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Article

Efficient Synthesis of Thiazole-Fused Bisnoralcohol Derivatives as Potential Therapeutic Agents

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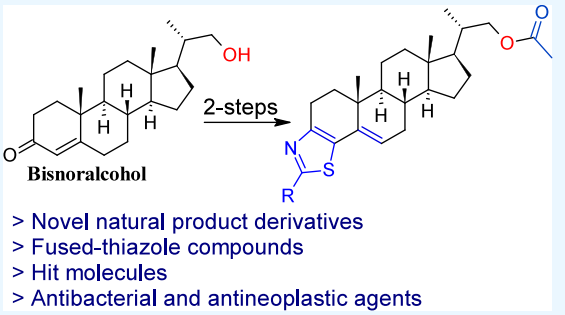
ABSTRACT: Thiazole derivatives are known for a wide range of therapeutic properties. Bisnoralcohol is an inexpensive natural product obtained by the biodegradation of sterols. This article describes an efficient synthesis of a library of thiazole-fused bisnoralcohol derivatives. These novel compounds have been studied for their antineoplastic and antibacterial properties, which led to the discovery of hit compounds with therapeutic potential. The antibacterial compound is noncytotoxic and nonhemolytic against cancer cell lines and sheep red blood cells, respectively. Several of the antineoplastic compounds showed activity against human cancer cell lines with growth inhibition at submicromolar concentration.



INTRODUCTION

Natural products and their derivatives are the leading sources of drugs and bioactive compounds.[1] These compounds cover the majority of drugs to treat cancer and bacterial infections.[2] Bisnoralcohol (BA) is an inexpensive biodegraded product of sterols. This steroidal compound has been used as a precursor for the synthesis of progestational and adrenocortical hormones.[3] BA has been utilized to synthesize several bioactive compounds such as ursodeoxycholic acid,[4] parathiosteroids,[5] FF-Mas,[6] squalamine,[7] and its analogues.[8] Qui et al. utilized BA to synthesize a series of degraders of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase that lowers cholesterol.[9]

The thiazole heterocycle is a privileged scaffold in drug discovery, and this nucleus is a cornerstone of several approved drugs and numerous natural products with a wide range of bioactivities.[10] Dasatinib, mirabegron, and edoxaban are widely used thiazole-containing approved drugs.[11] Due to several therapeutic uses of thiazole derivatives, a number of synthetic methodologies have been reported to synthesize these compounds.[12] A myriad number of thiazole derivatives have been reported that show antibacterial and antineoplastic properties.[13] In our effort to develop domino reactions to synthesize small molecule heterocycles, we have reported the synthesis of thiazoline and thiazole derivatives and fused thiazoles with natural products such as cholestenone, androstenone, progesterone, and nootkatone (Figure 1) (**1**−**4**).[14] We have found thiazole-fused androstenones (**1**) and ethisterones (**2**) as potent growth inhibitors of cancer cell lines including melanoma and renal cancer cell line panels.[15] Thiazole-fused nootkatones (e.g., **3**) are found to be potent growth inhibitors of drug-resistant bacteria such as methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant enterococci (VRE).[16] Encouraged by these results, we envisaged the synthesis of fused thiazole-bisnoralcohol derivatives as potential cytotoxic agents and these compounds will be less lipophilic than androstenone and ethisterone derivatives.

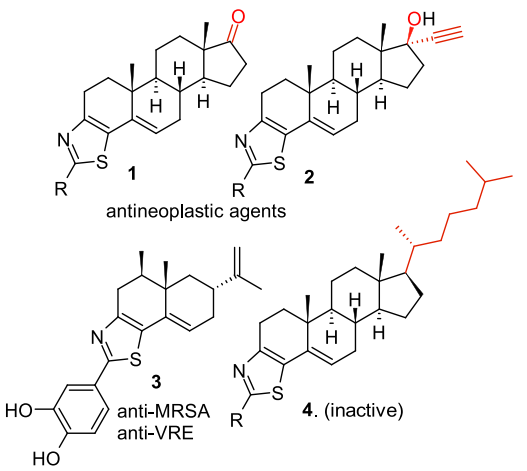


Figure 1. Fused thiazole derivatives as therapeutically important compounds.

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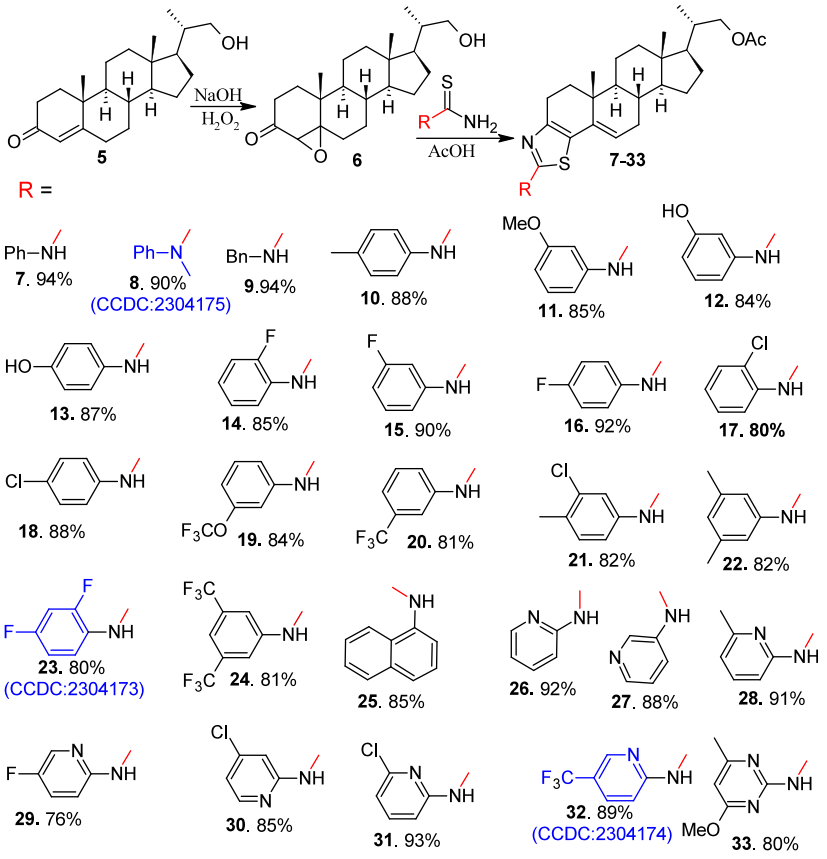
Scheme 1. Synthesis of Fused Thiazolo-Bisnoralcohol Derivatives (7−33) by Reacting Substituted Thioureas with Epoxy-Bisnoralcohol (6), Which Was Synthesized from Commercially Available Bisnoralcohol (5)



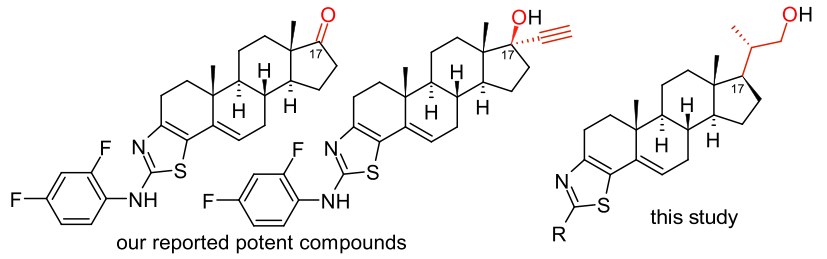


Figure 2. Design of potential antineoplastic compounds based on our previous results.

