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Short communication

Synthesis and evaluation of new quinazolone derivatives of nalidixic acid as potential antibacterial and antifungal agents

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Abstract

In continuation of our work on synthesis of biheterocycles carrying the biodynamic heterocyclic systems at position 3, a series of new nalidixic acid derivatives having quinazolones moiety were synthesised to achieve enhanced biological activity and wide spectrum of activity. Nalidixic Acid was first converted into its acid chloride using thionyl chloride as an acylating agent at laboratory temperature. Later it was converted to methyl ester. Nalidixoyl chloride formed vigorously reacts with methanol to give a methyl ester of nalidixic acid. The ester on addition of hydrazine hydrate furnished nalidixic acid hydrazide. Appropriate anthranilic acid was refluxed with acetic anhydride to form Benzoxazine/Acetanthranil. 5-iodo-derivative of anthranilic acid was prepared and also utilised to obtain 6-iodo-Benzoxazine/Acetanthranil. Also, 6-nitro-Benzoxazine/Acetanthranil was obtained by nitration of acetanthranil using conc. H_2SO_4 and fuming HNO_3. Equimolar proportions of the appropriate synthesised acetanthranils and nalidixic acid hydrazide in the presence of ethanol were refluxed to synthesise quinazolones. Elemental analysis and IR spectra confirmed nalidixic acid hydrazide formation. The structures of the compounds obtained have been established on the basis of Spectral (IR, ^{1}H NMR and mass) data. The current study also involves in vitro antimicrobial screening (using Agar dilution and Punch well diffusion method) of synthesised quinazolone derivatives bearing nalidixic acid moiety on randomly collected microbial strains. The derivatives **Ga (NAH)**, **Gb (QN)** and **Gd (NiQNA)** showed marked inhibitory activity against enteric pathogen like *Aeromonas hydrophila*, a causative agent of diarrhoea in both children as well as adults. Among the respiratory pathogens included in study, derivative **Gd (NiQNA)** was found to be active against *Streptococcus pyogenes*. No significant inhibitory activity was seen by any of synthesised derivatives against *Coagulase negative Staphylococcus.* Derivative **Ga (NAH)** was found to show very high activity against the *Candida* colonies and derivative **Gd (NiQNA)** was also found to exhibit inhibitory activity against *Candida albicans*; a normal flora of the human body which plays an important role in causing opportunistic infections in immunocompromised hosts. *Proteus vulgaris,* a gram-negative bacteria included in our study was found to be inhibited by derivative **Gb (QN)**.

Keywords: Proteus vulgaris; *Streptococcus pyogenes*; *Candida albicans*; *Aeromonas hydrophila*; *Coagulase negative Staphylococcus*; Nalidixic acid hydrazide (NAH); Quinazolinone derivative of nalidixic acid (QN); Iodo-quinazolinone derivative of nalidixic acid (IQN); Nitro-quinazolinone derivatives of nalidixic acid (NIQNA); Urinary tract infections

1. Introduction

Nalidixic acid (1,8-naphthyridine derivative) was introduced for the treatment of urinary tract infections in 1963. It failed to achieve adequate concentrations in the plasma or tissues for the treatment of systemic infections following oral or potential administration but got concentrated in the urine, where they could be effective for eradicating urinary tract infections.

Structure activity relationship (SAR) of compounds based on nalidixic acid have led to a large group of synthetic antibacterial agents collectively known as the quinolones [1]. Resistance was found to emerge rapidly, even while on therapy. These agents inhibit DNA synthesis during bacterial replication. This effect may result from interference with DNA gyrase activity [2].

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Nalidixic acid is known to be effective against indole positive *Proteus* in urinary tract infections. Failure observed in patients may be due to reinfection from the prostate gland [2].

