Stereospecific ligands and their complexes. Part VIII. Antimicrobial activity of palladium(II) complexes with *O*,*O*'-dialkyl esters of (*S*,*S*)-ethylenediamine-*N*,*N*'-di-2-(4-methyl)-pentanoic acid*

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Abstract

Palladium(II) complexes (**1–4**) of general formula [$PdCl_2$(R_2-*S,S*-eddp)] with bidentate *N,N'*--ligands, *O,O'*-dialkyl esters (R = ethyl, *n*-propyl, *n*-butyl and *n*-pentyl), of (*S,S*)-ethylenediamine-*N,N'*-di-2-(4-methyl)pentanoic acid (*S,S*-eddp) were prepared and characterized by microanalysis, infrared and UV/Vis spectroscopy. The ligands and its complexes were tested for their *in vitro* antimicrobial activity against 15 species of bacteria and fungi. Testing is performed by the microdilution method, with the minimum inhibitory concentration (MIC) and the minimum microbicidal concentration (MMC) being determined. The MIC values were in range from 4.9 μg cm^{-3} to > 5000 μg cm^{-3} while MMC values ranged from 78 μg cm^{-3} to > 5000 μg cm^{-3}. Palladium(II) complexes [$PdCl_2$(**L*n***)] (***n***, **1–4**) have statistically significant higher activity than the corresponding ligands. Complex **4** displayed the strongest activity among all tested compounds.

Keywords: palladium(II) complexes, antimicrobial activity, electronic absorption spectra.

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In parallel with the rapid development of a wide range of antibacterial agents since the 1940s, bacteria have proven to be extremely adept at developing resistance to each new employed agent. The rapidly increasing incidence of bacterial resistance to antimicrobial agents has become a serious problem worldwide. Resistance mechanisms have been identified and described for all the known antibiotics currently available for clinical use [1].

Numerous complexes based on palladium(II) ion have been synthesized and their different biological activities have been documented [2–4]. The impact of different palladium complexes on the growth and metabolism of various groups of microorganisms has been studied. Garoufis *et al.* [5] reviewed numerous scientific papers on anti-viral, antibacterial and antifungal activity of palladium(II) complexes with different types of ligands (sulfur and nitrogen donor ligands, Schiff base ligands and drugs as ligands). There are other papers in the literature showing different intensity of palladium complexes activity on various species of bacteria and fungi [6–11].

In our earlier work we have reported on the synthesis, characterization and antimicrobial activity of palladium(II) complexes with some alkyl esters of (*S,S*)--ethylenediamine-*N*,*N*'-di-2-propanoic acid [12].

This study is focused on the *in vitro* antimicrobial activity of four R_2edda-type ligand precursors: *O*,*O*'-diethyl- (**L1**·2HCl) *O*,*O*'-dipropyl- (**L2**·2HCl) *O,O'*-dibutyl- (**L3**·2HCl) *O*,*O*'-dipentyl-(*S,S*)-ethylenediamine-*N*,*N*'-di-2-(4-methyl)-pentanoate dihydrochlorides (**L4**·2HCl) and their corresponding palladium(II) complexes: dichloro(*O*,*O*'-diethyl-(*S,S*)-ethylenediamine-*N*,*N*'-di-2-(4-methyl)-pentanoate)palladium(II), (**1**), dichloro(*O*,*O*'-dipropyl-(*S*,*S*)-ethylenediamine-*N*,*N*'-di-2-(4-methyl)pentanoate)palladium(II) (**2**), dichloro(*O*,*O*'-dibutyl-(*S*,*S*)--ethylenediamine-*N*,*N*'-di-2-(4-methyl)-pentanoate)palladium(II) (**3**), dichloro(*O*,*O*'-dipentyl-(*S*,*S*)-ethylenediamine-*N*,*N*'-di-2-(4-methyl)-pentanoate)palladium(II) (**4**).

EXPERIMENTAL

Chemistry

All the ligands **L1**·2HCl–**L4**·2HCl and corresponding palladium(II) complexes **1–4** were prepared using appropriate modifications of known methods [13–16].

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The other reagents were obtained commercially and used without further purification.

