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Synthesized novel N10-alkyl substituted acridine-9-one derivatives' antimicrobial activity

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Introduction

A significant class of chemotherapeutic medications and dyes, acrylidine and its derivatives are bacteriostatic and bactericidal to both Gram-positive and Gram-negative organisms. 1–2. A derivative of acridine, acridin-9-(10H)-one is an intriguing tricyclic nitrogen-containing heterocyclic molecule that was initially utilized to treat malaria in the 19th century3. Alkaloids containing acridone have been identified in the bark and leaves of a number of trees, including Melicopoe fareana and Evodia xanthoxyloids. Melicopicine, melicopidine, and eroxanthine4 are significant alkaloids discovered in the woods of Queensland, Australia. Numerous biological actions, including antileishmanial, anticancer, anti-HIV, and antibacterial properties, have been identified for acridone. Acridone5 is also linked to poor bioavailability and medication resistance. Some more recent N10-alkyl substituted acridin-9-ones were created by us. Tetrabutyl ammonium bromide was used as a phase transfer catalyst to perform N10-alkylation with 1-bromo-3-chloropropane in the first series and 1,2-dichloroethane in the second series. Later, using anhydrous acetonitrile as a solvent and refluxing for varying lengths of time in the presence of potassium carbonate, both were derivatized with distinct secondary amines in accordance with scheme (5a-5e, 6a-6b). The produced compounds' spectrum data are good and consistent with the suggested structures. Every synthetic molecule was tested for antibacterial activity (6–9).

