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# Click 1,4-regioselective synthesis, characterization, and antimicrobial screening of novel 1,2,3-triazoles tethering fluorinated 1,2,4-triazole and lipophilic side chain

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**Abstract** Base-catalyzed alkylation of 5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**3**) with one or two equivalents of propargyl bromide in presence of triethylamine as catalyst selectively produced the thiopropargylated 1,2,4-triazole **7** in 90 % yield. Under the same reaction conditions, 4-ethyl-5-(4-fluorophenyl)-3-(prop-2-yn-1-ylthio)-1,2,4-triazole (**8**) was produced. Conversely, when the propargylation was carried out in presence of sodium bicarbonate, a mixture of $S,N^4$- (**24**) and $S,N^2$-bis(propargylated) triazoles (**25**) was obtained in 85 % overall yield. The click 1,3-dipolar cycloaddition reaction of the mono- (**7**, **8**) and/or bis(propargylated)-1,2,4-triazoles (**24**) with a variety of long-chain alkyl azides, conducted in *t*-BuOH:$H_2O$ (10:1) in presence of sodium ascorbate and copper(II) sulfate at room temperature, afforded the regioselective 1,4-disubstituted mono- (**14**–**18**) and bis(1,2,3-triazole) derivatives (**26**–**30**) containing a fluorinated 1,2,4-triazole moiety and a lipophilic side chain. The structures of the new products were elucidated by infrared (IR), $^1$H and $^{13}$C nuclear magnetic resonance (NMR), and mass spectrometry. They were also assessed in vitro for their antimicrobial potency against six bacteria (*Bacillus subtilis*, *Streptococcus pneumonia*, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumonia*) and two fungi (*Aspergillus fumigatus* and

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*Candida albicans*). The bioassay results revealed that some of the tested compounds displayed promising antimicrobial activity.

**Keywords** Click synthesis · 1,2,3-Triazoles · 1,2,4-Triazoles · Nonionic surfactants · Antimicrobial activity

## Introduction

In recent years, considerable effort has been focused on synthesis of nonionic surfactants due to their relatively simple synthesis and broad applications in biochemical research and drug delivery systems [1, 2]. They are also widely employed in industry as detergents, solubilizers, and emulsifiers [3]. Heterocyclic surfactants have commanded attention from many research groups around the world; however, progress with such compounds has been limited [4].

Five-membered nitrogen-containing heterocycles, including 1,2,4-triazoles and 1,2,3-triazoles, have gained significant importance in modern heterocyclic chemistry, promising a wide range of medicinal applications, e.g., as antiinflammatory [5], anti-human immunodeficiency virus (HIV) [6], anticancer [7, 8], antibacterial [9], antifungal [10], antiviral [11], and antimalarial agents [12]. Triazole derivatives attached to long hydrocarbon chains form nonionic surfactants, being the most frequently used surfactants in medicinal and industrial chemistry [13].

Recently, synthesis of 1,2,3-triazoles that provide a connection between two pharmacophores has provided an ideal opportunity for design of novel classes of bifunctional bioactive molecules [14, 15]. In fact, there is growing interest in the concept of so-called click chemistry for construction of five-membered nitrogen-containing heterocycles, namely 1,2,3-triazoles, via 1,3-dipolar cycloaddition of terminal alkynes with organoazides [16, 17]. Cu(I)-catalyzed Huisgen ligation of azides and alkynes has been shown to be the most elegant and efficient approach for selective preparation of the regioisomer 1,4-disubstituted 1,2,3-triazole [18, 19].

Presence of a fluorine atom in a given drug structure often dramatically increases its thermal stability and lipophilicity, resulting in an enhanced transportation rate of the drug to an active site [20, 21]. In addition, some fluorinated drugs bearing a 1,2,3-triazole nucleus have generated much research interest in view of their important applications in medicinal chemistry and agrochemistry [22–25].

We describe herein the design and synthesis of a new series of regioselective mono- and bis-1,2,3-triazoles tethering a fluorinated 1,2,4-triazole scaffold and long alkyl side chains as novel classes of nonionic surfactants via a Cu-catalyzed azide–alkyne cycloaddition reaction. The antimicrobial activities of the propargylated 1,2,4-triazoles and their respective 1,2,3-triazoles were also evaluated.

## Results and discussion

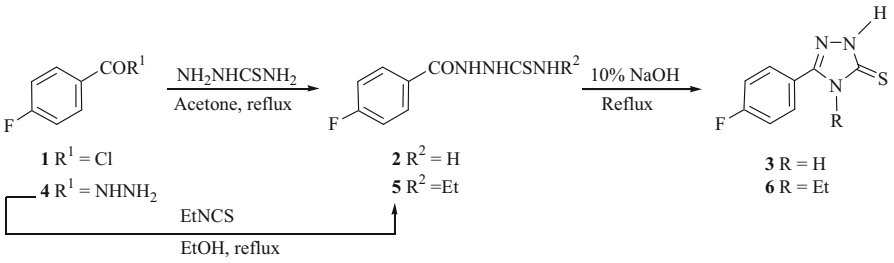
The synthetic protocol adopted for construction of novel 1,2,3-triazole nonionic surfactants with a 1,2,4-triazole tether linked through a flexible methylene spacer involved Huisgen 1,3-dipolar cycloaddition of propargylated 1,2,4-triazoles and

long-chain alkyl azides. The procedure for synthesis of the key intermediate 5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**3**) was based on a modified literature method [26] in which the reaction of 4-fluorobenzoylchloride (**1**) with thiosemicarbazide in refluxing acetone afforded the unsubstituted acylthiosemicarbazide **2** which was thermally cyclized to **3** in 90 % yield in presence of aqueous NaOH (Scheme 1). Furthermore, the 4-ethyl-5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**6**) was previously prepared in lower yield by initial treatment of 4-fluorobenzoylchloride (**1**) with ethylthiosemicarbazide to yield the corresponding ethylthiosemicarbazide **5**, followed by subsequent intramolecular ring closure in aqueous $NaHCO_3$ at 100 °C [27]. In the present study, synthesis of compound **6** was achieved in higher yield (89 %) by condensation of 4-fluorobenzohydrazide (**4**) with ethylisothiocyanate in refluxing ethanol to furnish the corresponding thiosemicarbazide **5** in 92 % yield. The latter compound was cyclized in basic medium (10 % NaOH) to obtain the 4-ethyl-1,2,4-triazole-3-thione (Scheme 1).

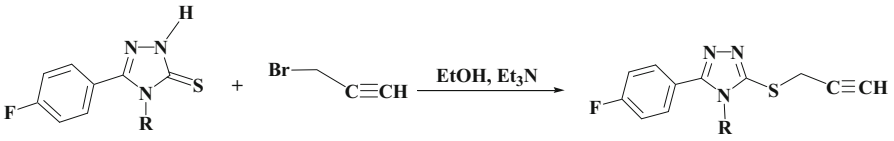
The structures of the substituted triazoles **3** and **6** were established based on their IR, $^{1}$H NMR, and $^{13}$C NMR spectra. Their IR spectra displayed the main characteristic absorptions at 3267–3382 $cm^{-1}$ (NH), 1289–1295 $cm^{-1}$ (C=S), and 1610–1620 $cm^{-1}$ (C=N). Moreover, proton NMR analysis confirmed the absence of signals attributed to the NH groups of their corresponding thiosemicarbazides **2**, **5** and the appearance of characteristic triazole-NH singlets at $\delta$ 13.83–13.95 ppm, thus confirming the presence of the two triazoles in thione form. The phenyl protons resonated at their normal chemical shifts at $\delta$ 7.40–7.87 ppm, and the characteristic ethyl protons appeared as a triplet at $\delta_H$ 1.14 ppm and a quartet at $\delta$ 4.00–4.05 ppm.

