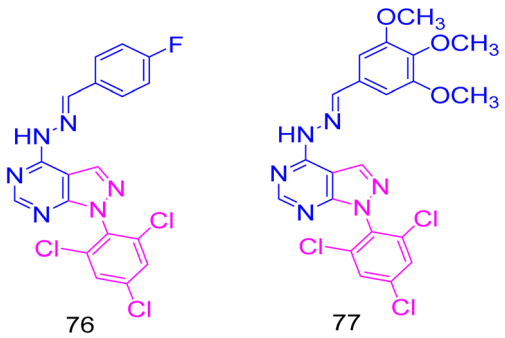


R_1, R_2 =Morpholin-4-yl, Piperdin-1-yl; Ar_1 = C_6H_5, 4-Meo-C_6H_5, 4-Cl-C_6H_5 , 4-CF_3-C_6H_5; Ar_2 = 4-Meo-C_6H_5, 2-Meo-C_6H_5, 3,4-$(MeO)_2$-C_6H_4, 4-$(Me)_2N$-C_6H_5, 4-CF_3-C_6H_5

Scheme 35 Preparation of [1,2,3]triazolo[4,5-*d*]pyrimidine from (arylazo)-6-aminouracil derivatives using copper(II) acetate in pyridine.

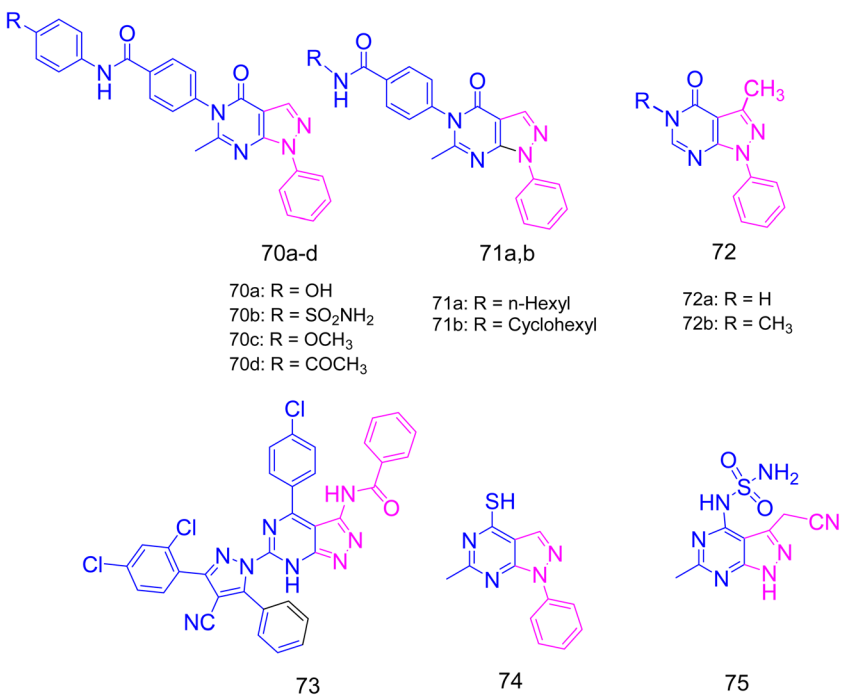


Scheme 36 Pyrazolo[3,4-*d*]pyrimidine derivatives as anticancer agents.

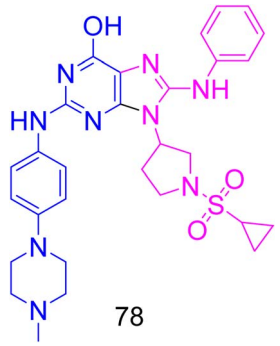


Scheme 38 Pyrazolo[3,4-*d*]pyrimidine derivatives as anticancer agents.

substitution of pyrimidine-5-carboxaldehyde (**61**) with (*R*)-alaninol, affording the corresponding pyrimidine derivative **62.** Subsequent condensation of compound **62** with hydrazine hydrate promoted intramolecular cyclization, leading to the formation of the fused [1,2,3]triazolo[4,5-*d*]pyrimidine derivative **63** in good yield (Scheme 33).[104]



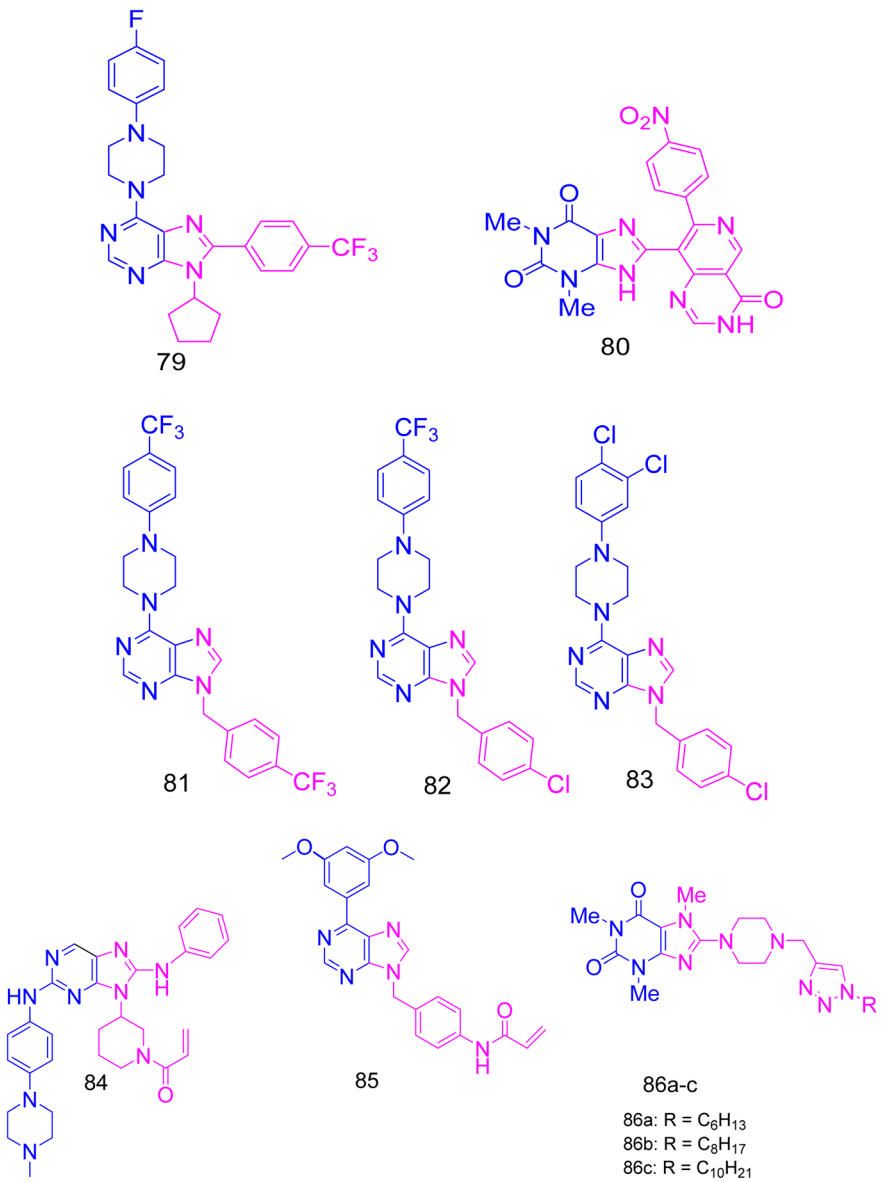
Scheme 37 Pyrazolo[3,4-*d*]pyrimidine derivatives as anticancer agents.



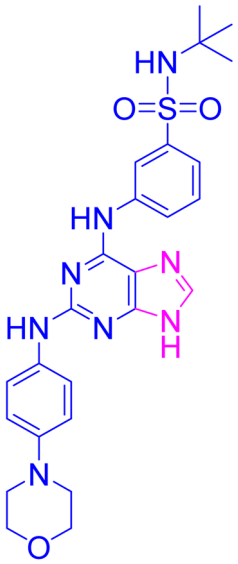
Scheme 39 Substituted-purine derivatives as anticancer agents.

In addition, hydrazino-substituted pyrimidines have been employed as versatile building blocks for the construction of [1,2,3]triazolo[4,5-*d*]pyrimidine derivatives, where oxidative or nitrosative cyclization promotes the formation of the fused triazole ring. In this context, Debnath *et al.* explored a method for converting 1,3-dimethyl-5-(arylazo)-6-aminouracil derivatives **64** into novel 1,3-dimethyl-8-arylazapurin-2,6-dione derivatives **65** through an oxidative transformation in the presence of copper ions under alkaline conditions. The reaction proceeds efficiently in a dimethylformamide–water medium, affording the desired fused heterocyclic products in good yields (Scheme 34).[111]

Subsequently, Alexander K. Eltyshev and co-workers investigated several oxidizing systems for the conversion of 5-(arylazo)-6-aminouracil derivatives **66** into 8-arylazapurine derivatives **67**. A variety of oxidants, including oxygen,

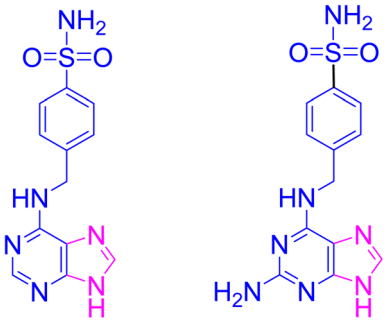


Scheme 40 Substituted-purine derivatives as anticancer agents.



87

Scheme 41 Substituted-purine derivatives as anticancer agents.



