Design, Synthesis, Characterization and Biological Evaluation of Some Novel Schiff Bases containing Quinoline Derivatives for Antioxidant and Anti-Inflammatory Activities

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Abstract

In this study, novel Schiff bases containing quinoline derivatives were synthesized, characterized and evaluated for their anti-inflammatory and antioxidant potential. In the process of synthesis, the first step involves the synthesis of quinoline derivatives by reacting substituted anilines with dimethylformamide (DMF) and phosphorus oxytrichloride ($PoCl_3$) under reflux conditions whereas the second step includes the condensation of the quinoline derivatives with metformin using a grinding technique at room temperature to form Schiff bases. The synthesized compounds were characterized using various analytical techniques such as melting point, TLC and spectroscopic methods (FT-IR, NMR and mass spectrometry). The anti-inflammatory potential was evaluated using the protein denaturation method and results reveals that QM 1 and QM 3 exhibited the highest inhibition rates at 1000 µg/mL, with values comparable to the standard drug. The antioxidant activity was assessed using the H_2O_2 scavenging assay and the compound QM 1 shows the strongest antioxidant potential.

Additionally, molecular docking study against Protein Arginine Deiminase (PAD4) suggests that QM 2 and QM 4 exhibited the strongest binding affinity that highlights their potential for inflammation-related therapeutic applications. The synthesized compounds were also evaluated for drug-likeness based on Lipinski's rule of five and the synthesized compounds indicate their potential as orally bioavailable drug candidates. In conclusion, the synthesized quinoline derivatives (QM 1, QM 3 and QM 2) show promising anti-inflammatory and antioxidant activities and could serve as lead compounds for further development in the treatment of inflammation and oxidative stress-related diseases.

Keywords: Quinoline derivatives, synthesis, antioxidant activity, anti-inflammation activity, docking studies, Lipinski's rule.

Introduction

Quinolines are the heterocyclic aromatic compounds that contain benzene ring fused to a pyridine ring. Quinoline is the fundamental scaffold in medicinal chemistry due to its diverse biological activities. Quinolines exhibit diverse pharmacological properties and hence they have attracted substantial interest in medicinal chemistry[15]. Quinoline is an essential pharmacophore in many bioactive molecules that includes clinically approved drugs such as chloroquine, quinine and moxifloxacin[8]. The quinoline core structure exhibits excellent binding affinity with biological targets such as enzymes, receptors and DNA. This quinoline core structural framework allows its versatile functionalization that enables the development of quinoline derivatives with diverse biological activities[9]. These compounds have been extensively investigated for their potential applications as anticancer, antibacterial, antifungal, antiviral and anti-inflammatory agents.

The imbalance between reactive oxygen species (ROS) production and the body's ability to detoxify these reactive intermediates causes oxidative stress in the body. These ROS can cause significant cellular damage by targeting lipids, proteins and DNA. The prolonged oxidative stress can cause diverse pathological conditions such as neurodegenerative disorders (Alzheimer's and Parkinson's disease), cardiovascular diseases, cancer and diabetes[10]. The antioxidants play a crucial role in neutralizing the ROS generated and protect cells from oxidative damage by donating electrons to free radicals and thereby prevent chain reactions that lead to cellular damage. The quinoline derivatives have shown promising antioxidant properties by scavenging free radicals and enhance the activity of endogenous antioxidant enzymes[6].

Inflammation is a protective response of the immune system against harmful stimulations such as pathogens, toxins, or

injuries. The acute inflammation is essential for tissue repair and immune defense whereas the chronic inflammation is associated with various diseases such as rheumatoid arthritis, inflammatory bowel disease, asthma and cancer[3]. The inflammatory response involves the activation of immune cells that release pro-inflammatory cytokines such as TNF-α, IL-1β, IL-6 and inflammatory mediators such as prostaglandins and leukotrienes[1]. The nonsteroidal anti-inflammatory drugs and corticosteroids are commonly prescribed to mitigate the inflammation.

However, the treatment associated with these compounds leads to possible side effects such as gastrointestinal ulcers, cardiovascular risks and immunosuppression.

Thus, there is a significant need to develop safer and more effective anti-inflammatory agents. Quinoline derivatives exhibit potent anti-inflammatory activities by modulating key inflammatory pathways[5]. The structural flexibility of quinoline allows for the incorporation of diverse moieties that exhibit potential antioxidant, inflammatory properties and make them excellent candidates for developing new therapeutic agents to combat various disorders.