RESULTS AND DISCUSSION

Synthesis of Fused Thiazolo-Bisnoralcohol. The reaction of commercially available BA (**5**) was treated with $NaOH/H_2O_2$ to get the epoxyketone derivative (**6**). This product is formed in a quantitative yield. The reaction of the epoxyketone (**6**) with different thiourea derivatives formed fused thiazole compounds (Scheme 1). We used acetic acid as a promoter and solvent for this reaction. *N*-Phenyl (**7**) and *N*-methyl-*N*-phenyl (**8**) substituted thioureas reacted efficiently with the electrophile (**6**) to form products in 94 and 90% yield, respectively. In addition to the fused thiazole formation, the hydroxyl group was acetylated in the reaction condition. A *N*-benzyl product (**9**) was formed efficiently in 94% isolated yield. Electron-donating groups such as methyl (**10**), methoxy (**11**), and hydroxy (**12** and **13**) groups on the phenyl of thiourea formed the products in good yields. Fluoro phenyl derivatives (**14**, **15**, and **16**) were obtained in ∼90% yield. Ortho (**17**) and para (**18**) chlorophenyl substituted compounds were obtained under the reaction conditions in similar average yields. Trifluoromethoxy and trifluoromethyl phenyl thiourea formed the products **19** and **20** efficiently by reacting with epoxy-bisnoralcohol (**6**). Disubstituted *N*-phenyl thioureas reacted with the electrophile to give the corresponding products (**21**, **22**, **23**, and **24**) without compromising the average yield. *N*-Naphthyl aminothiazole (**25**) formed in 85% yield by using the standard conditions. 2-Pyridyl and 3-pyridyl substituted compounds (**26** and **27**, respectively) were formed in an average of 90% yield. Substituted pyridyl compounds (**28**, **29**, **30**, **31**, and **32**) were obtained in 76 to 93% yields. Finally, yet importantly, the reaction of substituted *N*-pyrimidine thiourea with the electrophile formed the product **33** in 80% yield. Thus, we can conclude that this methodology is robust to synthesize the fused thiazoles of bisnoralcohol with different thiourea derivatives. These compounds have been thoroughly characterized by ^1H and ^{13}C NMR spectroscopy. Furthermore, single-crystal X-ray diffraction of three compounds, **8** (CCDC: 2304175), **23** (CCDC: 2304173), and **32**

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Table 1. Cytotoxicity Data of Hit Compounds (16, 20, and 23)[a]

		16		20		23	
panel	cell lines	GI_{50}	TGI	GI_{50}	TGI	GI_{50}	TGI
CNS cancer	SF-295	1.74	5.23	1.03	2.78	1.80	12.2
	SF-539	1.91	12.8	1.33	5.25	5.33	26.4
	SNB-75	9.7	>100	0.52	1.02	13.6	93.0
renal cancer	786-0	2.49	3.46	1.19	4.08	2.99	17.3
	A498	12.8	62.9	2.58	8.95	19.2	>100
	RXF393	0.60	3.51	0.30	1.14	0.85	3.39
breast cancer	HS 578T	3.02	26.6	0.62	5.82	2.25	23.6
	BT-549	3.72	27.8	2.2	37	3.64	20.2

[a]GI50 is the concentration that causes 50% growth inhibition, and TGI is the total growth inhibition in μM.

(CCDC: 2304174), further confirms the structure of these compounds (Figures S86−S90 and Table S1).

Cytotoxicity Studies. Previously, we have found thiazolo-androstenone and thiazolo-ethisterone derivatives as potent antineoplastic agents, particularly against melanoma and renal cancer cell lines.[14,17] The structure of bisnoralcohol is similar to androstenone and ethisterone derivatives except for some differences on carbon-17. We were expecting the primary alcohol of bisnoralcohol part would increase not only the favorable pharmacological properties but also the solubility of the designed compounds compared to our previous fused thiazole derivative (Figure 2).

For preliminary antineoplastic studies, we submitted these novel compounds to the Development Therapeutics Program (DTP) of the National Cancer Institute. We found several hit compounds with significant activity against many cancer cell lines.

As shown in Table 1, 4-fluorophenyl substituted compound (**16**) inhibited the growth of central nervous system (CNS) cancer cell lines SF-295 and SF-539 with 50% growth inhibition (GI_{50}) values of 1.74 and 1.91 μM, respectively. The total growth inhibition (TGI) value of this compound (**16**) was 5.23 μM against the SF-295 cell line (Figure S91). The trifluoromethyl substituted compound (**20**) was very effective in inhibiting the growth of CNS cancer cell lines with GI_{50} and TGI values as low as 1.03 and 2.78 μM, respectively (Figure S92). 2,4-Difluorophenyl derivative (**23**) showed potent GI_{50} and moderate TGI values against the SF-295 cell line. SF-295, SF-539, and SNB-75 are human glioblastoma cell lines (Figure S93). These results are very significant as glioblastoma is one of the most malignant types of CNS cancer, and despite advances in cancer treatment, this cancer is mostly incurable.[18] These compounds have significant potential to cross the blood brain barrier (BBB) because of their molecular weight (around 500 Da), optimum number of hydrogen bond formation, and lipid solubility.[19] These compounds are very effective against the renal cancer cell line panel, as shown in Table 1. The RXF393 cell line showed the most susceptibility against hit compounds with GI_{50} values at sub-μM concentration and TGI values in low single digits. These compounds (**16**, **20**, and **23**) are also effective in inhibiting the growth of breast cancer cell lines with GI_{50} values as low as 0.62 μM and with moderate TGI values. HS 578T and BT-549 are triple-negative breast cancer (TNBC) cell lines, and finding treatment for this cancer is very challenging.[20] Thus, these hit compounds will be a very good starting point for developing antineoplastic agents targeting triple negative breast cancer.

Antimicrobial Studies. In continuation of our efforts to find potent antibacterial agents,[21] we tested these novel compounds for their potential antibacterial properties. Most of these compounds did not show any significant antibacterial properties at 64 μg/mL concentration. Nonetheless, one of these compounds inhibited the growth of *S. aureus* strains with a minimum inhibitory concentration (MIC) of 32 μg/mL (Table 2). This compound (**24**) also inhibited the growth of *Staphylococcus epidermidis* moderately.

Table 2. Antimicrobial Properties of the Hit Compound 24[a]

	Sa23	Sa99	SaN	Se	Ef12	Ef99	Efm21	Bs
24	32	32	64	64	>64	>64	>64	>64
Van	1	4	1	2	2	>64	>64	0.25

[a]Antibiotic-susceptible *S. aureus* ATCC 25923 (Sa23), antibiotic-resistant *S. aureus* ATCC 700699 (Sa99), *S. aureus* Newman (SaN), *S. epidermidis* ATCC 700296 (Se), antibiotic-susceptible *E. faecalis* ATCC 29212 (Ef12), *E. faecalis* ATCC 51299 (Ef99)), vancomycin-resistant *E. faecium* ATCC 700221 (Efm21), *B. subtilis* ATCC 6623 (Bs). Vancomycin (Van) was used as a positive control. DMSO (2.5%) and growth media were used as negative controls.

Other tested strains of bacteria such as *Enterococcus faecium*, *Enterococcus faecalis*, and *Bacillus subtilis* were unaffected by these compounds. Thus, this compound is a moderate growth inhibitor of staphylococci strains of bacteria, which is very significant as *S. aureus* is an important bacterium of SPEAKS (*S. aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Acinetobacter baumannii*, *Klebsiella pneumoniae*, and *Streptococcus pneumoniae*) pathogens.[22]

Cytotoxicity and Hemolysis Studies. Furthermore, this compound (**24**) did not show any noticeable activity against 60 cancer cell lines at 10 μM concentration (Figure S94). We tested the possible hemolytic activity at 16, 32, and 64 μg/mL concentrations of the moderately antistaphylococci compound **24**.

As can be seen in Figure 3, this compound (**24**) did not cause any hemolysis up to the maximum tested concentration, 64 μg/mL. Thus, compound **24** is a noncytotoxic, non-hemolytic, and moderately active antibacterial agent. Compounds showing activity against CNS, renal, and breast cancer cell lines did not show any noticeable activity against the tested bacterial strains. Therefore, these compounds are not generally toxic to human cell lines and bacteria, rather they work on specific molecular targets.

