SCHOOL OF MATHEMATICAL AND NATURAL SCIENCES

DEPARTMENT OF CHEMISTRY

DESIGN, SYNTHESIS AND BIOLOGICAL EVALUATION OF NOVEL TETRA-SUBSTITUTED QUINOLINE-3-CARBOXAMIDES DERIVATIVES

Dissertation submitted in fulfilment of the requirements for the degree

Master of Science

By

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MARCH 2020

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Abstract

Quinolines are well known naturally occurring heterocyclic compounds with nitrogen as a heteroatom. Quinolines are also one of the major classes of naturally occurring compounds and the interest in their chemistry is due to the wide range of their biological activities.

The objective of the project was the synthesis of novel tetra-substituted quinoline-3-carboxamides and subsequent transformation to other novel derivatives and evaluation of their biological activities against malaria and cytotoxicity. In achieving the objective, 2-chloroquinoline-3-carbaldehyde analogues **54A-G** were synthesised from the reaction of acetanilides **53A-G** and acetic acid. Knoevenagal reaction of 2-chloroquinoline-3-carbaldehydes **54A-G** with thiazolidinedi-2,4-one **62** provided 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A-G** which then underwent nucleophilic substitution reaction with sodium azide and afforded (Z)-5-((tetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56A-F**. (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** was synthesised from the reaction of (Z)-5-((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56D** and ethyl-2-(2-chloroacetamido) acetate **65**. The structures of the compounds were characterised by 1D NMR (^{1}H, ^{13}C, and DEPT 135), IR spectroscopy, elemental analysis and high-resolution mass spectroscopy.

Novel selected synthesised quinoline compounds were evaluated of *in vitro* for two biological assays; namely anti-malarial activity and cytotoxicity. The anti-malaria activities of the novel quinoline compounds against 3D7 strain of the malaria parasite *Plasmodium falciparum* displayed that 2,6-dichloroquinoline-3-methylene thiazolidinedi-2,4-one **55C**, (Z)-5-((7-fluorotetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56B** and (Z)-5((7-ethoxytetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56F** are potential malaria drugs since they reduced the percentage parasite viability to 25.80, 12.40 and 20.40 respectively. These results were further substantiated by their IC_{50} values 0.40, 0.04 and 0.50 µg/mL. Compound **56B** displayed the highest cytotoxicity activity against human cervix adenocarcinoma cells displaying percentage viability of 14.22 %. Compounds **56F** and **56C** displayed moderate cytotoxicity activity at 56.60 and 59.81 % viability.

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Abbreviations

AcOH	acetic acid
Ar	aromatic
$CDCl_3$	deuterated chloroform
CHN	carbon, hydrogen, and nitrogen
DCM	dichloromethane
DMF	N, N-dimethylformamide
DMSO-d_6	deuterated dimethyl sulfoxide
d	doublet
dd	doublet of a doublet
EtOAc	ethyl acetate
EtOH	ethanol
FT-IR	Fourier-transform infrared
HeLa	human cervix adenocarcinoma
HCl	hydrochloric acid
HIV	human immunodeficiency virus
HRMS	high-resolution mass spectroscopy
H_2SO_4	sulphuric acid
IC_{50}	half maximal inhibitory concentration
J_{H-H} meta	J-coupling constant (meta coupling)
J_{H-H} ortho	J-coupling constant (ortho coupling)
KOH	potassium hydroxide
NaN_3	sodium azide
NH_2SO_3H	sulfamic acid

$N_2H_2.H_2O$	hydrazine hydrate
NMR	nuclear magnetic resonance
NaOEt	sodium ethoxide
MeOH	methanol
MHz	Mega Hertz (Hz)
$MgSO_4$	magnesium sulfate
OH	hydroxyl
S	singlet
SAR	structure activity relationship
$SOCl_2$	thionyl chloride
TB	tuberculosis
TBSOTf	tert-butyldimethylsilyl triflate
TLC	thin layer chromatography

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CHAPTER 1

1. Introduction and literature review

1.1 General introduction

Infectious and chronic disease are the two main types of diseases that are causing mortality and morbidity globally. The study of organic chemistry provides information about the past, present and future molecules and their interaction with the environment. The progress of medicine during the past century has been driven by chemistry, clinical sciences, and drug research than any other factor.[1] The pharmaceutical and commercial use of heterocyclic compounds in terms of synthesis, isolation, and biological studies is of interest to organic and medicinal chemists.[2] Numerous drug moieties in the pharmaceutical industry incorporate heterocyclic compounds which is used for the treatment of malaria, tuberculosis, cancer and many others.

Organic synthesis is a compound-creating activity often focused on biologically active small molecules. The study on the chemistry of quinoline-3-carbaldehyde derivatives makes use of various types of reactions such as the Vilsmeier-Haack, Gould-Jacobs, and Mannich reactions to synthesise potential biologically active compounds. A review of the chemistry of quinolines and their biological activities are described below.

1.2 Quinolines

Quinolines are bicyclic heterocyclic aromatic compounds derived from alkaloids and display a wide biological spectrum.[3] Quinoline **1** is a nitrogen-containing heterocyclic compound comprised of a benzene ring fused to pyridine ring at two adjacent carbons as shown in Figure 1.[4]

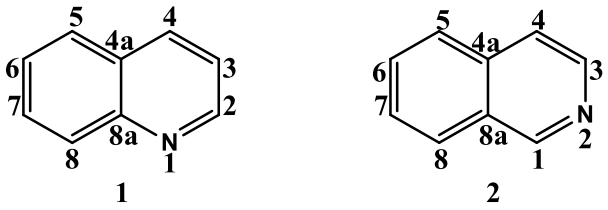


Figure 1: Chemical structure and nomenclature of quinoline.

The quinoline moiety **1** is isomeric to isoquinoline **2**.[5] Isoquinoline **2** is the simplest naturally occurring basic substance possessing fused benzene and pyridine ring with the nitrogen atom at position number two as shown in Figure 1.[6] Isoquinoline **2** derivatives have effectively shown a good biological spectrum such as anticancer, antimalarial, antitumor, antituberculosis, etc.[7]

1.3 Sources of Quinolines

Mankind is exposed to infectious diseases due to the presence or growth of pathogenic biological agents (virus, bacteria, fungi, protozoa, etc) in a host organism. The greatest concern with infectious disease especially malaria and tuberculosis are the alarming rate at which resistance against clinically used drugs develops. This promulgates a degree of urgency for the discovery of affordable and effective drugs.[8] One approach in overcoming some of the problems involves the use of the so-called hybrid drugs, which comprises the incorporation of two or more pharmacophores in a single molecule with intention to exert multi-drug action.[9]

Quinoline **1** occurs from two major sources namely; natural occurrence (plants and animals) and synthetic products. In the following discussion two sources of occurrence of quinolines will be discussed namely; natural and synthetic products. The biological activities of the selected compounds will also be briefly reviewed.

1.4 Naturally occurring quinolines and their biological activities

Most plants with bioactive natural products have been used for millennia as drugs. Numerous naturally occurring bioactive compounds and their derivatives have become drugs of central importance and represent a high percentage of the drugs used today such as antimalarial, antibiotics, and statins are promoted examples.[10, 11]

Heterocyclic compounds play a prominent role in designing new classes of structural entities of medicinal importance with potential new mechanisms of action. These heterocyclic compounds are well known to possess different pharmacological properties such as antimalarial, antiinflammatory, anticancer, antituberculosis, antiHIV, antiinflammatory, etc. Quinoline nucleus occurs in several natural compounds and pharmacologically active substances displaying a broad range of activities.[12]

Alkaloids are natural products that contain heterocyclic nitrogen atoms and are basic in character. Quinine **3** in Figure 2 was isolated from the bark of the *Cinchona tree* and

has been used for centuries for the treatment of malaria and was synthesised naturally by a large number of organisms including animals, plants, bacteria, and fungi. Naturally, fire products isolated from medicinal plants were alkaloids and were first obtained from plant materials in the early years of the 19th century, and it was found that they contain nitrogen.[13]

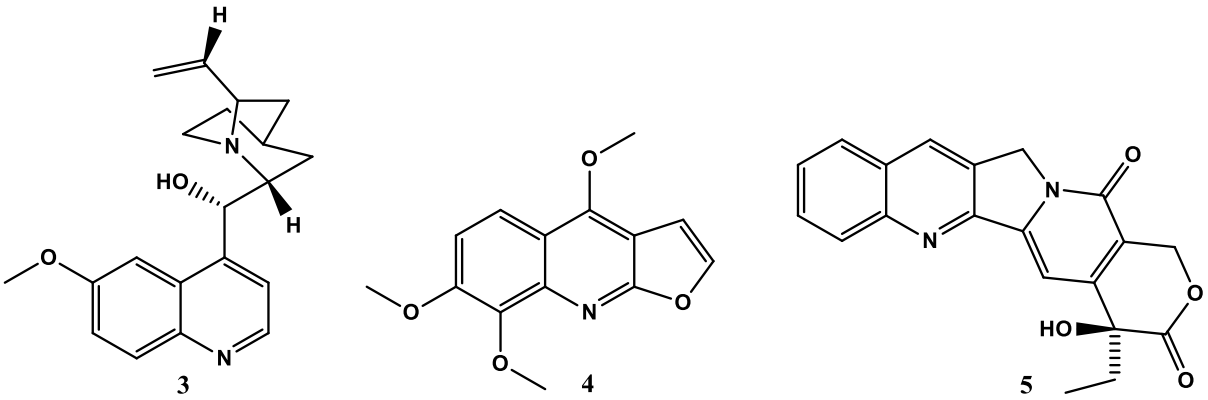
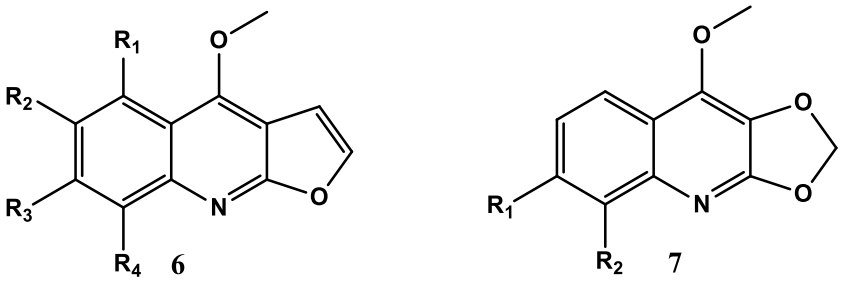


Figure 2: Quinoline alkaloids drugs used as anti-malarial, anti-convulsant and anti-cancer agents

The alkaloid skimmianine **4** was obtained from a perennial herb *Haplophyllum laeviusculum*, a family of Rutaceae that grows naturally around the Mediterranean section of Europe and in Western Asia up to Siberia. It was also endemic in West Azarbaijan Province of Iran locally known as Suddabby miyaneey. Many species of this genus are used for the treatment of herpes, warts, stomach-ache, erysipelas, toothache, skin diseases, and in the treatment of testicular cancer. A vast majority of quinoline alkaloids were also obtained from lowered plants isolated from animals and microorganisms, an example to that is an alkaloid skimmianine **4** in Figure 2 which was used as anticonvulsant and muscle relaxant.[14] Camptothecin **5** in Figure 2 was isolated from *Camptotheca* acuminate decene. The S configuration at C-20 of *Camptothecin* **5** shows excellent anti-cancer activity but the R isomer is inactive.[15]

The fuoroquinoline alkaloid is very common within the Rutaceae family and is the main alkaloid in the roots of *Dictamnus Albus* responsible for genetic alteration.[16] Fuoroquinolines alkaloids **6A-6E** shown in Figure 3 were isolated and used for the treatment of skin diseases such as eczema, atopic dermatitis, and psoriasis.[17]



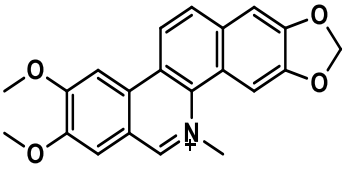
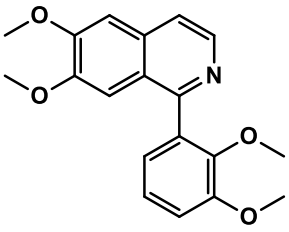
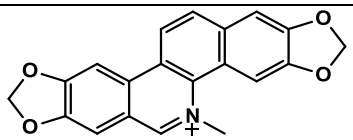
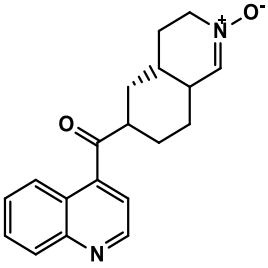
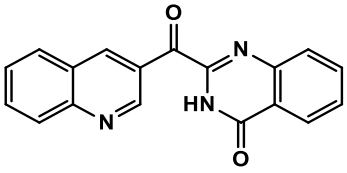
7A, R_1 = R_2 = OMe, 7B, R_1 = H, R_2 = OMe

Compound **6**	R_1	R_2	R_3	R_4
A	H	H	H	H
B	H	H	OH	H
C	H	H	H	OH
D	H	H	OMe	H
E	H	H	OH	OMe

Figure 3: Fluoroquinoline alkaloid derivatives used for the treatment of skin diseases.

Quinoline alkaloids in Figure 3 were also isolated from *Sebastiania corniculate* and 4-methoxy-1,3-dioxolo **7A** was obtained together with its derivative **7B** were found to be of chemotaxonomic importance.[18]

Table 1: Naturally occurring quinoline alkaloids.

Compound	Biological properties	Structure	Source	References
Nitidine **8**	Anti-inflammatory, anti-cancer and anti-tumor		*Zanthoxylum nitidum*	19
Papaverine **9**	Anti-fungal, anti-oxidant and anti-viral		*Opium*	20
Sanguinarine **10**	Skin cancer		*Sanguinaria canadensis*	21
Cincholenine **11**	Anti-malaria		*Cinchona ledgeriana*	22
Quinazoline-quinoline **12**	Cytotoxic, DNA topoisomerasenII inhibitory		*Peganum nigellastrum*	23

1.2 Synthetic quinoline derivatives and their biological activities

Drug discovery efforts from plants have been evolving continuously in response to a number of recent technological advances such as the development of chromatographic methods that allow reproducible and fast purification steps for diverse compound classes, the availability of sensitive spectroscopic methods permitting the structural characterisation of samples in microgram quantities, efficient chemical methods that permit the synthesis, derivatisation, and optimisation of bioactive lead compounds.[24]

The vast range of biological properties associated with quinoline derivatives has led to substantial research devoted to the synthesis of quinoline derivatives with emphasis on their potential medicinal properties.[25] The activities of these compounds are briefly reviewed below.

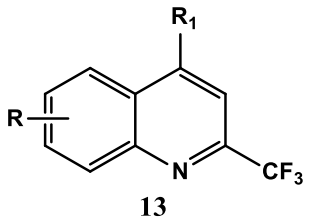
1.2.1 Anticancer Activity

Cancer is the world's top fatal disease, causing millions of deaths worldwide. The disease is predicted to increase if nothing is done to restrain it.[26]

Almost all of the chemotherapy drugs currently available on the market cause severe side effects including an inability to fight infection due to reduced natural immune responses in cancer patients.[27]

Anticancer drugs work on the affected cells thereby eradicating the disease, however, not all types of cancer are curable.[28] Cancer can be termed as a lifestyle disease due to the negative lifestyle of the populations as they live longer. According to the World Health Organisation (WHO), in the next forthcoming years, the mortality rates due to cancer will increase twice its current percentage.[29]

Quinoline derivatives are found to act as anticancer agents targeting topoisomerases (*topo*) *I* and *II* with marked efficiency in solid tumour. There is a growing interest in the synthesis of novel quinoline based molecules in the search of new therapeutic agents. The following quinoline derivatives **13A-13C** shown in Figure 4 exhibit some good activity against cancer.[30]



13

Compound	R	R_1
A	CF_3	Ph
B	OMe	Heteroaryl
C	Cl	Morpholine

Figure 4: Quinoline derivatives with anticancer activities.

1.2.2 Antimalarial Activity

Malaria is a potential blood-borne, deadly tropical disease caused by the parasites of the genus *Plasmodium*, which is transmitted via the bites of the infected mosquitoes. In spite, of the intensive efforts to combat malaria, the incidence of malaria has not reduced, especially in the tropical and subtropical areas.[31]Moreover, the *P. falciparum* strain is resistant to currently available drugs on the market and still spreading in the endemic areas. *In vitro* and *in vivo* resistance even against the most recently introduced artemisinin-based combination therapy (ACT) have also been demonstrated as therapy for uncomplicated *P. falciparum* infections.[32]

The quinoline **1** scaffold is prevalent in a variety of pharmacologically active synthetic and biologically active compounds. Quinoline derivatives are historically among the most important antimalarial drugs ever used.[33]

Chemotherapy still offers the best solution to drug resistance. Novel strategies in antimalarial treatment include the optimisation of therapies with available drugs, repurposing of medicines, developing analogues of existing drugs and the evaluation and use of drug-resistant reverse .[34]

Hence, the development of new chemotherapeutic treatment for malaria disease is urgently needed. [35]

Effective antimalarial agents are currently available such as chloroquine **14**, primaquine **15**, amoediaquine **16**, aablaquine **17,** mefloquine **18**, and quinacrine **19** in

Figure 5. Nevertheless, the development of new antimalarial agents is still required against resistant Plasmodium species.[36]

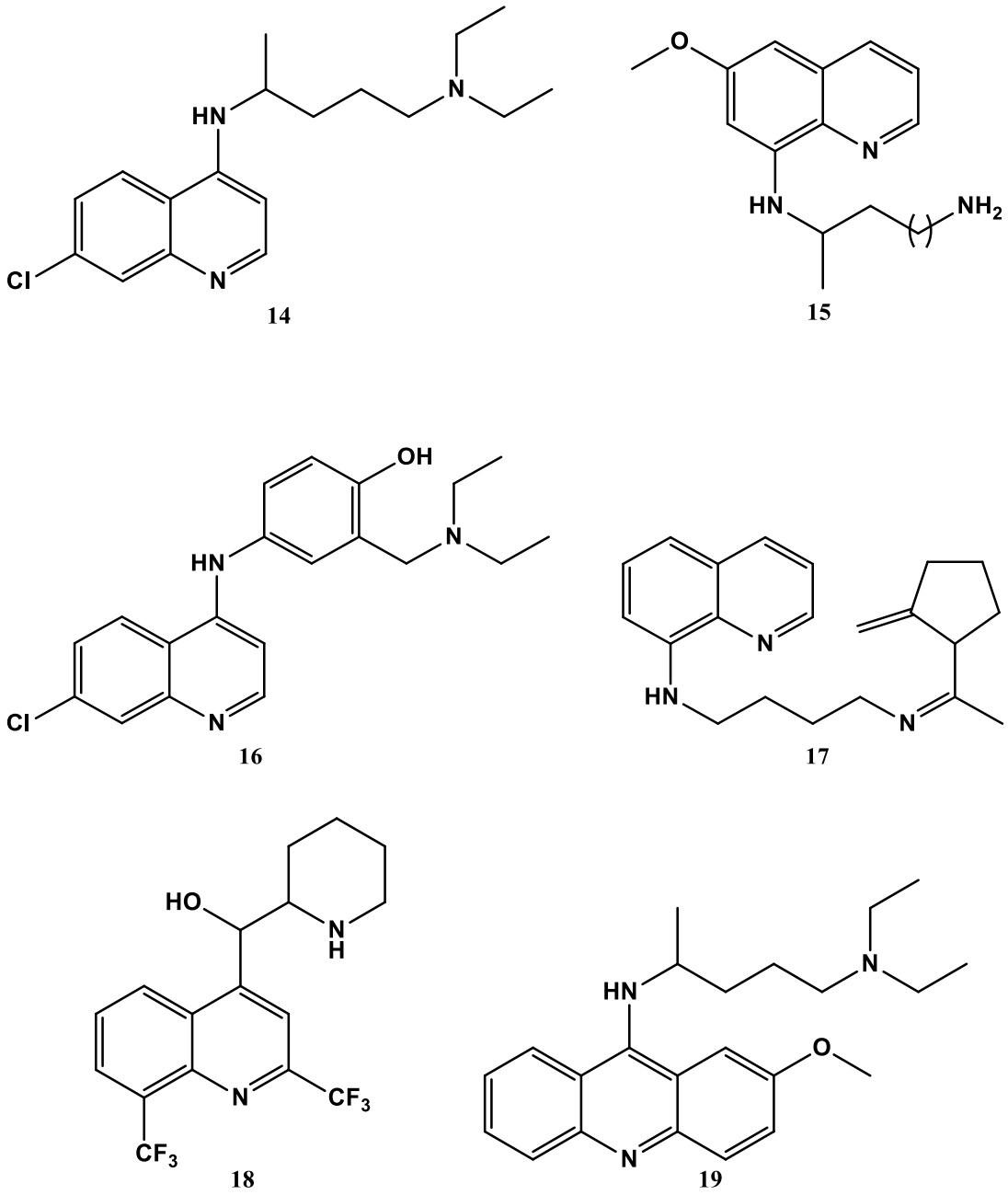


Figure 5. Quinoline drugs with antimalarial activities.

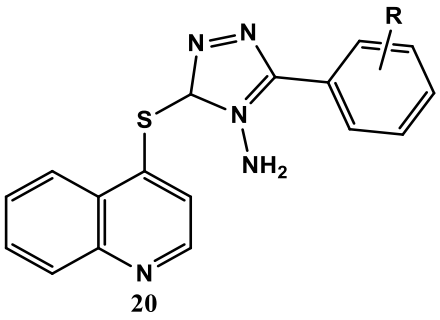
1.2.3 Antimicrobial Activity

Fungal and bacterial infections affect millions of people globally and are associated with high rates of mortality. An antimicrobial is an agent that kills microorganisms or inhibits its growth.[37]

The innovative research for antibiotics has improved humankind's health status by confining life-threatening infections. However, the emergence and spread of bacterial resistance represent a severe global problem. As a result of infectious microbial diseases remain a challenging problem globally because microbes resist therapy than any other form of life.[38]

Anti-infective chemotherapy is the science of administering chemical agents to treat infectious diseases. The use of anti-infective agents can be credited with saving more human lives than any other area of medicinal therapy discovered to date.[39]

The development of new antimicrobial agents is one of the fundamental goals in medicinal chemistry. The search for the discovery and development of strong and selective antimicrobial agents is of great interest with the quinoline derivatives being vital in drug design.[40] A number of quinolines with interesting antimicrobial properties are produced by various *pseudomonads* and other microorganisms. Some new N-[benzildine-2-(quinolin-8-yloxy)] **20** were successfully synthesized as antimicrobial agents in Figure 6.[41]



20

Compound	**R**
A	OCH_3
B	$COCH_3$
C	$(CH_3)_2N$

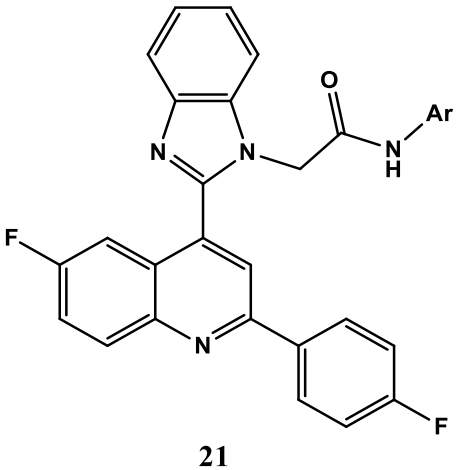
Figure 6. Quinoline derivatives with antimicrobial activities.

1.2.4 Antiinflammatory Activity

Inflammation is a biological response of tissue in attempting self-protection against harmful stimuli caused by a mechanical, biological agent or by an aberrant response.[42] Generally, normal inflammation is rapid and self-limiting but, aberrant resolution and prolonged inflammation cause various chronic disorders[43] such as atherosclerosis, diabetes, cardiovascular and cancer.[44] The search for drugs capable of disrupting the inflammation process has become a pivotal issue in scientific research, especially with the reference to the use of natural substances and the reduction of undesirable side effects[45]

Non-steroidal antiinflammatory drugs (NSAIDs) are widely used for the treatment of inflammation, fever, and pain. Since NSAIDs inhibit both isoforms of cyclooxygenase (COX) (constitute COX-1 responsible for cytoprotective effects and induce COX-2 responsible for inflammatory effects). The quinoline framework became visible as a new template for drug design and the identification of novel antiinflammatory agents. They attract attention owing to their diverse array of pharmacological properties including the ability to target several causes of inflammation.[46]

N-(1H-benzo[d] imidazole-2-yl)-6-fluoro-2-(4-fluorophenyl) quinoline acetamides **21** in Figure 7 was synthesised and were found to be active against inflammation.[47]



21

Compound	**R**
A	C_6H_5
B	$C_6H_4CH_3$
C	$C_6H_4OCH_3$

Figure 7. Quinoline derivatives with antiinflammatory activities.

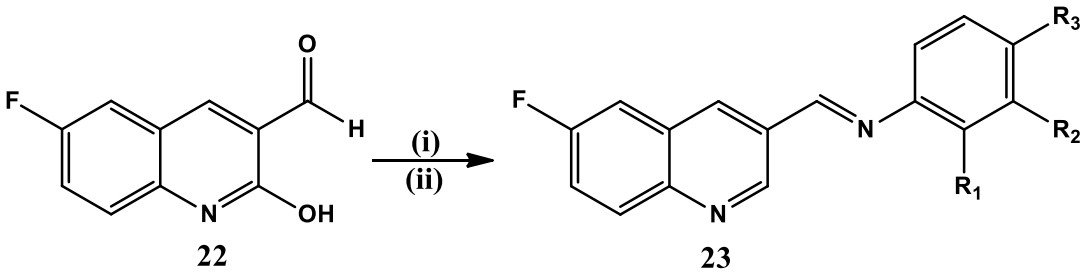
1.2.5 Anti-tuberculosis Activity

Tuberculosis (TB) is a disease of the ancient past caused by infection with members of the *Mycobacterium tuberculosis* complex, which includes *M. tuberculosis (Mtb), M. africanum, M. Bovis, M. caprae, M. microti, M. pinnipedi, and M. canneti*, but the Mtb organism is the main pathogen infecting more than one-third of the world's population. It's an airborne disease affecting millions of people each year being the third leading cause of death globally, after the HIV.[48]

The ascending multidrug resistance among the pathogens has urged the urgency for the development of new agents that can shorten the lengthy TB therapy as well as inhibits the targets involved in persistence.[49] The lengthy treatment, lack of drug supply, underdeveloped health care and appropriate drug prescription or dose leads to increasing the risk of resistance development. All the existing drugs on the market have acquired resistance and cross-resistance.[50]

Tuberculosis is a treatable contagious disease and despite the availability of useful drugs, it continues to kill millions of people each year globally. The two factors responsible for this are; (i) patient non-compliance to existing drug regimens which resulted in the emergence of single drug-resistant strains to all major antituberculosis drugs, (ii) emergence of multidrug-resistant TB (MDR-TB) which is defined as the disease caused by the strains of Mycobacterium tuberculosis resistant to two main initial-line anti-TB drugs, and (iii) association of human immunodeficiency virus (HIV) with TB is the leading cause of mortality among patients who are HIV-positive.[51]

Recently, the quinoline nucleus has gathered immense attention among chemists as well as biologists as it is one of the key building elements for many naturally occurring compounds.[52] The quinoline derivatives in Scheme 1 demonstrated very good anti-tuberculosis activities.



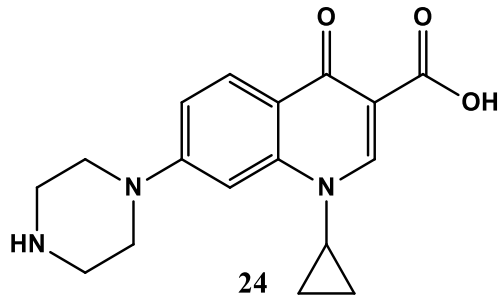
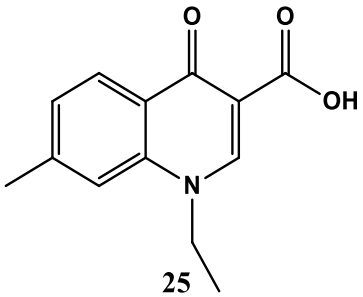
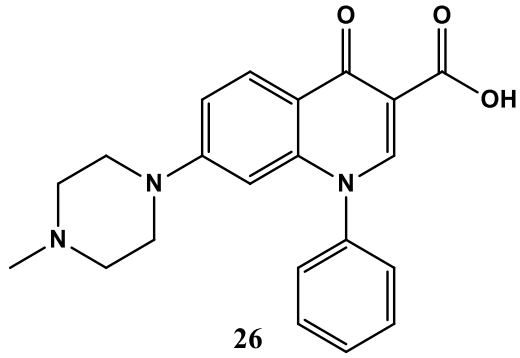
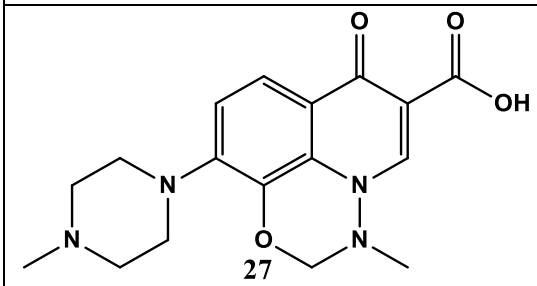
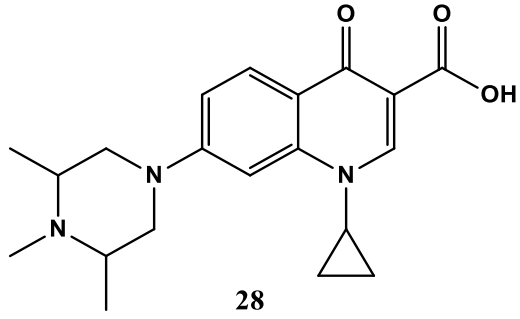
22 23

R_1	R_2	R_3
H	H	F
F	Cl	H
CF_3	H	H
H	CF_3	H
H	CF_3	CN

Reagents: (i) Substituted aniline, (ii) EtOH, reflux, 8 hrs.

Scheme 1: Quinoline derivatives with antituberculosis activity.

Table 2. Quinoline drugs with different activities.