The synthesis of quinoline derivatives involves various classical and modern synthetic approaches that allow diverse structural modifications to enhance their biological activity. The literature suggests that several quinoline derivatives were reported by involving classical reaction methodologies such as Conrad–Limpach[16], Friedländer[4], Gould–Jacobs[14], Doebner–von Miller[7], Skraup[13], Pfitzinger[2] and ultrasound irradiation reactions, or greener procedures[11,12]. The literature suggests that there is always a room to synthesise new compounds and investigate their biological applications. Hence, this study focused to synthesize quinoline derivatives.

Material and Methods

Chemicals and reactants: The chemicals used in the study such as salicylic acid, sodium hydroxide, ethyl acetate, n-hexane, chloroform and ethanol were procured from S.D. Fine Chem Limited, Mumbai. Laboratory reagent grade acetic anhydride and sulfuric acid were procured from Merck Chemicals, Mumbai. The reagent grade chemicals utilized in synthesis process such as phosphorus oxychloride ($PoCl_3$), dimethylformamide (DMF) and substituted anilines were procured from Merck Chemicals, Mumbai. The metformin active pharmaceutical ingredient with 98.75 % purity was obtained as gift sample from Lupin Ltd, Hyderabad, India.

Instruments: The ^{1}H and ^{13}C NMR analysis of synthesized Quinoline derivatives was conducted on Avance-III HD 500 MHz NMR spectrometer (Bruker, USA). The analysis utilizes deuterated chloroform as diluent and the chemical shifts were noticed as ppm on the δ scale. Tetramethylsilane (TMS) was utilized as internal reference standard and spectral integration was performed through TopSpin software (version 3.5). The mass spectral analysis was performed on 230 V Portable Mass Spectrometer (Raman Instruments Pvt. Ltd., India). The functional group analysis of synthesized Quinoline derivatives was evaluated using Shimadzu (Japan) Fourier transform infrared spectroscopy (FTIR). The Digital pH meter (S1326, Systronics, India), Melting Point Apparatus (SA MPA, S A Instruments, India), UV cabinet (model 124, Droplet Equipment's, India), Heating mantle (ISKO®, India) and Digital weighing balance (LCGC, Mumbai) were utilized in the study. TLC Aluminum plates were purchased from S.D. Fine Chemical Laboratories, Mumbai.

Synthesis of Schiff bases containing Quinoline Derivatives:

Step 1: Synthesis of Quinoline Derivatives: Accurately weighed substituted aniline (0.1 mol) was taken in a round-bottomed flask, followed by the addition of dimethylformamide (0.1 mol, 7.7 mL) and phosphorus oxytrichloride (0.01 mol, 0.2 mL). The reaction mixture was refluxed for 4 to 5 hours. After completion of the reaction, the round-bottomed flask was removed from the water bath and allowed to cool. The reaction mixture was then poured into crushed ice, leading to the precipitation of the product. The obtained precipitate was filtered, air-dried and subsequently recrystallized using ethanol to obtain the purified product.

Step 2: Synthesis of Quinoline-Metformin Derivatives (Grindstone Technique): Equimolar concentrations of metformin and the substituted quinoline product from the first step were accurately weighed and taken in a mortar. To this, 20 mL of methanol or ethanol was added and the mixture was ground continuously in one direction for 20 minutes. The resulting product was then added to crush ice, leading to precipitation.

The precipitate was filtered, air-dried and recrystallized for further purification. The completion of the reaction was confirmed by monitoring the melting point and thin-layer chromatography (TLC) analysis. The synthesis scheme was presented in figure 1 and table 1 represents the compounds synthesized in the study.

Characterization of synthesized Quinoline derivatives: The structural integrity and purity of the synthesized Schiff bases containing quinoline derivatives were performed using various analytical techniques. The solubility of the synthesized compounds was tested in different solvents and the solubility characteristics were recorded.

The purity of synthesized compounds was evaluated by determining melting points of the compounds. This was evaluated by adopting open capillary tube method and the compounds with pure crystals exhibit sharp and well-defined melting points. TLC was employed as a crucial analytical technique to confirm the formation of new compounds and to assess their purity.