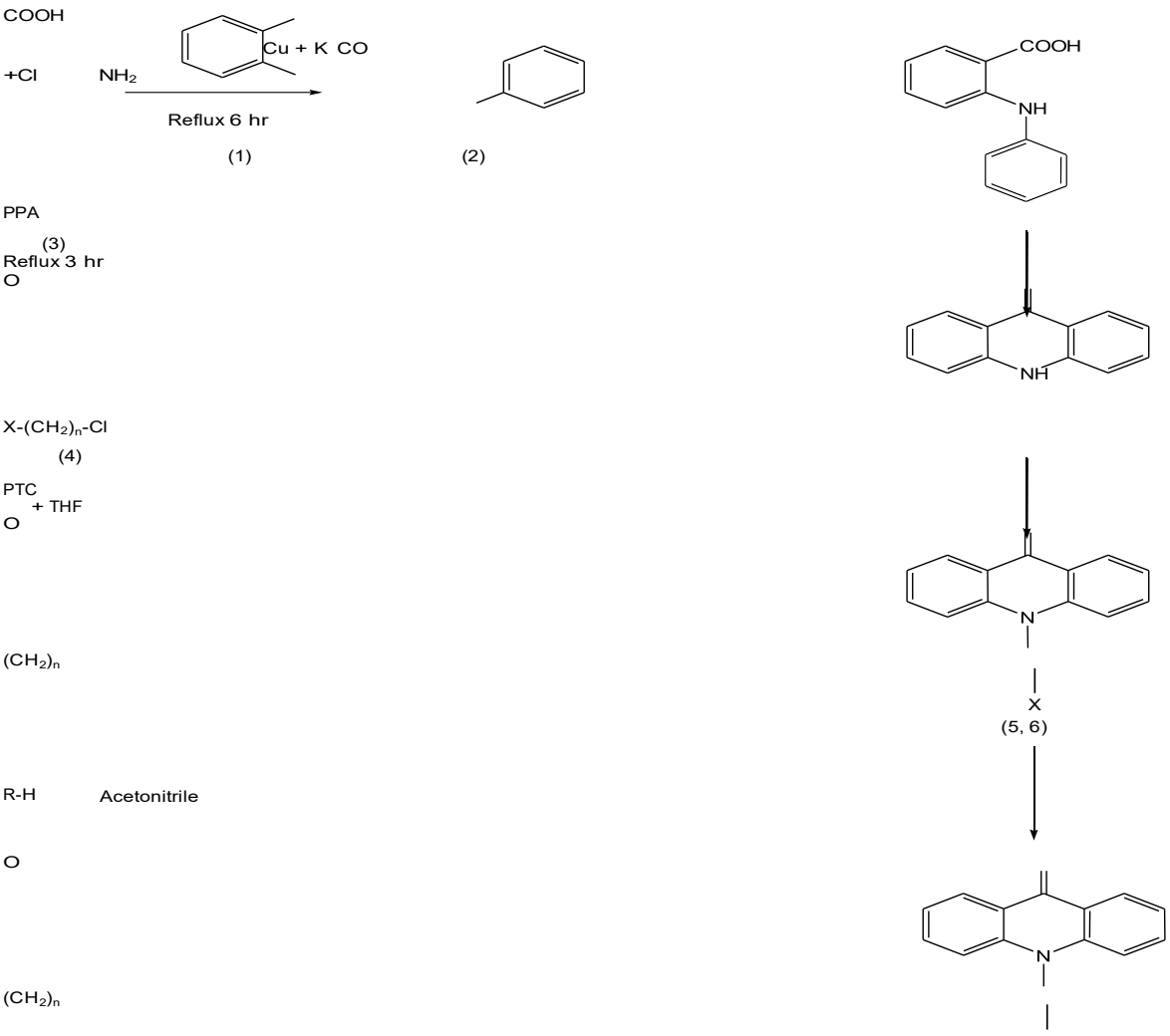
Materials and Methods

Aldrich, Hi-Media, Merck, Sigma, and Ranbaxy were the suppliers of the AR and LR grade chemicals and reagents utilized in this experiment. The open capillary method was used to determine the uncorrected melting points of the produced compounds. Deuterated chloroform and dimethyl sulfoxide were the solvents employed in the FTIR-8400S, SHIMADZU, and 1HNMR spectral analyses, which were performed using the instruments Amx-400 and FTIR-8400S, respectively. LCMS 2010A, SHIMADZU, provided the mass spectrum data.
The method described in literature 10–11 was used to manufacture N10-Alkylated Acridine-9-Ones.

Method of preparation of N-phenyl anthranilic acid (3):

Ullmann condensation:

Aniline (11.8 ml, 0.128 mol), copper metal (0.5 g), and o-chlorobenzoic acid (20 g, 0.128 mol) were combined in a 500 ml round-bottom flask. Isomyl alcohol (100 ml) was then added while stirring. Dry potassium carbonate (20g) was gradually added to this mixture while being stirred, and the reaction mixture was then allowed to reflux for six hours at 135–140°C in a light liquid paraffin oil bath.



The combination was put in two liters of hot water and acidified with strong hydrochloric acid after the isoamyl alcohol was extracted using steam distillation. The bluish-black precipitate that resulted was filtered, washed with hot water, and collected. The crude acid was dissolved in a 10% sodium hydroxide aqueous solution, boiled with activated charcoal present, and filtered. Strong hydrochloric acid was used to acidify the filtrates, resulting in a light yellowish precipitate.It was washed with hot water. The crude acid was recrystallized from aqueous methanol to obtain a pale yellow solid.

Scheme of Synthesis

Compound	X	n	R
5	Cl	3	--
5a	Cl	3	N,N-diethylamine
5b	Cl	3	N,N dimethylamine
5c	Cl	3	N,N diisopropylamine
5d	Cl	3	N,N diphenylamine
5e	Cl	3	pyrrolidin-2-one
6	Cl	2	--
6a	Cl	2	N,N-diethylamine
6b	Cl	2	N,N dimethylamine

Method of preparation of acridin-9-one (4):

Polyphosphoric acid (180g, 0.5327mol) was introduced to a 500ml round-bottom flask containing N-phenyl anthranilic acid (18g, 0.084mol), thoroughly shaken, and refluxed on a water bath at 100°C for three hours. The reaction was complete when the color turned yellow. After that, it was added to two liters of hot water and treated with a 25% ammonia solution to make it alkaline. After filtering and washing with hot water, the yellow precipitate was gathered. Acetic acid was used to recrystallize the crude acridin-9(10H)-one.

Method of preparation of 10-(3′-chloropropyl) acridin-9-one (5):

At room temperature, 500 milliliters of iodine flask were filled with 67.5 grams of potassium hydroxide and 5 grams of tetrabutyl ammonium bromide as a catalyst. After adding acridin-9(10H)-one (10g, 0.051mol) to this mixture while stirring, 200ml of tetrahydrofuran was added gradually, and the reaction mixture was agitated for 30 minutes. After adding 20 milliliters of bromo-3-chloro propane (0.127 mol) gradually, the reaction mixture was agitated for 40 hours at room temperature. Chloroform was used to remove the aqueous layer after tetrahydrofuran was evaporated. After three water washes, the hloroform layer was dried over anhydrous sodium sulfate and rotavaporated. Using the solvent system chloroform: acetone (8:1), column chromatography was used to purify the crude 10-(3'-chloropropyl) acridin-9-one, yielding a yellow solid.

