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SYNTHESIS AND CHARACTERIZATION OF SOME NEW DIORGANOBISMUTH (III) ARYLOXYACETATES AND THEIR ANTIMICROBIAL SCREENING

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Abstract- The present manuscript deals the synthesis of some novel organobismuth (III) aryloxyacetates which have a pyramidal geometry as resulted by their characterization with the help of different instrumental techniques along with their spectral analysis. The compounds are also screened for their antibacterial and antifungal studies against different bacterial and fungal strains. It was much surprising that these compounds plays significant role in control of growth of bacterial and fungal strains.

Introduction

It was found in literature that acetates in organic derivatives of group 15 elements could behave as monodentate, bidentate or as bridging ligands depending on the oxidation state (+3 or +5); physical state of the compound (solid or solution) and the various substituents present on the acetate group itself [1-10]. It has been found that major influences in determining mode of bonding, geometry and stability of the acetate system in general are the absence of ligand atom trans to an organic group; the preservation of the preferred angle of 120° at bounded oxygen atom and maximizing of secondary bonding via a bridging for acetate. Another important group of ligand which has not attracted much attention, but closely resembles to carboxylates is aryloxyacetates. However unlike acetate, aryloxyacetates has one extra donor site i.e. Ax-O-, in addition to the -C=O functionality. It is noteworthy that despite such a great variety of structural possibilities, as discussed above exhibited by organometallic carboxylates and by the variety of metals for which such derivatives have been synthesized, corresponding aryloxyacetates are mainly confirmed to transition metal derivatives [11] with an occasional reference on organotin and organoantimony [12] derivatives. A perusal of the literature further reveals that the complexes of aryloxyacetic acids in case of organotin possess intramolecular O--->Sn coordination; particularly for alkyl tin derivatives and the compounds are monomeric in solid state. In sharp contrast to this triaryltin derivatives having pentacoordination, were found to be polymeric with bridging carboxylic group [13-19]. On the basis of ultraviolet and infrared spectra it has been concluded that aryloxyacetate behave as monodentate ligand towards bismuth. The present manuscript deals the synthesis and characterization of some novel organobismuth (III) aryloxyacetates for their antibacterial and antifungal studies against different bacterial and fungal strains.

Experimental

The diorganobismuth (III) chloride, R_2BiCl was prepared by the redistribution reaction as per reported earlier [20]. The syntheses of some representative compounds are as follows.

Reaction of $(C_6H_5)_2Bi(III)Cl$ with $(C_6H_5OCH_2COOH)$ (1)

In the stirring solution of diphenylbismuth(III)chloride (0.398gm;1mmol), phenoxyacetic acid (0.152gm;1mmol) was added in presence of triethylamine (1 ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A flocculent white precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off. The filtrate on concentration gave a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of $(C_6H_5)_2Bi(III)Cl$ with (*p*-$CH_3C_6H_4OCH_2COOH$) (2)

In the stirring solution of diphenylbismuth(III)chloride (0.398gm;1mmol), *p*-methyl phenoxyacetic acid (0.166gm;1mmol) was added in presence of triethylamine (1ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A flocculent white precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off. The filtrate on concentration afforded a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of $(C_6H_5)_2Bi(III)Cl$ with (*p*-$ClC_6H_5OCH_2COOH$) (3)

In the stirring solution of diphenylbismuth(III)chloride (0.398gm;1mmol), *p*-chloro phenoxyacetic acid (0.186gm;1mmol) was added in presence of triethylamine (1ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A white colour precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off and the filtrate on concentration in vaccum afforded a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of $(C_6F_5)_2Bi(III)Cl$ with $(C_6H_5OCH_2COOH)$ (4)

In the stirring solution of bis(pentafluorophenyl) bismuth(III)chloride (0.578gm;1mmol), phenoxyacetic acid (0.152gm;1mmol) was added in presence of triethylamine (1 ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A flocculent white precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off. The filtrate on concentration gave a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of $(C_6F_5)_2Bi(III)Cl$ with (*p*-$CH_3C_6H_4OCH_2COOH$) (5)

In the stirring solution of bis(pentafluorophenyl)bismuth(III)chloride (0.578gm;1mmol), *p*-methyl phenoxyacetic acid (0.166gm;1mmol) was added in presence of triethylamine (1ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A flocculent white precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off. The filtrate on concentration afforded a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of $(C_6F_5)_2Bi(III)Cl$ with (*p*-$ClC_6H_5OCH_2COOH$) (6)