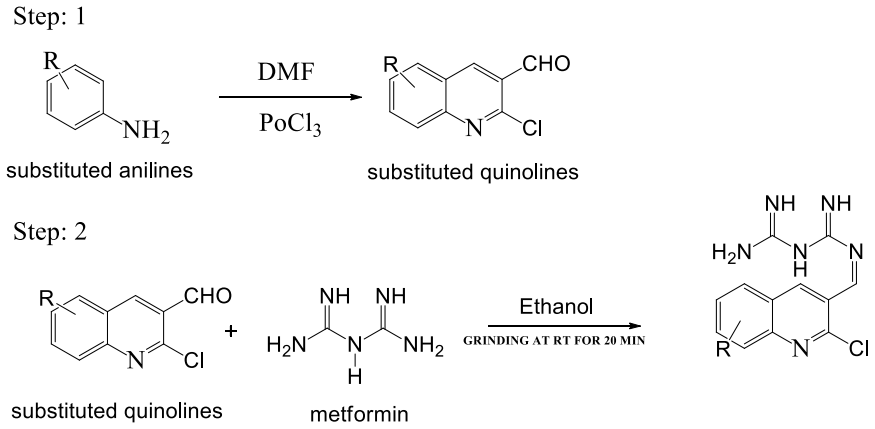
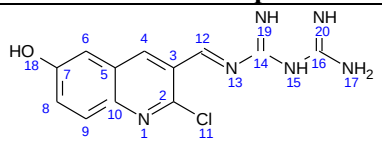
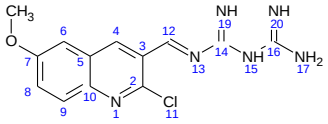
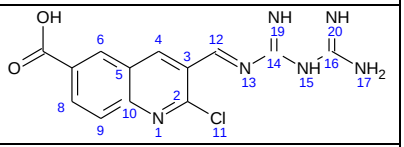
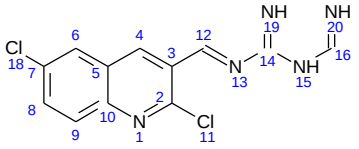


Figure 1: Schematic representation for the synthesis of Quinoline derivatives

Table 1
Details of Quinoline derivatives synthesized in the study

Compound	R	IUPAC NAME	Structure of compound
QM 1	H	*N-[(E)-(2-chloro-6-hydroxyquinolin-3-yl)methylidene]imidodicarbonimidic diamide*	
QM 2	OCH_3	*N-[(E)-(2-chloro-6-methoxyquinolin-3-yl)methylidene]imidodicarbonimidic diamide*	
QM 3	COOH	*3-{(E)-[(N-carbamimidoylcarbamimidoyl)imino]methyl}-2-chloroquinoline-6-carboxylic acid*	
QM 4	Cl	*1-carbonoimidoyl-3-[(E)-(2,6-dichloroquinolin-3-yl)methylidene]guanidine*	

Each compound's retention factor (Rf) value was determined as a characteristic parameter for evaluating the compound purity and the analysis was performed by mixing n-hexane and ethylacetate in 8:2 and 7:3 (v/v) as mobile phase.

Spectral characterization: The synthesized compounds were subjected to structural characterization using FT-IR, NMR and mass spectroscopy studies. The correlation and interpretation of spectral results confirmed the molecular structures of the synthesized quinoline derivatives.

***In vitro* antioxidant activity of synthesized compounds**: The *in vitro* antioxidant assay was performed to evaluate the free radical scavenging activity of quinoline derivatives synthesized in the study. A sample solution was prepared by dissolving 10 mg of the test compound in a 10 mL volumetric flask and make up the volume with phosphate buffer. The hydrogen peroxide (H_2O_2) solution was then added to initiate the reaction. The test samples were prepared at different concentrations by dissolving the required amount of the sample in 10 mL of phosphate buffer solution, labeled as the standard solution.

From this solution, 0.1 mL of the sample was taken and mixed with 0.6 mL of H_2O_2, referred to as standard dilution B. Serial dilutions were then carried out from this standard solution to obtain different concentrations: 200 µg/mL, 400 µg/mL, 600 µg/mL, 800 µg/mL and 1000 µg/mL. The samples were incubated at 70°C for 5 minutes and subsequently cooled for 10 minutes. The absorbance of the reaction mixture was measured at 230 nm using spectrophotometer. The percentage antioxidant activity was calculated using the formula:

$$\% \text{ actviity} = \frac{\text{Absorbance of control} - \text{Absorbance of test sample}}{\text{Absorbance of control}} \times 100$$

