

Chemistry and Materials Research ISSN 2224-3224 (Print) ISSN 2225-0956 (Online) Vol.3 No.1, 2013



X-ray crystallographic and structural studies of (benzothiazol-2-yl)ethanesulphonamide, and its antimicrobial properties

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Abstract

N-(benzothiazol-2-vl)ethanesulphonamide (ES2ABT) was synthesized by the condensation of 2-aminobenzothiazole and ethanesulphonylchloride in acetone at 130 °C. The resulting crude precipitates were recrystallized in absolute ethanol. ES2ABT was characterized using X-ray crystallography, mass spectrometry, elemental microanalysis, UV/VIS spectrophotometry, infra red, proton and ¹³C NMR spectroscopies. The antimicrobial tests of the compound were carried out on both multi-resistant bacterial strains isolated under clinical conditions and cultured species using agar-well diffusion method. The multi-resistant bacterial strains used were Escherichia coli, Proteus species, Pseudomonas aeroginosa and Staphylococcus aureus which were isolated from dogs. The culture species were Pseudomonas aeruginosa (ATCC 27853), Escherichia Coli (ATCC 25922) Staphylococcus aureus (ATCC 25923), and the fungi, Candida krusei (ATCC 6258) and Candida albicans (ATCC 90028). The tests were both in vitro and in vivo. Thus the Inhibition Zone Diameter (IZD), the Minimum Inhibitory Concentration (MIC), and the Lethal and Effective Concentrations (LC50 and EC50) were determined. The antimicrobial activity of the compound was compared with those of Ciprofloxacin and trimethoprim-sulphamethoxazole as antibacterial agents and Fluconazole as an antifungal drug. The compound showed varying activity against the cultured typed bacteria and fungi used. However, ES2ABT was less active than the antibacterial standard drugs used but not Fluconazole which did not show any activity against Candida krusei (ATCC 6258). The Lethal Concentration (LC₅₀) is 338.80 ± 28.6 ppm. This is within the permissible concentrations.

Key words: N-(benzothiazol-2-yl)ethanesulphonamide, antimicrobial, in vivo, in vitro

1. INTRODUCTION

The search for potent anti-infective agents occupies an important position in science more so now with the upsurge in disease diversity and declining sensitivity of the implicated organisms to available agents^(1, 2). Interest in the coordination chemistry of thiazole and its derivatives with metal ions has risen due to the important role they play in biological systems⁽³⁾. Thiazoles are known to posses antitubercular, hypotensive and hypothermic activities^(4, 5). Studies have shown that the metal complexes of sulfa drugs promote rapid healing of skin disorder, for instance, silver(I)sulfadiazine and zinc(II)sulfadiazine are used to treat burns in humans and animals respectively⁽⁶⁾.

Mercury(II), and copper(II) complexes of 6-methyl-2-aminobenzothiazole have been shown to have a high activity against *Aspergillus niger, Alternaria alternate, Curvularia plunata* and *Penicillium fumculorus*⁽⁷⁾. Obasi *et al* have previously studied some sulfonyl derivatives of 2-aminothiazole, and the results obtained showed that the compounds were significantly active against *Staphylococcus aureaus* and *Escherichia colt*⁽⁸⁾. Some novel N-(Benzothiazol-2-yl)ethanamides were also synthesized and characterized by Obasi *et al*, which were screened *in vitro* and *in vivo* for antibacterial activity. The compounds were very stable and showed high antibacterial activities against both gram-positive and gram-negative bacteria⁽⁹⁾. The present work is aimed at synthesizing and characterizing new derivative of 2-aminobenzothiazole, and investigating how the structural difference affects their antimicrobial activities when compared with conventional sulfonamides.



