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**SYNTHESIS, CHARACTERIZATION AND ANTIBACTERIAL EVALUATION OF
DIBENZALACETONE AS A MONOCARBONYL CURCUMIN ANALOGUE
AGAINST ESCHERICHIA COLI**

BY

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**A REPORT SUBMITTED TO THE FACULTY OF SCIENCE AND EDUCATION IN
PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF A
BACHELOR'S OF SCIENCE WITH EDUCATION OF BUSITEMA UNIVERSITY.**

MAY, 2026

DECLARATION

I **Namuganza Hadija** declare that this research project is my original work and has not been submitted to any other institution for academic credit.

Signature 

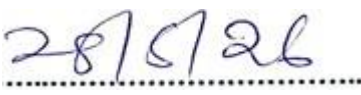
Date... 02/09/2026

APPROVAL

This research project has been submitted with the approval of the supervisor.

Supervisor's name : **Andima Moses**

Signature : 

Date : 

DEDICATION

I dedicate this book to my beloved father Mr. NGOBI JUMA for the support, care rendered to me work from the start up to the end. It's my pleasure to thank him for the so much efforts and support he has given me to see my success. May the almighty Allah bless him.

I also dedicate this work to my beloved friends most especially Mr. Kajubi Paul, Mr. Bogere sharif, Ms. Nerima Gloria, Ms. Mukyala Mariam, Ms. Mbwaali Judith, Mr. Nabongho Noah et al who have supported me emotionally, financially, physically and for always being available throughout this course.

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ABSTRACT

Curcumin is a naturally occurring polyphenolic compound isolated from turmeric and is known for its antimicrobial, antioxidant, anti-inflammatory, and anticancer properties. However, the practical application of curcumin has been limited due to its poor bioavailability and chemical instability. This research focused on the synthesis of monocarbonyl curcumin analogues and evaluation of their antimicrobial activity against *Escherichia coli*. The synthesized analogue was purified by recrystallization and characterized using ultraviolet-visible spectroscopy and the absorbance was found to be 0.485 at wavelength of 330nm, melting point determination and its melting point was found to be 119⁰ C and Fourier Transform Infrared Spectroscopy (FTIR) for functional group analysis. Antimicrobial activity was determined using agar well diffusion and minimum inhibitory concentration methods.

The synthesized monocarbonyl curcumin analogue exhibited a yellow-orange crystalline appearance with a relatively sharp melting point indicating purity. UV-visible spectroscopy showed characteristic absorption peaks associated with conjugated systems while FTIR analysis confirmed the presence of aromatic rings, hydroxyl groups, carbonyl groups, and alkene functionalities. Antimicrobial evaluation demonstrated inhibitory effects against *Escherichia coli*. The synthesized analogue exhibited zones of 2.25, 7.25 and 10.50mm at concentrations of 0.25, 0.5 and 1.0mg/L respectively. The percentage yield of the synthesized analogue was found to be 85%. The results indicated that structural modification of curcumin improves stability and enhances antimicrobial activity.

The study concludes that monocarbonyl curcumin analogues are promising antimicrobial agents and may contribute to development of novel therapeutic compounds against resistant bacterial strains.

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ABBREVIATIONS

ABBREVIATION	MEANING
AMR	Antimicrobial resistance
WHO	World Health Organization
MIC	Minimum Inhibitory Concentration
FTIR	Fourier Transform Infra-Red
UV-Vis	Ultra Visible
MHA	Muller Hinton Agar

CHAPTER ONE: INTRODUCTION

1.1 Background.

Curcumin is a naturally occurring polyphenolic compound isolated from the rhizomes of turmeric (*Curcuma longa*). It has been widely used in traditional medicine for centuries, particularly in Asian countries, due to its diverse pharmacological properties. (Chongshan Dai 1, 2022) These include anti-inflammatory, antioxidant, anticancer, and antimicrobial activities. The biological activity of curcumin is largely attributed to its unique chemical structure, which consists of two aromatic rings connected by a conjugated β -diketone system. (Upadhyaya, 2021) This conjugated system allows electron delocalization, making curcumin highly reactive toward biological targets.

Antimicrobial resistance (AMR) has become one of the greatest global public health threats of the twenty-first century. The World Health Organization (WHO) recognizes AMR as a major challenge because bacteria, fungi, viruses, and parasites are increasingly developing resistance to medicines that were once effective in treating infections. (Marcin Górski¹, 2022) As a result, common infectious diseases are becoming more difficult to treat, leading to prolonged illness, increased healthcare costs, treatment failure, and higher mortality rates. It is estimated that antimicrobial-resistant infections contribute to millions of deaths worldwide each year, and if effective interventions are not implemented, the burden is expected to increase significantly in the coming decades.

Among the pathogens contributing to antimicrobial resistance, *Escherichia coli* (*E. coli*) is one of the most clinically important. It is a Gram-negative bacterium commonly found in the intestinal tract of humans and animals, where it normally exists as part of the natural microbiota. (Oglah, 2020) However, pathogenic strains of *E. coli* cause a wide range of infections including urinary tract infections, diarrhoeal diseases, neonatal meningitis, wound infections, septicemia, and foodborne illnesses. The increasing emergence of multidrug-resistant *E. coli* strains has reduced the effectiveness of commonly used antibiotics, making treatment more difficult and increasing the risk of severe disease and death.

The high resistance of Gram-negative bacteria such as *E. coli* is largely attributed to their unique cell wall structure. Unlike Gram-positive bacteria, Gram-negative bacteria possess an outer membrane containing lipopolysaccharides that acts as an additional permeability barrier.

This outer membrane restricts the entry of many antimicrobial agents while specialized efflux pumps actively remove toxic compounds from the bacterial cell. (Syeda Nazish Sohaib, 2025) Furthermore, many Gram-negative bacteria produce enzymes such as β -lactamases that destroy antibiotics before they reach their target sites. These protective mechanisms make Gram-negative bacteria particularly difficult to eliminate using conventional antibiotics.

The increasing failure of conventional antibiotics has renewed scientific interest in natural products as alternative sources of antimicrobial agents. Natural products have historically contributed significantly to drug discovery, with many clinically useful antibiotics, anticancer drugs, and anti-inflammatory agents originating from plants, microorganisms, and marine organisms. Plant-derived secondary metabolites are particularly attractive because they possess diverse chemical structures capable of interacting with multiple biological targets while often exhibiting relatively low toxicity.

One of the most extensively studied natural products is curcumin, a yellow polyphenolic compound isolated from the rhizomes of *Curcuma longa* (turmeric). Curcumin has been used for centuries in traditional medicine, particularly in Asian countries, because of its anti-inflammatory, antioxidant, antimicrobial, antiviral, anticancer, and wound-healing properties. The biological activities of curcumin are largely attributed to its unique chemical structure, which consists of two aromatic rings connected by a conjugated β -diketone chain. This highly conjugated system allows extensive electron delocalization, enabling the molecule to interact with proteins, enzymes, nucleic acids, and reactive oxygen species.

