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Synthesis and Antiurease & Antioxidant Activities of *Bis*-Schiff Bases of Isophthalaldehyde

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Bis-Schiff bases of iosophthalaldehyde were synthesized by reacting isophthalaldehyde with hydrazine hydrate in 1:2 ratio in ethanol for 2-3 h under reflux. Product of first step was again reacted in 1:2 ratio with different aldehyde or acetophenone yielding *bis*-Schiff bases (**1-16**). All compounds were characterized by EI-MS and ^{1}H NMR. All analogs were tested against antioxidant and urease inhibition. Few compounds were found to showed potent antiurease and antioxidant potential.

Keywords: Schiff bases, Isophthalaldehyde, Urease inhibition, Antioxidant potential.

INTRODUCTION

Urease plays a key role in certain human and animal diseases contributing in kidney stone, polynephritis, peptic ulcer, gastric cancers and other ailments [1]. Urease shows its vitality in nitrogen metabolism during plant germination [2,3]. Carbon dioxide and ammonia are formed from urea hydrolysis in presence of urease [4-7]. Phyto-pathogenesis and loss of ammonia results due to excessive decomposition of urea by topsoil urease [8]. Many Schiff bases inhibit urease by decelerating the formation of NH_4^+ thus enhance the soil fertility [9]. Due to oxidative stress, antioxidant defense system causing the formation of reactive oxygen species [10]. Antioxidants are helpful in healing of blood vessels membranes, normalizing the blood flow to heart and brain as an anticancer agent, preventing Alzheimer disease and dementia preventing through radical scavenging [11-13]. Free radicals also contribute in pathologies of artheriosclerosis, malaria and rheumatoid arthritis and play a vital role in neurodegenerative disease and in aging, antioxidants are used to reduce their activity [14,15]. Well known antioxidant is glutathione present in deficient amount result in lipid peroxidation, DNA strand breaks and membrane damage occur [16]. Proteases, nucleases and protein kinesis mechanisms are also associated with it. Oxidative damages have also take part in human diseases. However, its contribution to illness differs from case to case. In case of atheriosclerosis, it evident from literature with the chain-breaking antioxidant probucol and from epidemiological work suggests that oxidative damage promotes plaque development [17].

Our research group had previously reported various novel classes' of DPPH radical scavengers [18]. In view of our previous work we have synthesized *bis*-Schiff based of isophthalaldehyde (**1-16**) and evaluated for their DPPH radical and antiurease potential.

EXPERIMENTAL

^{1}H NMR spectra were recorded on AVANCE AV 300 M Hz spectrometers. NMR spectra were recorded by using DMSO as solvent. TMS was used as internal standard. Finingan MAT-311A spectrometer was used for electron impact mass spectra (EI-MS) analysis. CsI was used as an internal standard for mass analysis. Column chromatography was performed on silica gel (E. Merck, type 60, 70-230 mesh). Pre-coated silica gel aluminum plates (Kieselgel 60, 2020 and 0.5 mm thick, E.

Merck, Germany) were used for TLC analysis. UV lamp of wavelength 254 and 365 nm was used to visualize the spot in chromatogram.

General procedure for synthesis of compounds 1-16: Schiff bases were synthesized by refluxing isophthalaldehyde with hydrazine hydrate in 1:2 in ethanol by refluxing for 2-3 h. Reaction completion was monitored by TLC. After completion of reaction the mixture was cooled, concentrated and washed with *n*-hexane to obtain the pure product. This product in second step is further reacted with different aromatic aldehydes/acetophenone (1:2) in refluxing ethanol for 2-3 h. Reaction completion was monitored by TLC. These Schiff bases (**1-16**) were then washed with ethyl alcohol and *n*-hexane to obtain pure products

3,3′-(1E,1′E)-[(2Z,2′E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene)]*bis*(methan-1-yl-1-ylidene)diphenol (1): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 9.7 (s, 2H, OH), 8.8 (s, 1H, H-2), 8.7 (s, 2H, H-2), 8.6 (s, 2H, H-7/7), 8.5 (s, 2H, H-8/8), 8.0 (m, 2H, H-5′/5), 7.7 (m,1H, H-5), 7.5 (s, 2H, H-2/2), 7.3 (d, $J_{4',5'/6',5'}$ = 6 Hz, 4H, H-4/6/4/6), 6.9 (d, $J_{4,5/6,5}$ = 3 Hz, 2H, H-4/6); EI-MS *m/z* (rel. int. %): 527 (M^+, 42), 329 (100), 313 (56), 299 (73), 196 (51).