2. EXPERIMENTAL

2.1 Reagents and apparatus

The compound, N-(benzothiazol-2-yl)ethanesulphonamide (ES2ABT) was prepared based on our modified method from that by Sprague *et al*⁽¹⁰⁾. All reagents were of analytical grade and were used as supplied except otherwise stated. A UV-Visible spectrum (200 - 800 nm in DMSO) of the compound was obtained on UV-2550 UV-VIS Spectrophotometer, (SHIMADZU). FTIR spectra of the compound were run as Nujol mulls on FTIR-84005 FTIR Spectrophotometer, (SHIMADZU). ¹³C and ¹H NMR spectra were recorded on Bruker-BioSpin 500 MHz NMR Spectrometer (UK) using DMSO and CDCl₃ as solvents respectively. The proton NMR peaks were observed at 500 MHz whereas the carbon-13 spectra were observed at 125 MHz. Elemental microanalysis was carried out using LECO-CHNS 932 microanalysis apparatus, the mass spectrometric analysis was carried out using a Thermo Finnigan LCQ DUO instrument using an electrospray ion trap method. X-ray crystallography was carried out at 123 K (-150 °C) on a Nonius KappaCCD single-crystal diffractometer, using graphite monochromated Mo-Kα radiation, all at the Department of Pure and Applied Chemistry, University of Strathclyde, Scotland, UK.

2.2 Synthesis of N-(benzothiazol-2-yl)ethanesulphonamide(ES2ABT)

To a solution of 2-aminobenzothiazole (3.0 g; 20 mmole) in acetone (15 mL) was added ethanesulphonylchloride (5 mL; 20 mmole) with stirring. The mixture was refluxed for 30 min at 130 °C. A milkish white precipitate was formed on refluxing which was collected and recrystallised from absolute ethanol. The yield was 80.2%. The melting point is 170-172 °C.

N-(benzothiazol-2-yl)ethanebenzenesulfonamide

Scheme 1: Synthesis of N-(benzothiazol-2-yl)ethanesulphonamide(ES2ABT)

2.3 X-Ray analysis

Crystals obtained were collected, coated in mineral oil and mounted on glass fibres. Data were collected at 123 K on a Nonius Kappa CCD diffractometer using graphite monochromated Mo-K α radiation. The heavy atom positions were determined by Patterson methods and the remaining atoms located in the difference electron density maps. Data were solved using Shelx 97 program⁽¹¹⁾ and SIR 92 program⁽¹²⁾ using the graphical interface Wingx⁽¹³⁾. All non-hydrogen atoms are anisotropic. The hydrogen atoms are placed as a mixture of independent and constrained refinement in the calculated positions around the parent atoms. A summary of the crystallographic parameters are given in Table 1.

2.4 Antimicrobial properties

2.4.1 In vitro Tests

Multi-resistant bacterial strains isolated under clinical conditions and Typed strains (ATCC Cultures) were used in the study. The bacterial strains used were Escherichia coli strains (E. Coli Strain 1 and E. Coli Strain 15), Proteus species strains (Proteus spp strains 25, Proteus spp strains 26), Pseudomonas aeroginosa strains 34 and multi-resistant Staphylococcus aureus (SR) strain. The bacteria Typed strains (ATCC Cultures) used were Pseudomonas aeruginosa (ATCC 27853), Escherichia Coli (ATCC 25922), Staphylococcus aureus (ATCC 25923). Fungi Typed strains (ATCC Cultures) used were Candida krusei (ATCC 6258) and Candida albicans (ATCC 90028). The Typed



strains were obtained from Bioresources Development and Conservation Program (BDCP), International Centre for Ethnomedicine and Drug Development (IntaceEED), Nsukka, Nigeria.

The antibacterial and antifungal activities of the compound, ES2ABT against these multi-resistant bacteria were determined using the agar well diffusion method as described by Chah et al⁽¹⁴⁾. Mueller-Hinton agar plates were inoculated with 0.1 mL of 3 h broth culture of the test bacteria. Using a cork borer, wells (7 mm in diameter and 2.5 mm deep) were bored into the inoculated agar. The test compound was solubilized in 20% v/v dimethyl sulfoxide (DMSO) and 0.05 mL of the compound at a concentration of 20 mg/mL was delivered into the wells. One of the wells contained 20% v/v DMSO and served as control. The plates for antibacterial screening were incubated at 37 °C for 24 h while the fungi were incubated at 30 °C for 48 h and assessment of activity was based on the measurement of the diameter of inhibition zone (IZD) around the wells. The test was performed in triplicates, mean IZD was recorded to the nearest whole millimetre.

