American International Journal of Research in Science, Technology, Engineering & Mathematics

Available online at http://www.iasir.net

ISSN (Print): 2328-3491, ISSN (Online): 2328-3580, ISSN (CD-ROM): 2328-3629

AIJRSTEM is a refereed, indexed, peer-reviewed, multidisciplinary and open access journal published by International Association of Scientific Innovation and Research (IASIR), USA

(An Association Unifying the Sciences, Engineering, and Applied Research)

Spectral and Antifungal Studies on Nickel Phenylbiguanide Complexes with Different Anions

Bina Rani[a] and R.K. Prasad[b]

[a]Department of Chemistry, Magadh Mahila College, Patna University, Patna-800 001, Bihar, INDIA

[b]Department of Chemistry, Science College, Patna University, Patna-800 005, Bihar, INDIA

Abstract: *Isomeric cis trans Nickelphenylbiguanide salts of composition have been isolated and characterized from the studies of their colour, magnetic susceptibilities, infrared vibrations and electronic absorption spectra. These complexes have been found to be effective in controlling the growth of aspergillius niger in PDA (Potato Dextrose Agar) and SDA (Sabourand and Dextrose Agar) mediums. Some metal complexes of phenylbiguanide with anions: S_2O_3, SO_3, BrO_3 and NO_2 have been synthesized and isolated in the pure state. These metal complexes have been found effective in inhibiting the growth of fungi named, Aspergillus niger and Aspergillus versicolor. The metal complexes of phenylbiguanide ligand were dissolved in DMSO and were tested with Dilution test method and growths were separated and examined under different concentrations of the afore said complexes in PDA (Potato Dextrose Agar) and SDA (Sabourand and Dextrose Agar) medium.*

Keywords: *Phenylbiguanide, Potato Dextrose Agar, Sabourand and Dextrose Agar and Aspergillus Niger*

I. Introduction:

Biguanide can refer to a molecule or to a class of drugs based upon this molecule. Biguanide can function as oral anti-hyperglycemic drugs used for diabetes mellitus or prediabetes treatment. They are also used as antimalarial drugs[10-14].

Biguanides have a replaceable hydrogen atom in a potentially imino group and also has a donor nitrogen (amino) atom for chelation, thus behaving as bidentate ligand. In formula V suggested by Ray[15], it has been shown that in the inner metallic complexes formed by biguanide, it is the hydrogen atom of one of the imino groups (=NH) of biguanide, which is replaced by the metal atom to form a primary valence bond.

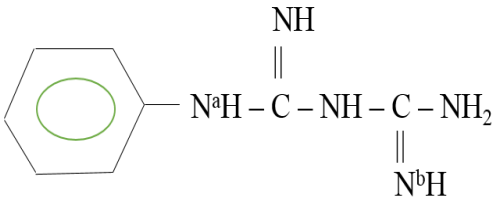

Fig 1. For Phenylbiguanide we can represent the bonding N by labelling it as a and b.

II. Experimental

A. Phenybiguanide hydrochloride (Melting Point 244 – 247^{0}C) $[C_8H_{11}N_5.HCl]$

Phenylbiguanide hydrochloride was prepared by refluxing alcoholic solution of both aniline hydrochloride $C_6H_5NH_2.HCl$ and dicyandiamide $C_2H_4N_4$ in round bottom flask on water bath. This process continues for about 2 hours (Smolka and Friedrich Monatch, 1888, 9, 227). The crystals get deposited on cooling the mixture obtained. The crystals were filtered and recrystallized with hot water.

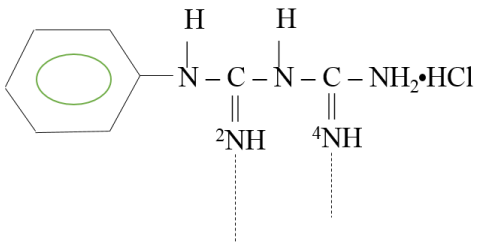

Fig 2. Phenylbiguanide hydrochloride

B. **Bis(phenylbiguanidium) nickel(II)thiosulphate**
$[Ni(PhBigH)_2]S_2O_3$

It was obtained from an excess of conc. solution of sodium thiosulphate and that of complex chloride. The yellow crystalline precipitate was washed & dried as usual.