1-((E)-((E)-(4-Nitrobenzylidene)hydrazono)methyl)-3-((Z)-((E)-(4-nitrobenzylidene)hydrazono)methyl)benzene (2): Yield: 82 %; ^{1}H NMR (DMSO-d_6 ppm): δ 8.9 (d, $J_{4,5/6,5}$ = 3.9 Hz, 2H, H-4/6), 8.8 (s, 1H, H-2), 8.5 (s, 2H, H-7/7′), 8.4 (d, $J_{3',2'/5'/6'}$ = 8.4 Hz, 4H, H-3′/5′/3′/5′), 8.2 (d, $J_{2',3'/6',5'/2'/3'/6',5'}$ = 8.7 Hz, 4H, H-2′/6′/2′/6′), 8.1 (s, 2H, H-8/8′), 7.7 (m, 1H, H-5); EI-MS *m/z* (rel. int. %): 497 (M^+, 42), 299 (95), 283 (74), 196 (41), 154 (100).

5,5'-(1E,1'E)-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene))*bis*(methan-1-yl-1-ylidene)*bis*(2-methoxyphenol) (3): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 9.3 (s, 2H, OH), 8.8 (s, 1H, H-2), 8.7 (s, 2H, H-7/7′), 8.6 (s, 2H, H-8/8′), 8.4 (s, 2H, H-2′/2′), 8.0 (m, 1H, H-5), 7.4 (d, $J_{4,5/6,5}$ = 14.4 Hz, 2H, H-4/6), 7.2 (d, $J_{6',5'/6',5'}$ = 8.1 Hz, 2H, H-6′/6′) 7.1 (d, $J_{5',6'/5',6'}$ = 9 Hz, 2H, H-5′/5′), 3.8 (s, 6H, OCH_3); EI-MS *m/z* (rel. int. %): 430 (M^+, 42), 400 (89), 313 (100), 299 (53), 196 (48).

4,4'-(1E,1'E)-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene))*bis*(methan-1-yl-1-ylidene)dibenzene-1,2,3-triol (4): Yield: 82 %; ^{1}H NMR (DMSO-d_6 ppm): δ 11.2 (s, 2H, OH), 9.7 (s, 4H, OH), 8.8 (s, 2H, H-7/7′), 8.7 (s, 2H, H-8/8′), 8.5 (d, $J_{4,5/6,5}$ = 10.8 Hz, 2H, H-4/6), 8.1 (s, 1H, H-2), 7.7 (m, 1H, H-5), 6.99 (d, $J_{6',5'/6',5'}$ = 8.7 Hz, 2H, H-6′/6′), 6.5 (d, $J_{5',6'/5',6'}$ = 6.6 Hz, 2H, H-5′/5′); EI-MS *m/z* (rel. int. %): 370 (M^+, 42), 299 (100), 287 (25), 154 (74), 126 (38).

1-((E)-((E)-(2,4-Dichlorobenzylidene)hydrazono)-methyl)-3-((Z)-((E)-(2,4-dichloro benzylidene)hydrazono)-methyl)benzene (5): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 8.9 (s, 2H, H-3′/3′), 8.8 (s, 2H, H-7/7′), 8.4 (s, 2H, H-8/8′), 8.2 (d, $J_{5',6'/5',6'}$ = 8.7 Hz, 2H, H-5′/5′), 8.1 (s,1H, H-2), 7.9 (m, 1H, H-5), 7.8 (d, $J_{6',5'/6',5'}$ = 1.8 Hz, 2H, H-6′/6′), 7.6 (d, $J_{4,5/6,5}$ = 8.7 Hz, 2H, H-4/6); EI-MS *m/z* (rel. int. %): 571 (M^+, 42), 374 (89), 343 (100), 196 (37), 154 (61).

