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Synthesis, antimicrobial activity and molecular docking of di- and triorganotin(IV) complexes with thiosemicarbazide derivatives

Claudia Huedo^a, Franca Zani^b, M. Antonia Mendiola^a, Sayantan Pradhan^c, Chittaranjan Sinha^c, Elena López-Torres^{a*}

^a Departamento de Química Inorgánica, Universidad Autónoma de Madrid, C/ Francisco Tomás y Valiente 7, 28049, Madrid, Spain.

^b Dipartimento di Farmacia, Università degli Studi di Parma, Parco Area delle Scienze 27/A, 43124 Parma, Italy.

^c Department of Chemistry, Jadavpur University, Kolkata, 700 032, India

(E. López-Torres) E-mail: elena.lopez@uam.es; Phone: +34 91 497 2376.

ELT: 0000-0002-6826-0146

MAM: 0000-0003-2753-8209

Abstract

Six organotin(IV) complexes with two ligands derived from 2,3-butanedione and thiosemicarbazide have been synthesized and fully characterized by several spectroscopic techniques, including ¹¹⁹Sn NMR and single crystal X-ray diffraction. Reactions of the ligand diacetyl-2-(thiosemicarbazone)-3-(3-hydroxy-2-naphthohydrazone), L¹H₂, with SnR₂Cl₂ (R=Me, Bu, Ph) lead to the obtaining of complexes **1-3** with general formula [SnR₂L¹] (R=Me **1**, R=Bu **2**, R=Ph **3**), in which the ligand is doubly deprotonated and behaves as a N₂SO donor, whereas from the reactions of diacetyl-2-thiosemicarbazone, HATs, with the same organotin precursors any complex could be isolated. By contrast, reaction of HATs with SnR₃Cl induces the

ligand cyclization to form a 1,2,4-triazine-3-thione that binds to the metal as a monoanionic donor in a mono or bidentate manner to form compounds **4-6** with formula $[\text{SnR}_3\text{L}^2]$ (R=Me **4**, R=Bu **5**, R=Ph **6**). The antimicrobial activity of the ligands and the six complexes was tested towards bacteria and fungi, including clinical isolated strains. The results show that the ligands are devoid of activity, except HATs that displays activity against *Bacillus subtilis*. Conversely, the complexes exhibit good antimicrobial properties against Gram positive and negative bacteria, yeasts and moulds. The best results are obtained for complexes $[\text{SnBu}_3\text{L}^2]$ **5** and $[\text{SnPh}_3\text{L}^2]$ **6**, indicating that their more lipophilic nature could play an important role in the ease of microbial cell penetration. In some cases, these complexes display similar or higher activity than that of ampicillin and miconazole, used as antibacterial and antifungal positive controls, respectively. Docking study with DHPS protein (*S. aureus*) has shown that out of six drugs, the compound **6** has the best binding affinity (-8.5 Kcal/mol).

Keywords: thiosemicarbazones, hydrazones, organotin(IV) complexes, , antimicrobial activity, molecular docking

Introduction

Treating infectious diseases caused by bacteria and fungi is an important and challenging public health problem.^[1] In the majority of the cases these infections affect patients with decreased immunological system, neoplastic disorders or undergoing organ transplantation.^[2] In addition, common pathogens and new pathogenic species with intrinsic primary resistance are rapidly developing secondary resistance to the current antimicrobial agents.^[3] Therefore, multidrug resistance to therapeutic antibiotics constitutes a serious public health threat, since the prevalence of extremely resistant bacteria such as methicillin-resistant *Staphylococcus aureus*, fluoroquinolone-resistant *Pseudomonas aeruginosa*, fluoroquinolone-resistant *Enterococcus faecalis* or

vancomycin-resistant *Enterococci* has enormously increased in some hospitals, resulting in higher rate of mortality and morbidity.^[4-6] In order to prevent this serious medical problem, the search of new antimicrobial agents is an interesting topic to pursue, in particular the synthesis and characterization of metal complexes with bioactive ligands. The biological activities of the metal complexes usually differ from those of either the ligand or the metal ion, and the results obtained till date show that structural factors, which will modulate the antimicrobial activity, strongly depends on the metal ion.

Organotin(IV) derivatives have been widely investigated because of their structural diversity and biological activity, for example as bactericides, fungicides, acaricides, antifouling and anti-tumor agents.^[7-17] It is well established that the biocide activity of organotin(IV) complexes depends on a delicate balance between structure, coordination number, extent of alkylation and nature of the organic groups bonded to the tin atom.^[18] Despite the intense research on the toxicology of organotin(IV) compounds, their mode of action is not completely understood, although some general features seem to be widely accepted. The activity of organotin(IV) compounds is dependent on both the covalently bonded organic groups and the nature of the other ligands.^[19] Trialkyl and triaryl tin(IV) compounds are generally more toxic than diorganotin(IV) compounds, and monosubstituted ones are the less toxic.¹¹ However, the order of toxicity depends also on the microorganism, and differs from strain to strain.^[20] On the other hand, inhibitory activity generally increases in the order Me < Bu < Ph, which is attributed to lipophilicity that allows the drug to cross the microorganism membrane.^[19,21]

Hydrazones and thiosemicarbazones have received much attention over the last decades due to their fascinating structural versatility^[22-27] together with their outstanding therapeutic activities, such as antitumor, antiviral, antiprotozoal,

antibacterial or antifungal agents.^[28-35] As a continuation of our studies on the synthesis and antimicrobial activity of organotin(IV) complexes with thiosemicarbazone/hydrazone ligands,^[36-39] we have synthesized two ligands, one mixed thiosemicarbazone/hydrazone ligand, L¹H₂, and one mono(thiosemicarbazone), HATs, and their complexes with SnR₂Cl₂ and SnR₃Cl (R=Me, Bu, Ph), respectively. In a previous work we have found that introduction of a lipophilic pendant group in the terminal amine of the thiosemicarbazone do not increase the antimicrobial activity of the complexes,^[39] so we decided to introduce a NH₂ group, which is less lipophilic but has less steric hindrance to evaluate if the ligand size could have any influence in the antimicrobial properties. We also used tri- and diorganotin(IV) precursors to modify the overall lipophilicity of the derivatives. The compounds are fully characterized by several spectroscopic techniques including ¹¹⁹Sn NMR in solution and in the solid state and some of them by single crystal X-ray diffraction. The *in vitro* antimicrobial activity of the ligands and their corresponding organotin(IV) complexes was evaluated against several strains of bacteria and fungi. Furthermore, molecular docking was also carried out, since computational aided drug discovery is a useful rapid and economic procedure and has been recognized to be more effective than the conventional wet lab drug discovery scheme.^[40] It involves docking of the drug-like molecule into a protein target followed by applying a scoring function to estimate the likelihood that the compound will bind to the protein with high affinity.^[41] In this present work DHPS (dihydrofolatesynthetase) has been focused as drug target. Drugs inhibit bacterial synthesis of dihydrofolic acid by binding to DHPS and decreasing the synthesis of bacterial nucleotides and DNA, which are essential for bacterial survival.^[42]

Materials and methods

Microanalyses were carried out using a LECO CHNS-932 Elemental Analyzer. IR spectra in the 4000-400 cm^{-1} range were recorded as KBr pellets on a Jasco FT/IR-410 spectrophotometer. The ESI mass spectra in positive mode were recorded on a Q-STAR PULSAR I instrument using a hybrid analyzer QTOF (Quadrupole time-of-flight). ^1H , ^{13}C and ^{119}Sn NMR spectra were recorded on a spectrometer Bruker AVIII HD-300 MHz using DMSO- d_6 as solvent and TMS (^1H and ^{13}C) or SnMe_4 (^{119}Sn) as internal reference. ^{119}Sn CP/MAS NMR spectra were recorded at 298 K in a Bruker AV400WB spectrometer equipped with a 4 mm MAS (magic-angle spinning) NMR probe and obtained using a cross-polarization pulse sequence using spinning rates of 10-14 KHz, pulse delays of 30 s, contact times of 8 ms and two-pulse phase-modulated high power proton decoupling. Chemical shifts are reported relative to SnMe_4 , using tin(IV) oxide as a secondary reference. Molar conductivity was measured using a freshly prepared DMF solution (ca. 10^{-3} M) at 25 °C with a Crison EC-Meter BASIC 30+ instrument.

Synthesis of the compounds

All the chemicals were purchased from standard commercial sources and used as received. Scheme 1 summarizes the reactions described in this section and includes the atom labeling used for spectra assignment.

Synthesis of diacetyl-2-thiosemicarbazone, HATs. To a solution of 2.00 g (21.94 mmol) of thiosemicarbazide in 40 mL of deionized water with 10 drops of conc. hydrochloric acid was added 5.6 mL (65.80 mmol) of 2,3-butanedione and the mixture was stirred at room temperature for 1 h. The white solid obtained was filtered off, washed with cold water and methanol and vacuum-dried. Yield 3.39 g, 97%. Found: C, 37.65; H, 5.61; N, 26.32; S, 20.05. $\text{C}_5\text{H}_9\text{N}_3\text{OS}$ requires C, 37.72; H, 5.70; N, 26.41; S,

20.10. ^1H NMR (300 MHz, DMSO- d_6 , ppm): 10.58 (s, 1H, NH), 8.71 (s, 1H, NH_2), 8.10 (s, 1H, NH_2), 2.46 (s, 3H, CH_3CO), 1.97 (s, 3H, CH_3CN). IR (KBr): ν/cm^{-1} 3399s, 3326s and 3182s $\nu(\text{NH})$, 1685s $\nu(\text{CO})$, 1587s $\delta(\text{HNH})$, 852m thioamide IV. MS (ESI^+): m/z 160.1 $[\text{M}+\text{H}]^+$.