88 89

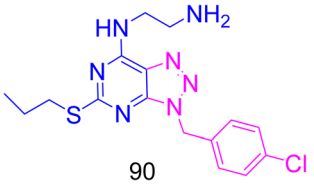
Scheme 42 Substituted-purine derivatives as anticancer agents.

potassium persulfate, sodium perborate, and *N*-bromosuccinimide, were evaluated for this transformation. Among the tested systems, the combined use of copper(II) acetate in pyridine was found to be the most effective oxidizing system, providing the desired azapurine derivatives with efficiency under the optimized reaction conditions (Scheme 35).[112–114]

Biological activity

Anticancer

Salem *et al.* and Nassar *et al.* reported the design and synthesis of novel pyrazolo[3,4-*d*]pyrimidine derivatives as structural analogues of classical antifolate agents, including methotrexate (MTX), pralatrexate, and pemetrexed (PMX). *In vitro* biological evaluation demonstrated that compounds **68** and **69** exhibit potent inhibitory activity against dihydrofolate reductase (DHFR), along with significant antiproliferative effects across



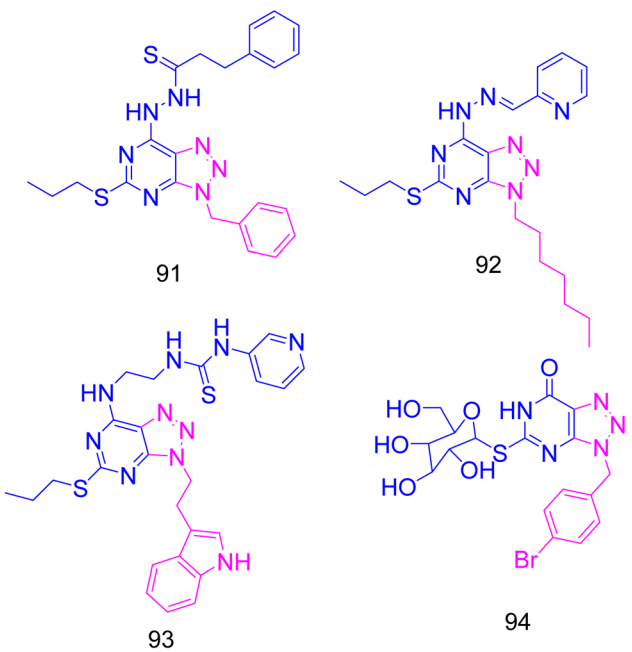
90

Scheme 43 [1,2,3]Triazolo[4,5-*d*]pyrimidine derivatives as anticancer agents.

a panel of human cancer cell lines compared to the reference compound sorafenib (Scheme 36).[115–117]

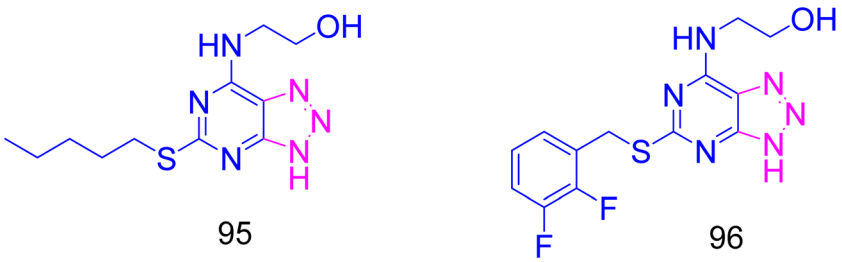
In addition, numerous studies have focused on the design, synthesis, and biological evaluation of diverse pyrazolo[3,4-*d*]pyrimidine derivatives for their anticancer potential. These compounds have been screened against a panel of human cancer cell lines, including HCT-116 (colon, EGFR mutant), HT-29 (colon, EGFR wild-type), MCF-7 and MDA-MB-231 (breast), HepG2 (hepatocellular), HGC-27 (gastric), and A549 (lung). Several derivatives, namely **70a–d**, **71a**, **71b**, **72a**, **72b**, **73**, **74**, and **75**, exhibited significant antiproliferative activity against selected cell lines, in some cases comparable to or exceeding that of standard reference drugs such as *sorafenib*, *erlotinib*, and *doxorubicin* (Scheme 37).[43,118–122]

Moreover, Nemr *et al.* reported that compounds **76** and **77** exhibit pronounced inhibitory activity against cyclin-dependent kinase 2 (CDK2) and related isoforms. Molecular interaction studies revealed that these derivatives bind efficiently within the CDK active site, adopting binding modes comparable to those of endogenous substrates or well-established CDK inhibitors (Scheme 38).[123]

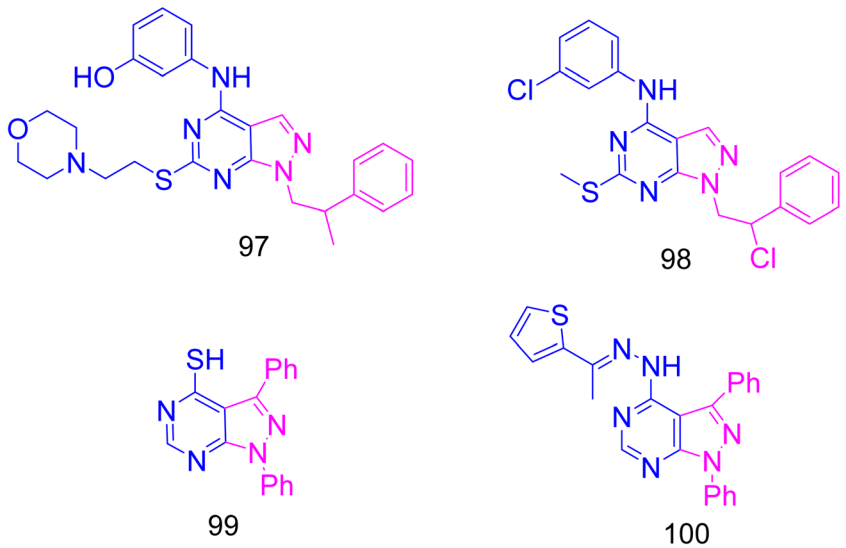


91 92 93 94

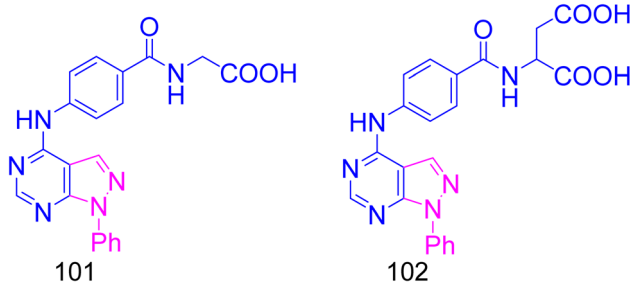
Scheme 44 [1,2,3]Triazolo[4,5-*d*]pyrimidine derivatives as anticancer agents.



Scheme 45 [1,2,3]Triazolo[4,5-*d*]pyrimidine derivatives as anticancer agents.



Scheme 46 Pyrazolo[3,4-*d*]pyrimidine derivatives as antimicrobial agents.



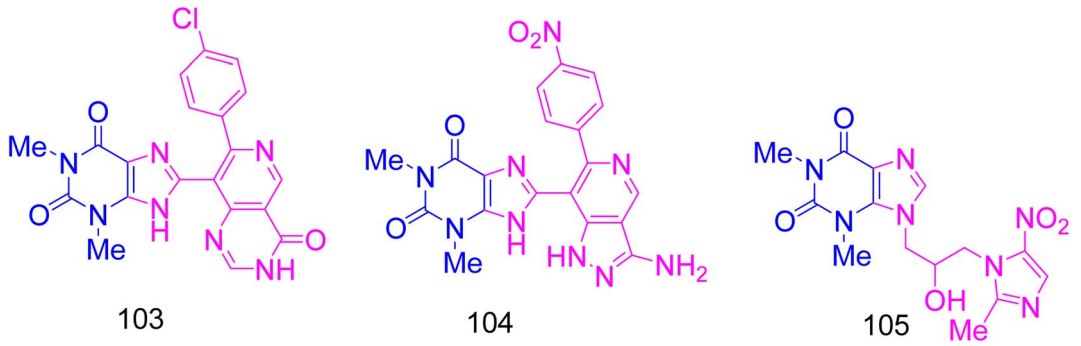
Scheme 47 Pyrazolo[3,4-*d*]pyrimidine derivatives as antimicrobial agents.

Lei *et al.* designed and synthesized a series of novel 9-heterocyclyl-substituted 9*H*-purine derivatives as potent inhibitors targeting the mutant $EGFR^{L858R/T790M/C797S}$ tyrosine kinase. Among the synthesized compounds, compound **78** exhibited remarkable antiproliferative activity against the HCC827 and H1975 cell lines and demonstrated strong inhibitory activity against $EGFR^{L858R/T790M/C797S}$ (Scheme 39).[98]

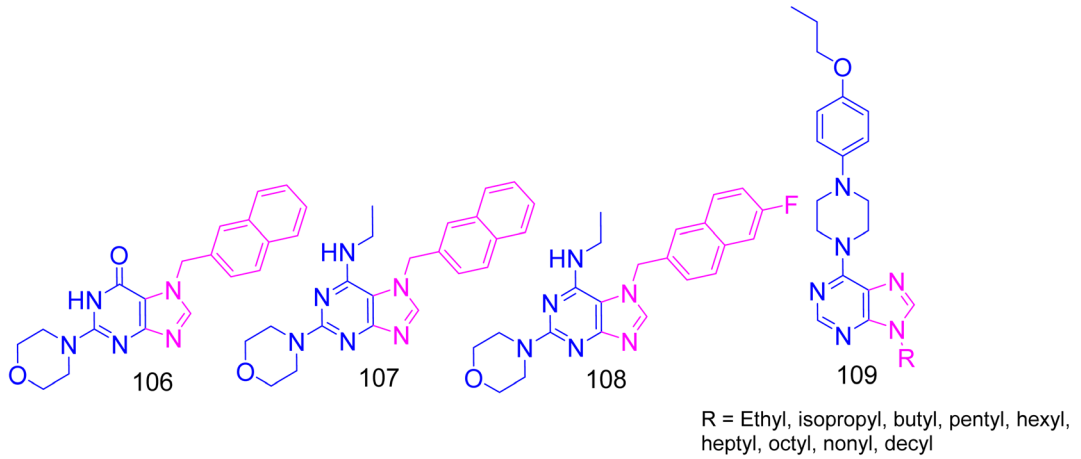
In addition, numerous studies have reported the evaluation of purine derivatives against a broad spectrum of cancer cell lines, including HUH7 (liver), MCF7 (breast) and T47D (breast), HCT-116 (colon), A549 (lung), HeLa (cervical), Panc-1 (pancreatic), HCC827 and H1975 (lung). Structure–activity relationship (SAR) analysis revealed that compounds **79–85** and **86a**, **86b** represent the most potent derivatives, exhibiting significant antiproliferative activity across the tested cell lines. Notably, their IC_{50} values were comparable to those of established anticancer agents such as camptothecin (CPT), cladribine, fludarabine, fexagratinib (AZD4547), and 5-fluorouracil (5-FU) (Scheme 40).[80,97,124–127]

Also, a series of novel 9*H*-purine-2,6-diamine derivatives was designed, synthesized, and biologically evaluated as dual inhibitors targeting Janus kinase 2 (JAK2) and the second bromodomain of BRD4 [BRD4(BD2)]. Both *in vitro* and *in vivo* studies demonstrated their potential therapeutic relevance. Among them, compound **87** displayed promising inhibitory activity against both targets, exhibiting favorable IC_{50} values for BRD4(BD2) and JAK2 (Scheme 41).[128]

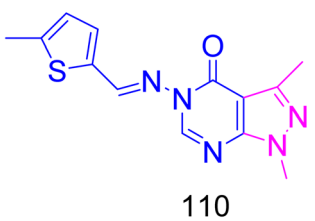
Sivakrishna Narra and Nethaji Munirathinam reported the synthesis and biological evaluation of sulfonamide-decorated *N*6-benzylaminopurine derivatives **88** and **89** for their anticancer potential. *In silico* studies indicated that compound **89** exhibits a higher binding affinity toward the ATP-binding sites of cyclin-dependent kinases CDK1, CDK2, and CDK4 compared to roscovitine, a well-established CDK inhibitor. Furthermore, cytotoxicity assays demonstrated that



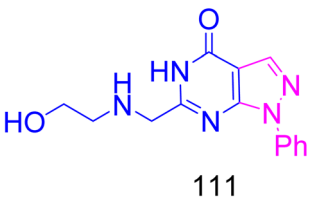
Scheme 48 Purine derivatives as antimicrobial agents.