4,4'-(1E,1'E)-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene))*bis*(methan-1-yl-1-ylidene)diphenol (6): Yield: 83 %; ^{1}H NMR (DMSO-d_6 ppm): δ 10.0 (s, 2H, OH), 8.8 (d, $J_{4,5/6,5}$ = 4.2 Hz, 2H, H-4/6), 8.7 (s, 1H, H-2), 8.64 (s, 2H, H-7/7′), 8.5 (s, 2H, H-8/8′), 8.0 (d, $J_{2',3'/6',5'/2',3'/6',5'}$ = 7.5 Hz, 4H, H-2′/6′/2′/6′), 7.8 (m, 1H, H-5), 6.9 (d, $J_{3',2'/5',6'/3',2'/5',6'}$ = 9.9 Hz, 4H, H-3′/5′/3′/5′); EI-MS *m/z* (rel. int. %): 370 (M^+, 42), 147 (100), 212 (20), 120 (62), 82 (42), 65 (87).

1-((E)-((E)-(2,4-Dichlorobenzylidene)hydrazono)-methyl)-3-((Z)-((E)-(2,4-dichloro benzylidene)hydrazono)-methyl)benzene (7): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 8.9 (d, $J_{3',4'/3',4'}$ = 10.5 Hz, 2H, H-3′/3′), 8.8 (s, 2H, H-7/7′), 8.8 (s, 2H, H-8/8′), 8.5 (s, 1H, H-2), 8.2 (d, $J_{6',5'/6',5'}$ = 7.5 Hz, 2H, H-6′/6′), 8.05 (d, $J_{4,5/6,5}$ = 5.7 Hz, 2H, H-4/6), 7.9 (m, 2H, H-4′/4′), 7.81 (m, 2H, H-5′/5′), 7.7 (m, 1H, H-5); EI-MS *m/z* (rel. int. %): 541 (M^+, 42), 343 (34), 313 (100), 196 (36), 154 (65).

1-((E)-((E)-(3,4-Dimethoxybenzylidene)hydrazono)-methyl)-3-((Z)-((E)-(3,4-dimethoxy benzylidene)hydrazono)-methyl)benzene (8): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 8.8 (s, 2H, H-7/7′), 8.6 (s, 2H, H-8/8′), 8.4 (s, 1H, H-2), 8.0 (s, 2H, H-2′/2′),7.6 (m, 1H, H-5), 7.5 (d, $J_{5',6'/5',6'}$ = 11.4 Hz, 2H, H-5′/5′), 7.3 (d, $J_{6',5'/6',5'}$ = 7.8 Hz, 2H, H-6′/6′), 7.1 (d, $J_{4,5/6,5}$ = 8.1 Hz, 2H, H-4/6), 3.8 (s, 12H, OCH_3); EI-MS *m/z* (rel. int. %): 511 (M^+, 42), 313 (100), 283 (69), 196 (23), 154 (44).

2,2'-(1E,1'E)-1,1'-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene))*bis*(ethan-1-yl-1-ylidene)diphenol (9): Yield: 80 %; ^{1}H NMR (DMSO-d_6 ppm): δ 9.97 (s, 2H, OH), 8.8 (S, 1H, H-2), 8.0 (S, 2H, H-7/7′), 7.8 (d, $J_{3',4'/6',5'/3',4'}$ = 8.4 Hz, 4H, H-3′/6′/3′/6′), 7.5 (d, $J_{4,5/6,5}$ = 7.2 Hz, 2H, H-4/6),7.2 (m, 1H, H-5), 7.4 (m, 4H, H-4′/5′/4′/5′), 2.3 (s, 6H, CH_3); EI-MS *m/z* (rel. int. %): 526 (M^+, 42), 298 (67), 283 (58), 196 (27), 154 (100).