Compound	Biological property	References
 24	Inhibitory activity	53
 25	Murine gammaherpesvirus MVH-68 type	54
 26	Antibiotic	55
 27	Topoisomerase II	56
 28	Antimicrobial	57

1.3 Classical methods for the synthesis of quinolines

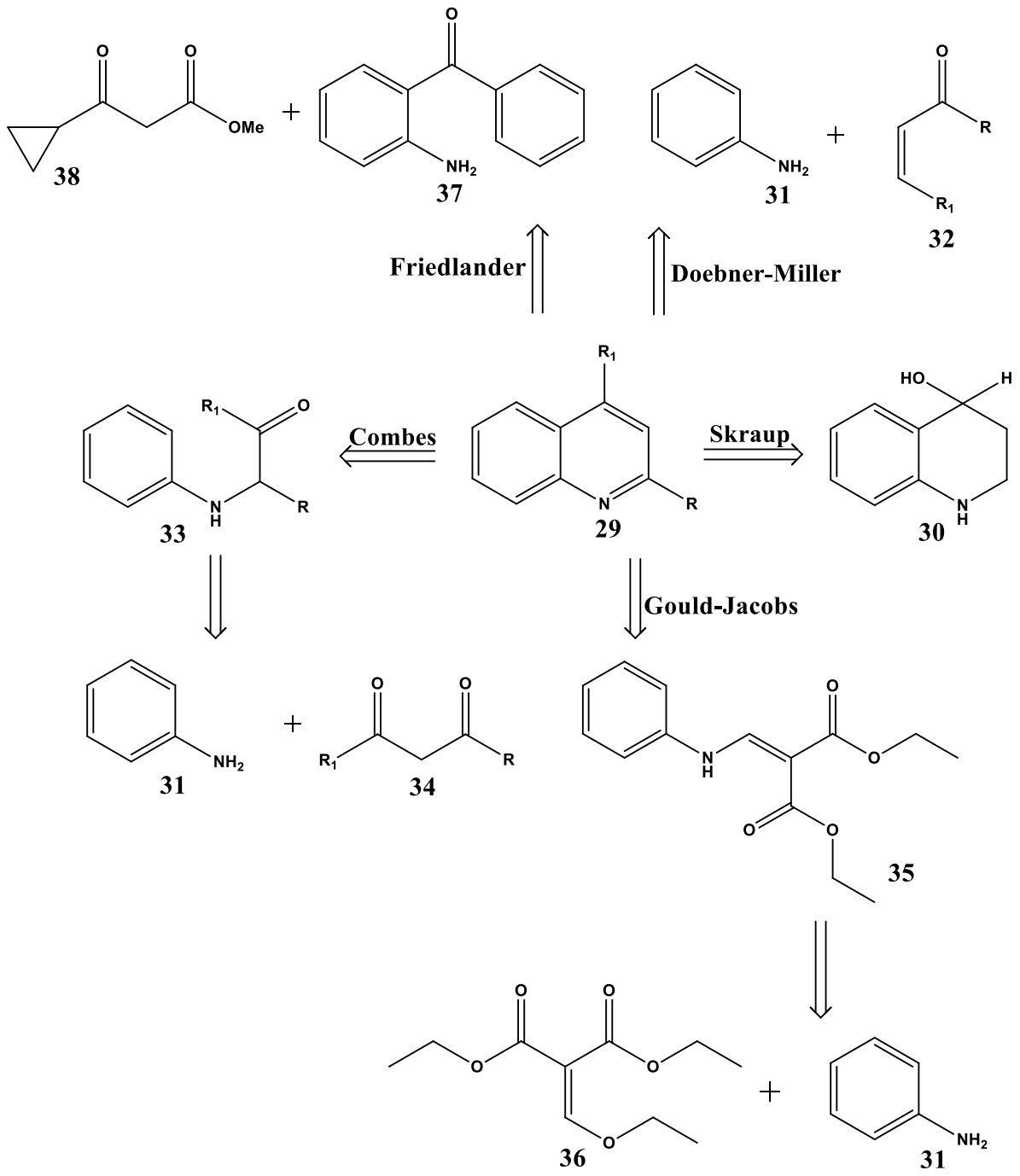
The great attention paid by researchers to the study of quinoline derivatives is explained by the broad range of biological activities highlighted above. Owing to such significance, there has been increasing interest in the development of efficient methodologies for the synthesis of quinoline derivatives. A number of established protocols exist for the synthesis of quinoline compounds which are broken down into classes based on the substitution pattern of the starting materials.[58]

Thus, it is of importance to develop novel preparations for substituted quinolines. Although the previous synthesis for substituted quinolines such as the Knorr synthesis (acid-catalysis)[59], Skraup, Döbner-von Miller, Conrad-Limpach, Friedlaender and Pfitzinger syntheses are still considered as the most useful, they require harsh reaction conditions and the yields are unsatisfactory in most cases.[60-61]

The retrosynthesis strategy in Scheme 2 illustrates the disconnection approach to quinolines. Various methods have been developed for the synthesis of quinolines such as the Combes, Skraup, Friedlander reaction and many other methodologies. Most of these methodologies use aniline as the starting material.[62]

In the following discussion, five different methods used for the synthesis of quinoline derivatives will be briefly reviewed, namely: Combes, Doebner-Miller, Friedlander, Gould-Jacobs and the Skraup reaction.

1.3.1 Retrosynthesis strategy to quinolines

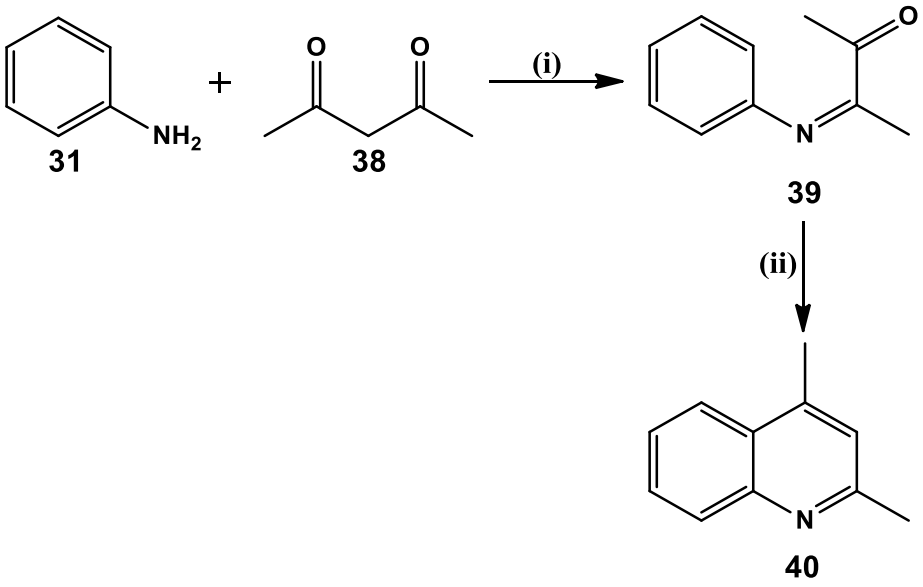


R = R_1 = H, CH_3

Scheme 2: Retrosynthetic strategies to quinolines.

1.3.2 Combes quinoline synthesis

The Combes quinoline synthesis involves condensation of 1,3-dicarbonyl compound with aniline to give the amino enone in high yield, which is cyclized using concentrated acid such as H_2SO_4 to afford 2,4-dimethylquinoline as shown in Scheme 2. In order to synthesize 4-unsubstituted quinoline, a 1,3-keto aldehyde guarantees the regioselectivity as depicted in Scheme 3.[63-65]



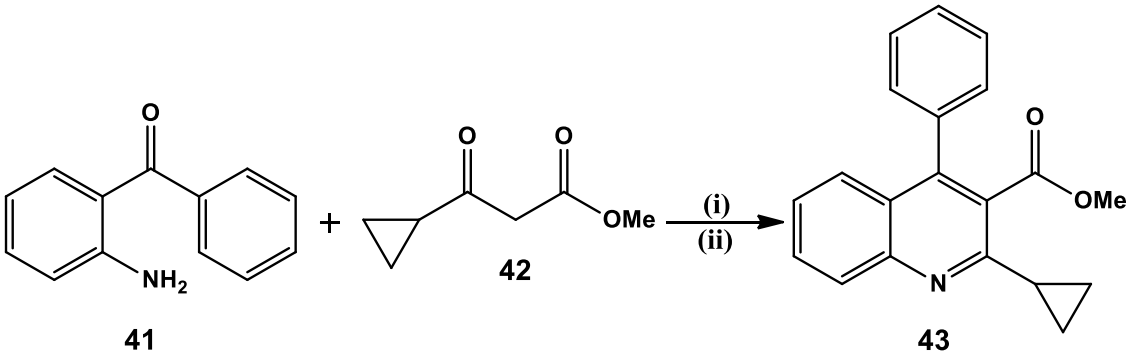
Reagents: (i) Heat, condensation, (ii) Conc. H_2SO_4

Scheme 3: Combes Synthesis.

1.3.3 Friedlander quinoline synthesis

The Friedlander reaction is one of the most simple and straightforward approaches for the synthesis of polysubstituted quinoline derivatives. It involves the reaction of an aromatic *ortho*-amino aldehyde, an aldehyde or ketone, and an alpha methylene functionality. Friedlander reactions are generally carried out by refluxing either aqueous or alcoholic solution of reactants in the presence of a base or by heating a mixture of the reactants at high temperatures ranging from 150 to 220 ˚C in the absence of a catalyst. Under thermal or basic catalysis conditions, ortho-aminobenzophenone fails to react with simple ketones such as cyclohexanone, deoxybenzoin, and β-keto esters.[66]

Treatment of 2-aminobenzophenone **41** with 3-cyclopropyl-3-oxopropionic acid methyl ester **42** in the presence of 5 mol % of NH_2SO_3H catalyst resulted in the formation of 4-phenyl-2-cyclopropylquinoline-3-benzophenone **43** in 95 % yield under solvent-free conditions as shown in Scheme 4.[67]



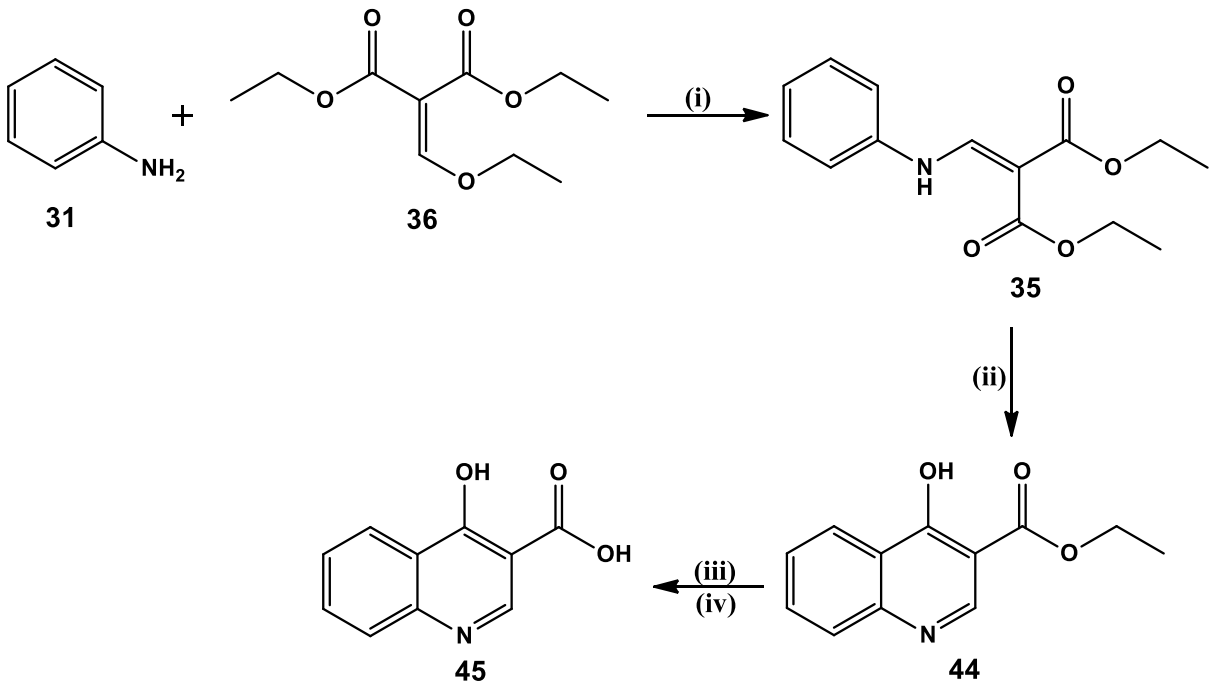
Reagents: (i) NH_2SO_3H, 70 ˚C, (ii) Solvent-free

Scheme 4: Friedlander Synthesis.

1.3.4 Gould-Jacobs quinoline synthesis

The Gould-Jacobs quinoline synthesis is one of the straight-forward method used for the synthesis of quinoline derivatives which showed good antimicrobial activity.[68]

4-hydroxyquinoline-3-carboxylic acid **45** was synthesised starting from aniline **31** and diethyl-2-(ethoxymethylene) malonate **36** by the Gould-Jacobs reaction via cyclisation of the intermediate anilinomethylenemalonate **35** followed by hydrolysis and decarboxylation to obtain 4-hydroxyquinoline-3-carboxylate **44** as illustrated in Scheme 5.[69]

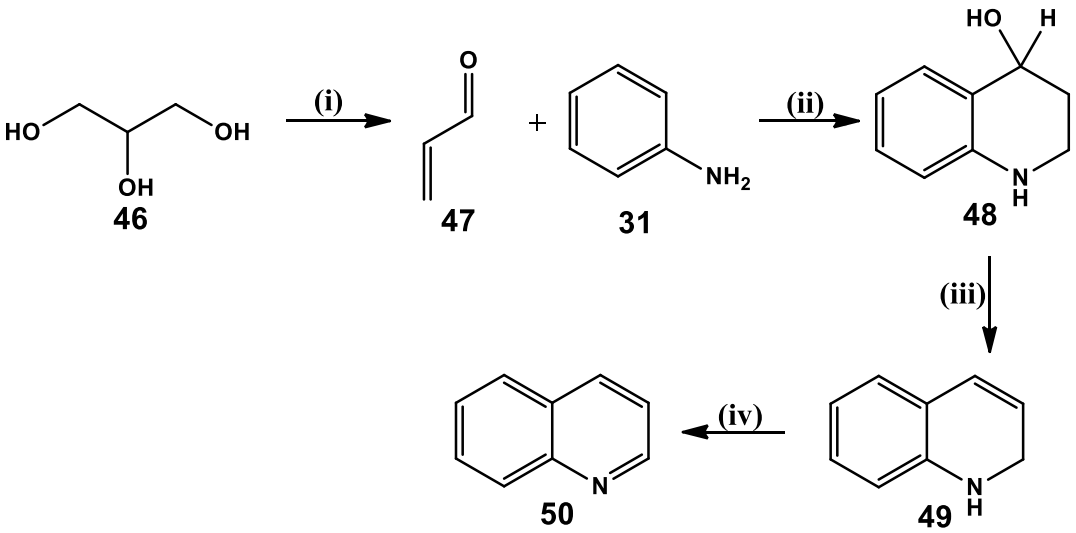


Reagents: (i) EtOH, 80 ^{o}C, (ii) $(C_6H_5)_2O$, 250 ^{o}C, (iii) NaOH, (iv) HCl

Scheme 5: Gould-Jacobs Synthesis.

1.3.5 Skraup quinoline synthesis

The Skraup quinoline synthesis involves the dehydration of glycerine **46** to acrolein **47**, using concentrated sulfuric acid which is then reacted with aniline **31** via conjugate addition forming an intermediate **48**. Dehydration results in ring closure forming 1,2-dihydroquinoline **49**, followed by oxidation to quinoline **50** as shown in Scheme 6. A limitation to this technique results if aniline contains a meta substituent, an isomeric mixture of quinoline is hence formed making it difficult to separate.[70-74]

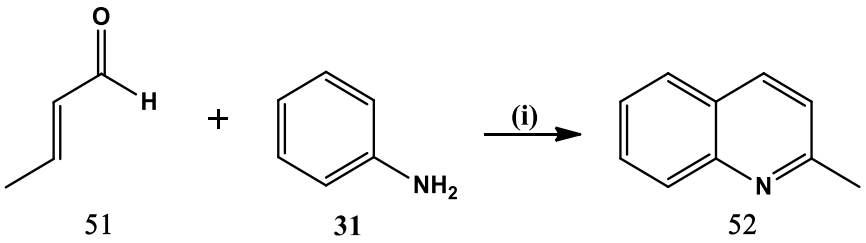


Reagents: (i) H_2SO_4, (ii) Heat, (ii) $-H_2O$, (iv) $+PhNO_2$, $-PhNO_2$.

Scheme 6. Skraup Synthesis.

1.3.6 Döebner-Miller quinoline synthesis

The Doebner-von Miller and Skraup reactions can be arbitrarily combined by replacing the previously used glycerol with α,β-unsaturated aldehydes and ketones and heated with aniline **31** in the presence of an acid catalyst.[75] The first stage of the Döebner-Miller reaction is a crotonic condensation of an aldehyde **51** with aniline **31**, which results in the formation of an α,β-unsaturated 2-methyl quinoline **52** as shown in Scheme 7.[75-76]



Reagents: (i) H_2SO_4

Scheme 7: Döebner-Miller Synthesis.

1.4 Chemistry of 2-chloroquinoline-3-carbaldehyde

2-chloroquinoline-3-carbaldehydes are a group of quinoline derivatives characterised by the attachment CHO functional group to the C-3 position and the chlorine atom at the C-2 position of the quinoline structure.[77] Crucial pharmacological properties have been associated with 2-chloroquinoline-3-carbaldehyde derivatives. These compounds possess a broad spectrum of biological activity such as antimicrobial, antimalarial, antitumor, anti-inflammatory and antiparasitic activity.[78, 79] The main objective of the present investigation was to synthesise novel tetra-substituted quinoline-3-carboxamides derivatives and their biological evaluations.

CHAPTER 2

2. Problem Statement

The quinoline moiety is well-represented in structural natural products, many of which exhibit pharmacological activities.[80] Over the years, several modified approaches have been adopted for the synthesis and isolation of quinolines, then screened for a wide range of the biological spectrum. However, 2-chloroquinoline-3-carbaldehydes derivatives are isolated in limited amounts from natural products and methods to synthesise them and other compounds are in great demand. The search for new methodologies for the synthesis of novel quinoline derivatives continues to be of great interest to organic chemists. In recent years, 2-chloroquinoline-carbaldehydes have attracted considerable attention as highly reactive compounds that serve as the valuable moiety for the incorporation of heterocycles.[81] Heterocycles play a pivotal role in the design of physiological and pharmacologically active compounds. However, there are few methods that make use of mild reaction conditions, short reaction times and cheaper reagents.[82]

The growing mortality rates of incurable diseases such as diabetes, cancer, and HIV-AIDS have been of great concern to organic and medicinal chemists globally. The resistance of antimicrobial agents is a major problem because of the fact microbes acquire the ability to resist antimicrobial drugs by undergoing genetic alterations either by mutations or gene transfer within or between species that allow microbes to defend themselves against the antimicrobial agents. Hence, the discovery of new antimicrobials has assumed critical pivotal to combat the fungal and bacterial infections. This present investigation will focus on the synthesis of novel tetra-substituted quinoline-3-carboxamides derivatives and conduct their biological evaluations as potential antimalarial and cytotoxic agents.

CHAPTER 3

3. Aim and Objectives

The general aim of the study was the synthesis of various novel tetra-substituted quinoline-3-carboxamides, subsequent transformation to other novel compounds and biological evaluation as potential antimalaria and cytotoxicity agents.

The specific objectives of the study were the following:

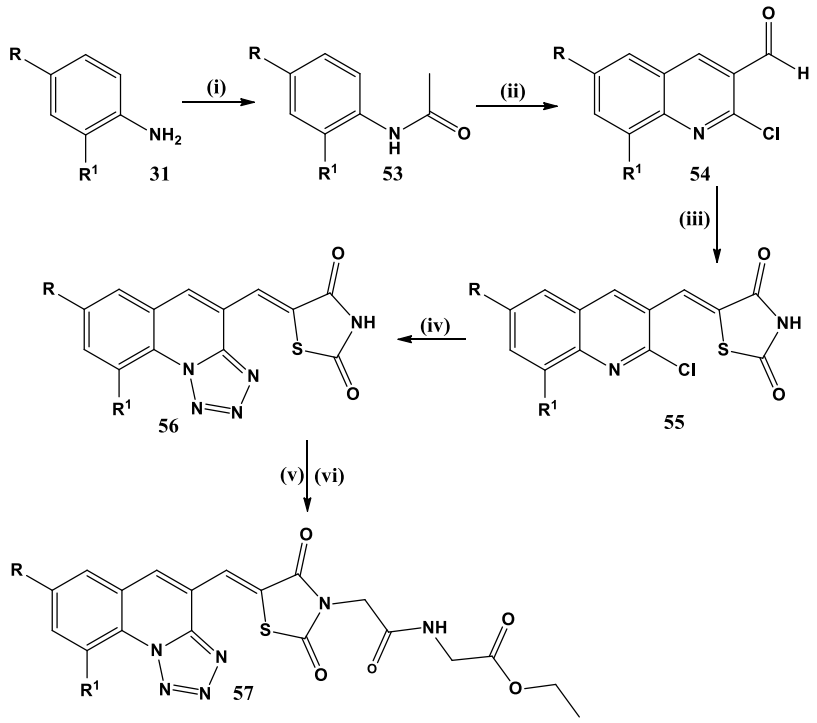
- To synthesise various substituted acetanilides from anilines
- To synthesise various 2-chloroquinoline-3-carbaldehydes from acetanilides
- To transform various 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones from 2-chloroquinoline-3-carbaldehydes
- To transform various (Z)-5-((tetrazolo [1,5a] quinolin-4-yl) methylene) thiazolidinedi-2,4-ones from 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones
- To transform various (Z)-ethyl-2-(2-(2,4-dioxo-5-(tetrazolo [1,5a] quinolin-4-ylmethylene) thiazolidin-3-yl) acetamido) acetates from (Z)-5-((tetrazolo [1,5a] quinolin-4-yl) methylene) thiazolidinedi-2,4-ones
- Biological evaluation of the selected novel target compounds as potential antimalarial and anticytotoxic agents.

CHAPTER 4

4. Results and Discussion

4.1. General

Scheme 8 outlines the proposed synthesis of target compounds (Z)-ethyl-2-(2-5((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**. The synthesised compounds were characterised by using the combination of various spectroscopic techniques namely, 1D NMR (^{1}H, ^{13}C and DEPT), CHN analysis, IR spectroscopy, and HRMS.



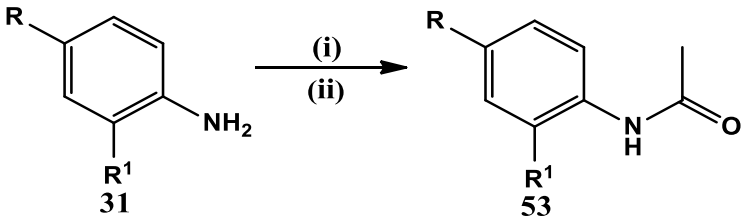
R	H	F	Cl	Br	I	OCH_2CH_3	Br
R^1	H	H	H	H	H	H	Cl

Reagents and conditions (i) Ac_2O, AcOH, reflux, 1-5 hrs, (ii) $POCl_3$, DMF 85 ^{o}C, heat, (iii) AcOH, thiazolidinedi-2,4-one, NaOAc, reflux, 8 hrs (iv) NaN_3, EtOH, 78 ^{o}C, reflux, 2 hrs (v) KOH, EtOH, stir 1 hr (vi) ethyl 2-(2-chloro acetamido) acetate, DCM, 40 °C reflux, 24 hrs.

Scheme 8: Synthesis of the target compounds (Z)-ethyl-2-(2-5((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.

4.1.1 Synthesis of acetanilide analogues 53A-G

The synthesis of acetanilides **53A-G** was conducted following the literature procedure in Scheme 9.[83] In this investigation, commercially available anilines **31** were reacted with acetic anhydride in the presence of glacial acetic acid and refluxed for about 1-5 hours. The resulting mixture was poured over crushed ice-water and the solid product was filtered, dried and recrystallised from methanol to afford the corresponding acetanilides analogues **53A-G** in high yields ranging from 88-94%.

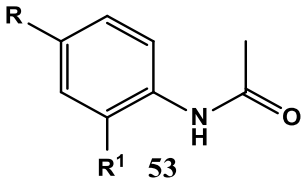


R	H	F	Cl	Br	I	OCH_2CH_3	Br
R^1	H	H	H	H	H	H	Cl

Reagents and conditions (i) Ac_2O, (ii) AcOH, 80 ^oC, reflux, 1-5 h.

Scheme 9: Synthesis of acetanilides analogues **53A-G**.

The NMR confirmed the synthesis of all the acetanilides **53A-G**. The experimental melting points of all the compounds were comparable with those reported in the literature.[83] The melting point and the percentage yields are summarised in table 3.



53

Table 3: Synthesised acetanilides analogues **53A-G**

Compound **53**	R	R^1	% Yield	Melting point (°C)	Lit. Melting point (°C)
A	H	H	93.33	114.1-114.9	114[83]
B	4-F	H	82.20	157.3-157.9	154[83]
C	4-Cl	H	88.68	180.1-180.8	178-179[83]
D	4-Br	H	88.24	168.3-169.8	168[83]
E	4-I	H	85.15	182.3-182.5	184[83]
F	4-OCH_2CH_3	H	90.48	115.2-116.8	114[83]
G	4-Br	2-Cl	89.42	154.4-155.1	151[83]

* *Isolated yields after recrystallisation*

These compounds were fully characterised by using 1 D NMR (^{1}H, ^{13}C NMR), FT-IR spectrum and their physical properties.

The assignment of the protons for compounds **53A-G** was done after consideration of the chemical shifts and coupling constants observed for signals in the ^{1}H NMR spectra.

The ^{1}H NMR spectra of the acetanilide analogues **53A-G** were characterised by the appearance of two singlets. One singlet accounting for three protons and confirming the presence of a methyl group, was observed at high field region ranging from 2.04-2.16 ppm. A second singlet accounting for one proton and confirming NH group in the low field between 7.44-8.36 ppm. For the 4-ethoxy acetanilide **53F** two more peaks were observed, a quartet accounting for two protons at high field region of 4.02 ppm and a triplet accounting for three protons also at a high field region of 1.38 ppm.

Spectroscopic data obtained from ^{13}C NMR was used to confirm the synthesis of the acetanilide analogues **53A-G**. The compounds were established by the appearance

of the characteristic carbonyl carbon (C=O) at low field region ranging from 168.19-168.95 ppm and the methyl carbon (CH_3) appeared at high field region ranging from 24.38-24.83 ppm indicating the introduction of the acyl group of the acetic anhydride resulting in the formation of acetanilide analogues **53A-G**.

For the ethoxy acetanilide **53F** the appearance of the methylene (CH_2) carbon at high field 63.66 ppm and the methyl (CH_3) carbon also at a high field in the aliphatic region 14.83 ppm, thus confirming the successful synthesis of the acetanilide framework.

The chemical structure of the acetanilide analogues **53A-G** was also supported by the FT-IR spectrometry. The stretching of the amine nitrogen (N-H) functionality ranged from 3209.89-3298.47 cm^{-1}, carbonyl carbon (C=O) functionality ranged from 1662.64-1699.29 cm^{-1} and the methene functionality (C=C) was observed in the range 1598.99-1662.64 cm^{-1}.

The ^{1}H NMR spectrum of the acetanilide **53A** in Figure 8 revealed two singlets signals corresponding to the (N-H) proton at 8.36 ppm and the singlet in the aliphatic region at 2.16 ppm integrating for three protons corresponding to the methyl carbon (CH_3). The aromatic protons ranged from 7.09 ppm-7.55 ppm corresponding to protons 2-H, 3-H, 4-H, 5-H, and 6-H.

The ^{13}C NMR spectrum of the acetanilide **53A** revealed a total of 6 carbons which was consistent with the compound as shown in Figure 9.

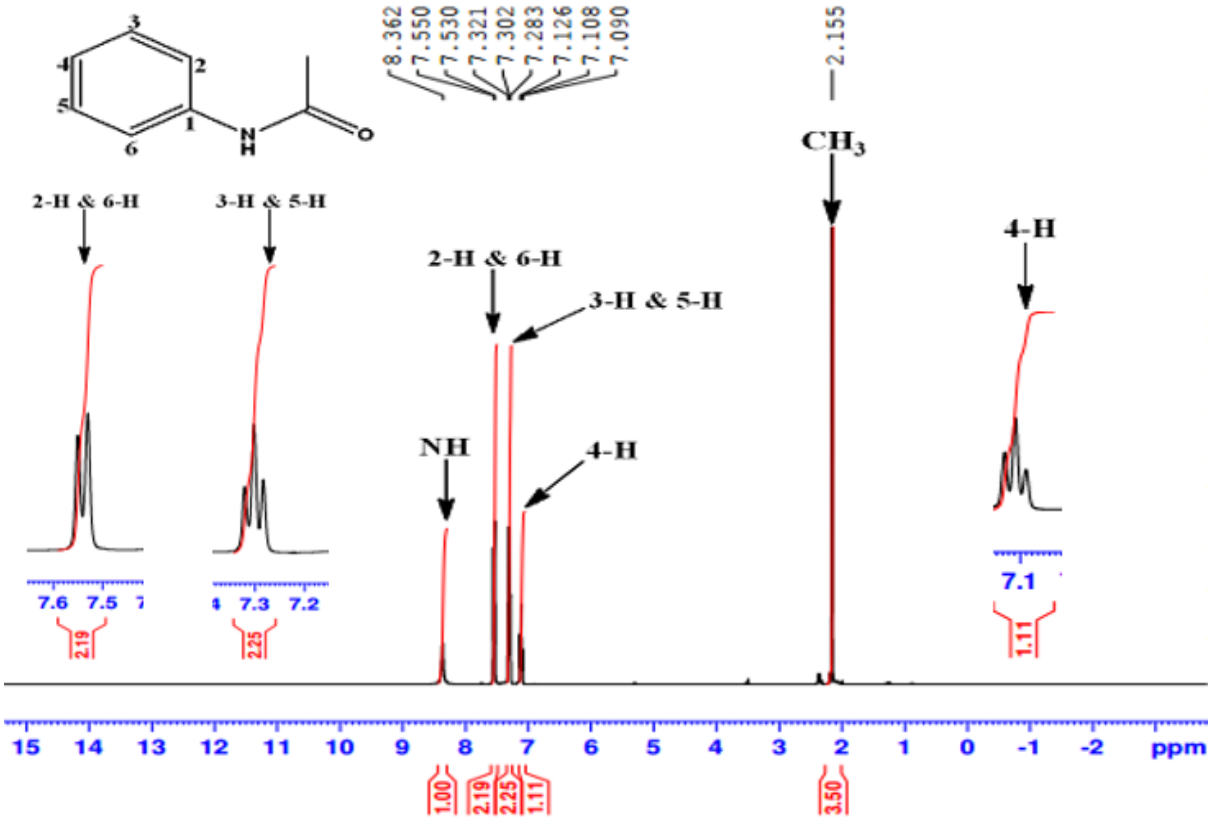


Figure 8: 400 MHz ^{1}H NMR spectrum of acetanilide **53A** in $CDCl_3$

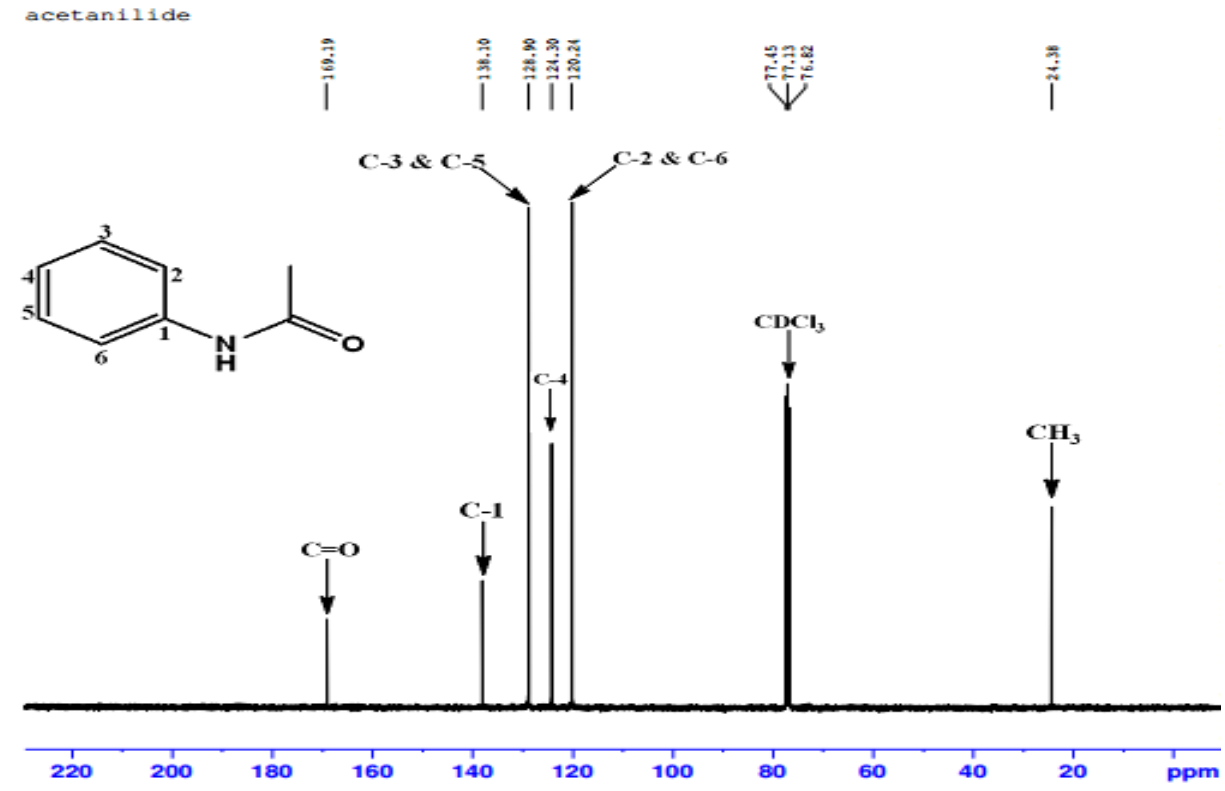


Figure 9: 100 MHz ^{13}C NMR spectrum of acetanilide **53A** in $CDCl_3$

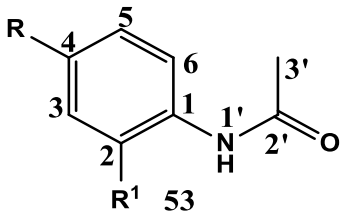
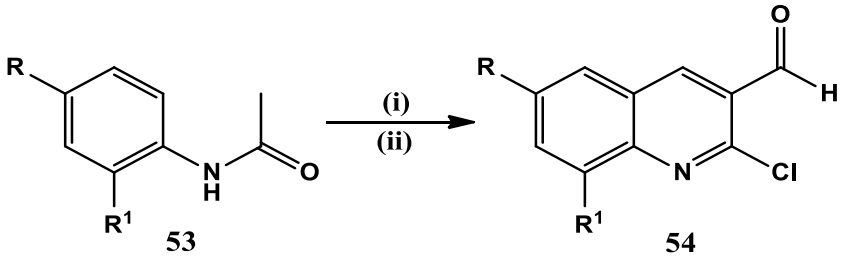


Table 4: 100 MHz ^{13}C NMR chemical shift values (ppm) of the acetanilides **53A-G**.