Signals related to ethyl carbons were observed at $\delta$ 13.32 and 30.64 ppm in the $^{13}$C NMR spectrum of compound **6**. Formation of triazoles **3** and **6** in thione form was further confirmed by the appearance of the C=S signal at $\delta$ 166.77–169.25 ppm.

The thiopropargylated triazole **7** was regioselectively prepared in satisfactory yield (90 %) according to our reported procedures [28–30] by refluxing the triazole **3** with one equivalent of propargyl bromide in presence of triethylamine as basic catalyst and ethanol as solvent (Scheme 2). The reaction completed in a very short period of time (30 min) with simple workup. Under the same basic reaction conditions ($Et_3N$, EtOH), propargylation of the ethyltriazole **6** furnished the corresponding 5-(4-fluorophenyl)-4-ethyl-1,2,4-triazole-3-thio(prop-2-yne) (**8**) in excellent yield (88 %) (Scheme 2).




**Scheme 1** Synthesis of 5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thiones **3** and **6**




**3** R = H
**6** R = Et

**7** R = H
**8** R = Et

**Scheme 2** Synthesis of the thiopropargylated 1,2,4-triazoles **7** and **8**

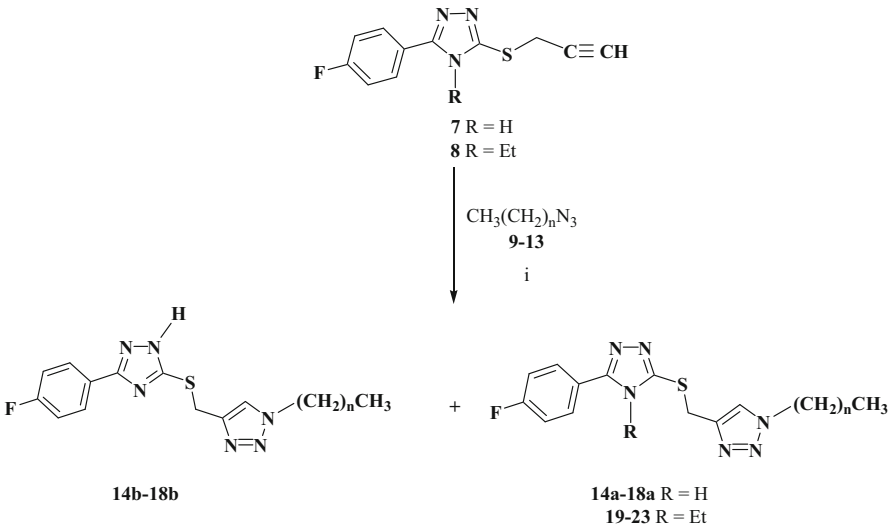
Formation of the thiopropargylated triazoles **7** and **8** was confirmed by their IR, $^{1}$H NMR, $^{13}$C NMR, and mass spectra. In the IR spectra, the sharp band at 3305–3310 cm$^{-1}$ clearly indicated the presence of acetylenic hydrogen ≡C–H, while the C≡C group appeared as a sharp band at 2149–2154 cm$^{-1}$.

The $^{1}$H NMR spectra of compounds **7** and **8** clearly indicated successful *S*-alkylation reaction via the appearance of two new singlets in the aliphatic region. Integration of the first singlet indicated the presence of two protons which appeared at $\delta$ 4.02–4.07 ppm relative to $SCH_2$, while the second singlet at $\delta$ 3.20–3.25 ppm possessed one proton by integration, being related to an sp-CH. In addition, the presence of one broad singlet at $\delta$ 14.53 ppm was attributed to one triazolic NH proton, confirming formation of the mono *S*-propargylated triazole **7**. The spectrum of compound **8** also showed the presence of ethyl protons as a triplet and multiplet at $\delta$ 1.20 and 4.02–4.07 ppm, respectively. The four aromatic protons resonated at their usual chemical shifts (7.38–8.04 ppm).

The $^{13}$C NMR analysis also revealed the incorporation of the propargyl residue by the appearance of diagnostic carbon signals at $\delta$ 20.41–21.85, 74.54–74.80, and 79.51–80.82 ppm, which were attributed to alkyne $SCH_2$ and C≡C groups, respectively. The signals observed at $\delta$ 115.98–164.57 ppm were associated with aromatic and C=N carbons.

A new series of regioselective 1,4-disubstituted mono-1,2,3-triazoles **14**–**23** containing fluorinated 1,2,4-triazoles and lipophilic side chains were successfully synthesized in 80–96 % yield via the Cu(I)-catalyzed click 1,3-dipolar cycloaddition reaction of 5-(4-fluorophenyl)-4*H*/ethyl-1,2,4-triazole-3-thio(prop-2-yne) (**8**, **9**) with freshly prepared azido long-chain alkyl **9**–**13**. The reaction was conducted at room temperature using sodium ascorbate and copper sulfate as catalysts and *t*-BuOH/$H_2O$ (10:1) as solvent (Scheme 3). The azides **9**–**13** were prepared via a metathesis reaction of the appropriate long-chain alkyl halides with sodium azide in acetone–water mixture according to a reported procedure [31].

Formation of compounds **14**–**23** was confirmed based on their spectroscopic data (IR, $^{1}$H NMR, $^{13}$C NMR, and mass). Their IR spectra clearly confirmed the absence of peaks belonging to C≡C at 2149–2154 cm$^{-1}$ and ≡CH at 3305–3310 cm$^{-1}$, thus confirming success of the cycloaddition reaction. The $^{1}$H NMR spectrum of compound **15** revealed the disappearance of the signal attributed to ≡C–H proton at $\delta$ 3.20 ppm of the precursor *S*-alkyne **7** and the appearance of one singlet at $\delta$ 8.00 ppm, which was assigned to the 1,2,3-triazole CH proton. The spectrum also contained two pairs of singlets for the $SCH_2$ protons at $\delta$ 4.43 and 4.52 ppm at ratio



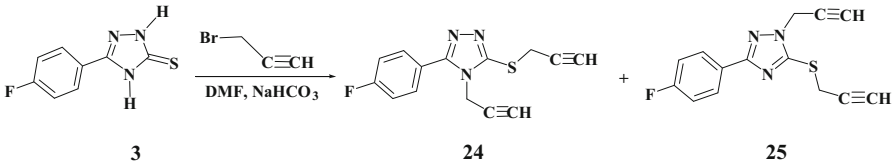

**Scheme 3** Regioselective synthesis of 1,4-disubstituted mono-1,2,3-triazoles **14**–**23** containing 1,2,4-triazole and lipophilic side chains

of 2:3, and for the triazolic NH proton at $\delta$ 14.16 and 14.51 ppm with the same ratio (2:3). This pairing of signals could presumably be due to the tautomerism in 1,2,4-triazole due to the mobility of the NH proton between the N-2 and N-4 nitrogen atoms. Consequently, the two tautomeric forms **15a** and **15b** may exist in equilibrium with predominance of form **15b**. Additionally, the $^{13}$C NMR spectrum revealed the disappearance of the two sp-carbons from their chemical shifts of 74.54 and 80.82 ppm, and the $SCH_2$-carbon resonated at 31.74 ppm. New signals also appeared in the aliphatic region, being assigned to the methylene and methyl carbons. All $sp^2$ carbons were observed in the aromatic region.