The minimum inhibitory concentration (MIC) of the test compound was determined using the agar dilution method as described by Ojo et al⁽¹⁵⁾. Two-fold serial dilutions of test compound were made in 20% v/v DMSO. One millilitre of each serial dilution was added to 19 mL of sterile Mueller-Hinton agar maintained at 45 °C, thoroughly mixed and poured into a sterile plate and the medium allowed to solidify. The final concentrations of the compound ranged from 20 mg/mL to 1.25 mg/mL. Amended media were incubated overnight at 37 °C to check for sterility. Overnight nutrient broth cultures of the test bacteria were adjusted to contain approximately 10⁸ cfu/mL and 0.025 mL of each of the test organisms was spot-inoculated on the amended culture media. Inoculated plates were incubated at 37°C for 24 h and observed for presence of visible growth. The minimum inhibition concentration was determined as the value of the lowest concentration that completely suppressed growth of the organisms.

2.4.2 In vivo Tests [Brine Shrimps Lethality Test (BSLT)]

The method of McLaughlin and coworkers was used to study the bioactivity of the synthesized compound⁽¹⁶⁾. Artemia salina eggs obtained from a pet shop in Davis California were incubated in natural sea water (from Bar Beach, Lagos, Nigeria) in a dam-well under room condition. About ten (10) 48 h- shrimp nauplii in 1mL of autoclaved sea water were put into Bijou bottles using a Pasteur pipette under a stereo-microscope with a light source. They were separated into 7 groups in triplicate. Increasing concentrations (10, 100, 1000 ppm) of the synthesized compound were added into each of the triplicate and distilled water was added into the control group. The nauplii were incubated at room temperature (37 °C) for 24 h after which the survivors in each well were counted. The results were analysed using Finney Probit Analysis (MS-DOS-Computer-Program) to determine the LC₅₀ at 95% confidence interval. Weak nauplii were noted as an indication of central nervous system depression.

3 RESULTS AND DISCUSSION

The equation of reaction for the syntheses of the compound, ES2ABT is represented in Scheme 1.

3.1 X-ray anlaysis of the ES2ABT

ES2ABT was structurally characterized using X-ray methods (Figure 1). The gross structural features of the compound are in good agreement with that of the other uncoordinated sulphonamides reported thus far (Table 1)⁽¹⁷⁻¹⁹⁾. Typical for these species the thioimidazole ring lies at an angle to the sulphonyl group. Remarkably there are only a few simple sulphonamide salts known in the crystallographic database. Indeed sulfonamides with ethyl substituent on the sulfur are not well established. The metrical parameters for the four representatives of the sulphonamides and their salts are shown in Table 1.

Figure 2 shows the skeletal framework for the sulphonamide with the numbering system for the sulphonamide employed in Table 2. Two forms have been reported, namely neutral compounds where N1 is protonated and salts where N1 is deprotonated. R is typically a substituted aryl ring apart from this study where it is an ethyl group.

Of the four sulphonamide complexes in the crystallographic data base, two are neutral (pTos and bz-OMe) and two are anionic (Naphthyl and pTol, Table 2). In all cases previously reported these species contain electron withdrawing group attached to sulphur. Thus it can be seen that as a result of deprotonating N1 the C1-N1 bond length increases and the N1-S1 bond decreases. Despite the electron donating properties of the ethyl substitutent, the metrical parameters for the compound reported here (Figure 1) are in good agreement with the data reported for the neutral species reported thus far.