Method of preparation of 10-[3'-(N, N-diethyl amino) propyl] acridin-9-one (5a):

Acetonitrile (200ml) was added to a 500ml round-bottom flask containing 10-(3'-Chloropropyl) acridin-9-one (5g, 0.0184mol) while stirring. This mixture was refluxed for 30 minutes at 60°C after potassium iodide (7.5g) and potassium carbonate (12.5g) were added. N, N-diethylamide (10 ml, 0.14 mol) was then gradually added, and the mixture refluxed for eighteen hours. Chloroform was used to remove the contents after they had cooled and been diluted with water. After three water washes, the chloroform layer was dried over anhydrous sodium sulfate and rotavaporated. Using the solvent system chloroform:acetone (8:1), the crude product was refined by column chromatography to provide a light yellow solid of 10-[3'-(N, N-diethylamino) propyl] acridin-9-one.

Method of preparation of 10-[3'-(N, N-dimethylamino) propyl] acridin-9-one (5b):

The procedure used for 5a was repeated with 5g (0.0184mol) of 5, 7.5g of KI, 12.5g of K_2CO_3 and 10g (0.222mol) of N, N dimethylamine by refluxing 16 hr. The product was purified by column chromatography to give a light yellow solid.

Method of preparation of 10-[3'-(N, N-diisopropylamino) propyl] acridin-9-one (5c):

5g (0.0184mol) of 5, 7.5g of KI, 12.5g of K_2CO_3, and 10ml (0.099mol) of N, N diisopropylamine were added, and the refluxing process was repeated for 20 hours. Column chromatography was used to purify the product, yielding a yellow solid.

Method of preparation of 10-[3'-(N, N-diphenylamine) propyl] acridin-9-one (5d):

5g (0.0184mol) of 5, 7.5g of KI, 12.5g of K_2CO_3, and 10g (0.059mol) of N, N diphenylamine were refluxed for 19 hours using the same method as for 5a. Column chromatography was used to purify the product, yielding a light yellow solid.

Method of preparation of 10-[3'-(N-pyrrolidin-2''-one) propyl] acridin-9-one (5e):

5g (0.0184mol) of 5, 7.5g of KI, 12.5g of K2CO3, and 10ml (0.1175mol) of pyrrolidin-2-one were refluxed for 21 hours in the same manner as for 5a. Column chromatography was used to purify the product, yielding a light yellow solid.

Method of preparation of 10-(2′-chloroethyl) acridin-9-one (6):

Using 10g (0.051mol) of acridin-9-one, 5g of tetrabutyl ammonium bromide, and 1,2-dichloroethane (20ml, 0.2021mol), compound 6 was created in its pure form using the same method as compound 5. Column chromatography was used to purify the crude product, resulting in a solid that was yellowish gray in color.

Method of preparation of 10-[2'-(N, N-diethylamino) ethyl] acridin-9-one (6a):

Using 5g (0.0194mol) of 6, 7.5g KI, 12.5 g K2CO3, and 10ml (0.14mol) of N,N diethylamine, the experimental procedure described for 5a was repeated. Column chromatography was used to purify the product, yielding a light yellow solid.

Method of preparation of 10-[2'-(N, N-dimethylamino) ethyl] acridin-9-one (6b):

Using 5g (0.0194mol) of 6, 7.5g KI, 12.5 g K2CO3, and 10g (0.222mol) of N,N dimethylamine, the experimental procedure used for 5b was repeated. Column chromatography was used to purify the product, yielding a yellowish grey crystal.

Antibacterial activity:

At a concentration of 100µg/ml, the cup-plate method was used to assess the synthetic compounds' antibacterial activity against the gram-positive bacteria Staphylococcus aureus and Bacillus subtilis as well as the gram-negative bacteria Escherichia coli, Pseudomonas aeuroginosa, and Shigella. Nutrient Agar medium was used to subculture the microorganisms. For twenty-four hours, the petridishes were incubated at 37ºC. The standards were ciprofloxacin (30 mcg/disc) (Std.2) and ampicillin (10 mcg/disc) (Std.1). Table 2 presents the findings.

Antifungal activity:

The antifungal activity of the synthesized compounds was carried out against the fungus Candida albicans and Aspergillus niger at 100µg/ml concentration. The Sabouraud Dextrose Agar medium was used to subculture the fungi. Fluconazole (10 mcg/disc) (Std. 1), Amphotericin B (100 units/disc) (Std. 2), and Clotrimazole (100 mcg/disc) (Std. 3) were used in the cup-plate method of testing for fungal susceptibility. The petridishes were kept at 25ºC for 48 hours. Table 2 presents the findings.