***In vitro* anti-inflammatory activity of synthesized compounds**: The albumin denaturation inhibition assay was

performed to evaluate the anti-inflammatory potential of quinoline derivatives synthesized in the study. The egg albumin solution was prepared by dissolving 1 g of egg albumin in 100 mL volumetric flask and make up the volume with distilled water. The synthesized quinoline derivative test compounds were prepared in different concentrations and 1 mL of each test solution was mixed with 1 mL of a 1% aqueous solution of egg albumin. The pH of the reaction mixture was adjusted to 6.8 using glacial acetic acid. The samples were then incubated at 72°C for 5 min and then cool the solution for 10 min. This procedure produces turbidity in the solution and the absorbance of the samples was measured at 660 nm using spectrophotometer. The percentage inhibition of protein (albumin) denaturation was calculated using the formula:

$$\% \text{ Inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of test sample}}{\text{Absorbance of control}} \times 100$$

***In silico* docking and QSAR studies**: The docking and QSAR studies were conducted to evaluate the potential anti-inflammatory activity of the synthesized compounds. The protein arginine deiminase was selected as the target enzyme for molecular docking studies. The docking parameters such as binding energy and IC_{50} values were studied to assess the binding affinity and inhibitory potential of the compounds. Additionally, molecular descriptors such as Log P (partition coefficient), hydrogen bond donors (HBD), hydrogen bond acceptors (HBA), topological polar surface area (TPSA) and ADME (absorption, distribution, metabolism and excretion) properties were analyzed to determine the pharmacokinetic and drug-likeness characteristics of the synthesized derivatives.

Results and Discussion

In this study, quinoline-based derivatives were synthesized in two sequential steps. In the first step, substituted anilines were reacted with DMF and $PoCl_3$ under reflux conditions to yield substituted quinolines. This transformation follows the Vilsmeier-Haack reaction mechanism in which DMF reacts with $PoCl_3$ to generate an iminium ion, which acts as an electrophile and facilitates the formylation of the aniline derivative. The intermediate undergoes cyclization and subsequent oxidation to form quinoline core structure. In the second step, the synthesized substituted quinolines were subjected to a condensation reaction with metformin in ethanol under grinding conditions at room temperature. This reaction involves the formation of a Schiff base where the aldehyde functional group of the quinoline derivative reacts with the amine groups of metformin through nucleophilic addition, followed by dehydration to form a stable imine linkage. The progress of reaction was monitored by TLC and initial purity of the compound was assessed through melting point analysis. The results achieved were tabulated in table 2.

Characterization of synthesized quinoline derivatives: The spectral characterization of quinoline derivatives was performed using FT-IR, NMR and mass spectroscopy studies.

QM 1: White powder; *m.f.*: $C_{12}H_{11}ON_6Cl$; ^{1}H NMR (500 MHZ, CD_3OD, δ, ppm): 5.53 (1H, s), 7.19 (2H, s), 7.65 (1H, dd, *J* = 7.4, 1.9 Hz), 7.70 (1H, ddd, *J* = 1.9, 1.3, 0.4 Hz), 8.09 (1H, dt, *J* = 7.4, 0.4 Hz), 8.56 (2H, s), 8.76 (1H, dd, *J* = 1.3, 0.4 Hz), 9.50 (1H, s); ^{13}C NMR (500 MHZ, CD_3OD, δ, ppm): 108.7 (1C, s), 121.3 (1C, s), 127.6 (1C, s), 130.3 (1C, s), 130.7 (1C, s), 136.6 (1C, s), 145.7 (1C, s), 147.6 (1C, s), 155.3 (1C, s), 157.9 (1C, s), 159.4 (1C, s), 159.6 (1C, s); FT-IR (ν, cm^{-1}): 3341.55 (Aromatic N-H Stretch), 2812.95 (Aromatic C-H Stretch), 1511.77 (C=C Stretch), 1466.33 (C=N Stretch).

QM 2: White powder; *m.f.*: $C_{13}H_{13}ON_6Cl$; ^{1}H NMR (500 MHZ, CD_3OD, δ, ppm): 3.80 (3H, s), 5.51 (1H, s), 7.20 (2H, s), 7.64 (1H, dd, *J* = 6.4, 1.8 Hz), 7.69 (1H, ddd, *J* = 1.8, 1.3, 0.4 Hz), 8.07 (1H, dt, *J* = 6.4, 0.4 Hz), 8.51 (2H, s), 8.76 (1H, dd, *J* = 1.3, 0.4 Hz), 9.50 (1H, s); ^{13}C NMR (500 MHZ, CD_3OD, δ, ppm): 55.5 (1C, s), 106.4 (1C, s), 126.5 (1C, s), 127.6 (1C, s), 129.8 (1C, s), 130.7 (1C, s), 136.6 (1C, s), 145.7 (1C, s), 147.6 (1C, s), 157.9 (1C, s), 158.7 (1C, s), 159.4 (1C, s), 159.6 (1C, s); FT-IR (ν, cm^{-1}): 2917.12 (Aromatic N-H Stretch), 2838.81 (Aromatic C-H Stretch), 1514.79 (C=C Stretch), 1454.65 (C=N Stretch).