In the stirring solution of bis(pentafluorophenyl) bismuth(III)chloride(0.578gm;1mmol), *p*-chloro phenoxyacetic acid (0.186gm;1mmol) was added in presence of triethylamine (1ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A white colour precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off and the filtrate on concentration in vaccum afforded a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of (*p*-$FC_6H_4)_2Bi(III)Cl$ with $(C_6H_5OCH_2COOH)$ (7)

In the stirring solution of bis(*p*-fluorophenyl) bismuth(III)chloride (0.434gm;1mmol), phenoxyacetic acid (0.152gm;1mmol) was added in presence of triethylamine (1 ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A flocculent white precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off. The filtrate on concentration gave a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of (*p*-$FC_6H_4)_2Bi(III)Cl$ with (*p*-$CH_3C_6H_4OCH_2COOH$) (8)

In the stirring solution of bis(*p*-fluorophenyl)bismuth(III)chloride (0.434gm;1mmol), *p*-methyl phenoxyacetic acid (0.166gm;1mmol) was added in presence of triethylamine (1ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A flocculent white precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off. The filtrate on concentration afforded a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Reaction of (*p*-$FC_6H_4)_2Bi(III)Cl$ with (*p*-$ClC_6H_5OCH_2COOH$) (9)

In the stirring solution of bis(*p*-fluorophenyl)bismuth(III)chloride (0.434gm;1mmol), *p*-chloro phenoxyacetic acid (0.186gm;1mmol) was added in presence of triethylamine (1ml) in benzene and was stirred in anhydrous oxygen free, nitrogen conditions for 6h, followed by refluxing for 2h to ensure completion of the reaction. A white colour precipitate of $Et_3H.HCl$ (M.P.240°C) was formed which was filtered off and the filtrate on concentration in vaccum afforded a light brown solid which was recrystallised by petroleum ether (40°-60°C).

Antibacterial activity

The antibacterial activity of these organobismuth (III) compounds was determined by disc diffusion method [21]. In this technique, the filter paper (Whatman No. 1) sterile discs of 5 mm diameter, impregnated with the test compounds (10 μg/ml of ethanol) were placed on the nutrient agar plate at 37°C for 24 hrs. The inhibition zones around the dried impregnated discs were measured after 24 hrs. The activity was classifieds as 'highly active' (diameter > 14 mm); "moderately active" (diameter = 10-14 mm) and 'slightly active' (diameter = 6-10). The diameter less than 6 mm was regarded as inactive.

Antifungal activity

The antifungal activity of these compounds was tested by agar diffusion method [22] using two concentrations of the test compound, *viz*, 50 and 100 μg/ml against *Aspergillus flavus* and *Aspergillus niger*. The one ml of each organobismuth compound was poured into a petri dish having about 20-25 ml of molten potato dextrose agar medium of. As the medium gets solidify, petri dishes were inoculated separately with the fungal isolates and kept at 26°C for 96 hrs in incubator. All the values (% inhibition) were recorded after 96 hrs and their % inhibition was calculated.

Results and Discussion

All the reactions were conducted at room temperature and the products were recrystallised from petroleum ether (40-60°C) or in benzene. The complexes are off-white to light brown solids and obtained as a sticky mass which on treatment with sodium in dry benzene gets solidified and subsequently crystallized with benzene/pet-ether. The complexes are fairly stable on air and moisture and have sharp melting point. There is no regular trend of the melting point of the complexes and they melt without decomposition. Complexes are also soluble in chloroform and acetonitrile. They can be stored at room temperature without decomposition for several weeks. The molar conductance value of 10^{-3} M solution of these compounds were recorded in methanol and found in the range of 15-25 Ohm^{-1} $mole^{-1}$ cm^2 indicating the absence of ionic species in solution.