Infrared spectra were recorded by a Perkin-Elmer FTIR 31725-X and Perkin-Elmer Spectrum One FTIR spectrophotometer using the KBr pellet technique (4000–400 cm^{-1}). Electronic apsorption spectra were recorded on a Rayleigh UV – 9200 UV/Vis spectrometer using $CHCl_3$ solutions (1×10^{-2} mol dm^{-3}) of the complexes. Elemental microanalyses for C, H and N were performed by standard methods.

Preparation of *O,O'*-dialkyl esters of [(*S,S*)-H_4eddl]Cl_2, L1·2HCl–L4·2HCl

The esters were prepared using the esterification reaction previously described [16]. Thionyl chloride (4 cm^3, 55 mmol) was introduced into a flask containing 50 cm^3 of corresponding ice cooled alcohol (ethyl, *n*-propyl, *n*-butyl or *n*-pentyl; anhydrous conditions) for 1 hour. After addition of the 2 g (5.54 mmol) [(*S,S*)-H_4eddl]Cl_2 the reaction mixture was refluxed for 16 h, filtered off and the filtrate was left for a few days in a refrigerator at 4 °C. The esters were recrystallized from the hot alcohol used for each reaction.

L1·2HCl·H_2O. Yield, 1.12 g (48.78%). Anal. Calcd. for $C_{18}H_{38}N_2O_4Cl_2{\cdot}H_2O$: C, 49.65; H, 9.25; N, 6.43%. Found: C, 49.73; H, 8.79; N, 6.44%. IR (cm^{-1}): 3457, 2975, 2719, 2622, 2527, 2403, 1737, 1546, 1476, 1216, 1066, 1015, 803.

L2·2HCl·0.5H_2O. Yield, 0.98 g (40.90%). Anal. Calcd. for $C_{20}H_{42}N_2O_4Cl_2{\cdot}0.5H_2O$: C, 52.86; H, 9.54; N, 6.16%. Found: C, 52.74; H, 9.21; N, 6.36%. IR (cm^{-1}): 3453, 2966, 2721, 2621, 2528, 2407, 1735, 1547, 1472, 1210, 1065, 927, 802.

L3·2HCl·H_2O. Yield, 1.02 g (39.38%). Anal. Calcd. for $C_{22}H_{46}N_2O_4Cl_2{\cdot}H_2O$: C, 53.75; H, 9.84; N, 5.69%. Found: C, 53.29; H, 9.48; N, 5.78%. IR (cm^{-1}): 3455, 2963, 2716, 2619, 2526, 2405, 1736, 1546, 1470, 1209, 1062, 925, 803.

L4·2HCl·H_2O. Yield, 0.96 g (35.05%). Anal. Calcd. for $C_{24}H_{50}N_2O_4Cl_2{\cdot}H_2O$: C, 55.47; H, 10.08; N, 5.39%. Found: C, 55.32; H, 9.83; N, 5.14%. IR (cm^{-1}): 3452, 2960, 2726, 2623, 2525, 2401, 1736, 1543, 1470, 1211, 1066, 956, 798.

Preparation of the palladium(II) complexes, 1–4

Complexes were obtained by mixing $K_2[PdCl_4]$ (0.2 g, 0.61 mmol) and equimolar amount of the **L1**·2HCl·H_2O (0.267 g, 0.61 mmol), **L2**·2HCl·H_2O (0.277 g, 0.61 mmol), **L3**·2HCl·H_2O (0.301 g, 0.61 mmol) or **L4**·2HCl·H_2O (0.318 g, 0.61 mmol) esters. During the two hours of stirring, 10 cm^3 of water solution of LiOH (0.0293 g, 1.22 mmol) was added in small portions to the reaction mixture. Within this period, pale yellow precipitates of the complexes **1–4** were obtained, filtered off, washed with cold water, ethanol and ether and air dried.

1·0.5H_2O. Yield, 0.26 g (80.00%). Anal. Calcd. for $C_{18}H_{36}N_2O_4Cl_2Pd{\cdot}0{,}5H_2O$: C, 40.73; H, 7.02; N, 5.27%. Found: C, 40.83; H, 6.93; N, 5.44%. IR (cm^{-1}): 3088, 2960, 2872, 1739, 1468, 1370, 1239, 1194, 1143, 1024, 934, 849, 774.