1-((E)-((E)-(1-(3-Nitrophenyl)ethylidene)hydrazono)-methyl)-3-((Z)-((E)-(1-(3-nitro phenyl)ethylidene)hydrazono)methyl)benzene (10): Yield: 81 %; ^{1}H NMR (DMSO-d_6 ppm): δ 8.8 (s, 2H, H-2′/2′), 8.7 (d, 2H, $J_{4',5'/4',5'}$ = 6.3 Hz, H-4′/4′), 8.4 (s, 2H, H-7/7′), 8.3 (m, 2H-5′/5′), 8.1 (d, $J_{6',5'/6',5'}$ = 8.7 Hz, 2H, H-6′/6′),8.0 (s, 1H, H-2), 7.8 (d, $J_{4,5/6,5}$ = 7.8 Hz, 2H, H-4/6), 7.68 (m, 1H, H-5),2.4 (s, 6H, CH_3); EI-MS *m/z* (rel. int. %): 550 (M^+, 42), 317 (100), 196 (37), 154 (72), 126 (37).

4,4'-(1E,1'E)-1,1'-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene))*bis*(ethan-1-yl-1-ylidene)dibenzene-1,3-diol (11): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 13.5 (s, 2H, H-7/7′) 8.8 (s, 1H, H-2),8.7 (s, 2H, H-3′/3′), 8.5 (m, 1H, H-5), 7.7 (d, $J_{4,5/6,5}$ = 7.5 Hz, 2H, H-4/6), 7.6 (d, $J_{5,6'/5',6'}$ = 7.8 Hz, 4H, H-5′/6′/5′/6′), 2.41 (s, 6H, CH_3); EI-MS *m/z* (rel. int. %): 550 (M^+, 42), 317 (100), 196 (37), 154 (72), 126 (37).

4,4'-(1E,1'E)-1,1'-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diylidene))*bis*(ethan-1-yl-1-ylidene)diphenol (12): Yield: 82 %; ^{1}H NMR (DMSO-d_6 ppm): δ 9.7 (s, 2H, OH), 8.8 (s, 2H, H-7/7′), 8.5 (s, 1H, H-2), 8.0 (m, 1H, H-5), 7.8 (d, 4H, $J_{3',2'/5',6'/3',2'/5',6'}$ = 8.4 Hz, H-3′/5′/3′/5′) 7.7 (d, 4H, $J_{2',3'/6',5'/2',3'/6'/5'}$ = 8.4 Hz, H-2′/6′/2′/6′), 6.8 (d, 2H, $J_{4,5/6,5}$ = 8.4 Hz, H-4/6), 2.9 (s, 6H, H-CH_3); EI-MS *m/z* (rel. int. %): 511 (M^+, 42), 313 (100), 283 (69), 196 (23), 154 (44).

1-((E)-((E)-(1-(4-(Piperidin-4-yl)phenyl)ethylidene)-hydrazono)methyl)-3-((Z)-((E)-(1-(4-(piperidin-4-yl)-

phenyl)ethylidene)hydrazono)methyl)benzene (13): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 8.8 (s, 1H, H-2), 8.5 (s, 2H, H-7/7′), 8.0 (s, 2H, NH), 7.8 (d, $J_{4,5/6,5}$ = 7.5 Hz, 2H, H-4/6), 7.7 (d, $J_{2',3'/6',5'/2',3'/6',5'}$ = 8.7 Hz, 4H, H-2′/6′/2′/6′), 7.6 (m, 1H, H-5), 6.9 (d, $J_{3',2'/5',6'/3',2'/5',6'}$ = 8.4 Hz, 4H, H-3′/5′/3′/5′), 2.3 (s, 6H, CH_3), 1.5 (s, 16H, H-pipridno); EI-MS *m/z* (rel. int. %): 511 (M^+, 42), 313 (100), 283 (69), 196 (23), 154 (44).