Despite its promising therapeutic potential, curcumin suffers from several major limitations that hinder its practical application. One of the most significant challenges is its poor chemical stability under physiological conditions. (Oglah, 2020) The β -diketone moiety undergoes keto–enol tautomerism, which contributes to rapid degradation, especially at neutral and alkaline pH. In addition, curcumin exhibits low water solubility, poor absorption, and rapid metabolism in the body, leading to low bioavailability. These limitations have motivated researchers to design and synthesize structural analogues that retain the beneficial properties of curcumin while overcoming its shortcomings.

Synthetic curcumin analogues are modified derivatives designed to improve stability, solubility, and biological activity. (Oglah, 2020) These analogues often involve modification of the central β -diketone moiety, substitution on the aromatic rings, or incorporation of

heterocyclic structures. For instance, replacing the β -diketone with a monocarbonyl group eliminates keto–enol tautomerism, thereby enhancing chemical stability. Similarly, the introduction of heterocyclic rings can improve binding interactions with biological targets and increase resistance to metabolic degradation.

To overcome these limitations, medicinal chemists have developed numerous synthetic curcumin analogues. Structural modification aims to improve chemical stability, biological activity, pharmacokinetic properties, and bioavailability while retaining the beneficial pharmacological effects of natural curcumin. Among these derivatives, monocarbonyl curcumin analogues have attracted considerable attention because replacing the β -diketone moiety with a single carbonyl group eliminates keto-enol tautomerism, thereby enhancing chemical stability. (Oglah, 2020)

Dibenzalacetone is one of the simplest and most widely studied monocarbonyl curcumin analogues. It is synthesized through Claisen-Schmidt condensation between benzaldehyde and acetone under basic conditions. Besides being chemically stable, dibenzalacetone possesses an extended conjugated system that contributes to antioxidant and antimicrobial activities. Previous studies have demonstrated that dibenzalacetone and related monocarbonyl analogues exhibit improved antibacterial activity compared with natural curcumin, particularly because their increased hydrophobicity enhances interaction with bacterial membranes.

Several mechanisms have been proposed to explain the antibacterial activity of curcumin analogues. These compounds may disrupt bacterial cell membranes, generate reactive oxygen species that damage cellular components, inhibit bacterial cell division proteins such as FtsZ, interfere with DNA replication, and inhibit essential metabolic enzymes. Their multiple mechanisms of action reduce the likelihood of rapid development of bacterial resistance, making them promising candidates for future antimicrobial drug development.

Although numerous studies have investigated curcumin analogues internationally, relatively little information exists regarding the synthesis and biological evaluation of these compounds within Uganda. Most antimicrobial studies have focused on natural curcumin, while comparatively fewer investigations have evaluated simple monocarbonyl analogues such as dibenzalacetone using locally available laboratory facilities. This knowledge gap limits the development of affordable antimicrobial agents that could potentially supplement conventional antibiotics.

Therefore, this study synthesized dibenzalacetone as a monocarbonyl curcumin analogue using the Claisen-Schmidt condensation reaction, characterized the synthesized compound using melting point determination, UV-Visible spectroscopy, and Fourier Transform Infrared (FTIR) spectroscopy, and evaluated its antibacterial activity against *Escherichia coli*. The findings are expected to contribute to the growing body of knowledge on curcumin-derived antimicrobial agents and provide useful information for future medicinal chemistry research.

In recent years, there has been increasing interest in the antibacterial properties of curcumin and its analogues. The rise of antibiotic-resistant bacteria has become a major global health concern, necessitating the search for new and effective antimicrobial agents. Curcumin has demonstrated moderate antibacterial activity against both Gram-positive and Gram-negative bacteria, but its effectiveness is limited by its physicochemical properties. (Syeda Nazish Sohaib, 2025) Synthetic analogues, however, have shown enhanced antibacterial activity due to improved structural features that facilitate interaction with bacterial cells.

Escherichia coli is a Gram-negative bacterium commonly used as a model organism in microbiological studies. It is also a significant pathogen responsible for various infections, including urinary tract infections, diarrhea, and septicemia. The outer membrane of Gram-negative bacteria like *E. coli* presents a barrier to many antimicrobial agents, making them inherently more resistant. Therefore, evaluating the antibacterial activity of curcumin analogues against *E. coli* provides valuable insights into their potential as therapeutic agents. (Marcin Górski¹, 2022)

This study focuses on the synthesis of selected curcumin analogues and the evaluation of their antibacterial activity against *Escherichia coli*. By modifying the chemical structure of curcumin, it is expected that the resulting analogues will exhibit improved stability and enhanced antibacterial properties. The findings of this study may contribute to the development of novel antimicrobial agents and provide a deeper understanding of the relationship between chemical structure and biological activity.

1.2 Statement of the Problem

The increasing prevalence of antimicrobial-resistant *Escherichia coli* poses a serious threat to effective disease management in Uganda. Commonly used antibiotics are becoming less

effective, leading to prolonged illness, increased healthcare costs, and higher risk of complications. Although curcumin has demonstrated antimicrobial properties, its natural form has limited effectiveness against Gram-negative bacteria such as E- coli. Furthermore, there is limited research in Uganda focusing on the synthesis and antimicrobial evaluation of curcumin analogues. This gap in knowledge necessitates research into curcumin-based compounds that may offer improved antibacterial activity against E-coli.

1.3 Justification of the Study

This study is justified for several reasons. First, E. coli infections are common in Uganda and are increasingly difficult to treat due to antimicrobial resistance. Identifying alternative antimicrobial agents could help reduce dependence on conventional antibiotics. Second, curcumin is derived from turmeric, a plant that is locally available and culturally accepted in Uganda. Third, synthesizing curcumin analogues provides an opportunity to improve the antimicrobial properties of curcumin through chemical modification. Finally, the study will contribute to undergraduate training by providing practical experience in organic synthesis, compound characterization, and microbiological testing.

1.4 Objectives of the Study

1.4.1 General Objective

To synthesize curcumin analogues and evaluate their antimicrobial effects against *Escherichia coli*.

1.4.2 Specific Objectives

- I. To synthesize selected curcumin analogues using standard organic synthesis methods.
- II. To purify and characterize the synthesized curcumin analogues.
- III. To determine the antimicrobial activity of the synthesized curcumin analogues against *Escherichia coli*.

1.6 Scope of the Study

The study will focus on laboratory synthesis of a limited number of curcumin analogues and their in vitro antimicrobial evaluation against E. coli. The study will not involve animal or human testing and will be conducted within the available laboratory facilities.