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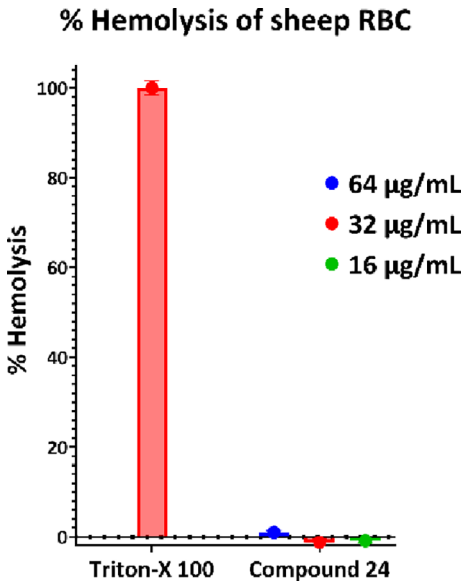


Figure 3. Hemolytic studies of the hit compound **24**.

EXPERIMENTAL METHODS

General Methods. All reactions were carried out in the standard air atmosphere in round-bottom flasks. All reacting materials, reagents, and solvents were bought from Fischer Scientific (Hanover Park, IL, USA) and Oakwood Chemical (Estill, SC, USA). No chemicals were further purified. The infrared (IR) spectral data were obtained in a Nicolet iS 10 FTIR spectrometer (Thermo Fisher Scientific Inc., Waltham, MA, USA) in KBr pellets. Proton (^{1}H) and carbon (^{13}C) spectra were recorded on a JEOL with 400 MHz for ^{1}H, 101 MHz for ^{13}C, and 100 MHz for fluorine decoupled ^{13}C in DMSO-d_6 and $CDCl_3$ solvent. ^{1}H NMR spectra are described in chemical shifts (δ, ppm), and multiplicity are designated as follows: s = singlet, d = doublet, t = triplet, q = quartet, dd = doublet of doublets, ddd = doublet of doublets of doublets, td = triplet of doublets, m = multiplet. A Bruker Apex II-FTMS was used to obtain high resolution mass spectroscopy (HRMS) data.

*Synthesis of Thiazolo-Bisnoralcohol. Synthesis of Epoxy-Bisnoralcohol (**6**).* Bisnoralcohol (1 mmol), dichloromethane (3 mL), methanol (4 mL), 10% NaOH (0.28 mL), and 30% H_2O_2 (0.56 mL) were combined and stirred for 12 h in a round-bottom flask. The solvent was evaporated to get the solid product. Methanol was added to precipitate solid impurities. Filtration followed by evaporating methanol with the hexane azeotrope gave pure epoxy-bisnoralcohol (**6**) as a white powder (**6**).

*Synthesis of Thiazolo-Bisnoralcohol (**7**−**33**).* Epoxy-bisnoralcohol (0.5 mmol) and thiourea derivative (0.55 mmol) were taken in acetic acid (5 mL) solvent in a round-bottom flask. The mixture was heated for 8 h at 100 °C. Following the completion of the reaction, sodium bicarbonate was added slowly to quench some of the acidic acid, and then, water was added to precipitate the product. To get a pure product, the precipitate was thoroughly washed with water and vacuum-dried. In case of impurities, recrystallization with acetonitrile gave pure products.

Cytotoxic Studies. For cytotoxicity screening of the thiazolo-bisnoralcohol compounds, we performed a resazurin cell viability assay in an SK MEL 5 melanoma cell line.[23] We seeded 6000 cells on each well of the 96-well plate that was incubated for 24 h at 37 °C in the presence of 5% CO_2. Each of the compound was used in triplicate at 50 μM concentration with proper controls incubating for an additional 24 h. Resazurin solution was added to a final concentration of 25 μg/mL in each well and incubated for 4 h. Fluorescence of reduced resazurin was measured at 560 nm excitation and 600 nm emission using a Synergy HTX Multimode plate reader (BioTek, Winooski, VT, USA). Cell viability was calculated in comparison to the vehicle control (1% DMSO).

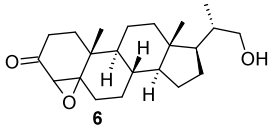
Antimicrobial Studies. Compounds were screened against various bacterial strains at 64 μg/mL concentration. Compounds at single concentration were incubated with bacterium strain diluted to approximately 10^6 CFU/mL in the cation-adjusted Muller Hinton broth (CAMHB) in triplicates in a transparent polystyrene 96-well plate and then incubated for 18−20 h, after which the wells were observed for visual turbidity. The compound wells that did not exhibit visible turbidity were scored as active compounds against the bacterial strain and were subsequently tested to determine the minimum inhibitory concentration (MIC).

The MIC values of active compounds against various bacterial strains were determined following the CLSI micro-dilution technique. Briefly, compounds were diluted 2-fold down the column of a 96-well plate using CAMHB media, and then approximately 10^6 CFU/mL test bacteria were added to each well. The plate was incubated for 18−20 h at 35 °C, after which the wells were examined for visible turbidity. The minimum concentration of the compound that exhibited no visible turbidity was recorded as MIC. The MIC was confirmed in three different experiments on different days.

Hemolysis Studies. The hemolysis assay to determine the hemolytic activity of the compounds was carried out as described previously with few modifications.[24] Briefly, 5% sheep RBCs were incubated with various concentrations of the compound for 30 min at 37 °C in a transparent polystyrene 96-well plate. Following incubation, the plate with samples was centrifuged at 500g for 5 min to a pellet intact RBCs. The supernatant was then carefully transferred to a new 96-well plate, and the absorbance was spectrophotometrically measured at 540 nm using the BioTek Epoch microplate spectrophotometer. The percent of RBC hemolysis was quantified relative to that of the positive control samples lysed with 1% Triton-X 100. All samples were run in triplicates.

EXPERIMENTAL DATA

(1*S*,2*R*,11*S*,12*S*,15*R*,16*S*)-15-[(1*S*)-2-Hydroxy-1-methyl-ethyl]-2,16-dimethyl-7-oxapentacyclo-[9.7.0.0^{2,8}.0^{6,8}.0^{12,16}]octadecan-5-one (6).



6

White powder (294 mg, 85%); IR (KBr pellet, cm^{-1}) 3408, 2942, 1712, 1603, 1445; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 3.61−3.58 (m, 1H), 3.34−3.30 (m, 1H), 2.94 (s, 1H), 2.40−2.05 (m, 3H), 1.97 (d, *J* = 12.6 Hz, 1H), 1.83−1.70 (m, 4H), 1.61−1.46 (m, 4H), 1.40−0.96 (m, 16H), 0.68 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 207.1, 70.4, 67.9, 62.7, 55.6, 52.4, 46.4, 42.8, 39.3, 38.8, 37.2, 35.1, 32.6, 30.5, 29.9, 27.7, 26.2, 24.4, 21.6, 19.0, 16.8, 12.1. HRMS (ESI-FTMS) Mass

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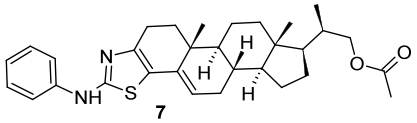
(m/z): calcd for $C_{22}H_{34}O_3$ $[M + H]^+$ = 347.2580, found 347.2571.

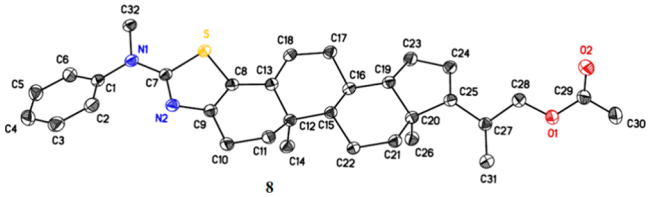
[(2*R*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-Anilino-2,18-dimethyl-8-thia-6-azapentacyclo[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]-icosa-5(9),6,10-trien-17-yl]propyl] Acetate (7).