The bisalkylation on the triazole nitrogen atom depends on the ability of these basic catalysts ($Et_3N$, $NaHCO_3$) to deprotonate the triazolic N–H group. In this context, all trials attempting to prepare the *S*,*N*-bis(propargylated) triazoles by using two equivalents of $Et_3N$ were unsuccessful. In contrast, alkylation of **3** with 2.2 equivalents of propargyl bromide in refluxing dimethylformamide (DMF) using $NaHCO_3$ as base for 10 h produced a mixture of *S*,$N^4$- and *S*,$N^2$-bis(propargylated)triazoles **24** as major product, and **25** as minor product in overall yield of 85 % (Scheme 4). These results are in accordance with those reported in our previous work concerning regioselective dialkylation of unsubstituted 1,2,4-triazole-3-thione, where the *S*,$N^4$- and *S*,$N^2$-bis(alkylated)triazoles were also formed with preferential formation of the *S*,$N^4$-isomer in high yield [28–30].

The reactions were monitored by thin-layer chromatography (TLC), where the bis(propargylated) triazoles could be identified by their $R_f$ values ($R_f^{N\text{-}4} > R_f^{N\text{-}2}$).

$^1$H NMR analysis was used to differentiate between the N-2 and N-4 isomers, where the $–CH_2$-$N^2$ protons should be deshielded relative to the $–CH_2$-$N^4$ protons.




3 24 25

**Scheme 4** Synthesis of *S*,$N^4$- and *S*,$N^2$-bis(propargylated) triazoles **24** and **25**

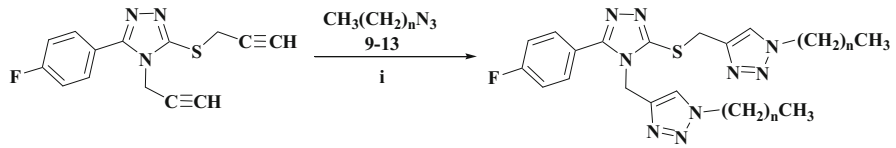
Due to the similar $R_f$ values of the two isomers, the isomeric ratio was determined from the $^1$H NMR spectrum of the crude mixture prior to separation by column chromatography, which was very tedious. The proportion of the *S*-,$N^4$-isomer **24** was much higher than that of the *S*-,$N^2$ isomer (80:20). Moreover, in the $^{13}$ C NMR spectrum, the appearance of the characteristic propargyl groups ($SCH_2$, $NCH_2$, and C≡C) as two sets of signals provided definitive proof of formation of a mixture of *S*,$N^4$- and *S*,$N^2$-bis(propargylated) triazoles **24**, **25**.

The absence of the NH stretch at 3284–3348 $cm^{-1}$ in the IR spectra of compounds **24** and **25**, and the appearance of the characteristic C≡C and ≡C–H bands at 2145–2153 and 3298–3306 $cm^{-1}$, respectively, confirmed the incorporation of two alkyne side chains.

The $NCH_2$- protons appeared as a doublet at $\delta$ 4.98 ppm in the $^1$H NMR spectrum of compound **24**, resonating at lower field than for the isomeric structure **25** at $\delta$ 5.70 ppm. Their $^1$H NMR spectra confirmed the presence of two alkyne protons (≡CH) as two triplets at $\delta$ 2.29 and 2.47 ppm for **24** and as one triplet at $\delta$ 2.29 ppm for **25**.

The presence of characteristic signals in the $^{13}$C NMR spectrum of compound **24** at $\delta$ 22.78 and 38.64 ppm, attributed to $SCH_2$ and $NCH_2$, respectively, confirmed the presence of two alkyne side chains. Conversely, compound **25** showed these carbons at $\delta$ 22.32 and 89.18 ppm, respectively.

The regioselective synthesis of the *S*,$N^4$-bis(1,4-disubstituted 1,2,3-triazoles) **26–30** carrying fluorinated 1,2,4-triazole and lipophilic side chains was performed by a copper-catalyzed click reaction between the *S*,$N^4$-bis(propargylated) triazole **24** and the appropriate long-chain alkyl azides **9–13** in aqueous *t*-BuOH (Scheme 5). The synthesis required regular stirring for 16 h at room temperature to afford the desired 1,2,3-triazoles **26–30** in 84–95 % yield.




24 26-30

i : $CuSO_4$, Na-ascorbate, *t*-BuOH, $H_2O$ (10:1)
n = 9, 10, 11, 13, 15

**Scheme 5** Synthesis of 1,4-disubstituted *S*,$N^4$-bis-1,2,3-triazoles **26–30** containing 1,2,4-triazole and lipophilic side chains

The formation of the $S,N^4$-bis(1,2,3-triazoles) **26–30** was supported by IR, $^1$H NMR, $^{13}$C NMR, and mass spectrometric analyses. In the IR spectrum of compound **29**, the disappearance of the C≡C and ≡C–H absorption bands at 2145 and 3306 cm$^{-1}$, respectively, confirmed involvement of the alkyne linkage in the formation of the 1,2,3-triazole ring. Its $^1$H NMR spectrum showed the appearance of two =C–H protons at $\delta$ 8.06 ppm, confirming the presence of two 1,2,3-triazole moieties. The $SCH_2$ and $NCH_2$ protons resonated as two singlets at $\delta$ 4.59 and 5.39 ppm, respectively. The spectrum also revealed the presence of a triplet at $\delta$ 0.88 ppm, which was attributed to the two terminal methyl groups of the lipophilic side chain. The methylene protons resonated at their appropriate aliphatic region (see "Experimental" section).

Further elucidation of the chemical structure of compound **29** was obtained from its $^{13}$C NMR spectrum, which exhibited signals at $\delta$ 44.41 and 50.64 ppm, which are characteristic of $SCH_2$ and $NCH_2$ carbons, respectively. New signals also appeared in the aliphatic region, being assigned to the lipophilic side-chain carbons. The spectrum also revealed the disappearance of the sp carbons of its starting material **24**.

## Antimicrobial screening

Three Gram-positive bacterial strains (*Bacillus subtilis*, *Streptococcus pneumonia*, and *Staphylococcus aureus*), three Gram-negative bacteria (*Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumonia*), and two fungi (*Aspergillus fumigatus* and *Candida albicans*) were selected as pathogenic strains to evaluate the antimicrobial activity of the synthesized compounds by the broth dilution method [32, 33].

The antimicrobial activity, expressed as minimum inhibitory concentration (MIC) values (Table 1), demonstrated that most of the screened compounds exhibited excellent inhibitory activity with MIC values of 4–8 μg/mL. 5-(4-Fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**3**) and 4-ethyl-5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**7**) displayed moderate to good antibacterial activity towards all bacterial strains with MIC values of 16–31.25 μg/mL, and good to excellent antifungal activity with MIC values of 8–16 μg/mL. Moreover, the mono- (**7**, **8**) and bis(propargylated)-1,2,4-triazoles (**24**) were found to be more potent against Gram-negative bacterial strains with MIC of 16 μg/mL, showing moderate to no antifungal activity against both fungal species.

The antimicrobial screening results for the mono-1,2,3-triazoles **14–23** demonstrated that all of the screened compounds exhibited good to excellent antibacterial activity against Gram-positive bacteria with MIC values of 8–16 μg/mL, and moderate to good inhibitory activity against Gram-negative bacteria with MIC values of 16–31.25 μg/mL.

From the antibacterial bioassay results, it can be concluded that the bis-1,2,3-triazoles **26–30** were more effective against all of the bacterial strains with MIC values of 4–16 μg/mL. Among these, the bis-1,2,3-triazoles **28–30** including $C_{12}$–$C_{16}$ alkyl side chains exhibited promising antibacterial activity with MIC of 4–8 μg/mL against all of the bacterial strains.