Nalidixic acid is particularly active against the majority of gram-negative organisms that infect urinary tract especially *E. coli*. It is often effective against other coliform bacteria such as *Klebsiella* and *Enterobacter aerogenosa*. *Brucella* species and some strains of *Salmonella* and *Shigella* are also sensitive. However, most *Pseudomonas sp.* are resistant to nalidixic acid. This drug is ineffective against gram-positive bacteria including *Staphylococcus* and *Enterococcus fecalis* (formerly, *Streptococcus fecalis*). Studies have indicated that nalidixic acid is effective against 99% strains *of E. coli*, 98% of *P. mirabilis*, 92% of *Klebsiella* and *Enterobacter* and 80% of other coliform species [3].

1,8-Naphthyridines possess diverse biological activities and there are numerous reports that highlight their chemistry and use. They are also found to possess interesting antibacterial activity.

Quinazolones have been reported to have wide-ranging biological activities including antitubercular [4], fungicidal [5], antimalarial [6], anticancer and anti-HIV agents [6], antiviral [7] and antibacterial activity [8].

Structural variations bring about new physical and biological properties. The molecular manipulation of a promising lead compound is still a major line of approach for the discovery of new drugs. Molecular manipulation involves the efforts to combine separate groups having similar activity in one compound by eliminating or substituting new moiety to a parent lead compound.

In the present work, antimicrobial activity associated with both quinazolines and nalidixic acid moieties prompted us to synthesise some nalidixic acid derivatives carrying the biodynamic heterocyclic systems (quinazolones) at position-3 with an objective to obtain biheterocycles of enhanced biological activities. The compounds so obtained were screened for antifungal and antibacterial activities.

2. Antimicrobial screening [9]

All the synthesised quinazolone derivatives of nalidixic acid were screened for antimicrobial activity using the following methods.

2.1. Agar dilution method

Reference preparations of quinazolone derivatives of nalidixic acid were prepared in diluent DMSO (Dimethyl Sulphoxide) at 10 × concentrations, starting with a stock concentration of 30 and 600 μg ml^{-1} and covered a full range of 30–600 μg ml^{-1}. This method was convenient and economical on pipette use. MHA (Muller Hinton Agar) was used as a medium for bacterial screening [9] whereas SDA (Sabouraud's Dextrose Agar) Broth was used for fungal screening. Study derivatives were incorporated into liquefied agar medium at 45–50 °C, mixed and poured in petridishes and allowed to solidify. A series of petriplates were prepared with increasing concentrations of the drug. With the help of inoculating applicator, as many as 14 different strains were spot inoculated on each plate. After overnight incubation, minimum inhibitory concentration (MIC) end point was determined by placing plate against a dark background and observing the lowest concentration of derivatives inhibiting visible growth. The MIC of each derivative was recorded in μg ml^{-1}. Wherever two or more colonies persisted beyond the end point or growth was present in higher concentration and not in lower concentration, the test was repeated.

2.2. The punch well/cup plate diffusion method

In this technique petridishes of agar medium plate were prepared by pouring melted agar inoculated with a variety of microorganisms. After the agar settled, cups were made in the agar petridishes. Test solutions were prepared using the following concentrations based on results obtained in agar dilution method.

Derivative **Ga** (NAH) in concentrations of 30, 100, 200, 300 μg ml^{-1}, derivative **Gb** (QN) in concentrations of 50, 100, 150, 200, 250, 300 μg ml^{-1}, derivative **Gc** (IQN) in concentrations of 20, 40, 60, 80, 100 μg ml^{-1}, derivative **Gd** (NiQNA) in concentrations of 50 100, 150, 200, 250, 300 μg ml^{-1} were made.

Isolates of clinically obtained strains used are mentioned below:

- *Aeromonas hydrophila*, *Escherichia coli*, *Klebsiella aerogenes*, *Enterococci* (COPS) *Coagulase positive Staphylococcus aureus* (CONS) *Coagulase negative Staphylococcus*, *Streptococcus pyogenes*, *Candida albicans*, *Salmonella typhimurium*, *Shigella flexneri*, *Vibrio cholerae*, *Acinetobacter* sp., *Proteus vulgaris*.