7

Gray solid (474 mg, 94%); IR (KBr pellet, cm^{-1}) 3446, 2935, 1730, 1603; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.34−7.23 (m, 4H), 7.06 (t, J = 7.0 Hz, 1H), 5.45 (s, 1H), 4.06−4.04 (m, 1H), 3.77 (dd, J = 10.3, 7.8 Hz, 1H), 2.65−2.63 (m, 2H), 2.20−2.16 (m, 1H), 2.09 (s, 2H), 2.05−1.96 (m, 3H), 1.81−1.61 (m, 6H), 1.51−0.97 (m, 14H), 1.02−0.97 (m, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 177.0 171.6, 163.6, 142.2, 139.9, 135.9, 129.5, 123.4, 118.9, 118.5, 69.6, 56.5, 52.7, 47.9, 42.6, 39.5, 36.6, 35.9, 34.1, 31.6, 27.8, 24.3, 22.9, 21.6, 21.4, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{31}H_{40}N_2O_2S$ $[M + H]^+$ = 505.2883, found 505.2880.

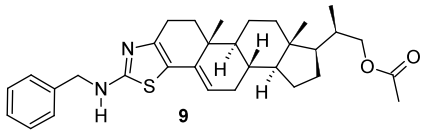
[(2*R*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(*N*-methylanilino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (8).



8

Off-white powder (466 mg, 90%); IR (KBr pellet, cm^{-1}) 3445, 2935, 1729, 1597, 1519, 1451; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.41−7.35 (m, 4H), 7.24−7.22 (m, 1H), 5.27 (s, 1H), 4.06 (dd, J = 10.4, 2.4 Hz, 1H), 3.76 (dd, J = 10.2, 7.9 Hz, 1H), 3.50 (s, 3H), 2.70−2.66 (m, 2H), 2.13−1.95 (m, 6H), 1.81−1.60 (m, 6H), 1.46−1.00 (m, 14H), 0.70 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.5, 167.1, 146.5, 146.0, 136.8, 129.8, 126.6, 125.4, 120.6, 117.8, 77.3, 69.6, 56.6, 52.7, 48.1, 42.6, 40.2, 39.6, 36.5, 35.9, 34.5, 31.7, 31.6, 27.8, 24.4, 21.4, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{42}N_2O_2S$ $[M + H]^+$ = 519.3039, found 519.3040.

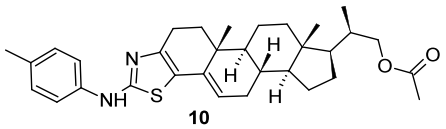
[(2*R*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(Benzylamino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (9).



9

Off-white powder (487 mg, 94%); IR (KBr pellet, cm^{-1}) 3370, 2929, 1736, 1556, 1453; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.34−7.27 (m, 5H), 5.33 (s, 1H), 4.44 (s, 2H), 4.06 (dd, J = 10.7, 3.1 Hz, 1H), 3.76 (dd, J = 10.5, 7.6 Hz, 1H), 2.59 (d, J = 6.0 Hz, 2H), 2.18−2.91 (m, 7H), 1.84−1.58 (m, 6H), 1.50−1.00 (m, 14H), 0.71 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 166.9, 145.7, 137.7, 136.8, 128.8, 127.8, 127.7, 120.0, 117.6, 77.3, 69.6, 56.6, 52.7, 49.8, 48.1, 42.6, 39.6, 36.6, 35.9, 34.4, 31.7, 27.8, 24.4, 24.2, 21.4, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{42}N_2O_2S$ $[M + H]^+$ = 519.3039, found 519.3039.

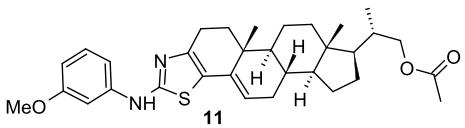
[(2*R*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(4-methylanilino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (10).



10

Brick red powder (456 mg, 88%); IR (KBr pellet, cm^{-1}) 3446, 2943, 1735, 1514; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.25 (d, J = 3.0 Hz, 1H), 6.88 (s, 2H), 5.45 (s, 1H), 4.06 (d, J = 10.5 Hz, 1H), 3.76 (t, J = 7.8 Hz, 1H), 2.62 (d, J = 6.6 Hz, 2H), 2.30 (s, 3H), 2.18−2.13 (m, 1H), 2.04−1.93 (m, 7H), 1.83−1.59 (m, 6H), 1.50−1.00 (m, 14 H), 0.71 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 163.4, 144.7, 137.9, 136.5, 133.2, 130.0, 119.9, 119.4, 118.3, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.6, 35.9, 35.7, 34.4, 31.6, 27.8, 24.4, 23.9, 21.4, 21.1, 20.9, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{42}N_2O_2S$ $[M + H]^+$ = 519.3039, found 519.3050.

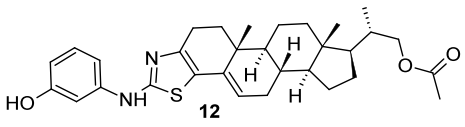
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(3-Methoxyanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (11).



11

Red gray powder (454 mg, 85%); IR (KBr pellet, cm^{-1}) 3429, 2942, 1730, 1705, 1603, 1523; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.25−7.20 (m, 1H), 6.93 (t, J = 2.2 Hz, 1H), 6.85 (dd, J = 8.0, 1.8 Hz, 1H), 6.59 (dd, J = 8.2, 2.2 Hz, 1H), 5.46−5.44 (m, 1H), 4.07 (dd, J = 10.7, 3.3 Hz, 1H), 3.80 (s, 3H), 3.78−3.75 (m, 1H), 2.69−2.65 (m, 2H), 2.21−2.15 (m, 1H), 2.05−1.96 (m, 5H), 1.82−1.59 (m, 6H), 1.48−1.01 (m, 14H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 161.9, 160.6, 145.0, 141.5, 136.5, 130.3, 120.8, 118.7, 110.9, 108.5, 104.4, 69.7, 56.5, 55.4, 52.7, 48.0, 42.6, 39.6, 36.6, 35.9, 34.4, 31.7, 31.6, 27.8, 24.4, 24.1, 21.4, 21.2, 18.8, 17.3, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{42}N_2O_3S$ $[M + H]^+$ = 535.2989, found 535.2982.

[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(3-Hydroxyanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (12).



12

Light black powder (437 mg, 84%); IR (KBr pellet, cm^{-1}) 3273, 2941, 1734, 1716, 1605; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.13 (t, J = 8.0 Hz, 1H), 6.77−6.75 (m, 2H), 6.58−6.56 (m, 1H), 5.40 (s, 1H), 4.06 (dd, J = 10.5, 3.0 Hz, 1H), 3.75 (dd, J = 10.3, 7.8 Hz, 1H), 2.61−2.60 (m, 2H), 2.16−1.92 (m, 7H), 1.80−1.58 (m, 6H), 1.48−0.96 (m, 15H), 0.69 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 177.8, 171.8, 163.6, 157.7, 140.4, 135.5, 130.5, 119.5, 118.7, 111.4, 109.8, 105.8, 69.7, 56.4, 52.7, 47.9, 42.6, 39.5, 36.6, 35.9, 33.8, 31.6, 27.7, 24.4, 22.3, 21.9, 21.4, 21.1, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{31}H_{40}N_2O_3S$ $[M + H]^+$ = 521.2832, found 521.2845.

https://doi.org/10.1021/acsomega.3c09721
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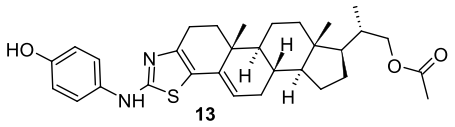
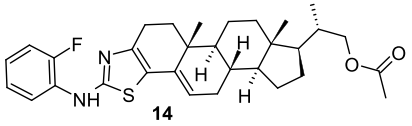
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(4-Hydroxyanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (13).