QM 3: White powder; *m.f.*: $C_{13}H_{11}O_2N_6Cl$; ^{1}H NMR (500 MHZ, CD_3OD, δ, ppm): 5.53 (1H, s), 7.24 (2H, s), 7.87 (1H, dt, *J* = 6.9, 0.5 Hz), 8.21 (1H, dd, *J* = 6.9, 1.5 Hz), 8.51 (1H, ddd, *J* = 1.7, 1.5, 0.5 Hz), 8.59 (2H, s), 8.89 (1H, dd, *J* = 1.7, 0.5 Hz), 9.61 (1H, s); ^{13}C NMR (500 MHZ, CD_3OD, δ, ppm): 127.6 (1C, s), 128.6 (1C, s), 128.7 (1C, s), 130.3 (1C, s), 130.9 (1C, s), 130.7 (1C, s), 136.6 (1C, s), 145.7 (1C, s), 147.6 (1C, s), 157.9 (1C, s), 159.4 (1C, s), 159.6 (1C, s), 166.9 (1C, s); FT-IR (ν, cm^{-1}): 3402.43 (Aromatic N-H Stretch), 3070.66 (Aromatic C-H Stretch), 1561.63 (C=C Stretch), 1450.47 (C=N Stretch).

Table 2
Characteristics of quinoline derivatives synthesized in the study

S.N.	Compound	R	Molecular formula	Molecular weight	M.P (ºC)	Yield %	Rf value
1	QM 1	H	$C_{12}H_{11}ON_6Cl$	290.74	191ºC -193ºC	92%	0.51
2	QM 2	OCH_3	$C_{13}H_{13}ON_6Cl$	304.00	165ºC -168ºC	75%	0.43
3	QM 3	COOH	$C_{13}H_{11}O_2N_6Cl$	381.71	201ºC -203ºC	85%	0.61
4	QM 4	Cl	$C_{12}H_{10}Cl_2N_6$	309.154	179ºC -180ºC	80%	0.36

QM 4: White powder; *m.f.*: $C_{12}H_{10}Cl_2N_6$; ^{1}H NMR (500 MHZ, CD_3OD, δ, ppm): 5.55 (1H, s), 7.20 (2H, s), 7.75 (1H, s), 7.92 (1H, dd, *J* = 8.3, 1.5 Hz, 8.20 (1H, dt, *J* = 8.3, 0.5 Hz), 8.27 (1H, td, *J* = 1.5, 0.4 Hz), 8.54 (2H, s), 8.80 (1H, dd, *J* = 1.6, 0.5 Hz), 9.60 (1H, s); ^{13}C NMR (500 MHZ, CD_3OD, δ, ppm): 111.6 (1C, s), 126.6 (1C, s), 127.6 (1C, s), 129.6 (1C, s), 130.7 (1C, s), 131.4 (1C, s), 131.9 (1C, s), 136.6 (1C, s), 145.7 (1C, s), 147.6 (1C, s), 157.9 (1C, s), 159.4 (1C, s); FT-IR (ν, cm^{-1}): 3433.31 (Aromatic N-H Stretch), 3117.11 (Aromatic C-H Stretch), 1542.81 (C=C Stretch), 1430.51 (C=N Stretch).

The synthesized quinoline derivatives (QM 1, QM 2, QM 3 and QM 4) were evaluated for their anti-inflammatory potential using the protein denaturation method. This study was conducted within 200 to 1000 µg/mL concentration range utilizing ascorbic acid as standard for activity comparison. The results indicate that QM 1 exhibits the highest activity among the synthesized compounds and shows 70.46% inhibition at 200 µg and reaching 88.69% at 1000 µg. The assay potential was exhibited to be very close to standard drug that shows 94.16% inhibition at the highest concentration. QM 3 demonstrates significant activity with an inhibition of 19.59% at 200 µg and increasing to 93.89% at 1000 µg whereas QM 4 shows moderate to high activity and shows 28.81% activity at 200 µg and reaches to 87.56% at 1000 µg.