Synthesis of diacetyl-2-(thiosemicarbazone)-3-(3-hydroxy-2-naphthohydrazone),

L^1H_2 . To a suspension of 0.51 g (3.20 mmol) of HATs in 25 mL of absolute ethanol with 10 drops of conc. HCl, a suspension of 0.65 g (3.20 mmol) of 3-hydroxy-2-naphthohydrazide in 15 mL of the same solvent was added. The mixture was stirred for 5 h. at room temperature. The beige solid formed was filtered off, washed with methanol and vacuum-dried. Yield 0.90 g, 90%. Found: C, 55.85; H, 5.13; N, 20.31; S, 9.27. $\text{C}_{16}\text{H}_{17}\text{N}_5\text{O}_2\text{S}$ requires C, 55.96; H, 4.99; N, 20.41; S, 9.32. ^1H NMR (300 MHz, DMSO- d_6 , ppm): 11.75 (s, 1H, H_{2a}), 11.60 (s, 1H, H_5), 10.28 (s, 1H, H_2), 8.63 (s, 1H, H_6), 8.44 (s, 1H, H_{1a}), 7.97 (d, 1H, H_8 , $^3J=8.3$ Hz), 7.93 (s, 1H, H_{1b}), 7.75 (d, 1H, H_{11} , $^3J=8.3$ Hz), 7.48 (t, 1H, H_{10} , $^3J=7.6$ Hz), 7.35 (m, 2H, H_9+H_{13}), 2.25 (s, 3H, H_{15}), 2.23 (s, 3H, H_{16}). ^{13}C NMR (300 MHz, DMSO- d_6 , ppm): 179.5 (C_1), 161.7 (C_4), 153.0 (C_{14}), 152.6 (C_2), 148.6 (C_3), 136.3 (C_{12}) 133.1 (C_7), 129.4 (C_6), 128.9 (C_5) 127.7 (C_8), 126.2 (C_{10}), 124.4 (C_{11}), 121.1 (C_9), 111.2 (C_{13}), 12.0 (C_{15}), 11.7 (C_{16}). IR (KBr): ν/cm^{-1} 3422m $\nu(\text{OH})$, 3241m and 3145m $\nu(\text{NH})$, 1638m $\nu(\text{CO})$, 1621w $\nu(\text{CN})$, 1499m thioamide II, 848w thioamide IV. Crystals suitable for X-ray analysis were obtained by recrystallization in DMSO.

Synthesis of L^1H_2 derivatives

$[\text{SnMe}_2\text{L}^1]$, 1. To a suspension of 150 mg (0.44 mmol) of L^1H_2 and 37 mg (0.88 mmol) of lithium hydroxide monohydrated in 10 mL of absolute ethanol, a solution of 96 mg (0.44 mmol) of SnMe_2Cl_2 in the same solvent was added and the mixture was stirred

under reflux for 1 h. The orange solid formed was filtered off, washed with ethanol and vacuum-dried. Yield 197 mg, 92%. Found, C, 44.30; H, 4.25; N, 14.46; S, 6.66; $\text{SnC}_{18}\text{H}_{21}\text{N}_5\text{O}_2\text{S}$ requires C, 44.09; H, 4.32; N, 14.29; S, 6.53. ^1H NMR (300 MHz, DMSO-d_6 , ppm): 13.40 (s, 1H, H_{2a}), 8.52 (s, 1H, H_6), 7.92 (d, 1H, H_8 , $^3J=8.3$ Hz), 7.73 (d, 1H, H_{11} , $^3J=8.3$ Hz), 7.46 (t, 1H, H_9 , $^3J=8.2$ Hz), 7.30 (t, 1H, H_{10} , $^3J=7.4$ Hz), 7.25 (s, 1H, H_{13}), 7.11 (s, 2H, $\text{H}_{1a}+\text{H}_{1b}$), 2.42 (s, 3H, H_{15}), 2.41 (s, 3H, H_{16}), 0.66 (s, 6H, SnMe_2 , $^2J(^{119}\text{Sn}-^1\text{H})=60$ Hz). ^{13}C NMR (300 MHz, DMSO-d_6 , ppm): 178.3 (C_1), 170.3 (C_4), 156.7 (C_{14}), 150.2 (C_2), 146.7 (C_3), 136.5 (C_{12}), 130.5 (C_7), 129.2 (C_6), 128.1 (C_5), 127.1 (C_8), 126.1 (C_{10}), 123.3 (C_{11}), 121.3 (C_9), 110.5 (C_{13}), 19.8 (Me), 16.2 (C_{15}), 15.6 (C_{16}). IR (KBr): ν/cm^{-1} 3375w $\nu(\text{OH})$, 3265w and 3167w $\nu(\text{NH})$, 1638w $\nu(\text{CO})$, 1594w $\nu(\text{CN})$, 1520m thioamide II, 818w thioamide IV. MS (ESI^+): m/z 492.04 $[\text{M}+\text{H}]^+$. Λ_{M} ($\Omega^{-1}\text{cm}^2 \text{mol}^{-1}$, DMF): 2.3. Recrystallization in ethanol yielded crystals suitable for single crystal X-ray diffraction.

[SnBu₂L¹], 2. This complex was obtained following the same procedure described for the synthesis of **1** but adding 133 mg (0.44 mmol) of SnBu_2Cl_2 (orange, 180 mg, 72%). Found: C, 50.47; H, 5.90; N, 12.37; S, 5.51; $\text{SnC}_{24}\text{H}_{33}\text{N}_5\text{O}_2\text{S}$ requires C, 50.18; H, 5.79; N, 12.20; S, 5.57. ^1H NMR (300 MHz, DMSO-d_6 , ppm): 13.40 (s, 1H, H_{2a}), 8.52 (s, 1H, H_6), 7.92 (d, 1H, H_8 , $^3J=7.9$ Hz), 7.73 (d, 1H, H_{11} , $^3J=8.1$ Hz), 7.46 (t, 1H, H_9 , $^3J=7.4$ Hz), 7.30 (t, 1H, H_{10} , $^3J=7.4$ Hz), 7.25 (s, 1H, H_{13}), 7.12 (s, 2H, $\text{H}_{1a}+\text{H}_{1b}$), 2.42 (s, 6H, $\text{H}_{15}+\text{H}_{16}$), 1.06 (m, 12H, $\text{CH}_2\text{-Bu}$), 0.69 (t, 6H, $\text{CH}_3\text{-Bu}$, $^3J=7.1$ Hz). ^{13}C NMR (300 MHz, DMSO-d_6 , ppm): 178.9 (C_1), 170.3 (C_4), 156.7 (C_{14}), 150.9 (C_2), 147.0 (C_3), 136.5 (C_{12}), 130.4 (C_7), 129.2 (C_6), 128.1 (C_5), 127.1 (C_8), 126.1 (C_{10}), 123.4 (C_{11}), 121.3 (C_9), 110.5 (C_{13}), 36.1 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 27.5 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 26.1 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 16.2 (C_{15}), 15.5 (C_{16}), 14.6 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$). IR (KBr): ν/cm^{-1} 3466w $\nu(\text{OH})$, 3267w and 3123w $\nu(\text{NH})$, 1640w $\nu(\text{CO})$, 1594w $\nu(\text{CN})$, 1524m

thioamide II, 746w thioamide IV. MS (ESI⁺): m/z 576.16 [M+H]⁺. Λ_M ($\Omega^{-1}\text{cm}^2\text{mol}^{-1}$, DMF): 4.7.

[SnPh₂L¹], **3**. This complex was obtained following the same procedure described for the synthesis of **1** but adding 150 mg (0.44 mmol) of SnPh₂Cl₂ (orange, 223 mg, 83%). Found: C, 54.66; H, 4.34; N, 11.56; S, 5.34. SnC₂₈H₂₅N₅O₂S requires C, 54.73; H, 4.10; N, 11.41; S, 5.21. ¹H NMR (300 MHz, DMSO-d₆, ppm): 13.05 (s, 1H, H_{2a}), 8.58 (s, 1H, H₆), 7.92 (d, 1H, H₈, ³*J*=7.9 Hz), 7.75 (d, 1H, H₁₁, ³*J*=8.0 Hz), 7.60-7.55 (m, 3H, H_{1a}+H_{1b}+H₁₀), 7.48 (t, 1H, H₉, ³*J*=8.2 Hz), 7.38 (s, 1H, H₁₃), 7.37-7.10 (m, 10H, SnPh₂), 2.38 (s, 6H, H₁₅+H₁₆). ¹³C NMR (300 MHz, DMSO-d₆, ppm): 178.5 (C₁), 170.7 (C₄), 156.5 (C₁₄), 155.5 (Ph), 150.8 (C₂), 145.9 (C₃), 136.6 (C₁₂), 133.8 (Ph), 130.6 (C₇), 129.3 (C₆), 128.3 (Ph), 128.2 (Ph), 127.8 (C₅), 127.2 (C₈), 126.2 (C₁₀), 123.6 (C₁₁), 120.6 (C₉), 110.8 (C₁₃), 16.3 (C₁₅), 15.7 (C₁₆). IR (KBr): ν/cm^{-1} 3439w $\nu(\text{OH})$, 3268w and 3315w $\nu(\text{NH})$, 1636w $\nu(\text{CO})$, 1522m thioamide II, 821w thioamide IV. MS (ESI⁺): m/z 616.08 [M+H]⁺, 583.03 [M-Ph]⁺. Λ_M ($\Omega^{-1}\text{cm}^2\text{mol}^{-1}$, DMF): 3.2.

Complexes **1-3** were also obtained from the triorganotin(IV) derivatives SnR₃Cl in the presence of two equivalents of lithium hydroxide.