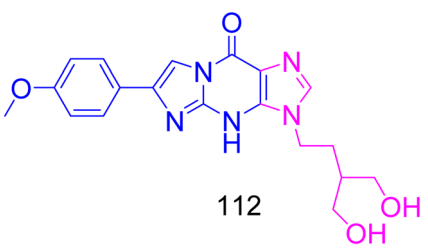
Scheme 49 Purine derivatives as antimicrobial agents.



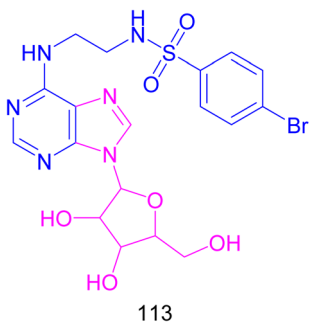
Scheme 50 Pyrazolo[3,4-*d*]pyrimidine derivative as an antiviral agent.



Scheme 51 Pyrazolo[3,4-*d*]pyrimidine derivative as an antiviral agent.

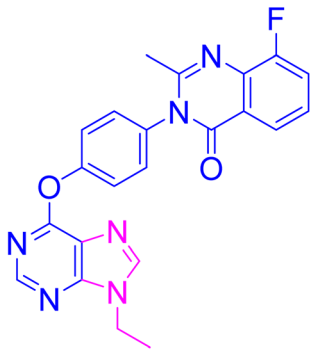


Scheme 52 Purine derivative as an antiviral agent.



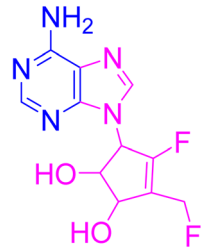
Scheme 53 Purine derivative as an antiviral agent.

compound **89** showed enhanced antiproliferative activity relative to both compound **88** and roscovitine. Collectively, these findings suggest that compound **89** represents



114

Scheme 54 Purine derivative as an antiviral agent.



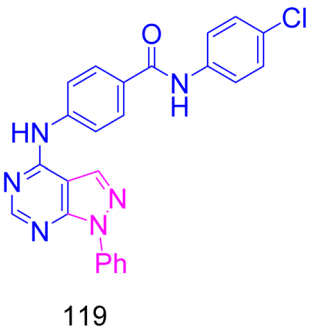
115

Scheme 55 Purine derivative as an antiviral agent.

a promising lead scaffold for the development of novel anti-cancer agents (Scheme 42).[129]

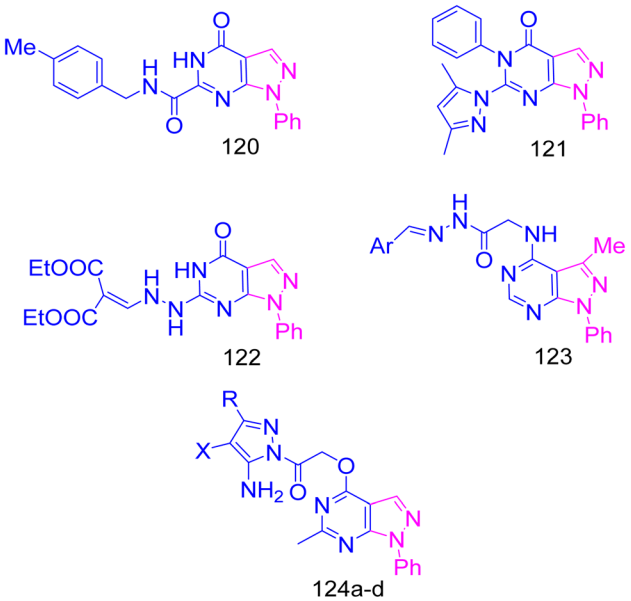
Liu *et al.* reported the design and synthesis of novel [1,2,3]triazolo[4,5-*d*]pyrimidine derivatives as inhibitors of ubiquitin-specific peptidase 28 (USP28), exhibiting activity in the low micromolar range. Among them, compound **90** demonstrated potent inhibitory activity against USP28 and displayed high selectivity over related targets, including USP7 and lysine-specific demethylase 1 (LSD1), with IC_{50} values exceeding 100 μmol L^{-1} (Scheme 43).[38]

Several authors reported the design and synthesis of novel [1,2,3]triazolo[4,5-*d*]pyrimidine derivatives, which were subsequently evaluated for their antiproliferative activity against a panel of human cancer cell lines, including MGC-803 (gastric), NCI-H1650 and PC9 (lung), PC-3 (prostate), and EC9706 (esophageal) cells. Among these compounds, derivatives **91–94**



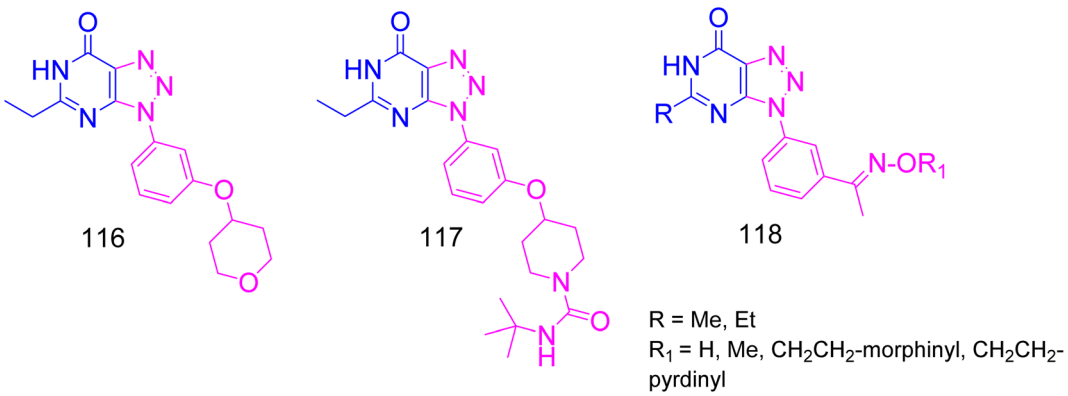
119

Scheme 57 Pyrazolo[3,4-*d*]pyrimidine as an anti-inflammatory agent.



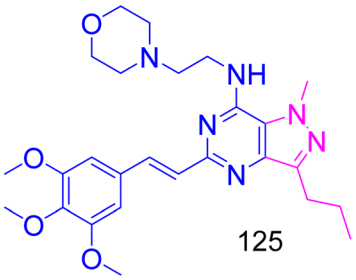
a: R=H , X= CN
b R=Me, X= CN
c: R=H , X= COOEt
d: R=Me , X= COOEt

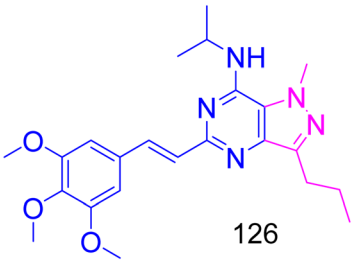
Scheme 58 Pyrazolo[3,4-*d*]pyrimidines as anti-inflammatory agents.



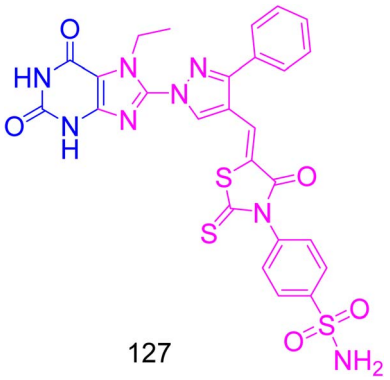
R = Me, Et
R_1 = H, Me, CH_2CH_2-morphinyl, CH_2CH_2-pyrdinyl

Scheme 56 [1,2,3]Triazolo[4,5-*d*]pyrimidine derivative as an antiviral agent.

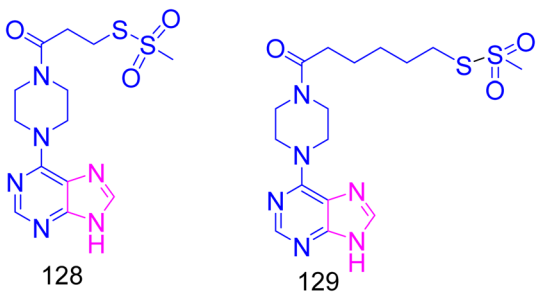




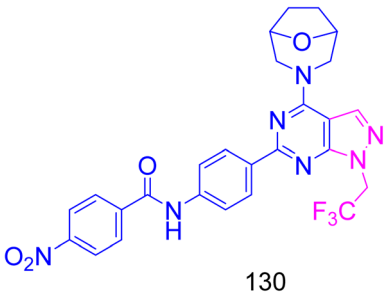
Scheme 59 Pyrazolo[4,3-*d*]pyrimidines as anti-inflammatory agents.



Scheme 60 Purine derivative as an anti-inflammatory agent.

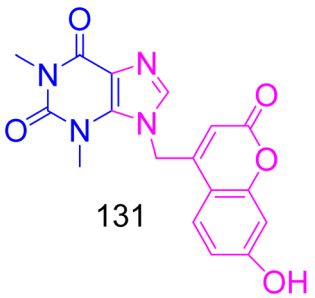


Scheme 61 Purine derivatives as anti-inflammatory agents.

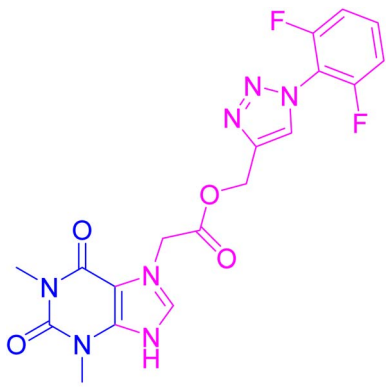


Scheme 62 Pyrazolo[3,4-*d*]pyrimidine as an anti-diabetic agent.

displayed the most potent anticancer activity when compared to the effect of the commonly used anticancer agent doxorubicin (Scheme 44).[34,37,105,106]



Scheme 63 Purine hybrid as an antioxidant agent.

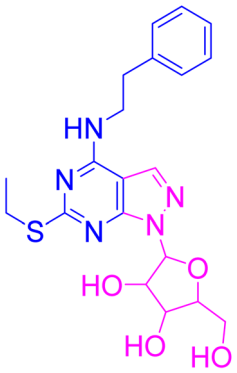


Scheme 64 Inhibitory activity of the purine derivative against acetylcholinesterase.