2. Yield, 0.30 g (89.45%). Anal. Calcd. for $C_{20}H_{40}N_2O_4Cl_2Pd$: C, 43.68; H, 7.33; N, 5.09%. Found: C, 43.33; H, 7.12; N, 5.03%. IR (cm^{-1}): 3143, 2961, 2875, 1739, 1468, 1389, 1241, 1195, 1144, 1057, 946, 845, 775.

3. Yield, 0.33 g (93.19%). Anal. Calcd. for $C_{22}H_{44}N_2O_4Cl_2Pd$: C, 45.72; H, 7.67; N, 4.84%. Found: C, 45.49; H, 7.78; N, 4.77%. **IR** (cm^{-1}): 3133, 2960, 2872, 1740, 1466, 1369, 1241, 1197, 1143, 1089, 938, 844, 736.

4·2.5H_2O. Yield, 0.29 g (72.72%). Anal. Calcd. for $C_{24}H_{48}N_2O_4Cl_2Pd{\cdot}2.5H_2O$: C, 44.27; H, 8.20; N, 4.30%. Found: C, 43.79; H, 8.01; N, 4.57%. **IR** (cm^{-1}): 3139, 2959, 2870, 1741, 1466, 1369, 1229, 1197, 1129, 1070, 971, 730.

In vitro antimicrobial assay

Test substances

The tested compounds were dissolved in DMSO and then diluted into nutrient liquid medium to achieve a concentration of 10% DMSO. An antibiotic, doxycycline (Galenika A.D., Belgrade) was dissolved in nutrient liquid medium, a Mueller–Hinton broth (Torlak, Belgrade, Serbia), while an antimycotic, fluconazole (Pfizer Inc., USA) was dissolved in Sabouraud dextrose broth (Torlak).

Test microorganisms

Antimicrobial activity of four ligands and corresponding palladium(II) complexes was tested against 15 microorganisms including five strains of pathogenic bacteria (standard and clinical strains): *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 29212, *Pseudomonas aeruginosa* ATCC 27853, *Proteus mirabilis* (clinical isolate) and *Escherichia coli* (clinical isolate); six species of pathogenic fungi: moulds *Aspergillus fumigatus* PMFKG-F23, *Aspergillus flavus* PMFKG-F24, *Aspergillus restrictus* PMFKG-25, *Aspergillus niger* PMFKG-26 and *Aspergillus niger* ATCC 16404 and one yeast species *Candida albicans* (clinical isolate); four species of probiotics: *Lactobacillus plantarum* PMFKG-P31, *Bacillus subtilis* IP 5832 PMFKG-P32, *Bifidobacterium animalis* subsp. *lactis* PMFKG-P33 and *Saccharomyces boulardii* PMFKG-P34. All clinical isolates were a generous gift from the Institute of Public Health, Kragujevac, Serbia. The other microorganisms were provided from a collection held by the Microbiology Laboratory, Faculty of Science, University of Kragujevac, Serbia.

Suspension preparation

Bacterial suspensions and yeast suspension were prepared by the direct colony method. The colonies were taken directly from the plate and were suspended in 5 cm^3 of sterile 0.85% saline. The turbidity of initial suspension was adjusted by comparing with 0.5 McFarland's standard (0.5 cm^3 1.17% w/v $BaCl_2 \cdot 2H_2O$ + 99.5 cm^3 1% w/v H_2SO_4) [17]. When adjusted to the turbidity of the 0.5 McFarland's standard, bacteria suspension contains about 10^8 colony forming units (CFU) cm^{-3} and suspension of yeast contains 10^6 CFU cm^{-3}. 1:100 dilutions of initial suspension were additionally prepared into sterile 0.85% saline. The suspensions of fungal spores were prepared by gentle stripping of spore from slopes with growing aspergilli. The resulting suspensions were 1:1000 diluted in sterile 0.85% saline.