Synthesis of HATs derivatives

[SnMe₃L²], **4**. To a suspension of 150 mg (0.94 mmol) of HATs and 40 mg (0.95 mmol) of LiOH.H₂O in 10 mL of absolute ethanol, a solution of 188 mg (0.94 mmol) of SnMe₃Cl in 2 mL of the same solvent was added. The mixture was stirred under reflux for 3 days. Then the solvent was completely removed at reduced pressure and a brown oil was obtained. A pale orange solid was obtained by addition of hexane to a solution of the oil in diethyl ether. This solid was filtered off and vacuum-dried. Yield 221 mg, 79%. Found: C, 31.49; H, 4.95; N, 13.87; S, 10.59. SnC₈H₁₅N₃S requires C, 31.60; H,

4.98; N, 13.83; S, 10.52. ^1H NMR (300 MHz, DMSO- d_6 , ppm): 2.46 (s, 3H, H_4), 2.36 (s, 3H, H_5), 0.52 (s, 9H, SnMe_3 , $^2J(^{119}\text{Sn}-^1\text{H})=64$ Hz). ^{13}C NMR (300 MHz, DMSO- d_6 , ppm): 174.5 (C_1), 158.6 (C_2), 152.4 (C_3), 21.6 (C_4), 18.9 (C_5), 0.5 (Me). IR (KBr): ν/cm^{-1} 1537w $\nu(\text{CN})$, 1505w thioamide II, 771s thioamide IV. MS (ESI^+): m/z 631.00 $[2\text{M}+\text{Na}]^+$, 469.97 $[\text{Sn}_2\text{Me}_6\text{L}^2]^+$, 342.94 $[\text{M}+\text{K}]^+$, 327.99 $[\text{M}+\text{Na}]^+$, 238.03 $[\text{SnMe}_3\text{CSN}_2]^+$, 164.97 $[\text{SnMe}_3]^+$. Λ_{M} ($\Omega^{-1}\text{cm}^2\text{mol}^{-1}$, DMF): 1.9. Slow evaporation of a solution in CH_3Cl afforded crystals suitable for single crystal X-ray diffraction.

$[\text{SnBu}_3\text{L}^2]$, 5. This complex was obtained following the same procedure described for the synthesis of **4** but adding 0.25 mL (0.94 mmol) of SnBu_3Cl . The mixture was refluxed for 48 h and the solvent was removed at reduced pressure until a brown oil was obtained. Yield 361 mg, 89%. Found: C, 47.65; H, 7.88; N, 10.01; S, 7.27. $\text{SnC}_{17}\text{H}_{33}\text{N}_3\text{S}$ requires C, 47.45; H, 7.74; N, 9.77; S, 7.44. ^1H NMR (300 MHz, DMSO- d_6 , ppm): 2.35 (s, 3H, H_4), 2.07 (s, 3H, H_5), 1.57 (m, 2H, $\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 1.27 (sex, 2H, $\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$, $^3J=7.3$ Hz), 1.14 (m, 2H, $\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 0.83 (t, 3H, $\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$, $^3J=7.3$ Hz). ^{13}C NMR (300 MHz, DMSO- d_6 , ppm): 174.0 (C_1), 158.6 (C_2), 152.5 (C_3), 30.9 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 28.5 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$, $^2J(^{119}\text{Sn}-^{13}\text{C})=106$ Hz), 26.9 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$, $^1J(^{119}\text{Sn}-^{13}\text{C})=295$ Hz), 21.5 (C_4), 19.8 ($\text{CH}_2\text{-CH}_2\text{-CH}_2\text{-CH}_3$), 18.8 (C_5). MS (ESI^+): m/z 718.26 $[\text{Sn}_2\text{Bu}_6\text{L}^1]^+$, 454.14 $[\text{M}+\text{Na}]^+$. Λ_{M} ($\Omega^{-1}\text{cm}^2\text{mol}^{-1}$, DMF): 3.4.

$[\text{SnPh}_3\text{L}^2]$, 6. This complex was obtained following the same procedure described for the synthesis of **4** but adding 364 mg (0.94 mmol) of SnPh_3Cl . The mixture was stirred under reflux for 1 h. The reddish orange solid was filtered off, washed with ethanol and vacuum-dried. Yield 375 mg, 73%. Found: C, 56.20; H, 4.42; N, 8.82; S, 6.41. $\text{SnC}_{23}\text{H}_{21}\text{N}_3\text{S}$ requires C, 56.34; H, 4.32; N, 8.58; S, 6.53. ^1H NMR (300 MHz, DMSO- d_6 , ppm): 7.76 (m, 5H, Ph), 7.39 (m, 10H, Ph), 2.37 (s, 3H, H_4), 2.21 (s, 3H, H_5). ^{13}C

NMR (300 MHz, DMSO- d_6 , ppm): 173.4 (C_1), 159.4 (C_2), 152.9 (C_3), 136.5 (Ph, $^3J(^{119}\text{Sn}-^{13}\text{C})=87$ Hz), 129.5 (Ph), 128.9 (Ph, $^2J(^{119}\text{Sn}-^{13}\text{C})=120$ Hz), 21.4 (C_4), 18.9 (C_5). IR (KBr): ν/cm^{-1} 1592w $\nu(\text{CN})$, 1479m thioamide II, 857w thioamide IV. MS (ESI $^+$): m/z 989.10 $[2\text{M}+\text{Na}]^+$, 838.06 $[\text{Sn}_2\text{Ph}_6\text{L}^1]^+$, 514.04 $[\text{M}+\text{Na}]^+$, 351.01 $[\text{SnPh}_3]^+$. Λ_{M} ($\Omega^{-1}\text{cm}^2 \text{mol}^{-1}$, DMF): 3.8. Slow evaporation of a solution in DMSO yielded crystals suitable for single crystal X-ray diffraction.

The reactivity of HATs with SnR_2Cl_2 under different reaction conditions was also explored, but in all the cases the starting materials were recovered.

X-ray crystallography

Data for compound $[\text{1EtOH}]\text{EtOH}$ were acquired using an Agilent Supernova Dual diffractometer equipped with an EosS2 CCD plate detector using a mirror monochromator (Cu K_{α} radiation $\lambda = 1.54184$ Å) and for L^1H_2 , 4 and 6 were acquired using a Bruker Kappa Apex-II diffractometer equipped with an Apex-II CCD area detector using a graphite monochromator (Mo K_{α} radiation, $\lambda = 0.71073$ Å). The substantial redundancy in data allows empirical absorption corrections (SADABS)^[43] to be applied using multiple measurements of symmetry-equivalent reflections. The raw intensity data frames were integrated with the SAINT program, which also applied corrections for Lorentz and polarization effects.^[44] The software package SHELXTL version 6.10 was used for space group determination, structure solution and refinement. The structures were solved by direct methods (SHELXS-97),^[45] completed with difference Fourier syntheses, and refined with full-matrix least squares using SHELXL-2014 minimizing $\omega(F_0^2 - F_c^2)$. Weighted R factors (R_w) and all goodness of fit S are based on F^2 ; conventional R factors (R) are based on F .^[46] All non-hydrogen atoms were refined with anisotropic displacement parameters. C-H hydrogen atoms were

placed onto calculated positions and refined riding on their parent atom and those on nitrogen and oxygen were located in a difference Fourier map and their coordinates and isotropic thermal parameters subsequently refined. All scattering factors and anomalous dispersion factors are contained in the SHELXTL 6.10 program library.

CCDC 1562728-1562731 for L^1H_2 , [1EtOH]EtOH, 4 and 6 respectively contain the supplementary crystallographic data for this paper. These data can be obtained free of charge from the Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

Antimicrobial activity

The *in vitro* antimicrobial properties of the designed ligands and their corresponding organotin(IV) complexes were tested against a wide spectrum of microorganisms by determining their minimum inhibitory concentrations (MICs) by means of the serial double dilution procedure.^[47] The antibacterial activity was detected against Gram positive (*Bacillus megaterium* BGSC 7A2, *Bacillus subtilis* ATCC 6633, *Sarcina lutea* ATCC 9341, *Staphylococcus aureus* ATCC 25923, *Staphylococcus epidermidis* ATCC 12228, clinical isolate of *Streptococcus agalactiae*) and Gram negative (*Escherichia coli* ATCC 8739, *Haemophilus influenzae* ATCC 19418, *Proteus vulgaris* ATCC 13315, *Pseudomonas aeruginosa* ATCC 9027, *Salmonella typhimurium* ATCC 14028) bacteria. For the antifungal bioassay, yeasts (*Candida albicans* ATCC 10231, *Candida tropicalis* ATCC 1369, *Saccharomyces cerevisiae* ATCC 9763 and clinical isolates of *Candida guilliermondii*, *Candida parapsilosis* and *Cryptococcus neoformans*) and moulds (*Aspergillus niger* ATCC 6275, clinical isolates of *Aspergillus flavus*, *Aspergillus fumigatus*, *Epidermophyton floccosum*, *Trichophyton interdigitalis*, *Trichophyton mentagrophytes*, *Trichophyton rubrum*, *Trichophyton soudanense*) were used as test microorganisms.

Compounds were dissolved in dimethyl sulfoxide and diluted in the media (Haemophilus test medium for *Haemophilus influenzae*, Mueller Hinton broth for the other bacteria, Sabouraud liquid medium for fungi) (Oxoid, Basingstoke, UK) so as to achieve the concentration range of 200-0.0015 $\mu\text{g mL}^{-1}$. In all cases, DMSO never exceeded the amount of 1% v/v and, to ensure that DMSO had no effect on microbial growth, control tubes with media supplemented with the solvent were also maintained in each experiment. Aliquots of the bacterial and fungal suspensions were mixed with the chemicals to obtain an inoculum size of $5 \cdot 10^5$ colony forming units/mL and $5 \cdot 10^3$ cells/mL, respectively. Ampicillin and miconazole were tested under the same conditions as antibacterial and antifungal reference drugs, respectively. After an incubation period of 24 h at 37 °C for bacteria and of 48 h at 30 °C for fungi, the minimum inhibitory concentrations (MIC, $\mu\text{g mL}^{-1}$) were recorded as the lowest concentrations of compound that inhibit visible growth of the tested microorganisms.