All the derivatives were compared with Ampicillin, nalidixic acid and Fluconazole for antibacterial and antifungal activity.

3. Results

The derivatives showed marked activity against *A. hydrophila*, *C. negative Staphylococcus*, *S. pyogenes*, *C. albicans*, and *P. vulgaris* with agar dilution method (see Tables 1–2). Only those strains, which were found to be susceptible to the test derivatives, have been utilised for cup plate diffusion method and the results are included in Tables 3–5.

Derivative **Ga (NAH)**, **Gb (QN)** and **Gd (NiQNA)** are found to have good inhibitory effect at concentrations of 300, 200 and 200 μg ml^{-1}, respectively; on one of the enteric pathogen *A. hydrophila,* the causative agent of diarrhoea in both adults and children (see Table 3).

Among the respiratory pathogens included in our study, derivative **Gd (NiQNA)** is found to have high activity against *S. pyogenes* at 300 μg ml^{-1} (see Table 3).

Table 1
Agar dilution method results of synthesised new derivatives of nalidixic acid

Strain used	Derivative **(NAH)** Ga				Ampicillin	Derivative **(QN)** Gb					
	Concentrations in μg ml^{-1}					Concentrations in μg ml^{-1}					
	30	100	200	300	10	50	100	150	200	250	300
A. hydrophila	−	+	−	+	+	+	+	+	+	+	+
E. coli	−	−	−	−	+	−	−	−	−	−	−
K. aerogenes	−	−	−	−	+	−	−	−	−	−	−
Enterococi	−	−	−	−	+	−	−	−	−	−	−
S. aureus	−	−	−	−	+	−	−	−	−	−	−
C. negative Staphylococcus	−	−	+	−	+	−	−	−	−	−	−
Streptococcus	−	+	+	+	+	−	−	−	−	−	−
C. albicans	−	−	+	+	+	−	−	−	−	−	−
S. typhimurium	−	−	−	−	−	−	−	−	−	−	−
S. flexneri	−	−	−	−	−	−	−	−	−	−	−
V. cholerae	−	−	−	−	−	−	−	−	−	−	−
Acenitobacter sp.	−	−	−	−	−	−	−	−	−	−	−
P. vulgaris	−	−	−	−	−	−	−	+	+	−	−

(−) Represents no inhibition of growth/resistant; (+) represents inhibition of growth/susceptible.

Table 2
Agar dilution method results of synthesised new derivatives of nalidixic acid

Strain used	Derivative **(IQN)** Gc concentrations in μg ml^{-1}					Ampicillin	Derivative **(NIQNA)** Gd concentrations in μg ml^{-1}					
	20	40	60	80	100	10	50	100	150	200	250	300
A. hydrophila	−	−	−	−	−	+	+	+	+	+	+	+
E. coli	−	−	−	−	−	+	−	−	−	−	−	−
Klebsiella aerogenes	−	−	−	−	−	+	−	−	−	−	−	−
Enterococi	−	−	−	−	−	+	−	−	−	−	−	−
Coagulase positive Staphylococcus aureus	−	−	−	−	−	+	−	−	−	−	−	−
C. negative Staphylococcus	−	−	−	−	−	+	−	−	−	−	−	−
Streptococcus	−	−	−	−	−	+	+	+	+	+	+	+
C. albicans	−	+	+	+	−	+	+	+	+	+	+	+
S. typhimurium	−	−	−	−	−	+	−	−	−	−	−	−
S. flexneri	−	−	−	−	−	+	−	−	−	−	−	−
V. cholerae	−	−	−	−	−	+	−	−	−	−	−	−
Acenitobacter sp.	−	−	−	−	−	+	−	−	−	−	−	−
P. vulgaris	−	−	−	−	−	+	−	−	−	−	−	−

Table 3
Punch well/cup plate diffusion method results of synthesised new derivatives of nalidixic acid