Found: N=26.50%, Ni=11.17%, S_2O_3=21.53%
$[Ni(PhBigH^+)_2]S_2O_3$ requires: N=26.63%, Ni=11.18%, S_2O_3=21.33%

C. **Bis(phenylbiguanidium) nickel(II)suphite**
$[Ni(PhBigH)_2](SO_3).H_2O$

A concentrated solution of sodium sulphite in excess was added to a hot concentrated solution of complex chloride. On cooling, the yellow crystals of Bis(phenylbiguanidium) nickel(II)suphite gradually separated out.

Found: Ni=11.53% and SO_3^{-2}=15.65%
$[Ni(PhBigH)_2](SO_3).H_2O$ requires: Ni=11.50% and SO_3^{-2}=15.75%

D. **Bis(phenylbiguanidium) nickel(II)bromate**
$[Ni(PhBigH)_2](BrO_3)_2$
It was obtained as sparingly soluble, orange coloured crystals from a conc. solution of potassium bromate and a hot conc. solution of the complex chloride $[Ni(PhBigH^+)_2]Cl_2$.

Found: Ni=8.72%, BrO_3^- =37.92
$[Ni(PhBigH)_2](BrO_3)_2$ requires: Ni=8.77%, BrO_3^- =38.27%

E. **Bis(phenylbiguanidium) nickel(II) nitrite monohydrate**
$[Ni(PhBigH)_2](NO_2)_2.H_2O$

It was obtained from a conc. solution of $NaNO_2$ (sodium nitrite) and that of the complex chloride $[Ni(PhBigH^+)_2]Cl_2$. It forms sparingly soluble orange yellow crystals.

Found: Ni=11.15%, NO_2^- =18.0%
$[Ni(PhBigH)_2](NO_2)_2.H_2O$ requires: Ni=11.23%, NO_2^- =17.60%

F. **Preparation of PDA(Potato Dextrose Agar)**
Potato tubers were taken peeled off and weighed 200g. It was chopped into small pieces and transferred to a beaker containing about 100ml of distilled water and boiled for 20minutes and filtered with muslin cloth. 20g Dextrose, 15g agar and 2g peptone were added into the extract and gently heated. The filterate so obtained was made to 1 litre. The pH of the solution was maintained at 5.6 by using 1N HCl or NaOH and kept in Erlenmeyer flask. This solution so obtained was PDA medium and autoclaved at 121^0C for 20minutes before using.

G. **Preparation of SDA (Sabourand and Dextrose Agar Medium)**
It was prepared by combining the ingredients water, dextrose, agar, peptone & antibiotics separately, in many different variations. In the case of using premix, the proper amount (around 70gms) was mixed with one litre of water and heated to dissolve the agar. pH of the medium was adjusted with one molar solution of hydrochloric acid to lower pH. The pH was maintained at 5.5. The medium was then autoclaved and stored at room temperature. The medium can be used to inoculate with fungal spores and mycelium inhibition growth was counted by usual method. The observation of the study is represented in the Table A.

Results and Discussion
Nickel phenylbiguanide hydrates and its salts – the nickelphenylbiguanidium chloride or Bis(phenylbiguanide) nickel(II) chloride was prepared which was then used to prepare further other complexes. The anhydrous nickel phenylbiguanidium have proved to be monomeric by ebullioscopic measurements. Bis(phenylbiguanidium) nickel(II)thiosulphate was obtained from an excess of conc. solution of sodium thiosulphate and that of complex chloride. A concentrated solution of sodium sulphite in excess with hot concentrated solution of complex chloride was used to generate Bis(phenylbiguanidium) nickel(II)suphite. Similarly, Bis(phenylbiguanidium)

nickel(II)bromate was obtained using potassium bromate while Bis(phenylbiguanidium) nickel(II) nitrite monohydrate was obtained using sodium nitrite.

The planar configuration of the 4 coordinated inner metallic complexes of bivalent Ni is now been well established. Evidences in support of this are, derived not only from the classical methods of stereochemistry but also from a substantial mass of physical data, specially relating to X-ray measurement of crystalline salts.