**Table 1** Antimicrobial activity of compounds **3–30** expressed as MIC (μg/mL)

| Compound | Gram-positive organisms[a] | | | Gram-negative organisms[b] | | | Fungi[c] | |
|---|---|---|---|---|---|---|---|---|
| | *Sp* | *Bs* | *Sa* | *Pa* | *Ec* | *Kp* | *Af* | *Ca* |
| **3** | 16 | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 8 | 8 |
| **6** | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 | 8 | 16 |
| **7** | 31.25 | 31.25 | 31.25 | 16 | 16 | 16 | 62.5 | 62.5 |
| **8** | 31.25 | 62.5 | 62.5 | 16 | 16 | 16 | 125 | 125 |
| **14** | 16 | 16 | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 |
| **15** | 16 | 16 | 16 | 16 | 16 | 16 | 31.25 | 31.25 |
| **16** | 8 | 16 | 16 | 16 | 16 | 16 | 31.25 | 31.25 |
| **17** | 8 | 8 | 8 | 16 | 16 | 16 | 31.25 | 31.25 |
| **18** | 8 | 8 | 8 | 16 | 16 | 16 | 31.25 | 31.25 |
| **19** | 16 | 16 | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 |
| **20** | 16 | 16 | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 |
| **21** | 8 | 16 | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 |
| **22** | 8 | 8 | 16 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 |
| **23** | 8 | 8 | 8 | 31.25 | 31.25 | 31.25 | 31.25 | 31.25 |
| **24** | 62.5 | 62.5 | 62.5 | 16 | 16 | 16 | 62.5 | 62.5 |
| **25** | 62.5 | 62.5 | 62.5 | 31.25 | 16 | 31.25 | 62.5 | 62.5 |
| **26** | 8 | 16 | 16 | 16 | 16 | 16 | 31.25 | 31.25 |
| **27** | 8 | 8 | 8 | 16 | 16 | 16 | 31.25 | 31.25 |
| **28** | 8 | 4 | 4 | 8 | 4 | 8 | 31.25 | 16 |
| **29** | 8 | 4 | 4 | 8 | 4 | 8 | 31.25 | 16 |
| **30** | 4 | 4 | 4 | 8 | 4 | 8 | 31.25 | 16 |
| Ciprofloxacin | ≤5 | ≤1 | ≤5 | ≤5 | ≤1 | ≤1 | – | – |
| Fluconazole | – | – | – | – | – | – | ≤1 | ≤1 |

[a] *MIC* minimum inhibitory concentration

[b] Gram-positive bacteria: *Streptococcus pneumonia* (RCMB 010010, *Sp*), *Bacillus subtilis* (RCMB 010067, *Bs*), *Staphylococcus aureus* (RCMB 010025, *Sa*)

[c] Gram-negative bacteria: *Pseudomonas aeruginosa* (RCMB 010043, *Pa*), *Escherichia coli* (RCMB 010052, *Ec*), *Klebsiella pneumonia* (RCMB 010058, *Kp*)

[d] Fungi: *Aspergillus fumigatus* (RCMB 02568, *Af*), *Candida albicans* (RCMB 05036, *Ca*)

On the other hand, the preliminary results of the antifungal activity assays revealed that, among the synthesized 1,2,3-triazole derivatives, the greatest antifungal inhibition was displayed by compounds **26–30** against all of the fungal species with MIC values of 16–31.25 μg/mL.

From the results of the antimicrobial activity studies and the structure–activity relationship, it can be concluded that propargylation of the 1,2,4-triazoles **3** and **6** to the corresponding propargylated-1,2,4-triazole derivatives **7**, **8**, and **24** resulted in lower inhibitory activities against Gram-positive bacterial strains and fungal species. In addition, the clubbing of a 1,2,4-triazole moiety with a 1,2,3-triazole carrying long alkyl side chain was found to confer excellent antibacterial activity compared with their propargylated precursors.

## Experimental

### Apparatus and analysis

Melting points were measured on a melt-temp apparatus (SMP10) and are uncorrected. TLC was performed on precoated silica gel (Kieselgel, 0.25 mm, 60 F254, Merck, Germany), and spots were visualized by ultraviolet (UV) light absorption using a developing solvent system of ethyl acetate/hexane. IR spectra were measured using a PerkinElmer 1430 series Fourier-transform infrared (FTIR) spectrometer from potassium bromide pellets. $^{1}$H NMR spectra were recorded using an Advance Bruker NMR spectrometer at 400–600 MHz, whereas $^{13}$C NMR spectra were obtained on the same instrument at 100–150 MHz using tetramethylsilane (TMS) as internal standard. Mass spectra were obtained on a Finnegan mass spectrometer (MAT312) for electron ionization mass spectroscopy (EIMS); High-resolution mass spectroscopy (HRMS) was carried out using a Thermo Finnegan MAT 95XP mass spectrometer.

### Synthesis and characterization of 1,2,4-triazole precursors 3 and 6

*Synthesis of 3-fluorophenyl thiosemicarbazide (**2**)*

A mixture of 4-fluorobenzoyl chloride (**1**) (10 mmol) and thiosemicarbazide (10 mmol) in acetone (100 mL) was heated under reflux for 2 h. The solid thus obtained on cooling was recrystallized from ethanol to give compound **2** in 90 % yield; M.p.: 202–204 °C (lit. [26] 205 °C).

*Synthesis and characterization of 5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**3**)*

To 10 mmol of compound **2**, 100 mL of 10 % aqueous sodium hydroxide solution was added, followed by heating under reflux for 6 h. The reaction mixture was cooled and filtered, and the obtained filtrate was acidified with hydrochloric acid. The crude triazole thus obtained was filtered and recrystallized from ethanol to give compound **3** in 87 % yield; M.p.: 173 °C (lit. [26] 174 °C). IR (KBr) ($cm^{-1}$): 3267–3382 (NH), 3067 (CH-Ar), 1610 (C=N), 1578 (C=C), 1290 (C=S). $^{1}$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 7.60–7.87 (m, 4H, Ar–H), 13.83, 13.89 (2bs, 2H, 2 × NH). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 122.84, 127.52, 129.05, 131.92, 133.45, 137.18, 149.63, 162.52, 165.61 (Ar–C, C=N), 169.25 (C=S).

*Synthesis of 1-(4-fluorobenzoyl)-4-ethylthiosemicarbazide (**5**)*

A mixture of 4-fluorobenzohydrazide (**4**) (10 mmol) and ethyl isothiocyanate (10 mmol) in ethanol (50 mL) was heated under reflux for 6 h. The solid thus obtained on cooling was recrystallized from ethanol to give compound **5** in 92 % yield; M.p.: 201–202 °C (lit. [27] 203 °C).

*Synthesis of 4-ethyl-5-(4-fluorophenyl)-2,4-dihydro-1,2,4-triazole-3-thione (**6**)*

To 10 mmol of compound **5**, 100 mL of 10 % aqueous sodium hydroxide solution was added, followed by heating under reflux for 6 h. The reaction mixture was cooled and filtered, and the obtained filtrate was acidified with hydrochloric acid. The crude triazole thus obtained was filtered and recrystallized from ethanol to give compound **6** in 89 % yield; M.p.: 242–243 °C (lit. [27] 244 °C); IR (KBr) ($cm^{-1}$): 3284–3348 (NH), 3032 (CH-Ar), 1620 (C=N), 1559 (C=C), 1295 (C=S). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 1.14 (t, 3H, $J = 4$ Hz, $CH_3$), 4.00–4.05 (q, 2H, $J = 4$, $J = 8$ Hz, $CH_2$), 7.40–7.46 (m, 2H, Ar–H), 7.74–7.79 (m, 2H, Ar–H), 13.95 (s, 1H, NH); $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 13.32 ($CH_3$), 30.64 ($CH_2$), 116.09, 116.31, 122.61, 122.64, 131.16, 131.25, 150.28, 162.11, 164.58 (Ar–C, C=N), 166.77 (C=S).