Aeromonas hydrophila Inhibition zone (in mm) against												*Streptococcus pyogenes* Inhibition zone (in mm) against											
(NAH) **Ga**		(QN) **Gb**		(IQN) **Gc**		(NiQNA) **Gd**		Nalidixic acid		Standard Ampicillin		(NAH) **Ga**		(QN) **Gb**		(IQN) **Gc**		(NiQNA) **Gd**		Nalidixic acid		Standard Ampicillin	
Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm	Conc. μg / ml	mm
30	9	50	10	20	-	50	10	30	-	10	14	30	-	50	-	20	-	50	10	30	-	10	30
100	6	100	10	40	-	100	10					100	9	100	-	40	-	100	11				
200	8	150	10	60	-	150	10					200	10	150	-	60	-	150	11				
300	14	200	14	80	-	200	14					300	10	200	-	80	-	200	13				
		250	10	100	-	250	10							250	-	100	-	250	14				
		300	9			300	10							300	-			300	30				

(-) represents no inhibition of growth / Resistant

Table 4
Punch well/cup plate diffusion method results of synthesised new derivatives of nalidixic acid

Coagulase negative Staphylococcus Inhibition zone (in mm) against												*Candida albicans* Inhibition zone (in mm) against											
(NAH) **Ga**		(QN) **Gb**		(IQN) **Gc**		(NiQNA) **Gd**		Nalidixic acid		Standard Ampicillin		(NAH) **Ga**		(QN) **Gb**		(IQN) **Gc**		(NiQNA) **Gd**		Nalidixic acid		Standard Fluconazole	
Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm
30	-	50	-	20	-	50	-	30	-	10	14	30	16	50	-	20	-	50	10	30	-	25	32
100	-	100	-	40	-	100	-					100	10	100	-	40	11	100	11				
200	-	150	-	60	-	150	-					200	10	150	-	60	12	150	11				
300	7	200	-	80	-	200	-					300	30	200	-	80	-	200	12				
		250	-	100	-	250	-							250	-	100	-	250	10				
		300	-			300	-							300	-			300	13				

(-) represents no inhibition of growth / Resistant

Table 5
Punch well/cup plate diffusion method results of synthesised new derivatives of nalidixic acid

Proteus vulgaris Inhibition zone (in mm) against											
(NAH) **Ga**		(QN) **Gb**		(IQN) **Gc**		(NiQNA) **Gd**		Nalidixic acid		Standard Ampicillin	
Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm	Conc. µg / ml	mm
30	-	50	-	20	-	50	-	30	18	10	14
100	-	100	-	40	-	100	-				
200	-	150	14	60	-	150	-				
300	-	200	7	80	-	200	-				
		250	-	100	-	250	-				
		300	-			300					

(-) represents no inhibition of growth / Resistant

Derivative **Ga (NAH)** shows inhibition zone of 7 mm at 300 µg ml^{-1} against *C. negative Staphylococcus* that is insufficient to inhibit the growth appreciably (see Table 4).

Derivative **Ga (NAH)** shows very high activity at 300 µg ml^{-1} comparable to standard drug Fluconazole (25 µg ml^{-1}) against *C. albicans*. It also has moderate activity at concentrations of 30, 100, 200 µg ml^{-1}. Derivatives **Gc (IQN)** and **Gd (NiQNA)** are found to have moderate activity against *C. albicans* at almost all concentrations studied (see Table 4).