4 Physical Properties and Elemental microanalysis of the compound

Table 3 shows some physical properties of the compound. The melting point of the compound, ES2ABT is 170-172 °C and it is milky coloured. The compound, ES2ABT is amorphous. The result of the elemental microanalysis of the compound is recorded in Table 3. The amount of carbon, hydrogen and nitrogen calculated theoretically correspond to a reasonable extent with the experimental result.

The mass spectral result of the compound was in agreement with that expected.

5 Electronic Spectra

The electronic spectrum of the compound is shown in Table 4. Two bands were observed for the compound at 209.8 nm and 264.0 nm. They are due to $\pi \rightarrow \pi^*$ and $n \rightarrow \pi^*$ transitions.

6 IR Spectra of the ES2ABT

The IR spectrum of the compound is shown in Table 4. The broad peak at 3484 cm⁻¹ was assigned N-H stretching vibration for the compound, ES2ABT. The strong peaks at 2975 cm⁻¹, 2870 cm⁻¹, 2758 cm⁻¹ were assigned to C-H stretching vibration. The strong peaks at 1650 cm⁻¹, 1625 cm⁻¹ in the compound were assigned to C=C stretching vibration of aromatic ring. Two strong peaks in the compound at 1555 cm⁻¹, 1478 cm⁻¹ are assigned to C=N stretching vibration of benzothiazole ring. A strong peak was observed at 1304 cm⁻¹ in the compound, ES2ABT. This peak was assigned SO₂ stretching vibration. Weak peak at 836 cm⁻¹ was assigned to C-H bending vibration of substituted benzene ring. The strong peak at 640 cm⁻¹ was assigned C-S-C stretching vibration of the thiazole ring.

7 ¹H and ¹³C NMR Spectral Data

The 1 H and 13 C NMR Spectral Data of the compound is shown in Table 4. The peak at 9.99 ppm (1H, s) is assigned to N-H proton. The peak at 7.89 ppm (4H, m) is assigned to benzothiazole protons. The peak at 1.20 ppm (2H, q) is assigned to $-\text{CH}_2$ (methylene) protons. The peak at 1.11 ppm (3H, t) is assigned to $-\text{CH}_3$ (methyl) protons.

The peaks to a large extent are in agreement with our expectations. Peak at 169.6 ppm is assigned benzothiazole ring carbon (C7). Peak at 128.1 ppm is assigned benzothiazole ring carbon (C5). Peak at 139.6 ppm is assigned benzothiazole ring carbon (C6). Peak at 124.8 ppm is assigned benzothiazole ring carbon (C1). Peak at 123.7 ppm is assigned benzothiazole ring carbon (C2) while peak at 123.2 ppm is assigned benzothiazole ring carbon (C3). Peak at 124.1 ppm is assigned benzothiazole ring carbon (C4). However peak at 48.2 ppm is assigned methelene carbon (C8), and the peak at 46.0 ppm is assigned methyl carbon (C9).

8 Antimicrobial activity of the compound

The antimicrobial activities of the compound are recorded in Tables 5 and 6.

Table 5 showed the activities against multi-resistant bacterial strains isolated under clinical conditions. The Inhibitory Zone Diameter (IZD) in mm and Minimum Inhibitory Concentration (MIC) in mg/mL of the compounds were determined. Two strains each of E. Coli (E.Coli strain 1 and E. Coli strain 15), and Proteus species (Proteus spp strains 25 and Proteus spp strains 26), Pseudomonas aeroginosa strains 34 and multi-resistant Staphylococcus aureus (SR) strain, all isolated from dogs at clinical conditions were used. Ciprofloxacin and trimethoprim-sulphamethoxazole were used as the standard drugs. We determined the MIC on the concentration range 0.125-10 mg/mL. We discarded concentrations above 10 mg/mL. Based on this, the compound showed activity against two of the tested multi-resistant bacterial strains- E. Coli Strain 15 and Pseudomonas aeroginosa strains 34 with MIC of 10 mg/mL and IZD of 11 mm, and MIC of 10 mg/mL and IZD of 10 mm respectively.