4-((E)-((Z)-(3-((E)-((Z)-((2,4 Dihydroxyphenyl)(phenyl)-methylene)hydrazono)methyl) benzylidene)hydrazono)-(phenyl)methyl)benzene-1,3-diol (14): Yield: 84 %; ^{1}H NMR (DMSO-d_6 ppm): δ 12.2 (s, 2H, OH), 10.7 (s, 2H, OH), 7.6 (s, 2H, H-3′/3′), 7.5 (d, $J_{,2'/3'/,6',5'/2',3'/6',5'}$ = 2.4 Hz, 4H, H-2′/6′/2′/6′), 7.4 (m, 4H, H-3′/5′/3′/5′), 7.41 (s, 1H, H-2), 7.37 (d, 2H, $J_{5',6'/5',6'}$ = 8.4 Hz, H-5′/5′), 7.2 (d, $J_{6',5'/5'/6',5'}$ = 8.4 Hz, 2H, H-6′/6′), 7.1 (m, 2H, H-4′/4′), 7.1 (m, 2H, H-4′/4′), 6.9 (d, $J_{4,5/6,5}$ = 2.4 Hz, 2H, H-4/6), 6.35 (m, 1H, H-5); EI-MS *m/z* (rel. int. %): 511 (M^+, 42), 313 (100), 283 (69), 196 (23), 154 (44).

1-((E)-((E)-(Naphthalen-2-ylmethylene)hydrazono)-methyl)-3-((Z)-((E)-(naphthalen-2-ylmethylene)hydrazono)methyl)benzene (15): Yield: 82 %; 1H NMR (DMSO-d_6 ppm): δ 8.4 (s, 4H, H-7/7′/2′/2′), 8.3 (s, 2H, H-8/8′/2), 8.2 (s, 1H, H-2), 8.1 (d, *J* = 6 Hz, 2H, H-4/6), 7.8 (m, 1H, H-5), 8.0 (d, *J* = 6.5 Hz, 6H, H-3′/3′/8′/8′/7′/7′), 7.9 (d, $J_{5,6}$ = 7 Hz, 4H, H-4′/4′/6′/6′), 7.8 (m, 2H, H-5′/5′); EI-MS *m/z* (rel. int. %): 438 (M^+, 54), 416 (60), 365 (69), 289 (35), 156 (42).

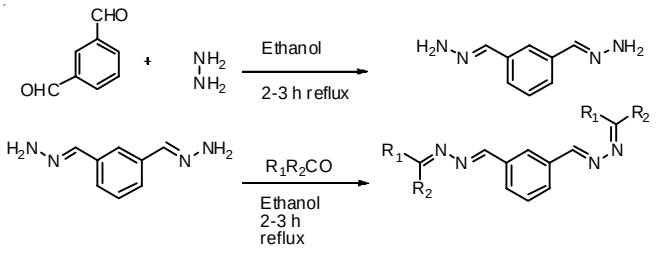
2,2'-(1E,1'E)-((2Z,2'E)-(1,3-Phenylene*bis*(methan-1-yl-1-ylidene))*bis*(hydrazine-2,1-diyli dene))*bis*(methan-1-yl-1-ylidene)dibenzene-1,3-diol (16): Yield: 83 %; ^{1}H NMR (DMSO-d_6 ppm): δ 11.2 (s, 2H, OH), 10.6 (s, 2H, OH), 7.6 (s, 2H, H-3′/3′), 7.5 (d, 4H, $J_{,2'/3'/,6',5'/2',3'/6',5'}$ = 2.4 Hz, 4H, H-2′/6′/2′/6′), 7.43 (m, 4H, H-3′/5′/3′/5′), 7.41 (s, 1H, H-2), 7.37 (d, $J_{5',6'/5',6'}$ = 8.4 Hz, 2H, H-5′/5′), 7.2 (d, $J_{6',5'/5'/6',5'}$ = 8.4 Hz, 2H, H-6′/6′), 7.1 (m, 2H, H-4′/4′), 6.9 (d, $J_{4,5/6,5}$ = 2.4 Hz, 2H, H-4/6), 6.35 (m, 1H, H-5), 1.8 (s, 6H, CH_3); EI-MS *m/z* (rel. int. %): 541 (M^+, 42), 413 (100), 383 (69), 186 (23), 144 (44).