13

Light black powder (87%, 452 mg); IR (KBr pellet, cm^{-1}) 3331, 2942, 1734, 1716, 1540, 1513; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.03−7.01 (m, 2H), 6.78−6.76 (m, 2H), 5.34 (s, 1H), 4.06−4.04 (m, 1H), 3.77−3.73 (m, 1H), 2.59 (s, 2H), 2.15−1.91 (m, 3H), 1.80−1.59 (m, 6H), 1.44−0.91 (m, 15H), 0.69 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.8, 166.8, 154.8, 139.5, 135.5, 131.5, 123.0, 119.4, 117.5, 116.7, 69.7, 56.4, 52.7, 47.9, 42.6, 39.5, 36.6, 35.9, 33.8, 31.6, 27.7, 24.3, 22.4, 22.1, 21.4, 21.1, 18.7, 17.2, 12.0. HRMS (ESI-FTMS) Mass (*m*/*z*): calcd for $C_{31}H_{40}N_2O_3S$ [M + H]$^+$ = 521.2832, found 521.2841.

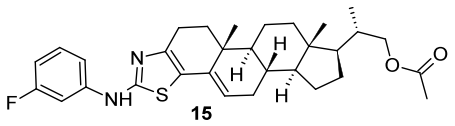
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(2-Fluoroanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (14).



14

Light yellow powder (444 mg, 85%); IR (KBr pellet, cm^{-1}) 3328, 2941, 1733, 1539; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.70 (t, *J* = 8.1 Hz, 1H), 7.14−7.08 (m, 2H), 7.03−7.00 (m, 1H), 5.44 (s, 1H), 4.06 (dd, *J* = 10.5, 3.2 Hz, 1H), 3.76 (dd, *J* = 10.5, 7.6 Hz, 1H), 2.67−2.63 (m, 2H), 2.20−2.14 (m, 1H), 2.10−1.96 (m, 5H), 1.84−1.60 (m, 6H), 1.51−0.96 (m, 14H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 176.7, 171.6, 162.8, 153.2 ($^1J_{C-F}$ = 245.6 Hz), 143.7, 136.1, 128.6 ($^3J_{C-F}$ = 11.1 Hz), 124.7 ($^4J_{C-F}$ = 3.8 Hz), 123.9 ($^3J_{C-F}$ = 7.2 Hz), 120.3, 119.3 ($^2J_{C-F}$ = 38.1 Hz), 115.8 ($^2J_{C-F}$ = 19.3 Hz), 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.6, 35.9, 34.2, 31.7, 31.6, 27.8, 24.4, 23.3, 21.4, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (*m*/*z*): calcd for $C_{31}H_{39}FN_2O_2S$ [M + H]$^+$ = 523.2789, found 523.2797.

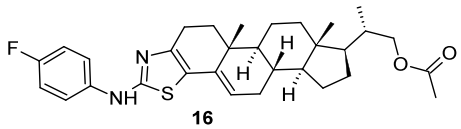
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(3-Fluoroanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (15).



15

Off-white powder (470 mg, 90%); IR (KBr pellet, cm^{-1}) 3349, 2937, 1733, 1716, 1616; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.26−7.22 (m, 1H), 7.09 (d, *J* = 10.5 Hz, 1H), 7.01 (d, *J* = 8.0 Hz, 1H), 6.71 (t, *J* = 8.1 Hz, 1H), 5.47 (s, 1H), 4.07−4.05 (m, 1H), 3.78−3.74 (m, 1H), 2.65−2.63 (m, 2H), 2.20−1.95 (m, 6H), 1.82−1.59 (m, 6H), 1.50−1.00 (m, 14H), 0.71 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 177.4, 171.6, 163.5 ($^1J_{C-F}$ = 245.1 Hz), 143.4, 141.7 ($^3J_{C-F}$ = 10.6 Hz), 136.0, 130.7 ($^3J_{C-F}$ = 9.6 Hz), 120.3, 119.2, 113.5 ($^4J_{C-F}$ = 2.90 Hz), 109.5 ($^2J_{C-F}$ = 21.7 Hz), 105.0 ($^2J_{C-F}$ = 25.5 Hz), 69.6, 56.5, 52.7, 48.0, 42.6, 39.5, 36.6, 35.9, 34.1, 31.6, 27.7, 24.3, 23.3, 21.8, 21.4, 21.1, 18.7, 17.2, 12.0. HRMS (ESI-FTMS) Mass (*m*/*z*): calcd for $C_{31}H_{39}FN_2O_2S$ [M + H]$^+$ = 523.2789, found 523.2790.

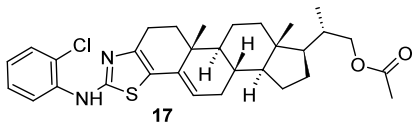
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(4-Fluoroanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (16).



16

Dark red powder (480 mg, 92%); IR (KBr pellet, cm^{-1}) 3346, 2942, 1734, 1716, 1506; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.25−7.22 (m, 2H), 7.05−7.01 (m, 2H), 5.42−5.40 (m, 1H), 4.06 (dd, *J* = 10.7, 3.4 Hz, 1H), 3.76 (dd, *J* = 10.7, 7.6 Hz, 1H), 2.63−2.61 (m, 2H), 2.15−1.95 (m, 6H), 1.73−1.58 (m, 6H), 1.37−1.00 (m, 14H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 177.4, 171.6, 164.4, 159.1 ($^1J_{C-F}$ = 243.2 Hz), 142.8, 136.3, 136.0, 120.8 ($^3J_{C-F}$ = 8.2 Hz), 118.9, 116.3 ($^2J_{C-F}$ = 23.1 Hz), 69.6, 56.5, 52.7, 47.9, 42.6, 39.5, 36.6, 35.9, 34.1, 31.7, 27.8, 24.4, 23.0, 21.8, 21.4, 21.2, 18.8, 17.3, 12.1. HRMS (ESI-FTMS) Mass (*m*/*z*): calcd for $C_{31}H_{39}FN_2O_2S$ [M + H]$^+$ = 523.2789, found 523.2796.

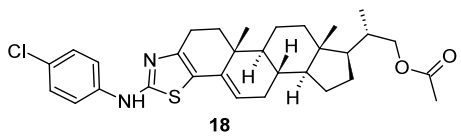
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(2-Chloroanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (17).



17

Gray powder (431 mg, 80%); IR (KBr pellet, cm^{-1}) 3404, 2941, 1737, 1596, 1533, 749; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 8.04−8.02 (m, 1H), 7.36−7.34 (m, 1H), 7.3−7.2 (m, 1H), 6.93 (t, *J* = 7.6 Hz, 1H), 5.48 (s, 1H), 4.08−4.05 (m, 1H), 3.76 (t, *J* = 8.6 Hz, 1H), 2.72−2.69 (m, 2H), 2.20−2.16 (m, 1H), 2.04−1.93 (m, 5H), 1.81−1.61 (m, 6H), 1.51−0.96 (m, 14H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 159.9, 145.7, 136.9, 136.4, 129.5, 128.0, 122.7, 122.2, 121.8, 119.3, 117.9, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.7, 35.9, 34.4, 31.8, 31.6, 27.8, 24.4, 24.2, 21.4, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (*m*/*z*): calcd for $C_{31}H_{39}ClN_2O_2S$ [M + H]$^+$ = 539.2493, found 539.2504.

[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(4-Chloroanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (18).

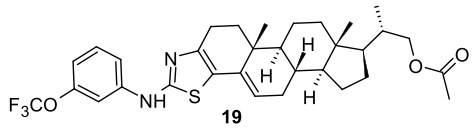


18

Gray powder (474 mg, 88%); IR (KBr pellet, cm^{-1}) 3420, 2939, 1738, 1703, 1592, 1529, 639; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.26−7.25 (m, 4H), 5.43 (s, 1H), 4.07−4.04 (m, 1H), 3.76 (dd, *J* = 10.2, 7.9 Hz, 1H), 2.63 (t, *J* = 6.8 Hz, 2H), 2.20−2.09 (m, 1H), 2.04−1.95 (m, 5H), 1.84−1.59 (m, 6H), 1.47−0.96 (m, 14H), 0.73−0.69 (m, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 162.2, 144.1, 138.9, 136.2, 129.5, 128.0, 120.4, 119.7, 119.1, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.6, 35.9, 34.2, 31.7, 31.6, 27.8, 24.4, 23.7, 21.4, 21.2, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (*m*/*z*): calcd for $C_{31}H_{39}ClN_2O_2S$ [M + H]$^+$ = 539.2493, found 539.2492.

https://doi.org/10.1021/acsomega.3c09721
ACS Omega 2024, 9, 23283−23293

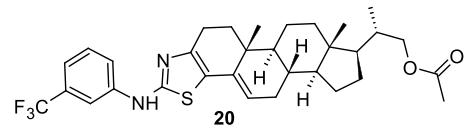
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(3-methylanilino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (19).