Nucleus **53**	**53A** **R = H** **R^1 = H**	**53B** **R = F** **R^1 = H**	**53C** **R = Cl** **R^1 = H**	**53D** **R = Br** **R^1 = H**	**53E** **R = I** **R^1 = H**	**53F** **R=OCH_2CH_3** **R^1 = H**	**53G** **R = Br** **R^1 = Cl**
C-1	138.10	159.47	138.71	139.11	139.59	131.01	133.85
C-2	120.24	121.18	120.94	114.96	121.63	122.03	123.43
C-3	128.90	115.50	126.97	131.91	137.75	114.65	122.86
C-4	124.30	157.09	128.98	121.33	86.75	155.72	116.26
C-5	128.90	115.70	126.97	131.91	137.75	114.65	130.74
C-6	120.24	121.10	120.94	114.96	137.75	122.03	131.41
C=O	168.69	168.62	168.90	168.95	168.95	168.69	168.33
CH_3	24.38	24.27	24.41	24.46	24.51	24.19	24.80
OCH_2CH_3	-	-	-	-	-	CH_2 = 63.66 CH_3 = 14.83	-

4.1.2 Synthesis of 2-chloroquinoline-3-carbaldehyde analogues 54A-G

The synthesised acetanilide analogues **53A-G** were subjected to Vilsmeier-Haack reaction by first preparing a mixture of DMF and $POCl_3$ at 0-5 ^oC for 0.5 h maintaining the temperature in an ice-acetone water bath. Acetanilide analogues **53A-G** were slowly added to the above mixture and refluxed for 24 h and then poured into crushed ice-water and stirred for 0.5 hour as shown in Scheme 10.[83] The resulting solid was filtered, dried and recrystallised from ethyl acetate to afford the corresponding 2-chloroquinoline-3-carbaldehyde analogues **54A-G** in low to moderate yield ranging from 10.80-39.94 %.



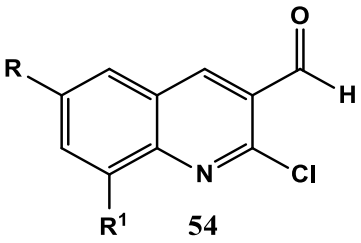
53 54

R	H	F	Cl	Br	I	OCH_2CH_3	Br
R^1	H	H	H	H	H	H	Cl

Reagents and conditions: (i) DMF-$POCl_3$, 85^oC, heat, 24 h.

Scheme 10: Synthesis of 2-chloroquinoline-3-carbaldehyde analogues

The NMR spectroscopy was used to confirm the formation of all the 2-chloroquinoline-3-carbaldehydes **54A-G**. The experimental melting point of all the compounds were comparable with those reported in the literature[83]. The melting point and the percentage yield are summarised in table 5.



54

Table 5: Synthesised 2-chloroquinoline-3-carbaldehyde analogues **54A-G**.

Compound **54**	R	R^1	% Yield	Melting point (^{0}C)	Lit. Melting point (^{0}C)
A	H	H	38.94	144.3-145.8	143[83]
B	6-F	H	24.62	337.6-338.4	339[83]
C	6-Cl	H	10.80	138.8-139.6	138[83]
D	6-Br	H	17.21	187.1-187.7	189[83]
E	6-I	H	18.70	170.8-171.8	169[83]
F	6-OCH_2CH_3	H	19.86	179.2-180.7	178[83]
G	6-Br	8-Cl	34.19	117.8-118.3	-

**Isolated yield after recrystallisation*

These compounds were fully characterised by using 1 D NMR (^{1}H, ^{13}C NMR), FT-IR spectrum and their physical properties.

The ^{1}H NMR spectra of the 2-chloroquinoline-3-carbaldehydes **54A-G** were characterised by the disappearance of a singlet proton at high field region 2.04-2.16 ppm of the methyl carbon (CH_3) and a singlet proton corresponding to the amine (N-H) in the low field at 7.44-8.36 ppm indicating the disappearance of the acetanilides **53A-G**. The appearance of the aldehydic proton (CHO) at 10.56 ppm and the singlet proton (4-H) in the region between 8.55-8.75 ppm in the ^{1}H NMR spectra.

For the 6-ethoxy 2-chloroquinoline-3-carbaldehyde **54F** the appearance of the secondary carbon (CH_2) proton in the high field at 1.53 ppm and the methyl (CH_3) singlet proton also at a high field in the aliphatic region 4.19 ppm.

Spectroscopic data obtained from ^{13}C NMR was used to confirm the number of carbons present in each of the 2-chloroquinoline-3-carbaldehydes **54A-G**. The compounds were established by the disappearance of the characteristic carbonyl carbon (C=O) at low field region ranging from 168.19-168.95 ppm and the disappearance of the methyl carbon (CH_3) in the high field region ranging from 24.38-24.83 ppm. One aldehydic carbonyl carbon (CHO) was observed in the low field region ranging from 172.52-189.42 ppm, thus, confirming the successful synthesis of 2-chloroquinoline-3-carbaldehydes **54A-G**.

The chemical structure of the 2-chloroquinoline-3-carbaldehydes **54A-G** was also supported by the FT-IR spectrometry. The stretching of the carbaldehyde (CHO) functionality was observed at the range of 1681.39-1738.98 cm^{-1} and the methene functionality (C=C) in the range 1635.64-1668.03 cm^{-1}.

6-Bromo-2,8-dichloroquinoline-3-carbaldehyde **54G** was synthesised as a novel compound.

The ^{1}H NMR spectrum of the 2-chloroquinoline-3-carbaldehyde **54A** in Figure 10 revealed one singlet signals corresponding to the aldehyde (CHO) proton at 10.58 ppm. The aromatic protons were observed in the range 8.10 ppm-8.78 ppm.

The ^{13}C NMR spectrum of the 2-chloroquinoline-3-carbaldehyde **54A** revealed a total of 10 carbons which was consistent with the compound as shown in Figure 11.

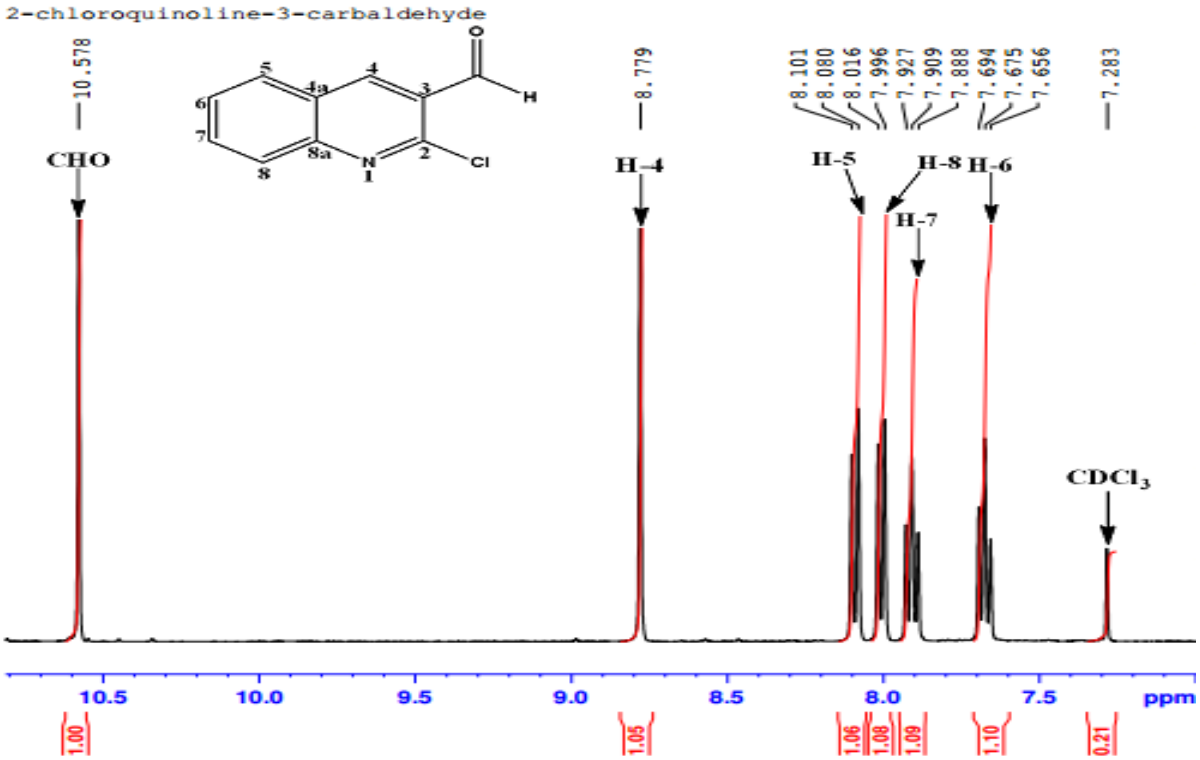


Figure 10: 400 MHz ^{1}H NMR spectrum of 2-chloroquinoline-3-carbaldehyde **54A** in $CDCl_3$.

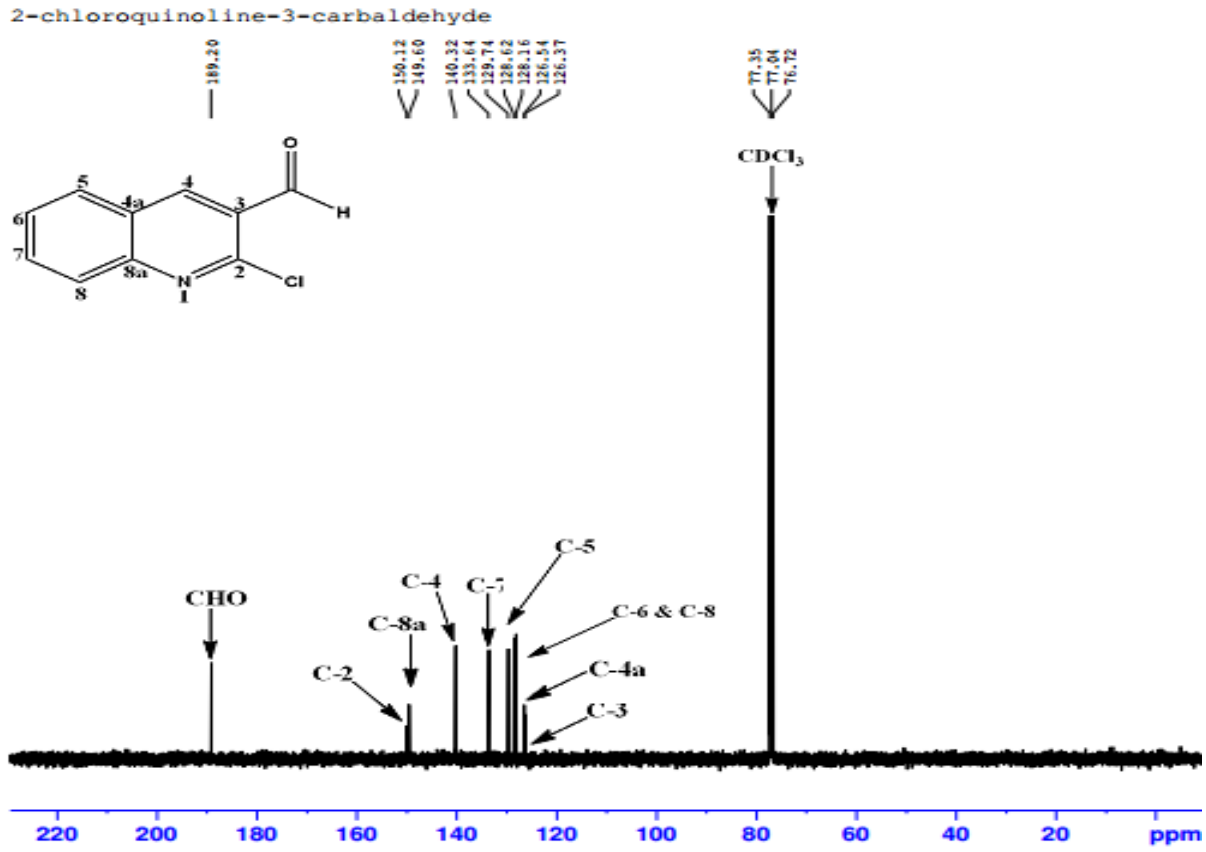


Figure 11: 100 MHz ^{13}C NMR spectrum of 2-chloroquinoline-3-carbaldehyde **54A** in $CDCl_3$.

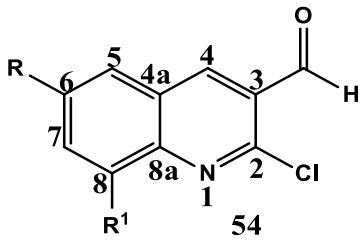


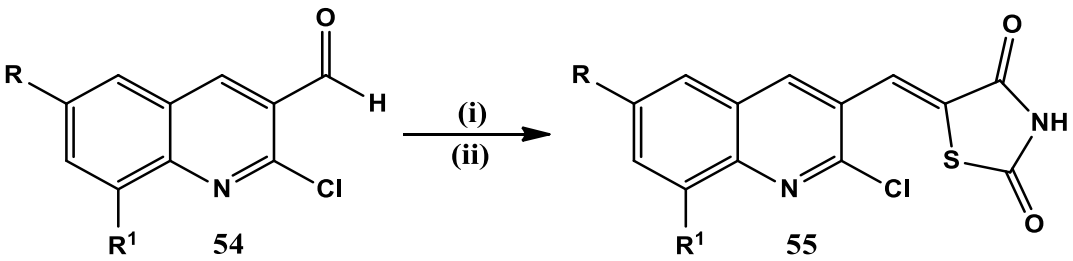
Table 6: 100 MHz ^{13}C NMR chemical shift values (ppm) of the 2-chloroquinoline-3-carbaldehydes **54A-G**, *(**54A**, **54F** and **54G** in $CDCl_3$ and **54B, 54C, 54D, 54E** in DMSO-d_6).

Nucleus **54**	**54A** **R = H** **R^1= H**	**54B** **R = F** **R^1= H**	**54C** **R = Cl** **R^1 = H**	**54D** **R = Br** **R^1 = H**	**54E** **R = I** **R^1 = H**	**54F** **R=OCH_2CH_3** **R^1 = H**	**54G** **R = Br** **R^1 = Cl**
C-2	150.12	159.78	150.32	150.44	150.56	147.51	156.57
C-3	126.37	127.37	127.02	127.63	126.86	126.33	-
C-4	140.32	139.53	139.19	139.09	142.19	138.60	132.81
C-4a	126.54	146.65	127.15	127.01	128.10	127.77	129.87
C-5	129.74	112.76	128.10	131.48	138.15	107.04	128.73
C-6	128.16	162.29	134.09	122.14	93.67	158.13	121.11
C-7	133.64	123.96	134.42	136.96	138.85	126.83	131.85
C-8	128.62	131.21	130.13	130.18	130.03	129.83	134.82
C-8a	149.60	149.45	147.89	148.11	148.45	145.68	-
CHO	189.20	188.93	188.80	188.76	188.74	189.45	172.52
OCH_2CH_3	-	-	-	-	-	CH_2 = 64.15 CH_3 = 14.61	-

** Isolated yield after recrystallisation*

4.1.3 Synthesis of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues 55A-G.

The synthesis of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones **55A-G** was conducted using the modification of the literature procedure as shown in Scheme 11. In this investigation, 2-chloroquinoline-3-carbaldehydes **54A-G** were subjected to thiazolidinedi-2,4-one, AcOH and anhydrous sodium acetate and refluxed the mixture for 8 hours and poured the resulting mixture in crushed ice-water. The resulting solid was filtered, dried and recrystallised from DMF to afford the corresponding 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55A-G** in moderate yield ranging from 19.42-53.49 %.[84]



R	H	F	Cl	Br	I	OCH_2CH_3	Br
R^1	H	H	H	H	H	H	Cl

Reagents and conditions: (i) Thiazolidinedi-2,4-one, (ii) AcOH, 110°C, reflux, 8 hrs

Scheme 11: Synthesis of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55A-G**.

Most of the synthesised 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55A-G** are novel, as such 1D NMR (^{1}H, ^{13}C, DEPT), CHN analysis, HRMS and IR spectroscopic methods were used to confirm the structures of these compounds.

The NMR spectroscopy confirmed the structures of all the synthesised 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55A-G**. The melting point and the percentage yield are summarised in table 7.

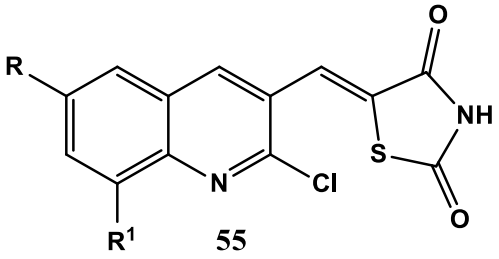


Table 7: Synthesised 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55A-G**.

Compound **55**	**R**	**R^1**	% Yield	Melting point (°C)	Lit. Melting point (°C)
A	H	H	44.19	219.3-220.0	221[84]
B	6-F	H	41.38	336.7-337.5	-
C	6-Cl	H	42.57	349.5-350.4	-
D	6-Br	H	53.49	345.2-345.6	-
E	6-I	H	27.36	317.4-318.4	-
F	6-OCH_2CH_3	H	40.32	287.9-288.4	-
G	6-Br	8-H	19.42	128.8-129.3	-

**Isolated yield after recrystallisation*

These compounds were fully characterised by using 1 D NMR (^{1}H, ^{13}C NMR, DEPT), CHN analysis, HRMS, and IR spectroscopy.

The ^{1}H NMR spectra of the 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** was characterised by the disappearance of the singlet proton 4-H at the low field 10.58 ppm of the carbaldehyde proton (CHO) and carbonyl carbon (C=O) signal at 189.20 ppm of the 2-chloroquinoline-3-carbaldehyde **54A**. The appearance of the broad signal at 12.26 ppm of the N-H from thiazolidinedi-2,4-one, methylene proton 6'-H at 10.25 ppm in the ^{1}H NMR spectra, two carbonyl carbons (C=O) signals at 161.90, 160.82 and methylene carbon (C-6', ArCH=C) signal at 190.25 in the ^{13}C NMR spectra, respectively, confirmed the formation of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones **55A-G**.

The ^{1}H NMR and ^{13}C NMR of the six novel 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones **55B-G** were in agreement with 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A**. The broad signal of the N-H from the thiazolidinedi-2,4-

one appeared at the range of 10.00-12.37 ppm and the methylene proton H-6' signal at 8.37-10.25 ppm in the ^{1}H NMR spectra.

Only ten carbons of the 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones **55B-G** of the expected thirteen carbons appeared in the ^{13}C spectra NMR. One carbonyl (C=O) and two quarternary carbons were missing from the ^{13}C NMR spectra attributed to the fact that unsaturated compounds tend to have peaks overlapping with each other, and that the quaternary carbons are of low intensity in nature and needs longer running periods (as in this case they were run for thirty-six hours) still couldn't appear on the ^{13}C NMR. The other reasons are that the ^{13}C NMR signals are slow relaxation and broad signals. The broad signals are due to one or more quadrupolar nucleus, the intermediate rate exchange processes (tautomeric equilibrium) and the C=N bond could be so in this case.[85] Other methods of analysis such as CHN and HRMS analysis were used in this regard to confirm the successful synthesis of the six novels 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55B-G**.

The ^{1}H NMR spectrum of the 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** in figure 12 revealed two singlet signals corresponding to the N-H proton at 12.26 ppm and the other singlet corresponding to the methylene proton 6'-H at 10.25 ppm. The aromatic protons were observed in the range of 7.64-8.50 ppm for 5-H, 6-H, 7-H and 8-H respectively.

The ^{13}C NMR spectrum of the 2-chloroquinoline-3-carbaldehyde **55A** revealed a total of 13 carbons which was consistent with the compound as shown in Figure 13.

The IR spectrum of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** revealed the amine stretch at 3093.05 cm^{-1}, nitrile (ArC=N) stretch at 2626.56 cm^{-1} and the alkene (ArC=C) stretch at 1619.46 cm^{-1}.

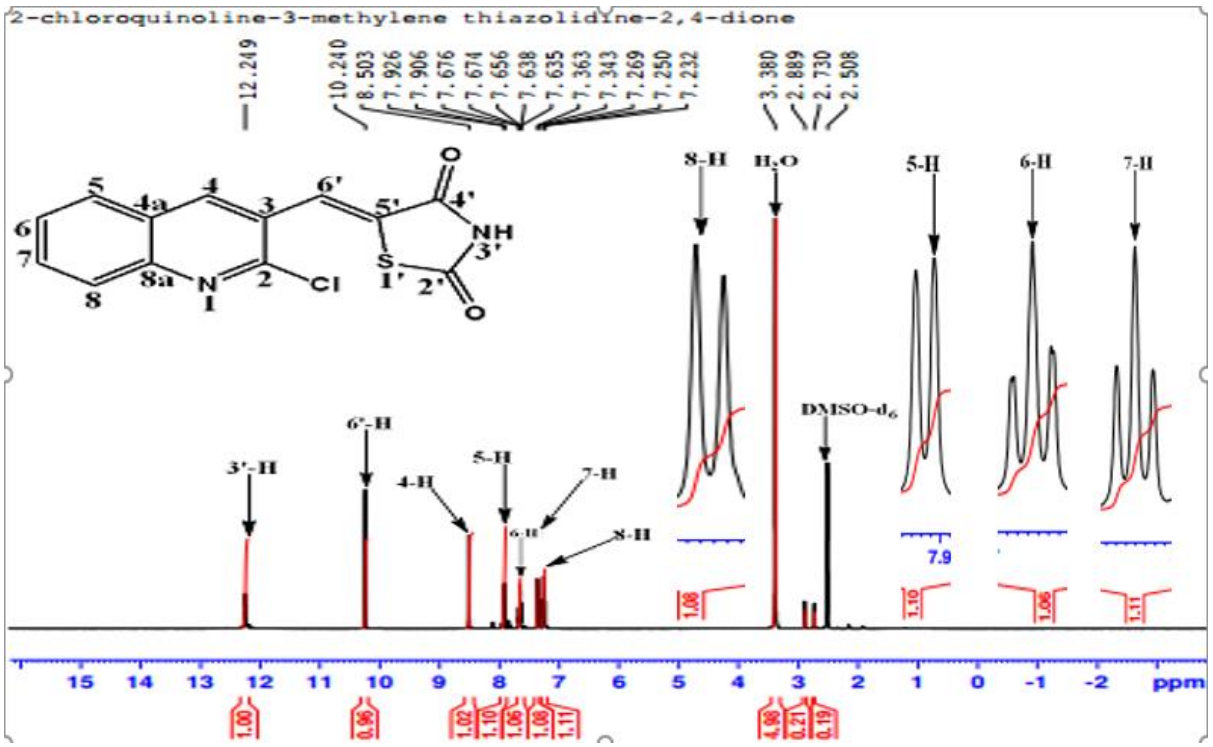


Figure 12. 400 MHz ^{1}H NMR spectrum of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** in DMSO-d_6.

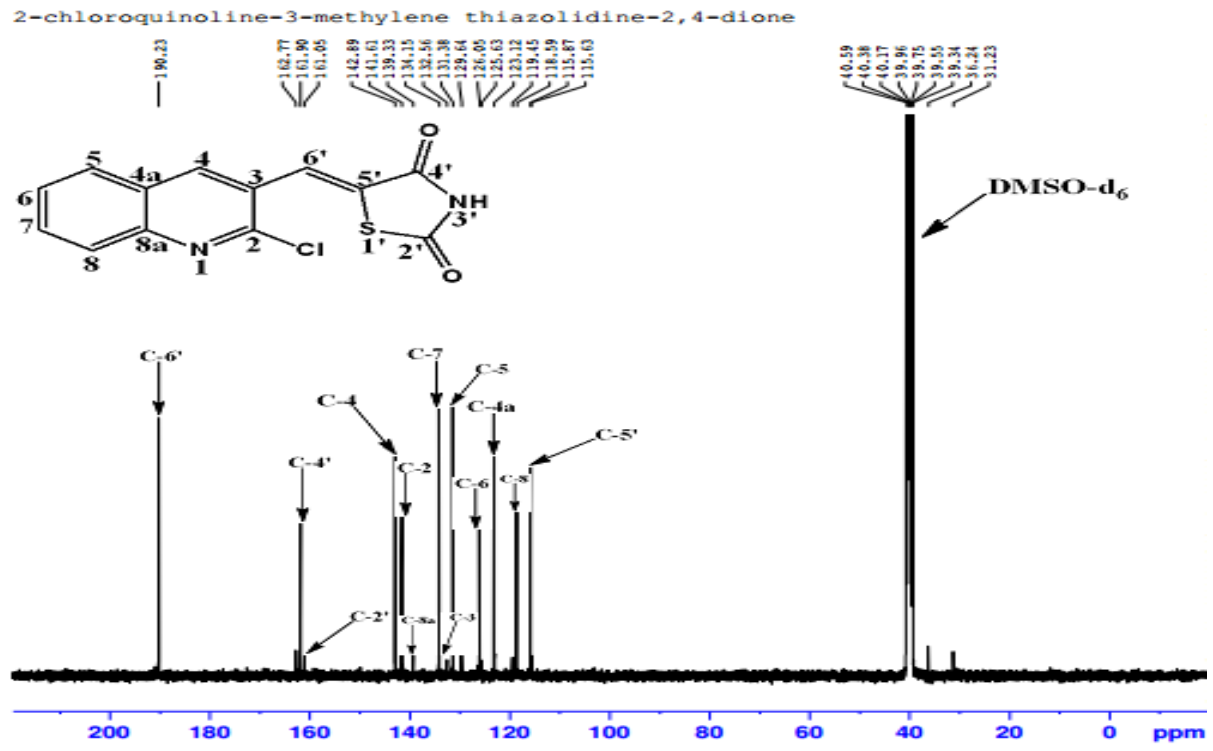


Figure 13: 100 MHz ^{1}H NMR spectrum of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** in DMSO-d_6.

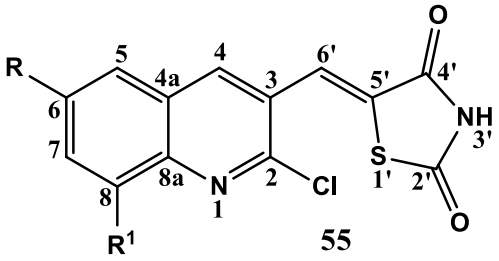


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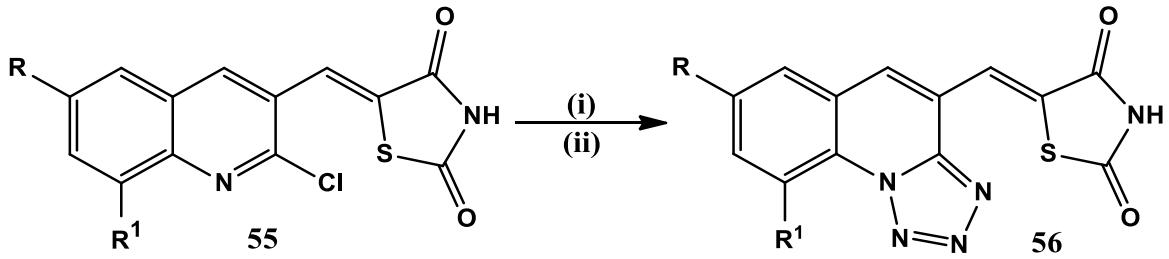
Table 8. 100 MHz ^{13}C NMR chemical shift values (ppm) of the 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones **55A-G** in DMSO-d_6.

Nucleus **55**	**55A** R = H R^1= H	**55B** R = F R^1 = H	**55C** R = Cl R^1 = H	**55D** R = Br R^1 = H	**55E** R = I R^1 = H	**55F** R=OCH_2CH_3 R^1 = H	**55G** R = Br R^1 = Cl
C-2	141.61	156.36	140.22	-	141.53	136.34	134.40
C-3	132.56	119.21	126.84	126.83	142.33	119.16	-
C-4	142.89	141.99	141.73	141.67	141.79	142.28	132.04
C-4a	119.45	126.87	119.69	120.23	120.75	-	-
C-5	125.63	115.60	117.83	118.04	126.06	112.22	124.98
C-6	123.32	158.74	-	114.53	86.23	154.19	116.58
C-7	134.35	117.94	133.82	136.40	139.06	117.24	131.10
C-8	118.59	122.57	129.96	133.00	118.01	124.45	-
C-8a	139.33	138.40	126.90	140.51	140.87	126.11	124.82
C-6’	190.25	190.22	190.12	190.09	190.06	190.36	160.97
C-2’	162.77	161.61	161.66	161.64	161.62	161.53	-
C-4’	161.05	-	-	-	-	-	-
C-5’	115.63	-	-	-	-	-	-
CH_2	-	-	-	-	-	64.02	-
CH_3	-	-	-	-	-	15.00	-

4.1.4 Synthesis of novel (Z)-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues 56A-G

The synthesis of the novel (Z)-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-ones **56A-G** was conducted as outlined in Scheme 12. In this investigation, 2-chloroquinoline-3-methylene thiazolidinedi-2,4-ones **55A-G** were subjected to NaN_3, EtOH and AcOH and refluxed the mixture for 2 hours and thereafter cooled and poured the resulting mixture into cold EtOH-DMF (3:2).[86] The resulting solid was filtered, dried and recrystallised from EtOH-DMF (3:2) to afford the corresponding novel (Z)-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues **56A-G** in moderate to excellent yield ranging from 36.11-88.57 %.



R	H	F	Cl	Br	I	OCH_2CH_3	Br
R^1	H	H	H	H	H	H	Cl

Reagents and conditions: (i) NaN_3, (ii) EtOH-AcOH, reflux, 2 hrs

Scheme 12: Synthesis of novel (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues **56A-G**.

The NMR spectroscopy confirmed the synthesis of all the novel (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues **56A-G**. The melting points and the percentage yields are summarised in table 9.

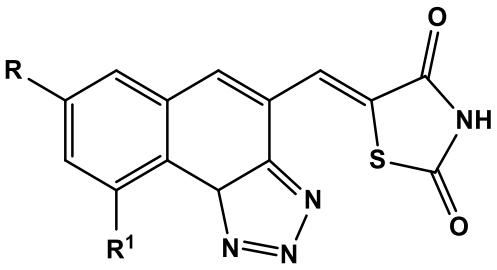


Table 9. Synthesised (Z)-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues **56A-G**.