1.7 Significance of the Study

The increasing emergence of antibiotic-resistant bacteria poses a serious global public health challenge. Many commonly used antibiotics are gradually losing their effectiveness due to misuse, overuse, and microbial resistance. This situation creates an urgent need to explore alternative and affordable antimicrobial agents, especially in developing countries like Uganda.

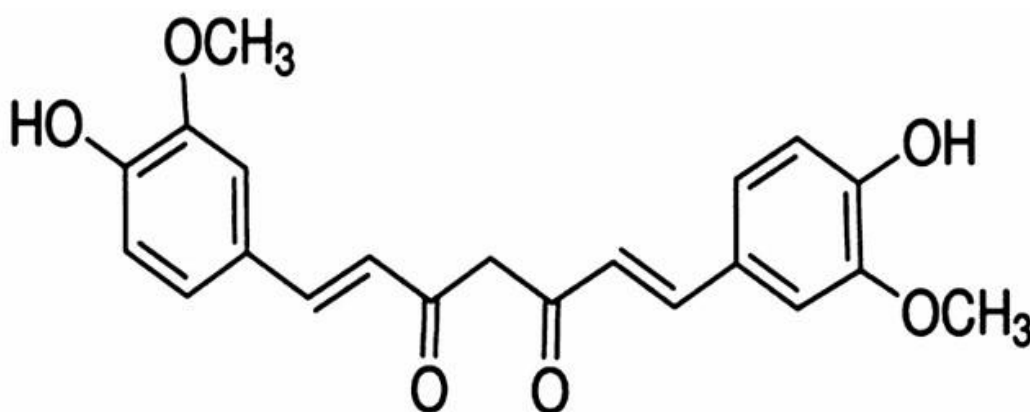
CHAPTER TWO: LITERATURE REVIEW

2.1 The chemistry of curcumin

The IUPAC name of curcumin is (1E, 6E)-1,7-bis (4- hydroxy-3-methoxyphenyl)-1,6heptadiene-3,5-dione with chemical formula and molecular weight as $C_{21}H_{20}O_6$ and 368.38 g/mol, respectively. (Upadhyaya, 2021)

Curcumin (diferuloylmethane, $C_{21}H_{20}O_6$) is a bis- α , β -unsaturated β -diketone: two feruloyl (4hydroxy-3-methoxyphenyl) groups connected by a seven-carbon heptadienedione linker. This molecule exists in keto-enol tautomeric equilibrium, the diketo form dominates in acidic/neutral solution and in cell membranes, while the enol form is favored in basic conditions. It has four reactive sites: the two phenolic OH groups, the methoxy groups, the central active methylene carbon, and the conjugated diketone system itself. (Dalia R. Emam 1, 2017)

That β -diketone core is also the source of curcumin's biggest liabilities. It's chemically unstable (degrades rapidly at physiological pH), poorly water-soluble, rapidly metabolized, and behaves as a **PAINS compound** (pan-assay interference substance) it covalently labels proteins, chelates metals, generates redox activity, aggregates, and disrupts membranes in ways that can produce false-positive bioactivity readings in lab assays. (Dalia R. Emam 1, 2017)



Structure of curcumin.

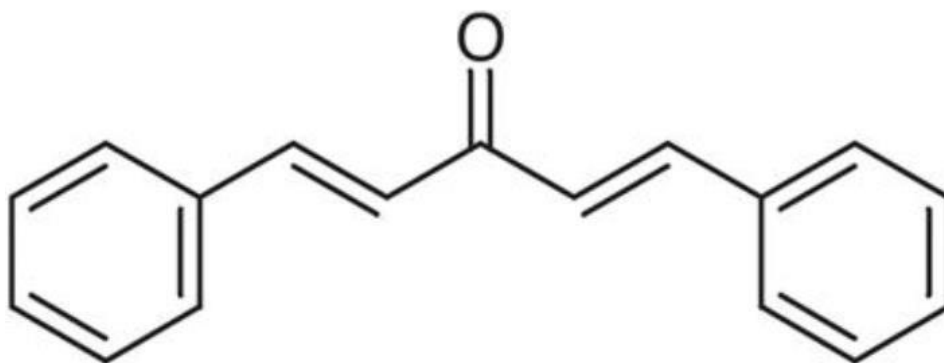
The presence of phenolic hydroxyl groups contributes to curcumin's antioxidant properties, as these groups can donate hydrogen atoms to neutralize free radicals. The methoxy groups on the aromatic rings influence electron density and enhance resonance stabilization. The conjugated double bonds allow curcumin to absorb visible light, one in the visible region of the spectrum with highest absorption from 410 to 430 nm and another band is in the ultraviolet region with highest absorption at 265 nm, giving it its characteristic yellow color. (Upadhyaya, 2021) Chemically, curcumin is reactive due to its electrophilic α , β -unsaturated carbonyl groups, which can undergo nucleophilic addition reactions such as Michael addition. This reactivity plays a significant role in its interaction with biological molecules, including proteins and enzymes. However, the β -diketone system is chemically unstable, especially in alkaline conditions where hydrolysis and degradation occur rapidly. This instability is one of the main reasons for the development of synthetic analogues.

2.2 Synthetic Curcumin Analogues

Several classes of synthetic curcumin analogues have been developed to overcome the limitations of natural curcumin. These include monocarbonyl analogues, chalcone analogues, piperidone-based analogues, heterocyclic analogues and hydrogenated analogues. These compounds are more stable and exhibit strong antioxidant properties, although their antibacterial activity may differ from that of curcumin. (Kim, 2018)

Monocarbonyl analogues are among the most studied derivatives. In these compounds, the β -diketone moiety is replaced with a single carbonyl group, resulting in improved stability and resistance to hydrolysis. These analogues retain the α,β -unsaturated system, which is essential for biological activity. (Polaquini, 2021)

Figure 2.1. Structure of monocarbonyl curcumin analogue



Chalcone analogues are simpler structures consisting of two aromatic rings linked by an α,β unsaturated carbonyl system. These compounds are known for their ease of synthesis and significant biological activities, including antimicrobial effects. Their planar structure facilitates interaction with biological targets. Piperidone-based analogues incorporate a heterocyclic ring in the central linker, resulting in increased rigidity and enhanced binding affinity. The presence of nitrogen in the ring also allows for additional interactions such as hydrogen bonding. Heterocyclic analogues, such as pyrazoles, are formed by cyclization reactions involving curcumin or its intermediates. These compounds exhibit improved chemical stability and often show enhanced biological activity due to the presence of heteroatoms. Hydrogenated analogues, such as tetrahydro curcumin, are produced by reducing the double bonds in the conjugated system. These compounds are more stable and exhibit strong antioxidant properties, although their antibacterial activity may differ from that of curcumin.