19

Off-white powder (493 mg, 84%); IR (KBr pellet, cm^{-1}) 3533, 2941, 1718, 1611, 1524; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.41 (s, 1H), 7.81 (s, 1H), 7.46−7.44 (m, 1H), 7.34 (t, J = 8.0 Hz, 1H), 6.82 (d, J = 7.3 Hz, 1H), 5.33 (s, 1H), 3.93−3.90 (m, 1H), 3.60−3.56 (m, 1H), 2.58 (s, 2H), 2.10−1.86 (m, 3H), 1.66−1.49 (m, 6H), 1.34−1.22 (m, 3H), 1.11−0.87 (m, 11H), 0.61 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ 171.0, 159.8, 149.4, 146.0, 143.1, 136.5, 130.9, 120.9, 120.6 ($^1J_{C-F}$ = 256.2 Hz), 119.1, 116.0, 113.0, 109.3, 69.1, 56.3, 52.6, 47.9, 42.5, 39.5, 36.4, 35.8, 34.3, 31.6, 31.5, 27.6, 24.5, 24.4, 21.4, 21.2, 19.0, 17.4, 12.2. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{39}F_3N_2O_3S$ [M + H]$^+$ = 589.2706, found 589.2703.

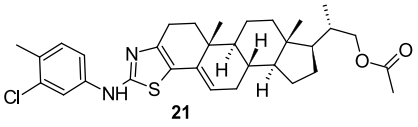
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(3-methylanilino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (20).



20

Dark red powder (463 mg, 81%); IR (KBr pellet, cm^{-1}) 3325, 2943, 1737, 1716, 1566, 1524; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 7.55 (s, 1H), 7.51−7.48 (m, 1H), 7.41 (t, J = 7.9 Hz, 1H), 7.26−7.25 (m, 1H), 5.47 (s, 1H), 4.07−4.05 (m, 1H), 3.79−3.74 (m, 1H), 2.66−2.65 (m, 2H), 2.20−1.95 (m, 6H), 1.82−1.59 (m, 6H), 1.48−1.00 (m, 14H), 0.71−0.66 (m, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 177.3, 171.7, 161.9, 143.6, 140.7, 136.0, 131.8 ($^2J_{C-F}$ = 32.3 Hz), 130.1, 123.9 ($^1J_{C-F}$ = 272.6 Hz), 121.0, 119.5, 119.3 ($^3J_{C-F}$ = 4.3 Hz), 114.5 ($^3J_{C-F}$ = 4.3 Hz), 69.7, 56.5, 52.7, 48.0, 42.6, 39.5, 36.7, 35.9, 34.1, 31.7, 27.7, 24.4, 23.4, 21.7, 21.4, 21.1, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{39}F_3N_2O_2S$ [M + H]$^+$ = 573.2757, found 573.2765.

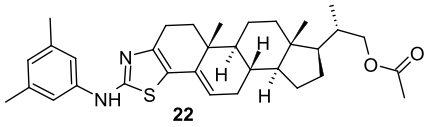
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(3-Chloro-4-methyl-anilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (21).



21

Off white powder (453 mg, 82%); IR (KBr pellet, cm^{-1}) 3244, 2942, 1736, 1607, 1540, 1524, 638; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 7.29 (s, 1H), 7.17−7.15 (m, 1H), 7.08−7.05 (m, 1H), 5.46 (s, 1H), 4.07−4.05 (m, 1H), 3.79−3.74 (m, 1H), 2.63−2.62 (m, 2H), 2.31 (s, 3H), 2.20−2.16 (m, 1H), 2.09−1.95 (m, 2H), 1.83−1.59 (m, 6H), 1.48−1.00 (m, 14H), 0.70 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 177.0, 171.6, 163.3, 138.9, 135.9, 135.0, 131.6, 130.9, 119.4, 119.2, 119.0, 117.0, 69.6, 56.5, 52.7, 47.9, 42.6, 39.5, 36.7, 35.9, 34.1, 31.7, 27.8, 24.4, 23.0, 21.6, 21.4, 21.2, 19.5, 18.8, 17.3, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{32}H_{41}ClN_2O_2S$ [M + H]$^+$ = 553.2650, found 553.2659.

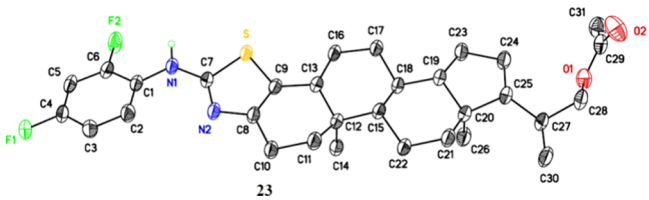
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(3,5-Dimethylanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (22).



22

Light red powder (436 mg, 82%); IR (KBr pellet, cm^{-1}) 3301, 2944, 1723, 1603, 1571, 1514; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 6.88 (s, 2H), 6.71 (s, 1H), 5.45 (s, 1H), 4.08−4.05 (m, 1H), 3.76 (t, J = 7.8 Hz, 1H), 2.63−2.61 (m, 2H), 2.30−2.14 (m, 5H), 2.09−1.95 (m, 8H), 1.81−1.62 (m, 6H), 1.51−1.00 (m, 14H), 0.67−0.74 (3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 176.9, 171.6, 164.0, 141.9, 139.7, 139.4, 136.0, 125.5, 118.9, 116.3, 77.3, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.7, 35.9, 34.0, 31.7, 27.8, 24.4, 22.7, 21.6, 21.5, 21.4, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{33}H_{44}N_2O_2S$ [M + H]$^+$ = 533.3196, found 533.3194.

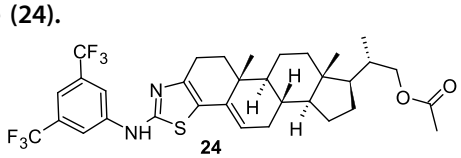
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(2,4-Difluoroanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (23).



23

Brick red powder (432 mg, 80%); IR (KBr pellet, cm^{-1}) 3345, 2942, 1724, 1563, 1545, 1524; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 8.03−7.98 (m, 1H), 6.89−6.85 (m, 2H), 5.44 (d, J = 4.2 Hz, 1H), 4.07 (dd, J = 10.7, 3.2 Hz, 1H), 3.77 (dd, J = 10.5, 7.6 Hz, 1H), 2.70−2.66 (m, 2H), 2.20−2.15 (m, 1H), 2.05−1.97 (m, 5H), 1.85−1.60 (m, 6H), 1.52−1.01 (m, 15H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 162.0, 158.2 ($^1J_{C-F}$ = 245.1 Hz), 153.0 ($^1J_{C-F}$ = 247.6 Hz), 144.9, 136.2, 127.7, 125.2 ($^3J_{C-F}$ = 11.1 Hz), 121.1 ($^3J_{C-F}$ = 7.7 Hz), 119.2, 111.4 ($^2J_{C-F}$ = 21.7 Hz), 104.4 ($^2J_{C-F}$ = 23.1 Hz), 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.7, 35.9, 34.3, 31.7, 31.6, 27.8, 24.4, 23.9, 21.4, 21.1, 18.8, 17.2, 12.1. ^{19}F-decoupled ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 160.8, 157.7, 152.3, 145.8, 136.4, 125.3, 121.8, 120.3, 119.2, 111.3, 104.0, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.7, 35.9, 34.4, 31.7, 31.6, 27.8, 24.4, 24.3, 21.4, 21.1, 18.8, 17.2, 12.1 HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{31}H_{38}F_2N_2O_2S$ [M + H]$^+$ = 541.2695 found 541.2704.