Electronic Spectra

A. UV spectra

For Bis(phenyldibiguanidium)nickel(II)chloride

250nm CT + $^1A_{1g} \longrightarrow {}^1E_g$

340nm CT

For Bis(phenyldibiguanidiium)nickel (II) sulphite

260nm CT + $^1A_{1g} \longrightarrow {}^1E_g$

340nm CT

For Bis(phenyldibiguanidiium)nickel (II) thiosulphate

440nm $^1A_{1g} \longrightarrow {}^1B_{1g}$

340nm & 360nm CT

240nm CT + $^1A_{1g} \longrightarrow {}^1E_g$

For Bis(phenyldibiguanidiium)nickel (II) bromate

240nm CT + $^1A_{1g} \longrightarrow {}^1E_g$

B. IR spectra

K_1 (Phenyldibiguanidine hydrochloride)

Band Position (in cm^{-1})	Assignment
3550.8	N-H (w)
3419.1	ν (NH_2) + ν (N-H)
3377.6	ν (NH_2) + ν (N-H)
3287.7	N-H(str)(H-bonded)
3165.0	C-H(str) in aromatic ring
2915.5	ν (C-H)
2718.6 **2508.8** **2455.6** **2369.7**	N-H(str) in>N^+H
1748.1	N-H (scissoring)
1626.4	N-H def for NH_2^+, C=N(str)
1544.3	N-H def for NH_3^+, C=N(str)
1291.4 **1218.8**	C-N str in aromatic amines In plane bending bands of ring C-H bond
1068.9	C-N str
831.0 **771.3** **678.6**	Out of plane bending of the ring C-H bond
576.7	N-C-N (def)
520.1	N-C-N (def)

K_2 Bis(Phenyldibiguanidium)nickel(II)chloride

Band Position (in cm^{-1})	Assignment
3595.9	N-H (wagging) and ν(H_2O)
3384.7	ν (NH_3) & ν (N-H)
3302.9	ν N-H (H-bonded)
3021.6	ν (C-H) in aromatic group
2401.4	N-H(str) in>N^+H
1586.8	ν C=C(str), C-N(str)
1519.7	C=C(str)

1293.5	In plane bending bands of ring C-H bond C-N(str) in aromatic amines
1216.2 **1068.7** **928.7**	In plane bending bands of ring C-H bond C-N(str) in aliphatic amines
757.9	C-H def in mono substituted aromatic compounds
670.5	Out of plane C-H bending in benzene ring
503.5	ν Ni-N

K_3 Bis(phenyldibiguanidium)nickel(II) thiosulphate

Band Position (in cm^{-1})	Assignment
3428.7	ν (N-H) & ν (NH_2)
3022.3	ν (C-H) in aromatic group
2367.6	ν N-H in>N^+H
1652.5	Str C=C in aromatic nuclei
1523.0	Str C=C in aromatic nuclei
1427.0	N-H def
1216.7 **928.8**	str C-N in aromatic amines In plane bending of C-H in phenyl ring
764.8 **671.2**	Phenyl ring C-H out of plane bending
536.1	N-C-N (def)
498.9	ν (Ni-N)

K_4 Bis(phenyldibiguanidium)nickel(II) bromate

Band Position (in cm^{-1})	Assignment
3376.5	ν (N-H) & ν (NH_2)
3022.1	ν (C-H) in aromatic group
2367.8	ν (N-H) in>N^+H
1657.7	Str C=C in aromatic nuclei
1520.3	Str C=C in aromatic nuclei
1429.3	N-H def
1216.9 **930.0**	str C-N in aromatic amines In plane bending of C-H bond in phenyl ring
766.6 **671.9**	Phenyl ring C-H out of plane bending
497.7	ν (Ni-N)