In contrast, QM 2 displays the weakest anti-inflammatory potential among the compounds synthesized and exhibited only 12.21% inhibition at 200 µg and 54.96% at 1000 µg. This indicates that this compound possesses less effective response compared to the other compounds. The results suggest a concentration-dependent increase in inhibition for all compounds and confirms their potential as anti-inflammatory agents. Among them, QM 1 and QM 3 showed the most promising activity suggesting that they could be further explored for therapeutic applications in inflammation-related conditions. Table 3 presents the results observed in this study and figure 2A presents the comparative results noticed in this study.

The H_2O_2 scavenging assay method was utilized to evaluate the antioxidant activity of the synthesized quinoline derivatives (QM 1, QM 2, QM 3 and QM 4) and this study utilizes ascorbic acid as a reference. The result indicates a concentration-dependent increase in antioxidant activity for all the synthesized compounds and reflects the ability to scavenge hydrogen peroxide. Among the compounds synthesized, QM 1 exhibits the highest activity, with 29.86% inhibition at 200 µg/mL and progressively increases to 80.30% at 1000 µg/mL. QM 3 also demonstrates strong antioxidant potential and shows 24.73% inhibition at 200 µg/mL and activity increases to 67.66% at 1000 µg/mL. QM 4 shows an initial inhibition of 23.69% at 200 µg/mL and increases to 61.04% at 1000 µg/mL indicating a moderate antioxidant response whereas QM 2 displays the weakest activity, with only 16.09% inhibition at 200 µg/mL and 42.40% at the highest concentration.

The results suggest that QM 1 possesses the strongest antioxidant potential among the synthesized compounds whereas QM 2 exhibits the least activity. These findings indicate that QM 1 and QM 3 could be promising candidates for further investigation as potential antioxidant agents. Table 4 presents the results observed in this study and figure 2B presents the comparative results noticed in this study.

Table 3
Anti-inflammatory activity results of quinoline derivatives synthesized in this study

S.N.	% Inhibition*					
	Concentration in µg/mL	QM 1	QM 2	QM 3	QM 4	Standard
1	200	70.46±0.204	12.21±0.101	19.59±0.115	28.81±0.113	68.64±0.435
2	400	72.28±0.157	38.46±0.097	49.40±0.187	56.87±0.224	75.02±0.421
3	600	76.84±0.114	44.94±0.238	66.63±0.236	65.78±0.269	80.47±0.204
4	800	81.40±0.198	47.67±0.098	79.67±0.229	72.22±0.105	86.87±0.113
5	1000	88.69±0.135	54.96±0.223	93.89±0.367	87.56±0.415	94.16±0.321

*Average ± standard deviation (n = 3)

Table 4
Antioxidant activity results of quinoline derivatives synthesized in this study

S.N.	% Inhibition*					
	Concentration in µg/mL	QM 1	QM 2	QM 3	QM 4	Standard
1	200	29.85±0.115	16.09±0.201	24.72±0.201	23.69±0.104	39.75±0.145
2	400	39.65±0.098	20.93±0.231	36.30±0.165	31.05±0.226	54.09±0.198
3	600	55.40±0.113	31.26±0.108	52.99±0.141	44.73±0.251	76.46±0.141
4	800	70.32±0.204	38.81±0.114	58.61±0.112	58.35±0.314	88.01±0.232
5	1000	80.30±0.198	42.39±0.121	67.65±0.309	61.04±0.332	98.77±0.254

*Average ± standard deviation (n = 3)

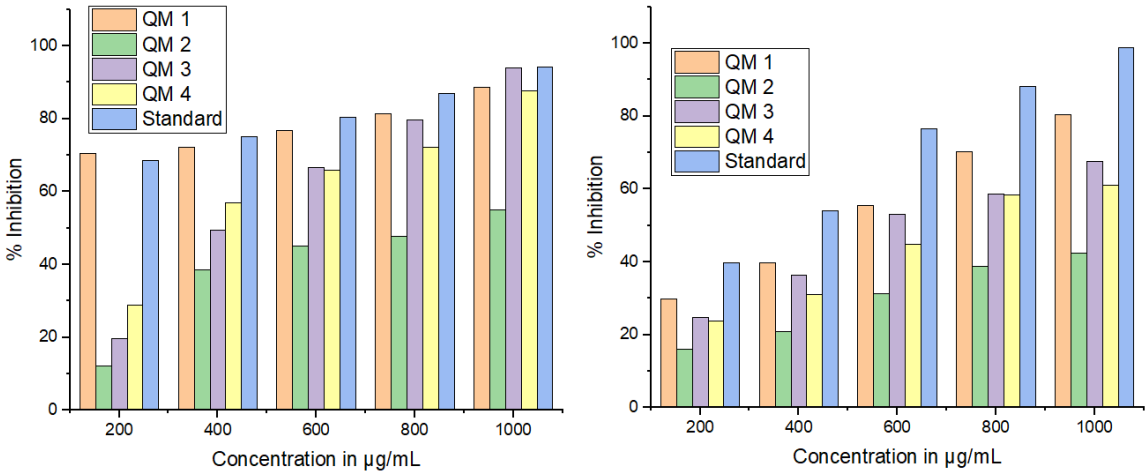
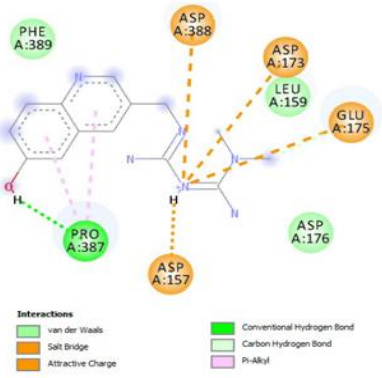