Microdilution method

Antimicrobial activity was tested by determining the minimum inhibitory concentration (MIC) and minimum microbicidal concentration (MMC) using microdilution method with resazurin [18]. The 96-well plates were prepared by dispensing 100 µL of nutrient broth, Mueller–Hinton broth for bacteria and Sabouraud dextrose broth for fungi and yeasts, into each well. A 100 µL aliquot from the stock solution of tested compound (concentration of 10000 µg/cm^3) was added into the first row of the plate. Then, twofold, serial dilutions were performed by using a multichannel pipette. The obtained concentration range was from 5000 to 2.44 µg cm^{-3}. 10 µL of diluted bacterial, yeast suspension and suspension of spores was added to each well to give a final concentration of 5×10^5 CFU cm^{-3} for bacteria and 5×10^3 CFU cm^{-3} for fungi and yeast. Finally, 10 µL resazurin solution was added to each well inoculated with bacteria and yeast. Resazurin is an oxidation–reduction indicator used for the evaluation of microbial growth. It is a blue non-fluorescent dye that becomes pink and fluorescent when reduced to resorufin by oxidoreductases within viable cells. The inoculated plates were incubated at 37 °C for 24 h for bacteria, 28 °C for 48 h for the yeast and 28 °C for 72 h for fungi. MIC was defined as the lowest concentration of tested substance that prevented resazurin color change from blue to pink. For fungi, MIC values of the tested substance were determined as the lowest concentration that visibly inhibited mycelia growth.

Doxycycline and fluconazole were used as a positive control. Solvent control test was performed to study the effect of 10% DMSO on the growth of microorganism. It was observed that 10% DMSO did not inhibit the growth of microorganism. Also, in the experiment, the concentration of DMSO was additionally decreased because of the twofold serial dilution assay (the working concentration was 5% and lower). Each test included growth control and sterility control. All tests were performed in duplicate and MICs were constant.

Minimum bactericidal and fungicidal concentration was determined by plating 10 µL of samples from wells, where no indicator color change was recorded, on nutrient agar medium. At the end of the incubation period the lowest concentration with no growth (no colony) was defined as minimum microbicidal concentration.

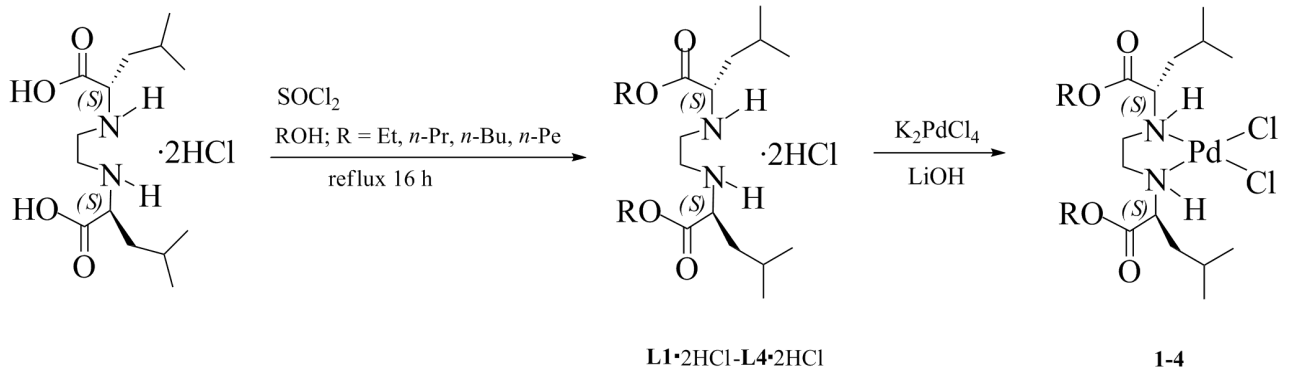
Statistical analysis

All statistical analyses were performed using SPSS package. The results for continued variables were presented as mean ± SD. The Student paired t-test was used to compare concentrations between ligands and palladium(II) complexes. Differences between groups (compounds, microorganisms) were analyzed using 1-factor ANOVA. In all cases P values < 0.05 were considered statistically significant.