K_5 Bis(phenyldibiguanidium)nickel(II) sulphite

Band Position (in cm^{-1})	Assignment
3454.2	ν (N-H) & ν (NH_2)
3022.9	ν (C-H) in aromatic group
2402.9	ν (N-H) in>N^+H
2369.8	ν (N-H) in>N^+H
2258.3	ν (N-H) in>N^+H
1631.5	Str C=C in aromatic nuclei
1524.0	Str C=C in aromatic nuclei
1475.8	N-H def
1430.0	N-H def
1216.8	str C-N in aromatic amines In plane bending of C-H bond
1026.0 **928.5**	str C-N in aliphatic amines In plane bending of C-H bond
763.8 **671.2**	Phenyl ring C-H out of plane bending
555.1	N-C=N
497.8	ν (Ni-N)

K_6 Bis(phenyldibiguanidium)nickel(II) bromate

Band Position (in cm^{-1})	Assignment
3459.7	ν (NH) & ν (NH_2)
3369.4	ν (NH) & ν (NH_2)
3021.6	ν (C-H) in aromatic group
2401.3	N-H str in>N^+H
2361.7	N-H str in>N^+H
1609.8	Str C=C in aromatic nuclei
1509.3	Str C=C in aromatic nuclei
1426.8	N-H def
1216.0	str C-N in aromatic amines In plane bending of C-H bond
1032.0 **929.9**	str C-N in aliphatic amines In plane bending of C-H bond
767.3 **670.8**	Phenyl ring C-H out of plane bending
500.4	ν (Ni-N)

The metal complexes of phenylbiguanide were dissolved in DMSO using the concentration which was most effective from Dilution test method. The inoculation of fungus was done in PDA and SDA medium at 25^0C and checked daily for a week. The fungus are Aspergillusniger and Aspergillusversicolor. Both are from ascomycetes group. The MIC was then calculated i.e. the minimum inhibition concentration of the fungus. During this process it was found that the complexes was effective in controlling 100% of the fungal growth if the concentration was raised to 800µg/ml to 1000µg/ml. The solution was used in the ratio 1:10 and the concentration of the solution was 400µg/ml, 200µg/ml and 100µg/ml. Refer to table I for **Micelle count inhibition of fungal growth**.

	% Inhibition of fungal growth		
Metal complex	**Concentration (µg/ml)**	**PDA Medium**	**SDA Medium**
Bis(phenylbiguanidium)nickel(II)chloride	400 µg/ml	84.01%	82.09%
	200 µg/ml	65.72%	62.37%
	100 µg/ml	44.36%	42.27%
Nickelphenylbiguanide	400 µg/ml	85.3%	84.70%
	200 µg/ml	60.51%	57.92%
	100 µg/ml	40.17%	38.72%

Table I Percentage inhibition of growth of fungus Aspergillusniger at indicated dose

References

[1]. Chohan, Z.H. and Rauf, A. (1996). Some Biologically Active Mixed Ligand Complexes of Co(II), Cu(II) and Ni(II) with ONO, NNO and SNO Donor Nicotinoylhydrazine-Derived Ligands. *Synthesis and Reactivity in Inorganic and Metal-Organic Chemistry.* 6, 591-604.

[2]. Bonire, J.J., Ayoko, G.A., Olurinola, P.F., Ehinmidu, J.O., Jalil, N.S.N. and Omachi, A.A. (1998). Syntheses and Antifungal Activity of some organotin(IV)carboxylates. *Metal-Based Drugs.* 5 (4), 233 - 236 and references therein

[3]. Chohan, Z.H. and Rauf, A. (1996). Some Biologically Active Mixed Ligand Complexes of Co(II), Cu(II) and Ni(II) with ONO, NNO and SNO Donor Nicotinoylhydrazine-Derived Ligands. *Synthesis and Reactivity in Inorganic and Metal-Organic Chemistry.* 6, 591-604. Coates, L.V., Brain, D.J., Kerridge, K.H., Marcus, F.J., and Tattershall, K. (1957). *Journal of Pharmacy andPharmacology*, 9, 855.

[4]. Gershon, H. (1974). Antifungal Activity of Bischelates of 5-, 7-, and 5,7-halogenated 8-quinols with copper(II).Determination of the long and short aces of the pores in the fungal spore wall. *Journal of Medicinal Chemistry*. 17, 824-827.