IR Spectra

As expected infrared absorptions inherent to pheny1and fluorophenyl groups bound to bismuth do no differ appreciably and hence not discussed. The Infrared absorptions having diagnostic value for organobismuth aryloxyacetates, related to the ligand, have been identified which on preliminary stage indicates the mode of bonding with aryloxy ligand. The characteristic v(OH) absorption band of ligands which appeared around 3400cm^{-1} in the free ligand, was found missing in the newly synthesized complexes. A medium strong intensity band appearing at 1690-1700 cm^{-1} can confidently be assigned to v_{asy}(OCO) mode while comparatively weaker band in the range 1380-1400 cm^{-1} can be attributed to v_{sym} (OCO) band. The deformation mode as a medium intensity band was found in the range 780-815 cm^{-1}. The absorption associated with the bismuth-oxygen appears in the range between 400-430 cm^{-1} and the absorption due to bismuth-carbon corresponding to y-mode occurs in the range 450-470cm^{-1}. These values clearly indicated the formation of organobismuth (III) aryloxyacetates. The comparison of IR spectra of the compounds with those of respective ligands in solid and solution states did not show any significant shift in v_{asy}(C=O), v_{sym} (C-O) and v(C-O-C) deformation bands which in turn showed the lack of coordination with bismuth through -C=O or C-O-Ar center of the ligand. Since the separation observed in the present compounds is fairly large (2300cm^{-1}), monomolecular constitution seems to be most plausible where bismuth would be have a coordination number three. This observation is in sharp contrast to organotin complexes of aryloxyacetates which have been found to be polymeric involving carboxylic bridges. In addition to this intermolecular interaction involving the ethereal oxygen has also been demonstrated in some cases particularly.

^{1}HNMR Spectra

^{1}H NMR spectra of these compounds was recorded in $CDCl_3$ using TMS as an internal reference at 25°C. The disappearance of OH proton signals (89.1 ppm) present in the ligand clearly indicates the formation of aryloxyacetate derivatives. The appearance of singlet for -CH_3 protons at 84.85 ppm showed that the ligand is in one plane. The pheny1 protons for both the derivatives appear as multiplets in the range δ7.80-7.20 ppm.

UV Spectra

The electronic spectra obtained for representative compounds were recorded in chloroform in the range 200-400 nm. The UV absorption due to COO group appears at 274±6 and 294±2 are due to aryloxy moieties. Since there was no significant change in absorption peaks of the ligands indicates that -C=O and -C-O-Ar centre of aryloxyacetates are not coordinated to bismuth in any of the compounds. This also lends support to the fact that aryloxyacetates behave as monodentate ligand towards bismuth in +3 oxidation state.

Based on IR, NMR and UV spectral analysis data, it may tentatively be concluded that aryloxyacetate under the present study behave as monodentate ligand and these compounds have three coordinate pyramidal structures.

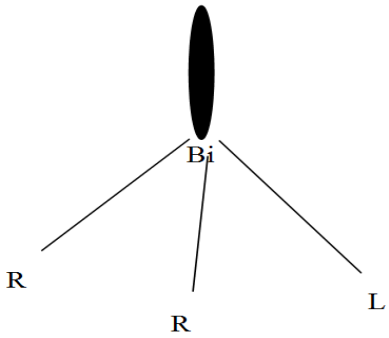

Fig.1- L=Aryloxyacetate Group

Antibacterial activity

All the diorganobismuth (III) aryloxyacetates were tested for antibacterial activity against three bacterial strains *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Klebsiela pneumoniae* using 10 μg/ml concentration of test compound. All the compounds show higher to moderate activity against the bacterial strains. It was found that compounds having fluoro and pentafluorophenyl ring are more effective because of their water and lipid solubility. The fluorine containing compounds may generally form complexes with metaloenzymes, particularly those which responsible in basic physiology such as *cytochrome oxidase*. These compounds may react with peptidoglycan layer of bacterial cell wall and damage it by penetrating in such a manner that the phenyl ring gets entered inside the cell by puncturing it followed by death of bacterial cell. Some times these compounds in low concentration may cause bacteriostatic condition by slow down the growth of bacteria.

Antifungal Activity

The antifungal activity of all these compounds was tested against *Aspergillus flavus* and *Aspergillus niger* using 50 and 100 μg/ml concentration. The activity of these compounds was found variable at 50μg/ml concentration

but at higher concentration all the compounds show high activity against fungal strains. Presence of phenyl and pentafluorophenyl ring along with bismuth in +3 oxidation state is considered for fungal activity. The role of different aryloxyacetates as ligands was also commendable. These compounds generally damage the fungal strains by puncturing the cell wall similarly as in case of bacteria. Water and lipid solubility also increases the activity due to presence of fluorine.