RESULTS AND DISCUSSION

Chemistry

All the esters (**L1**·2HCl–**L4**·2HCl) of (*S*,*S*)-ethylenediamine-*N*,*N*'-di-2-(4-methyl) pentanoic acid and corresponding palladium(II) complexes (**1–4**) were prepared using appropriate modifications of known methods [13–16] (Scheme 1). These esters are not soluble



L1·2HCl-**L4**·2HCl **1-4**

*Scheme 1. The synthesis of the esters **L1**·2HCl–**L4**·2HCl and corresponding palladium(II) complexes **1–4**.*

in chloroform, but rather in water, methanol, dimethyl sulfoxide and hot alcohols used in individual esterification reactions. The complexes are soluble in chloroform and dimethyl sulfoxide, but not in water.

Infrared spectra

IR spectra of the ligands and corresponding complexes are similar to those of the previously reported compounds [19,20]. Absorption bands for secondary amino groups were found for **1–4** at 3088, 3143, 3133 and 3139 cm^{-1}, respectively, indicating coordination *via* nitrogen atoms. The same bands infrared spectra of the ligands can be observed; **L1**·2HCl–**L4**·2HCl:ν($R_2NH_2^+$) at 3457, 3453, 3455 and 3452 cm^{-1}. All other specific bands were found at similar positions for the ligands and corresponding complexes, thus indicating that no other atom was coordinated to the palladium center (see Experimental part).

Electronic absorption spectra

The electronic spectra of palladium(II) complexes **1–4** are very similar to each other and their absorption maxima are summarized in Table 1.

Complex	λ / nm	ε / $M^{-1}cm^{-1}$	Reference
1	393	161.1	This work
2	399	156.1	
3	399	143.9	
4	399	146.2	
$[PdCl_4]^{2-}$	474	161	[21]
$[Pd(H2O)_4]^{2+}$	378	83	[21]

In all complexes (**1–4**) palladium(II) ions have a square-planar geometry. For palladium(II) complexes with D_{4h} molecular geometry, spin-forbidden d-d transitions ($^1A_{1g} \rightarrow {}^1A_{2g}$) are expected in the region 350–500 nm [21,22]. The solutions of all complexes were very similar in color, yellow-orange. The almost equal position (Table 1) of the absorption bands in UV/Vis spectra of the complexes **1–4** confirmed square-planar environment of the palladium(II) ion. The shift of the Pd(II) d-d transition band from 474 nm for $[PdCl_4]^{2-}$ complex [21] to ~ 400 nm for complexes **1–4** can be explained as the exchange of two Cl^- ligands with **L1–L4** ligands, The decreasing of the molar absorption coefficients (Table 1) with increasing of the number of C atoms in the alkyl group is expected, and can be explained with decreasing of the ligand field strength around palladium(II) ion.

***In vitro* antimicrobial studies**

The results of *in vitro* testing of antibacterial and antifungal activities of the four palladium(II) complexes and their ligands are shown in Tables 2 and 3. For comparison, MIC and MMC values of doxycycline and fluconazole are also listed in Table 2 and 3.

The tested palladium(II) complexes and ligands showed different degrees of antimicrobial activity in relation to the tested species of microorganisms. The intensity of antimicrobial action varied depending on the group of microorganisms and on the type of the compounds.

Antimicrobial activity of tested compounds was evaluated by determining MICs and MMCs in relation to the 15 species of microorganisms. MIC values were in range from 4.9 to > 5000 μg cm^{-3} while MMC values were from 78 to > 5000 μg cm^{-3}. Generally, the palladium(II) complexes were more active than their ligands ($p < 0.0005$ for MIC and MMC). By comparing the activity of ligand and corresponding complex, it was remarked that complexes **1** and **4** were statistically more active than their ligands **L1** and **L4** ($p = 0.004$ for MIC and $p = 0.005$ for MMC; $p = 0.001$ for MIC and $p < 0.0005$ for MMC, respectively). If we observe the ligands between themselves there was no statistically significant difference. On the contrary, there was statistically significant difference between palladium(II) complexes: complexes **1** and **4** are more active than complexes **2** and **3**. The complex **4** displayed the strongest activity among the all tested compounds ($p < 0.05$). MICs for complex **4** were from 39.1 to 1250 μg cm^{-3} and MMCs from 39.1 to > 2500 μg cm^{-3}.