Compound **4**	R	R^1	% Yield	Melting point (°C)	Lit. Melting point (°C)
A	H	H	76.42	313.8-314.2	-
B	7-F	H	60.95	240.9-241.8	-
C	7-Cl	H	36.11	359.6-360.9	-
D	7-Br	H	88.57	346.3-347.1	-
E	7-I	H	67.86	320.1-320.7	-
F	7-OCH_2CH_3	H	48.33	291.0-291.5	-

**Isolated yield after recrystallisation*

These novel compounds were fully characterised by using 1D NMR (^{1}H, ^{13}C NMR, DEPT 135), CHN, HRMS and IR spectroscopic methods were used to confirm the structures of these compounds.

The ^{1}H NMR spectra of the (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** was characterised by the appearance of the broad signal at 12.36 ppm of the N-H from thiazolidinedi-2,4-one, methylene signal (6'-H, ArCH=C) at 10.21 ppm in the ^{1}H NMR spectra. In the ^{13}C spectra one carbonyl carbon (C=O) signal at 161.66 ppm, one methylene carbon (C-6', ArCH=C) signal at 190.10 ppm, and the shifting of the C-3 signal from 140.49 ppm-142.33 ppm towards the high field region in the ^{13}C NMR spectra was observed respectively and confirmed the formation of novel (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues **56A-F**.

Although the ^{1}H NMR confirmed the successful synthesis of (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D**, three carbons (one carbonyl carbon (C=O) and two quarternary carbons) of the thirteen expected carbons

were missing in the ^{13}C NMR spectra as discussed in the previous discussion (refer to 4.1.3) above. The successful synthesis of this compound was confirmed by the ^{1}H, ^{13}C NMR, CHN analysis, DEPT 135, HRMS and IR spectroscopy.

The ^{1}H NMR and ^{13}C NMR of the five novel (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-ones **56A**, **56B**, **56C**, **56E**, and **56F** was in agreement with (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D**. The broad signal of the 6'-H (N-H) of the thiazolidinedi-2,4-one appeared at the range of 12.17-12.36 ppm and the methylene signal (6'-H, ArCH=C) at 10.20 ppm-10.24 ppm in the ^{1}H NMR spectra.

Only ten carbons of the (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-ones **56A**, **56B**, **56C**, **56E**, and **56F** of the expected thirteen carbons appeared in the ^{13}C NMR spectra. One carbonyl and two quarternary carbons were missing from the ^{13}C spectra attributed to the fact that unsaturated compounds tend to have peaks overlapping with each other, and that the quaternary carbons are of low intensity in nature and needs longer period (in this case they were run for three days) still couldn't appear on the ^{13}C NMR. The other reasons are that the ^{13}C NMR signals are slow relaxation and broad signals. The broad signals are due to one or more quadrupolar nucleus, the intermediate rate exchange processes (tautomeric equilibrium) and the C=N bond could be so in this case.[87] Other methods of analysis such as CHN and HRMS analysis were used in this regard to confirm the successful synthesis of the five novels (Z)-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56A**, **56B**, **56C**, **56E**, and **56F**.

The ^{1}H NMR spectrum of the (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** in Figure 13 revealed one singlet signal corresponding to the N-H proton at 12.36 ppm and the other singlet corresponding to the methylene proton (6'-H, ArCH=C) 10.25 ppm. A singlet at 8.46 ppm was observed for proton 5-H. A doublet at 8.17 ppm (J_{H-H} meta = 2.00 Hz) corresponds to the 6-H obtaining its splitting pattern from proton 8-H. A doublet of a doublet at 7.79 ppm (J_{H-H} meta and ortho = 2.40 Hz and 8.80 Hz) corresponding to the 8-H was observed obtaining its splitting pattern from proton 6-H and 9-H. A doublet at 7.30 ppm (J_{H-H} ortho was observed for proton 9-H which obtained its splitting pattern from proton 8-H as shown in Figure 14.

The assignments of the carbons were done using DEPT 135 and ^{13}C NMR. The methylene carbon (C-6') was observed at 190.10 ppm, though it's region was not included in DEPT 135. The aromatic carbons (CH, ArC) were observed at the following chemical shifts, 118.05 ppm (C-6), 132.99 ppm (C-9), 136.42 ppm (C-8) and 141.17 ppm (C-5) as shown in Figure 15.

The IR spectrum of (Z)-5-((7-bromotetrazolo [1,5a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** revealed the amine (N-H) stretch at 3132.57, nitrile (ArC=N) stretch at 2025.12 cm^{-1} and alkene (ArC=C) stretch at 1609.58 cm^{-1} as shown in Figure 17.

The ^{13}C NMR spectrum of the (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** revealed a total of ten carbons which was consistent with the compound as shown in Figure 16.

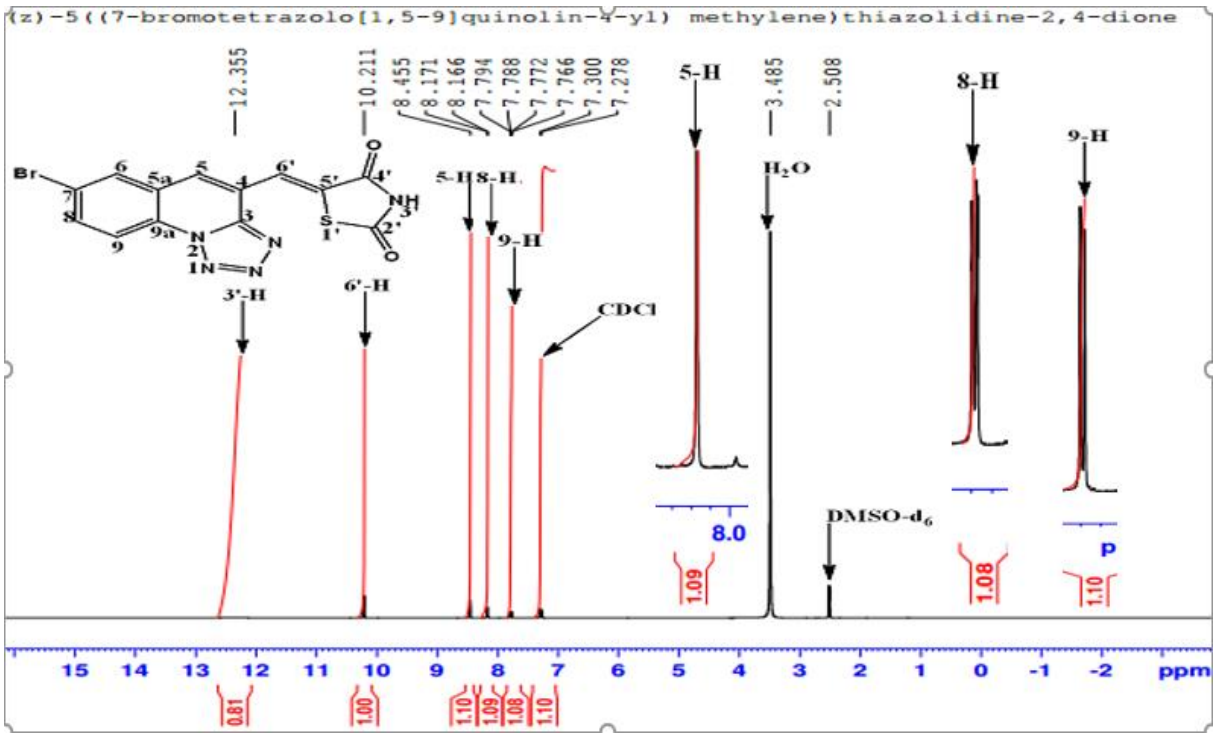


Figure 14: 400 MHz ^{1}H NMR spectrum of (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** in DMSO-d_6.

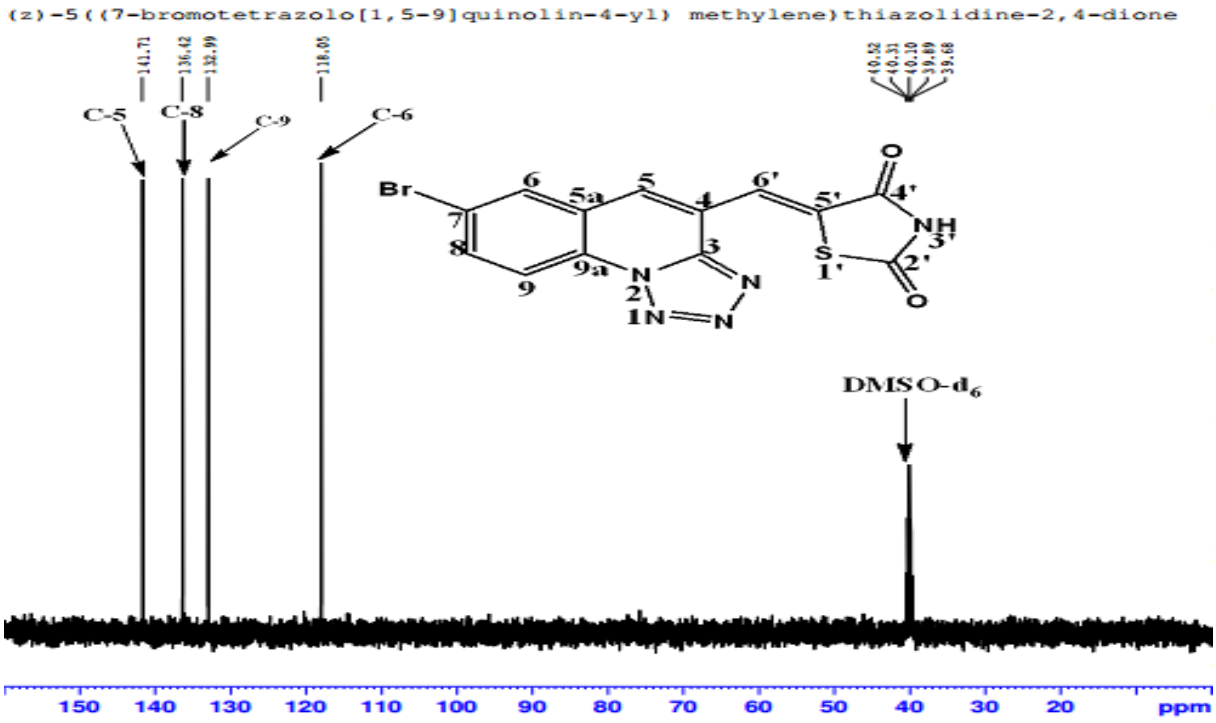


Figure 15: DEPT 135 spectrum of (Z)-5-((7-bromotetrazolo [1,5a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D**.

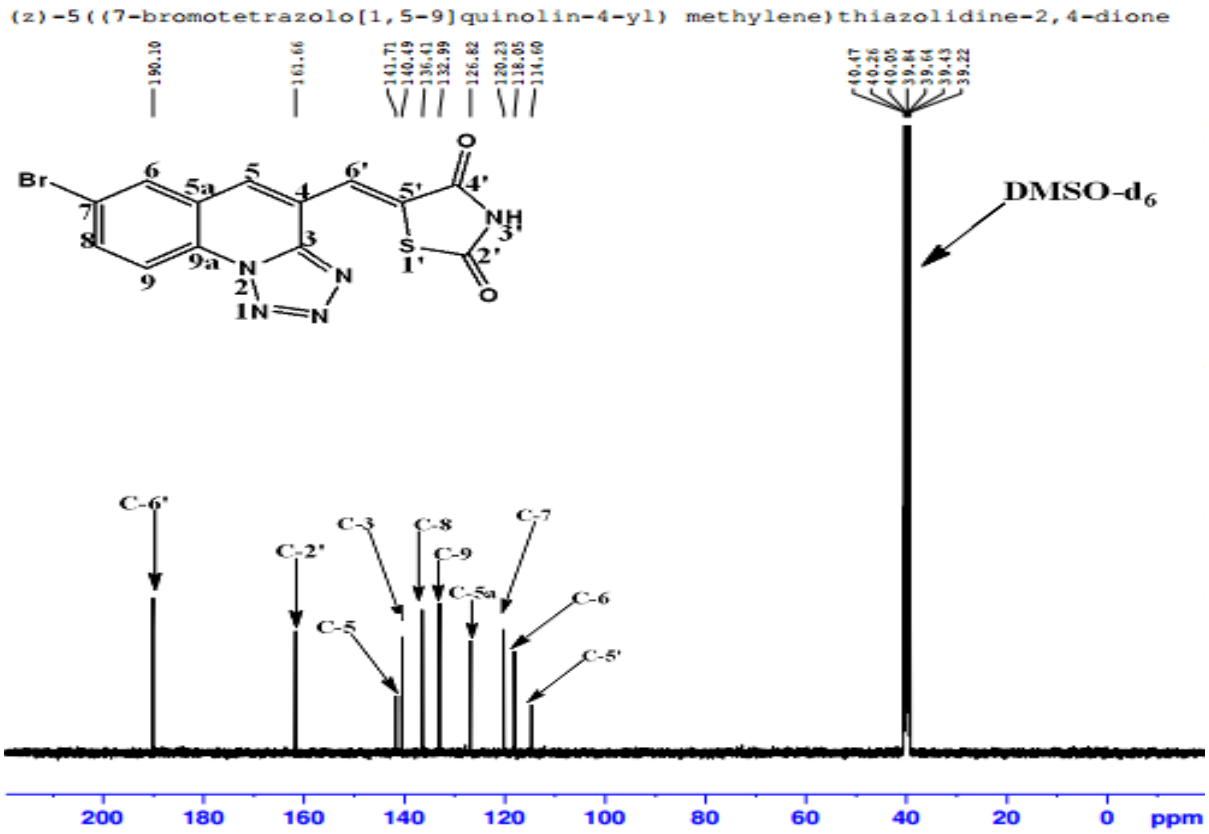


Figure 16: 100 MHz ^{1}H NMR spectrum of (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** in DMSO-d_6.

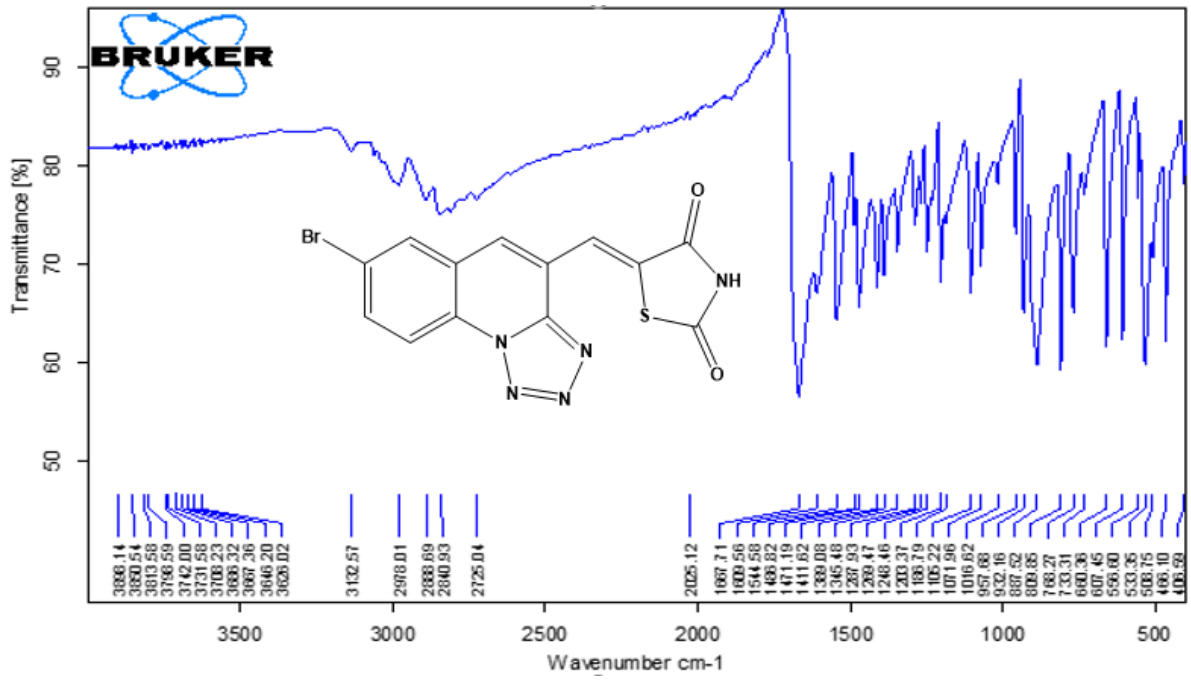


Figure 17: IR spectrum of (Z)-5-((7-bromotetrazolo [1,5a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D**.

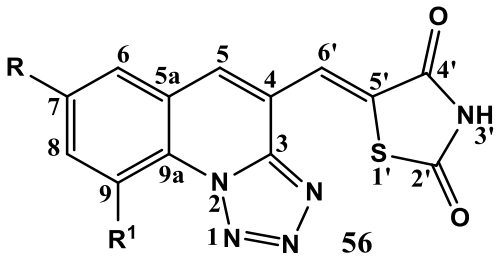
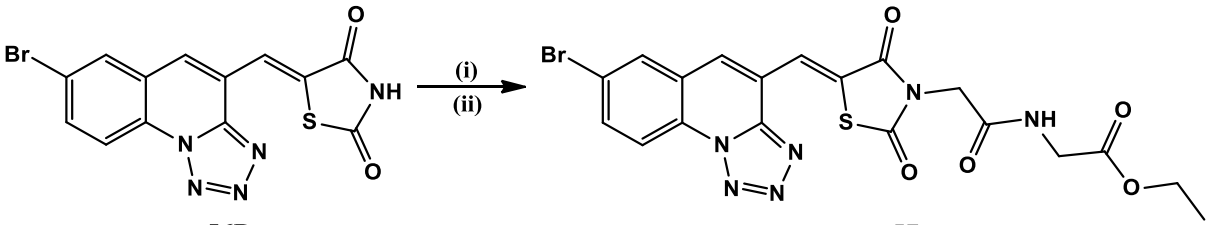


Table 10. 100 MHz ^{13}C NMR chemical shift values (ppm) of the (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56A-F** in DMSO-d_6.

Nucleus **56**	**56A** **R = H** **R^1 = H**	**56B** **R = F** **R^1 = H**	**56C** **R = Cl** **R^1 = H**	**56D** **R = Br** **R^1 = H**	**56E** **R = I** **R^1 = H**	**56F** **R=OCH_2CH_3** **R^1 = H**
C-3	141.60	156.35	140.92	140.49	140.86	136.34
C-4	115.87	138.42	126.59	-	120.76	119.18
C-5	142.89	142.00	141.51	141.71	139.04	142.34
C-5a	-	126.84	126.59	126.82	-	-
C-6	131.37	115.57	118.31	118.05	141.61	112.23
C-7	123.12	158.73	129.86	120.23	86.22	154.21
C-8	134.15	117.98	133.67	136.41	141.80	117.26
C-9	118.59	122.56	-	132.99	118.05	124.48
C-9a	126.06	-	-	-	126.57	126.11
C-1’	190.37	190.24	190.41	190.10	190.09	190.37
C-2’	-	-	119.76	114.60	-	-
C-3’	-	-	-	-	-	-
C-4’	161.89	161.65	162.17	161.66	161.66	161.55
CH_2	-	-	-	-	-	64.04
CH_3	-	-	-	-	-	14.99

4.1.5 Synthesis of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate 57

The synthesis of novel (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate was conducted using (Z)-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56D** which was reacted with KOH in the presence of EtOH and refluxed to generate salt intermediate which was subjected to ethyl-2-(2-chloroacetamido) acetate in dry DMF as shown in Scheme 13, and refluxed the resulting mixture for 24 hours and thereafter cooled, and evaporated the solvent under reduced pressure[87]. The resulting solid was dried and recrystallised from DMF to afford the corresponding novel (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** in low yield of 38.89 %.



56D 57

Reagents and conditions: (i) KOH, EtOH, (ii) ethyl-2-(2-chloroacetamido) acetate, DMF, 40 °C reflux, 24 hrs

Scheme 13: Synthesis of novel (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.

The NMR spectroscopy confirmed the synthesis of novel (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** with low yield.

The experimental melting point of the compound ranged from 320.5-321.8 °C.

This compound was fully characterised using 1 D NMR (^{1}H, ^{13}C NMR, DEPT), CHN analysis, HRMS and IR spectroscopy.

Only (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** was successfully synthesised from the

corresponding (Z)-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56D**.

The ^{1}H NMR spectra of the (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** was characterised by the disappearance of the broad singlet proton at 12.36 ppm of the 3'-H from thiazolidinedi-2,4-one moiety of the quinoline in the ^{1}H NMR spectra. In the aliphatic region, at 1.21 ppm a triplet integrated to three protons arising from the neighbouring 12'-H proton appeared corresponding to protons on carbon C-13', a singlet at 3.37 ppm integrated to two protons arising from the 7'-H protons appeared corresponding to protons on C-7', a doublet at 3.88 ppm integrated to one proton arising from neighbouring 9'-H (NH) appeared corresponding to a proton on carbon C-10', and a singlet at 5.04 ppm integrated to one proton arising from 9'-H (NH) proton appeared corresponding to a proton on the nitrogen (N-9').

The data obtained from DEPT 135 of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** in Figure 18 revealed a total of nine carbons, (five methylene (C=C) carbons, three CH_2 carbons, and one CH_3 carbon), consistent with the target molecule. The carbon observed at 60.98 ppm correspond to the carbon C-12' next to the oxygen heteroatom, it is deshielded since it is directly attached to oxygen heteroatom. The carbons observed at 44.84 ppm and 41.44 ppm are more shielded since they are directly bonded to nitrogen heteroatom (N-3 and N-9') and the carbonyl carbon (C-8' and C-11'). The observed carbon at 14.51 is more shielded since it is further away from the oxygen heteroatom. The other five carbons correspond to four aromatic carbons (CH) from 141.64 ppm to 118.05 ppm and one methylene carbon on position six prime (C-6') at 190.15 ppm.

Only sixteen carbons of the (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **56D** of the expected nineteen carbons appeared in the ^{13}C NMR spectra. One carbonyl and two quarternary carbons were missing from the ^{13}C spectra attributed to the fact that unsaturated compounds tend to have peaks overlapping with each other, and that the quaternary carbons are of low intensity in nature and needs longer period (in this case they were run for three days) still couldn't appear on the ^{13}C NMR. The other reasons are that the ^{13}C NMR

signals are slow relaxation and broad signals. The broad signals are due to one or more quadrupolar nucleus, the intermediate rate exchange processes (tautomeric equilibrium) and the C=N bond could be so in this case.[85] Other methods of analysis such as CHN and HRMS analysis were used in this regard to confirm the successful synthesis of the novel (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **56D**.

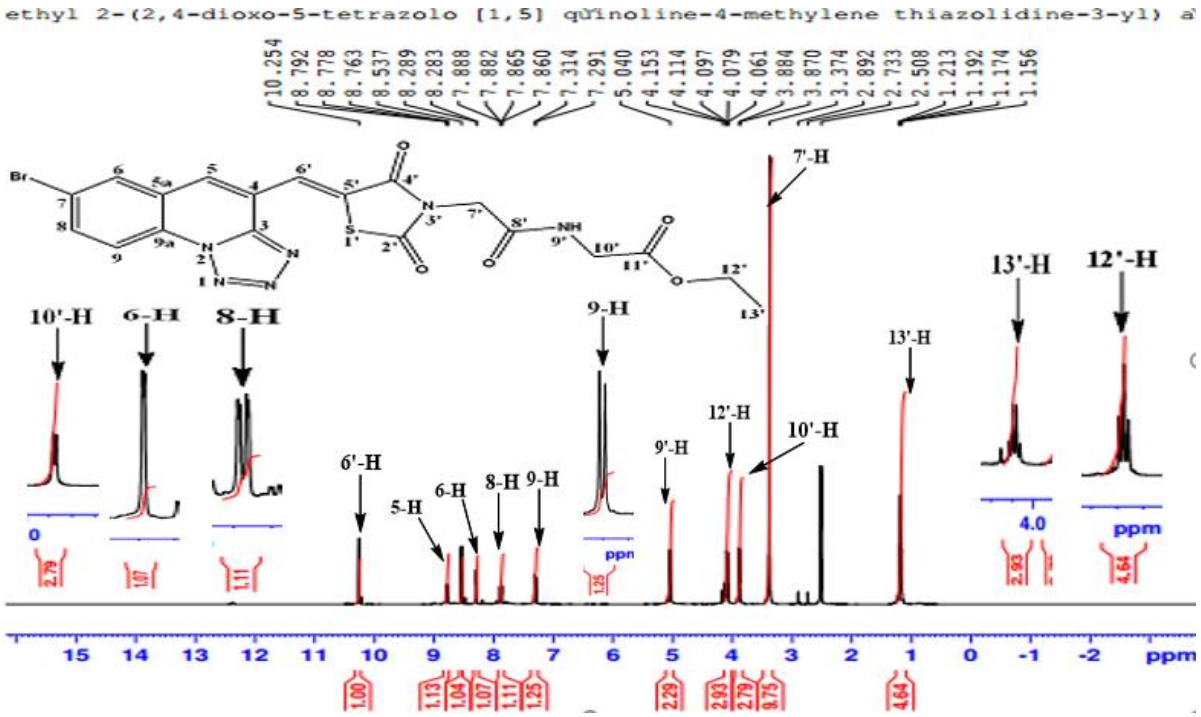


Figure 18: 400 MHz ^{1}H NMR spectrum of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.

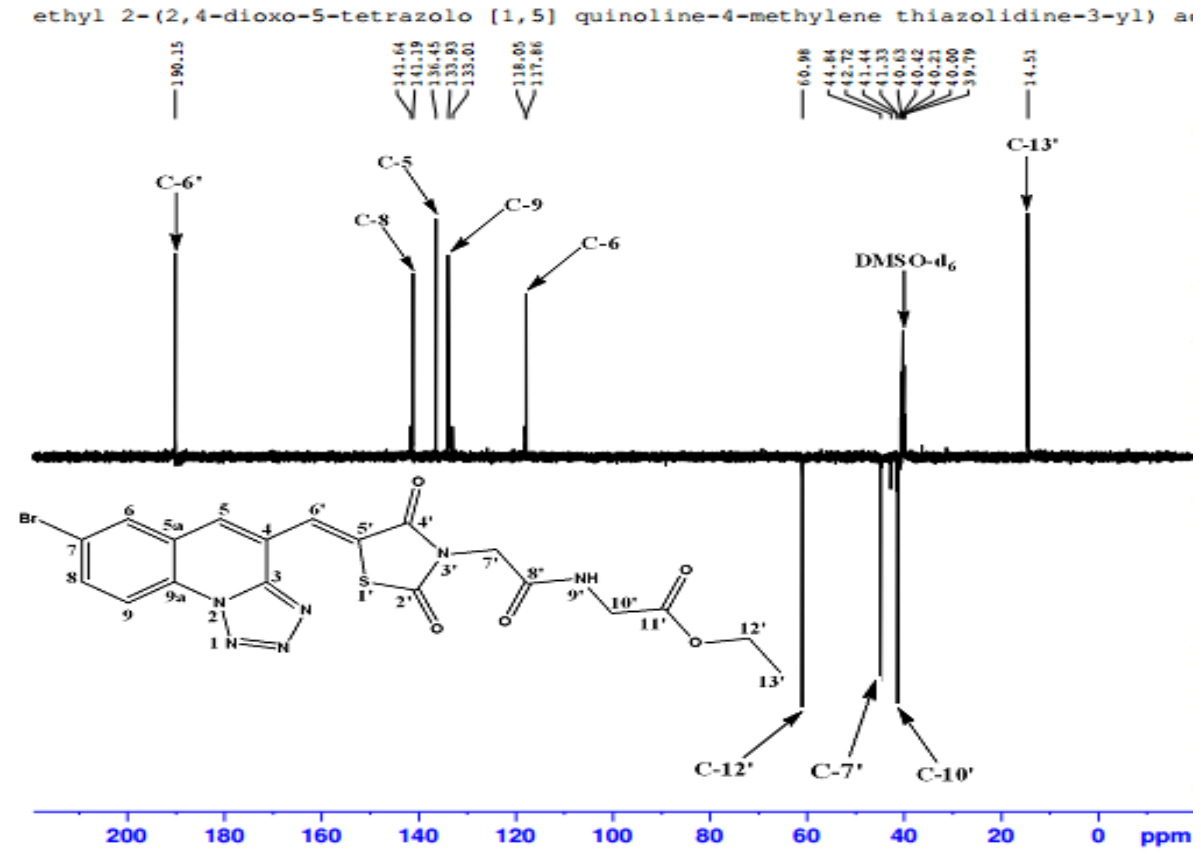


Figure 18: DEPT 135 spectrum of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.

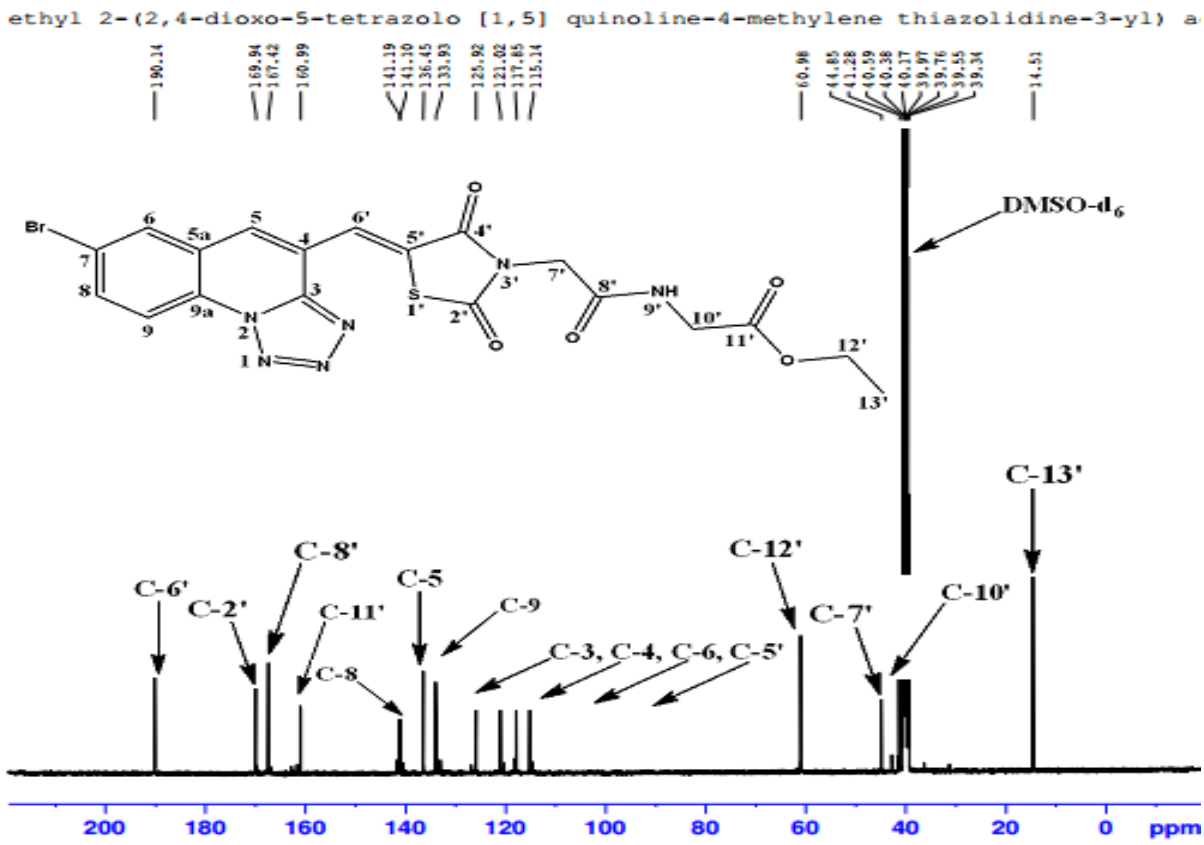


Figure 18: 100 MHz ^{13}C NMR spectrum of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.