*General procedure for synthesis of mono-thiopropargylated 1,2,4-triazoles (**7**, **8**)*

To a solution of compound **3** and/or **6** (10 mmol) in ethanol (50 mL) and triethylamine (10 mmol), propargyl bromide (10 mmol) was added with stirring, followed by heating under reflux for 1 h. Ethanol was removed under reduced pressure, and the resulting crude product was recrystallized from ethanol to afford the desired product.

*5-(4-Fluorophenyl)-3-[(prop-2-ynyl)thio]-4H-1,2,4-triazole (**7**)* Yield 90 %; M.p.: 140–142 °C. IR (KBr) ($cm^{-1}$): 3373 (NH), 3305 (≡CH), 3029 (Ar–H), 2154 (C≡C), 1603 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 3.20 (s, 1H, ≡CH), 4.02 (s, 2H, $SCH_2$), 7.35–7.38 (m, 2H, Ar–H), 8.04 (dd, 2H, $J = 6$, 12 Hz, Ar–H), 14.53 (bs, 1H, NH). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 20.41 ($SCH_2$), 74.54 (≡CH), 80.82 (C≡C), 116.75, 129.06, 162.91, 164.57 (Ar–C, C=N); HRMS (ESI) 234.0496 [M + 1].

*4-Ethyl-5-(4-fluorophenyl)-3-(prop-2-yn-1-ylthio)-1,2,4-triazole (**8**)* Yield 88 %; M.p.: 254–255 °C. IR (KBr) ($cm^{-1}$): 3310 (≡CH), 3084 (Ar–H), 2149 (C≡C), 1614 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 1.20 (t, 3H, $J = 4$ Hz, $CH_3$), 3.25 (t, 1H, $J = 4$ Hz, ≡CH), 4.02–4.07 (m, 4H, $SCH_2$, $NCH_2$), 7.38–7.44 (m, 2H, Ar–H), 7.70–7.74 (m, Ar–H). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 15.10 ($CH_3$), 21.85 ($SCH_2$), 39.82 ($NCH_2$-overlapped with DMSO-$d_6$), 74.80 (≡CH), 79.51 (C≡C), 115.98, 116.19, 123.69, 123.72, 130.82, 130.90, 148.77, 154.23, 161.81, 164.28 (Ar–C, C=N); HRMS (ESI) 262.0776 [M + 1].

*General procedure for synthesis of mono-1,2,3-triazoles (**14–23**)*

To a solution of compound **7** and/or **8** (1.5 mmol) in a 10:1 mixture of *tert*-butanol and water (10 mL), $CuSO_4$ (0.8 mmol), Na-ascorbate (1.1 mmol), and the appropriate long-chain alkyl azide **9–13** (2 mmol) were added with stirring. Then, stirring was continued for 30 h at room temperature. The consumption of the

starting materials was monitored using TLC. Water was added to the reaction mixture, and the product was then extracted three times with dichloromethane and dried over sodium sulfate. Removal of the solvent in vacuum gave the title compounds **14**–**23**, which were crystallized from ethanol.

*1-Decyl-4-(((5-(4-fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1H-1,2,3-triazole (**14**)* Yield 96 %; M.p.: 140–141 °C. IR (KBr) ($cm^{-1}$): 3324 (NH), 3048 (Ar–H), 1610 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.9 (t, 3H, $J = 6$ Hz, $CH_3$), 1.27–1.33 (m, 14H, 7 × $CH_2$), 1.92 (bs, 2H, $NCH_2CH_2$), 4.37 (bs, 4H, $NCH_2$, $SCH_2$), 7.14 (t, 2H, $J = 6$ Hz, Ar–H), 7.29 (bs, 2H, Ar–H), 7.58 (s, 1H, NH), 8.10 (s, 1H, CH-1,2,3-triazole). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.09 ($CH_3$), 22.65, 26.48, 28.94, 29.23, 29.34, 29.45 ($CH_2$), 30.15 ($SCH_2$), 31.84 ($NCH_2$), 115.61, 115.75, 124.11, 128.37, 130.56, 131.56, 149.48, 153.03, 161.37, 163.52 (Ar–C, C=N); MS (EI) 416.23 [$M^+$].

*4-(((5-(4-Fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1-undecyl-1H-1,2,3-triazole (**15**)* Yield 93 %; M.p.: 124–125 °C. IR (KBr) ($cm^{-1}$): 3347 (NH), 3072 (Ar–H), 1604 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.85 (t, 3H, $J = 6$ Hz, $CH_3$), 1.17–1.26 (m, 16H, 8 × $CH_2$), 1.73 (bs, 2H, $NCH_2CH_2$), 4.29 (t, 2H, $J = 6$ Hz, $NCH_2$), 4.43 (s, 1.2H, $SCH_2$), 4.52 (s, 0.8H, $SCH_2$), 7.29 (t, 0.8H, $J = 6$ Hz, Ar–H), 7.40 (t, 1.2H, $J = 6$ Hz, Ar–H), 8.00–8.03 (m, 3H, Ar–H, CH-1,2,3-triazole), 14.16 (s, 0.4H, NH), 14.51 (s, 0.6H, NH). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.41 ($CH_3$), 26.20, 28.78, 29.15, 29.26, 29.38, 29.39, 30.09 ($CH_2$), 31.74 ($SCH_2$), 55.35 ($NCH_2$), 116.14, 116.77, 120.87, 123.92, 125.28, 128.35, 128.97, 130.05, 148.21, 153.44, 161.05, 162.16 (Ar–C, C=N); MS (EI) 430.33 [$M^+$].

*1-Dodecyl-4-(((5-(4-fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1H-1,2,3-triazole (**16**)* Yield 95 %; M.p.: 117–118 °C. IR (KBr) ($cm^{-1}$): 3318 (NH), 3018 (Ar–H), 1600 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.85 (t, 3H, $J = 6$ Hz, $CH_3$), 1.05–1.35 (m, 18H, 9 × $CH_2$), 1.74 (bs, 2H, $NCH_2CH_2$), 4.30 (bs, 2H, $NCH_2$), 4.43 (s, 2H, $SCH_2$), 7.38 (bs, 2H, Ar–H), 8.03 (bs, 3H, Ar–H, CH-1,2,3-triazole), 14.17 (s, 0.4H, NH), 14.50 (s, 0.6H, NH). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.40 ($CH_3$), 22.55, 26.29, 28.78, 29.16, 29.38, 29.26, 29.37, 29.45, 30.07 ($CH_2$), 31.75 ($SCH_2$), 49.92 ($NCH_2$), 116.27, 116.63, 124.01, 128.51, 128.70, 130.12, 148.36, 153.43, 161.29, 162.78 (Ar–C, C=N); MS (EI) 444.35 [$M^+$].

*4-(((5-(4-Fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1-tetradecyl-1H-1,2,3-triazole (**17**)* Yield 84 %; M.p.: 134–135 °C. IR (KBr) ($cm^{-1}$): 3351 (NH), 3037 (Ar–H), 1615 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.85 (dd, 3H, $J = 6$, 12 Hz, $CH_3$), 1.17–1.25 (m, 22H, 11 × $CH_2$), 1.73 (t, 2H, $J = 6$ Hz, $NCH_2CH_2$), 4.29 (t, 2H, $J = 6$ Hz, $NCH_2$), 4.46 (s, 2H, $SCH_2$), 7.37 (bs, 2H, Ar–H), 7.99–8.03 (m, 3H, Ar–H, CH-1,2,3-triazole), 14.19 (s, 0.4H,NH), 14.53 (s, 0.6H, NH). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.49 ($CH_3$), 22.55, 26.19, 28.77, 29.17, 29.25, 29.36, 29.43, 29.47, 29.49, 30.08 ($CH_2$), 31.82 ($SCH_2$), 49.74 ($NCH_2$), 115.82, 116.35,

124.48, 128.90, 129.62, 130.32, 149.08, 153.77, 161.45, 162.51 (Ar–C, C=N); MS (EI) 472.37 [$M^+$].