Van Hoof *et al.* and Omanakuttan *et al.* reported some triazolo[4,5-*d*]pyrimidine analogues displaying dual antagonistic activity against CCR7 and CXCR2. Among them, lead compounds **95** and **96** revealed promising activity. Structure–activity relationship studies revealed that shortening or elongation of the length of the aliphatic group at the 2-thio-position was detrimental to enhancing the CCR7 antagonistic potency and selectivity, and in both cases, resulted in compounds exhibiting low micromolar activity as CCR7 and CXCR2 antagonists (Scheme 45).[104,130]

Antimicrobial activity

Several studies have investigated the antimicrobial potential of pyrazolo[3,4-*d*]pyrimidine derivatives against both Gram-



133

Scheme 65 Pyrazolo[3,4-*d*]pyrimidine as an antiplatelet agent.

positive and Gram-negative bacteria. The results demonstrated that compounds **97–99** exhibited nearly complete inhibition of *Staphylococcus aureus* and *Escherichia coli* growth, respectively. Additionally, compound **100** displayed the highest antibacterial activity against *Bacillus subtilis* and *Pseudomonas aeruginosa*, showing strong efficacy with low minimum inhibitory concentration (MIC) values (Scheme 46).[131–133]

Given the critical role of dihydrofolate reductase (DHFR) inhibition in treating bacterial and protozoal infections, Ibrahim *et al.* evaluated a series of pyrazolo[3,4-*d*]pyrimidine derivatives for their ability to inhibit bacterial DHFR. These compounds were further assessed both *in vitro* and *in vivo* for their antibacterial activity against multiple bacterial strains. Among the tested derivatives, compounds **101** and **102** exhibited potent antibacterial effects, demonstrating low minimum inhibitory concentrations (MICs) against *Pseudomonas aeruginosa* and *Staphylococcus aureus* (Scheme 47).[134]

Maddila *et al.* and Hu *et al.* investigated the antimicrobial potential of novel purine derivatives against both Gram-positive and Gram-negative bacteria. *In vitro* studies revealed that compounds **103** and **104** exhibited superior antibacterial activity against *Escherichia coli* and *Klebsiella pneumoniae* compared to the reference drugs norfloxacin and ciprofloxacin, respectively. Additionally, a purine–metronidazole hybrid (compound **105**) demonstrated significant inhibitory activity, with an MIC value of 6 μM, approximately fourfold more potent than norfloxacin (MIC of 25 μM) (Scheme 48).[79,135]

Finger *et al.* and Nadaf *et al.* synthesized a series of substituted purine derivatives and evaluated their *in vitro* antimycobacterial activity against *Mycobacterium tuberculosis* H37Rv, along with their binding interactions with *M. tuberculosis* enoyl reductase. The results demonstrated that 6*H*-purin-6-ones (**106–108**) and alkyl-substituted purines (**109**) exhibited potent antimycobacterial activity against both drug-sensitive and drug-resistant *M. tuberculosis* strains, while displaying limited cytotoxicity (Scheme 49).[136,137]

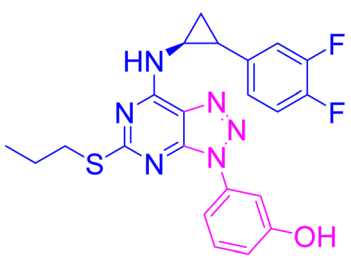
Antiviral

Additionally, pyrazolo[3,4-*d*]pyrimidine derivative **110** was evaluated for antiviral activity against tobacco mosaic virus (TMV). The results demonstrated that compound **110** exhibited remarkable inactivation potential, with a half-maximal effective concentration (EC_{50}) of 53.65 μg mL^{-1}, significantly more potent than the reference antiviral ribavirin (EC_{50} = 150.45 μg mL^{-1}) (Scheme 50).[138]

Alseud *et al.* designed and synthesized a novel series of pyrazolo[3,4-*d*]pyrimidines (HCQ-PPs) as potential anti-SARS-CoV-2 agents. Among the synthesized derivatives, compound **111** exhibited the highest antiviral activity, surpassing that of remdesivir. These findings highlight compound **111** as a promising lead for further optimization and development as an anti-SARS-CoV-2 therapeutic (Scheme 51).[139]

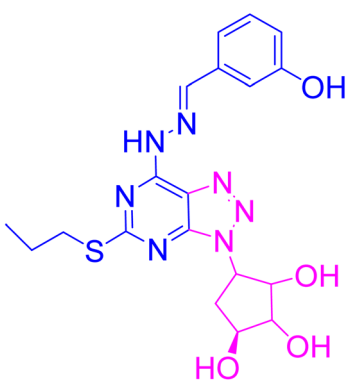
Mohammed *et al.* synthesized a series of tricyclic penciclovir (PCV) derivatives and evaluated their antiviral activity, particularly against herpes simplex virus (HSV). Among them, derivative **112** demonstrated potent antiviral effects, with EC_{50} values of 1.5, 0.8, and 0.8 μM against HSV-1, HSV-2, and HSV-1 TK + VZV Oka strains, respectively. These activities are comparable to or better than the reference antivirals cidofovir (EC_{50} = 2.0, 2.0, and 5.0 μM) and acyclovir (EC_{50} = 0.9, 0.9, and 100 μM) (Scheme 52).[140]

A series of purine nucleoside derivatives incorporating sulfa ethylamine moieties was designed, synthesized, and evaluated



134

Scheme 66 1,2,3-Triazolo[4,5-*d*]pyrimidine as an antiplatelet agent.



135

Scheme 67 Novel 8-azapurine derivative as potent antiplatelet agent compared with ticagrelor.

for antiviral activity against tobacco mosaic virus (TMV), cucumber mosaic virus (CMV), and potato virus Y (PVY). Several derivatives exhibited notable antiviral effects, with compound **113** showing particularly strong protective activity against CMV and PVY, surpassing that of the reference antiviral agent pingyangmycin (Scheme 53).[141]

Furthermore, Deng *et al.* reported the synthesis of novel purine–quinazoline hybrids and evaluated their antiviral activity against tobacco mosaic virus (TMV). *In vivo* bioassays revealed that several purine–quinazoline derivatives exhibited higher antiviral efficacy than the commercial antiviral ribavirin. Notably, compound **114**, containing an 8-fluoroquinazoline moiety, demonstrated curative and protective activities of 65.2% and 60.2%, respectively, surpassing those of ribavirin (50.1% and 57.2%) (Scheme 54).[142]

On the other hand, high-throughput virtual screening identified compound **115** as a promising candidate, supported by molecular dynamics simulations and compliance with Lipinski's rule of five. These results suggest that compound **115** may possess potential as a broad-spectrum antiviral agent against diarrhea-causing viruses (Scheme 55).[143]

Gómez-Sanjuan and colleagues identified a series of 3-aryl-[1,2,3]triazolo[4,5-*d*]pyrimidin-7(6*H*)-ones as selective inhibitors of chikungunya virus (CHIKV) replication. To enhance their physicochemical properties and antiviral potency, a new series of derivatives was synthesized. Among them, compounds **116**, **117**, and **118** demonstrated markedly improved inhibitory activity against various CHIKV strains (Scheme 56).[144,145]

Anti-inflammatory

Somakala *et al.* synthesized a series of *N*-substituted 4-((1-phenyl-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-yl)amino)benzamides and evaluated their *in vitro* anti-inflammatory activity. Among the derivatives, compound **119** exhibited notable anti-inflammatory potential, achieving 83.73% inhibition compared to the standard drug diclofenac sodium (78.05%). Furthermore, compound **119** demonstrated reduced ulcerogenic liability and decreased lipid peroxidation relative to the standard (Scheme 57).[146]

Similarly, several studies have reported the synthesis of pyrazolo[3,4-*d*]pyrimidine derivatives bearing diverse functional groups, which have been evaluated for their anti-inflammatory activity *via in vitro* COX-1/COX-2 inhibition assays. Preliminary results indicated that derivatives **120–123** and **124a–d** exhibited higher COX-1/COX-2 selectivity indexes (SI values) compared to the reference drugs celecoxib and diclofenac sodium (Scheme 58).[147–150]

In other studies, Shi *et al.* and Wang *et al.* synthesized a series of novel pyrazolo[4,3-*d*]pyrimidine analogues and evaluated their anti-inflammatory activity. Structure–activity relationship (SAR) analysis revealed that the introduction of a 3-morpholinopropan-1-amine moiety, a substituted 3,4,5-trimethoxystyryl group at C-5, and an alkylamine at C-7 of the pyrazolo[4,3-*d*]pyrimidine scaffold significantly enhanced anti-inflammatory effects. Among the synthesized derivatives, compounds **125** and **126** were identified as the most potent candidates (Scheme 59).[151,152]

Afifi *et al.* evaluated the anti-inflammatory activity of a series of designed purine-pyrazole derivatives against 15-lipoxygenase (15-LOX). All the tested compounds demonstrated potent 15-LOX inhibitory activity, comparable to the reference inhibitors zileuton, quercetin, and meclofenamate sodium. Among them, purine derivative **127** exhibited the highest potency (Scheme 60).[153]

Pournara *et al.* designed and synthesized 6-piperazinyl-substituted purine analogues incorporating nitric oxide (NO)-donor furoxan and hydrogen sulfide (H_2S)-donor moieties. The anti-inflammatory activity of these derivatives was evaluated by measuring their inhibitory effects on lipopolysaccharide (LPS)-induced IL-1β secretion in human aortic smooth muscle cells (HAoSMCs). The results demonstrated that the compounds bearing [(methylsulfonyl)thio]propanoyl and [(methylsulfonyl)thio]hexanoyl groups (compounds **128** and **129**, respectively) effectively suppressed LPS-induced IL-1β release in HAoSMCs (Scheme 61).[154]

Anti-diabetic activity

The pyrazolo[3,4-*d*]pyrimidine derivative **130**, featuring a bicyclic moiety and a benzamide functionality, exhibited superior anti-diabetic activity, with an IC_{50} value of 1.60 ± 0.48 μM, compared to the reference drug acarbose (IC_{50} = 1.73 ± 0.05 μM) (Scheme 62).[155]

Anti-oxidant activity

Mangasuli *et al.* synthesized a series of coumarin–purine hybrids using a microwave-assisted method and evaluated their *in vitro* antioxidant activity. Among the synthesized derivatives, compound **131** exhibited the highest activity, which is attributed to the presence of a hydroxyl (–OH) group, and demonstrated superior antioxidant potential compared to the standard ascorbic acid (Scheme 63).[156]

Anti-Alzheimer's activity

Sharma *et al.* synthesized three series of caffeine-based triazoles and evaluated their *in vitro* inhibitory activity against acetylcholinesterase (AChE) and β-site amyloid precursor protein cleaving enzyme-1 (BACE-1) at 10 μM. The results indicated that all the derivatives exhibited moderate to potent inhibition. Notably, purine derivative **132** demonstrated dual inhibitory potential against both AChE and BACE-1. Its activity was comparable to the reference compounds cryptolepine and rivastigmine for AChE, and β-secretase inhibitor IV for BACE-1 (Scheme 64).[157]