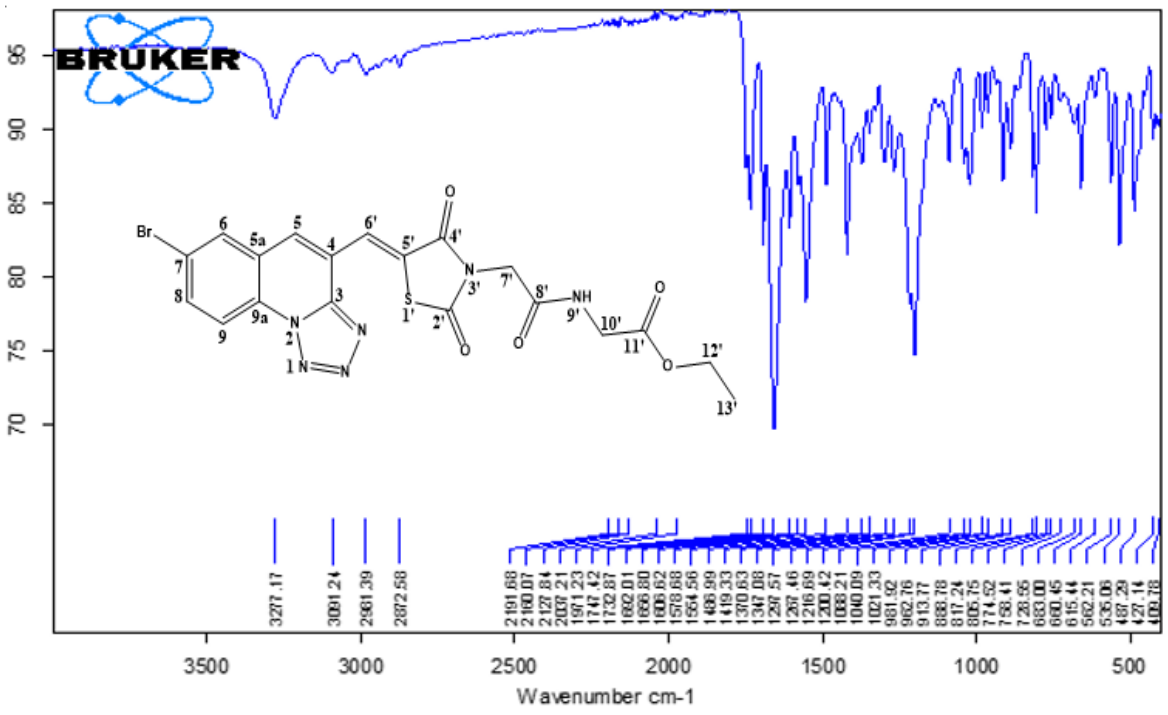
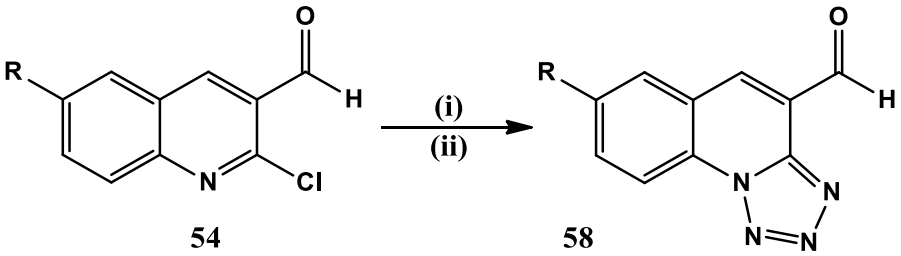


Figure 18: IR spectrum of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.

4.1.6 Synthesis of tetrazolo (1,5-a) quinoline-4-carbaldehyde analogues 58A-B

The synthesis of tetrazolo [1,5-a] quinoline-4-carbaldehyde **58A-B** was conducted using 2-chloroquinoline-3-carbaldehyde analogues **54A-B** which were subjected to glacial acetic acid in ethanol and sodium azide, refluxed the mixture to the completion for about 12 hours as shown in Scheme 14. The resulting mixture was poured in crushed ice-water and filtered the resulting solid, dried and recrystallised from DMF: Ethanol to afford the corresponding tetrazolo [1,5-a] quinoline-4-carbaldehyde analogues[86] **58A-B** in 70.36-88.66 % yield.



R = H, F

Reagents and conditions: (i) AcOH, EtOH, (ii) DMF, 78^{o}C, reflux, 2 hrs

Scheme 14: Synthesis of tetrazolo [1,5-a] quinoline-4-carbaldehyde analogues

The NMR spectroscopy confirmed the synthesis of all the tetrazolo [1,5-a] quinoline-3-carbaldehyde analogues **58A-B**.

These compounds were fully characterised by using 1 D NMR (^{1}H, ^{13}C NMR, DEPT), and IR spectrum and their physical properties.

Two tetrazolo [1,5-a] quinoline-4-carbaldehyde analogues **58A-B** were successfully synthesised from the corresponding 2-chloroquinoline-3-carbaldehyde analogues **58A-B**.

The ^{1}H NMR spectra of the tetrazolo [1,5-a] quinoline-4-carbaldehydes **58A-B** were characterised by the shift of the singlet proton (4-H) at high field region between 8.55 ppm-8.75 ppm to the low field down quite a bit in the region between 8.63 ppm-8.78 ppm and a singlet proton corresponding to the aldehydic proton (CHO) shift up quite a bit in the low field at 10.56-10.87 ppm to 10.12-00 ppm indicating the disappearance of **54A-B**. The aldehydic proton (CHO) resonates at 10.28 ppm-10.58 ppm and the (4-

H) proton at 9.03-9.07 ppm in the ^{1}H NMR spectrum established the formation of tetrazolo [1,5-a] quinoline-4-carbaldehyde.

The ^{1}H NMR spectrum of the tetrazolo [1,5-a] quinoline-4-carbaldehyde **58A** in Figure 16 revealed two singlet signals corresponding to the (CHO) proton at 10.42 ppm and the other singlet corresponding to the methine carbon (4-H) 9.03 ppm. The aromatic protons were observed between 7.69 ppm-8.78 ppm.

The ^{13}C NMR spectrum of the tetrazolo [1,5-a] quinoline-4-carbaldehyde **58A** revealed a total of 10 carbons which was consistent with the compound as shown in Figure 22.

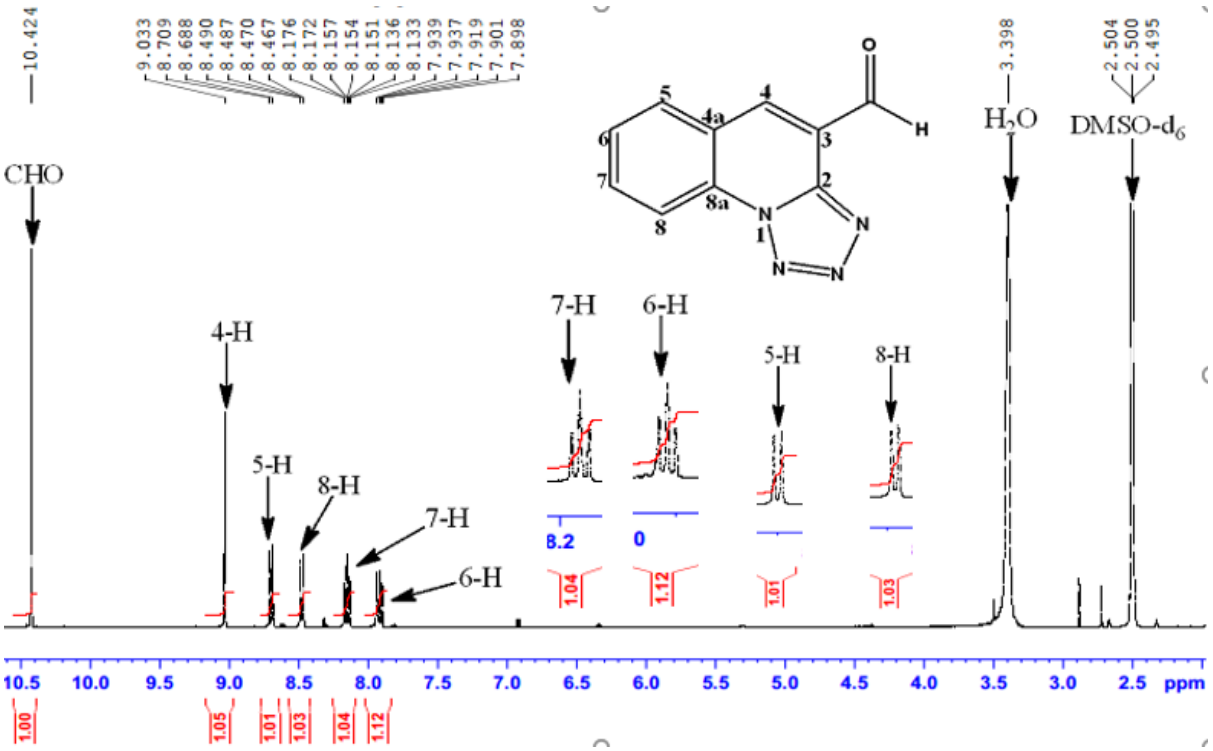


Figure 22: 400 MHz ^{1}H NMR spectrum of tetrazolo [1,5-a] quinoline-3-carnbaldehyde **58A** in DMSO-d$_6$.

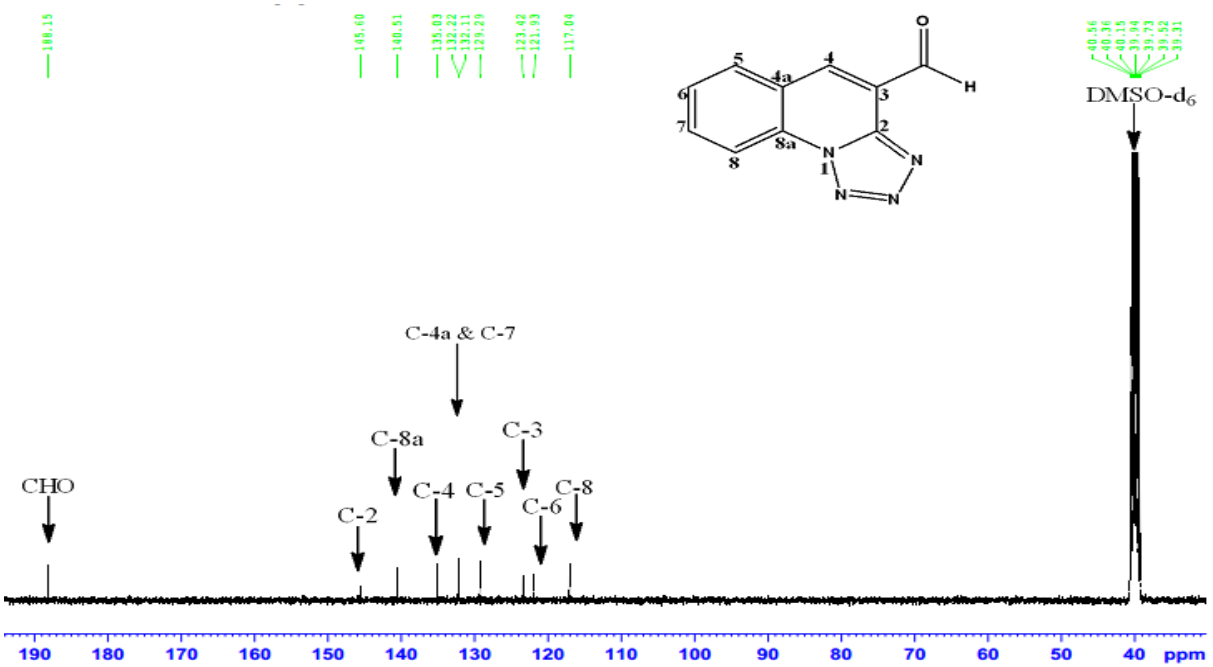
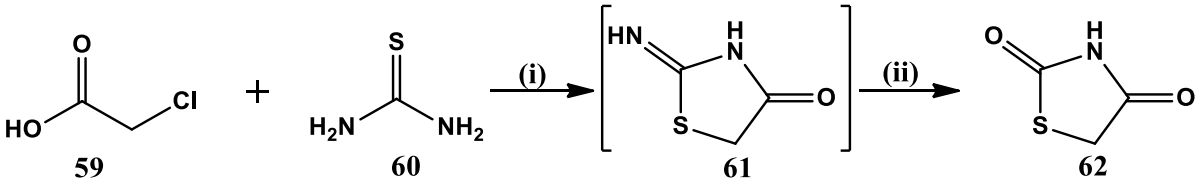


Figure 23: 100 MHz ^{1}H NMR spectrum of tetrazolo [1,5-a] quinoline-4-carbaldehyde **58A** in DMSO-d$_6$.

4.1.7 Synthesis of thiazolidinedi-2,4-one 62

The synthesis of thiazolidinedi-2,4-one **62** was conducted using the adapted method in Scheme 15. In this investigation, commercially available chloroacetic acid **59** was stirred with water and thiourea **60** for 30 minutes between 0 °C – 5 °C to yield the intermediate 2-iminothiazolidine-4-one **61** which was subjected to concentrated hydrochloric acid and refluxed for 12 hours.[88] After the completion of the reaction, the resulting mixture was poured over crushed ice-water and the resulting solid was filtered, dried and recrystallised from methanol to afford the corresponding thiazolidinedi-2,4-one **62** in 87.62 % yield.



Reagents and conditions: (i) water, 0 °C – 5 °C, (ii) Conc.HCl

Scheme 15: Synthesis of thiazolidinedi-2,4-one **62**.

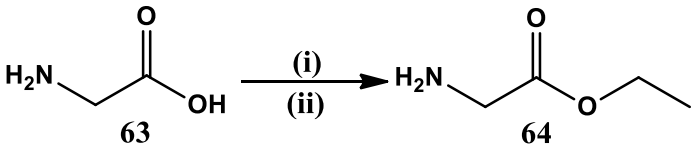
The NMR spectroscopy confirmed the synthesis of thiazolidinedi-2,4-one **62**. The afforded yield was excellent and was compared with the data reported in the literature.[88] The experimental melting point of the compound was consistent with that reported in the literature.

This compound was fully characterised by using 1 D NMR (^{1}H, ^{13}C NMR, DEPT), FT-IR spectroscopy and its physical properties.

The ^{1}H NMR spectrum thiazolidinedi-2,4-one **62** revealed two singlet signals corresponding to 3-H (NH) proton at 12.02 ppm and the other singlet corresponding to the methine proton 5-H (CH_2) 4.14 ppm. The ^{13}C NMR revealed three carbons, two carbonyl carbons at 174.32 for the C-4 and C-2 at 173.57 ppm and the methine carbon (CH_2) at 36.26 ppm).

4.1.8 Synthesis of ethyl-2-amino acetate 64

The synthesis of glycine ester **64** was conducted using the adapted method in Scheme 16. In this investigation, commercially available 2-amino acid **63** was stirred with EtOH between 0 °C – 5 °C to which $SOCl_2$ was added dropwise and refluxed for 12 hours[89]. After the completion of the reaction, the resulting solid was filtered, dried and recrystallised from EtOAc to afford the corresponding glycine ester **64** in 87.82 % yield.



Reagents and conditions: (i) water, 0 °C – 5 °C, (ii) Conc.HCl

Scheme 16: Synthesis of ethyl-2-amino acetate ester **64**.

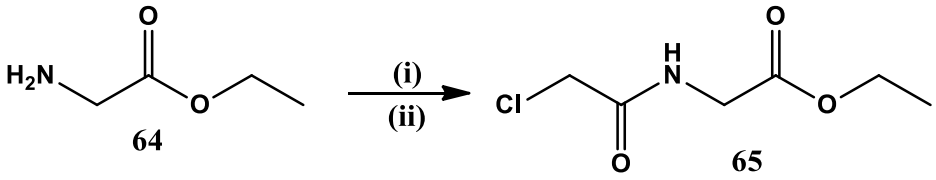
The NMR spectroscopy confirmed the purity of glycine ester **64**. The afforded yield was excellent and was compared with the data reported in the literature. The experimental melting point of all the compounds were consistent with those reported in the literature.[89]

This compound was fully characterised by using 1 D NMR (^{1}H, ^{13}C NMR, DEPT), IR spectroscopy and its physical properties.

The ^{1}H NMR spectrum glycine ester **64** revealed two singlet signals corresponding to 2-H protons at 4.07 ppm and the other singlet corresponding to the NH_2 protons 1-H at 3.13 ppm, one signal corresponding to the quartet protons 4-H at 4.23 and one signal of the triplet proton H-5 at 1.21 ppm. The carbon observed at 40.19 ppm correspond to the carbon C-2 (CH_2) next to the nitrogen heteroatom, it is deshielded since it is directly attached to nitrogen heteroatom. The carbon observed at 36.30 ppm is more shielded since it's directly bonded to oxygen heteroatom. The observed carbon at 13.18 is more shielded since it is further away from the oxygen heteroatom. The other carbon observed at 168.15 ppm corresponds to the carbonyl carbon (C=O) (C-3).

4.1.9 Synthesis of ethyl-2-(2-chloroacetamido) acetate 65

The synthesis of ethyl-2-(2-chloroacetamido) **65** was conducted using the modified method in Scheme 17. In this investigation, synthesised glycine ester **64** was stirred with KOH and water. DCM and chloroacetyl chloride were added to the mixture generated above between 0 °C – 5 °C and refluxed for 12 hours.[87] After the completion of the reaction, the resulting mixture was extracted with EtOAc, dried with $MgSO_4$ and recrystallised from EtOAc to afford the corresponding ethyl-2-(2-chloroacetamido) **65** in 50.24 % yield.



Reagents and conditions: (i) KOH, water, (ii) DCM, 2-chloroacetyl chloride, 0 °C – ambient temperature.

Scheme 17: Synthesis of ethyl-2-(2-chloroacetamido) acetate

The NMR spectroscopy confirmed the synthesis of ethyl-2-(2-chloroacetamido) acetate **65**. The afforded yield was excellent and was compared with the data reported in the literature. The experimental melting point of ethyl-2-(2-chloroacetamido) acetate **65** was consistent with that reported in the literature.[87]

This compound was fully characterised by using 1 D NMR (^1H, ^{13}C NMR, DEPT), IR spectroscopy and its physical properties.

The ^1H NMR spectrum ethyl-2-(2-chloroacetamido) acetate **65** revealed two singlet signals corresponding to 2-H protons at 4.09 ppm and the other singlet corresponding to the NH proton 4-H at 7.16 ppm, one signal corresponding to the doublet protons H-5 (CH_2) at 4.07 ppm which integrate with the NH proton and one signal of the triplet proton H-8 at 1.31 ppm one signal corresponding to the quartet protons 7-H at 4.26. The carbon observed at 61.74 ppm correspond to the carbon C-7 (CH_2) next to the oxygen heteroatom, it is deshielded since it is directly attached to oxygen heteroatom. The carbons observed at 42.34 ppm corresponding to C-2 and C-5 at 41.54 ppm are more shielded since they are directly bonded to chlorine (halogen), nitrogen heteroatom and the carbonyl carbon (C=O). The observed carbon at 14.10 is more

shielded since it is further away from the oxygen heteroatom. The other carbonyl carbons observed at 168.15 ppm and 166.28 ppm correspond to the carbonyl carbon (C=O) (C-6) and (C=O) (C-3).

The initially proposed reaction scheme was modified due to some difficulties encountered while synthesising the intermediates of this project. Gould-Jacobs method for the synthesis of tetra-substituted quinoline-3-carboxamides derivatives couldn't cyclise anilinomethylene malonate to 4-hydroxyquinoline-3-carboxylate, which resulted in the modification of this project.

4.2 Biological evaluations

4.2.1 pLDH Malaria Assay

The synthesised compounds were evaluated for their *in vitro* anti-plasmodial activity against the 3D7 strain of the malaria parasite, *Plasmodium falciparum*, using chloroquine as a reference drug. As a general rule compounds that displayed percentage parasite viability greater than 50 % at a compound of 20 µg/mL are considered inactive, those that display percentage parasite viability between 20 % and 50 % are considered marginally active and compounds with percentage viability less than 20 % are considered active. The percentage parasite viability results after exposure of the plasmodium to the synthesised compounds is summarised in Table 11 and Figure 24. Out of all thirteen compounds tested, compounds **56F, 55C** and **56B** displayed the highest activity against *Plasmodium falciparum* at 20.40, 25.80 and 12.40 %, respectively. The remaining compounds displayed more than 50 % parasite viability with **56A** and **55F** showing no activity at all.

Chloroquine, an existing drug treatment for malaria, was used as a control drug standard. The concentration used was 20 µg/mL.

A selection of 13 compounds containing the quinoline moiety were tested for anti-plasmodial activity.

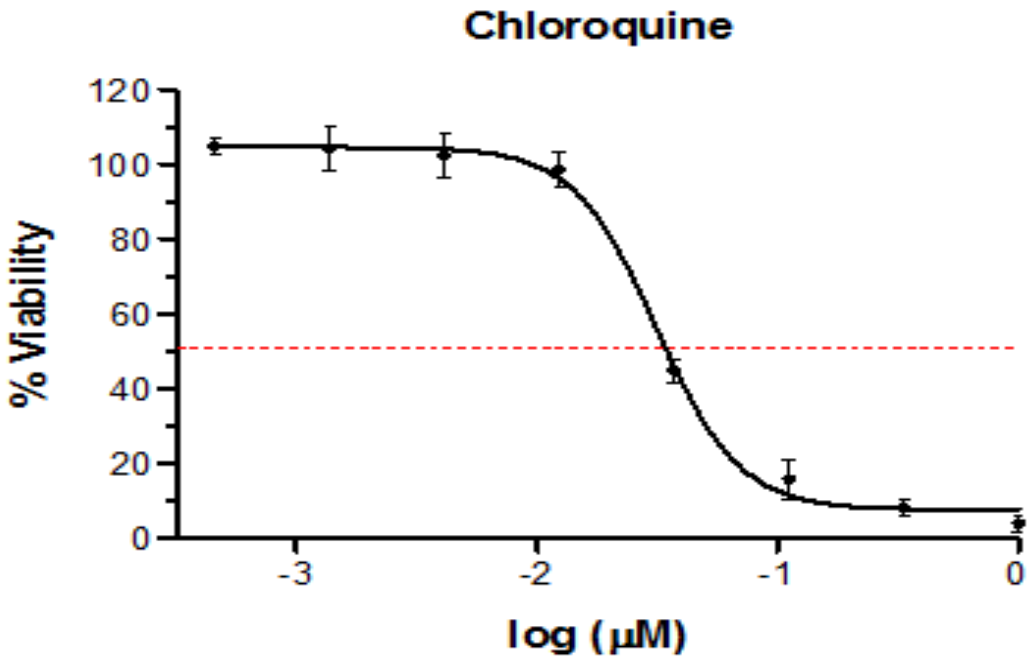


Figure 24: Chloroquine standard at 0.03 µM

Table 11: Summary of the pLDH assay of synthesised novel quinoline compounds at 20 µg/mL and of the standard chloroquine incubated for 48 h.

Compounds	% parasite viability	S.D
Chloroquine	0.00	0.00
57	89.90	4.20
56A	102.60	3.10
56F	20.40	2.40
55F	101.40	5.90
55D	98.90	6.40
56C	92.20	5.70
55E	85.20	2.10
55B	94.90	3.30
56D	97.30	5.60
55C	25.80	5.40
56E	96.30	4.80
56B	12.40	1.40
55G	93.30	5.50

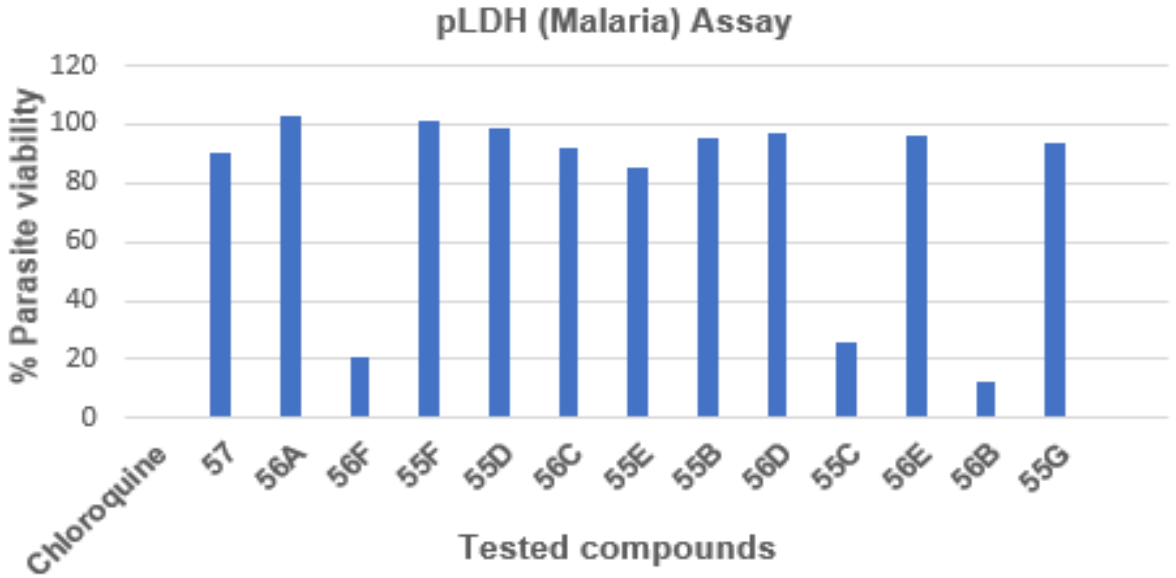
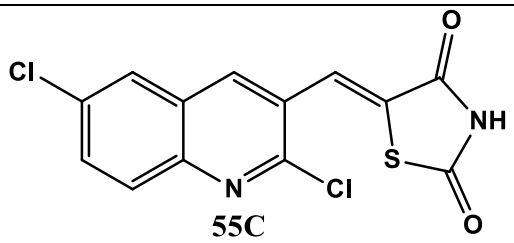
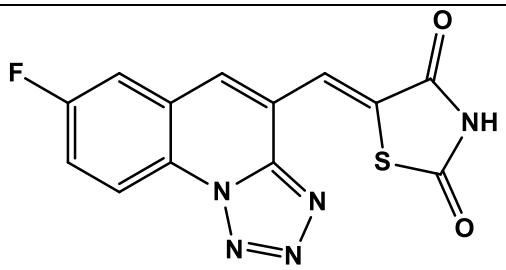
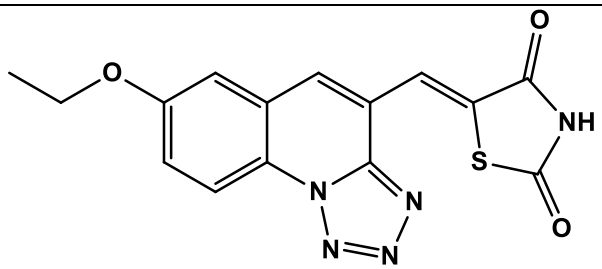


Figure 25: Activity of the synthesised novel quinoline compounds at 20 µg/mL and of the standard chloroquine incubated for 48 h

Table 12: Active synthesised novel quinoline compounds

Active compounds
 55C
 56B
 56F

Plasmodium falciparum susceptibility to compounds **56F**, **55C** and **56B** was evaluated by determining the half maximal inhibitory concentration (IC_{50}) values (Table 13). The IC_{50} value was taken as the concentration of compound (**56F**, **55C**, and **56B**) needed to reduce the absorbance ration by 50 %, therefore the lower the concentration displayed in IC_{50}, the more active the compound will be to the parasite plasmodium falciparum. Compound **56B** displayed the highest activity against *Plasmodium falciparum* with an IC_{50} value of 0.04 µg/mL.

Table 13: IC_{50} values displayed by quinoline compounds

Compound	IC_{50}
56F	0.50
55C	0.40
56B	0.04

4.2.2 Cytotoxicity Assay

The compounds evaluated for pLDH Malaria Assay were also screened for cytotoxicity activity. The results of the cytotoxicity activities of compounds are represented in figure A. Cytotoxicity was determined according to the percentage (%) of viability where compounds displaying % viability below 50 % are toxic, the smaller the value in % viability the more toxic the compound. Compounds displaying cytotoxicity above 50 % are classified non-toxic.

Emetine was used as a control drug standard. Concentration of drug used was 20 µg/mL.

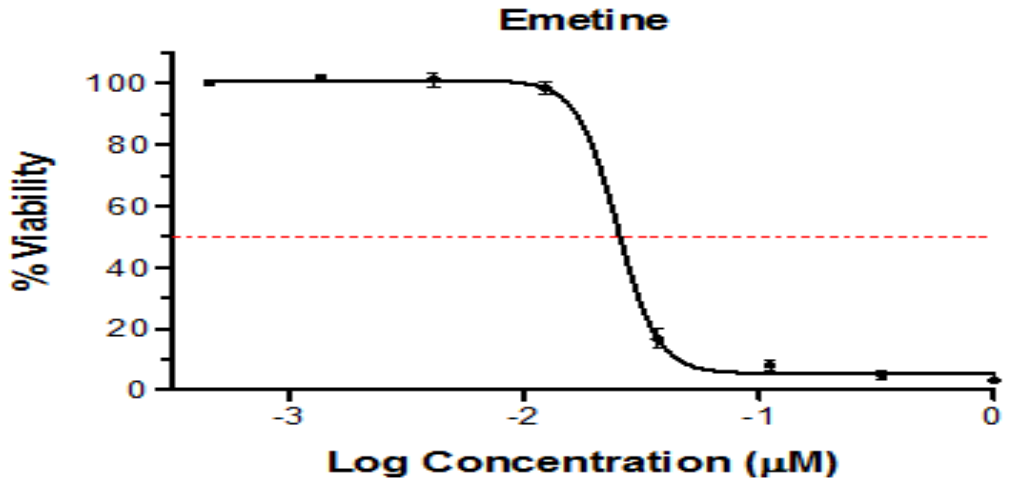


Figure 26: Emetine standard at 0.025 µM

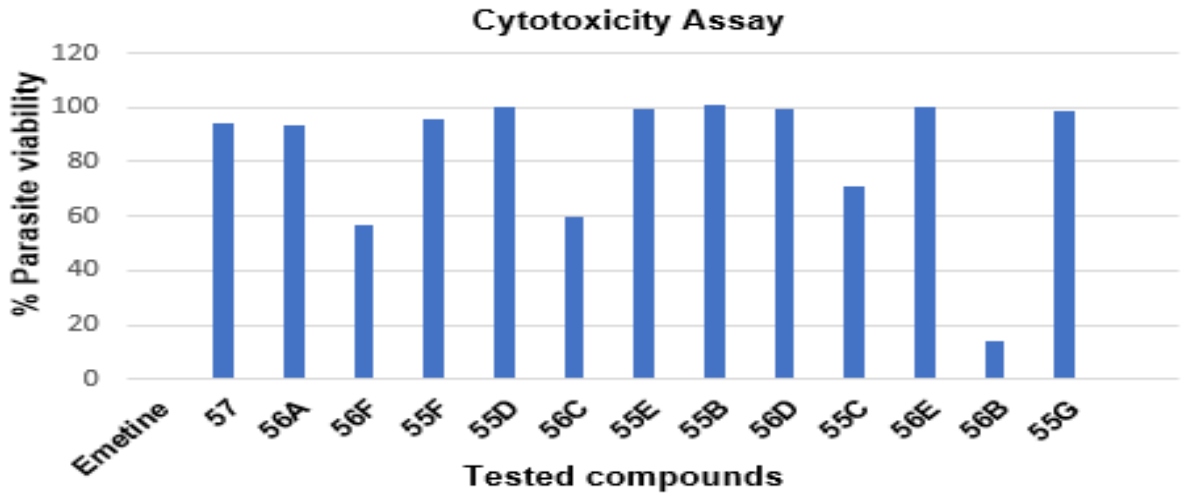
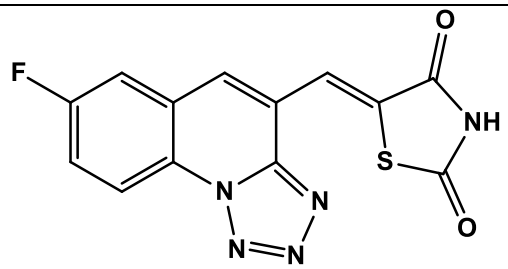
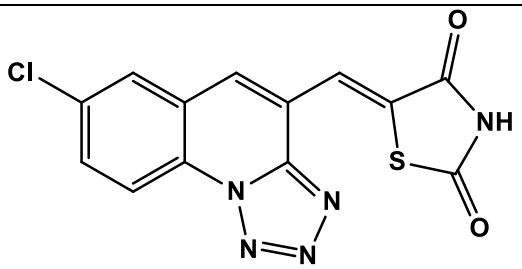
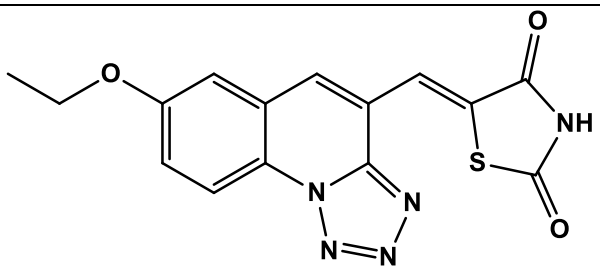


Figure 27: Cytotoxicity activity of the synthesised novel quinoline compounds against HeLa cells.