2.3 Methods of preparation

There are three major synthetic routes for preparing curcumin analogues, the traditional Claisen–Schmidt condensation, microwave-assisted synthesis, and solvent-free synthesis. They differ significantly in reaction speed, energy demands, environmental impact, and overall efficiency.

The classical Claisen–Schmidt condensation is a liquid-phase, base- or acid-catalyzed aldol reaction where an aromatic aldehyde condenses with a ketone in standard organic solvents like ethanol or methanol under reflux. While it provides a well-understood, predictable chemical pathway, it suffers from severe limitations: long reaction times ranging from 2 to 24 hours, it gives moderate yields due to competing side reactions, there is high solvent consumption generating hazardous chemical wastes, and the necessity for lengthy post-reaction purification steps such as;

1. Microwave-assisted synthesis. This dramatically improves upon these drawbacks by using electromagnetic radiation to directly excite dipolar molecules and ions within the reaction mixture, inducing rapid localized heating via dipolar polarization and ionic conduction. This uniform thermal energy transfer shortens reaction times from hours down to just minutes or seconds while minimizing side-product formation to deliver substantially higher yields and cleaner reaction profiles. However, it requires specialized, high-pressure microwave reactors to prevent thermal degradation, and scaling up the reaction presents engineering challenges due to the limited penetration depth of microwave radiation.
2. Solvent-free synthesis. This embodies green chemistry principles by eliminating volatile organic solvents entirely, reacting the aldehydes, ketones, and solid catalysts directly through mechanochemical grinding in a mortar and pestle or ball mill, or via mild heating of neat reagents. This approach offers outstanding atom economy, drastically reduces environmental waste, and simplifies work-up protocols, often requiring only a quick water or ethanol wash. Its primary challenges include handling viscous or solid masses that can hinder mass transfer and proper mixing, as well as managing localized heat buildup during exothermic reactions without a solvent heatsink.

2.4 Biological activities of Curcumin and Its Analogues.

a. Antibacterial Effects

The antibacterial activity of curcumin analogues, particularly monocarbonyl derivatives and heterocycle-substituted variants, relies on multi-target cellular disruption. A primary target is the bacterial cell division protein FtsZ (Filamenting temperature-sensitive mutant Z); analogues bind to FtsZ, inhibiting its GTPase activity and preventing the assembly necessary for bacterial cytokinesis. (Oglah, 2020) Additionally, these compounds directly interact with lipid bilayers to destabilize bacterial cell membranes, causing loss of membrane potential, cellular leakage, and rapid cell lysis across both Gram-positive and Gram-negative species. Many analogues also impair quorum sensing communication, thereby inhibiting biofilm formation and virulence factor expression. Some heterocycle-bearing analogues can even serve as photosensitizers in photodynamic therapy, generating localized reactive oxygen species upon light exposure to eradicate resistant pathogens. (Marcin Górski¹, 2022)

Curcumin has been reported to exhibit antibacterial activity against a wide range of microorganisms. Its mechanism of action is believed to involve disruption of bacterial cell membranes, inhibition of protein synthesis, and interference with nucleic acid function. (Oglah, 2020) (Marcin Górski¹, 2022) However, it is noted that the effectiveness is limited by poor solubility and instability. Synthetic curcumin analogues have demonstrated enhanced antibacterial activity compared to natural curcumin. Monocarbonyl analogues, in particular, show increased potency due to their improved stability and electrophilic nature, which allows them to interact more effectively with bacterial enzymes. (Polaquini, 2021) Chalcone derivatives have also shown significant antibacterial activity, often attributed to their ability to inhibit bacterial enzymes and disrupt metabolic pathways. Piperidone-based analogues exhibit strong activity due to their rigid structure and enhanced binding interactions. Heterocyclic analogues, such as pyrazoles, have been found to possess broad-spectrum antimicrobial activity. The presence of nitrogen atoms in these structures enhances their ability to interact with biological targets. (Chongshan Dai ¹, 2022)

b. Antioxidant Effects

While natural curcumin undergoes rapid hydrolytic degradation due to its central beta-diketone linker, synthetic analogues replace or modify this core while optimizing electron-donating groups, such as phenolic hydroxyl and methoxy substituents. (Oglah, 2020) These structural refinements allow analogues to act as direct scavengers of free radicals including superoxide, hydroxyl, and peroxy species by donating hydrogen atoms or electrons from their conjugated aromatic systems. On a molecular level, monocarbonyl analogues potentially stimulate the Nrf2/ARE (Nuclear Factor Erythroid 2–Related Factor 2) pathway, triggering the nuclear translocation of Nrf2 and boosting the expression of endogenous protective enzymes such as superoxide dismutase, catalase, and glutathione peroxidase. (Oglah, 2020) Furthermore, these analogues minimize lipid peroxidation within cell membranes and chelate redox-active metal ions, effectively suppressing Fenton reactions that generate tissue-damaging ROS.

c. Anticancer Effects

The anticancer potency of curcumin analogues is markedly higher than that of natural curcumin due to improved cellular uptake and extended systemic half-lives. Analogues execute tumor cell clearance primarily by triggering apoptosis through both mitochondrial (intrinsic) and death-receptor (extrinsic) pathways; they upregulate pro-apoptotic proteins like Bax and Bad, downregulate anti-apoptotic proteins like Bcl-2, and induce caspase-3/9 cleavage. Concurrently, they disrupt key oncogenic survival pathways by inhibiting I kappa B kinase (IKK) to prevent NF- kappa B activation, blocking STAT3 phosphorylation, and suppressing the PI3K/Akt/mTOR pathway. At the cell-cycle level, analogues downregulate cyclin B1 and surviving to induce phase cell-cycle arrest. Finally, they restrict tumor progression and metastasis by downregulating matrix metalloproteinases (MMP-2 and MMP-9) and suppressing vascular endothelial growth factor (VEGF) expression to block angiogenesis. (Oglah, 2020)

CHAPTER THREE: METHODOLOGY

3.1 Research design

This study employed an experimental laboratory-based design to synthesize curcumin analogues and evaluate their antimicrobial activity against E-coli.

3.2 Study area

The research was conducted in the Chemistry Laboratory of Busitema University, which is equipped for chemical synthesis and compound characterization.