Antiplatelet aggregation

A series of novel adenosine derivatives was synthesized, with compound **133** identified as the reference molecule. Based on this lead structure, new derivatives can be rationally designed by introducing favorable substituents at specific positions. In particular, incorporating bulky electron-withdrawing groups at

position 6 in region A, such as F, Cl, Br, or $-NO_2$, and stronger hydrogen-bond-donor groups at the C-5′ position, such as –COOH or $-CH_2OH$, is predicted to enhance biological activity (Scheme 65).[158]

Also, a series of 1,2,3-triazolo[4,5-*d*]pyrimidines, structurally related to ticagrelor, was synthesized and evaluated as potential antiplatelet agents. Among the derivatives, compound **134** demonstrated notable antiplatelet activity (Scheme 66).[159]

Moreover, a series of 8-azapurine derivatives was synthesized and their antiplatelet activity was evaluated both *in vitro* and *in vivo* using ticagrelor as a reference. Structure–activity relationship (SAR) analysis and molecular docking studies were also performed. Among the synthesized compounds, derivative **135** exhibited the highest activity (IC_{50} = 0.20 μM), approximately fourfold more potent than ticagrelor (IC_{50} = 0.74 μM). Furthermore, treatment with compound **135** resulted in shorter bleeding times, reduced blood loss, and lower acute toxicity compared to ticagrelor (Scheme 67).[57]

Conclusions

Building on the foundation established in Part I, this review (Part II) highlights the expansion of azolo[*d*]pyrimidine scaffolds through the incorporation of multi-nitrogen azole moieties. The fusion of these azole rings onto the pyrimidine core affords structurally diverse regioisomeric systems, including pyrazolo[3,4-*d*]pyrimidines, pyrazolo[4,3-*d*]pyrimidines, imidazolo[4,5-*d*]pyrimidines (purines), and [1,2,3]triazolo[4,5-*d*]pyrimidines (8-azapurines). Over the past decade, extensive studies have demonstrated their broad biological potential, including anticancer, antimicrobial, antiviral, anti-inflammatory, antioxidant, neuroprotective, anti-diabetic, and antiplatelet activities. Despite these promising advances, several critical challenges and research gaps remain. First, most reported compounds are evaluated primarily through *in vitro* assays, with a relative scarcity of *in vivo* studies and clinical validation, thereby restricting translational potential. Second, mechanistic insights at the molecular level remain limited for many derivatives, hindering rational drug design.

Future research should therefore focus on expanding biological evaluation to include *in vivo* models and pharmacokinetic studies, improving target selectivity and safety profiles and integrating green chemistry approaches and advanced synthetic strategies to access novel derivatives with higher structural complexity.

In conclusion, azolo[*d*]pyrimidine derivatives represent highly versatile and privileged scaffolds in medicinal chemistry. Addressing the current limitations and research gaps will be essential to fully exploit their potential and accelerate the development of next-generation therapeutic agents targeting diverse disease pathways.

Author contributions

Aymn E. Rashad: investigation, writing – original draft, review and editing. Ahmed H. Shamroukh: investigation, writing – original draft, review and editing.

Conflicts of interest

The authors declare no conflicts of interest.

Data availability

The authors confirm that no new experimental data were generated in this study. Data presented in tables and figures are derived from previously published studies, all of which are appropriately cited in the reference list. The authors confirm that this study was carried out using publicly available data.

Acknowledgements

The authors of this work wish to thank the authorities of the National Research Centre for their support in carrying out this study.

References

1 A. E. Rashad, T. El-Malah and A. H. Shamroukh, *Curr. Org. Chem.*, 2024, **28**(16), 1244–1264, DOI: 10.2174/0113852728306820240515054401.
2 S. P. Vemuluri, V. L. Somarapu and L. Eppakayala, *Results Chem.*, 2024, **7**, 101403, DOI: 10.1016/j.rechem.2024.101403.
3 T. Manna, S. Maji, M. Maity, *et al.*, *Mol. Divers.*, 2025, **29**, 817–848, DOI: 10.1007/s11030-024-10870-4.
4 A. S. Limaye, P. Rananaware, A. Ghosh, T. Rajashekarreddy, N. Raghavendrarao, V. Brahmkhatri, R. V. Hegde and R. B. Dateer, *ACS Appl. Bio Mater.*, 2024, **7**, 1790–1800, DOI: 10.1021/acsabm.3c01191.
5 M. S. Raghu, C. B. Pradeep Kumar, K. Yogesh Kumar, M. K. Prashanth, F. Alharethy and B.-H. Jeon, *Bioorg. Med. Chem. Lett.*, 2024, **103**, 129707, DOI: 10.1016/j.bmcl.2024.129707.
6 C. Li, X. Tian, Z. Huang, X. Gou, B. Yusuf, C. Li, Y. Gao, S. Liu, Y. Wang and T. Yang, *J. Med. Chem.*, 2023, **66**, 2699–2716, DOI: 10.1021/acs.jmedchem.2c01647.
7 V. Kamat, D. A. Barretto, B. Poojary, A. Kumar, V. B. Patil and S. Hamzad, *Bioorg. Chem.*, 2024, **143**, 107085, DOI: 10.1016/j.bioorg.2024.107085.
8 E. Korkusuz, Y. Sert, S. Arslan, H. Aydın, İ. Yıldırım, Y. Demir, İ. Gülçin and İ. Koca, *Arch. Pharm.*, 2024, **357**, 2300634, DOI: 10.1002/ardp.202300634.
9 M. Sayed, A. M. Sayed, A. A. El-Rashedy, A. A. Saddik, A. T. Alsaggaf, A. M. K. ElDean, R. Hassanien and M. Ahmed, *Curr. Org. Chem.*, 2024, **28**, 56–64, DOI: 10.2174/0113852728289430231227042754.
10 F. N. Gallo, A. B. Marquez, D. M. Fidalgo, A. Dana, M. Dellarole, C. C. García and M. Bollini, *Eur. J. Med. Chem.*, 2024, **272**, 116465, DOI: 10.1016/j.ejmech.2024.116465.
11 N. Irshad, A.-U. Khan, Alamgeer, S.-U.-D. Khan and M. S. Iqbal, *Biomed. Pharmacother.*, 2021, **139**, 111567, DOI: 10.1016/j.biopha.2021.111567.

12 L. Ravindar, S. A. Hasbullah, K. P. Rakesh, S. Raheem, N. Ismail, L. Y. Ling and N. I. Hassan, *Bioorg. Med. Chem. Lett.*, 2024, **114**, 129992, DOI: 10.1016/j.bmcl.2024.129992.
13 S. M. Awad, Y. M. Zohny, S. A. Ali, S. Mahgoub and A. M. Said, *Molecules*, 2018, **23**, 2913, DOI: 10.3390/molecules23112913.
14 M. Myriagkou, E. Papakonstantinou, G.-E. Deligiannidou, A. Patsilinakos, C. Kontogiorgis and E. Pontiki, *Molecules*, 2023, **28**, 3913, DOI: 10.3390/molecules28093913.
15 N. Polish, M. Nesterkina, N. Marintsova, A. Karkhut, I. Kravchenko, V. Novikov and A. Khairulin, *Acta Chim. Slov.*, 2020, **67**, 934–943, DOI: 10.17344/acsi.2020.5938.
16 M. A. Martina, L. Giannessi and M. Radi, *Eur. J. Org Chem.*, 2023, e202201288, DOI: 10.1002/ejoc.202201288.
17 J. Kaźmierczak-Barańska, K. Halczuk and B. T. Karwowski, *Nutrients*, 2025, **17**, 2206, DOI: 10.3390/nu17132206.
18 T. Xu, F. Li, Z. Feng, C. Dang, Y. Yang, J. Wang, C.-X. Zang, X.-Q. Bao, S.-S. Yu, D. Zhang and R.-B. Wang, *Eur. J. Med. Chem.*, 2025, **284**, 117213, DOI: 10.1016/j.ejmech.2024.117213.
19 D. Durga Devi, S. Manivarman and S. Subashchandrabose, *Karbala Int. J. Mod. Sci.*, 2017, **3**, 18–28, DOI: 10.1016/j.kijoms.2017.01.001.
20 P. Sagar, Y. S. Ozarde, V. S. Vichare, P. P. Mehta and R. V. Gadhave, *Mini-Rev. Med. Chem.*, 2026, DOI: 10.2174/0113895575417710251128043828.
21 S. Smaoui, M. Messaad, I. Dhouib, B. Gargouri and B. Khemakhem, *J. Mol. Struct.*, 2026, **1354**, 144912, DOI: 10.1016/j.molstruc.2025.144912.
22 A. Ragab, M. S. Abusaif, H. M. Selim, O. K. M. Riad, M. H. Helal, A. M. Ali, Y. A. Ammar and G. E. Ahmed, *Bioorg. Chem.*, 2025, **165**, 108977, DOI: 10.1016/j.bioorg.2025.108977.
23 A. Ragab, R. Ayman, M. A. Salem, Y. A. Ammar and M. S. Abusaif, *Eur. J. Med. Chem.*, 2025, **290**, 117499, DOI: 10.1016/j.ejmech.2025.117499.
24 A. A. Gaber, M. Sharaky, A. A. Elmaaty, M. M. Hammouda, A. A. Mourad, S. Y. Elkhawaga and A. A. Al-Karmalawy, *Future Med. Chem.*, 2023, **15**, 1773–1790, DOI: 10.4155/fmc-2023-0156.
25 F. Ran, L. Dong, Y. Liu and G. Zhao, *Lett. Drug Des. Discov.*, 2024, **21**, e020623217579, DOI: 10.2174/1570180820666230602093051.
26 A. A. Al-Karmalawy, A. A. Elmaaty, A. Y. M. Alabdali, S. Shaaban, A. O. A. Khatib, M. H. Abdellattif, A. A. Alnjaa, M. Sharaky, W. M. Eldehna and A. A. Gaber, *New J. Chem.*, 2025, **49**, 6025–6035, DOI: 10.1039/D5NJ00367A.
27 S. Bai, S. Wan, R. Wu, M. Li, S. Tang, F. Wang, L. Chen and X. Lv, *J. Mol. Struct.*, 2026, **1353**, 144715, DOI: 10.1016/j.molstruc.2025.144715.
28 Z. Huang, J. Ge, Y. Chen, F. Zou, Q. Liu, B. Wang, Y. Zhou, C. Wu, W. Wang, A. Wang, J. Liu and Q. Liu, *J. Med. Chem.*, 2025, **68**, 21394–21411, DOI: 10.1021/acs.jmedchem.5c01334.
29 M. E. Khalifa, *J. Mol. Struct.*, 2021, **1229**, 129843, DOI: 10.1016/j.molstruc.2020.129843.
30 K. M. Shaik, K. Pandurangan, T. R. Allaka, *et al.*, *Sci. Rep.*, 2025, **15**, 26853, DOI: 10.1038/s41598-025-95669-5.
31 F. Farooqui, A. R. Khan, M. A. Khan, M. Nasibullah and J. A. Ansari, *Future Med. Chem.*, 2026, **18**, 103–112, DOI: 10.1080/17568919.2025.2594966.
32 N. Rana, P. Grover and H. Singh, *Curr. Top. Med. Chem.*, 2024, **24**, e260124226420, DOI: 10.2174/0115680266290152240110074034.
33 A. Ragab, *RSC Adv.*, 2025, **15**, 3607–3620, DOI: 10.1039/D4RA08271K.
34 A. M. Mohamed, M. N. El-Bayaa, D. H. Elnaggar, N. A. Abdel-Hafez, S. F. Mohamed and W. A. El-Sayed, *Polycycl. Aromat. Compd.*, 2024, **44**, 1470–1494, DOI: 10.1080/10406638.2023.2197603.
35 E. Kabir and M. Uzzaman, *Results Chem.*, 2022, **4**, 100606, DOI: 10.1016/j.rechem.2022.100606.
36 Z.-H. Li, J.-L. Ma, G.-Z. Liu, X.-H. Zhang, T.-T. Qin, W.-H. Ren, T.-Q. Zhao, X.-H. Chen and Z.-Q. Zhang, *Eur. J. Med. Chem.*, 2020, **187**, 111989, DOI: 10.1016/j.ejmech.2019.111989.
37 C. Xu, W. Zhou, G. Dong, H. Qiao, J. Peng, P. Jia, Y. Li, H. Liu, K. Sun and W. Zhao, *Bioorg. Chem.*, 2020, **105**, 104424, DOI: 10.1016/j.bioorg.2020.104424.
38 Z. Liu, T. Zhao, Z. Li, K. Sun, Y. Fu, T. Cheng, J. Guo, B. Yu, X. Shi and H. Liu, *Acta Pharm. Sin. B*, 2020, **10**, 1476–1491, DOI: 10.1016/j.apsb.2019.12.008.
39 S. S. Ragab, N. E. Ibrahim, M. S. Abdel-Aziz, A. A. Elrashedy and A. K. Allayeh, *Results Chem.*, 2023, **6**, 101163, DOI: 10.1016/j.rechem.2023.101163.
40 A. Elkamhawy, M. M. Al-Sanea, C. Song, T. Sim and E. J. Roh, *Bull. Korean Chem. Soc.*, 2015, **36**, 1863–1873, DOI: 10.1002/bkcs.10363.
41 Kh. R. A. Abdellatif and R. B. Bakr, New advances in synthesis and clinical aspects of pyrazolo[3,4-d]pyrimidine scaffolds, *Bioorg. Chem.*, 2018, **78**, 341–357, DOI: 10.1016/j.bioorg.2018.03.032.
42 A. E. Kassab, Pyrazolo[3,4-*d*]pyrimidine scaffold: A review on synthetic approaches and EGFR and VEGFR inhibitory activities, *Arch. Pharm.*, 2023, **356**, e2200424, DOI: 10.1002/ardp.202200424.
43 M. El Hafi, E. H. Anouar, S. Lahmidi, M. Boulhaoua, M. Loubidi, A. S. Alanazi, I. Filali, M. Hefnawy, L. El Ghayati, J. T. Mague, *et al.*, *Molecules*, 2024, **29**, 5020, DOI: 10.3390/molecules29215020.
44 M. A. Abdelgawad, N. A. A. Elkanzi, A. A. Nayl, A. Musa, N. H. Alotaibi, W. A. A. Arafa, S. M. Gomha and R. B. Bakr, *Arab. J. Chem.*, 2022, **15**, 103781, DOI: 10.1016/j.arabjc.2022.103781.
45 I. M. Salem, S. A. El Zohny, M. A. Hayallah and A. M. Hayallah, *Sphinx J. Pharm. Med. Sci.*, 2025, **10**, 19–35.
46 C. M. Marshall, J. G. Federice, C. N. Bell, P. B. Cox and J. T. Njardarson, *J. Med. Chem.*, 2024, **67**, 11622–11655, DOI: 10.1021/acs.jmedchem.4c01122.
47 S. Cruz and J. Portilla, *Chemistry*, 2025, **7**, 106, DOI: 10.3390/chemistry7040106.