The minimum bactericidal concentrations (MBCs) and the minimum fungicidal concentrations (MFCs) were determined by subculturing on fresh sterile medium 100 μL of culture from each sample remained clear and incubating the obtained mixtures at 37 °C for 24 h (bacteria) and at 30 °C for 48 h (fungi). MBC and MFC values represented the lowest concentration of compound ($\mu\text{g mL}^{-1}$) needed for the reduction of the initial inoculum of 99.9%.

Complexes **5** and **6**, exhibiting strong inhibition of *A. niger*, were further on screened for their antifungal activity by mean of serial plate dilution method against several human pathogenic moulds freshly isolated from pathological material. Each fungal strain was suspended in physiologic saline and streaked onto the surface of Sabouraud Dextrose Agar (Oxoid, Basingstoke, UK) plates containing 200-0.0015 $\mu\text{g mL}^{-1}$ of compound. MICs were determined after incubation of the plates at 25 °C for 14

days. The antifungal activity was compared with that of miconazole used as standard drug.

Each experiment was carried out in triplicate under sterile conditions and the reported results were obtained from three independent measurements.

Docking procedure

The three-dimensional (3D) structures of the metal complexes which are essential for docking has been obtained from optimized structure of density functional theory (DFT) computation using B3LYP method and LanL2DZ basis set by using Gaussian 09 and Gauss View 5.0.9. [48-50] The drug-relevant properties of the complexes have been analyzed following Lipinski's rule of five.[51] According to the rule, molecules possessing drug-like properties should have a molecular weight lower than 500 Dalton, high lipophilicity (LogP below 5), less than five hydrogen bond donors and less than ten hydrogen bond acceptors.

Protein structure of DHPS of *Staphylococcus aureus* has been retrieved from protein data bank (PDB id: 1AD4) and only A chain is used for docking from A and B chains. The protein (DHPS) and ligand (metal complexes) structures have been modified by Autodock Tools [52] The A chain of *S. aureus* DHPS is modified by removing water and bound ligand. Missing amino acids have been checked and hydrogen bonds have been added to the protein structure. Center Grid box x:36.056, y:4.39, z:36.65 and number of points in x,y,z dimensions are considered as 30x30x30 Å³ respectively and grid spacing has been taken as 0.3750 Å. Ligands have been prepared by adding Gasteiger charges, detecting root and choosing torsions from torsion tree of Autodock Tools panel. [53] Docking procedure has been performed by using Lamarckian genetic algorithm. [54]

Docking of metal complexes has been carried out with AutoDock 4.2.6 on windows platform with 8 GB RAM and Intel I 5 processor. ^[52]

Results and discussion

Synthesis

The mono(thiosemicarbazone) ligand HATs, resulting from the condensation in water of 2,3-butanedione and thiosemicarbazide, was synthesized. The excess of dione prevents from double condensation and the ligand was obtained completely pure and in quantitative yield. The synthesis of L^1H_2 was achieved by reaction of HATs with 3-hydroxy-2-naphthohydrazide to yield the desired ligand in good yield.

The reactivity of these two ligands against organotin(IV) derivatives has been explored (Scheme 1). Reactions of SnR_2Cl_2 ($R=Me, Bu, Ph$) with the dissymmetric ligand L^1H_2 in absolute ethanol and under reflux for one hour yield complexes **1-3**, with general formula $[SnR_2L^1]$, in good yield. Two equivalents of lithium hydroxide were added to induce ligand deprotonation. It is worth to note that, similarly to the ligand with cyclohexyl and in contrast with the isopropyl derivative,^[39] symmetrisation of the ligand to give the bis(hydrazone) complexes does not occur.

Regarding the reactions of HATs with SnR_3Cl ($R=Me, Bu, Ph$) in basic medium, *in situ* cyclization of the organic precursor takes place to yield a 1,2,4-triazine-3-thione that behaves as a singly deprotonated donor and coordinates to the tin in a mono or bidentate manner to form complexes **4-6** with formula $[SnR_3L^2]$. Surprisingly, it was not possible to isolate any complex from the reactions with SnR_2Cl_2 , although several reaction conditions were changed, such as solvent (THF, acetonitrile), pH (with and without lithium hydroxide), temperature (reflux and room temperature) and reaction time (from 1 to 72 hours).

The elemental analysis of all the complexes shows a 1:1 metal to ligand ratio, as well as the absence of chlorides, confirming the double and mono deprotonation of ligands L^1H_2 and L^2H , respectively. In addition, the elemental analysis data of complexes **4-6** are in agreement with the cyclization of HATs to yield the 1,2,4-triazine-3-thione.

The mass spectra of complexes **1-2** only show the peak corresponding to $[M+H]^+$ and in complex **3** also to $[M-Ph]^+$. By contrast, in the spectra of complexes **4-6** a number of peaks corresponding to different fragmentations and associations are observed, but in all of them the presence of the pseudo molecular ion $[M+Na]^+$ is present.

Crystal structures

The molecular structure of four compounds could be determined by single crystal X-ray diffraction. The crystallographic and refinement data are collected in Table 1 and selected bond distances are listed in Tables 2 and 3. The crystals of $[1EtOH]EtOH$ were very small and could not be measured in a standard equipment, but they could be measured in a Supernova diffractometer. Due to its tiny size the final R value is high (0.1356) and the structure presents other refinement problems, but the crystal structure is strongly supported by all the spectroscopic and spectrometric techniques used, as well as by the crystal structures previously reported for analogous complexes bearing NH^iPr or $NHCy$ instead of NH_2 .^[39]

The asymmetric unit of L^1H_2 is made up by one ligand (Figure 1) and one DMSO molecule held together in the crystal packing by hydrogen bonds. The ligand core can be considered planar, with a maximum deviation of 0.37 Å for S1. The disposition around the C2-C3 bond is *trans*, as well as the C=N and C=S groups of the thiosemicarbazone branch, whereas the C=N and C=O of the hydrazone limb are in *cis*

disposition. Therefore, formation of complexes, in which the ligand acts as a N₂SO chelate, takes place via two 180° rotations around C2-C3 and C1-N2 bonds. The ligand shows some charge delocalization and most of the distances are intermediate between those of single and double bonds. There is one intramolecular hydrogen bond between N5 and the OH and a hydrogen bond between the C=O group with the OH of another molecule that leads to the formation of infinite chains running parallel to *c* axis, that are linked by N1-H1B...S1 hydrogen bonds to form sheets in the *ac* plane. Finally, these sheets are connected through hydrogen bonds with the DMSO molecule to give a 3D network.

When complex **1** is recrystallized in ethanol two solvent molecules are incorporated, one coordinated to the metal and the other as a crystallization molecule to give [1EtOH]EtOH. Coordination of a donor solvent such as H₂O, EtOH, DMF or DMSO was previously observed in related complexes.^[39] The ligand is doubly deprotonated and binds to the tin as a N₂SO tetradentate chelate, giving rise to the formation of three five-member chelate rings that confers great stability to the complex. The tin atom has distorted pentagonal bipyramid geometry with the methyl groups occupying the axial positions (Figure 2). Although the ligand is doubly deprotonated, bond distances within the complex are similar to those of the free ligand and only can be observed a significant enlargement of C=S and C=O bonds due to their coordination to the metal. The ligand core is strongly deviated from planarity, with a maximum deviation from the least-squares plane defined by S1-C1-N2-N3-C2-C3-N4-N5-C4-O1 of 0.2544 Å for S1. There is an intramolecular hydrogen bond between N5-H5 and O2. Moreover, exists a hydrogen bond between the OH of the coordinated ethanol molecule and the C=O group of another unit that links the molecules in dimers that are cross-

linked through an extended hydrogen bond network involving N2, the NH₂ group and two OH groups (from naphthol and ethanol of crystallization) to form a 3D architecture.

The crystal structures of complexes **4** and **6** confirm the cyclization of HATs to yield the 1,2,4-triazine-3-thione. In both complexes the ligand behaves as a monoanionic donor, but its coordination mode is different. Analysis of the bond distances reveals that in both compounds the ligand is the thiol tautomer. In complex **4** the distance between N1 and the tin atom is 3.098 Å, so it can be considered that the triazine formally binds only through the sulphur atom and that the metal is in a C₃S coordination environment (Figure 3). The value of the τ_4 parameter is 0.90, corresponding to a distorted tetrahedral arrangement ($\tau_4=0$ for square-planar and $\tau_4=1$ for tetrahedral).^[55] The steric congestion imposed by the five methyl groups prevents from π - π stacking.

In complex **6** the Sn1-N3 bond distance, although long, is in the range of other compounds found in the literature,^[56,57] so the ligand coordinates to the metal in a bidentate manner, giving rise to the formation of a four-member chelate ring, coordination mode that has been previously observed in related complexes.^[38,58,59] The metal is in a C₃NS coordination environment (Figure 4) with a value of the τ_5 parameter of 0.68, corresponding to a strongly distorted trigonal bipyramid environment ($\tau_5=0$ for square-pyramid and $\tau_5=1$ for trigonal bipyramid).^[60] In this complex there are not π - π stacking, but there is a CH- π interaction between the triazine ring and H5B, with a distance of 2.953 Å, that links the molecules into dimers.

IR spectroscopy

In the IR spectra of all the complexes can be observed the bands corresponding to the organic groups attached to the tin ion, confirming the presence of the organotin(IV)

moieties. In the IR spectra of complexes **1-3** a decrease in the number of $\nu(\text{NH})$ bands due to ligand deprotonation is observed. The bands corresponding to $\nu(\text{C=O})$, $\nu(\text{C=N})$ and $\nu(\text{C=S})$ are shifted with respect to the free ligand, suggesting coordination of these groups to the metal. In the spectra of complexes **4** and **6** cyclization of HATs can be clearly observed since the bands belonging to $\nu(\text{C=O})$ and $\delta(\text{HNNH})$ have disappeared. Moreover, deprotonation is also confirmed by the absence of any bands assignable to $\nu(\text{NH})$. Comparison with the ligand to establish its coordination mode was not possible since the uncoordinated triazine was not isolated.