Table 6 showed activities of the compound against Typed Strains (ATCC Cultures) microorganisms. The bacteria cultures used are Pseudomonas aeruginosa (ATCC 27853), Escherica coli (ATCC 25922) and Staphylococcus aureus (ATCC 25923). The fungi, Candida krusei (ATCC 6258) and Candida albicans (ATCC 90028) were also used. As with the multi-resistant bacteria Strains, the Inhibitory Zone Diameter (IZD) in mm and Minimum Inhibitory Concentration (MIC) in mg/mL of the compounds were determined. Ciprofloxacin, and trimethoprim-sulphamethoxazole were used as the antibacterial standard drugs while Fluconazole disk was used as antifungal standard drugs.

The MIC was determined majorly on the concentration range of 0.125 - 10 mg/mL. The compound, ES2ABT showed activity against all Typed Strains (ATCC Cultures) microorganisms. The compound, ES2ABT was active



against the fungi- Candida albicans (ATCC 90028) tested with MIC of 10 mg/mL and IZD of 13 mm. It was also active against Candida krusei (ATCC 6258) with MIC of 10 mg/mL and IZD of 13.

The result showed that the compound has good antifungal properties. Fluconazole is primarily fungistatic but can be fungicidal against certain organisms in dose-dependent manner. Fluconazole was only active against the typed strain Candida albicans (ATCC 90028) but not against C. Krusei tested strains. This was confirmed from literature⁽²⁰⁾. We can conclude that the compound showed some degree of activity against the tested microorganisms which to a large extent can be compared with the standard drugs used. Since the standard antifungal drug used did not show activity against the Candida krusei (ATCC 6258), we can say that the compound, ES2ABT was more active that the fluconazole.

8.1 Lethal Concentration (LC₅₀) and Effective Concentration (EC₅₀)

The result of the Cytotoxic tests viz; Lethal Concentration (LC_{50}) and Effective Concentration (EC_{50}) is recorded in Table 4.

The result showed that compound showed high level of bioactivity against 48 h-nauplii. The compound showed lethal concentration of 338.80 ± 28.6 ppm with EC_{50} of 33.9 ppm. BSLT is a rapid, inexpensive and single bioassay for testing bioactivity of natural and synthetic products, which in most cases correlates reasonably well with cytotoxicity and antitumor properties of the products. The results of Brine Shrimps Lethality Test (BSLT) established that the compound and the complexes are very potent bioactive compounds. EC_{50} value for general bioactivity is approximately one tenth of the value is the LC_{50} in BSLT. The surviving nauphii were dull and inactive, which may be a sign of Central Nervous System (CNS) depression.

9 CONCLUSION

N-(benzothiazol-2-yl)ethanesulphonamide was synthesized. The compound was characterized using X-ray crystallography, mass spectrometry, elemental microanalysis, UV/VIS spectrophotometry, infra red, proton and 13 C NMR spectroscopies. The spectral analyses confirmed the structure of the compound. The antimicrobial tests of the compound were carried out on both multi-resistant bacterial and fungal strains isolated under clinical conditions and cultured species using agar-well diffusion method. The tests were both in vitro and in vivo. The antimicrobial activities of the compound were compared with those of ciprofloxacin and trimethoprim-sulphamethoxazole as antibacterial agents and fluconazole as an antifungal drug. The compound, N-(benzothiazol-2-yl)ethanesulphonamide showed reasonable activity in that it was active against all of the typed strains used-both the bacteria and the fungi, and multi-resistant E. Coli Strain 15 and Pseudomonas aeroginosa strains 34. Since the standard antifungal drug (fluconazole) used did not show activity against the Candida krusei (ATCC 6258), we can conclude that the compound, ES2ABT was more active than the fluconazole and can be recommended for preclinical screening. The Lethal Concentration (LC₅₀) was within the permissible concentrations.