48 M. T. M. Nemr, M. A. Abdelaziz, M. Teleb, A. E. Elmasry and Y. A. A. M. Elshaier, *Mol. Divers.*, 2025, **29**, 4765–4785, DOI: 10.1007/s11030-024-11016-2.
49 C. M. Diaz-Montero, E. G. Holvey-Bates, P. A. Rayman, Y. Parker, D. J. Lindner, G. R. Stark and S. De, *Front. Oncol.*, 2026, **15**, 1745967, DOI: 10.3389/fonc.2025.1745967.
50 A. Aboelezz and S. H. Mahmoud, *Medicines*, 2026, **13**, 6, DOI: 10.3390/medicines13010006.
51 Z. Y. Chen, Q. R. Li, L. H. Liao, X. L. Chen, X. L. Xiao, H. Jin, Y. Li, L. N. Wang, C. Liang, Z. Fan, T. F. Yue, C. Y. Yang, X. Q. Luo, Y. L. Tang, L. B. Huang and X. L. Zhang, *Pediatr. Res.*, 2026, DOI: 10.1038/s41390-025-04728-0.
52 J. M. Herrerías, J. M. Esteban, A. M. Caballero-Plasencia, *et al.*, *Dig. Dis. Sci.*, 2003, **48**, 986–991, DOI: 10.1023/A:1023020201005.
53 M. Maciejczyk and M. Pyrka, *Eur. Biophys. J.*, 2023, **52**, 545–557, DOI: 10.1007/s00249-023-01672-x.
54 A. Khandazhinskaya, B. Eletskaya, A. Mironov, I. Konstantinova, O. Efremenkova, S. Andreevskaya, T. Smirnova, L. Chernousova, E. Kondrashova, A. Chizhov, *et al.*, *Int. J. Mol. Sci.*, 2023, **24**, 15421, DOI: 10.3390/ijms242015421.
55 S. Sardar, J. S. C. C. Martins, T. C. Sousa, A. S. M. Silva, M. A. Pinto, F. F. Silveira, T. B. Silva, R. R. F. França, L. C. S. Pinheiro, N. Boechat, *et al.*, *Pathogens*, 2026, **15**, 324, DOI: 10.3390/pathogens15030324.
56 T. Felicetti, M. C. Pismataro, V. Cecchetti, O. Tabarrini and S. Massari, *Curr. Med. Chem.*, 2022, **29**, e260521193605, DOI: 10.2174/0929867328666210526120534.
57 Z. Zhao, Y. Wang, N. Tian, H. Yan and J. Wang, *RSC Med. Chem.*, 2021, **12**, 1414–1427, DOI: 10.1039/d1md00128k.
58 W. Tashie, H. P. de Koning, N. O. Duah-Quashie and N. B. Quashie, *Front. Parasitol.*, 2025, **4**, 1634209, DOI: 10.3389/fpara.2025.1634209.
59 Y. Zhou, W. Wang, C. Lv, J. Wu, S. Lv, L. He, X. Zhao, X. Qin, B. Tian, R. Chen, C. Guo, L. Huang and H. Yu, *Bioorg. Med. Chem.*, 2026, **134**, 118550, DOI: 10.1016/j.bmc.2026.118550.
60 N. Kumar, K. Jangid, V. Kumar, A. R. Dwivedi, V. Kumar, B. Devi, T. Arora, J. Parkash and V. Kumar, *RSC Adv.*, 2026, **16**, 13369, DOI: 10.1039/D5RA08679E.
61 V. Papesch and R. M. Dodson, *J. Org. Chem.*, 1965, **30**, 199–203, DOI: 10.1021/jo01012a046.
62 S. Senda, K. Hirota, T. Asao and Y. Yamada, *J. Chem. Soc., Chem. Commun.*, 1977, 556–557, DOI: 10.1039/C39770000556.
63 K. Hirota, Y. Yamada, T. Asao and S. Senda, *J. Chem. Soc., Perkin Trans. 1*, 1982, 277–284, DOI: 10.1039/P19820000277.
64 X. Li, J. Su, Y. Yang, W. Lian, Z. Deng, Z. Yang, G. Chen, B. Zhang, C. Dong, X. Liu, L. Li, Z. Wang, Z. Hu, Q. Xu and X. Deng, *Eur. J. Med. Chem.*, 2020, **207**, 112755, DOI: 10.1016/j.ejmech.2020.112755.
65 Y. Sun, Y. Sun, L. Wang, T. Wu, W. Yin, J. Wang, Y. Xue, Q. Qin, Y. Sun, H. Yang, D. Zhao and M. Cheng, *Eur. J. Med. Chem.*, 2022, **238**, 114424, DOI: 10.1016/j.ejmech.2022.114424.
66 F. A. Al-Joufi, M. S. Alwethaynani, H. Sarfraz, F. Asif, R. Hussain, A. Sardar, Y. Khan, F. Hassan, A. Y. Binsaleh, N. Al-Hoshani, R. N. Hassan and M. A. Mojally, *J. Mol. Struct.*, 2026, **1359**, 145424, DOI: 10.1016/j.molstruc.2026.145424.
67 Y. Wang, S. Wan, Z. Li, Y. Fu, G. Wang, J. Zhang and X. Wu, *Eur. J. Med. Chem.*, 2018, **155**, 210–228, DOI: 10.1016/j.ejmech.2018.05.054.
68 J. A. Lee, H. R. Kim, H. J. Son, N. Shin, S. H. Han, C. S. Cheong, D. J. Kim and O. Hwang, *Exp. Neurol.*, 2020, **332**, 113387, DOI: 10.1016/j.expneurol.2020.113387.
69 Y. Fu, Y. Wang, S. Wan, Z. Li, G. Wang, J. Zhang and X. Wu, *Molecules*, 2017, **22**, 542, DOI: 10.3390/molecules22040542.
70 Y. Yin, C.-J. Chen, R.-N. Yu, L. Shu, Z.-J. Wang, T.-T. Zhang and D.-Y. Zhang, *Bioorg. Chem.*, 2020, **98**, 103720, DOI: 10.1016/j.bioorg.2020.103720.
71 E. S. Ofitserova, A. A. Shklyarenko, V. A. Tafeenko, *et al.*, *Russ. J. Gen. Chem.*, 2025, **95**, 3509–3516, DOI: 10.1134/S1070363225605691.
72 Z. Ebrahimpour, A. Shiri, M. Bakavoli, S. S. Mohammad, M. Bahreini and F. Oroojalian, *Heterocycl. Commun.*, 2016, **22**, 49–53, DOI: 10.1515/hc-2015-0145.
73 C. Wang, H. Liu, Z. Song, Y. Ji, L. Xing, X. Peng, X. Wang, J. Ai, M. Geng and A. Zhang, *Bioorg. Med. Chem. Lett.*, 2017, **27**, 2544–2548, DOI: 10.1016/j.bmcl.2017.03.088.
74 F. A. Ragab, Y. M. Nissan, E. M. Seif, A. Maher and R. K. Arafa, *Bioorg. Chem.*, 2020, **96**, 103621, DOI: 10.1016/j.bioorg.2020.103621.
75 S. El-Kalyoubi and F. Agili, *Molecules*, 2016, **21**, 1714, DOI: 10.3390/molecules21121714.
76 A. V. Komkov, A. O. Chizhov, A. S. Shashkov and I. V. Zavarzin, *Russ. Chem. Bull.*, 2018, **67**, 1088–1099, DOI: 10.1007/s11172-018-2185-5.
77 A. El-Mekabaty, H. A. Etman, A. Mosbah and A. A. Fadda, *ChemistrySelect*, 2020, **5**, 4856–4861, DOI: 10.1002/slct.202001006.
78 S. Maddila, J. Valand, H. Bandaru, K. Yalagala and P. Lavanya, *J. Heterocycl. Chem.*, 2016, **53**, 319–324, DOI: 10.1002/jhet.2407.
79 S. Maddila, M. Momin, L. Palakondu and C. V. Rao, *J. Saudi Chem. Soc.*, 2016, **20**, 173–177, DOI: 10.1016/j.jscs.2012.06.008.
80 M. F. Polat, İ. D. Şahin, P. Kul, R. C. Atalay and M. Tuncbilek, *Bioorg. Med. Chem. Lett.*, 2024, **106**, 129775, DOI: 10.1016/j.bmcl.2024.129775.
81 F. Doğanç, A. S. Aydın, E. Şahin and H. Göker, *J. Mol. Struct.*, 2023, **1272**, 134200, DOI: 10.1016/j.molstruc.2022.134200.
82 R. Bansal, G. Kumar, S. Rohilla, K.-N. Klotz, S. Kachler, L. C. Young and A. L. Harvey, *Drug Dev. Res.*, 2016, **77**, 241–250, DOI: 10.1002/ddr.21317.
83 R. Yadav, R. Bansal, S. Rohilla and K.-N. Klotz, *Bioorg. Chem.*, 2016, **65**, 26–37, DOI: 10.1016/j.bioorg.2016.01.003.
84 S. Rohilla, R. Bansal and K.-N. Klotz, *Bioorg. Chem.*, 2019, **87**, 601–612, DOI: 10.1016/j.bioorg.2019.03.032.
85 S. El-Kalyoubi and F. Agili, *Molecules*, 2020, **25**, 5205, DOI: 10.3390/molecules25215205.
86 S. El-Kalyoubi and F. Agili, *Arab. J. Chem.*, 2022, **15**, 103669, DOI: 10.1016/j.arabjc.2021.103669.