[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-(3,5-Dimethylanilino)-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (24).



24

Light red powder (518 mg, 81%); IR (KBr pellet, cm^{-1}) 3322, 2943, 1708, 1523, 1473; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 7.88 (s, 2H), 7.45 (s, 1H), 5.52 (s, 1H), 4.07 (dd, J = 10.8, 3.2 Hz, 1H), 3.77 (dd, J = 10.7, 7.7 Hz, 1H), 2.73−2.69 (m, 2H), 2.23−2.17 (m, 1H), 2.05−1.98 (m, 5H), 1.83−1.61 (m, 6H), 1.53−1.00 (m, 14H), 0.73 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.7, 159.9, 145.0, 141.8, 136.0, 133.0 ($^2J_{C-F}$ =

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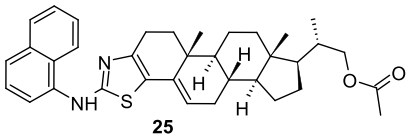
33.2 Hz), 126.0 ($^1J_{C-F}$ = 272.6 Hz), 122.3, 120.2, 117.0, 115.3 ($^3J_{C-F}$ = 7.2 Hz), 69.7, 56.5, 52.7, 48.0, 42.6, 39.5, 36.7, 35.9, 34.2, 31.7, 31.6, 27.8, 24.4, 24.0, 21.4, 21.2, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{33}H_{38}F_6N_2O_2S$ $[M + H]^+$ = 641.2631, found 641.2628.

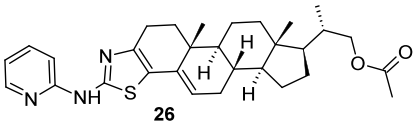
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(1-naphthylamino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (25).



25

Light red powder (471 mg, 85%); IR (KBr pellet, cm^{-1}) 3389, 2941, 1735, 1547; ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.06 (s, 1H), 7.86−7.85 (m, 1H), 7.72−7.68 (m, 2H), 7.48−7.44 (m, 3H), 5.31 (s, 1H), 4.06−4.04 (m, 1H), 3.77−3.73 (m, 1H), 2.54−2.52 (m, 2H), 2.13−1.98 (m, 6H), 1.89−1.59 (m, 6H), 1.44−0.98 (m, 14H), 0.70 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 165.4, 144.9, 136.5, 134.6, 128.6, 127.9, 126.6, 125.9, 125.7, 123.8, 121.7, 120.4, 119.1, 118.3, 114.6, 77.3, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.6, 35.9, 34.3, 31.7, 27.8, 24.3, 23.9, 21.4, 21.1, 18.8, 17.2, 12.1; HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{35}H_{42}N_2O_2S$ $[M + H]^+$ = 555.3039, found 555.3047.

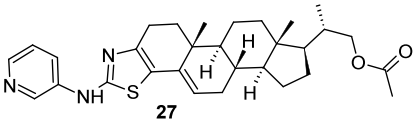
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(2-pyridylamino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (26).



26

Black solid (464 mg, 92%); IR (KBr pellet, cm^{-1}) 3259, 2940, 1738, 1605, 1540; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 8.31 (d, J = 4.8 Hz, 1H), 7.57 (t, J = 7.8 Hz, 1H), 6.92−6.84 (m, 2H), 5.65 (s, 1H), 4.06 (dd, J = 10.5, 3.2 Hz, 1H), 3.76 (dd, J = 10.5, 7.8 Hz, 1H), 2.66−2.65 (m, 2H), 2.22−2.16 (m, 1H), 2.08−1.96 (m, 5H), 1.83−1.59 (m, 6H), 1.51−0.95 (m, 14H), 0.71 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 159.3, 151.1, 146.8, 140.4, 137.8, 136.2, 122.3, 119.0, 116.8, 111.4, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.5, 35.9, 34.2, 31.7, 31.6, 27.8, 24.4, 22.7, 21.5, 21.2, 18.7, 17.3, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{30}H_{39}N_3O_2S$ $[M + H]^+$ = 506.2836, found 506.2828.

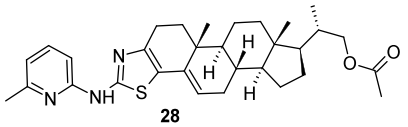
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-(3-pyridylamino)-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (27).



27

Gray powder (444 mg, 88%); IR (KBr pellet, cm^{-1}) 2940, 1737, 1524, 1037; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 8.57 (s, 1H), 8.26−8.25 (m, 1H), 7.89−7.86 (m, 1H), 7.28−7.25 (m, 2H), 5.45 (s, 1H), 4.06 (dd, J = 10.7, 3.1 Hz, 1H), 3.76 (dd, J = 10.5, 7.6 Hz, 1H), 2.68−2.65 (m, 2H), 2.20−2.16 (m, 1H), 2.09−1.96 (m, 5H), 1.84−1.60 (m, 6H), 1.52−0.97 (m, 14H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 161.4, 144.5, 143.5, 140.0, 137.2, 136.1, 124.8, 124.0, 121.1, 119.5, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.7, 35.9, 34.2, 31.7, 31.6, 27.8, 24.4, 23.7, 21.4, 21.2, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{30}H_{39}N_3O_2S$ $[M + H]^+$ = 506.2835, found 506.2839.

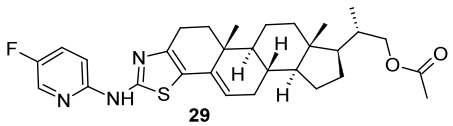
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-[(6-methyl-2-pyridyl)amino]-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (28).



28

Gray powder (472 mg, 91%); IR (KBr pellet, cm^{-1}) 3418, 3261, 2941, 1736, 1611, 1533, 1456; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 7.46 (t, J = 7.7 Hz, 1H), 6.72−6.70 (m, 2H), 5.66−5.65 (m, 1H), 4.06 (dd, J = 10.5, 3.2 Hz, 1H), 3.76 (dd, J = 10.5, 7.6 Hz, 1H), 2.67−2.66 (m, 2H), 2.51 (s, 3H), 2.24−2.16 (m, 1H), 2.11−1.96 (m, 5H), 1.84−1.60 (m, 6H), 1.52−0.95 (m, 14H), 0.70 (m, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 159.2, 156.3, 150.3, 140.5, 138.1, 136.4, 122.3, 118.8, 116.0, 107.9, 69.6, 56.5, 52.7, 48.0, 42.5, 39.6, 36.5, 35.9, 34.2, 31.7, 31.6, 27.8, 24.4, 23.8, 22.3, 21.5, 21.2, 18.7, 17.3, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{31}H_{41}N_3O_2S$ $[M + H]^+$ = 520.2992, found 520.2998.

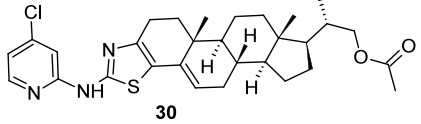
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-[(5-Fluoro-2-pyridyl)amino]-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (29).



29

Dark red solid (400 mg, 76%); IR (KBr pellet, cm^{-1}) 3426, 3263, 2940, 1737, 1620, 1548, 1493; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 8.16 (s, 1H), 7.36−7.32 (m, 1H), 6.93−6.90 (m, 1H), 5.63 (s, 1H), 4.07−4.04 (m, 1H), 3.75 (t, J = 8.0 Hz, 1H), 2.64−2.63 (m, 2H), 2.21−1.95 (m, 6H), 1.80−1.58 (m, 6H), 1.49−1.29 (m, 3H), 1.24−0.98 (m, 11H), 0.70 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 159.5, 155.2 ($^1J_{C-F}$ = 247.6 Hz), 147.6, 140.0, 136.0, 133.7 ($^2J_{C-F}$ = 25.5 Hz), 125.9 ($^2J_{C-F}$ = 20.7 Hz), 121.8, 119.2, 112.2 ($^3J_{C-F}$ = 3.8 Hz), 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.5, 35.9, 34.1, 31.7, 31.6, 27.8, 24.4, 22.6, 21.4, 21.1, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{30}H_{38}FN_3O_2S$ $[M + H]^+$ = 524.2741, found 524.2732.