Compound **56B** displayed the highest cytotoxicity activity against human cervix adenocarcinoma cells displaying percentage viability of 14.22 %. Compounds **56F** and **56C** displayed moderate cytotoxicity activity at 56.60 and 59.81 % viability.

Table 14: Toxic and moderate toxic compounds

Toxic compound	Moderate toxic compounds
 56B	 56C
	 56F

Compound **56F** showed good plasmodial activity and its cytotoxicity was moderate displaying viability of 56.60 % making it considerable drug candidate for *Plasmodium falciparum*. Compound **56B**, although very active against *Plasmodium falciparum*, it is toxic.

CHAPTER 5

5.1 Conclusion

The investigation into 2-chloroquinoline-3-carbaldehyde derivatives was conducted because of the various biological activities the quinoline scaffold possess and their potential to produce novel lead compounds for the treatment of various diseases. A series of 2-chloroquinoline-3-carbaldehyde analogues **54** was successfully synthesised and transformed into 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues **55** in moderate yield of 41.55 %. 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55** was successfully converted into novel (Z)-5-((tetrazolo [1,5-a] quinolin-4yl) methylene) thiazolidinedi-2,4-one **56** in excellent yield of 75.23 %. (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** was successfully synthesised from (Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56** in excellent yield of 77.03 %.

Thirteen synthesised compounds were evaluated for their *in vitro* biological activities against the 3D7 strain of the malaria parasite *Plasmodium falciparum*. Out of all thirteen compounds tested, compounds **56F, 55C** and **56B** displayed the highest activity against *Plasmodium falciparum* at 20.40, 25.80 and 12.40 %, respectively. The remaining compounds displayed more than 50 % parasite viability with **56A** and **55F** showing no activity at all. *Plasmodium falciparum* susceptibility to compounds **56F**, **55C** and **56B** was evaluated by determining the half maximal inhibitory concentration (IC_{50}) values (Table 13). The IC_{50} value was taken as the concentration of compound (**56F**, **55C**, and **56B**) needed to reduce the absorbance ration by 50 %, therefore the lower the concentration displayed in IC_{50}, the more active the compound will be to the parasite plasmodium falciparum. Compound **56B** displayed the highest activity against *Plasmodium falciparum* with an IC_{50} value of 0.04 µg/mL. Compound **56B** displayed the highest cytotoxicity activity against HeLa cells displaying percentage viability of 14.22 %. Compounds **56F** and **56C** displayed moderate cytotoxicity activity at 56.60 and 59.81 % viability. Futhermore, due to moderate cytotoxicity and high antimalarial

activity observed for compound **56F**, makes it a considerable drug candidate for *Plasmodium falciparum*. Compound **56B**, although very active against *Plasmodium falciparum*, it is toxic.

5.2 Future work

In the future work, different biological assays will be explored for the synthesised compounds, such as, antiinflammatory, anticancer and antituberculosis. Other activities in the future work will entail, but not limited to the following;

- Synthesis of (Z)-ethyl-2-(2-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate analogues
- Investigation of the metal cross-coupling of various (Z)-ethyl-2-(2-5-((tetrazolo [1,5-a] quinolin-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate analogues
- Structure-activity relationship (SAR) studies of the synthesised compounds

CHAPTER 6

6. Experimental

6.1 Materials

Commercially available anilines, acetic anhydride, glacial acetic acid, DMF, phosphorus oxychloride, sodium azide, ethanol, thiazolidine-2,4-dione, ethyl 2-(2-chloro acetamido) acetate, phenylboronic acids and other reagents and solvents used were purchased from Sigma Aldrich and Merck. All reagents were analytically pure and were used without any further purification. All reactions were carried out using oven-dried glassware and the reactions were monitored by thin-layer chromatography (TLC). TLC plates were visualized under UV light (λ = 254-365 nm). Synthesized compounds were purified by distillation or recrystallization.

6.2 Instrumentation

6.2.1 Infrared Spectroscopy and Elemental analysis

IR spectra were collected using a Perkin-Elmer 1420 spectrophotometer and were reported in wavenumber (cm^{-1}). CHN analysis was done at Stellenbosch University.

6.2.2 Nuclear Magnetic Resonance (NMR) spectroscopy

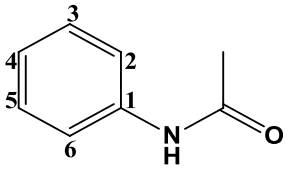
Bruker 400 MHz NMR spectrometer was used for determination of 1D NMR spectra. Chemical shifts (δ) are reported in parts per million (ppm). ^{1}H shifts are referenced to $CDCl_3$ (δ = 7.26 ppm), DMSO-d_6 (δ = 2.5 ppm) and residual water in DMSO-d_6 (δ = 3.44 ppm). ^{13}C shifts are referenced to $CDCl_3$ (δ = 77.16 ppm) and DMSO-d_6 (δ = 39.52 ppm). Splitting patterns were represented as follows: s for singlet; d for doublet; t for triplet; q for the quartet, bs for broad singlet and m for multiplet. The coupling constants (J-values) were reported in hertz (Hz).

6.2.2 Melting point

All melting points were determined on a Buchi melting points B-540 apparatus using open capillary tubes and were uncorrected.

6.3 Synthesis of acetanilide analogues[83] 53A-G

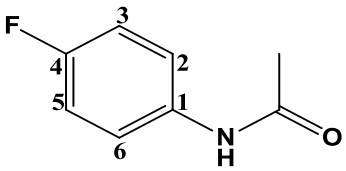
N-phenylacetamide 53A



A solution of aniline **31A** (1.01 mL, 7.69 mmol) was added to a 50 mL round bottom flask. To the above, acetic anhydride (0.77 mL, 7.69 mmol) was added dropwise with continuous stirring and added glacial acetic acid (0.77 mL, 13.46 mmol). The reaction was heated under reflux with continuous stirring for 5 hours. The mixture was cooled and poured into 50 mL of ice water. The resulting crystals were filtered and allowed to dry and recrystallised from methanol to afford N-phenylacetamide **53A** (1.26 g, 93.33 %) as a white solid, m.p 114.1-114.9 ºC, (Lit.,[83] 114 ºC); IR ν_{max}/cm^{-1} 3290.04, (N-H), 1681.14 (C=O), 1618.51 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 8.36 (bs, 1H, NH), 7.55 (d, 2H, J_{H-H} ortho = 8.00 Hz, 2-H and 6-H, ArH), 7.32 (t, 2H, J_{H-H} = 8.00 Hz, 3-H,5-H, ArH), 7.13 (t, 1H, J_{H-H} orth

o = 7.20 Hz, 4-H, ArH), 2.16 (s, 3H, CH_3); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 169.19 (C=O), 138.10 (C-1, ArC), 128.90 (C-3 and C-5, ArC), 124.30 (C-4, ArC), 120.24 (C-2 and C-6, ArC), 24.18 (CH_3).

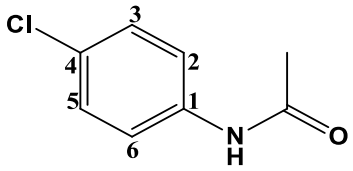
4-fluoroacetanilide 53B



The experimental procedure employed for the synthesis of **53A** was followed using 4-fluoroaniline **31B** (1.01 mL, 7.69 mmol), acetic anhydride (0.76 mL, 7.69 mmol) and glacial acetic acid (0.76 mL, 13.46 mmol). Work-up afforded a crude solid which was recrystallized from methanol to afford 4-fluoroacetanilide **53B** (0.97 g, 82.20 %) as a

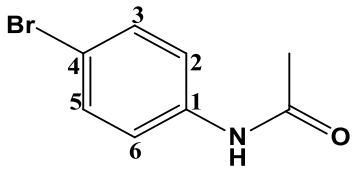
white solid; m.p 157.3-157.9 ºC (Lit.,[83] 154 ºC); IR ν_{max}/cm^{-1} 3209.89 (N-H), 1694.74 (C=O), 1659.23 (C=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 9.98 (s, 1H, NH), 7.62 (d, 2H, J_{H-H} ortho = 3.60 Hz, 3-H and 5-H, ArH), 7.15 (d, 2H, J_{H-H} ortho = 3.60 Hz, 2-H and 6-H, ArH), 2.04 (s, 3H, CH_3); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 168.62 (C=O), 159.47 (C-4, ArC), 136.18 (C-1, ArC), 121.18 (C-2 and C-6, ArC), 115.72 (C-3 and C-5, ArC), 24.27 (CH_3).

4-chloroacetanilide 53C



The experimental procedure employed for the synthesis of **53A** was followed using 4-chloroaniline **31C** (1.06 g, 6.25 mmol), acetic anhydride (0.63 mL, 6.25 mmol) and glacial acetic acid (0.63 mL, 10.94 mmol). Work-up afforded a crude solid which was recrystallized from methanol to afford 4-chloroacetanilide **53C** (0.94 g, 88.68 %) as a white solid; m.p 180.1-180.8 ºC (Lit.,[83] 178-179 ºC); IR ν_{max}/cm^{-1} 3259.70 (N-H), 1662.64 (C=O), 1606.70 (C=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 10.09 (bs, 1H, NH), 7.65 (d, 2H, J_{H-H} ortho = 10.00 Hz, 2-H and 6-H, ArH), 7.36 (d, 2H, J_{H-H} ortho = 10.00 Hz, 3-H and 5-H, ArH), 2.06 (s, 3H, CH_3); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 168.90 (C=O), 138.71 (C-1, ArC), 128.98 (C-3 and C-5, ArC), 126.97 (C-4, ArC), 120.94 (C-2 and C-6, ArC), 24.41 (CH_3).

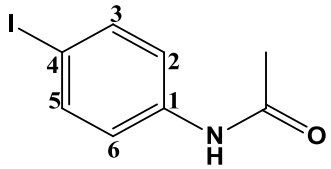
4-bromoacetalinide 53D



The experimental procedure employed for the synthesis of **53A** was followed using 4-bromoaniline **31D** (1.02 g, 4.76 mmol), acetic anhydride (0.48 mL, 4.76 mmol) and glacial acetic acid (0.48 mL, 8.33 mmol). Work-up afforded a crude solid which was recrystallized from methanol to afford 4-bromoacetanilide **54D** (0.90 g, 88.24 %) as a white solid; m.p 168.3-169.8 ºC (Lit.,[83]168 ºC); IR ν_{max}/cm^{-1} 3288.63 (N-H), 1664.57 (C=O), 1598.99 (C=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 10.07 (bs, 1H, NH), 7.57

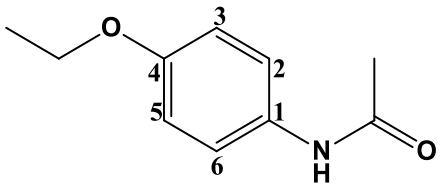
(d, 2H, J_{H-H} ortho = 9.20 Hz, 2-H and 6-H, ArH), 7.48 (d, 2H, J_{H-H} ortho = 10.00 Hz, 3-H and 5-H, ArH), 2.04 (s, 3H, CH_3); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 168.95 (C=O), 139.11 (C-1, ArC), 131.91 (C-3 and C-5, ArC), 121.33 (C-2 and C-6, ArC), 114.96 (C-4, ArC), 24.46 (CH_3).

4-iodoacetanilide 53E



The experimental procedure employed for the synthesis of **53A** was followed using 4-iodoaniline **31E** (1.01 g, 3.45 mmol), acetic anhydride (1 mL, 3.45 mmol) and glacial acetic acid (0.38 mL, 6.73 mmol). Work-up afforded a crude solid which was recrystallized from methanol to afford 4-iodoacetanilide **53E** (0.86 g, 85.15 %) as a white solid; m.p 182.3-183.5 ºC (Lit.,[83]184 ºC); IR ν_{max}/cm^{-1} 3298.47 (N-H), 1689.21 (C=O), 1627.03 (C=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 10.04 (bs, 1H, NH), 7.64 (d, 2H, J_{H-H} ortho = 11.60 Hz, 3-H and 5-H, ArH), 7.44 (d, 2H, J_{H-H} ortho = 8.80 Hz, 2-H and 6-H, ArH), 2.04 (s, 3H, CH_3); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 168.95 (C=O), 139.59 (C-1, ArC), 137.75 (C-3 and C-5, ArC), 121.63 (C-2 and C-6, ArC), 86.75 (C-4, ArC), 24.51 (CH_3).

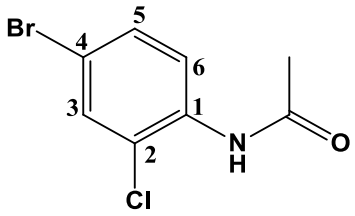
4-ethoxyacetanilide 53F



The experimental procedure employed for the synthesis of **53A** was followed using 4-ethoxyaniline **31F** (1.05 mL, 6.25 mmol), acetic anhydride (0.63 mL, 6.25 mmol) and glacial acetic acid (0.63 mL, 10.94 mmol). Work-up afforded a crude solid which was recrystallized from methanol to afford 4-ethoxyacetanilide **53F** (0.95 g, 90.48 %) as a white solid; m.p 115.2-116.8 ºC (Lit.,[83] 114 ºC); IR ν_{max}/cm^{-1} 3275.13 (N-H), 1699.29 (C=O), 1653.00 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 9.78 (bs, 1H, NH), 7.48 (d, 2H, J_{H-H} ortho = 12.16 Hz, 2-H and 6-H, ArH), 6.86 (d, 2H, J_{H-H} ortho = 12.20 Hz, 3-H and 5-H, ArH), 3.98 (q, 2H, J_{H-H} = 6.96 Hz, CH_2), 2.00 (s, 3H, CH_3), 1.31 (t, 3H, J_{H-H} = 6.92

Hz, CH_3); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 172.94 (C=O), 159.49 (C-4, ArC), 137.62 (C-1, ArC), 125.74 (C-2 and C-6, ArC), 119.50 (C-3 and C-5, ArC), 68.25 (CH_2), 28.98 (CH_3), 19.89 (CH_3).

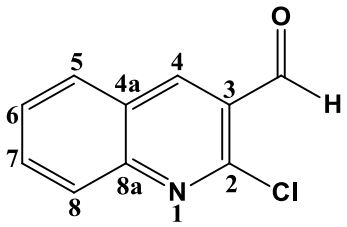
4-bromo-2-chloromoacetanilide 53G



The experimental procedure employed for the synthesis of **53A** was followed using 4-bromo-2-chloroaniline **31G** (1.04 g, 10.90 mmol), acetic anhydride (0.42 mL, 4.17 mmol) and glacial acetic acid (0.42 mL, 7.29 mmol). Work-up afforded a crude solid which was recrystallized from methanol to afford 4-bromo-2-chloroacetanilide **53G** (0.93 g, 89.42 %) as a white solid; m.p 154.4-155.1 ºC (Lit.,[83] 151 ºC); IR ν_{max}/cm^{-1} 3277.06 (N-H), 1697.36 (C=O), 1662.64 (C=C); ^1H NMR: ($CDCl_3$, 400 MHz): δ_H 8.18 (d, 1H, J_{H-H} ortho = 8.80 Hz, 2-H, ArH), 7.62 (bs, 1H, NH), 7.45 (d, 1H, J_{H-H} meta = 2.40 Hz, 5-H, ArH), 7.31 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 8.80 Hz, 3-H, ArH), 2.16 (s, 3H, CH_3); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 168.33 (C=O), 133.85 (C-1, ArC), 131.41 (C-5, ArC), 130.74 (C-3, ArC), 123.43 (C-6, ArC), 122.86 (C-2, ArC), 116.26 (C-4, ArC), 24.80 (CH_3).

6.4 Synthesis of 2-chloroquinoline-3-carbaldehyde analogues[83] 54A-G

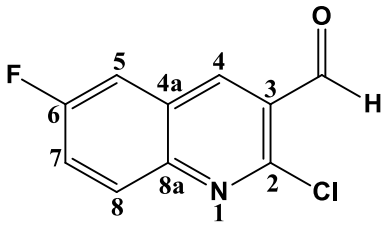
2-chloroquinoline-3-carbaldehyde 54A



Dry DMF (9.60 mL, 125 mmol) was transferred to a two-neck round bottom flask under inert nitrogen gas and cooled to 0 ºC in an ice bath. $POCl_3$ (32.20 mL, 350 mmol) was added dropwise with continuous stirring while maintaining the temperature below 10 ºC. N-phenylacetamide **53A** (6.76 g, 50 mmol) was added to the above mixture and heated at 85ºC under reflux for 24 hours. After completion, the mixture was cooled to

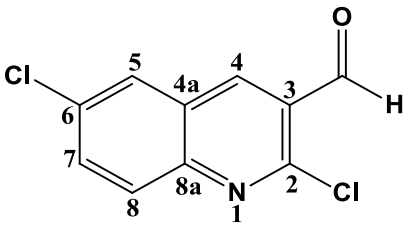
room temperature and poured into 200 mL of ice water and stirred for 1 hour below 10ºC. The resulting pale-yellow precipitate was filtered off and washed with 100 mL water and allowed to dry. The crude product was recrystallized from 30 mL ethyl acetate to afford 2-chloroquinoline-3-carbaldehyde **54A** (3.76 g, 38.94 %) as a pale-yellow solid, m.p 144.3-145.8 ºC, (Lit.,[83] 143 ºC); IR ν_{max}/cm^{-1} 1681.39 (CHO), 1662.64 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.58 (s, 1H, CHO), 8.78 (s, 1H, 4-H, ArH), 8.10 (d, 1H, J_{H-H} ortho = 8.40 Hz, 5-H, ArH), 8.02 (d, 1H, J_{H-H} ortho = 8.00 Hz, 8-H, ArH), 7.93 (t, 1H, J_{H-H} ortho = 7.20 Hz, 7-H, ArH), 7.69 (t, 1H, J_{H-H} ortho = 7.60 Hz, 6-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 189.20 (C=O), 150.12 (C-2, ArC), 149.60 (C-8a, ArC), 140.32 (C-4, ArC), 133.64 (C-7, ArC), 129.74 (C-5, ArC), 128.62 (C-6, ArC), 128.16 (C-8, ArC), 129.74 (C-4a, ArC), 126.37 (C-3, ArC). Anal. Calc. for $C_{10}H_6ClNO$: C 62.68; H 3.16; N 7.31 Found: C 66.50; H 2.01; N 7.35

6-fluoroquinoline-3-carbaldehyde 54B



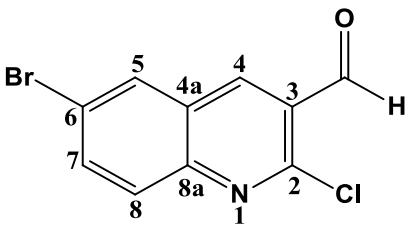
The experimental procedure employed for the synthesis of **54A** was followed using 4-fluoroacetanilide **53B** (10.48 g, 50 mmol), DMF (9.60, 125 mmol) and $POCl_3$ (32.20 mL, 350 mmol). Work-up afforded a crude solid which was recrystallized from ethyl acetate to afford 6-fluoro-2-chloroquinoline-3-carbaldehyde **54B** (2.58 g, 24.62 %) as a pale-yellow solid, m.p 337.6-338.4 ºC, (Lit.,[83] 71 ºC); IR ν_{max}/cm^{-1} 1703.14 (CHO), 1651.07 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.49 (s, 1H, CHO), 8.64 (s, 1H, 4-H, ArH), 8.04 (d, 1H, J_{H-H} ortho = 5.20 Hz, 8-H, ArH), 7.61 (d, 1H, J_{H-H} meta = 2.80, 5-H, ArH), 7.59 (dd, 1H, J_{H-H} meta, ortho = 2.80 and 4.00Hz Hz, 7-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 188.93 (C=O), 162.29 (C-6, ArC), 159.78 (C-2, ArC), 149.45 (C-8a, ArC), 146.65 (C-4a, ArC), 139.53 (C-4, ArC), 131.21 (C-8, ArC), 127.37 (C-3, ArC), 123.96 (C-7, ArC), 112.76 (C-5, ArC).

2,6-dichloroquinoline-carbaldehyde 54C



The experimental procedure employed for the synthesis of **54A** was followed using 4-chloroacetanilide **53C** (11.30 g, 50 mmol), DMF (9.60, 125 mmol) and $POCl_3$ (32.20 mL, 350 mmol). Work-up afforded a crude solid which was recrystallized from ethyl acetate to afford 6-chloro-2-chloroquinoline-3-carbaldehyde **54C** (1.22 g, 10.80 %) as a pale-yellow solid, m.p 138.8-139.6 ºC, (Lit.,[83] 138 ºC); IR ν_{max}/cm^{-1} 1734.01 (CHO), 1653.00 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.48 (s, 1H, CHO), 8.59 (s, 1H, 4-H, ArH), 7.95 (d, 1H, J_{H-H} ortho = 9.20 Hz, 8-H, ArH), 7.89 (d, 1H, J_{H-H} meta = 2.00 Hz, 5-H, ArH), 7.75 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 9.20 Hz, 7-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 188.80 (C=O), 150.32 (C-2, ArC), 147.89 (C-8a, ArC), 139.19 (C-4, ArC), 134.42 (C-7, ArC), 134.09 (C-6, ArC), 130.13 (C-8, ArC), 128.10 (C-5, ArC), 127.15 (C-4a, ArC), 127.02 (C-3, ArC). Anal. Calc. for $C_{10}H_5Cl_2NO$: C 53.13; H 2.23; N 6.20 Found: C 53.16; H 1.51; N 6.00

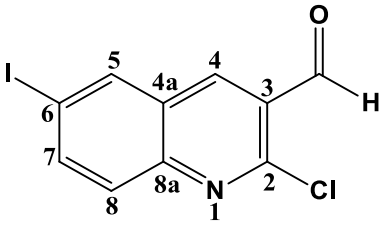
6-bromo-2-chloroquinoline-3-carbaldehyde 54D



The experimental procedure employed for the synthesis of **54A** was followed using 4-bromoacetanilide **53C** (13.54 g, 50 mmol), DMF (9.60, 125 mmol) and $POCl_3$ (32.20 mL, 350 mmol). Work-up afforded a crude solid which was recrystallized from ethyl acetate to afford 6-bromo-2-chloroquinoline-3-carbaldehyde **54D** (2.33 g, 17.21 %) as a pale-yellow solid, m.p 187.1-187.7 ºC, (Lit, [83] 189 ºC); IR ν_{max}/cm^{-1} 1716.65 (CHO), 1647.21 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.48 (s, 1H, CHO), 8.59 (s, 1H, 4-H, ArH), 8.07 (s, 1H, 5-H, ArH), 7.87 (d, 2H, J_{H-H} ortho, meta = 1.20 Hz, 7-H and 8-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 188.76 (C=O), 150.44 (C-2, ArC), 148.11 (C-

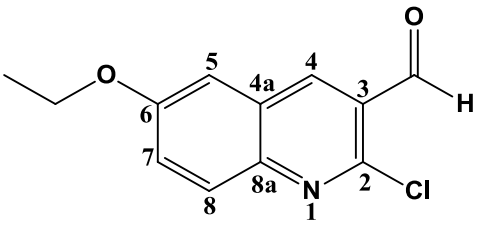
8a, ArC), 139.09 (C-4, ArC), 136.96 (C-7, ArC), 131.48 (C-5, ArC), 130.18 (C-8, ArC), 127.63 (C-3, ArC), 127.01 (C-4a, ArC), 122.14 (C-6, ArC). Anal. Calc. for $C_{10}H_5BrClNO$: C 44.40; H 1.86; N 5.18 Found: C 47.57; H 1.46; N 5.13

6-iodo-2-chloroquinoline-3-carbaldehyde 54E



The experimental procedure employed for the synthesis of **54A** was followed using 4-iodoacetanilide **53E** (15.88 g, 50 mmol), DMF (9.60, 125 mmol) and $POCl_3$ (32.20 mL, 350 mmol). Work-up afforded a crude solid which was recrystallized from ethyl acetate to afford 6-iodo-2-chloroquinoline-3-carbaldehyde **54E** (2.97 g, 18.70 %) as a pale-yellow solid, m.p 170.8-171.8 ºC, (Lit, [83] 169 ºC); IR ν_{max}/cm^{-1} 1727.41 (CHO), 1635.64 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.47 (s, 1H, CHO), 8.55 (s, 1H, 4-H, ArH), 8.29 (d, 1H, J_{H-H} meta = 1.60 Hz, 5-H, ArH), 8.04 (dd, 1H, J_{H-H} ortho, meta = 8.80 and 2.00 Hz, 7-H, ArH), 7.23 (d, 1H, J_{H-H} ortho = 8.80 Hz, 8-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 188.74 (C=O), 150.56 (C-2, ArC), 148.45 (C-8a, ArC), 142.19 (C-4, ArC), 138.85 (C-7, ArC), 138.15 (C-5, ArC), 130.03 (C-8, ArC), 128.10 (C-4a, ArC), 126.86 (C-3, ArC), 93.67 (C-6, ArC). Anal. Calc. for $C_{10}H_5ClINO$: C 37.83; H 1.59; N 4.41 Found: C 40.57; H 1.09; N 4.26

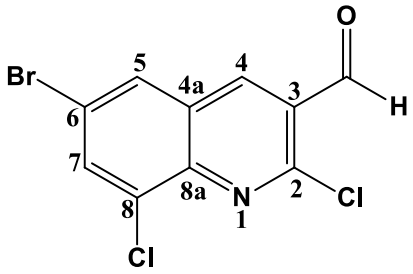
6-ethoxy-2-chloroquinoline-3-carbaldehyde 54F



The experimental procedure employed for the synthesis of **54A** was followed using 4-ethoxyacetanilide **53F** (11.78 g, 50 mmol), DMF (9.60, 125 mmol) and $POCl_3$ (32.20 mL, 350 mmol). Work-up afforded a crude solid which was recrystallized from ethyl acetate to afford 6-ethoxy-2-chloroquinoline-3-carbaldehyde **54F** (2.34 g, 19.86 %) as a pale-yellow solid, m.p 179.2-180.7 ºC, (Lit, [83] 178 ºC); IR ν_{max}/cm^{-1} 1738.78 (CHO),

1657.71 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.53 (s, 1H, CHO), 8.60 (s, 1H, 4-H, ArH), 7.95 (d, 1H, J_{H-H} ortho = 9.20 Hz, 8-H, ArH), 7.52 (dd, 1H, J_{H-H} meta, ortho = 2.80 Hz and 9.20 Hz, 7-H, ArH), 7.16 (d, 1H, J_{H-H} meta = 2.40 Hz, 5-H, ArH), 4.19 (q, 2H, J = 6.80 Hz, CH_2), 1.53 (t, 3H, J = 6.80 Hz, CH_3); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 189.42 (C=O), 158.13 (C-6, ArC), 147.51 (C-2, ArC), 145.68 (C-8a, ArC), 138.60 (C-4, ArC), 129.82 (C-8, ArC), 127.77 (C-4a, ArC), 126.33 (C-3, ArC), 126.82 (C-7, ArC), 107.04 (C-5, ArC), 64.15 (CH_2), 14.60 (CH_3). Anal. Calc. for $C_{12}H_{10}ClNO_2$: C 61.16; H 4.28; N 5.94 Found: C 65.17; H 3.80; N 5.89

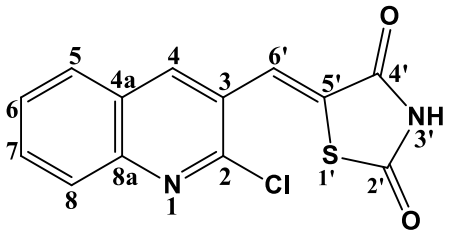
6-bromo-2,8-dichloroquinoline-3-carbaldehyde 54G



The experimental procedure employed for the synthesis of **54A** was followed using 6-chloro-4-bromoacetanilide **53G** (15.24 g, 50 mmol), DMF (9.60, 125 mmol) and $POCl_3$ (32.20 mL, 350 mmol). Work-up afforded a crude solid which was recrystallized from ethyl acetate to afford 6-bromo-2,8-dichloroquinoline-3-carbaldehyde **54G** (5.21 g, 34.19 %) as a pale-yellow solid, m.p 117.8-118.3 ºC; IR ν_{max}/cm^{-1} 1729.26 (CHO), 1668.03 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 8.53 (s, 1H, CHO), 7.92 (d, 1H, J_{H-H} meta = 2.00 Hz, 4-H, ArH), 7.70 (d, 1H, J_{H-H} meta = 1.60 Hz, 5-H, ArH), 7.58 (d, 1H, J_{H-H} meta = 8.40 Hz, 7-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 172.52 (C=O), 156.57 (C-2, ArC), 134.82 (C-8, ArC), 132.81 (C-4, ArC), 131.85 (C-7, ArC), 129.87 (C-4a, ArC), 128.73 (C-5, ArC), 121.11 (C-6, ArC).

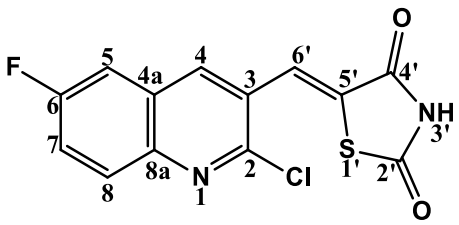
6.5 Synthesis of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one analogues[84] 55A-G

2-chloroquinoline-3-methylene thiazolidinedi-2,4-one 55A



2-chloroquinoline-3-carbaldehyde **54A** (1.41 g, 7.40 mmol) was added in a 100 mL round bottom flask to which thiazolidinedi-2,4-one (g, 7.40 mmol) was added. Anhydrous sodium acetate (0.08 g, 7.40 mmol) and 10 mL of glacial acetic acid were added to the above mixture and refluxed for 12 hours. After the completion of the reaction, monitored by TLC, the mixture was cooled to room temperature and filtered the yellow precipitate formed and further washed with 50 mL water. The crude product was recrystallized from 10 mL DMF to afford 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** (0.95 g, 44.19 %) as yellow solid, m.p 307.9-308.0 ºC (Lit,[84] 221 ºC); IR ν_{max}/cm^{-1} 3093.05 (N-H), 2626.56 (ArC=N), 1619.46 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.26 (bs, 1H, NH), 10.25 (s, 1H, ArCH=C), 8.52 (s, 1H, 4-H, ArH), 7.94 (d, 1H, J_{H-H} ortho = 7.20 Hz, 5-H, ArH), 7.69 (t, 1H, J_{H-H} ortho = 8.40 Hz, 7-H, ArH), 7.38 (d, 1H, J_{H-H} ortho = 8.00 Hz, 8-H, ArH), 7.28 (t, 1H, J_{H-H} ortho = 7.20 Hz, 6-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.25 (C-2'), 161.90 (C-4'), 134.82 (C-8, ArC), 132.81 (C-4, ArC), 131.85 (C-7, ArC), 129.87 (C-4a, ArC), 128.73 (C-5, ArC), 121.11 (C-6, ArC). Anal. Calc. for $C_{13}H_7ClN_2O_2S$: C 53.71; H 2.43; N 9.64. Found: C 79.75; H 3.26; N 8.63

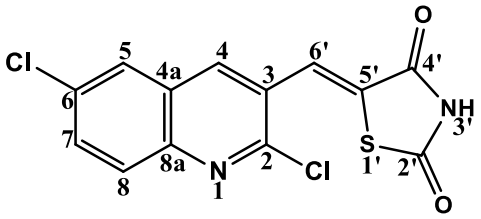
6-fluoroquinoline-3-methylene thiazolidinedi-2,4-one 55B



The experimental procedure employed for the synthesis of **55A** was followed using 6-fluoroquinoline-3-carbaldehyde **55B** (1.58 g, 7.50 mmol), thiazolidinedi-2,4-one (0.88

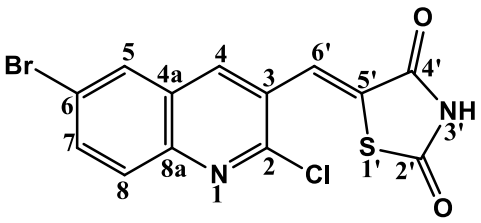
g, 7.50 mmol) was added. Anhydrous sodium acetate (0.08 g, 7.50 mmol) and 10 mL of glacial acetic acid. Work-up afforded a solid crude product which was recrystallized from DMF to afford 6-fluoroquinoline-3-methylene thiazolidinedi-2,4-one **55B** (0.96 g, 41.38 %) as a yellow solid, m.p 336.7-337.5 ºC; IR ν_{max}/cm^{-1} 3509.48 (N-H), 2237.04 (ArC=N), 1619.89 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.33 (bs, 1H, NH), 10.24 (s, 1H, ArCH=C), 8.49 (s, 1H, 4-H, ArH), 7.83 (dd, 1H, J_{H-H} meta, ortho = 2.80 Hz and 8.80 Hz, 7-H, ArH), 7.61 (d, 1H, J_{H-H} meta = 2.80 Hz, 5-H, ArH), 7.40 (d, 1H, J_{H-H} ortho = 4.80 Hz, 8-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.22 (C-1'), 161.61 (C-4'), 158.74 (C-6, ArC), 156.36 (C-2, ArC), 141.99 (C-4), 138.40 (C-8a, ArC), 126.87 (C-4a, ArC), 122.57 (C-8, ArC), 119.21 (C-3, ArC), 117.94 (C-7, ArC), 115.60 (C-5, ArC).