3.3 Materials and reagents

3.3.1 Chemicals for synthesis

- Acetone (97%)
- Sodium hydroxide (50%)
- Benzaldehyde
- Ethanol (solvent, 97%)
- Hydrochloric acid (1M) □ Distilled water

3.3.2 Microbiological testing materials

- Sterile petri dishes, pipettes and culture tubes
- Standard strain of Escherichia coli
- Nutrient agar and broth
- Mueller-Hinton agar

3.3.3 Equipments

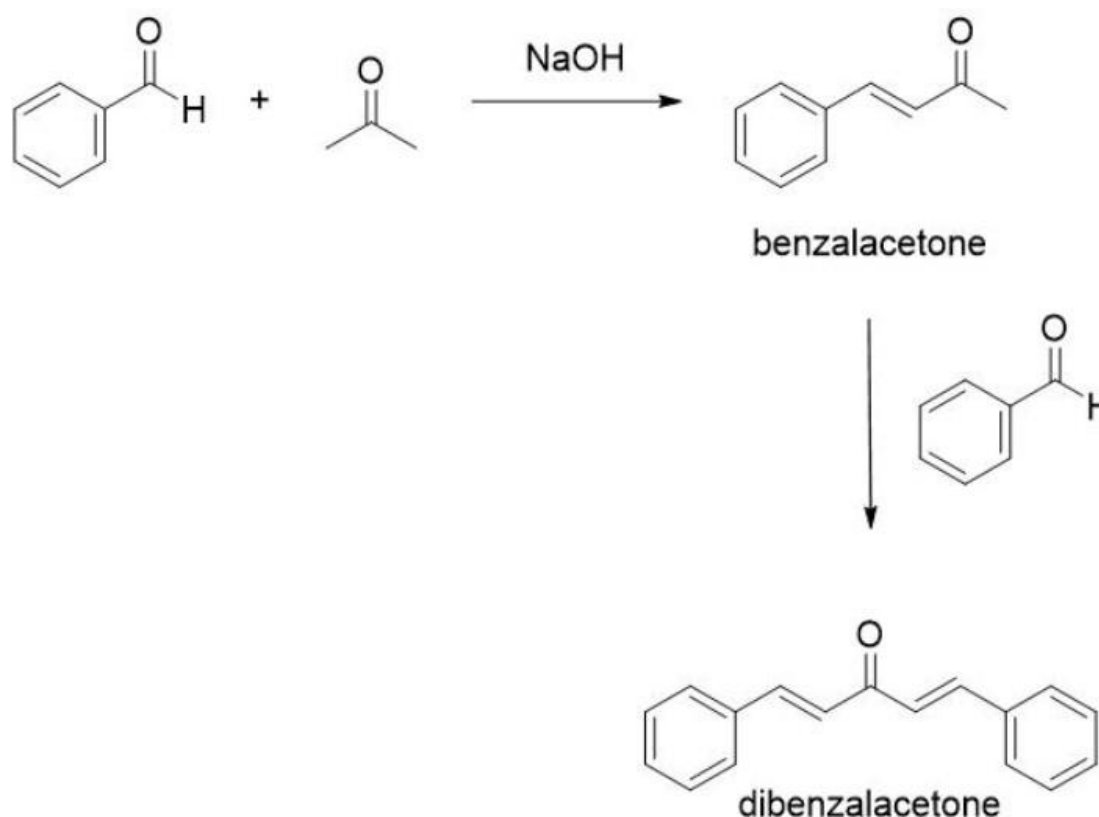
- Rotary evaporator
- UV-Vis spectrometer
- Incubator (37⁰C)
- FTIR spectrometer
- Round-bottom flask
- Heat source
- Stirrer
- Melting point apparatus
- Filter paper

3.4 Chemical synthesis of curcumin analogues.

3.4.1 Synthesis of Monocarbonyl Analogues.

100.0 mmol of benzaldehyde was dissolved in 20mls of ethanol in a round bottomed flask. 50mmol of acetone was added to the solution with continuous stirring. 5mls of dilute sodium hydroxide solution (50%) was then added dropwise while stirring continuously. The reaction mixture was cooled in an ice bath to control the rate of reaction and minimize side reactions. After complete addition of sodium hydroxide solution, the resulting mixture was left to proceed for 5 hours at room temperature. During this time, a yellow precipitate gradually formed, indicating product formation. The reaction mixture was then poured into cold distilled water to enhance precipitation. The solid product was collected by vacuum filtration, washed with cold water to remove residual base and impurities, and dried. The crude product was purified by recrystallization using ethanol to obtain pure monocarbonyl curcumin analogue. (Polaquini, 2021)

FIGURE 3.1 Equation of reaction leading to the formation of monocarbonyl analogue.