10 Acknowledgement

Dr. L. N. Obasi is grateful to Dr. J. Reglinski of the Department of Pure and Applied Chemistry, University of Strathclyde, Scotland, UK for useful discussions, and the use of some equipment in the course of this work. He is also grateful to Prof. Alexander Gray and his team at SIPBS, also at the University of Strathclyde, Scotland, UK for hosting him, and to the University of Nigeria and the ETF for sponsorship to the UK for a research visit that enhanced this work.

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Table 1: X-ray crystallographic parameter of the compound

Molecular formula	$C_9H_{10}N_2O_2S_2\\$
M	242.31
Crystal system	Triclinic
Space group	P-1
a/Å	4.7298(7)
$b/ m \AA$	9.9806(13)
c/Å	11.4264(14)
α / $^{\rm o}$	99.868(11)
β/°	101.343(11)
$\gamma/^{\rm o}$	96.281(11)
$V/\text{Å}^3$	515.29(12)
Z	2
μ (Mo-Ka)/cm ⁻¹	0.496



T/\mathbf{K}	123
Reflections measured	4814
Unique reflections	2514
Observed reflections	1973
R	0.048
R'	0.104

Supporting Inforation

Details of the x-ray crystal structure determination may be obtained from CCDC, 12 Union Road, Cambridge, CB2 1EZ, UK (fax +44 1223 336033; e-mail deposit@ccdc.cam.ac.uk or www.http://ccdc.cam.ac.uk) on request quoting the depository number CCDC 871872

Table 2: The metrical parameters for the five structurally characterized sulphonamides R = pTos and bz-OMe are netrual compounds. R = Naphthyl and pTol are anionic by virtue of the deprotonation of N1. esd's are not available for R = Naphthyl and pTol

	C(1)-N(1)	N(1)-S(1)	S(1)-O(1)	S(1)-O(2)	C(1)-N(1)-	N(1)-	N(1)-	O(1)-
	C(1)-IV(1)	11(1)-5(1)	3(1)-0(1)	3(1)-0(2)	S(1)	S(1)-O(1)	S(1)-O(2)	S(1)-O(2)
PTos ¹⁷	1.294(7)	1.609(5)	1.440(4)	1.428(4)	123.1(4)	105.1(3)	110.5(3)	118.8(3)
Bz-OMe ¹⁸	1.330(3)	1.614(2)	1.439(2)	1.431(2)	120.7(2)	105.3(1)	111.8(1)	118.0(1)
Naphthyl*19	1.3497	1.5837	1.4457	1.4338	122.25	106.00	112.53	117.05
pTol* ¹⁹	1.3543	1.5802	1.4517	1.4426	122.01	106.26	112.86	116.14
ES2ABT	1.326(3)	1.616(2)	1.446(2)	1.439(2)	119.0(2)	106.5(1)	111.5(1)	117.2(1)



Table 3: Physical properties, mass spectroscopy, and elemental microanalysis of the compound, ES2ABT

Sample	Molecular Mass (gmol ⁻¹) Calc. Found			%C	%]	H	%N		
			Calc.	Calc. Found		Calc. Found		Found	
	242.02	243.13	44.62	47.45	4.16	3.98	11.57	11.09	
ES2ABT	Melting P	Point (°C)	C	blour	Texture				
	170-172		N	lilky	Amorphous				

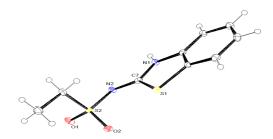


Figure 1: The X-ray crystals structure of ES2ABT. Thermal ellisoids are shown at 30%

Figure 2: The skeletal framework for the sulphonamide with the numbering system for the sulphonamide employed in Table 2.