[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-[(4-Chloro-2-pyridyl)amino]-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (30).



30

Light yellow powder (459 mg, 85%); IR (KBr pellet, cm^{-1}) 3420, 3265, 2939, 1738, 1703, 1592, 1478; ^{1}H NMR (400 MHz, $CDCl_3$-d) δ 8.20−8.18 (m, 1H), 6.93−6.84 (m, 2H), 5.65 (s, 1H), 4.06−4.05 (m, 1H), 3.79−3.74 (m, 1H), 2.65 (s, 2H), 2.20−1.99 (m, 6H), 1.80−1.62 (m, 6H), 1.50−1.33 (m, 2H), 1.24−0.99 (m, 12H), 0.70 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-d) δ 171.6, 158.9, 152.2, 147.8, 145.0, 141.0, 136.1, 123.0, 119.3, 117.1, 111.0, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6,

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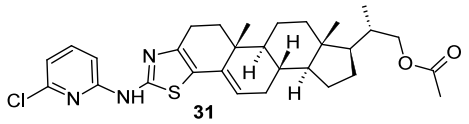
36.5, 35.9, 34.2, 31.7, 27.8, 24.4, 22.8, 22.0, 21.4, 21.1, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{30}H_{38}ClN_3O_2S$ $[M + H]^+$ = 540.2446, found 540.2436.

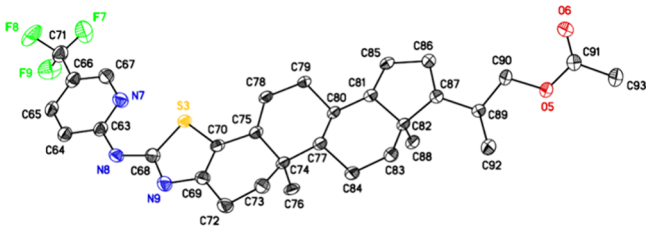
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-[(6-Chloro-2-pyridyl)amino]-2,18-dimethyl-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (31).



31

Off-white powder (506 mg, 93%); IR (KBr pellet, cm^{-1}) 3415, 3229, 2939, 1739, 1599, 1531, 1457; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 7.51−7.49 (m, 1H), 6.85−6.80 (m, 2H), 5.69 (s, 1H), 4.05 (s, 1H), 3.76−3.75 (m, 1H), 2.65 (s, 2H), 2.19−2.00 (m, 6H), 1.79−1.63 (m, 6H), 1.47−0.99 (m, 14H), 0.71 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 158.6, 151.3, 148.8, 141.5, 139.9, 136.1, 123.5, 119.5, 116.0, 109.1, 69.6, 56.5, 52.7, 48.0, 42.6, 39.6, 36.5, 35.9, 35.8, 34.2, 31.7, 27.8, 24.4, 23.2, 21.4, 21.1, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{30}H_{38}ClN_3O_2S$ $[M + H]^+$ = 540.2446, found 540.2443.

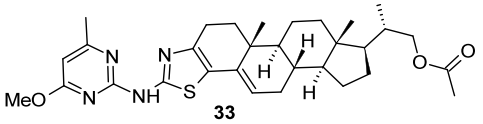
[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-2,18-Dimethyl-7-[(5-methyl-2-pyridyl)amino]-8-thia-6-azapentacyclo-[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (32).



32

White powder (510 mg, 89%); IR (KBr pellet, cm^{-1}) 3452, 2941, 1736, 1611, 1553, 1456, 1036; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 8.58 (s, 1H), 7.75−7.73 (m, 1H), 6.95−6.94 (m, 1H), 5.68 (s, 1H), 4.08−4.05 (m, 1H), 3.76 (t, J = 8.6 Hz, 1H), 2.66−2.64 (m, 2H), 2.23−1.97 (m, 6H), 1.79−1.60 (m, 6H), 1.51−1.30 (m, 3H), 1.25−0.99 (m, 11H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.6, 158.6, 153.4, 144.8 ($^3J_{C-F}$ = 4.3 Hz), 141.0, 136.0, 134. 7, 124.1 ($^1J_{C-F}$ = 272.2 Hz), 123.4, 119.6, 119.2 ($^2J_{C-F}$ = 33.2 Hz), 110.9, 56.5, 52.7, 48.0, 42.6, 39.6, 36.5, 35.9, 34.1, 31.7, 31.6, 27.8, 24.4, 22.7, 21.8, 21.5, 21.1, 18.7, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{31}H_{38}F_3N_3O_2S$ $[M + H]^+$ = 574.2709, found 574.2694.

[(2*S*)-2-[(1*S*,2*R*,13*S*,14*S*,17*R*,18*S*)-7-[(4-Methoxy-6-methyl-pyrimidin-2-yl)amino]-2,18-dimethyl-8-thia-6-azapentacyclo[11.7.0.0^{2,10}.0^{5,9}.0^{14,18}]icosa-5(9),6,10-trien-17-yl]propyl] Acetate (33).



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Light pink powder (440 mg, 80%); IR (KBr pellet, cm^{-1}) 3423, 2942, 1738, 1720, 1601, 1550; ^{1}H NMR (400 MHz, $CDCl_3$-*d*) δ 6.12 (s, 1H), 5.61 (s, 1H), 4.08−4.03 (m, 4H), 3.76 (dd, J = 10.5, 7.6 Hz, 1H), 2.92 (dd, J = 17.1, 4.9 Hz, 1H), 2.78−2.69 (m, 1H), 2.35 (s, 3H), 2.23−2.16 (m, 1H), 2.04−1.96 (m, 2H), 1.84−1.61 (m, 6H), 1.51−1.30 (m, 3H), 1.26−1.00 (m, 11H), 0.72 (s, 3H); ^{13}C NMR (101 MHz, $CDCl_3$-*d*) δ 171.5, 170.8, 157.6, 156.7, 144.4, 137.0, 123.9, 118.2, 99.1, 69.6, 56.5, 54.8 52.8, 48.1, 42.6, 39.6, 36.5, 35.9, 34.6, 31.8, 31.7, 27.8, 24.4, 23.8, 23.7, 21.5, 21.1, 18.8, 17.2, 12.1. HRMS (ESI-FTMS) Mass (m/z): calcd for $C_{31}H_{42}N_4O_3S$ $[M + H]^+$ = 551.3050, found 551.3059.

CONCLUSIONS

We have efficiently synthesized a series of novel compounds based on a fused thiazole with bisnoralcohol scaffold. These novel compounds did not show any *in vitro* cytotoxicity against human cancer cell lines except three compounds. These three compounds could be starting scaffolds to synthesize new compounds for lead optimization. One of the compounds showed moderate activity against staphylococci bacteria, which opens the new possibility of design and synthesis of novel compounds as antibacterial agents based on this scaffold. The hit compound did not cause hemolysis at 64 μg/mL concentration, which indicates that this compound would be therapeutically safe as an antibacterial agent.

ASSOCIATED CONTENT

Data Availability Statement

The data underlying this study are available in the published article and its online Supporting Information

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsomega.3c09721.

The Supporting Information is available free of charge at Copies of ^{1}H, ^{13}C, ^{19}F and Mass spectra, X-rya crystal and NCI data (DOCX)

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Author Contributions

Conceptualization, M.A.A.; methodology, S.R, H.R.KC, and S.A; resources, M.A.A; writing—original draft preparation, M.A.A.; writing—review and editing, S.R, H.R.KC, and S.A.; funding acquisition, M.A.A. All authors have read and agreed to the published version of the manuscript.

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Notes

The authors declare no competing financial interest.

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