87 S. A. El-Kalyoubi, H. A. M. Gomaa, E. M. N. Abdelhafez, M. Ramadan, F. Agili and B. G. M. Youssif, *Pharmaceuticals*, 2023, **16**, 716, DOI: 10.3390/ph16050716.
88 M. M. van der Walt and G. Terre'Blanche, *Bioorg. Med. Chem.*, 2015, **23**, 6641–6649, DOI: 10.1016/j.bmc.2015.09.012.
89 R. Harmse, M. M. van der Walt, J. P. Petzer and G. Terre'Blanche, *Bioorg. Med. Chem. Lett.*, 2016, **26**, 5951–5955, DOI: 10.1016/j.bmcl.2016.10.086.
90 D. Marx, L. M. Wingen, G. Schnakenburg, C. E. Müller and M. S. Scholz, *Front. Chem.*, 2019, **7**, 56, DOI: 10.3389/fchem.2019.00056.
91 S. Basu, D. A. Barawkar, V. Ramdas, M. Patel, Y. Waman, A. Panmand, S. Kumar, S. Thorat, M. Naykodi, A. Goswami, B. S. Reddy, V. Prasad, S. Chaturvedi, A. Quraishi, S. Menon, S. Paliwal, A. Kulkarni, V. Karande, I. Ghosh, S. Mustafa, S. De, V. Jain, E. R. Banerjee, S. R. Rouduri, V. P. Palle, A. Chugh and K. A. Mookhtiar, *Eur. J. Med. Chem.*, 2017, **134**, 218–229, DOI: 10.1016/j.ejmech.2017.04.014.
92 S. A. El-Kalyoubi, E. A. Fayed and A. S. Abdel-Razek, *Chem. Cent. J.*, 2017, **11**, 66, DOI: 10.1186/s13065-017-0294-0.
93 S. El-Kalyoubi, F. Agili, W. A. Zordok and A. S. A. El-Sayed, *Int. J. Mol. Sci.*, 2021, **22**, 10979, DOI: 10.3390/ijms22201097.
94 M. Pretze, C. Neuber, E. Kinski, B. Belter, M. Köckerling, A. Caflisch, J. Steinbach, J. Pietzsch and C. Mamat, *Org. Biomol. Chem.*, 2020, **18**, 3104–3116, DOI: 10.1039/D0OB00391C.
95 A. Kucukdumlu, M. Tuncbilek, E. B. Guven and R. C. Atalay, *Acta Chim. Slov.*, 2020, **67**, 70–82, DOI: 10.17344/acsi.2019.5196.
96 J. M. Orduña, N. del Río and M. J. Pérez-Pérez, *Org. Biomol. Chem.*, 2023, **21**, 5457–5468, DOI: 10.1039/D3OB00822C.
97 V. A. Verma, B. Halu, A. R. Saundane and R. S. Meti, *Polycycl. Aromat. Compd.*, 2022, **42**, 3694–3716, DOI: 10.1080/10406638.2020.1871384.
98 H. Lei, S. Fan, H. Zhang, Y. J. Liu, Y. Y. Hei, J. J. Zhang, A. Q. Zheng, M. Xin and S. Q. Zhang, *Eur. J. Med. Chem.*, 2020, **186**, 111888, DOI: 10.1016/j.ejmech.2019.111888.
99 Á. Lorente-Macías, I. Iañez, M. C. Jiménez-López, M. Benítez-Quesada, S. Torres-Rusillo, J. J. Díaz-Mochón, I. J. Molina and M. J. P. de las Infantas, *Arch. Pharm.*, 2021, **354**, 2100095, DOI: 10.1002/ardp.202100095.
100 M. Hisham, B. G. M. Youssif, E. E. A. Osman, A. M. Hayallah and M. Abdel-Aziz, *Eur. J. Med. Chem.*, 2019, **176**, 117–128, DOI: 10.1016/j.ejmech.2019.05.015.
101 T. Ma, Q.-S. Ma, B. Yu and H.-M. Liu, *Eur. J. Med. Chem.*, 2019, **174**, 76–86, DOI: 10.1016/j.ejmech.2019.04.044.
102 G. Chakraborty, R. Mondal, A. K. Guin and N. D. Paul, *Org. Biomol. Chem.*, 2021, **19**, 7217–7233, DOI: 10.1039/D1OB01154E.
103 M. Van Hoof, K. Boon, T. Van Loy, D. Schols, W. Dehaen and S. De Jonghe, *Eur. J. Med. Chem.*, 2022, **235**, 114268, DOI: 10.1016/j.ejmech.2022.114268.
104 V. K. Omanakuttan, M. Froeyen, S. Claes, B. Van Bosstraeten, K. Boon, D. Schols, L. Van Meervelt, T. Van Loy, W. Dehaen and S. De Jonghe, *Eur. J. Med. Chem.*, 2026, **306**, 118597, DOI: 10.1016/j.ejmech.2026.118597.
105 Z.-H. Li, X. Q. Liu, T. Q. Zhao, P. F. Geng, W. G. Guo, B. Yu and H. M. Liu, *Eur. J. Med. Chem.*, 2017, **139**, 741–749, DOI: 10.1016/j.ejmech.2017.08.042.
106 P. F. Geng, X. Q. Liu, T. Q. Zhao, C. C. Wang, Z.-H. Li, J. Zhang, H. M. Wei, B. Hu, L. Y. Ma and H. M. Liu, *Eur. J. Med. Chem.*, 2018, **146**, 147–156, DOI: 10.1016/j.ejmech.2018.01.031.
107 Z.-H. Li, L. Ding, Z. Li, Z. Wang, F. Suo, D. Shen, T. Zhao, X. Sun, J. Wang, Y. Liu, L. Ma, B. Zhao, P. Geng, B. Yu, Y. Zheng and H. Liu, *Acta Pharm. Sin. B*, 2019, **9**, 794–808, DOI: 10.1016/j.apsb.2019.01.001.
108 H. G. Du, M. W. Yu, C. E. Deng, C. H. Lu and S. L. Li, *J. Heterocycl. Chem.*, 2017, **54**, 436–449, DOI: 10.1002/jhet.2601.
109 Y. Wang, H. Yan, C. Ma and D. Lu, *Bioorg. Med. Chem. Lett.*, 2015, **25**, 4461–4463, DOI: 10.1016/j.bmcl.2015.09.002.
110 G. Zhang, H. Xiong, P. Yang, C. Lei, W. Hu, G. Cheng and H. Yang, *Chem. Eng. J.*, 2021, **404**, 126514, DOI: 10.1016/j.cej.2020.126514.
111 D. Debnath, A. Purkayastha, A. Kirillov, R. Ganguly and T. K. Misra, *J. Mol. Struct.*, 2017, **1150**, 118–126, DOI: 10.1016/j.molstruc.2017.08.087.
112 A. K. Eltyshev, P. O. Suntsova, K. D. Karmatskaia, O. S. Taniya, P. A. Slepukhin, E. Benassi and N. P. Belskaya, *Org. Biomol. Chem.*, 2018, **16**, 9420–9429, DOI: 10.1039/c8ob02644k.
113 A. K. Eltyshev, I. A. Agafonova, A. S. Minin, V. A. Pozdina, V. A. Shevirin, P. A. Slepukhin, E. Benassi and N. P. Belskaya, *Org. Biomol. Chem.*, 2021, **19**, 9880–9896, DOI: 10.1039/d1ob01801a.
114 A. K. Eltyshev, A. S. Minin, L. T. Smoliuk, E. Benassi and N. P. Belskaya, *Eur. J. Org Chem.*, 2020, 316–329, DOI: 10.1002/ejoc.201901582.
115 I. M. Salem, S. M. Mostafa, I. Salama, O. I. El-Sabbagh, W. A. H. Hegazy and T. S. Ibrahim, *Bioorg. Chem.*, 2022, **129**, 106207, DOI: 10.1016/j.bioorg.2022.106207.
116 I. M. Salem, S. M. Mostafa, I. Salama, O. I. El-Sabbagh, W. A. H. Hegazy and T. S. Ibrahim, *J. Enzyme Inhib. Med. Chem.*, 2023, **38**, 203–215, DOI: 10.1080/14756366.2022.2142786.
117 I. F. Nassar, M. T. A. Aal, W. A. El-Sayed, M. A. E. Shahin, E. G. E. Elsakka, M. M. Mokhtar, M. Hegazy, M. Hagras, A. A. Mandour and N. S. M. Ismail, *RSC Adv.*, 2022, **12**, 14865–14882, DOI: 10.1039/D2RA01968J.
118 M. Alswah, *Al-Azhar J. Pharm. Sci.*, 2021, **64**, 80–92, DOI: 10.21608/ajps.2021.187757.
119 N. A. A. M. Aziz, R. F. George, T. Nasr, K. El-Adl and G. F. Elmasry, *RSC Adv.*, 2026, **16**, 9590, DOI: 10.1039/D6RA00037A.
120 M. Mohamed, S. Fatahala, R. Abd El-Hameed and M. Mohamed, *Egypt. J. Chem.*, 2025, **68**, 71–98, DOI: 10.21608/ejchem.2024.299076.9897.
121 A. I. Hassaballah, A. M. AboulMagd, M. M. Hemdan, M. H. Hekal, A. A. El-Sayed and P. S. Farag, *RSC Adv.*, 2024, **14**, 1995–2015.