NMR spectroscopy

^1H and ^{13}C NMR chemical shifts of L^1H_2 , HATs and the six complexes are listed in the experimental section (the data for L^2H are not reported since the uncoordinated neutral triazine ligand could not be isolated). The ^1H NMR spectrum of L^1H_2 shows all the signals expected and both the chemical shifts and the integrals are in agreement with its structure. In the spectra of complexes **1-3** the signals corresponding to H_2 and H_5 have disappeared, confirming the ligand double deprotonation. The signal of the OH group is more deshielded than in the free ligand due to the coordination to the tin. Moreover, there is only one signal for H_{1a} and H_{1b} , whereas in the free ligand these two protons are not equivalent. Additional signals corresponding to the organic groups (Me, Bu and Ph) bonded to the tin can be observed for the three complexes. In the spectrum of complexes **4-6** the signals at 8.71 and 8.10 have disappeared due to the cyclization to form the triazine and the absence of any signals over 8.0 ppm indicates that the ligand is deprotonated. Similarly to complexes **1-3**, the presence of Me, Bu or Ph groups can also be observed.

The ^{13}C NMR spectra of complexes **1-3** shows that the signals attributable to C=O, C=N and C=S are shifted compared to the free ligand, suggesting coordination of these groups to the metal. In the spectra of complexes **4-6** the signal corresponding to the C=O group has disappeared, supporting the formation of the triazine. In the six complexes the signals of the organic groups can be also clearly observed.

^{119}Sn NMR chemical shift depends on the coordination number and is also very sensitive to the nature of the donor atoms bonded to the metal ion, so it is a useful tool to determine the chemical environment of the tin atom.^[61,62] The ^{119}Sn NMR spectra have been acquired both in solution and in the solid state (Table 4), except for complex **5** that was only studied in solution due to its oily nature, which has permitted to bridge the information gap between the structures found by single crystal X-ray diffraction and the behavior in solution. For complexes **1-3** the chemical shift found in the solid state corresponds to six coordination number, provided by one tetradentate ligand and two organic groups. In solution an increase in the coordination number from six to seven can be observed, due to the coordination of one DMSO molecule to the metal, which also occurs in the crystal structure of complex **1** in ethanol, in which a solvent molecule binds to the metal in the fifth equatorial position of a pentagonal bipyramid. In the case of complexes **4** and **6** the coordination number found in solution and in the solid state is the same, four for complex **4** and five for complex **6**, which is in agreement with the structures found by X-ray diffraction. For complex **5**, the value found in solution corresponds to tetra-coordination, so the structure must be similar to that of complex **1**, in which as a consequence of the high steric demand imposed by the butyl groups, the ligand is monodentate and therefore, the tin atom is in a tetrahedral arrangement. This coordination environment is also supported by the value of the C-Sn-C angle obtained by substitution of $^1J(^{119}\text{Sn}-^{13}\text{C})$ obtained from the ^{13}C NMR spectrum in the

corresponding Lockhart-Manders equation [$^1J(^{119}\text{Sn}-^{13}\text{C}) = 11.40 - 875$],^[63] which is 102.6°.

Antimicrobial activity

The *in vitro* antimicrobial activity of both ligands and their complexes is summarized in Table 5 with reference to antibacterial ampicillin and antifungal miconazole as standard drugs and it is clear that all the tin(IV) complexes were much more active than their corresponding ligands. In order to establish the stability of the complexes, we compared the ^{119}Sn NMR spectra of the compounds in DMSO and those obtained in DMSO+H₂O 1:2. For all the derivatives both are identical, suggesting that the complexes remain unaffected in the presence of water.

None of the ligands demonstrated antimicrobial activity up the concentration of 200 $\mu\text{g mL}^{-1}$, except HATs that inhibited the growth of *B. subtilis* at 100 $\mu\text{g mL}^{-1}$. By contrast, the organotin(IV) complexes exhibited excellent antibacterial activity against Gram positive bacteria, such as *B. subtilis* and *S. aureus*, and Gram negative *H. influenzae*. Tributyltin(IV) complex **5** and triphenyltin(IV) complex **6**, carrying the triazine moiety, have demonstrated to be the most active antibacterial agents, with minimum inhibitory concentrations ranging from 0.3 to 0.7 $\mu\text{g mL}^{-1}$. Also dibutyltin(IV) and diphenyltin(IV) complexes **2** and **3**, having the mixed thiosemicarbazone/hydrazone ligand, were found to possess high inhibitory properties against the same microorganisms (MIC 3 $\mu\text{g mL}^{-1}$ for *B. subtilis*, 12 $\mu\text{g mL}^{-1}$ for *H. influenzae* and 25 and 6 $\mu\text{g mL}^{-1}$, respectively, for *S. aureus*). Furthermore, both methyl-containing complexes **1** and **4** showed remarkable toxicity towards *B. subtilis* (MICs 6-12 $\mu\text{g mL}^{-1}$), but a poor inhibition of *S. aureus* and *H. influenzae* (MICs 100 $\mu\text{g mL}^{-1}$). Among bacteria, Gram positive *B. subtilis* was the most sensitive microorganism (MICs 0.3-12 $\mu\text{g mL}^{-1}$), whereas Gram negative *E. coli* was resistant to

all tested complexes, with the exception of trimethyltin(IV) derivative complex **4**, showing MIC value $200 \mu\text{g mL}^{-1}$. In addition to this, the triazine complexes **5** and **6** demonstrated very strong antifungal effects against *A. niger* mould at $0.03\text{-}0.3 \mu\text{g mL}^{-1}$ and against both *S. cerevisiae* and *C. tropicalis* yeasts at $1.5\text{-}3 \mu\text{g mL}^{-1}$. *A. niger* was found to be the most susceptible fungus, being also inhibited by complexes **2-4** with minimum inhibitory concentrations in the range $12\text{-}50 \mu\text{g mL}^{-1}$. A moderate antifungal activity was exhibited against *S. cerevisiae* by complex **2** at $25 \mu\text{g mL}^{-1}$. It is worth to note, that complexes **1-3** are much more active than derivatives with the ligands bearing NH^iPr or NHCy groups instead of NH_2 , which possess a poor antimicrobial potency,^[39] showing the great effect of the ligand backbone in the antimicrobial activity of the complexes.

As an extension of our study, we investigated the antimicrobial properties of the most active complexes **5** and **6** against a wide spectrum of bacterial and fungal human pathogenic strains (Table 6). Both complexes exhibited a strong toxicity against the tested microorganisms, and especially significant was the triphenyl derivative complex **6**, which showed the highest inhibitory effect. Its MIC values resulted to be in the range $0.15\text{-}0.7 \mu\text{g mL}^{-1}$ for Gram positive bacteria, Gram negative *P. aeruginosa*, yeasts *C. albicans* and *C. neoformans* and dermatophyte *E. floccosum*, while Gram negative *P. vulgaris*, *Aspergillus* and *Trichophyton spp.* were susceptible to concentrations of $1.5\text{-}6 \mu\text{g mL}^{-1}$.

In order to explore the kind of the detected antimicrobial activity, minimum bactericidal concentrations (MBC) and minimum fungicidal concentrations (MFC) were determined. Based on the results presented in Tables 5 and 6, it is found that MBC and MFC values are higher than the corresponding MICs for all the microorganisms tested, suggesting that these compounds act as bacteriostatic and fungistatic agents. In general,

new tested compounds demonstrated to possess similar or lower antimicrobial properties, when compared to ampicillin and miconazole positive controls. On the other hand, it is noteworthy that complexes **5** and **6** were more toxic than ampicillin in the inhibition of Gram positive *S. epidermidis* and Gram negative *P. aeruginosa*, as well as than miconazole against yeasts *S. cerevisiae* and *C. albicans* and against moulds *A. niger* and *A. flavus*.

Differential antimicrobial behaviour was noticeable among the studied complexes. Specifically, in both $[\text{SnR}_2\text{L}^1]$ and $[\text{SnR}_3\text{L}^2]$ series the inhibitory properties increase in the order $\text{Me} < \text{Bu} \leq \text{Ph}$, which could be related to an increase in the lipophilicity that allows the compounds to cross the microorganism membrane. In fact, complexes **1** and **4** containing the methyl group showed moderate activity, but the replacement of this substituent by the longer butyl chain (complexes **2** and **5**) enhanced both antibacterial and antifungal activity. In several cases, this toxicity improved for complexes **3** and **6**, where the alkyl chain system is replaced by the aromatic phenyl ring

It is also noted that the triorganotin(IV) complexes **4-6**, containing the triazine moiety, showed enhanced and more extensive antimicrobial activity with respect to the corresponding diorganotin(IV) **1-3**, carrying the thiosemicarbazone/hydrazone ligand. This could be due to the greater lipophilic character of the aromatic residue, which plays an important role in the antimicrobial properties of these complexes, but also to the smaller ligand size. According to these two factors $[\text{SnPh}_3\text{L}^2]$ **6** demonstrated to be the most potent as both antibacterial and antifungal agent.