Results and Discussion

N-phenyl anthranilic acid (3): 1658.67 (C=O str.); 3330.84 (N-H str.); 2500-3300 (C=O bend.); IR (KBr), CM-1: 3041.53 (C-H str. aromatic); 1514.02, 1452.30 (C=C str. aromatic). IR(KBr), CM-1: 3352.05 (N-H str.); 3097.47, 3062.75, 3031.89 (C-H str. aromatic); 1641.31 (C=O str.); 1535.23, 1473.51 (C=C str. aromatic); Acridin-9-one (4): (m.p. 340°C). 10-(3′-chloropropyl) acridin-9-one (5): (m.p. 180°C); IR(KBr), CM-1: 3070.46, 3018.39 (C-H str. aromatic); 2943.17, 2864.09 (C-H str. alkanes); 1610.45 (C=O str.); 1556.45, 1492.80 (C=C str. aromatic); 567.03, 543.89 (C-H rock methyl); 754.12 (C-H rock methyl); 1H NMR (CDCl3).

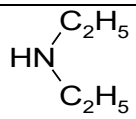
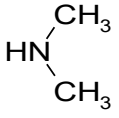
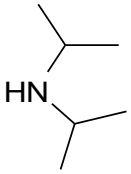
10-[3'-(N,N-diethylamino) propyl] acridin-9-one (5a): (m.p. 152°C); IR(KBr), CM-1: 3097.47, 3033.82 (C-H str. aromatic); 2993.32, 2948.96 (C-H str. alkanes); 1596.95, 1492.80 (C=C str. aromatic); 1631.67 (C=O str.); 752.19 (C-H rock, methyl); 1H NMR (CDCl3): 7.1 -8.5 {m, Ar-H (8H)}; 3.6 -4.0 {t,Ha (2H)}; 3.1 -3.4 {m, Hd (4H)}; 2.3 -2.0 {t, Hc (2H)}; 1.7-1.9 {m, Hb (2H)}; 0.8 -1.0 {s, He (6H)}; MS: (m/z) 308 (M+), M+1, M+2, 200, 181.

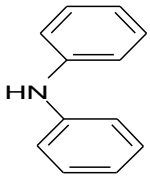
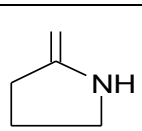
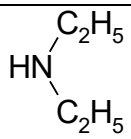
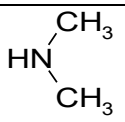
10-[propyl-3'-(N,N-dimethylamino)] (m.p. 220°C); IR(KBr), CM-1: 3097.47, 3031.89 (C-H str. aromatic); 2993.32, 2948.96 (C-H str. alkanes); 1598.88, 1531.37 (C=C str. aromatic);

1635.52 (C=O str.); 752.19 (C-H rock, methyl). IR(KBr), CM-1: 3095.54, 3033.82 (C-H str. aromatic); 2956.67, 2867.95 (C-H str. alkanes); 1598.88, 1494.73 (C=C str. aromatic); 1633.59 (C=O str.); 752.19 (C-H rock, methyl); 10-[3'-(N,N-diisopropylamino) propyl] acridin-9-one (5c): (m.p.170°C). 10-[3'-(N, N-diphenylamino) propyl] acridin-9-one (5d): (m.p. 240°C); IR (KBr), CM-1: 3099.39, 3031.89 (C-H str. aromatic); 2993.32, 2896.88 (C-H str. alkanes); 1598.88, 1488.94 (C=C str. aromatic); 1635.52 (C=O str.); 752.19 (C-H rock, methyl). Propyl 10-[3'-(N-pyrrolidin-2''-one)] IR (KBr), CM-1: 3095.54, 3033.82 (C-H str. aromatic); 2995.25, 2869.88 (C-H str. alkanes); 1596.95, 1494.73 (C=C str. aromatic); 1631.67 (C=O str.); 750.26 (C-H rock, methyl); acridin-9-one (5e): (m.p. 124°C). 10-(2′-chloroethyl) acridin-9-one (6): (m.p. 160°C); IR(KBr), CM-1:3045.39, 3010.67 (C-H str. aromatic); 2960.53, 2873.74 (C-H str. alkanes); 750.26 (C-H rock, methyl); 1573.81, 1487.01 (C=C str. aromatic); 673.11, 551.60 (C-Cl str.); 1625.88 (C=O str.); 1H NMR (CDCl3): ± 7.2-8.4 {m, Ar-H (8H)}; ± 3.5--3.9 {m,Hb (2H)}; ± 3.0--3.2 {m, Ha (2H); MS: (m/z) 257 (M+), M+1, M+2, with further peaks at 258, 259, 221, and 63. IR(KBr), CM-1: 3095.54, 3031.89 (C-H str. aromatic); 2991.39, 2898.81 (C-H str. alkanes); 1600.81, 1531.37 (C=C str. aromatic); 1635.52 (C=O str.); 750.26 (C-H rock, methyl); 10-[2'-(N, N-diethylamino) ethyl] acridin-9-one (6a): (m.p. 190°C).