*4-(((5-(4-Fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1-hexadecyl-1H-1,2,3-triazole (**18**)* Yield 80 %; M.p.: 122–123 °C. IR (KBr) ($cm^{-1}$): 3319 (NH), 3090 (Ar–H), 1611 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.85 (t, 3H, $J = 6$ Hz, $CH_3$), 1.17–1.26 (m, 26H, 13 × $CH_2$), 1.73 (t, 2H, $J = 6$ Hz, $NCH_2CH_2$), 4.29 (bs, 2H, $NCH_2$), 4.43 (s, 1.1H, $SCH_2$), 4.52 (s, 1H, $SCH_2$), 7.29 (s, 1H, Ar–H), 7.42 (d, 1.3H, Ar–H), 8.00–8.03 (d, 3H, Ar–H, CH-1,2,3-triazole), 14.16 (s, 0.4H, NH), 14.50 (s, 0.6H, NH). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.43 ($CH_3$), 22.55, 26.20, 28.77, 29.17, 29.25, 29.36, 29.42, 29.49, 30.08 ($CH_2$), 31.75 ($SCH_2$), 49.76 ($NCH_2$), 116.63, 116.90, 124.02, 128.79, 129.17, 130.87, 149.92, 153.44, 161.23, 162.80 (Ar–C, C=N); MS (EI) 500.44 [$M^+$].

*1-Decyl-4-(((4-ethyl-5-(4-fluorophenyl)-1,2,4-triazol-3-yl)thio)methyl)-1H-1,2,3-triazole (**19**)* Yield 85 %; M.p.: 87–88 °C. IR (KBr) ($cm^{-1}$): 3056 (Ar–H), 1607 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.86 (dd, 3H, $J = 6$, 12 Hz, $CH_3$), 1.13 (dd, 3H, $J = 6$, 12 Hz, $NCH_2CH_3$), 1.17–1.25 (m, 14H, 7 × $CH_2$), 1.71–1.79 (m, 2H, $NCH_2CH_2$), 3.87–3.92 (q, 3H, $J = 6$,12 Hz, $NCH_2CH_3$), 4.30 (q, 2H, $J = 6$ Hz, $NCH_2$), 4.49 (s, 2H, $SCH_2$), 7.43 (dd, 2H, $J = 6$, 12 Hz, Ar–H), 7.71 (ddd, 2H, $J = 6$, 12 Hz, Ar–H), 7.99 (s, 1H, CH-1,2,3-triazole). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.40 ($CH_3$), 15.44 ($NCH_2CH_3$), 22.54, 26.25, 28.51, 28.81, 29.1, 29.29, 29.35, 30.16 ($CH_2$), 31.73 ($SCH_2$), 39.98 ($NCH_2$-overlapped with DMSO-$d_6$), 49.76 ($NCH_2$), 116.49, 116.64, 124.02, 124.66, 131.25, 131.41, 143.00, 150.00, 154.49, 162.95, 164.55 (Ar–C, C=N); MS (ESI) 444.36 [$M^+$].

*4-(((4-Ethyl-5-(4-fluorophenyl)-1,2,4-triazol-3-yl)thio)methyl)-1-undecyl-1H-1,2,3-triazole (**20**)* Yield 83 %; M.p.: 91–92 °C. IR (KBr) ($cm^{-1}$): 3042 (Ar–H), 1602 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.85 (bt, 3H, $CH_3$), 1.10 (bt, 3H, $NCH_2CH_3$), 1.20–1.25 (m, 16H, 8 × $CH_2$), 1.74 (t, 3H, $J = 6$ Hz, $NCH_2CH_2$), 3.91–3.96 (q, 2H, $J = 6$, 12 Hz, $NCH_2CH_3$), 4.31 (t, 2H, $J = 6$ Hz, $NCH_2$), 4.50 (s, 2H, $SCH_2$), 7.40–7.46 (m, 2H, Ar–H), 7.70–7.77 (m, 2H, Ar–H), 8.01 (s, 1H, CH-1,2,3-triazole). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.53 ($CH_3$), 15.41 ($NCH_2CH_3$), 22.55, 26.23, 28.48, 28.82, 29.15, 29.29, 29.41, 30.17 ($CH_2$), 31.74 ($SCH_2$), 39.98 ($NCH_2$-overlapped with DMSO-$d_6$), 49.82 ($NCH_2$), 116.49, 116.63, 124.17, 131.28, 131.33, 162.68, 164.33 (Ar–C); MS (EI) 458.33 [$M^+$].

*1-Dodecyl-4-(((4-ethyl-5-(4-fluorophenyl)-1,2,4-triazol-3-yl)thio)methyl)-1H-1,2,3-triazole (**21**)* Yield 82 %; M.p.: 99–100 °C. IR (KBr) ($cm^{-1}$): 3076 (Ar–H), 1611 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.86 (dd, 3H, $J = 6$, 12 Hz, $CH_3$), 1.11 (t, 3H, $J = 6$ Hz, $NCH_2CH_3$), 1.20–1.27 (m, 18H, 9 × $CH_2$), 1.71–1.76 (m, 2H, $NCH_2CH_2$), 3.89–3.93 (q, 2H, $J = 6$ Hz, $NCH_2CH_3$), 4.32 (dd, 2H, $J = 6$, 12, $NCH_2$), 4.50 (s, 2H, $SCH_2$), 7.42 (dd, 2H, $J = 6$, 12, Ar–H), 7.72 (ddd, 2H, $J = 6$, 12, Ar–H), 8.01 (s, 1H, CH-1,2,3-triazole). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.41 ($CH_3$), 15.41 ($NCH_2CH_3$), 22.55, 26.23, 28.47, 28.82, 29.16, 29.29, 29.40,

29.46, 30.17 ($CH_2$), 31.75 ($SCH_2$), 39.98 ($NCH_2$-overlapped with DMSO-$d_6$), 49.82 ($NCH_2$), 116.49, 116.63, 124.18, 131.28, 131.34, 162.68, 164.33 (Ar–C, C=N); MS (EI) 472.43 [$M^+$].

*4-(((4-Ethyl-5-(4-fluorophenyl)-1,2,4-triazol-3-yl)thio)methyl)-1-tetradecyl-1H-1,2,3-triazole (**22**)* Yield 83 %; M.p.: 106–107 °C; IR (KBr) $\upsilon_{max}/cm^{-1}$ 3028 (Ar–H), 1605 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.86 (t, 3H, $J = 6$ Hz, $CH_3$), 1.13 (dd, 3H, $J = 6$, 12 Hz, $NCH_2CH_3$), 1.17–1.26 (m, 22H, 11 × $CH_2$), 1.71–1.76 (m, 2H, $NCH_2CH_2$), 3.89–3.93 (q, 2H, $J = 6$, 12 Hz, $NCH_2CH_3$), 4.32 (dd, 2H, $J = 6$, 12 Hz, $NCH_2$), 4.50 (s, 2H, $SCH_2$), 7.41 (dd, 2H, $J = 6$, 12 Hz, Ar–H), 7.72 (ddd, 2H, $J = 6$, 12 Hz, Ar–H), 8.00 (s, 1H, CH-1,2,3-triazole). $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.52 ($CH_3$), 15.48 ($NCH_2CH_3$), 22.55, 26.22, 28.80, 29.16, 29.28, 29.39, 29.46, 29.50, 30.17 ($CH_2$), 31.75 ($SCH_2$), 39.98 ($NCH_2$-overlapped with DMSO-$d_6$), 49.92 ($NCH_2$), 116.50, 116.64, 131.26, 131.32, 162.67, 164.32 (Ar–C, C=N); MS (EI) 500.39 [$M^+$].