Molecular docking

The newly synthesized compounds displayed excellent antibacterial activity, in particular triazine derivatives **4-6**. Complexes **5** and **6** exhibit high inhibition of *S. aureus* that can cause a wide range of illnesses from minor skin infections to life-

threatening diseases. Nowadays, *S. aureus* has become resistant to many commonly used antibiotics and only 2% of all *S. aureus* isolates are found to be sensitive to penicillin,^[64] so we choose DHPS from *S. aureus* to perform the docking studies. Dihydropteroate synthase (DHPS) is an enzyme involved in the bacterial folate synthesis pathway which is a crucial pathway for synthesizing amino acids. Protein structure of DHPS from *S. aureus* is downloaded and docked with complexes **1-6**. The complexes have hydrophilic and hydrophobic parts to bind through hydrogen bonding and different electrostatic interactions. Amongst the docked conformations, complex **6** has shown best binding affinity (-8.5 kcal/mol) and complex **5** has shown the second best binding affinity (-8.1 kcal/mol) and both have showed stronger binding affinity than ampicillin (-5.5 kcal/mol) (Table 7). These data are in agreement with the antimicrobial activity results, since complex **6** is the most active compound, followed by complex **5**, in all the microorganisms tested. Table 8 lists all the interactions between the DHPS amino-acids residues and complexes **5** and **6** and ampicillin. As can be observed, ampicillin has only hydrogen bond interactions (Figure 5), while complex **5** has also hydrophobic ones (Figure 6) and in complex **6** there are as well electrostatic interactions (Figure 7), resulting in a significantly enhanced binding affinity. Conversely to binding affinity, ampicillin displays better activity against *S. aureus* than complexes **5** and **6**, which could be due to these compounds do not follow Lipinski's filter due to their log p value (Table 7), indicating less 'drug-like' nature. The only compound that follows this filter is complex **1**, but although its binding affinity (- 6.7 kcal/mol) is also stronger than that of ampicillin, only shows a moderate antimicrobial activity against *S. aureus*.

Conclusions

Three complexes, with general formula $[\text{SnR}_2\text{L}^1]$ are obtained from the reaction of L^1H_2 with SnR_2Cl_2 ($\text{R}=\text{Me}, \text{Bu}, \text{Ph}$) in which the ligand is doubly deprotonated and acts as a tetradentate N_2SO chelating donor. Reaction of the ligand HATs with SnR_3Cl induces an intramolecular cyclization to yield three 1,2,4-triazine-3-thione derivatives with formula $[\text{SnR}_3\text{L}^2]$, in which the donor behaves as mono or bidentate, depending on the nature of the organic groups. All the complexes display good antimicrobial activity against bacteria and fungi. The best results are found for the triazine derivatives $[\text{SnR}_3\text{L}^2]$ ($\text{R}=\text{Bu}$ **5**, Ph **6**) that have demonstrated high antimicrobial efficacy against all the strains tested except for Gram negative *Escherichia coli* and *S. Typhimurium*. These two complexes exhibit their best potency against fungi (both yeasts and moulds) with MIC and MFC values in some cases lower than those of miconazole, used as positive control. Substitution of cyclohexyl or isopropyl by NH_2 enhance the antimicrobial activity of the complexes with thiosemicarbazone/hydrazone ligands, which could be related to the ligand size and not to its lipophilicity, which seems to be controlled in the complex by the number and nature of the organic groups attached to the tin atom. Molecular docking reveals that complexes **5** and **6** shows better affinity to DHPS protein of *S. aureus* than ampicillin, although their *in vitro* activity is slightly higher, what could be attributed to the fact they do not follow Lipinski's filter due to their $\log p$ values.

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References

- [1] K.E. Jones, N.G. Patel, M.A. Levy, A. Storeygard, D. Balk, J.L. Gittleman, P. Daszak, *Nature* **2008**, *451*, 990-993.
- [2] C. Nathan, *Nature* **2004**, *431*, 899-902.
- [3]. T.C. White, K.A. Marr, R.A. Bowden, *Clin. Microbiol. Rev.* **1998**, *11*, 382-402.
- [4] A. Dessen, A.M. Di Guilmi, T. Vernet, O. Dideberg, *Curr. Drug. Targets Infect. Disord.* **2001**, *1*, 63-77.
- [5] F.C. Tenovera, *Clin. Infect. Dis.* **2001**, *33*, S108-S115.
- [6] R. Leclercq, *Clin. Microbiol. Infect.* **2009**, *15*, 224-231.
- [7] L. Hu, H. Wang, T. Xia, B. Fang, Y. Shen, Q. Zhang, X. Tian, H. Zhou, J. Wu, Y. Tian, *Inorg. Chem.*, **2018**, *57*, 6340-6348
- [8] M. Gielen, M. Biesemans, R. Willen, *Appl. Organomet. Chem.* **2005**, *19*, 440-450.
- [9] M. Gielen, E.R.T. Tiekink in *Metallotherapeutic Drug and Metal-Based Diagnostic Agents: 50 Tin Complexes and Their Therapeutic Potential*, Wiley, New York, **2005**.
- [10] A.S.L. Barbosa, J.S. Guedes, D. Rozendo da Silva, S.M.P. Meneghetti, M.R. Meneghetti, A.E. da Silva, M. Vital de Araujo, M.S. Alexandre-Moreira, T. Mendonca de Aquino, J. Pinto de Siqueira Jr., R.S. Aquino de Araújo, R. Marques Duarte da Cruz, F.J. Bezerra Mendonça-Jr., *J. Inorg. Biochem.*, **2018**, *180*, 80-88.
- [11] T.S.B. Baul, *Appl. Organomet. Chem.* **2008**, *22*, 195-204.
- [12] M. Sirajuddin, V. McKee, M. Tariq, S. Ali, *Eur. J. Med. Chem.*, **2018**, *143*, 1903-1918.
- [13] T. Sedaghat, M. Yousefi, G. Bruno, H. Amiri Rudbari, H. Motamedi, V. Nobakht, *Polyhedron* **2014**, *79*, 88-96.

- [14] F. Wang, H. Yin, J. Cui, Y. Zhang, H. Geng, M. Hong, *J. Organomet. Chem.* **2014**, 759, 83-91.
- [15] L. Dawara, R.V. Singh, *App. Organomet. Chem.*, **2011**, 25, 643-652.
- [16] M. Nath, P.K. Saini, *Dalton Trans.*, **2011**, 40, 7077-7121.
- [17] K.T. Mahmudov, M.F.C. Guedes da Silva, M.N. Kopylovich, A.R. Fernandes, A. Silva, A. Mizar, A J.L. Pombeiro, *J. Organomet. Chem.* **2014**, 760, 67-73.
- [18] M.S. Sarma, A. Saha, A. Roy, *Appl. Organometal. Chem.* **2008**, 22, 369-377.
- [19] G. Yenişehirli, N.A. Öztaş, E. Şahin, M. Çelebier, N. Ancın, S.G. Öztaş, *Heteroat. Chem.* **2010**, 21, 373-385.
- [20] J.S. White, J.M. Tobin, J.J. Coney, *Can. J. Microbiol.* **1999**, 45, 541-554.
- [21] J.J. Cooney, S. Wuertz, *J. Ind. Microbiol.* **1989**, 4, 375-402.
- [22] J.S Casas, M.S. García-Tasende, J. Sordo, *Coord. Chem. Rev.* **2000**, 209, 197-261.
- [23] T.S. Lobana, R. Sharma, G. Bawa, S. Khanna, *Coord. Chem. Rev.* **2009**, 253, 977-1050.
- [24] E. López-Torres, M.A. Mendiola, *Dalton Trans.* **2009**, 7639-7647.
- [25] E. López-Torres, M.A. Mendiola, C.J. Pastor, B. Souto Pérez, *Inorg. Chem.* **2004**, 43, 5222-5230.
- [26] S. Rodríguez-Hermida, A.B. Lago, L. Cañadillas-Delgado, R. Carballo, E.M. Vázquez-López, *Cryst. Growth Des.* **2013**, 13, 1193-1205.
- [27] A. Castiñeiras, N. Fernández-Hermida, R. Fernández-Rodríguez, I. García-Santos, *Cryst. Growth Des.* **2012**, 12, 1432-1442.
- [28] P. Kumar, B. Narasimhan, *Mini Rev. Med. Chem.* **2013**, 13, 971-987.
- [29] R. Narang, B. Narasimhan, S. Sharma, *Curr. Med. Chem.* **2012**, 19, 569-612.

- [30] M.C. Rodríguez-Argüelles, P. Tourón-Touceda, R. Cao, A.M. García-Deibe, P. Pelagatti, C. Pelizzi, F. Zani, *J. Inorg. Biochem.* **2009**, *103*, 35-42 and references therein.
- [31] A.G. Quiroga, C. Navarro Ranninger, *Coord. Chem. Rev.* **2004**, *248*, 119-133.
- [32] G.L. Parrilha , J.G. da Silva, L.F. Gouveia, A.K. Gasparoto, R.P. Dias , W.R. Rocha, D.A. Santos, N.L. Speziali, H. Beraldo, *Eur. J. Med. Chem.* **2011**, *46*, 1473-1482 and references therein.
- [33] X. Shang, B. Zhao, G. Xiang, M.F.C. Guedes da Silva, A.J.L. Pombeiro, *RSC Adv.* **2015**, *5*, 45053-45060.
- [34] S.Y. Ebrahimipour, I. Sheikhshoaie, J. Simpson, H. Ebrahimnejad, M. Dusek, N. Kharazmia, V. Eigner, *New J. Chem.* **2016**, *40*, 2401-241
- [35] M. Mohamadi, Y.S. Ebrahimipour, J. Castro, M. Torkzadeh-Mahani, *Journal of Photochemistry and Photobiology, B: Biology* **2016**, *158*, 219-227.
- [36] E. López-Torres, F. Zani, M.A. Mendiola, *J. Inorg. Biochem.*, **2011**, *105*, 600-608.
- [37] A. Bacchi, A. Bonardi, M. Carcelli, P. Mazza, P. Pelagatti, C. Pelizzi, G. Pelizzi, C. Solinas, F. Zani, *J. Inorg. Biochem.* **1998**, *69*, 101-112 and references therein.
- [38] E. López-Torres, M.A. Mendiola, C.J. Pastor, J.R. Procopio, *Eur. J. Inorg. Chem.* **2003**, 2711-2717.
- [39] C. González-García, A. Mata, F. Zani, M.A. Mendiola, E. López-Torres, *J. Inorg. Biochem.* **2016**, *163*, 118-130.
- [40] E. Lionta, G. Spyrou, D.K. Vassilatis, Z. Cournia, *Curr. Top. Med. Chem.* **2014**, *14*, 1923-38.