2,6-dichloroquinoline-3-methylene thiazolidinedi-2,4-one 55C



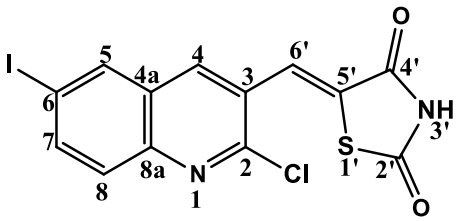
The experimental procedure employed for the synthesis of **55A** was followed using 2,6-dichloroquinoline-3-carbaldehyde **54C** (0.60 g, 3.10 mmol), thiazolidinedi-2,4-one (0.36 g, 3.10 mmol) was added. Anhydrous sodium acetate (0.08 g, 3.10 mmol) and 10 mL of glacial acetic acid. Work-up afforded a solid crude product which was recrystallized from DMF to afford 2,6-dichloroquinoline-3-methylene thiazolidinedi-2,4-one **55C** (0.43 g, 42.57 %) as a yellow solid, m.p 349.5-350.4 ºC; IR ν_{max}/cm^{-1} 3325.28 (N-H), 2308.79 (ArC=N), 1614.42 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.37 (bs, 1H, NH), 10.22 (s, 1H, ArCH=C), 8.48 (s, 1H, 4-H, ArH), 8.06 (d, 1H, J_{H-H} meta = 2.40, 5-H, ArH), 7.70 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 8.80 Hz, 7-H, ArH), 7.37 (d, 1H, J_{H-H} ortho = 8.80 Hz, 8-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.12 (C-1'), 161.66 (C-4'), 141.73 (C-4), 140.22 (C-2, ArC), 133.82 (C-7, ArC), 129.96 (C-8, ArC), 126.90 (C-8a, ArC), 126.84 (C-3, ArC), 119.69 (C-4a, ArC), 117.83 (C-5, ArC). Anal. Calc. for $C_{13}H_6Cl_2N_2O_2S$: C 48.02; H 1.86; N 8.62. Found: C 58.69; H 2.14; N 6.22

6-bromo-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one 55D



The experimental procedure employed for the synthesis of **55A** was followed using 6-bromo-2-chloroquinoline-3-carbaldehyde **54D** (1.89 g, 7.00 mmol), thiazolidinedi-2,4-one (0.82 g, 7.00 mmol), anhydrous sodium acetate (0.08 mg, 7.00 mmol) and 10 mL of glacial acetic acid. Work-up afforded a solid crude product which was recrystallized from DMF to afford 6-bromo-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55D** (1.38 g, 53.49 %) as a yellow solid, m.p 345.2-345.6 ºC; IR ν_{max}/cm^{-1} 3538.03 (N-H), 2229.01 (ArC=N), 1618.98 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.35 (bs, 1H, NH), 10.22 (s, 1H, ArCH=C), 8.46 (s, 1H, 4-H, ArH), 8.17 (d, 1H, J_{H-H} meta = 2.40, 5-H, ArH), 7.79 (dd, 1H, J_{H-H} meta, ortho = 8.80 and 2.40 Hz and 8.80 Hz, 7-H, ArH), 7.30 (d, 1H, J_{H-H} ortho = 8.80 Hz, 8-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.09 (C-1'), 161.64 (C-4'), 141.67 (C-4), 140.51 (C-8a, ArC), 136.40 (C-7, ArC), 133.00 (C-8, ArC), 126.83 (C-3, ArC), 120.23 (C-4a, ArC), 118.04 (C-5, ArC), 114.58 (C-6, ArC). Anal. Calc. for $C_{13}H_6BrClN_2O_2S$: C 42.24; H 1.64; N 7.58. Found: C 50.97; H 2.00; N 5.50

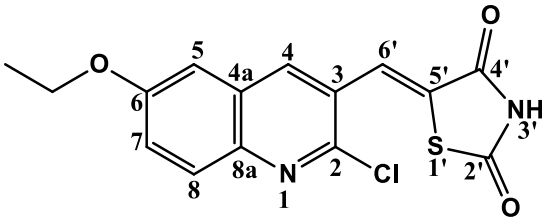
6-iodo-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one 55E



The experimental procedure employed for the synthesis of **54A** was followed using 6-iodo-2-chloroquinoline-3-carbaldehyde **55E** (1.51 g, 7.90 mmol), thiazolidinedi-2,4-one (0.92 g, 7.90 mmol), anhydrous sodium acetate (0.08 g, 7.90 mmol) and 10 mL of glacial acetic acid. Work-up afforded a solid crude product which was recrystallized from DMF to afford 6-iodo-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55E** (0.90 g, 27.36 %) as a yellow solid, m.p 317.4-318.4 ºC; IR ν_{max}/cm^{-1} 3523.95 (N-H), 2347.37 (ArC=N), 1606.70 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.31 (bs, 1H,

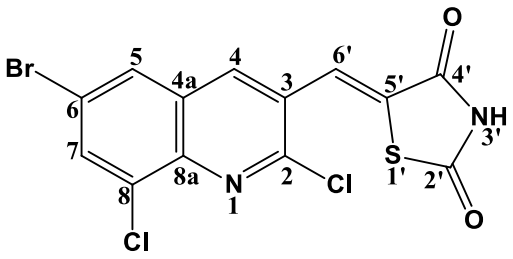
NH), 10.21 (s, 1H, ArCH=C), 8.42 (s, 1H, 4-H, ArH), 8.31 (d, 1H, J_{H-H} meta = 1.20, 5-H, ArH), 7.91 (dd, 1H, J_{H-H} meta, ortho = 1.60 Hz and 8.40 Hz, 7-H, ArH), 7.16 (d, 1H, J_{H-H} ortho = 8.80 Hz, 8-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.61 (C-1'), 161.62 (C-4'), 141.79 (C-4, ArC), 141.53 (C-2, ArC), 140.87 (C-8a, ArC), 139.06 (C-7, ArC), 126.60 (C-5), 120.75 (C-4a, ArC), 118.01 (C-8, ArC), 86.23 (C-6, ArC). Anal. Calc. for $C_{13}H_{16}ClIN_2O_2S$: C 37.48; H 1.45; N 6.72. Found: C 44.95; H 1.65; N 4.72

6-ethoxy-2-chloroquinoline-3-methylene thiazolidi-2,4-one 55F



The experimental procedure employed for the synthesis of **55A** was followed using 6-ethoxy-2-chloroquinoline-3-carbaldehyde **54F** (1.24 g, 6.50 mmol), thiazolidinedi-2,4-one (0.76 g, 6.50 mmol) , anhydrous sodium acetate (0.08 g, 6.50 mmol) and 10 mL of glacial acetic acid. Work-up afforded a solid crude product which was recrystallized from DMF to afford 6-ethoxy-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55F** (0.50 g, 40.32 %) as a yellow solid, m.p 287.9-288.4 ºC; IR ν_{max}/cm^{-1} 3429.06 (N-H), 2231.08 (ArC=N), 1624.06 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.16 (bs, 1H, NH), 10.23 (s, 1H, ArCH=C), 8.42 (s, 1H, 4-H, ArH), 7.44 (d, 1H, J_{H-H} meta = 1.60, 5-H, ArH), 7.30 (d, 1H, J_{H-H} meta, ortho = 2.40, 5-H and 7-H, ArH), 4.01 (q, 2H, J_{H-H} = 6.80 Hz, CH_2), 1.36 (t, 3H, J_{H-H} = 7.20 Hz, CH_3); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.36 (C-1'), 161.53 (C-4'), 154.19 (C-6, ArC), 142.28 (C-4), 136.34 (C-2, ArC), 126.11 (C-8a, ArC), 124.45 (C-8, ArC), 119.16 (C-3, ArC), 117.24 (C-7, ArC), 112.22 (C-5, ArC), 64.02 (CH_2), 15.00 (CH_3). Anal. Calc. for $C_{15}H_{11}ClN_2O_3S$: C 53.82; H 3.31; N 8.37. Found: C 71.64; H 4.09; N 6.43

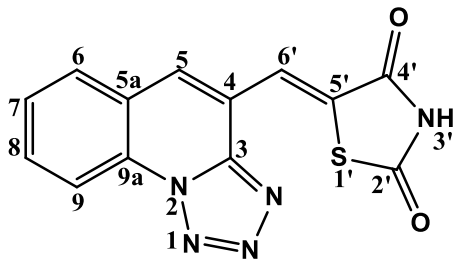
6-bromo-2,8-dichloroquinoline-3-methylene thiazolidinedi-2,4-one 55G



The experimental procedure employed for the synthesis of **55A** was followed using 6-bromo-2,8-dichloroquinoline-3-carbaldehyde **54G** (1.03 g, 3.39 mmol), thiazolidinedi-2,4-one (0.40 g, 3.39 mmol), anhydrous sodium acetate (0.04 g, 3.39 mmol) and 10 mL of glacial acetic acid. Work-up afforded a solid crude product which was recrystallized from DMF to afford 6-bromo-2,8-dichloroquinoline-3-methylene thiazolidinedi-2,4-one **55G** (0.20 g, 19.42 %) as a cream-white solid, m.p 128.8-129.3 ºC; IR ν_{max}/cm^{-1} 3242.34 (N-H), 2226.91 (ArC=C), 1662.64 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 10.00 (bs, 1H, NH), 8.37 (s, 1H, 6'-H, [ArCH=C]), 8.11 (s, 1H, 4-H, ArH), 7.78 (d, 1H, J_{H-H} meta = 2.00, 7-H, ArH), 7.56 (dd, 1H, J_{H-H} meta, ortho = 2.00 Hz and 8.80 Hz, 6-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 160.98 (C-6'), 134.30 (C-2, ArC), 132.04 (C-4, ArC), 131.10 (C-7, ArC), 124.98 (C-3, ArC), 124.82 (C-6, ArC), 116.58 (C-8, ArC).

6.6 Synthesis of (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one analogues[86] 56A-G.

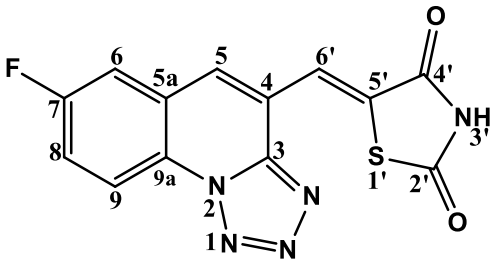
(Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one 56A



2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A** (1.03 g, 5 mmol) was transferred into a 100 mL round bottom flask, to which sodium azide (0.46 g, 8.70 mmol) was added followed by 1:10 AcOH and EtOH solution and the resulting mixture refluxed for 2 hours. After the completion of the reaction, monitored by TLC, the solid product was washed with ethanol (10 mL) and dried overnight. The crude product was

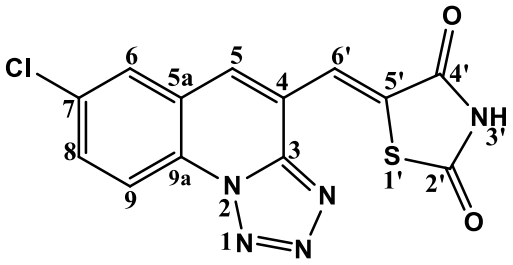
recrystallised from 2:3 DMF / EtOH to afford (Z)-5-(tetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56A** (0.81 g, 76.42 %) as a yellow solid, m.p 313.8-314.2 ºC; IR ν_{max}/cm^{-1} 3149.76 (N-H), 2829.57 (ArC=N), 1620.21 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.24 (bs, 1H, NH), 10.16 (s, 1H, C-6'), 8.50 (s, 1H, 5-H, ArH), 7.92 (d, 1H, J_{H-H} ortho = 8.00 Hz, 6-H, ArH), 7.67 (dd, 1H, J_{H-H} meta, ortho = 1.20 Hz and 8.40 Hz, 7-H, ArH), 7.36 (d, 1H, J_{H-H} ortho = 8.40 Hz, 8-H, ArH), 7.27 (d, 1H, J_{H-H} ortho = 7.60 Hz, 9-H ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.23 (C-6'), 161.89 (C-2'), 142.89 (C-5, ArC), 141.60 (C-3, ArC), 134.15 (C-8, ArC), 131.37 (C-6, ArC), 126.06 (C-9a, ArC), 123.12 (C-7, ArC), 118.59 (C-9, ArC), 115.87 (C-4, ArC). Anal. Calc. for $C_{13}H_7N_5O_2S$: C 52.52; H 2.37; N 23.56. Found: C 72.66; H 3.97; N 8.25

(Z)-5-((7-fluorotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one 56B



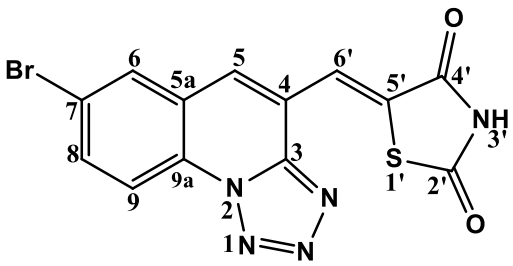
The experimental procedure employed for the synthesis of **56A** was followed using 6-fluoro-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55B** (1.03 g, 5 mmol), NaN_3 (0.43 g, 8.20 mmol) and 1:10 AcOH and EtOH solution. Work-up afforded a solid crude product which was recrystallized from 2:3 DMF and EtOH to afford (Z)-5-(7-fluorotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56B** (0.64 g, 60.95 %) as a yellow solid, m.p 240.9-241.8 ºC; IR ν_{max}/cm^{-1} 3523.95 (N-H), 2347.37 (ArC=N), 1604.77 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.20 (bs, 1H, NH), 10.23 (s, 1H, 6'-H), 8.48 (s, 1H, 5-H, ArH), 7.81 (d, 1H, J_{H-H} meta = 2.80 Hz, 9-H, ArH), 7.60 (d, 1H, J_{H-H} meta = 2.40 Hz, 6-H, ArH), 7.40 (d, 1H, J_{H-H} ortho = 2.80 Hz, 8-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.24 (C-6'), 161.65 (C-2'), 158.73 (C-7, ArC), 156.35 (C-3), 142.00 (C-5, ArC), 138.42 (C-4, ArC), 126.84 (C-5a, ArC), 122.56 (C-9, ArC), 119.21 (C-9a, ArC), 117.98 (C-8, ArC), 115.57 (C-6, ArC).

(Z)-5-((7-chlorotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one 56C



The experimental procedure employed for the synthesis of **56A** was followed using 2,6-dichloroquinoline-3-methylene thiazolidinedi-2,4-one **55C** (0.70 g, 3.50 mmol), NaN_3 (0.28 g, 3.70 mmol), and 1:10 AcOH and EtOH solution. Work-up afforded a solid crude product which was recrystallized from 2:3 DMF and EtOH to afford (Z)-5-(7-chlorotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56C** (0.26 g, 36.11 %) as a yellow solid, m.p 359.6-360.9 ºC; IR ν_{max}/cm^{-1} 3545.16 (N-H), 2310.72 (ArC=N), 1616.35 (ArC=C); ^1H NMR: (DMSO-d_6, 400 MHz): δ_H 12.27 (bs, 1H, NH), 10.23 (s, 1H, 6'-H), 8.47 (s, 1H, 5-H, ArH), 8.03 (d, 1H, J_{H-H} meta = 2.40 Hz, 6-H, ArH), 7.68 (d, 1H, J_{H-H} meta, ortho = 2.40 Hz and 8.80 Hz, 8-H, ArH), 7.37 (d, 1H, J_{H-H} ortho = 8.80 Hz, 9-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.41 (C-6'), 162.17 (C-2'), 141.51 (C-5, ArC), 140.92 (C-3, ArC), 133.67 (C-8, ArC), 129.86 (C-7, ArC), 126.79 (C-4, ArC), 126.59 (C-5a, ArC), 119.76 (C-9a, ArC), 118.31 (C-6, ArC). Anal. Calc. for $C_{13}H_7N_5O_2S$: C 47.07; H 1.82; N 21.11. Found: C 56.85; H 2.29; N 6.68

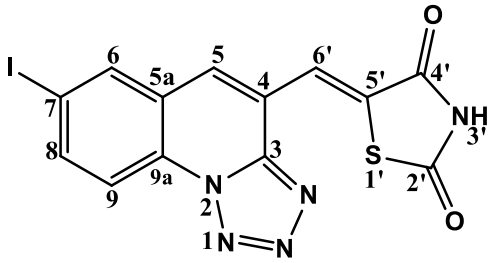
(Z)-5-((7-bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one 56D



The experimental procedure employed for the synthesis of **56A** was followed using 6-bromo-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55D** (1.03 g, 5.00 mmol), NaN_3 (0.36 g, 5.60 mmol), and 1:10 AcOH and EtOH solution. Work-up afforded a solid crude product which was recrystallized from 2:3 DMF:EtOH to afford (Z)-5-(7-

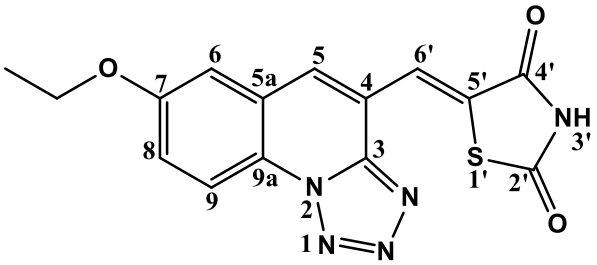
bromotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56D** (0.93 g, 88.57 %) as a yellow solid, m.p 346.3-347.1 ºC; IR ν_{max}/cm^{-1} 3132.57 (N-H), 2025.12 (ArC=C), 1609.58 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.26 (bs, 1H, NH), 10.21 (s, 1H, 6'-H), 8.46 (s, 1H, 5-H, ArH), 8.17 (d, 1H, J_{H-H} meta = 2.00 Hz, 6-H, ArH), 7.79 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 8.80 Hz, 8-H, ArH), 7.30 (d, 1H, J_{H-H} ortho = 8.80 Hz, 9-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.10 (C-6'), 161.55 (C-2'), 141.71 (C-5, ArC), 140.49 (C-3), 136.41 (C-8, ArC), 132.41 (C-9a, ArC), 126.82 (C-5a, ArC), 120.23 (C-7, ArC), 118.05 (C-6, ArC), 114.60 (C-4'). Anal. Calc. for $C_{13}H_7BrN_5O_2S$: C 41.51; H 1.61; N 18.62. Found: C 56.19; H 2.18; N 8.63

(Z)-5-((7-iodotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one 56E



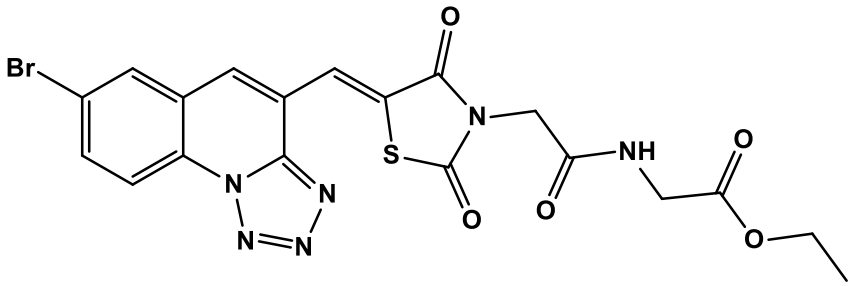
The experimental procedure employed for the synthesis of **56A** was followed using 6-iodo-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55E** (0.80 g, 4.10 mmol), NaN_3 (0.26 g, 3.34 mmol), and 1:10 AcOH and EtOH solution. Work-up afforded a solid crude product which was recrystallized from 2:3 DMF / EtOH to afford (Z)-5-((7-iodotetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56E** (0.57 g, 67.86 %) as a yellow solid, m.p 320.1-320.7 ºC; IR ν_{max}/cm^{-1} 3446.79 (N-H), 2227.78 (ArC=N), 1604.77 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.25 (bs, 1H, NH), 10.20 (s, 1H, 6'-H), 8.41 (s, 1H, 5-H, ArH), 8.30 (d, 1H, J_{H-H} meta = 1.60 Hz, 6-H, ArH), 7.90 (dd, 1H, J_{H-H} meta, ortho = 1.60 Hz and 8.40 Hz, 8-H), 7.15 (d, 1H, J_{H-H} ortho = 8.80 Hz, 9-H, ArH); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.09 (C-6'), 161.66 (C-2'), 141.40 (C-8, ArC), 141.61 (C-6), 140.86 (C-3, ArC), 139.04 (C-5, ArC), 126.57 (C-9a, ArC), 120.76 (C-4, ArC), 118.05 (C-9, ArC), 86.22 (C-7, ArC). Anal. Calc. for $C_{13}H_6IN_5O_2S$: C 36.90; H 1.43; N 16.55. Found: C 42.95; H 1.56; N 4.57

(Z)-5-((7-ethoxytetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one 56F



The experimental procedure employed for the synthesis of **56A** was followed using 6-ethoxy-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55F** (0.60 g, 3.00 mmol), NaN_3 (0.23 g, 4.23 mmol), and 1:10 AcOH and EtOH solution. Work-up afforded a solid crude product which was recrystallized from 2:3 DMF and EtOH to afford (Z)-5-(7-ethoxytetrazolo [1,5-a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one **56F** (0.29 g, 48.33 %) as a yellow solid, m.p 291.0-291.5 ºC; IR ν_{max}/cm^{-1} 3502.73 (N-H), 2017.54 ArC=N), 1624.06 (ArC=C); ^1H NMR: (DMSO-d_6, 400 MHz): δ_H 12.17 (bs, 1H, NH), 10.23 (s, 1H, 6'-H), 8.44 (s, 1H, 5-H, ArH), 7.45 (d, 1H, J_{H-H} meta = 1.60 Hz, 6-H, ArH), 7.34 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 9.20 Hz, 8-H, ArH), 7.31 (d, 1H, J_{H-H} ortho = 2.40 Hz, 9-H, ArH), 4.01 (q, 2H, J_{H-H} = 6.80 Hz, CH_2), 1.36 (t, 3H, J_{H-H} = 7.20 Hz, CH_3); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.37 (C-6'), 161.55 (C-2'), 154.21 (C-7, ArC), 142.34 (C-5), 136.34 (C-3, ArC), 126.11 (C-9a, ArC), 124.48 (C-9, ArC), 119.18 (C-4, ArC), 117.26 (C-8, ArC), 112.23 (C-6, ArC), 64.04 (CH_2), 14.99 (CH_3). Anal. Calc. for $C_{15}H_{11}N_5O_3S$: C 52.78; H 3.25; N 20.52. Found: C 71.40; H 4.18; N 6.32

6.7 Synthesis of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate[87] 57

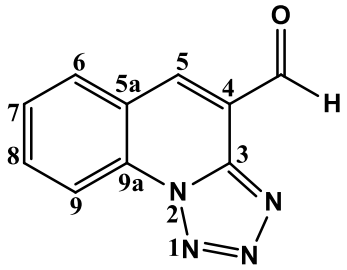


(Z)-5-((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56D** (0.40 g, 1.17 mmol) was added into 10mL EtOH in 100 mL round bottom flask with continuous stirring, KOH (0.07 g, 1.29 mmol) was added to the above resulting mixture, stirred at room temperature for 1 hour and warmed in water bath for 10 minutes. The resulting intermediate of solid potassium salt of (Z)-5-((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one was filtered and dried overnight. The solid potassium salt of (Z)-5-((7-bromotetrazolo [1,5a] quinolin-4-yl) methylene) thiazolidinedi-2,4-one (0.44 g, 1.05 mmol) in 10 mL DMF, ethyl-2-(2-chloroacetamido) acetate **65** (0.19 g, 1.05 mmol) was slowly added with continuous stirring. The resulting mixture was refluxed to the completion and monitored through TLC. After the completion of the reaction, the resulting mixture was poured into 20 mL ice cold water and filtered the solid product. The resultant solid product was recrystallised using DMF to afford (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57** (0.21 g, 38.89 %) as a yellow solid m.p 320.5-321.8 ºC; IR ν_{max}/cm^{-1} 3502.73 (N-H), 2017.54 ArC=N), 1624.06 (ArC=C); ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 10.25 (s, 1H, 6'-H [ArCH=C]), 8.79 (s, 1H, 5-H, ArH), 8.29 (d, 1H, J_{H-H} meta = 2.40 Hz, 6-H, ArH), 7.89 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 9.20 Hz, 8-H, ArH), 7.31 (d, 1H, J_{H-H} ortho = 9.20 Hz, 9-H, ArH), 5.04 (bs, 1H, 9'-H [N-H]), 4.15 (q, 2H, J_{H-H} = 6.80 Hz, 12'-H [CH_2]), 3.88 (d, 2H, J_{H-H} = 5.60 Hz, 10'-H [CH_2]), 3.37 (s, 2H, 7'-H [CH_2]), 1.36 (t, 3H, J_{H-H} = 7.20 Hz, 13'-H [CH_3]); ^{13}C NMR: (DMSO-d_6, 100 MHz): δ_C 190.37 (C-6', [ArCH=C]), 169.94 (C-2'), 167.42 (C-8'), 160.99 (C-11'), 141.19 (C-8, ArC), 141.10 (C-8, ArC), 136.45 (C-5, ArC), 133.93 (C-9, ArC), 125.92 (C-3, ArC), 121.02 (C-4, C=C), 117.85 (C-6, ArC), 115.14 (C-5', ArC), 60.98 (C-12', [CH_2]), 44.85 (C-7', [CH_2]), 41.28 (C-10', [CH_2]), 14.51 (C-

13', [CH_3]). Anal. Calc. for $C_{15}H_{11}N_5O_3S$: C 52.78; H 3.25; N 20.52. Found: C 71.40; H 4.18; N 6.32.

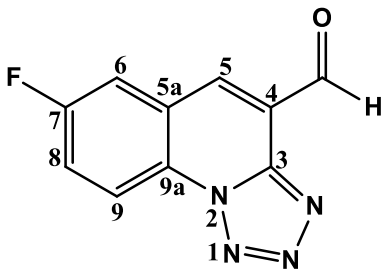
6.8 Synthesis of tetrazolo [1,5-a] quinoline-4-methylene analogues[86] 58A-B

Tetrazolo [1,5-a] quinoline-4-methylene 58A



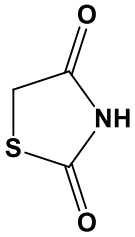
2-chloroquinoline-3-carbaldehyde **58A** (1.01 g, 4.90 mmol) was transferred into a 100 mL round bottom flask, to which sodium azide (0.46 g, 8.67 mmol) was added followed by followed by 1:10 AcOH, EtOH solution and refluxed the mixture for 2 hours. The progress of the reaction was monitored by TLC. After the completion of the reaction, the solid product was washed with 10 mL ethanol and dried overnight. The crude product was recrystallised from 2:3 DMF and EtOH to afford tetrazolo [1,5-a] quinoline-4-carbaldehyde **58A** (0.86 g, 88.66 %) as a cream-white solid, m.p 155.8-156.3 ºC, (Lit, [86] 155 ºC); IR ν_{max}/cm^{-1} 2223(C=N), 1685 (ArCHO), 1643 (C=C); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 10.58 (s, 1H, CHO), 8.78 (s, 1H, 4-H, ArH), 8.10 (d, 1H, J_{H-H} ortho = 8.40 Hz, 5-H, ArH), 8.02 (d, 1H, J_{H-H} ortho = 8.00 Hz, 8-H, ArH), 7.93 (t, 1H, J_{H-H} ortho = 7.20 Hz, 7-H, ArH), 7.69 (t, 1H, J_{H-H} ortho = 7.60 Hz, 6-H, ArH); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 189.20 (C=O), 150.12 (C-2, ArC), 149.60 (C-8a, ArC), 140.32 (C-4, ArC), 133.64 (C-7, ArC), 129.74 (C-5, ArC), 128.62 (C-6, ArC), 128.16 (C-8, ArC), 129.74 (C-4a, ArC), 126.37 (C-3, ArC).

7-fluorotetrazolo [1,5-a] quinoline-4-carbaldehyde 58B



The experimental procedure employed for the synthesis of **58A** was followed using 6-fluoro-2-chloroquinoline-3-carbaldehyde **58B** (1.03 g, 5.00 mmol), NaN_3 (0.43 g, 8.58 mmol) , 1:10 AcOH and EtOH solution. Work-up afforded a solid crude product which was recrystallized from 2:3 DMF and EtOH to afford 7-fluoro tetrazolo [1,5-a] quinoline-4-carbaldehyde **58B** (0.76 g, 70.37 %) as a pale-yellow solid, m.p 155.8-156.3 ºC; IR ν_{max}/cm^{-1} 2246 (C=N), 1708 (ArCHO), 1660 (C=C); ^1H NMR: ($CDCl_3$, 400 MHz): δ_H 10.28 (s, 1H, CHO), 8.63 (s, 1H, 4-H, ArH), 8.25 (d, 1H, J_{H-H} ortho = 8.40 Hz, 5-H, ArH), 8.09 (dd, 1H, J_{H-H} meta, ortho = 2.40 Hz and 8.00 Hz, 7-H, ArH), 7.93 (d, 1H, J_{H-H} ortho = 7.20 Hz, 8-H, ArH)); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 189.70 (C=O), 150.89(C-2, ArC), 149.20 (C-6, ArC), 139.89 (C-4, ArC), 132.94 (C-7, ArC), 130.46 (C-5, ArC), 129.01 (C-8a, ArC), 128.54 (C-8, ArC), 127.40 (C-4a, ArC), 126.78 (C-3, ArC).

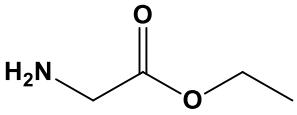
Synthesis of thiazolidinedi-2,4-one[88] 62



Water (15 mL) and chloroacetic acid (0.85 g, 9.00 mmol) was transferred to a round bottom flask with constant stirring. Thiourea (0.68 g, 9.00 mmol) was added into the above mixture and stirred for 30 minutes while the temperature was kept at 0-5ºC to form a white precipitate of 2-iminoithiazolidine-4-one. 15 mL of concentrated hydrochloric acid was then added to the above mixture and refluxed for 12 hours. The reaction was cooled in ice to form white solid crystals and further washed with 30 mL of water. The white crystals were allowed to dry overnight to afford thiazolidinedi-2,4-one **62** (0.92 g, 87.62 %) as white crystals, m.p 121.0-121.6 ºC, (Lit.,[91] 123-125 ºC);

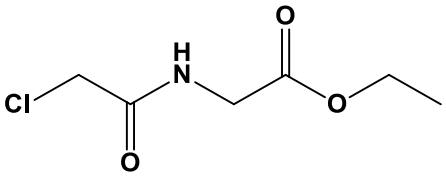
IR ν_{max}/cm^{-1} 3261.89 (N-H), 1718.03 (ArC=O), 1839.72 (C=O), 1188.04 (C=S) ; ^{1}H NMR: (DMSO-d_6, 400 MHz): δ_H 12.02 (s, 1H, NH), 4.14 (s, 2H, CH_2); ^{13}C NMR: ($DMSO_6$, 100 MHz): δ_C 174.32 (C-4), 173.57 (CH_2), 36.26 (CH_2).