*4-(((4-Ethyl-5-(4-fluorophenyl)-1,2,4-triazol-3-yl)thio)methyl)-1-hexadecyl-1H-1,2,3-triazole (**23**)* Yield 86 %; M.p.: 115–116 °C. IR (KBr) ($cm^{-1}$): 3067 (Ar–H), 1616 (C=N). $^1$H NMR (600 MHz, DMSO-$d_6$) $\delta$ 0.86 (t, 3H, $J = 6$ Hz, $CH_3$), 1.11 (t, 3H, $J = 6$ Hz, $NCH_2CH_3$), 1.16–1.24 (m, 26H, 13 × $CH_2$), 1.71–1.75 (m, 2H, $NCH_2CH_2$), 3.90–3.93 (q, 2H, $J = 6$, 12 Hz, $NCH_2$), 4.31 (dd, 2H, $J = 6$, 12 Hz, $NCH_2$), 4.50 (s, 2H, $SCH_2$), 7.41 (dd, 2H, $J = 6$, 12 Hz, Ar–H), 7.71 (m, 2H, Ar–H), 8.03 (s, 1H, CH-1,2,3-triazole); $^{13}$C NMR (150 MHz, DMSO-$d_6$) $\delta$ 14.41 ($CH_3$), 15.40 ($NCH_2CH_3$), 22.55, 26.23, 28.48, 28.81, 29.16, 29.28, 29.39, 29.44, 29.46, 29.49, 30.16 ($CH_2$), 31.75 ($SCH_2$), 39.98 ($NCH_2$-overlapped with DMSO-$d_6$), 49.86 ($NCH_2$), 116.49, 116.63, 131.28, 131.34, 162.69, 164.33 (Ar–C, C=N); MS (EI) 528.46 [$M^+$].

*General procedure for synthesis of S,N-bis(propargylated)-1,2,4-triazoles (**24**, **25**)*

To 10 mmol of compound **3** in 25 mL DMF, 22 mmol sodium bicarbonate and 22 mmol propargyl bromide were added, and the reaction mixture was heated under reflux for 8 h. After cooling, it was poured into ice water and the resulting solid was filtered and purified by column chromatography to afford a mixture of $S,N^4$- and $S,N^2$-bis(propargylated)-1,2,4-triazoles **24** and **25** in 85 % overall yield.

*5-(4-Fluorophenyl)-4-(prop-2-yn-1-yl)-3-(prop-2-yn-1-ylthio)-4H-1,2,4-triazole (**24**)* Obtained after separation by column chromatography (eluent hexane:EtOAc, 95:5) in 62 % yield; M.p.: 147–148 °C. IR (KBr) ($cm^{-1}$): 3306 (≡CH), 3043 (Ar–H), 2145 (C≡C), 1606 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 2.29 (t, 1H, $J = 4$ Hz, ≡CH), 2.47 (t, 1H, $J = 4$ Hz, ≡CH), 4.01 (d, 2H, $J = 4$ Hz, $SCH_2$), 4.98 (d, 2H, $J = 4$ Hz, $NCH_2$), 7.08–7.14 (m, 2H, Ar–H), 8.05–8.10 (m, 2H, Ar–H). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 22.78 ($SCH_2$), 38.64 ($NCH_2$), 72.72 (≡CH), 74.60 (≡CH), 75.76 (C≡C), 78.20 (C≡C), 115.43, 115.65, 126.71, 126.74,

128.27, 128.36, 150.81, 161.62, 162.42, 164.89 (Ar–C, C=N). HRMS (ESI) 272.0638 [M + 1].

*5-(4-Fluorophenyl)-2-(prop-2-yn-1-yl)-3-(prop-2-yn-1-ylthio)-4H-1,2,4-triazole (**25**)* Obtained after separation by column chromatography (eluent hexane:EtOAc, 95:5) in 13 % yield; M.p: 168–169 °C. IR (KBr) ($cm^{-1}$): 3298 (≡CH), 3054 (Ar–H), 2153 (C≡C), 1600 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 2.29 (t, 2H, $J = 4$ Hz, 2 × ≡CH), 4.07 (d, 2H, $J = 4$ Hz, $SCH_2$), 5.70 (d, 2H, $J = 4$ Hz, $NCH_2$), 7.08–7.17 (m, 2H, Ar–H), 8.06–8.09 (m, 2H, Ar–H). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 22.32 ($SCH_2$), 72.48 (≡CH), 78.23 (≡CH), 89.18 ($NCH_2$), 96.98 (C≡C), 115.44, 115.66, 126.58, 126.61, 128.38, 128.47, 151.60, 161.53, 162.47, 164.94 (Ar–C, C=N); HRMS (ESI) 272.0642 [M + 1].

*General procedure for synthesis and characterization of bis-1,2,3-triazoles (**26**–**30**)*

To a solution of compound **24** (1.5 mmol) in a 10:1 mixture of *tert*-butanol and water (10 mL), $CuSO_4$ (1.6 mmol), Na-ascorbate (2.2 mmol), and the appropriate long-chain alkyl azide **9**–**13** (4 mmol) were added with stirring. Stirring was then continued for 30 h at room temperature. The consumption of the starting materials was monitored using TLC. Water was added to the reaction mixture, then the product was extracted three times with dichloromethane and dried over sodium sulfate. Removal of the solvent in vacuum gave the title compounds **26**–**30**, which were crystallized from ethanol.

*1-Decyl-4-(((4-((1-decyl-1H-1,2,3-triazol-4-yl)methyl)-5-(4-fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)-methyl)-1H-1,2,3-triazole (**26**)* Yield 95 %; M.p.: 66–67 °C. IR (KBr) ($cm^{-1}$): 3067 (Ar–H), 1616 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 0.87 (t, 6H, $J = 4$ Hz, 2 × $CH_3$), 1.24–1.30 (m, 28H, 14 × $CH_2$), 1.82–1.88 (m, 4H, 2 × $NCH_2CH_2$), 4.24, 4.33 (2t, 4H, $J = 8$ Hz, 2 × $NCH_2CH_2$), 4.59 (s, 2H, $SCH_2$), 5.40 (s, 2H, $NCH_2$), 7.12 (dd, 2H, $J = 8$, 12 Hz, Ar–H), 7.58–7.74 (bs, 2H, Ar–H), 8.07 (s, 2H, CH-1,2,3-triazole). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 14.08 ($CH_3$), 22.65, 26.47, 26.52, 28.95, 28.97, 29.24, 29.34, 29.38, 29.46, 29.70, 30.14, 30.19, 31.85 ($CH_2$), 44.94 ($SCH_2$), 50.63 ($NCH_2$), 115.43, 115.65, 127.04, 128.11, 128.20, 162.35, 164.82 (Ar–C, C=N). HRMS (ESI): 638.4088 [M + 1].