- [41] P. Szymański, M. Markowicz, E. Mikiciuk-Olasik, *Int. J. Mol. Sci.* **2012**, *13*, 427-452.
- [42] R.J. Henry, *Bact. Rev.* **1943**, *7*, 175-262.
- [43] G. M. Sheldrick, SADABS Version 2.03, Program for Empirical Absorption Corrections, Universität Göttingen, Göttingen, Germany, **1997–2001**.
- [44] G.M. Sheldrick, SAINT+NT (Version 6.04) SAX Area-Detector Integration Program, Bruker AXS, Madison, WI, **1997–2001**.
- [45] G.M. Sheldrick, SHELXTL (Version 6.10) Structure Determination Package, Bruker AXS, Madison, WI, **2000**.
- [46] G.M. Sheldrick, *Acta Crystallogr. Sect. A* **1990**, *46*, 467.
- [47] J.H. Jorgensen, J.D. Turnidge in *Manual of Clinical Microbiology*, P.R. Murray, E.J. Baron, M.A. Pfaller, F.C. Tenover, R.H. Tenover (Eds.), American Society for Microbiology, Washington, DC, **1999**, pp. 1526-1554 and 1640-1652.
- [48] M.J. Frisch, G.W. Trucks, H.B. Schlegel, G.E. Scuseria, M.A. Robb, J.R. Cheeseman, G. Scalmani, V. Barone, B. Mennucci, G.A. Petersson, H. Nakatsuji, M. Caricato, X. Li, H.P. Hratchian, A.F. Izmaylov, J. Bloino, G. Zheng, J.L. Sonnenberg, M. Hada, M. Ehara, K. Toyota, R. Fukuda, J. Hasegawa, M. Ishida, T. Nakajima, Y. Honda, O. Kitao, H. Nakai, T. Vreven, J.A. Montgomery Jr, J.E. Peralta, F. Ogliaro, M. Bearpark, J.J. Heyd, E. Brothers, K.N. Kudin, V.N. Staroverov, R. Kobayashi, J. Normand, K. Raghavachari, A. Rendell, J.C. Burant, S.S. Iyengar, J. Tomasi, M. Cossi, N. Rega, J.M. Millam, M. Klene, J.E. Knox, J.B. Cross, V. Bakken, C. Adamo, J. Jaramillo, R. Gomperts, R.E. Stratmann, O. Yazyev, A.J. Austin, R. Cammi, C. Pomelli, J.W. Ochterski, R.L. Martin, K. Morokuma, V.G. Zakrzewski, G.A. Voth, P. Salvador, J.J. Dannenberg, S.

- Dapprich, A.D. Daniels, O. Farkas, J.B. Foresman, J.V. Ortiz, J. Cioslowski, D.J. Fox, Gaussian 09, revision b. 01, Gaussian. **2010**, 6492.
- [49] P.M. Gill, B.G. Johnson, J.A. Pople, M.J Frisch, *Chem. Phys. Lett.* **1992**, *197*, 499-505.
- [50] W.R. Wadt, P.J Hay, *J. Chem. Phys.* **1985**, *82*, 284-98.
- [51] C.A. Lipinski, F. Lombardo, B.W. Dominy, P.J. Feeney, *Adv. Drug. Deliv. Rev.* **2001**, *46*, 3-26.
- [52] G.M. Morris, R. Huey, W. Lindstrom, M.F. Sanner, R.K. Belew, D.S. Goodsell, A. J. Olson, *J. Comput. Chem.* **2009**, *16*, 2785-91.
- [53] J. Gasteiger, M. Marsili, *Tetrahedron*, **1980**, *36*, 3219-28.
- [54] G.M. Morris, D.D Goodsell, R.S. Halliday, R. Huey, W.E. Hart, R.K. Belew, A.J. Olson, *J. Comput. Chem.*, **1998**, *19*, 1639-62.
- [55] L. Yang, D.R. Powell, R.F. Houser, *Dalton Trans.* **2007**, 955-964.
- [56] A. Esparza-Ruiz, A. Peña-Hueso, I. Ramos-García, A. Flores-Parra, R. Contreras, *J. Organomet. Chem.* **2008**, *693*, 2739-2747.
- [57] A. Rodríguez, A. Sousa-Pedrares, J.A. García-Vázquez, J. Romero, A. Sousa, U. Russo, *Eur. J. Inorg. Chem.* **2007**, 1444-1456.
- [58] E. López-Torres, M.A. Mendiola, *Polyhedron* **2005**, *24*, 1435-1444.
- [59] E. López-Torres, U. Abram, *Inorg. Chem.* **2008**, *47*, 2890-2896.
- [60] A.W. Addison, T.N. Rao, J. Reedijk, J. van Rijn, G.C. Verschoor, *J. Chem. Soc. Dalton Trans.* **1984**, 1349-1356.
- [61] B. Wrackmeyer in *Fundamentals in tin chemistry, Tin chemistry: fundamentals, frontiers and applications*, A.G. Davies, M. Gielen, K.H. Pannel, E.R.T. Tiekink (Eds.), Wiley, Weinheim, **2008**, pp. 17-52.

- [62] H.C. Marsmann, F. Uhlig in *Further advances in germanium, tin and lead NMR, The chemistry of organic germanium, tin and lead compounds*, Z. Rappoport (Ed.), Wiley, Weinheim, **2002**, pp. 403-436.
- [63] T.P. Lockhart, W.F. Manders, *Inorg. Chem.* **1986**, 25, 892-895.
- [64] B.M. Chougala, S. Samundeeswari, M. Holiyachi, L.A. Shastri, S. Dodaman, S. Jalalpureb, S.R. Dixitd, S.D. Joshi, V.A. Sunag, *Eur. J. Med. Chem.* **2017**, 125, 101-116.

Table 1. Crystal data and structure refinement for **L¹H₂**, **[1EtOH]EtOH**, **4** and **6**.

| | L¹H₂.DMSO | [1EtOH]EtOH | 4 | 6 |
|--|--|---|---|--|
| Formula | C ₁₈ H ₂₃ N ₅ O ₃ S ₂ | SnC ₂₂ H ₃₃ N ₅ O ₄ S | SnC ₈ H ₁₅ N ₃ S | SnC ₂₃ H ₂₁ N ₃ S |
| M | 421.53 | 582.28 | 303.98 | 490.18 |
| Temperature/K | 130(2) | 150(2) | 296(2) | 296(2) |
| Crystal system | Monoclinic | Monoclinic | Monoclinic | Triclinic |
| Space group | <i>P2(1)/c</i> | <i>P2(1)/n</i> | <i>Cc</i> | <i>P</i> -1 |
| a/Å | 18.1657(5) | 7.436(3) | 13.7150(6) | 9.9826(2) |
| b/Å | 8.6439(2) | 20.806(13) | 10.2019(4) | 10.4816(2) |
| c/Å | 12.8464(3) | 16.848(13) | 10.0941(5) | 11.7310(2) |
| α/° | 90 | 90 | 90 | 89.4860(10) |
| β/° | 102.8400(10) | 99.99(7) | 118.082(2) | 65.6880(10) |
| γ/° | 90 | 90 | 90 | 80.6630(10) |
| U/ Å ³ | 1966.73(8) | 2567(3) | 1246.09(10) | 1101.50(4) |
| Z | 4 | 4 | 4 | 2 |
| D _c /Mgm ⁻³ | 1.424 | 1.507 | 1.620 | 1.478 |
| Absorption coefficient mm ⁻¹ | 0.301 | 8.983 | 2.184 | 1.267 |
| F(000) | 888 | 1192 | 600 | 492 |
| Goodness of fit on F ² | 1.127 | 0.880 | 1.180 | 1.217 |
| Reflections collected | 35855 | 17129 | 10175 | 18121 |
| Independent reflections | 4901 [R(int) = 0.0442] | 4526[R(int) = 0.3688] | 2529 [R(int) = 0.0417] | 4010[R(int) = 0.0359] |
| Final R1 and wR2[I>2σ(I)] | 0.0354, 0.0978 | 0.1365, 0.2516 | 0.0384, 0.0835 | 0.0290, 0.0744 |
| R indices (all data) | R1 = 0.0502 wR2 = 0.1162 | R1 = 0.2865, wR2 = 0.3471 | R1 = 0.0502, wR2 = 0.1211 | R1 = 0.0404, wR2 = 0.1084 |
| Residual electron density (min,max) (eÅ ⁻³) | -0.649, 0.702 | -1.392, 1.351 | -0.837, 1.075 | -0.773, 0.490 |

Table 2. Selected bond distances (Å) in L¹H₂ and complex [1EtOH]EtOH

| | L¹H₂ | [1EtOH]EtOH |
|-------------------|-----------------------------------|--------------------|
| C(1)-N(1) | 1.323(2) | 1.32(3) |
| C(1)-N(2) | 1.356(2) | 1.34(4) |
| C(1)-S(1) | 1.6947(16) | 1.73(2) |
| C(2)-N(3) | 1.292(2) | 1.35(3) |
| C(2)-C(3) | 1.477(2) | 1.47(3) |
| C(3)-N(4) | 1.292(2) | 1.26(3) |
| C(4)-O(1) | 1.2331(19) | 1.29(3) |
| C(4)-N(5) | 1.352(2) | 1.32(3) |
| C(14)-O(2) | 1.3713(19) | 1.41(3) |
| N(2)-N(3) | 1.3780(18) | 1.39(3) |
| N(4)-N(5) | 1.3730(19) | 1.34(3) |
| Sn-C(18) | - | 2.05(2) |
| Sn-C(17) | - | 2.07(2) |
| Sn-N(3) | - | 2.281(19) |
| Sn-N(4) | - | 2.34(2) |
| Sn-O(1) | - | 2.496(17) |
| Sn-O(3) | - | 2.534(17) |
| Sn-S(1) | - | 2.615(7) |

Table 3. Selected bond distances (Å) in complexes **4** and **6** and values for the τ_4 (**4**) and τ_5 (**6**) parameter.