Calculation for percentage yield

Mole ratio of dibenzalacetone: acetone=1:1

Moles of acetone= 0.05

Moles of dibenzalacetone=1/1*0.05= 0.05

Molar mass of dibenzalacetone= 234.3g

Theoretical yield of dibenzalacetone= 0.05* 234.3=11.715

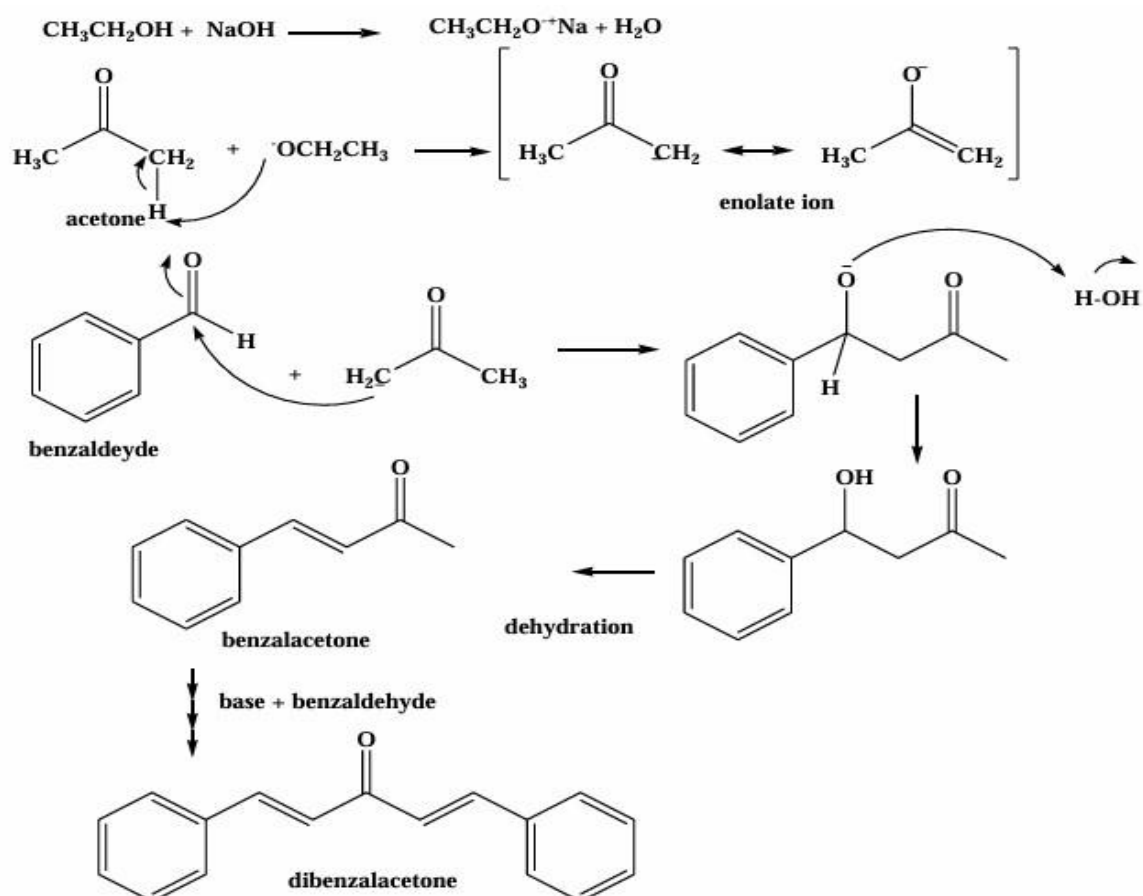
Actual yield= 10.5g

Percentage yield= actual yield/ theoretical yield * 100

$$=(10.5 \div 11.715) * 100 = 89.6\%$$

Therefore the percentage yield=89.6%

FIGURE 3.2 Mechanism for the reaction



3.5 Characterization of monocarbonyl analogue of curcumin.

3.5.1 Melting point determination

This method was used to test the purity and identity of the compound synthesized

A capillary tube was filled with a small amount of monocarbonyl analogue and placed in a melting point apparatus. The sample was monitored until complete melting. The temperature at which melting occurred was recorded.

UV -Visible spectroscopy

This method was used to determine the absorbance of monocarbonyl analogue.

Procedure

5mg of solid monocarbonyl analogue were weighed and transferred into a clean dry beaker

50ml of 98% ethanol were added to the beaker containing the solid monocarbonyl and the mixture shaken until the solid dissolved completely.

Two cuvettes, one was filled with sample solution and the other was filled with pure ethanol. The cuvette containing ethanol was used to run the blank scan the other cuvette containing the sample solution was used to run the scan at 330 nm.

The UV-Vis reading was taken and absorbance of the sample was recorded

3.5 Antibacterial Testing

Antibacterial activity will be evaluated using the agar well diffusion method.

1. 28 g of Mueller-Hinton agar (MHA) was dissolved in distilled 1000cm³ water and stirred. The resultant mixture was then heated to enable complete dissolution.
2. The resultant mixture was then sterilized at 121 °C and pressure 15 pi for 15 minutes.
3. The agar was then cooled, poured on sterilized petri dishes and allowed to solidify in a refrigerator.
4. . Wells are created in the agar using a sterile cork borer. Solutions of synthesized compounds are introduced into the wells.
5. Antibacterial activity was observed based on inhibition of bacterial growth around the disks.
6. The plates are incubated at 37°C for 24 hours. Zones of inhibition are measured in millimeters, and the results are recorded and compared. Larger zones indicate stronger antibacterial activity. (Dalia R. Emam 1, 2017)

CHAPTER FOUR: RESULTS AND DISCUSSIONS

4.1 Nature of monorcarbonyl curcumin analogue

The synthesized monorcarbonyl curcumin analogue appeared as yellow-orange crystals after recrystallization. The percentage yield obtained was satisfactory, indicating that the synthesis reaction proceeded efficiently. The yellow coloration suggested successful formation of a conjugated aromatic compound similar to curcumin derivatives reported in previous studies.



4.2 Melting point

The melting point of the purified compound was **119⁰C**, there was a very slight difference from the theoretical melting point which is between 112-114⁰C indicating high purity. Impure compounds generally show broad melting ranges because contaminants interfere with crystal lattice formation. The sharp melting point observed suggested effective purification through recrystallization.

Physical properties.

Property	Result
Yield(%)	89.6%
Colour	Pale yellow
Crystal form	Monoclinic
Melting point	119 ⁰ C

4.3 UV-Vis analysis

UV-visible spectroscopy showed strong absorption maxima within the visible region. At wavelength 330nm, dibenzalacetone showed an absorbance of **0.485**. This result is characteristic of compounds possessing extended conjugated π -electron systems. The absorption peak observed confirmed the presence of chromophoric groups responsible for the compound's colour and biological activity.

UV table

λ_{max}	Assignment
330nm	π - π^* transition of the conjugated system

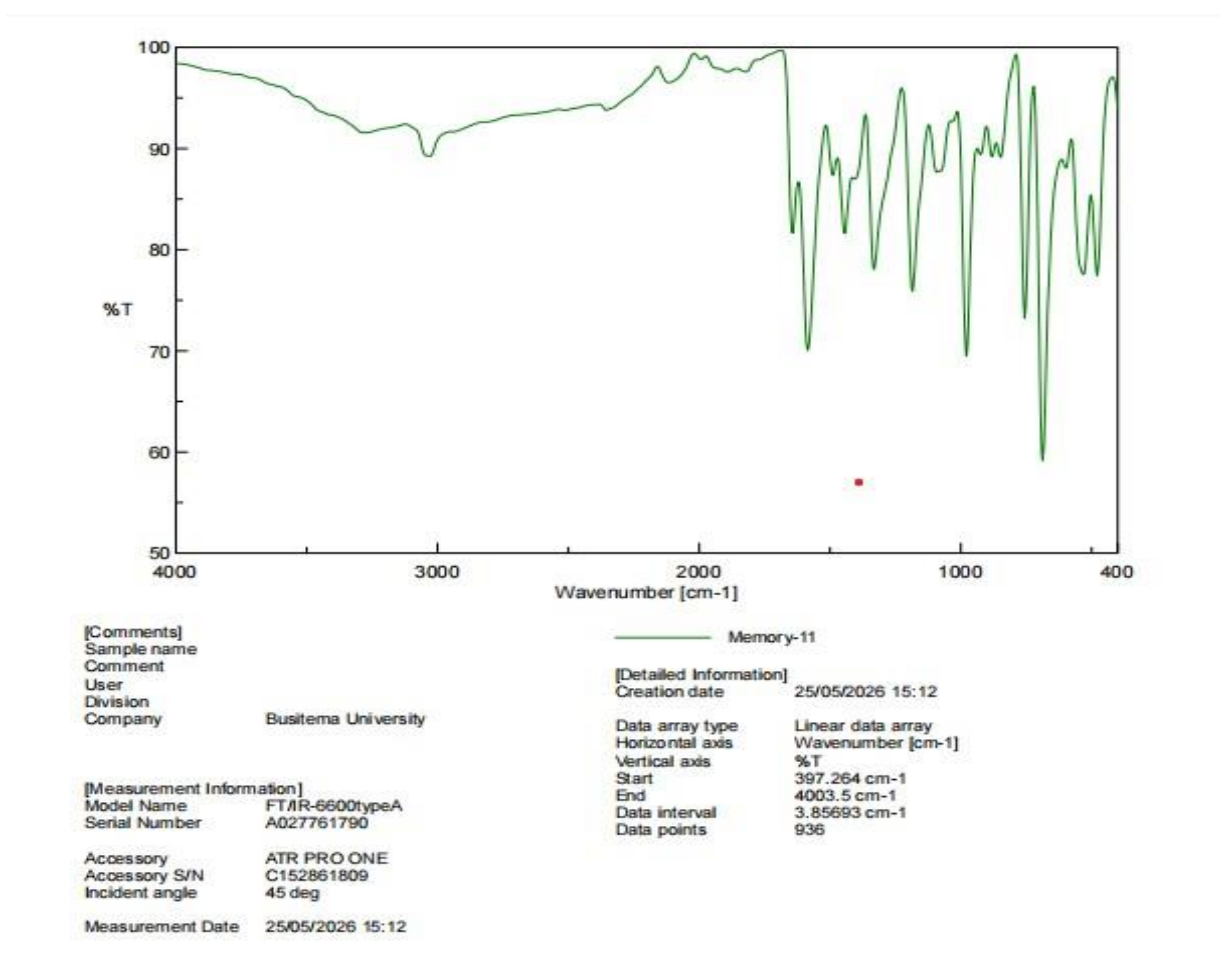
4.4 FTIR spectroscopy analysis.