Table 4: IR Spectra in cm $^{-1}$, UV/Visible spectral Result, ^{1}H & ^{13}C NMR Spectra of the ES2ABT in ppm, andLethal Concentration (LC $_{50}$) and Effective Concentration (EC $_{50}$) results in ppm (Cytotoxic test) of ES2ABT

		IR S	Spectra of th	e ES2ABT in	cm ⁻¹				
Ligand	√ N-H	√ C-H	√ C=C		√ C=N	√ SO ₂	δ C-H	√ C-S-C	
ES2ABT	3484 (br,w)	2975 (br) 1650 (s) 2870(sh) 1625(sh) 2758(sh)			1555.5(s) 1478 (s)	1304.6(s)	640 (s)		
	l	UV/Vi	sible spectra	l Result of E	SZABT	· L	I.		
λmax (nm)	10 ⁻³ € ₁	10 ⁻⁴ e ₂	10 ⁻⁵ € ₃	Assignme	nt				
209.8 264.0	1.92	2.45		π→π* n→ π*					
¹ H NMR	Spectra of the ES2	ABT in ppm			¹³ C NMR Spectra	of the ES2ABT	in ppm		
N-H protons			H ₃ C- (methyl) -CH ₂ protons (methyl) prot		6 2 S S S S S S S S S S S S S S S S S S	NH O S O S 8 H ₃ C ₉			
9.99 (1H, s)	7.89 (4H, m)	1.20 (2H,	q) 1.	11 (3H, t)	ES2ABT	Assignmen	ts		
					169.6	Benzothiazo	ole carbon (C1)	
	Concentration (LC ₅₀) in ppm (Cytotoxic te		Concentratio	n (EC ₅₀)	128.1	Benzothiazole carbon (C2)			
· Courts	Fb (Storovic to	,			139.6	Benzothiazo	ole carbon (C3)	
SAMPLE	LC ₅₀ (ppm)	EC ₅₀ (ppm)			124.8	Benzothiazo	ole carbon (C4)	
ES2ABT	338.80 ±28.6	33.9			123.7	Benzothiazo	ole carbon (C5)	
					123.2	Benzothiazo	ole carbon (C6)	

Legend: br= broad; m= medium; w= weak; s= strong; sh= shoulder



Table 5: Antimicrobial activity of the compound against multi-resistant bacterial strains isolated under clinical conditions

	Multi-resistant bacterial strains isolated from clinical conditions												
	Samples	Escherichia coli strains					Proteus species strains				s aeroginosa	multi-resistant	
S/n		E. Coli Strain 1 E. Coli Strain 15			Pr	Proteus spp Proteus spp			strains 34		Staphylococcus aureus (SR)		
						strains 25		Strains 26				strain	
		MIC MIC		IZD	MIC (mg/ml)	IZD	MIC	IZD (mm)	MIC (mg/ml)	IZD	MIC (mg/ml)		
		IZD	(mg/ml)	IZD	(mg/ml)	(mm)		(mm)	(mg/ml)			(mm)	
		(mm)		(mm)									
1	ES2ABT	00	00	11	10	00	00	00	00	10	10	00	00
2	Ciprofloxacin	00	0.05	00	0.05	00	0.05	25	0.05	27	0.05	00	0.05
3	Trimethoprim-		0.025		0.025		0.025		0.025		0.025		0.025
	sulphamethoxazole												

Table 6: Antimicrobial activity of the compound against Typed Strains (ATCC Cultures) microorganisms

	Typed strains (ATCC Cultures)												
S/n	Samples	Pseudon aerugina (ATCC 2	osa	Escherichia coli (ATCC 25922)		Staphylococcus aureus (ATCC 25923)		Candida krusei (ATCC 6258)		Candida albicans (ATCC 90028)			
		IZD (mm)	MIC (mg/ml)	IZD (mm)	MIC (mg/ml)	IZD (mm)	MIC (mg/ml)	IZD (mm)	MIC (mg/ml)	IZD (mm)	MIC (mg/ml)		
1	ES2ABT	11	10	12	10	13	10	13	10	13	10		
2	Ciprofloxacin	25	0.005	18	0.005	17	0.005	-	-	-	-		
3	Trimethoprim- sulphamethoxazole		0.025		0.025		0.025	-	-	-	-		
4	Fluconazole disk	-	-	-	-	-	-	00	00	20	10		

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