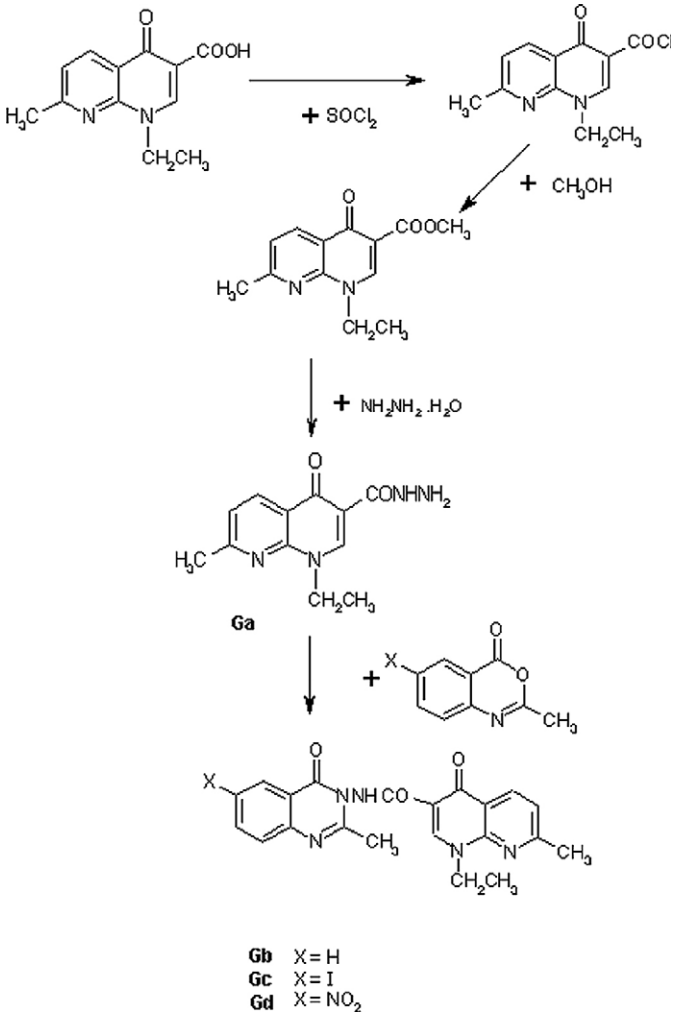
Gram negative bacteria *P. vulgaris* is inhibited by derivative **Gb (QN)** at 150 µg ml (see Table 5).

4. Experimental section

4.1. General

Nalidixic acid was obtained as a gift sample from CFL Pharma Limited, Curti, Ponda, Goa. All the melting points are uncorrected and were recorded in a Toshniwal melting point apparatus. TLC was carried out using Silica gel G procured from Merck. Solvent used was Chloroform: Acetone (4:1). Nitrogen % estimation was carried out for nalidixic acid Hydrazide by RSIC, CDRI, Lucknow. UV spectra were recorded on UV–Visible spectrometer (Shimadzu 160 IPC). IR

spectra of the compounds were recorded using KBr pellet method on an FTIR-8300 Shimadzu spectrophotometer. All the chemicals and reagents that where used were obtained in high quality. Mass spectra were provided by RSIC, IIT Madras, and were acquired on a MASSPEC system (msw/9629). H^1NMR was done on Instrument Joel Model: GSX 400.



Characterisation data of compounds **Ga**–**Gd**

Compound	Molecular weight	Nitrogen % found (Calc.)	Melting Point (° C)	R_f	Molecular formula
Derivative Ga (NAH)	246	22.9 (22.7)	218	0.46	$C_{12}H_{14}N_4O_2$
Derivative Gb (QN)	375		180	0.36	$C_{21}H_{19}N_5O_2$
Derivative Gc (IQN)	501		184	0.37	$C_{21}H_{19}N_5O_2I$
Derivative Gd (NiQNA)	521		271	0.44	$C_{21}H_{18}N_6O_4$

4.2. Preparation of nalidixic acid ester via nalidixoyl chloride from nalidixic acid [11]

1 g of pure nalidixic acid was accurately weighed into a 250 ml dry, clean, round bottom flask and 2 ml of thionyl chloride was added and closed in fuming cupboard chamber. The flask was kept aside for 15 min. Later 2 ml of methanol was added to the solution in the round bottom flask drop by drop and mixed thoroughly after each addition. The reaction should be carried out carefully in fuming cupboard chamber. The Nalidixoyl chloride formed, in situ, by the addition of Thionyl chloride to nalidixic acid reacted with methanol and was kept for refluxing. Time taken for complete conversion was 1 hour. This formed nalidixic acid ester.