| | 4 | 6 |
|------------------|-----------|------------|
| C(1)-N(1) | 1.350(15) | 1.343(6) |
| C(2)-N(1) | 1.309(17) | 1.319(6) |
| C(3)-N(2) | 1.318(11) | 1.328(6) |
| N(2)-N(3) | 1.346(14) | 1.346(5) |
| C(1)-N(3) | 1.323(15) | 1.322(5) |
| C(2)-C(3) | 1.415(17) | 1.403(7) |
| C(1)-S(1) | 1.756(12) | 1.748(5) |
| Sn(1)-C(6) | 2.141(14) | 2.129(4) |
| Sn(1)-C(7)/(12) | 2.124(13) | 2.140(4) |
| Sn(1)-C(8)/C(18) | 2.133(14) | 2.143(4) |
| Sn(1)-S(1) | 2.489(3) | 2.4464(12) |
| Sn(1)-N(3) | - | 2.856(4) |
| τ_4/τ_5 | 0.90 | 0.68 |

Table 4. Chemical shifts (ppm) observed in the ^{119}Sn NMR spectra of complexes **1-6** in DMSO- d_6 and in the solid state (CP/MAS).

| Compound | ^{119}Sn DMSO- d_6 | coordination number | ^{119}Sn CP/MAS | coordination number |
|--|--------------------------------------|---------------------|--------------------------|---------------------|
| [SnMe ₂ L ¹] (1) | -392.8 | 7 | -303.5 | 6 |
| [SnBu ₂ L ¹] (2) | -374.3 | 7 | -253.8 | 6 |
| [SnPh ₂ L ¹] (3) | -510.2 | 7 | -379.6 | 6 |
| [SnMe ₃ L ²] (4) | 0.5 | 4 | 48.9 | 4 |
| [SnBu ₃ L ²] (5) | 10.1 | 4 | - | - |
| [SnPh ₃ L ²] (6) | -166.3 | 5 | -91.9 | 5 |

Table 5. Antimicrobial activity, expressed as MIC ($\mu\text{g mL}^{-1}$) and, in brackets, as MBC ($\mu\text{g mL}^{-1}$) and MFC ($\mu\text{g mL}^{-1}$).

| Compound | Bacteria ^a | | | | Fungi ^b | | |
|--|-----------------------|---------------|---------------|---------------|--------------------|-------------|---------------|
| | BS | SA | EC | HI | CT | SC | AN |
| L¹H₂ | >200 | >200 | >200 | >200 | >200 | >200 | >200 |
| [SnMe₂L¹] (1) | 12 (25) | 100 (>200) | >200 | 100 (>200) | >200 | >200 | >200 |
| [SnBu₂L¹] (2) | 3 (6) | 25 (200) | >200 | 12 (100) | >200 | 25 (200) | 12 (200) |
| [SnPh₂L¹] (3) | 3 (25) | 6 (50) | >200 | 12 (100) | >200 | >200 | 50 (200) |
| HATs | 100 (200) | >200 | >200 | >200 | >200 | >200 | >200 |
| [SnMe₃L²] (4) | 6 (12) | 100 (>200) | 200 (>200) | 100 (>200) | >200 | >200 | 25 (200) |
| [SnBu₃L²] (5) | 0.3 (3) | 0.7 (6) | >200 | 0.7 (6) | 3 (12) | 1.5 (12) | 0.3 (3) |
| [SnPh₃L²] (6) | 0.3 (1.5) | 0.3 (1.5) | >200 | 0.7 (3) | 3 (6) | 1.5 (3) | 0.03 (0.7) |
| | | | | | | | |
| Ampicillin | 0.03 (0.3) | 0.15 (0.7) | 6 (12) | 0.07 (0.3) | - | - | - |
| Miconazole | - | - | - | - | 3 (25) | 12 (50) | 3 (25) |

^a Gram positive bacteria: *Bacillus subtilis* ATCC 6633 (BS) and *Staphylococcus aureus* ATCC 25923 (SA); Gram negative bacteria: *Escherichia coli* ATCC 8739 (EC) and *Haemophilus influenzae* ATCC 19418 (HI).

^b Yeasts: *Candida tropicalis* ATCC 1369 (CT) and *Saccharomyces cerevisiae* ATCC 9763 (SC); mould: *Aspergillus niger* ATCC 6275 (AN).

Table 6. Antimicrobial activity, expressed as MIC ($\mu\text{g mL}^{-1}$) and, in brackets, as MBC ($\mu\text{g mL}^{-1}$) and MFC ($\mu\text{g mL}^{-1}$), of complexes **[SnBu₃L²] 5** and **[SnPh₃L²] 6** against other microorganisms.

| Microorganisms | [SnBu ₃ L ²] (5) | [SnPh ₃ L ²] (6) | Ampicillin | Miconazole |
|---|---|---|-------------------|--------------|
| Gram positive bacteria | | | | |
| <i>Bacillus megaterium</i> BGSC 7A2 | 0.7 (1.5) | 0.15 (1.5) | 0.07 (0.15) | - |
| <i>Sarcina lutea</i> ATCC 9341 | 0.3 (3) | 0.15 (0.3) | 0.0015 (0.007) | - |
| <i>Staphylococcus epidermidis</i> ATCC 12228 | 1.5 (6) | 0.15 (0.7) | 3 (25) | - |
| <i>Streptococcus agalactiae</i> ^b | 0.7 (3) | 0.15 (0.3) | 0.03 (0.07) | - |
| Gram negative bacteria | | | | |
| <i>Proteus vulgaris</i> ATCC 13315 | 25 (100) | 3 (6) | 0.3 (0.7) | - |
| <i>Pseudomonas aeruginosa</i> ATCC 9027 | 6 (12) | 0.15 (3) | 100 (>200) | - |
| <i>Salmonella typhimurium</i> ATCC 14028 | 100 (>200) | >200 | 1.5 (3) | - |
| Yeasts | | | | |
| <i>Candida albicans</i> ATCC 10231 | 3 (50) | 0.7 (3) | - | 6 (25) |
| <i>Candida guilliermondii</i> ^b | 50 (100) | 12 (25) | - | 0.15 (3) |
| <i>Candida parapsilosis</i> ^b | 100 (200) | 12 (50) | - | 3 (12) |
| <i>Cryptococcus neoformans</i> ^b | 6 (50) | 0.3 (0.7) | - | 0.3 (0.7) |
| Moulds | | | | |
| <i>Aspergillus flavus</i> ^b | 6 | 3 | - | 50 |
| <i>Aspergillus fumigatus</i> ^b | 12 | 6 | - | 6 |
| <i>Epidermophyton floccosum</i> ^b | 0.3 | 0.3 | - | 0.015 |
| <i>Trichophyton interdigitalis</i> ^b | 3 | 1.5 | - | 1.5 |
| <i>Trichophyton mentagrophytes</i> ^b | 6 | 6 | - | 1.5 |
| <i>Trichophyton rubrum</i> ^b | 1.5 | 3 | - | 1.5 |
| <i>Trichophyton soudanense</i> ^b | 3 | 3 | - | 0.15 |

^b Clinical isolate

Table 7. Binding affinity and Lipinski's rule of complexes **1-6**.

| Complex | Binding affinity (kcal/mol) | Lipinski's filter | Cause of failing |
|-------------------|-----------------------------|-------------------|------------------|
| 1 | -6.7 | + | |
| 2 | -6.6 | - | Mol. wt. |
| 3 | -7.7 | - | Mol. wt. |
| 4 | -6.1 | - | Logp |
| 5 | -8.1 | - | Log p |
| 6 | -8.5 | - | Log p |
| Ampicillin | -5.5 | + | |

Table 8. Bond distances and interaction types of complexes **5**, **6** and ampicillin with *S. aureus* DHPS.

| Compound | Distance (Å) | Bond type | |
|---|--------------|---------------|----------------------------|
| [SnBu ₃ L ²] (5) | | | |
| A:GLN105:NE2 - P:C5:S1 | 3.31301 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:LYS203:NZ - P:C5:N1 | 3.22651 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:ARG239:NH1 - P:C5:N1 | 2.66416 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:ALA199 - P:C5:C6 | 2.45727 | Hydrophobic | Alkyl |
| A:ALA199 - P:C5:C12 | 3.92841 | Hydrophobic | Alkyl |
| P:C5:C5 - A:MET128 | 3.26018 | Hydrophobic | Alkyl |
| [SnPh ₃ L ²] (6) | | | |
| A:LYS203:NZ - P:C6:N1 | 3.34982 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:ARG239:NH1 - P:C6:N1 | 2.37229 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:ARG239:NH1 - P:C6 | 2.82399 | Electrostatic | π -Cation |
| A:ARG239:NH2 - P:C6 | 3.9122 | Electrostatic | π -Cation |
| A:ARG52:NE - P:C6 | 3.15855 | Hydrogen Bond | π -Donor Hydrogen Bond |
| A:ALA199 - P:C6:C6 | 3.12107 | Hydrophobic | Alkyl |
| P:C6:C5 - A:MET128 | 3.42281 | Hydrophobic | Alkyl |
| Ampicillin (AMP) | | | |
| A:ARG239:NH1 - P:AMP:O1 | 3.10326 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:ARG239:NH2 - P:AMP:O4 | 3.00411 | Hydrogen Bond | Conventional Hydrogen Bond |
| P:AMP:H12 - A:SER201:O | 1.6805 | Hydrogen Bond | Conventional Hydrogen Bond |
| P:AMP:H13 - A:GLN105:OE1 | 2.70167 | Hydrogen Bond | Conventional Hydrogen Bond |
| A:LYS203:CE - P:AMP:O1 | 3.61022 | Hydrogen Bond | Carbon Hydrogen Bond |
| A:HIS241:CE1 - P:AMP:O3 | 3.34736 | Hydrogen Bond | Carbon Hydrogen Bond |