FTIR spectroscopy confirmed the presence of key functional groups. Broad peaks corresponding to hydroxyl groups indicated phenolic OH functionalities. Strong carbonyl peaks confirmed the monocarbonyl structure. Aromatic C=C stretching vibrations demonstrated the aromatic nature of the compound while alkene peaks confirmed unsaturation within the molecular framework

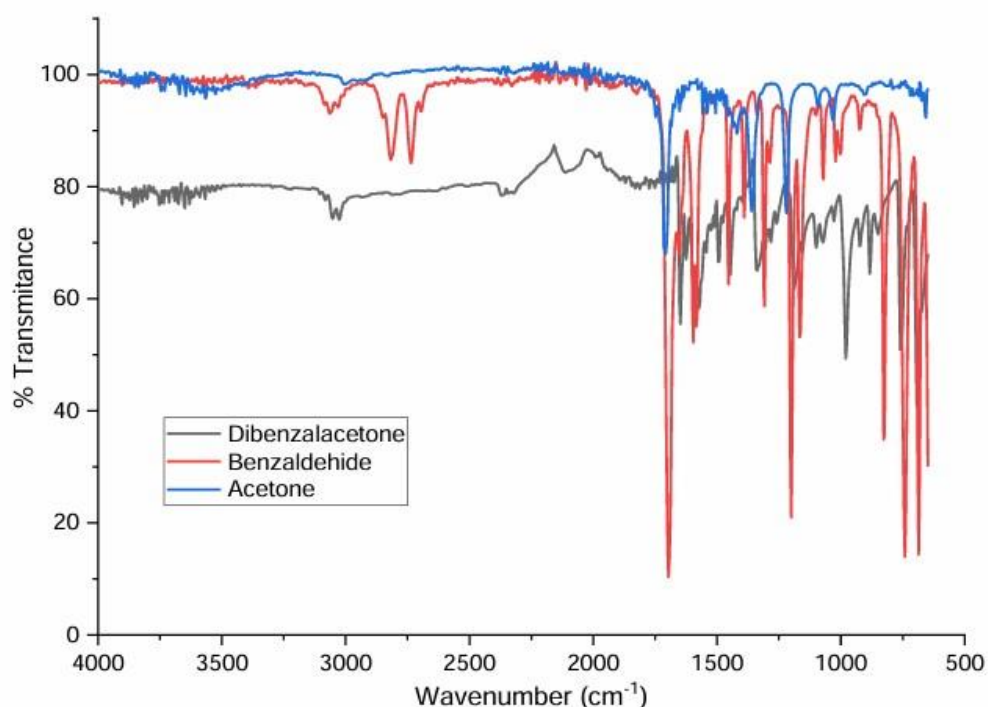
FTIR table

EXPECTED FUNCTIONAL GROUPS	EXPECTED WAVENUMBER
Alkene C=C	1000-1500cm ⁻¹
Carbonyl C=O	1700cm ⁻¹
Aromatic C=C	1500-1600cm ⁻¹
Aromatic C-H	2500-3200cm ⁻¹

FTIR spectrum of dibenzalacetone.



Overlay FTIR spectrum of benzaldehyde, acetone, and dibenzalacetone.



Based on the overlay spectrum between the synthesis product and its precursors, it can be seen that the synthesis results show a slightly different spectrum profile from the precursors. The synthesized compound had no aldehyde C-H group at 2817 and 2735 cm⁻¹. The C=O group in the synthesized compound is also slightly different in absorption compared to its constituent acetone. Because the synthesized compound is influenced by the group attached to the side chain of the C=O, there is a slight shift in the wavenumber. In addition, the resonance effect with the C=C bond causes the delocalization of pi electrons in both groups, so it will decrease the double bond character of C=O, which causes a decrease in absorption.