10-[2'-(N,N-dimethylamino) ethyl] acridin-9-one (6b): (m.p. 240°C); IR(KBr), CM-1: 3099.39, 3031.89 (C-H str. aromatic); 2993.32, 2896.88 (C-H str. alkanes); 1598.88, 1531.37 (C=C str. aromatic); 1635.52 (C=O str.); 752.19 (C-H rock, methyl); 1H NMR (CDCl3): 7.2-8.5 {m, Ar-H (8H)}; 3.2--3.4 {m,Ha (2H)}; ± 2.5-2.7 {m, Hb (2H)}; ±2.1-2.4{s,Hc(6H)}; MS: (m/z) 266 (M+), M+1, and other peaks are seen at 267, 199, 159, 71.

Table 1:
Physical Data of the Synthesized Compounds

Compound Code	n	R	Mol. Formula	% Yield	R_f*	M.P.
5	3	--	$C_{16}H_{14}NOCl$	46.69%	0.88	180°C
5a	3		$C_{20}H_{24}N_2O$	44.09%	0.75	152°C
5b	3		$C_{18}H_{20}N_2O$	48.45%	0.72	220°C
5c	3		$C_{22}H_{28}N_2O$	42.00%	0.68	170°C

5d	3		$C_{28}H_{24}N_2O$	48.39%	0.65	240°C
5e	3		$C_{20}H_{20}N_2O_2$	45.84%	0.70	124°C
6	2	--	$C_{15}H_{12}NOCl$	47.04%	0.93	160°C
6a	2		$C_{19}H_{22}N_2O$	49.04%	0.79	190°C
6b	2		$C_{17}H_{18}N_2O$	52.22%	0.74	240°C

*Stationary Phase : Silica Gel G
Mobile Phase : Chloroform: Acetone: 8:1

Antibacterial activity:

All investigated microorganisms (S. aureus, B. subtilis, E. coli, P. auroginosa, and Shigella) were susceptible to mild to moderate antibacterial activity from the majority of the compounds. The diameter of the zone of inhibition (Table 2) indicates that all of the drugs have demonstrated antibacterial activity. When compared to other derivatives, compound 5C was found to have the greatest activity against both Gram positive and Gram negative organisms, while compound 5d was found to have the least activity against fungal, Gram positive, and Gram negative organisms. Ciprofloxacin (30 mcg/disc) and 5c (100 µg/ml) both had antibacterial efficacy against B. subtilis and E. coli.

Antifungal activity:

Two fungus species were used to test the compounds' antifungal efficacy. The majority of the substances exhibited respectable antifungal activity against the two tested strains (C. albicans and A. niger). The antifungal activity of 5a (100 µg/ml) against C. albicans was higher than that of clotrimazole (100 mcg/disc), but comparable to that of fluconazole (10 mcg/disc) and amphotericin B (10 units/disc).

Table 2:
Biological Activity Data of the Synthesized Compounds

Compound Code	Zone of Inhibition (in mm)						
	B.subtilis	S.aureus	E.coli	P.aeuroginosa	Shigella	C.albicans	A.niger
5a	17	14	19	18	17	22	13
5b	19	15	22	22	19	18	4
5c	28	20	28	21	20	4	14
5d	13	12	2	5	7	3	3
5e	21	12	7	7	8	1	19
6	NI	NI	3	6	NI	4	4
6a	NI	NI	8	10	12	12	12

6b	NI	NI	21	18	10	7	11
Std 1	3	4	NI	9	6	23	17
Std 2	33	37	30	27	40	31	8
Std 3	--	--	--	--	--	18	23
Control	NI	NI	NI	NI	NI	NI	NI

Note : Average zone diameter in mm of triplicates
NI : No inhibition
Control : DMSO

Conclusion

When compared to the derivatives of N10-(2'-chloroethyl) acridin-9-one, the derivatives of N10-(3'-chloropropyl) acridin-9-one with different secondary amines have strong antibacterial activity. With these promising results, all of the produced chemicals can be investigated further for in-depth pharmacological and microbiological research to potentially produce more effective medications.

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