Figure 2: Comparative (A) Anti-inflammatory and (B) Antioxidant activity results of quinoline derivatives synthesized in this study



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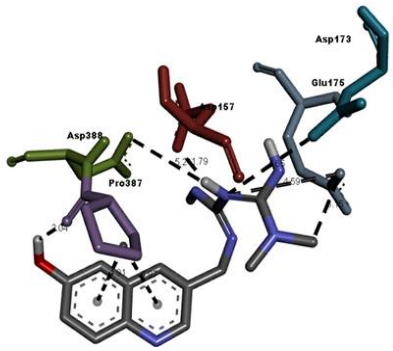

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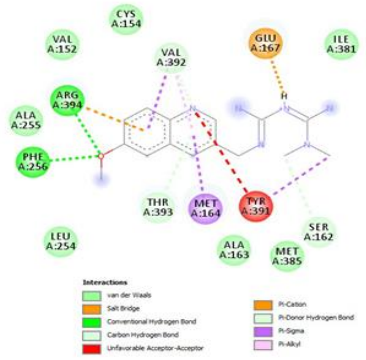

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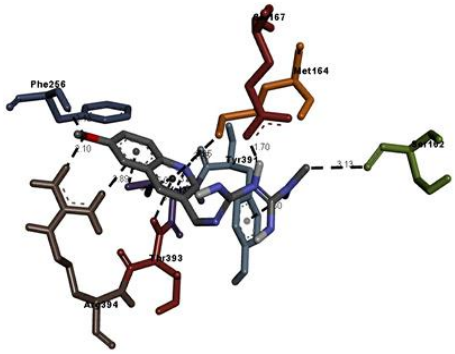

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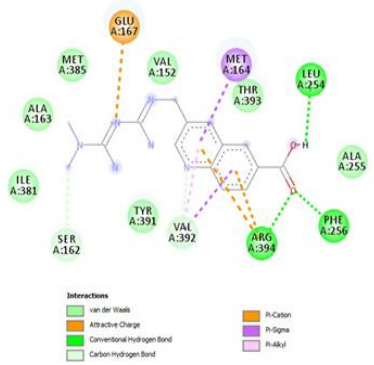

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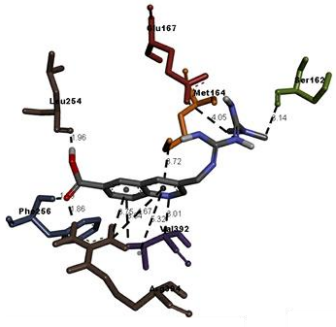

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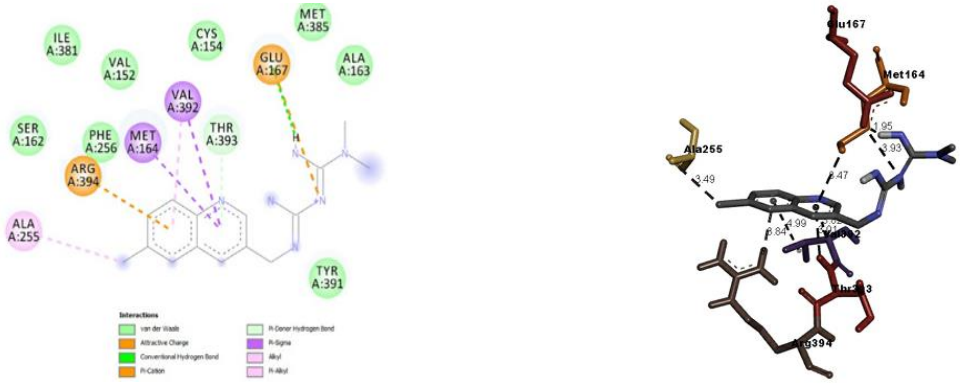