*4-(((5-(4-Fluorophenyl)-4-((1-undecyl-1H-1,2,3-triazol-4-yl)methyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1-undecyl-1H-1,2,3-triazole (**27**)* Yield 89 %; M.p.: 78–79 °C. IR (KBr) ($cm^{-1}$): 3086 (Ar–H), 1602 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 0.88 (t, 6H, $J = 4$ Hz, 2 × $CH_3$), 1.24–1.29 (m, 32H, 16 × $CH_2$), 1.82–1.88 (m, 4H, 2 × $NCH_2CH_2$), 4.25, 4.33 (2t, 4H, $J = 8$ Hz, 2 × $NCH_2CH_2$), 4.58 (s, 2H, $SCH_2$), 5.39 (s, 2H, $NCH_2$), 7.12 (dd, 2H, $J = 8$, 12 Hz, Ar–H), 7.77–7.82 (m, 2H, Ar–H), 8.07 (s, 2H, CH-1,2,3-triazole). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 14.09 ($CH_3$), 28.96, 28.98, 29.30, 29.34, 29.38, 29.52, 29.54, 30.14,

30.20, 31.89 ($CH_2$), 44.47 ($SCH_2$), 50.79 ($NCH_2$), 115.44, 115.65, 127.06, 127.08, 128.09, 128.17, 162.35, 164.82 (Ar–C, C=N); HRMS (ESI) 665.4533 [M + 1].

*1-Dodecyl-4-(((4-((1-dodecyl-1H-1,2,3-triazol-4-yl)methyl)-5-(4-fluorophenyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1H-1,2,3-triazole (**28**)* Yield 84 %; M.p.: 74–75 °C. IR (KBr) ($cm^{-1}$): 3029 (Ar–H), 1601 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 0.88 (t, 6H, $J = 4$ Hz, 2 × $CH_3$), 1.24–1.30 (m, 36H, 18 × $CH_2$), 1.82–1.88 (m, 4H, 2 × $NCH_2CH_2$), 4.26, 4.32 (2t, 4H, $J = 8$ Hz, 2 × $NCH_2CH_2$), 4.59 (s, 2H, $SCH_2$), 5.40 (s, 2H, $NCH_2$), 7.11 (dd, 2H, $J = 8$, 12 Hz, Ar–H), 7.67–7.79 (m, 2H, Ar–H), 8.07 (s, 2H, CH-1,2,3-triazole). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 14.10 ($CH_3$), 22.68, 26.48, 26.54, 28.96, 28.98, 29.33, 29.35, 29.39, 29.52, 29.60, 30.13, 30.18, 31.91 ($CH_2$), 44.58 ($SCH_2$), 50.66 ($NCH_2$), 115.44, 115.65, 127.01, 128.11, 128.20, 162.35, 164.83 (Ar–C, C=N); HRMS (ESI) 694.4680 [M + 1].

*4-(((5-(4-Fluorophenyl)-4-((1-tetradecyl-1H-1,2,3-triazol-4-yl)methyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1-tetradecyl-1H-1,2,3-triazole (**29**)* Yield 84 %; M.p.: 78–80 °C. IR (KBr) ($cm^{-1}$): 3090 (Ar–H), 1608 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 0.88 (t, 6H, $J = 4$ Hz, 2 × $CH_3$), 1.25–1.30 (m, 44H, 22 × $CH_2$), 1.81–1.89 (m, 4H, 2 × $NCH_2CH_2$), 4.23, 4.32 (2t, 4H, $J = 8$ Hz, 2 × $NCH_2CH_2$), 4.59 (s, 2H, $SCH_2$), 5.39 (s, 2H, $NCH_2$), 7.12 (dd, 2H, $J = 8$, 12 Hz, Ar–H), 7.65–7.75 (m, 2H, Ar–H), 8.06 (s, 2H, CH-1,2,3-triazole). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 14.11 ($CH_3$), 22.69, 26.46, 26.51, 28.96, 28.99, 29.35, 29.39, 29.53, 29.60, 29.65, 29.68, 30.17, 30.21, 31.93 ($CH_2$), 44.41 ($SCH_2$), 50.52, 50.64 ($NCH_2$), 115.44, 115.66, 127.06, 128.11, 128.20, 161.33, 162.35, 164.83 (Ar–C, C=N); HRMS (ESI) 750.5380 [M + 1].

*4-(((5-(4-Fluorophenyl)-4-((1-hexadecyl-1H-1,2,3-triazol-4-yl)methyl)-4H-1,2,4-triazol-3-yl)thio)methyl)-1-hexadecyl-1H-1,2,3-triazole (**30**)* Yield 90 %; M.p.: 82–84 °C. IR (KBr) ($cm^{-1}$): 3070 (Ar–H), 1615 (C=N). $^1$H NMR (400 MHz, DMSO-$d_6$) $\delta$ 0.88 (t, 6H, $J = 4$ Hz, 2 × $CH_3$), 1.24–1.30 (m, 52H, 26 × $CH_2$), 1.82–1.89 (m, 4H, 2 × $NCH_2CH_2$), 4.27, 4.32 (2t, 4H, $J = 8$ Hz, 2 × $NCH_2CH_2$), 4.58 (s, 2H, $SCH_2$), 5.40 (s, 2H, $NCH_2$), 7.09 (dd, 2H, $J = 8$, 12 Hz, Ar–H), 7.69–7.81 (m, 2H, Ar–H), 8.07 (s, 2H, CH-1,2,3-triazole). $^{13}$C NMR (100 MHz, DMSO-$d_6$) $\delta$ 14.11 ($CH_3$), 22.69, 26.50, 26.55, 28.97, 29.36, 29.40, 29.53, 29.61, 29.66, 29.69, 31.93 ($CH_2$), 44.50 ($SCH_2$), 50.87 ($NCH_2$), 115.43, 115.65, 127.01, 128.11, 128.19, 162.35, 164.82 (Ar–C, C=N); HRMS (ESI) 806.3018 [M + 1].

## Antimicrobial activity

### *Cell lines*

The newly synthesized 1,2,3-triazole derivatives **3**–**30** were evaluated for antimicrobial activity against several isolated clinical strains from the Regional Center for Mycology and Biotechnology (RCMB). Three Gram-positive bacteria (*Bacillus subtilis* RCMB 010067, *Streptococcus pneumonia* RCMB 010010, and

*Staphylococcus aureus* RCMB 010025), three Gram-negative bacteria (*Escherichia coli* RCMB 010052, *Pseudomonas aeruginosa* RCMB 010043, and *Klebsiella pneumonia* RCMB 010058), and two fungi (*Aspergillus fumigatus* RCMB 02568 and *Candida albicans* RCMB 05036) were selected for use in the assay.

*Antimicrobial evaluation using MIC assay*

Preliminary in vitro evaluation of antimicrobial activity was performed using the broth microdilution method [32, 33]. The minimum inhibitory concentration of the tested compounds was defined as the lowest concentration of each chemical compound in the tubes with no growth of inoculated bacteria/fungi. Thus, 10 mg of analyte was dissolved in dimethylsulfoxide (DMSO, 1 mL), then diluted in culture medium (Mueller–Hinton broth for bacteria and Sabouraud liquid medium for fungi). Additional dilutions with distilled water afforded different concentrations (1, 2, 4, 8, 16, 31.25, 62.5, 125, 250, and 500 mg/mL) of the tested compounds. The tubes were incubated at 37 °C for 24 h.

## Conclusions

We report on Huisgen click synthesis of novel bioactive 1,2,3-triazole nonionic surfactants carrying a 1,2,4-triazole scaffold. The approach involved first regioselective propargylation of the 1,2,4-triazole-3-thione in presence of an appropriate basic catalyst, followed by Cu(I)-catalyzed 1,3-dipolar cycloaddition reaction with several long-chain alkyl azides to afford corresponding *S*-mono- and $S,N^4$-bis(1,2,3-triazoles) tethering fluorinated 1,2,4-triazole and lipophilic side chains. The antimicrobial bioassay results revealed that the structural combination of 1,2,3-triazole, 1,2,4-triazole, and lipophilic side chains furnished novel, potentially active, antibacterial and antifungal agents.

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