[5]. Hadi, S. and Irawan, B. (2007). Synthesis, Characterization and The Antifungal Activity Test of Diphenyltin(IV) Complexes. Proceeding of International Conference on Chemical Sciences, Gadjah Mada University, 24 – 26 May 2007, pp. 21-216 (Life Sciences Section)

[6]. Hadi, S., Irawan, B., Efri. (2008). The Antifungal Activity Test of Some Organotin(IV) Carboxylates. Accepted for Publication at *Journal of Applied Sciences Research. **In Press.***

[7]. Mahmood, S., Ali, S., Bhatti, M.H., Mazhar, M., Iqbal, R. (2003). Synthesis, Characterization and Biological Applications of Organotin(IV) Derivatives of 2-(2-Fluoro-4-biphenyl)propanoic Acid, *Turkish Journal of Chemistry*, 27,657-666.

[8]. Pellerito, L. and Nagy, L. (2002). Organotin(IV)n+ complexes formed with biologically active ligands: equilibrium and structural studies, and some biological aspects, *Coordination Chemical Reviews* 224, 111 – 150 and references therein

[9]. Ruzika, A., Dostal, L., Jambor, R., Butcha, V., Brus, J., Cisarova, I., Holcapek, M., and Holecek, J. (2002). Structure and in vitro antifungal activity of [2,6-bis(dimethyl-iminomethyl)phenyl]diphenyltin(IV) compounds, *AppliedOrganometallic Chemistry*, 16(6), 315 – 322.
[10]. Hudson, A.T., M. Dickins, C. D. Ginger, W. E. Gutteridge, T. Holdich, D. B. Hutchinson, M. Pudney, A. W. Randall and V. S. Latter. 1991. 566C80; *a broad spectrum anti-infective agent with activity against malaria and opportunistic infections in AIDS patients*. Drugs Exp. Clin. Res. 17:427-435. [Pubmed]
[11]. Schafer, G. 1983. Biguanides; *A review of history, pharmacodynamics and therapy*, DiabeteMetab. 9:148-163. [Pubmed]
[12]. Basco, L. K., P. E. Pecoulas, C.M. Wilson, J. Le Bras and A. Mazabraud, 1995. *Point mutations in the dihydrfolatereducatese-thymidylate synthase gene and pyrimethamine and cycloguanil resistance in Plasmodium falciparum*. Mol. Biochem. Parasitol. 69:135-138, [Pubmed]
[13]. Bloland, P.B., P. N. Kazembe, W. M. Watkins, O. K. Doumbo, O. C. Nwanyanwu, and T. K. Ruebush. 1997. Malarone donation programme in Africa. Lancet 350:1624-1625. [Pubmed]
[14]. Radloff, P. D., J. Phillips, M. Nkeyi, D. Hutchinson, and P. D. Kremsner, 1996. *Atovaquone and Proguani for Plasmodium falciparum malaria*. Lancet. 347:1511-1514. [Pubmed]
[15]. P. Ray and H. Saha : J. Indian Chem. Soc. 26, 144 (1949)
[16]. Smolka abd Friedrich Monatch 1888, 9, 227.
[17]. Catherine E. Housecroft and Alan G. Sharpe, Inorganic Chemistry, 2nd Edition
[18]. B. Rathike : Ber., 12, 774 (1879)
[19]. Bina Rani, R.K. Prasad and Divya Singh, *AIJRSTEM 15 – 140* 9(1), December 2014 – February 2015, p – 77 – 80.
[20]. R.K. Prasad, Mrs. Bina Rani and Dhananjai Singh, *AIJRSTEM 14 – 744* © 8(2) September – November 2014, pp – 108 – 111.
[21]. R.K. Prasad, Mrs. Bina Rani and Madhu Kumari Gupta, *AIJRSTEM 14 – 329* 6(1) March – May 2014, pp – 92 – 96.
[22]. R.K. Prasad, Mrs. Bina Rani, *AIJRSTEM 15 – 713* 12(1) September – November 2015, pp 8 – 10.
[23]. R.K. Prasad, Mrs. Bina Rani, *AIJRSTEM 17- 122* December, 2016 to February, 2017, pp 57 - 62