4.3. Formation of nalidixic acid hydrazide [derivative Ga (NAH)] [10]

To the above formed methyl ester in solution, 2 ml of Hydrazine hydrate was added drop wise carefully through the side of the round bottom flask kept closed in fuming cupboard chamber. Evolution of heat took place with a violent reaction. The above mixture was then kept for refluxing for 4 hours. The contents of the flask were poured into a beaker containing ice cold water. A yellowish-orange coloured nalidixic acid hydrazide precipitated immediately.

4.4. Preparation of acetanthranil [11]

Appropriate anthranilic acid of weight 6.8 g (0.05 mol) was refluxed with 20 ml (0.21 mol) acetic anhydride for 1 hour. After excess acetic anhydride was distilled, the acetanthranil separated as solid mass and was used without further purification.

4.5. Preparation of 6-nitro-acetanthranil [12]

To 2-methyl-3,1-benzoxazine-4-one (6 g) dissolved in conc. Sulphuric acid (20 ml), was added fuming nitric acid (10 ml) keeping temperature below 75 °C. The reaction mixture was poured on ice (200 g), filtered, washed and recrystallised from glacial acetic acid (95%).

4.6. Preparation of 6-iodo-acetanthranil

This involves mainly two steps:

- Formation of Iodide derivative of anthranilic acid (Based on method developed by Klemme Carl J. and Hunter James H., November 16, 1940).
  With magnetic stirrer 5 g (0.184 mol) of recrystallised anthranilic acid (m.p. 145°) was dissolved in a solution of 3.38 g of stick potassium hydroxide in 100 cm^3 of water contained in 500 ml beaker. A solution of 9.3 g (0.184 mol) of Iodine in 50 cm^3 of water containing 4.95 g of stick potassium hydroxide was slowly run into a well-stirred potassium anthranilate solution. After 1 min, 20 cm^3 of Glacial acetic acid was quickly added and the reaction-mixture immediately diluted with 100 cm^3 of water. A dark precipitate began to appear almost at once; stirring was continued for 1 hour, during which time this

precipitate assumed a light brown colour. After standing undisturbed for 2 hours, excess iodine was removed by adding 5 cm^3 of 15% sodium bisulfite and thoroughly agitating. The mixture was allowed to stand a short while. The precipitate was collected, repeatedly washed with water and air-dried. Yield of crude product 8 g (86.8%).

- Acetanthranil formation by refluxing with acetic anhydride. [12].

  Appropriate 2-amino-5-iodobenzoic acid 3.2 g (0.0125 mol) was refluxed with acetic anhydride 10 ml (0.106 mol) for one and half-hour. After excess of acetic anhydride was distilled, the 5-iodoacetanthranil was separated as brown coloured crystalline product and was used without purification.

4.7. Quinazolones preparation

Equimolar proportions of the appropriate acetanthranils and nalidixic acid hydrazide in the presence of ethanol were heated under reflux for 8 hours to synthesise quinazolones [13].

4.7.1. Preparation of 2-methyl-3,1-quinazol-4-one derivatives of nalidixic acid [derivative Gb (QN)]

Interaction of 2-methyl-3, 1-benzoxazine-4-one with nalidixic acid hydrazide yielded dark orange brown coloured crystals of 3-N-(nalidixic amide)-2-methyl-3,1-quinazol-4-one.

4.7.2. 6-Iodo-2-methyl-3,1-quinazol-4-one derivatives of nalidixic acid [derivative Gc (IQN)]

Interaction of 2-methyl-6-iodo-3, 1-benzoxazine-4-one and nalidixic acid hydrazide yielded 6-iodo-2-methyl-3,1-quinazol-4-one derivatives of nalidixic acid. The product was obtained as flaky mass and was allowed to dry overnight.