G **H**

2D and 3D images showing synthesized quinoline derivatives binding with Protein Arginine Deiminase in which A and B for QM 1, C and D for QM 2, E and F for QM 3, G and H for QM 4

Figure 3: Representative molecular docking results of synthesized compounds with Protein Arginine Deiminase

The molecular docking study of the synthesized quinoline derivatives was performed against protein arginine deiminase to evaluate their binding affinity and inhibitory potential. This protein was chosen for this study because it plays pivotal role in inflammation and its potential as a therapeutic target for inflammatory and autoimmune diseases. The docking results are represented in terms of binding energy (in kcal/mol) and predicted IC_{50} values. The results in this study provide insights into the interaction strength and potential efficacy of the compounds. The lower binding energy indicates stronger binding affinity whereas the lower IC_{50} values suggest higher inhibitory potency.

Among the synthesized compounds, QM 2 exhibited the strongest binding affinity with a binding energy of -9.69 kcal/mol and the lowest predicted IC_{50} value of 79.37 nM indicates that this compound has the highest potential to inhibit the target protein studied. The compound QM 4 also shows strong binding with binding energy of -9.64 kcal/mol and a predicted IC_{50} value of 86.12 nM suggested significant inhibitory potential whereas QM 3 demonstrates a binding energy of -9.55 kcal/mol, with a predicted IC_{50} value of 99.42 nM. The compound QM 1 exhibits the least binding affinity among the tested compounds with a binding energy of -9.36 kcal/mol and a predicted IC_{50} of 137.41 nM suggested comparatively lower inhibitory potential. The results suggest that QM 2 and QM 4 could be prioritized for further experimental validation and optimization as potential therapeutic agents for the treatment of inflammation. The docking results were presented in figure 3.

The QSAR molecular descriptor values of the synthesized quinoline derivatives were studied to evaluate its ADME properties based on Lipinski's rule of five, which is a widely accepted guideline for predicting drug-likeness and oral bioavailability. The result suggests that all four synthesized compounds successfully passed the ADME assessment indicating their potential as orally bioavailable drug candidates. The Log P value that represents lipophilicity, ranges from 0.3479 (QM 4) to 0.9552 (QM 2) suggesting favorable hydrophobicity for membrane permeability. The number of H-bond donors varied between 3 and 4 whereas H-bond acceptor ranged from 7 to 8, both within the acceptable limits for drug-like molecules.

The number of rotatable bonds was achieved to be minimal (1–2) ensuring good molecular stability. The TPSA values were noticed to be within the range of 101.56 and 129.63 which supports good intestinal absorption and blood-brain barrier permeability. The synthezied compounds do not violate more than one of Lipinski's rules and hence are considered as drug-like and are ideal for further pharmacokinetic and pharmacodynamic studies.

Conclusion

The present study successfully synthesized novel quinoline-based derivatives as therapeutic agents for anti-inflammatory and antioxidant properties. An efficient and practical approach was adopted for the synthesis of quinoline-based derivatives. The synthesis reactions were carried out under mild conditions and utilize ethanol as a solvent in the condensation step enhancing the eco-friendliness of the synthesis by reducing solvent waste and energy consumption. The synthesized compounds were assessed for anti-inflammatory activity and results demonstrate that QM 1 and QM 3 exhibited the highest inhibitory effects that closely resemble the standard drug whereas QM 2 shows the weakest activity. The H_2O_2 scavenging assay for antioxidant activity reveals a concentration-dependent response in which QM 1 displays the highest radical scavenging ability followed by QM 3 and QM 4.

The molecular docking study against PAD4 highlights QM 2 as the most potent inhibitor. QM 4 also shows strong binding affinity suggesting their potential for further drug development in inflammation-related conditions. The QSAR molecular descriptor analysis confirms that all synthesized compounds complied with Lipinski's rule of five that ensures good oral bioavailability and drug-like properties. In conclusion, the study successfully identifies promising lead compounds (QM 1, QM 2 and QM 3) with significant anti-

inflammatory and antioxidant activities that make them potential candidates for further pharmacological and clinical investigations in the treatment of inflammation and oxidative stress-related diseases.

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