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Table-1 Physicochemical studies of organobismuth (III) aryloxyacetates

Sr. No.	Compounds	Formula	M.P. (°C)	Colour	Yield (%)	Solvent
1	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5)$	$C_{20}H_{17}O_3Bi$	190	Light Brown	60	Pet-ether
2	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	$C_{21}H_{19}O_3Bi$	156	Light Brown	75	Pet-ether
3	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	$C_{20}H_{16}O_3ClBi$	214	Light Brown	70	Pet-ether
4	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5)$	$C_{20}F_{10}H_7O_3Bi$	184	Light Brown	65	Pet-ether
5	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	$C_{21}F_{10}H_9O_3Bi$	176	Light Brown	60	Pet-ether
6	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	$C_{20}F_{10}H_6O_3ClBi$	204	Light Brown	60	Pet-ether
7	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5)$	$C_{20}F_2H_{15}O_3Bi$	192	Light Brown	60	Pet-ether
8	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	$C_{21}F_2H_{17}O_3Bi$	182	Light Brown	75	Pet-ether
9	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.Cl)$	$C_{20}F_2H_{14}O_3ClBi$	198	Light Brown	70	Pet-ether

Table-2 Analytical studies organobismuth (III) aryloxyacetates

Sr. No.	Compounds Formula	Formula weight	Elemental Analysis		IR Absorption frequencies		
			C (%)	H (%)	ν_{asy}(OCO)	ν_{sym}(OCO)	ν (Bi-C)
1	$C_{20}H_{17}O_3Bi$	514	46.69	3.30	1695	1395	411
2	$C_{21}H_{19}O_3Bi$	528	47.73	3.60	1694	1385	415
3	$C_{20}H_{16}O_3ClBi$	548.5	43.76	2.92	1694	1387	417
4	$C_{20}F_{10}H_7O_3Bi$	694	34.58	1.01	1696	1390	419
5	$C_{21}F_{10}H_9O_3Bi$	708	35.59	1.27	1700	1397	410
6	$C_{20}F_{10}H_6O_3ClBi$	728.5	32.94	0.82	1697	1400	425
7	$C_{20}F_2H_{15}O_3Bi$	550	43.64	2.72	1692	1406	428
8	$C_{21}F_2H_{17}O_3Bi$	564	44.68	3.01	1688	1394	422
9	$C_{20}F_2H_{14}O_3ClBi$	584.5	41.06	2.39	1694	1408	431

Table-3 Anti-bacterial activity of organobismuth (III) aryloxyacetates

Sr. No.	Compounds	Control	*Pseudomonas aeruginosa*	*Staphylococcus aureus*	*Klebsiela pneumoniae*
1	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5)$	–	+++	+++	++
2	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	–	++	++	++
3	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	–	+++	++	++
4	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5)$	–	++	++	++
5	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	–	++	++	+++
6	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	–	+++	++	++
7	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5)$	–	++	++	++
8	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	–	++	++	+++
9	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.Cl)$	–	+++	+++	++

+ = 6-10 mm; ++ = 10-14 mm; +++= >14 mm; – = Inactive

Table-4 Antifungal activity of organobismuth (III) aryloxyacetates at 50 μg/ml concentration

Sr. No.	Compounds	*Aspergillus flavus* Col. Dia. (mm)	% Inhibition	*Aspergillus niger* Col. Dia. (mm)	% Inhibition
1	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5)$	0.7	76.6	0.6	70.0
2	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	0.2	93.3	0.7	65.0
3	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	0.2	93.3	0.7	65.0
4	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5)$	0.5	83.3	0.4	80.0
5	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	0.2	93.3	0.7	65.0
6	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	0.2	93.3	0.7	65.0
7	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5)$	0.7	76.6	0.7	65.0
8	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	0.8	73.3	0.8	60.0
9	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.Cl)$	0.8	73.3	0.8	60.0
10	Control	3.0	–	2.0	–

Table-5 Antifungal activity of organobismuth (III) aryloxyacetates at 100 μg/ml concentration

Sr. No.	Compounds	*Aspergillus flavus* Col. Dia. (mm)	% Inhibition	*Aspergillus niger* Col. Dia. (mm)	% Inhibition
1	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5)$	0.1	96.7	0.4	80.0
2	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	0.2	93.3	0.3	75.0
3	$(C_6H_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	0.2	93.3	0.1	95.0
4	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5)$	0.1	96.7	0.1	95.0
5	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	0.4	86.7	0.2	90.0
6	$(C_6F_5)_2Bi(OOC.CH_2OC_6H_5.Cl)$	0.1	96.7	0.3	75.0
7	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5)$	0.2	93.3	0.3	75.0
8	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.CH_3)$	0.1	96.7	0.2	90.0
9	$(p\text{-}FC_6H_4)_2Bi(OOC.CH_2OC_6H_5.Cl)$	0.2	93.3	0.1	95.0
10	Control	3.0	–	2.0	–