4.7.3. 6-Nitro-2-methyl-3,1-quinazol-4-one derivatives of nalidixic acid [derivative Gd (NiQNA)]

Interaction of the appropriate 2-methyl-6-Nitro-3, 1-benzoxazine-4-one and nalidixic acid hydrazide yielded 6-nitro-2-methyl-3,1-quinazol-4-one derivatives of nalidixic acid.

5. Spectral data

5.1. Derivative Ga (NAH)

UV (MeOH): 322, 254, 216; **IR (KBr)**: 3309 cm^{-1}(amide NH bonded), 3232 cm^{-1} (Primary aromatic NH free), 3051 cm^{-1} (Aromatic –CH stretch), 1612 cm^{-1} (–C=O), 1496 cm^{-1} (Aromatic C=C), 1359 cm^{-1} (C–N stretch tertiary), 981 cm^{-1} (–CH deformation aromatic).

5.2. Derivative Gb (QN)

UV (MeOH): 304, 251, 220, 194; **IR (KBr)**: 3182 cm^{-1} (–NH–), 1691 cm^{-1} (–C=O) 1651 cm^{-1} (–N–C=O), 1595 cm^{-1} (–C=O), 1517 cm^{-1} (–C=N), 1375 cm^{-1} (C–N stretch tertiary), 962 (CH deformation aromatic).

5.3. Derivative Gc (IQN)

UV (MeOH): 325, 259, 225; **IR (KBr)**: 3234 cm^{-1} (–N–H), 1683 cm^{-1} (–C=O), 156 cm^{-1} (–N–C=O), 1598 cm^{-1} (–C=O), 1498 cm^{-1} (–C=N), 1375 cm^{-1} (C–N stretch tertiary), 962 cm^{-1} (CH deformation aromatic), 478 cm^{-1} (C–I); **^{1}H NMR (DMSO)**: 8 (MHz) (1 proton, CONH–), 7–8(MHz) (6H aromatic protons), 2(MHz) (CH_3, 3H aliphatic), 2(MHz) (CH_3, 3H aliphatic), 1(MHz) (C_2H_5, 5H aliphatic); **MS: m/z** 472 (M^+, 1%), 263 (100%), 215 (28%), 187 (24%), 160 (8%), 119 (26%), 91 (26%).

5.4. Derivative Gd (NiQNA)

UV (MeOH): 325, 256, 217, 203; **IR (KBr)**: 3419 cm^{-1} (–NH–), 2970 cm^{-1} (–CH), 1610 cm^{-1} (CONH), 1517 cm^{-1} (–C=N), 1442 cm^{-1} ($–NO_2$), 1334 cm^{-1} (C–N stretch tertiary).

6. Discussion

In a previous study, certain new nalidixic acid derivatives substituted with 1,2 pyrazolines were synthesised by Waheed and Khan [14] and showed antibacterial and analgesic activity. However, the present study is the first of its type, involving synthesis of quinazolone derivatives of nalidixic acid and screening for antibacterial and antifungal activity using 14 clinically isolated microbial strains.

The antibacterial activity data indicate that certain synthesised derivatives, e.g. derivatives **Ga** (NAH), **Gb** (QN) and **Gd** (NiQNA) exhibited inhibitory activity against *A. hydrophila*, which was resistant to pure nalidixic acid. Similarly, derivative **Ga** (NAH) and **Gd** (NiQNA) showed inhibitory activity against *S. pyogenes*, which was also resistant to pure nalidixic acid. However, no inhibitory activity was observed against *C. negative Staphylococcus* with both synthesised quinazolone derivatives and pure nalidixic acid.

Quinazolone ring substituted nalidixic acid moiety showed an inhibitory activity against *C. albicans*, which was found to be absent in the parent lead compound (pure nalidixic acid). A decreased inhibitory activity against *P. vulgaris* was noted in synthesised quinazolone derivatives of nalidixic acid as compared to pure nalidixic acid. Almost all the synthesised derivatives have displayed enhanced antimicrobial activity in comparison to pure nalidixic acid.

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