Synthesis of ethyl 2-amino acetate[89] 64



Glycine (2.11 g, 26.30 mmol) in EtOH (100 mL) was cooled in an ice bath. $SOCl_2$ (13.50 mL, 185.80 mmol) was added drop wise for 30 min to the above mixture. The reaction mixture was refluxed for 12 h. The white needle-like crystals formed upon cooling in an ice bath was collected by filtration and washed with cold EtOAc (20 mL) to afford ethyl 2-amino acetate **64** (2.38 g, 87.82 %) as white needles m.p 97.8-98.3 ºC, (Lit, [87] 99 ºC); IR ν_{max}/cm^{-1} 3468 (NH_2), 1753 ($CO_2CH_2CH_3$); ^{1}H NMR: (D_2O, 400 MHz): δ_H 4.07 (s, 2H, CH_2), 4.23 (q, 2H, J_{H-H} = 7.20 Hz, CH_2), 3.18 (bs, 2H, NH_2), 1.21 (t, 3H, J_{H-H} =7.20 Hz, CH_3); ^{13}C NMR: (D_2O, 100 MHz): δ_C 168.15(C-1), 36.30 (CH_2), 40.19 (C-2), 13.18 (CH_3).

Synthesis of ethyl 2-(2-chloroacetamido) acetate[87] 65



A solution of water (5 mL) and potassium hydroxide (1.68 g, 30 mmol) was transferred to a round bottom flask in an ice bath at 0 ºC, ethyl 2-amino acetate (1.18 g, 11.44 mmol) was added to the above mixture, stirred and added DCM 10 mL was added to the above. Chloroacetyl chloride (0.79 mL, 11.44 mmol) was added dropwise while maintaining the temperature at 0 ºC and further allowed the reaction mixture to stir at room temperature for 12 hours. The mixture was extracted with ethyl acetate (15 mL×3), dried the organic extracts with sodium sulphate and washed with sodium chloride. The solvent was evaporated and the crude product was recrystallized to afford ethyl 2-(2-chloroacetamido) acetate **65** (1.03 g, 50.24 %) m.p 98.1-99.3 ºC, (Lit, [92] 98 ºC); IR ν_{max}/cm^{-1} 3408 (N-H), 1753 ($CO_2CH_2CH_3$); ^{1}H NMR: ($CDCl_3$, 400 MHz): δ_H 7.16 (bs, 1H, NH), 4.26 (q, 2H, J_{H-H} = 7.20 Hz, CH_2), 4.09 (s, 2H, CH_2), 4.07 (d, 2H,

J_{H-H} = 7.20 Hz, C-2), 1.31 (t, 3H, J_{H-H} =7.20 Hz, CH_3); ^{13}C NMR: ($CDCl_3$, 100 MHz): δ_C 168.15(C-1), 166.28 (C=O), 61.74 (CH_2), 42.34 (CH_2), 41.54 (C-2), 14.10 (CH_3).

References

1. H C Kolb, K B Sharpless. *Drug Discovery Today*. **2003**, *8*, 1128-1137.

2. G W Kabalka, A R Mereddy. *Tetrahedron*. **2005**, *46*, 6315-6317.

3. D Habibi, O Marvi. *ARKIVOC*., **2006**, 8-15.

4 S N Pandaya, A Tyagi. *Int. J. Pharm. Sci*. **2011**, *3*, 53-61.

5. J P Michael. *Nat. Prod. Rep*. **2000**, *17*, 603-620.

6. R Pingaew, S Prachayasittiku, S Ruchirawat. *Molecules*. **2010**, *15*, 988-996

7. R Chaudhary, P Tripathi, A Singh. *J. Pharm. Res*. **2014**, *8(2)*, 220-226

8. H H Renate, M G Eric, L Carmen. *Bioorg. Med. Chem. Lett*. **2010**, *20*, 942-944

9. L F Tietze, H P Bell, S A Chandrasekhar. *Chem. Int. Ed*. **2003**,*42*, 3996-4028

10. G M Craag, D J Newman. *Bioch. Biophy. Acta (BBA)-Gen. Sub*. **2013**, 1830, 3670-3695.

11. G Brahmachari. Chemistry and pharmacologogy of naturally occurring bioactive compounds. CRC Pres. **2013**, 10-11

12. M Saxena, J Saxena, R Nema, D Singh, A Gupta. *J. Pharm. Phytochem*. **2013**, 1(6), 168-182.

13. K Ilango, P Valentina, K Shubhakar, M K Kathiravan. *Austin. J. A. Pharm. Chem*. **2015**, 2(4), 1048

14. G Diaz, I Luzia, M A N Diaz. Quinolines, Isoquinolines, Agustureine, congeneric alkaloids occurrence, chemistry and biological activity. Intech chapter 6.

15. S P Charvan, M S Venkatraman. *ARKIVOC. 2005*, 3, 165-169.

16. B Klier, O Schimmer. *Mutagenesis*. **1999**, 14(2), 181-185.

17. L V Mengyin, P Xu, Y Tian, J Liang, Y Gao, F Xu, Z Zhang, J Sun. *J. Ethnopharm*. **2015**, 171, 247-263

18. D N M Machado, S F P Junior, L M Conserva, R P L Lemose. *Biochem. Syst. Eco*. **2005**, 33, 555-558.

19. M Kang, H Ou, R Wang, W Liu, A Tang. *JBUO*. **2014**, 19(1), 130-136

20. M V Madhubabu, R Shankar, R Akula, U K S Kumar, M V B Rao. *Der Pharma Chem*. **2014,** 6(4), 50-56

21. A Croaker, G J King, J H Pyne, S Anoopkumar-Dukie, L Liu. *Int. J. Mol. Sci*. **2016**, *17*, 1414

22. G G Cheng, Y Zhang, X H Cai, M F Bao, J Gu, Y Li, N Liu, X D Luo. *Tetrahedron Lett*. **2013**, *54*, 4547-4550

23. Z Ma, Y Hano, T Nomura, Y Cheng. *Bioorg. Med. Chem. Lett*. **2004**, *14*, 1193-1196.

24. R Ikan. *Sel. Topics. Chem. Nat. Chem. W. Sci*. **2008**.

25. S M Tome, A M S Silva, C M M Santos. *Curr. Org. Synt*. **2014**, 11, 317-341.

26. Y Li, Z-Y Yang, J-C Wu, *Euro. J. Med. Chem*. **2010**, *45*, 5692-5701.

27. A Zablotskaya, I Segal, A Geronikaki, I Shestakova, V Mikolajeva, G Makarenkova. *Pharmacol Rep*. **2017**, *69*, 575-581.

28. P W Chiu, A C Chan, S Leung, H Leong, K Kwong, M K Li, A C Au-Yeung, S C Chung, E K Ng. *J. Gastroint. Surg*. **2005**, 9, 794.

29. S K J Shaikh, R R Kamble, S M Samagond, H C Devarajegowda, S R Dixit, S D Josh. *Euro. J. Med. Chem*. **2017**, *128*, 258-273.

30. H M Meshram, B C Reddy, D A Kumar, M Kalyan, P Ramesh, P Kavitha, J V Rao. *IJC-B*. **2012**, *51*, 1411-1416.

31. J D Bhatt, C J Chudasama, K D Patel. *Bioorg. Med. Chem*. **2015**, *23*, 7711-7716.

32. X Nqoro, N Tobeka, B A Aderibiigbe. *Molecules*. **2017**, *22*, 2268

33. K Kaur, M Jain, R P R Jain. *Euro. J. Med. Chem*. **2010**, *45*, 3245-3264

34. S Vandekeckhove, M D'hooge. *Bioorg. Med. Chem*. **2015**, *23*, 5098-5119

35. P B Madrid, J Sherill, A P Liou, J L Weisman, J L DeRisi, R K Guy. *Bioorg. Med. Chem. Lett*. **2005**, *15*, 1015-1018

36. M C Mandewale, B R Thorat, and R S Yamgar. *Der Pharma Chemica*. **2015**, *7(5)*, 207-215.

37. G D Brown, D W Denning, N A Gow, S M Levitz, M G Netea, T C White. *Sci. Trans. Med*. **2012**, 4, 162-165.

38. A R Bhat, T A Azam, I Choi, F Athar. *Euro. J. Med. Chem*. **2011**, *46*, 3158-3166.

39. P C Sharma, A Jain, S Jain. *Acta. Pol. Pharmaceutica-Drug. Res*. **2009**, 66(6), 587-604

40. K Sudeesh, R Gururaja. *Org. Chem. Curr. Res*. **2017**, 6(2), 183

41. C Amit, C Payal, K Kuldeep, S Mansimran, S Poonam, S Kuldeep. *Can O. Pharm J*. **2014**, *1(1)*, 1-12.

42. J W Zhao, D-S Chen, C S Deng, Q Wang, W Zhu, L Lin. *BMC. Comp. Alt. Med*. **2017**, 17, 7.

43. S Kumar, A K Pandey. *Sci. W. J*. **2013**, 13, 1-16.

44. M-H Pan, C-S Lai, C T Ho. *Food. Func*. **2010**, 1, 15-31.

45. D P de Sousa. *Molecules*. **2014**, *19*, 1459-1480.

46. M H Abdelranhman, B G M Ibrahim, Abd El G A Moustafa, L Tremblu, S N A Bukhari. *Euro. J. Med. Chem*. **2017**, *124*, 972-985.

47. S A El-Feky, H K Thabet, M T Ubeid. *J. Flourine Chem*. **2014**, *16*, 87-94.

48. B Tanwar, A Kumar, K P Yogeswari, D Sriram, A K Chakraborti. *Bioorg. Med. Chem. Lett.* **2016**, *26*, 5960-5966.

49. A Nayyar, S R Patel, M Shaokh, E Countinho. *Bioorg. Med. Chem*. **2009**, *44*, 2017-2029.

50. S R Patel, R Gangwal, A T Sangamwar, R Jain. *Euro. J. Med. Chem*. **2014**, *85*, 255-267.

51. A Nayyar, V Monga, A Malde, E Countinho, R Jain. *Bioorg. Med. Chem*. **2007**, *15*, 626-640.

52. M C Mandewale, B R Thorat, and R S Yamgar. *Der Pharma Chemica*. **2015**, *7(5)*, 207-215.

53. M Miliutina, S A Ejaz, S U Khan, V O Laroshenko, A Villinger, J Iqbal, P Langer. *Euro. J. Med. Chem*. **2017**, *126*, 408-420.

54. K Plevova, K Briestenska, F Colbert, J Mistrikova, V Milata, F R Leroux. *Tetrahedron Lett*. **2015**, *56*, 5112-5115.

55. I Rosendahl, J Siemens, R Kindler, J Groeneweg, J Zimmermann, S Czerwinski, M Lamshaft, V Laabs, B-M Wilke, H Vereecken, W Amelung. *J. Environ Qual*. **2012**, 41(4), 1275-1283.

56. A Naeem, S L Badshah, M Muska, N Ahmad, K Khan. *Molecules*. **2016**, *21(4)*, 268.

57. O M M Santos, J T J Freitas, E C L Cazeday, M B de Araujo, A C Doriquetto. *Molecules*. **2016**, *21*, 328.

58. V V Kouznetsov, L Y V Mendez, C M M Gomez. *Curr. Org. Chem.* **2005**, *9*, 141-161.

59. G Jones, A R Katritzky, C W Rees , E F V Scriven. *CHEC-III.* **1996**. *5*, 167.

60. C S Cho, B H Oh, J S Kim, T J Kim, S C Shim. *J. Chem. Soc. Chem. Commun*. **2000**, *19*, 1885.

61. M E Theclitou, L A Robinson. *Tetrahedron Lett*. **2002**, *43*, 3907.

62. J Safari, S H Banitaba, S S Samies. J. Chem. Sci. **2009**, 121(4), 481-484.

63. Z Song, DL Hughes. *J. Heterocycl.Chem.* **1993**, *30*, 17.

64. V P Lisimov. *Adv. Heterocycl. Chem*. **2006**, *91*, 189.

65. K Nakatani, S Samdo. *Tetrahedron Lett*. **1999**, *40*, 6029-6032.

66. D Yang, K Jiang, J Li, F Xu. *Tetrahedron*. **2007**, *63*, 7654-7658.

67. J S Yadav, P P Rao, D Sreenu, R S Rao, V N Kumar, K Nagaiah, A R Prasad. *Tetrahedron Lett.* **2015**, *46*, 7249-7253.

68. M A Khan, K Miller, K D Rainsford, Y Zhou. *Molecules*. **2013**, *18*, 3227-3240.

69. E Leyva, E Monreal, A Hernandez. *J. Fluorine. Chem*. **1999**, *94*, 7-10.

70. J J Eisch, T Dluzniewski, *J. Org. Chem.* **1989**, *54*, 1269-1274.

71. V V Kouznetsov, L Y V Mendez, C M M Gomez, *Curr. Org. Chem*. **2005**, *9*, 141-161.

72. S A Yamashkin, E A Oreshkina, *Chem. Heterocycl. Compd.* **2006**, *42*, 701-718.

73. G Gauti, S Perrozzi, R Riva. *Tetrahedron Asymmetry* **2002**, *13*, 2703-2706.

74. H Saggadi, D Luart, N Thiebault, I Polaert, L Estel, C Len, *Catal. Commun.* **2014**, *44*, 15-18.

75. Y Chao Wu, L Liu, H Jing Li, D Wang, Y Jun Chen. *J. Org. Chem*. **2006**, *71*, 6692-6595.

76. C Ludtke, A Haupt, M Wozniak, N Kulak. *J. Fluorine Chem*. **2017**, *193*, 98-105.

77. S T Selvi, V Nadaraj, S Mohan, R Sasi, M Heema. *Bioorg. Med. Chem*. **2006**, *14*, 3896.

78. R E Khidre, A A Abu-Hashem, M El-Shazly. *Euro. J. Med. Chem*. **2001**, *46*, 5057.

79. M Kidwai, S Saxena, M K-R Khan, S S Thukral. *Euro. J. Med. Chem*. **2005**, *40*, 816.

80. W Zhong, F Lin, R Chen, W Su. *Synthesis*. **2009**, *14*, 2333.

81. M Kidwai, K Singhal, R Thakur. *Lett. Org. Chem*. **2005**, *2*, 419.

82. A Chandra, B Singh, R S Khanna, R M Singh. *J. Org. Chem*. **2009**, *74*, 5664.

83.H Govender, C Mockstar, N A Koorbanally. *J. Heterocyclic. Chem*. **2018**, 55(4), 1-8.

84. S S P K Darsi, K S Kumar, B R Devi, A Naidu, P K Dubey. *Lett. Org Chem*. **2014**, 11(8), 551-555.

85. J T Urban. Nuclear Magnetic Resonance Studies of Quadrupolar Nuclei and Dipolar Field Effects. **2004**, University of Carlifonia.

86. S B Marganakop, R R Kamble, J Hoskeri, D J Prasad, G Y Meti. *Med Chem Res*. **2013**.

87. B R P Kumar, M Soni, S S Kumar, K Singh, M Patil, R B N Baig, L Adhikary. *Euro. J. Med. Chem*. **2011**, 46, 835-844.

88. K Kar, U Krithika, M P Basu, S S Kumar, A Reji, B R P Kumar. Bioorganic Chemistry. **2014**, 56, 27-33.

89. A N Maity, A C Shaikh, S Srimurugan. Chi-Ju Wu, C Chen, Shyne-Chu Ke. *Amino Acids*. **2012**, 42, 309-315.

Appendix

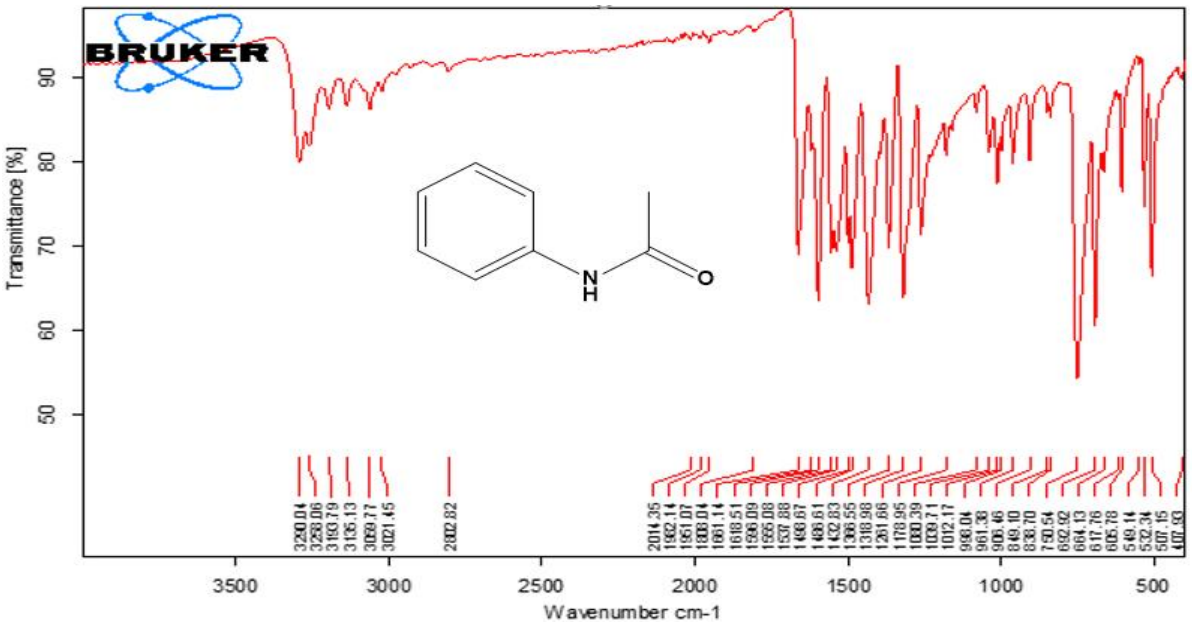


Figure 28. IR spectrum of acetanilide **53A**

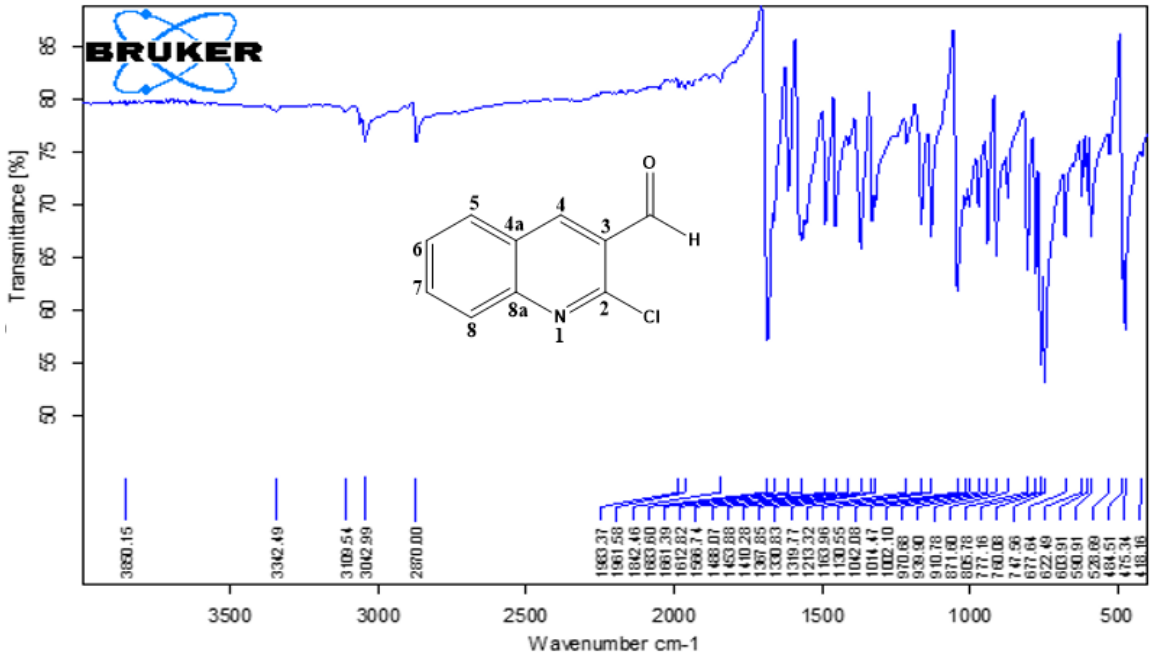


Figure 29. IR spectrum of 2-chloroquinoline-3-carbaldehyde **54A**

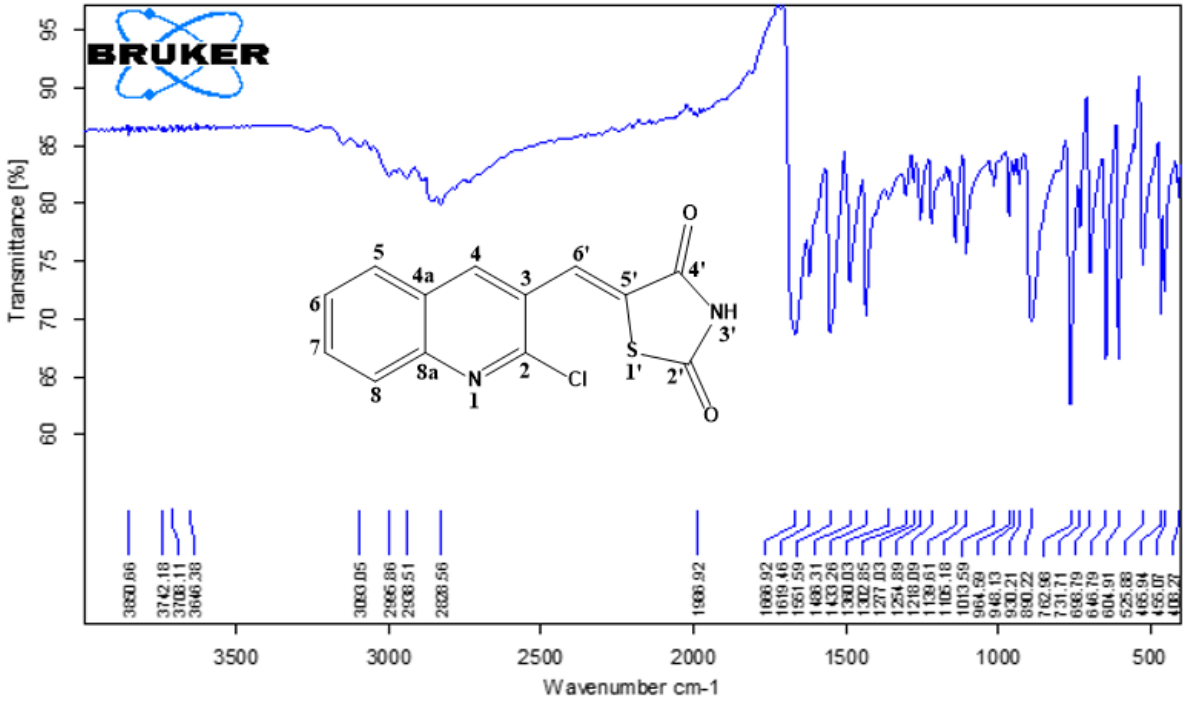


Figure 30: IR spectrum of 2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55A**

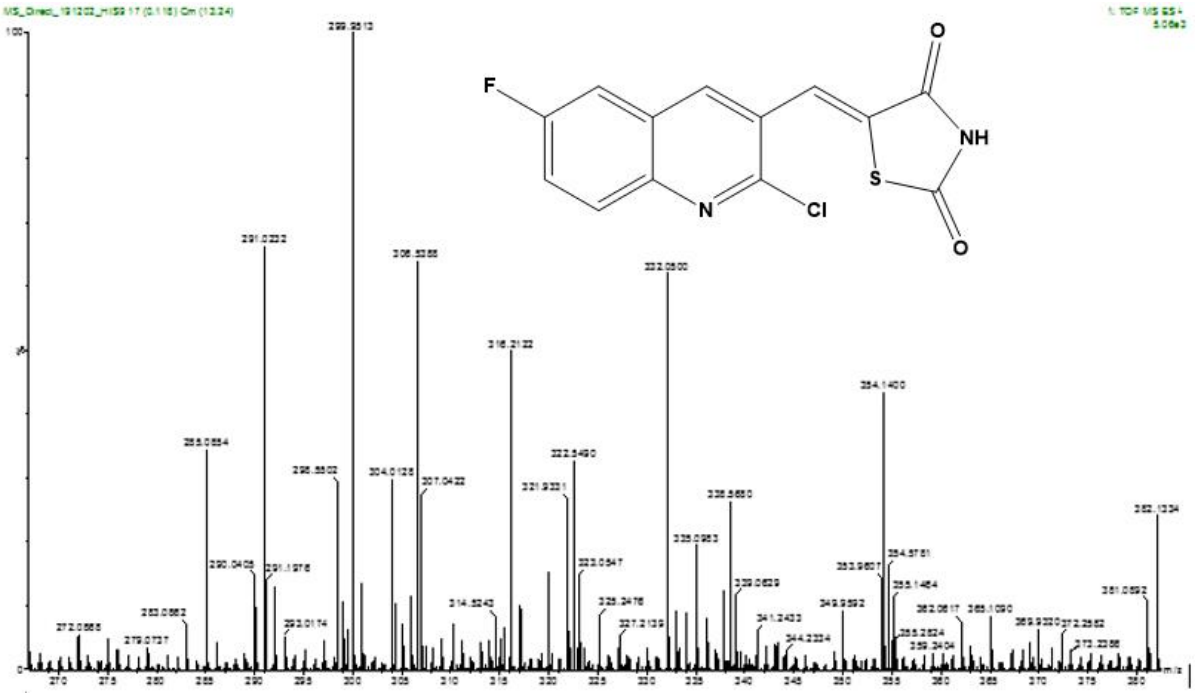


Figure 31: HRMS spectrum of 6-fluoro-2-chloroquinoline-3-methylene thiazolidinedi-2,4-one **55B.**

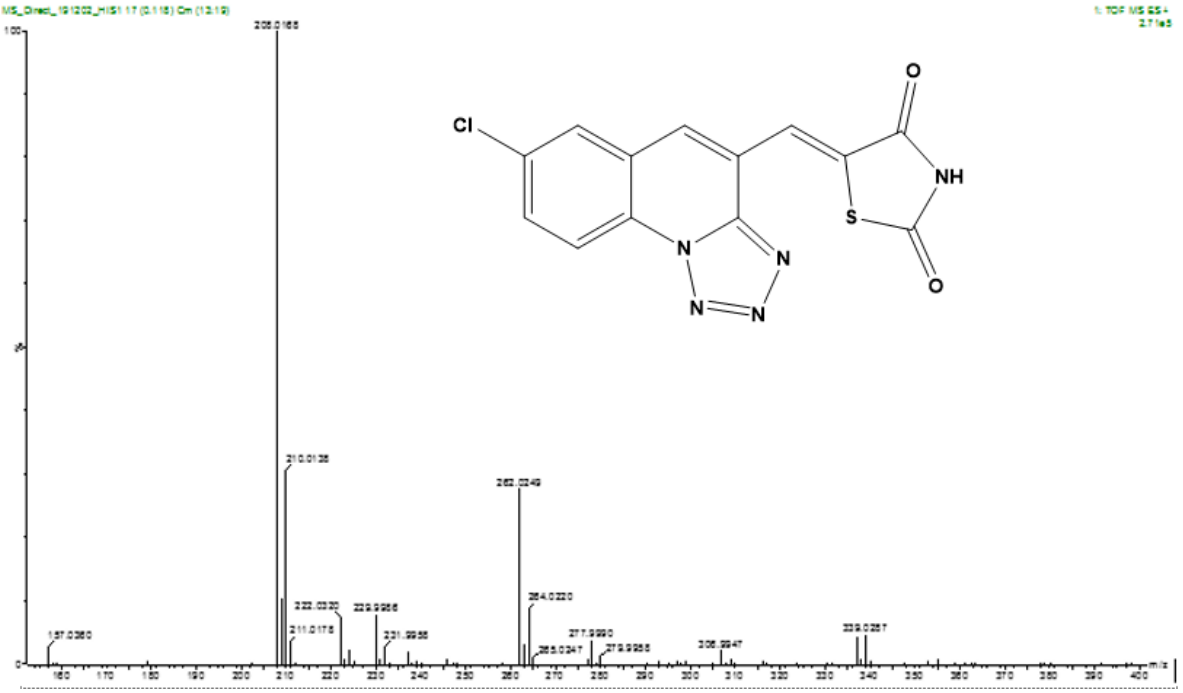


Figure 32. HRMS spectrum of (Z)-5-((7-chlorotetrazolo [1,5a] quinoline-4-yl) methylene) thiazolidinedi-2,4-one **56C**.

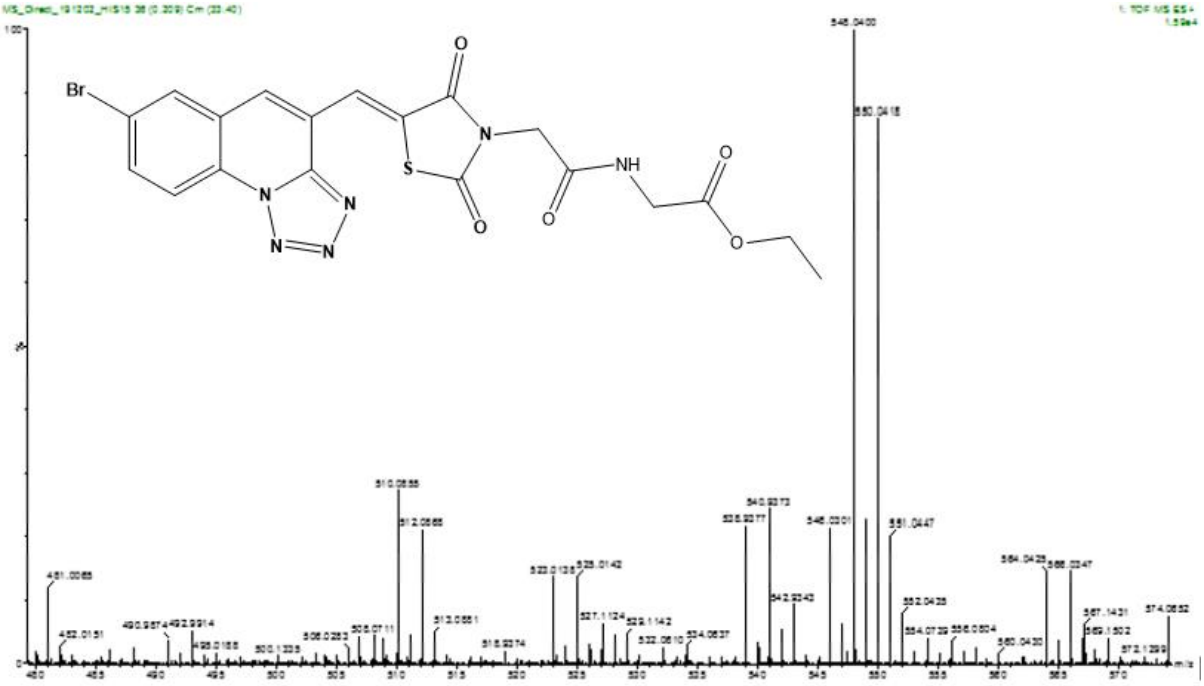


Figure 33: HRMS of (Z)-ethyl-2-(2-5-((7-bromotetrazolo [1,5a] quinoline-4-yl) methylene-2,4-dioxothiazolidin-3-yl) acetamido) acetate **57**.