Short Communication

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Biological Activity of 2,3-Di (Quinolyl-2)-6-Methyl Quinoxaline

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The biological activities of the nitrogen containing conjugated heterocyclic compounds are considerably used pharmaceutically as antiulcer, antimalarial, tubercluocidal and sedatives besides their uses as dyes in the textile industries, pesticides, stabilizers and inhibitors etc (Cheeseman 1963; Cheeseman and Werstuick 1978).

Thus quinoxalines likewise are also considered to have significant biological activities. Their reactions as well as their pharmacological actions, continue to stimulate many investigations for example 2-methylquinoxaline N,N-dioxides substituted in the 3-position (e.g. with amide, amidino and ester groups) are potent bacteriocides (Ley *et al* 1969, Ley *et al* 1970; Kasubick *et al* 1972). Antibiotics of the triostin and quinomycin series, isolated from the cultures of *Streptomyces aureus,* have been shown by degradative study to contain a quinoxaline-2-carboxylic acid residue and (Cheeseman 1963; Cheeseman and Werstuick 1968). Keeping this in view, the present research was conducted to evaluate its biological activities.

2,3-Di(quinolyl-2)-6-methyl quinoxaline was synthesized by condensation followed by ring closure reaction when 1,2-di (quinolyl-2)-1,2 ethanedione was treated with methyl and substituted *o*-phenylenediamine. The structure of the newly synthesized compound has been determined and characterised by ultraviolet, infrared, nuclear magnetic resonance and mass spectral data and is confirmed by elemental analysis. The biological screening of the compound was done using different standard techniques to determine its antibacterial, antifungal and cytotoxic activities.

Melting points were measured in an open capillaries with an electrothermal I A 9100 digital melting point apparatus. Infrared spectra were recorded on a Phillips PU 9714 spectrophotometer using infrared grade potassium bromide. Nuclear magnetic resonance (^1H) spectra were determined on "Varian 200MHz Gemini", "Bruker AC-200MHz FT-NMR" and "Bruker AM-500MHz FT-NMR" spectrometers in deuteriochloroform and are reported in parts per million downfield from tetramethyl silane (TMS) as the internal standard (δ scale). Mass spectra were obtained with E1 MAT 312, Varian MAT 111, Varian MAT 112 Hewlett Packard GC/MS 5890 spectrometer. Chemical analysis was performed in Austria and satisfactory results were obtained.

For the purpose of column chromatography, silica gel 60 (70-230mesh) from E. Merck AG was used. Eastman Kodak chromogram 13181 silica gel sheets with fluorescent indicator were used f ,2-di (quinoly1-2)-1,2 ethanedione was prepared by the oxidation of 1,2-di(quinoly1-2)-1,2-ethenediol which was synthesized by the condensation reaction of the quinoline-2-carboxaldehyde. Quinoline-2-carboxaldehyde in turn was prepared according to the literature procedure by the reaction of SeO_2 with the starting material i.e. 2-methyl quinoline (Kaplan 1941). The obtained heterocyclic carboxaldehyde was found to have properties similar to that given in the literature.

SeO_2 for the reaction was freshly prepared just before use by the method given by researchers (Blatt 1966). All crude reaction products were examined by thin layer chromatography with chloroform as developing solvent and compared with the starting material and reagents to follow the progress of the reactions.

The IR, NMR, MS and analytical data along with the purification procedures are given in each experiment separately.

Table 1

Antibacterial activity

Name of Bacteria	Clinical implications	zone inhibition (mm)	reference drug/zone of inhibition (mm)
Pseudomonas typhi	Typhoid fever. Salmonella food poisoning, localized infections etc.	7	Amoxicillin $(H_2O)_3$ /9 Ampicillin $(H_2O)_3$/12
Shigella boydii	Inflammation of GIT bacterial dysentry	10	Ampicillin $(H_2O)_3$/11 Ampicillin $(H_2O)_3$/11
Streptococcus pyogenes	Acute rheumatic fever, scarlet fever, sore throat, septic wound etc.	6	Amoxicillin $(H_2O)_3$/9 Ampicillin $(H_2O)_3$/8

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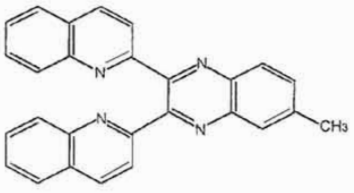
Table 2
Antifungal activity

Name of Fungi	Sample	Control	(mm) inhibition %	Reference	MIC µg/ml
Human pathogens					
Aspergillus niger	25	50	50.00	Miconazole	100
				Ketoconazole	100
Pseudallescheria boydii	45	80	43.70	Miconazole	100
				Ketoconazole	100
Trichophyton schoenleinii	50	80	37.00	Miconazole	100
				Ketoconazole	100
Animal pathogens					
Microsporum canis	20	56	64.20	Miconazole	100
				Ketoconazole	100
Trycophyton simii	75	80	6.25	Miconazole	100
				Ketoconazole	100
Plant pathogens					
Fusarium solanai var lycopersici (tomato)	45	80	43.70	Benlate	100
Fusarium oxysporum var lycopersici (tomato)	30	70	5.70	Benlate	100