Statistically significant difference on the level of MMCs at **L1** and **L4** related to complexes **1** and **4** is

Table 2. Antibacterial activity of tested ligands and corresponding palladium(II) complexes (μg cm^{-3})

Species	**L1**·2HCl		**1**		**L2**·2HCl		**2**		**L3**·2HCl		**3**		**L4**·2HCl		**4**		Doxycycline	
	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC
E. coli ATCC 25922	2500	2500	1250	1250	2500	5000	2500	2500	1250	2500	1250	2500	5000	5000	312.5	312.5	25	25
E. faecalis ATCC 29212	1250	1250	1250	1250	2500	>5000	625	>5000	156.3	>625	625	>5000	1250	5000	156.3	1250	50	50
P. aeruginosa ATCC 27853	5000	5000	1250	1250	2500	5000	1250	2500	2500	2500	2500	5000	5000	>5000	312.5	1250	12.5	50
P. mirabilis	5000	5000	1250	1250	>5000	>5000	2500	>5000	>5000	>5000	2500	>5000	5000	5000	1250	2500	50	>50
E. coli	625	>5000	625	1250	312.5	>2500	1250	>5000	1250	>5000	2500	>5000	625	>5000	312.5	>2500	12.5	12.5
B. animalis subsp. lactis	78	> 625	78	>625	>5000	>5000	1250	2500	5000	5000	2500	5000	5000	>5000	625	2500	0.024	>0.19
L. plantarum	9.8	78	4.9	>39.1	78	>625	78	2500	2500	>5000	78	5000	1250	>5000	312.5	>2500	0.012	>0.098
B. subtilis IP 5832	78	> 625	39.1	>312.5	625	>5000	78	>5000	2500	>5000	78	>5000	1250	>5000	312.5	>2500	0.003	0.024

Table 3. Antifungal activity of tested ligands and corresponding palladium(II) complexes (μg cm^{-3})

Species	**L1**·2HCl		**1**		**L2**·2HCl		**2**		**L3**·2HCl		**3**		**L4**·2HCl		**4**		Fluconazole	
	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC	MIC	MMC
S. boulardii	5000	5000	1250	1250	625	625	625	625	312.5	1250	625	625	625	1250	312.5	312.5	6.25	> 50
C. albicans	5000	>5000	2500	2500	1250	2500	1250	1250	1250	1250	625	1250	2500	2500	625	625	3.125	> 50
A. fumigatus	1250	>5000	2500	>5000	156.3	625	312.5	625	39.1	78	39.1	78	39.1	78	39.1	39.1	>500	>500
A. flavus	>5000	>5000	625	1250	312.5	2500	312.5	625	1250	1250	156.3	312.5	2500	2500	312.5	312.5	>500	>500
A. restrictus	>5000	>5000	312.5	625	156.3	625	156.3	312.5	1250	1250	312.5	625	1250	2500	39.1	39.1	>500	>500
A. niger	>5000	>5000	1250	>5000	625	1250	312.5	625	625	1250	312.5	312.5	1250	2500	312.5	625	>500	>500
A. niger ATCC 16404	>5000	>5000	1250	>5000	1250	2500	625	625	156.3	156.3	156.3	156.3	312.5	312.5	156.3	312.5	>500	>500

confirmed with analysing the difference of activity between ligands and corresponding complexes.

Due to the obtained results, the higher activity of the complexes, as compared to the ligands, can be understood in terms of chelation theory. This theory explains that a decrease in the polarizability of the palladium(II) ion could enhance the lipophilicity of the complexes [23].

According to different groups of microorganisms, the palladium(II) complexes demonstrated more potent inhibitory effects than ligands on the growth of pathogenic bacteria, probiotics and moulds ($p < 0.05$). Above stated, matches the results notified by Vasic *et al.* [12].

Being compared to the positive control, the palladium(II) complexes showed moderate to low antibacterial activity against tested standard and clinical strains of bacteria. The most sensitive was Gram-positive bacterium *Enterobacter faecalis* ATCC 29212 with MIC at 156.3 μg cm^{-3} for **L3** and **L4**. This finding is in accordance with the statement that Gram-positive bacteria were more sensitive to antimicrobial agents than Gram-negative bacteria. The main reason for that is chemical composition of their cell wall and presence of outer membrane in Gram-negative bacteria which is an effective barrier [24]. *Proteus mirabilis* was the most resistant. Tested compounds showed no effects or affected the growth of this bacterium at higher concentrations.