Urease inhibition assay: Khan *et al.* [19] procedure was followed for determining urease inhibition based on weatherburn procedure [17]. 5 µL solution of sample was mixed with 25 µL of urease solution; mixture was incubated for 15 min at 30 °C. In 96 well plates the aliquots were mixed with sample mixture of urea (100 mM) in buffer (40 µL) by re incubation. To wells 70 µL of alkali reagent (0.5 % w/v sodium hydroxide NaOH and 0.1 % sodium hypochlorate) and 50 µL of phenol reagent (1 % w/v phenol and 0.005 % w/v sodium nitroprusside) were mixed. Against the blank increase in absorbance was measured after 50 min at 630 nm. At pH 8.2 the reaction mixture volume were 200 µL. Thiourea were used as positive control. With help of formula the percentage inhibition were measured. Measurement were done triplicate for accuracy.

Antioxidant assay: Zonia *et al.* [20] method was used for measuring DPPH radical scavenging. In the ethanolic solution of DPPH (1 mmol/L, 0.5 mL) 5mL ethanol sample was added. At 30 °C the reaction mixtures were incubated for 0.5 h using UV-3000 spectrophotometer (Hitachi, Japan) the absorbance was measured against ethanol. Using DPPH and same amount of ethanol was used and measured on daily basis. DPPH solution were prepared freshly and stored in aluminium foiled flasks and measurements were done at dark at 4 °C. All the measurement were done triplicate.

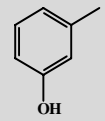
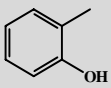
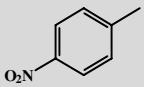
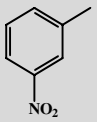
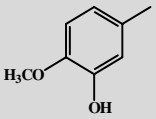
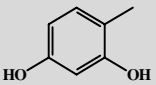
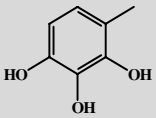
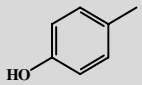
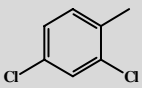
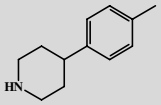
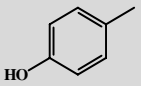
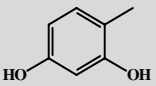
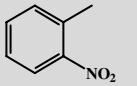
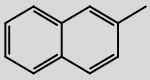
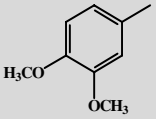
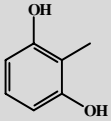
RESULTS AND DISCUSSION

Schiff bases were synthesized by refluxing isophthalaldehyde with hydrazine hydrate in 1:2 in ethanol by refluxing for 2-3 h. Reaction completion was monitored by TLC. After completion of reaction the mixture was cooled, concentrated and washed with *n*-hexane to obtain the pure product. This product in second step is further reacted with different aromatic aldehydes/acetophenone (1:2) in refluxing ethanol for 2-3 h. Reaction completion was monitored by TLC. These Schiff bases (**1-16**) were then washed with ethyl alcohol and *n*-hexane to obtain pure products (**Scheme-I**, Table-1).