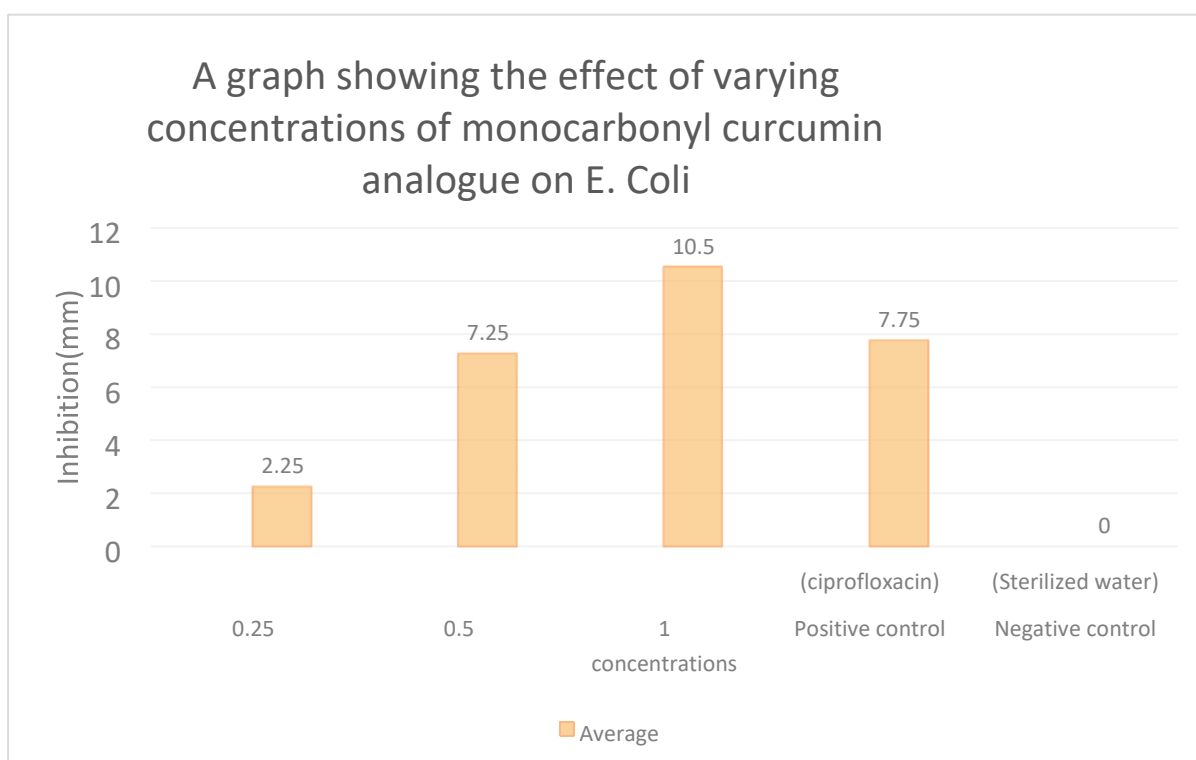
4.5 Bacterial activity

The antimicrobial assay showed that the synthesized analogue inhibited growth of *Escherichia coli*. The inhibition depended on the concentrations of curcumin analogue. Larger zones of inhibition were observed at higher concentrations. The antimicrobial effect was attributed to disruption of bacterial cell membranes, inhibition of protein synthesis, and interference with metabolic pathways.

The antibacterial activity of dibenzalacetone was assessed by measuring the diameter of the inhibition zone, which represents the largest clear area formed around the well, indicative of bacterial growth

inhibition. In this experiment, dibenzalacetone was tested against *Escherichia coli* (*E. coli*) at various concentrations: 1mg/L, 0.5mg/L, and 0.25mg/L, with ciprofloxacin at a concentration of 50 mg/L used as a positive control and distilled water as a negative control experiment.

Concentration(mg/L)	Inhibition zone (mm)				
	0.25	0.5	1	Positive control (ciprofloxacin)	Negative control (Sterilized water)
1	3.0	7.4	11.0	8.0	0.0
2	2.5	7.65	10.5	7.55	0.0
3	1.25	6.7	10.0	7.7	0.0
Average	2.25	7.25	10.5	7.75	0.0



At Cconcentration of 0.5mg/L, the compound's ability was comparable to the antibiotic ciprofloxacin. The cell wall that is most easily denatured is a cell wall composed of polysaccharides compared to a cell wall composed of phospholipids. Gram-negative bacteria have three layers: lipopolysaccharide, protein, and hydrophobic phospholipids. Dibenzalacetone compound has hydrophobic properties based on its solubility, so it will easily penetrate bacterial cell walls due to the same solubility properties bactericidal.

The minimum inhibitory concentration results showed that the analogue was effective even at relatively low concentrations. This suggests strong antibacterial potential and indicates that monocarbonyl modification improved biological activity compared to natural curcumin. The enhanced activity observed may be explained by improved stability of the monocarbonyl analogue. Removal of the β -diketone moiety reduces degradation and increases persistence under physiological conditions. Increased hydrophobicity may also facilitate penetration through Gram-negative bacterial membranes.

The findings agree with previous studies that reported improved antimicrobial activity among synthetic curcumin analogues. The analogue synthesized in this study demonstrated significant promise as an antimicrobial agent against *Escherichia coli*.

Another important observation was the relationship between structure and activity. The conjugated double bonds and aromatic substituents appear essential for biological activity. Hydroxyl groups may contribute to hydrogen bonding interactions with bacterial targets. Carbonyl groups also play important roles in electron delocalization and reactivity.

The results collectively demonstrate that monocarbonyl curcumin analogues possess improved physicochemical and antimicrobial properties compared to natural curcumin. These compounds may serve as templates for development of new antimicrobial drugs especially in the era of antibiotic resistance.

CHAPTER FIVE: CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

This study successfully achieved the synthesis of a monocarbonyl curcumin analogue through Claisen-Schmidt condensation reaction. The synthesized compound was purified by recrystallization and characterized using UV-visible spectroscopy, FTIR spectroscopy, and melting point determination. The characterization results confirmed successful synthesis of the desired analogue and indicated high purity of the final product.

The antimicrobial studies demonstrated that the synthesized monocarbonyl curcumin analogue possessed inhibitory activity against *Escherichia coli*. The activity increased with concentration, indicating dose-dependent antibacterial effects. The analogue exhibited improved stability and biological activity compared to natural curcumin due to structural modification.

The study confirms that monocarbonyl curcumin analogues are promising candidates for development of new antimicrobial agents. The findings contribute to ongoing research aimed at combating antimicrobial resistance and improving therapeutic efficacy of curcumin-derived compounds.

5.2 Recommendations

1. Further studies should be conducted using additional characterization techniques such as Nuclear Magnetic Resonance spectroscopy and Mass Spectrometry.
2. More bacterial strains including Gram-positive and multidrug resistant organisms should be investigated.
3. Toxicological studies should be conducted to assess safety of the synthesized compounds.
4. Structural modification of curcumin analogues should continue to improve potency and pharmacokinetic properties.
5. In vivo studies should be carried out to evaluate therapeutic potential under physiological conditions.
6. Combination studies involving curcumin analogues and conventional antibiotics should be explored to assess synergistic effects.

7. Pharmaceutical formulations should be developed to improve solubility and delivery of curcumin analogues.

REFERENCES

- BenchChem. (2025). Application Notes and Protocols for the One-Pot synthesis of chalcones. *BENCH CHEM*, 2-4.
- Chongshan Dai 1, 2. ,. (2022). The Natural Product Curcumin as an Antibacterial Agent:. *MDPI*, 2-5.
- Dalia R. Emam 1, *. A. (2017). Synthesis and Evaluation of Curcuminoid Analogues as antioxidant and antibacterial agents. *molecules*, 5.
- Kim, B. R. (2018). Design, synthesis and evaluation of curcumin analogues. *DOAJ*, 5-8.
- Marcin Górski1, B.-D. ,.-B.-E.-F. (2022). Antibacterial activity of curcumin. *DOAJ*, 5-6.
- Oglah, M. K. (2020). *Curcumin Analogues: Synthesis and Biological Activities*. ministry of higher education and scientific research.
- Polaquini, C. R. (2021). Antibacterial activity of a new monocarbonyl analog of curcumin. *Science direct*, 2-5.
- Syeda Nazish Sohaib. (2025). Evaluation of synergistic antibacterial activity and cytotoxicity of [W4KR5]-. *Chemical biology letters*, 1-3.
- Upadhyaya, S. S. (2021). Antibacterial activity of curcumin and its essential. *wiley*, 1-2.