The probiotics *Bifidobacterium animalis subsp. lactis*, *Lactobacillus plantarum* and *Bacillus subtilis* IP 5832 showed the highest sensitivity to the tested compounds. The best activity demonstrated **1**–**3** with MIC values between 4.9 to 78 μg cm^{-3}. The most sensitive was *Lactobacillus plantarum* (MIC was 4.9 μg cm^{-3}). Antimicrobial testing of newly synthesized complexes of palladium(II) on probiotics, done by Vasic *et al.*, led to similar results [12].

Activity of complexes **2**–**4** against tested moulds was statistically better than ligands and positive control fluconazole ($p < 0.05$). The obtained concentrations of palladium(II) complexes which inhibit the growth of moulds were from 39.1 to 625 μg cm^{-3}. The most sensitive was *Aspergillus fumigatus*. Although the activity of complexes against moulds is more significant than the positive control, fluconazole, the results in the paper Vasic *et al*. [12] on the same microorganisms are significantly better and go to the value of 0.49 μg cm^{-3}.

Compared to the moulds, yeast *Saccharomices boulardii* and *Candida albicans* did not indicate significant sensitivity to the tested compounds.

CONCLUSIONS

In this paper we report characterization of palladium(II) complexes **1**–**4** by microanalysis, infrared and UV/Vis spectroscopy and their antimicrobial activity. The results of antimicrobial activity showed that palladium(II) complexes have significantly higher activity than corresponding ligands. The antimicrobial activities varied depending on the group of microorganisms and the type of compounds. Tested ligands, with exceptions **L1** and **L2** on probiotics and **L3** and **L4** on *Aspergillus fumugatus*, showed very low antimicrobial activity. All palladium(II) complexes showed moderate antibacterial activity. The results obtained for *Aspergillus* species which are common in enviroment and which cause infection known as aspergillosis showed that complexes **2**–**4** reacted better than fluconazole.

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IZVOD

STEREOSPECIFIČNI LIGANDI I NJIHOVI KOMPLEKSI. VIII DEO. ANTIMIKROBNA AKTIVNOST PALADIJUM(II)-KOMPLEKSA SA *O*,*O*'-DIALKIL ESTRIMA (*S*,*S*)-ETILENDIAMIN-*N*,*N*'-DI-2-(4-METIL)-PENTANSKE KISELINE

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Sintetisani su paladijum(II)-kompleksi (**1**–**4**) sa bidentatnim *N*,*N*'-ligandima, *O*,*O*'-dialkil estrima (alkil = etil, *n*-propil, *n*-butil i *n*-pentil), od (*S*,*S*)-etilendiamin-*N*,*N*'-di-2-(4-metil)-pentanske kiseline i ispitani su na osnovu elementarne analize, infracrvene i UV/Vis spektroskopije. Ligandi i njihovi kompleksi su mikrobiološki testirani *in vitro* na 15 vrsta bakterija i gljiva. Testirani su mikrodilucionom metodom i određena je minimalna inhibitorna koncentracija (MIC) i minimalna mikrobicidalna koncentracija (MMC). MIC vrednosti su iznosile od 4,9 do > 5000 μg cm^{-3} dok su vrednosti za MMC iznosile od 78 do > 5000 μg cm^{-3}. Paladijum(II)-kompleksi [$PdCl_2$(**L*n***)] (***n***, **1**–**4**) pokazali su značajno veću aktivnost u odnosu na ligande. Od testiranih jedinjenja kompleks **4** pokazuje najveću aktivnost. Od mikroorganizama najveću otpornost prema testiranim supstancama pokazuju patogene bakterije, a najosetljivije su probiotske bakterije. Testirane gljive prema kompleksima **2**–**4** pokazuju veću osetljivost nego prema pozitivnoj kontroli, flukonazolu.

Ključne reči: Paladijum(II)-kompleksi • Antimikrobna aktivnost • Elektronski apsorpcioni spektri