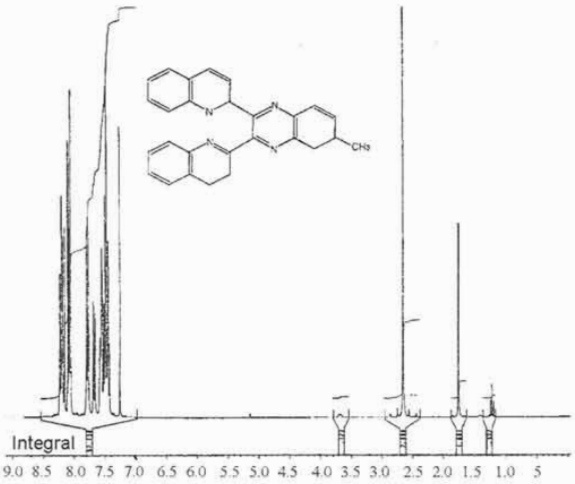
2,3-di (quinolyl-2)-6-methyl quinoxaline. To a solution of 2.0 mmole (0.2440gm) of 4-methyl 1,2-phenylenediamine in 20ml acetic acid, 2.0 mmoles of 1,2-di (quinolyl-2)-1,2 ethanedione (Kaban & Ansar 2002) was added and heated under reflux in an oil bath for an hour. The reaction mixture was cooled and 100mL of water was added and a beige colored emulsion was formed which was destroyed by adding 20% sodium hydroxide solution and the resulting precipitates were filtered. The impure product was then dissolved in alcohol & heated with active charcoal, filtered and reprecipitated with water and crystallized using ethyl alcohol yielding 0.51 gm (69%), m.p 2.334°C.

IR (Potassium bromide): 3080-3000 (aromatic, = C-H) 3000-2920 (methyl C-H) cm^{-1}. ^1H nmr (chloroform-D) δ: 2.64 (s, CH_3, 3H) 7.41-8.22 (m, aromatic, 15H). ms: m/z (relative intensity) 400 (M+2,2), 399(M+1,22), 398(M^+, 91),397 (M-1, 100), 199(22), 154(2), 128(10), 101(2). UV (chloroform): λmax 259.6, 399.2 nm.

Elemental analysis: Calculated for $C_{27}H_{18}N_4$; C=81.38, H=4.55, N=14.06 found C=81.30, H=4.89, N=14.19.



2,3-Di (quinolyl-2)-6-methylquinoxaline



^1H NMR spectrum of 2,3-di (quinolyl-2)-6-methylquinoxaline

Biological activity. Antimicrobial activities i.e. bactericidal as well as antifungal activities of 2,3-di (quinolyl-2)-6-methyl quinoxaline were investigated according to the standard procedures (Rahman *et al* 1999). The antibacterial activity (Table 1) was determined by agar well diffusion protocol whereas the fungicidal properties (Table 2) of the compound were screened against various pathogenic fungi using agar tube dilution method measuring diameter of zones(mm) showing inhibition and growth inhibition and calculated with reference to positive control. *In vivo* lethality to shrimp larvae was used to determine the cytotoxicity of the compound using the standard brine shrimp cytotoxicity bioassay procedure.

Cytotoxic activity. The brine shrimp lethality bioassay showed that the compound is moderately active and the LD_{50} is found to be 639.0581 µg/ml i.e. less than 1000 µg/ml.

Biological activity of the compound was determined by using standard procedures. It's bactericidal activity was found to be significant against *Shigella boydii* whereas it showed a moderate activity against *Pseudomonas typhi* and *Streptococcus pyogenes*. However it showed significant and moderate activities against many human, animal and plant pathogens and most significant against *Aspergillus niger, Microsporum canis* and *Fusarium solanai var lycopersici* (tomato). The cytotoxic activity of the compound was determined by brine shrimp lethality test which revealed its LD_{50} value to be 639.0581 µg/ml i.e. less than 1000 µg/ml enough to prove its moderate cytototoxic activity.

References

Blatt A H 1966 *Organic Synthesis*, John Wiley and Sons, Inc 12th ed, New York, Collective Vol 2 p. 510

Cheeseman G W H 1963 *Recent Advances in Quinoxaline Chemistry in Advances in Heterocyclic Chemistry*, A.R.Katritzky, ed, Academic Press, New York, Vol. 2 p.203-243

Cheeseman G W H, Werstiuk E S G 1978 *Quinoxaline Chemistry in Advances in Heterocyclic Chemistry,* eds A.R. Katritzky and A.J.Boulton, Academic press, News York, Vol 22 p.367-431.

Kaban Seniz, Ansar Nasir 2002 Synthesis of 2,3 di (quinol-8-yl) 6-methyl quinoxaline, *Pak. J. Sci. Ind. Res.* **45** (4) 243-245

Kaplan H 1941 *J Am Chem Soc* **63** 2655

Kasubick R V, Robertson R L, Demande Fr 1973, 2 132 378 (1972) *Chemical Abstr.*, **78** 159673

Ley K, Eholzer U, Metzger K G, Fritsche D 1969 South African Patent, 6806, 099, Chem Abstr **71** 91528.

Ley K Eholzer U, Metzger K G, Fritsche D 1970 South African Patent, 6906, 094, Chem Abstr **72** 3508.

Rahman Atta-ur, Choudhary M I, Thomson William J 1999 *Manual of Bioassay Techniques for Natural Product Research.* Harward Academic Press, Amsterdam pp 82-84.