122 Z. Ruzi, K. Bozorov, L. Nie, J. Zhao and H. A. Aisa, *Biomed. Pharmacother.*, 2022, **156**, 113948, DOI: 10.1016/j.biopha.2022.113948.
123 M. T. M. Nemr, A. Elshewy, M. L. Ibrahim, A. M. El Kerdawy and P. A. Halim, *Bioorg. Chem.*, 2024, **150**, 107566.
124 A. Kucukdumlu, M. Tuncbilek, E. B. Guven and R. C. Atalay, *Acta Chim. Slov.*, 2020, **67**, 70–82.
125 M. K. I. Attiogbe, H. Y. Zhao, J. Wang, T. T. Huang, P. P. Yan, Y. N. Liu, W. Li, L. Cao, S. Q. Zhang and Y. X. Cao, *Life Sci.*, 2024, **336**, 122308.
126 M. N. S. Rad, S. Behrouz, K. Shahbazkhani, M. Behrouz, E. Zarenezhad and A. Ghanbariasad, *RSC Adv.*, 2023, **13**, 24656–24673.
127 Y. Wang, Y. Pan, Z. Lv and S. Gou, *Eur. J. Med. Chem.*, 2024, **271**, 116415.
128 Y. Guo, Y. Zou, Y. Chen, D. Deng, Z. Zhang, K. Liu, M. Tang, T. Yang, S. Fu, C. Zhang, W. Si, Z. Ma, S. Zhang, B. Peng, D. Xu and L. Chen, *Bioorg. Chem.*, 2023, **132**, 106386, DOI: 10.1016/j.bioorg.2023.106386.
129 S. Narra and N. Munirathinam, *Results Chem.*, 2024, **8**, 101592, DOI: 10.1016/j.rechem.2024.101592.
130 M. Van Hoof, S. Claes, M. Proj, T. Van Loy, D. Schols, S. Gobec, W. Dehaen and S. De Jonghe, *Eur. J. Med. Chem.*, 2023, **251**, 115240, DOI: 10.1016/j.ejmech.2023.115240.
131 C. Greco, R. Catania, D. L. Balacco, V. Taresco, F. Musumeci, C. Alexander, A. Huett and S. Schenone, *Molecules*, 2020, **25**, 5354.
132 M. M. Hassaneen, F. M. Saleh, T. A. Abdallah, Y. Sh. Mohamed and E. M. Awad, *J. Heterocycl. Chem.*, 2020, **57**(2), 892–912, DOI: 10.1002/jhet.3835.
133 K. B. Alomari, A. L. Alanzy, A. S. Alharbi, N. M. Halawani, N. A. H. Alshammari, J. Qurban, S. Abu-Melha and N. M. El-Metwaly, *J. Heterocycl. Chem.*, 2025, **62**(11), 1642–1657, DOI: 10.1002/jhet.70090.
134 T. S. Ibrahim, I. M. Salem, N. A. Alhakamy, A. M. Al-Mahmoudy and W. A. H. Hegazy, *J. Microbiol. Biotechnol.*, 2025, **35**, e2508055, DOI: 10.4014/jmb.2508.08055.
135 Y. Hu, S. Hu, G. Pan, D. Wu, T. Wang, C. Yu, M. F. Ansari, R. R. Y. Bheemanaboina, Y. Cheng, L. Bai, C. Zhou and J. Zhang, *Bioorg. Chem.*, 2021, **114**, 105096, DOI: 10.1016/j.bioorg.2021.105096.
136 V. Finger, T. Kucera, R. Kafkova, L. Muckova, R. Dolezal, J. Kubes, M. Novak, L. Prchal, L. Lakatos, M. Andrs, M. Hympanova, J. Marek, M. Kufa, V. Spiwok, O. Soukup, E. Mezeiova, J. Janousek, L. Nevosadova, M. Benkova, R. R. A. Kitson, M. Kratky, S. Bősze, K. Mikusova, R. Hartkoorn, J. Roh and J. Korabecny, *Eur. J. Med. Chem.*, 2023, **258**, 115611, DOI: 10.1016/j.ejmech.2023.115611.
137 A. Q. A. Nadaf, M. S. Najare, M. Garbhagudi, S. Mantur, M. G. Sunagar, S. Gaonkar, S. Joshi and I. A. M. Khazi, *Chem. Biodivers.*, 2020, **17**(5), e2000053, DOI: 10.1002/cbdv.202000053.
138 Y. Wang, F. Xu, Y. Zhu, B. Song, D. Luo and G. Yu, *Bioorg. Med. Chem. Lett.*, 2018, **28**(17), 2979–2984.
139 K. Alseud and M. A. Abou-Salim, *Polycycl. Aromat. Compd.*, 2025, **45**(4), 597–618, DOI: 10.1080/10406638.2024.2413429.
140 A. F. Mohammed, G. Andrei, A. M. Hayallah, S. G. AbdelMoty, R. Snoeck and C. Simons, *Bioorg. Med. Chem.*, 2019, **27**, 1023–1033.
141 J. Zhang, F. He, J. Chen, Y. Wang, Y. Yang, D. Hu and B. Song, *J. Agric. Food Chem.*, 2021, **69**(20), 5575–5582, DOI: 10.1021/acs.jafc.0c06612.
142 Y. Deng, M. Chen, J. Yi and Y. Zheng, *Phytochem. Lett.*, 2024, **59**, 10–14, DOI: 10.1016/j.phytol.2023.11.003.
143 O. Ebenezer, M. A. Jordaan, N. Damoyi and M. Shapi, *Int. J. Mol. Sci.*, 2021, **22**(1), 171, DOI: 10.3390/ijms22010171.
144 A. Gómez-Sanjuan, A.-M. Gamo, L. Delang, A. Pérez-Sánchez, S. N. Amrun, R. Abdelnabi, S. Jacobs, E.-M. Priego, M.-J. Camarasa, D. Jochmans, P. Leyssen, L. F. P. Ng, G. Querat, J. Neyts and M.-J. Pérez-Pérez, *ACS Infect. Dis.*, 2018, **4**, 605–619, DOI: 10.1021/acsinfecdis.7b00219.
145 A. Gigante, A. Gomez-Sanjuan, L. Delang, C. Li, O. Bueno, A.-M. Gamo, E.-M. Priego, M.-J. Camarasa, D. Jochmans, P. Leyssen, E. Decroly, B. Coutard, G. Querat, J. Neyts and M.-J. Perez-Perez, *Antiviral Res.*, 2017, **144**, 216–222, DOI: 10.1016/j.antiviral.2017.06.003.
146 K. Somakala, S. Tariq and M. Amir, *Bioorg. Chem.*, 2019, **87**, 550–559, DOI: 10.1016/j.bioorg.2019.03.037.
147 N. Atatreh, A. M. Youssef, M. A. Ghattas, M. Al Sorkhy, S. Alrawashdeh, K. B. Al-Harbi, I. M. El-Ashmawy, T. I. Almundarij, A. A. Abdelghani and A. S. Abd-El-Aziz, *Bioorg. Chem.*, 2019, **86**, 393–400, DOI: 10.1016/j.bioorg.2019.02.014.
148 G. N. Tageldin, S. M. Fahmy, H. M. Ashour, M. A. Khalil, R. A. Nassra and I. M. Labouta, *Bioorg. Chem.*, 2018, **78**, 358–371, DOI: 10.1016/j.bioorg.2018.03.030.
149 E. K. A. Abdelall, P. F. Lamie, A. K. M. Ahmed and S. El-Nahass, *Bioorg. Chem.*, 2019, **86**, 235–253, DOI: 10.1016/j.bioorg.2019.01.031.
150 R. B. Bakr, A. A. Azouz, K. R. A. Abdellatif and J. Enzyme, *Inhib. Med. Chem.*, 2016, **31**(S2), 6–12, DOI: 10.1080/14756366.2016.1186018.
151 J. B. Shi, L. Z. Chen, B. S. Wang, X. Huang, M. M. Jiao, M. M. Liu, W. J. Tang and X. H. Liu, *J. Med. Chem.*, 2019, **62**, 4013–4031, DOI: 10.1021/acs.jmedchem.9b00039.
152 B. S. Wang, X. Huang, L. Z. Chen, M. M. Liu and J. B. Shi, *J. Enzyme Inhib. Med. Chem.*, 2019, **34**, 1121–1130, DOI: 10.1080/14756366.2019.1618291.
153 O. S. Afifi, O. G. Shaaban, H. A. Abd El Razik, S. A. Shams El-Dine, F. A. Ashour, A. A. El-Tombary and M. M. Abu-Serie, *Bioorg. Chem.*, 2019, **87**, 821–837, DOI: 10.1016/j.bioorg.2019.03.076.
154 D. T. Pournara, T. Fotopoulou and G. Paramel, *Chem. Biodivers.*, 2026, **23**(1), e02588, DOI: 10.1002/cbdv.202502588.
155 B. N. Reddy, R. Ruddarraju and G. Kiran, *J. Med. Chem. Drug Discov.*, 2019, **4**(23), 10072–10078.

156 S. N. Mangasuli, K. M. Hosamani and P. B. Managutti, *Heliyon*, 2019, **5**, e01131, DOI: 10.1016/j.heliyon.2019.e01131.
157 M. Sharma, A. Sharma, V. K. Nuthakki, S. Bhatt, U. Nandi and S. B. Bharate, *Drug Dev. Res.*, 2022, **83**, 1803–1821, DOI: 10.1002/ddr.21998.
158 S. Lia, X. F. Bao, C. Lu, C. Ren, G. Liu and H. Du, *Russ. J. Bioorg. Chem.*, 2020, **46**(3), 448–457, DOI: 10.1134/S1068162020030103.
159 E. Goffin, N. Jacques, L. Musumeci, A. Nchimi, C. Oury, P. Lancellotti and B. Pirotte, *Eur. J. Med. Chem.*, 2020, **208**, 112767, DOI: 10.1016/j.ejmech.2020.112767.