Scheme-I: Synthesis of *bis*-Schiff bases of isopthalaldehyde **1-16**

TABLE-1
SYNTHESIZED *BIS*-SCHIFF BASE ANALOGS OF ISOPTHALALDEHYDE (**1-16**)

Comp. No.	R_1	R_2	Comp. No.	R_1	R_2
1		H	**9**		CH_3
2		H	**10**		CH_3
3		H	**11**		CH_3
4		H	**12**		CH_3
5		H	**13**		CH_3
6		H	**14**		C_6H_6
7		H	**15**		H
8		H	**16**		Me

Urease inhibitory studies: Compounds **1-16** were subjected for urease inhibition. Out of sixteen, only three compounds showed potent inhibition. Compounds **4, 15** and **16** showed an excellent urease inhibition with IC_{50} values 18.3 ± 0.8, 13.8 ± 0.4 and 13.9 ± 0.2 μM respectively much better than the standard inhibitor thiourea with IC_{50} value 21.8 ± 0.11 μM (Table-2). Compounds **4,** 2,3,4-trihydroxy analog, **15** naphthyl analog and **16** 2,6-dihydroxy analog exhibit potent inhibition. Compound **4** and **16** have hydroxyl group on phenyl ring. Activity of compounds **4** and **16** might be attributed to hydroxyl groups. This hydroxyl group might be involved in hydrogen bonding with nickel of enzyme. Compound **15** have naphthalene moiety, activity of compound might be due to arene interaction. All other remaining compounds were found to be inactive.

TABLE-2
UREASE ACTIVITY OF *BIS*-SCHIFF BASES (**1-16**)

Compounds	IC_{50} ± SEM[a] (μ/M)	Compounds	IC_{50} ± SEM[a] (μ/M)
1	N.A	**9**	N.A
2	N.A	**10**	N.A
3	N.A	**11**	N.A
4	18.3 ± 0.8	**12**	N.A
5	N.A	**13**	N.A
6	N.A	**14**	N.A
7	N.A	**15**	13.8 ± 0.4
8	N.A	**16**	13.9 ± 0.2
Thiourea[c]	21.8 ± .0.11	–	–

SEM[a] is the standard error of the mean, NA[b] Not active, Thiourea[c] is standard inhibitor for urease inhibition

Antioxidant activity: Compounds **1-16** were subjected for DPPH radical scavenging activity. Out of sixteen, only three compounds were active. Compounds **4, 15** and **16** showed excellent to good DPPH radical scavenging activity with IC_{50} values 19.663 ± 0.8, 50.64 ± 4.7 and 92.821 ± 1.38 μM, respectively comparable to the standard inhibitor gallic acid with IC_{50} value 23.436 ± 0.43 μM (Table-3). Compounds **4,** 2,3,4-trihydroxy analog, **15** naphthyl analog and **16** 2,6-dihydroxy analog were active. Compound **4** and **16** have hydroxyl groups on phenyl ring, activity might be due to these hydroxyl groups which can stabilize the free radicals. Compound **15** has naphthalene moiety, activity might due to areneinteraction. Rests of the compound were not active against DPPH radical scavenging.

TABLE-3
ANTIOXIDANT ACTIVITY OF *BIS*-SCHIFF BASES (**1-16**)

Compounds	IC_{50} ± SEM[a] (μ/M)	Compounds	IC_{50} ± SEM[a] (μ/M)
1	N.A	**9**	N.A
2	N.A	**10**	N.A
3	N.A	**11**	N.A
4	19.663 ± 0.8	**12**	N.A
5	N.A	**13**	N.A
6	N.A	**14**	N.A
7	N.A	**15**	50.64 ± 4.7
8	N.A	**16**	92.821 ± 1.38
Gallic acid	23.436 ± 0.43	–	–

SEM[a] is the standard error of the mean, NA[b] Not active, Gallic acid is standard inhibitor for antioxidant inhibition

Conclusion

In present work analogs **1-16** were synthesized and subjected to antiurease inhibition and antioxidant potential. Three compounds showed potent urease inhibition and moderate antioxidant activity. Analogs **4**, **15** and **16** with IC_{50} values 18.3 ± 0.8, 13.8 ± 0.4 and 13.9 ± 0.2 μM respectively were found potent antiurease when compared with standard thiourea. The same analogs also showed antioxidant potential with IC_{50} values, 19.663 ± 0.8, 50.64 ± 4.7 and 92.821 ± 1.38 μM, respectively as compared with standard quercitin.

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