

Revolutionizing Wound Management

Revolutionizing Wound Management:

*Formulating Nanoparticles
with Tobacco Stem Bioactive
Compounds*

By

Kumud Bala and Yash Sharma

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TABLE OF CONTENTS

List of Tables	viii
List of Figures	ix
About the Authors	xiv
Preface	xvi
Acknowledgments	xviii
Abbreviation	xx
Abstract	xxii
CHAPTER 1: INTRODUCTIONS	1
CHAPTER 2: REVIEW OF LITERATURE	3
1. Medicinal Plants	3
2. <i>Nicotiana tabacum</i>	4
3. Role of Secondary Metabolite in Wound Healing	6
4. Flavonoids as a wound healer	7
5. Polyphenols as a wound healing agent	8
6. Tannin as Wound Healer	9
7. Antioxidants	10
8. Antioxidant Assays	12
9. Green Synthesis of Nanoparticles	14
10. Wound Healing	15
11. Angiogenesis	17
12. Mechanism of Angiogenesis	18
13. Molecular Mechanism	20
14. Different Models for Wound Healing	21

15. UV-Vis Spectrophotometer	33
16. Dynamic Light Scattering	37
17. Atomic Force Microscope	40
18. Scanning Electron Microscope (SEM)	42
19. Chromatographic Analysis	46
CHAPTER 3: MATERIAL AND METHODS	59
1. Source of Plant Collection and Authentication	59
2. Bacterial cells	59
3. Mouse fibroblast cells (L929)	59
4. Fertilized EGG	59
5. Preparation of Alcoholic extract	60
6. Isolation and Purification of Protein	60
7. Quantification of Protein	60
8. Preliminary Phytochemical Screening	60
9. Quantification of Phytochemicals	60
10. Chromatography techniques	61
11. Green Synthesis of Nanoparticle	62
12. Characterization of Silver Nanoparticles	63
13. Antimicrobial activity	63
14. Mode of Action of Nanoparticles on Bacterial Cells	64
15. Antioxidant activity	64
16. <i>In vitro</i> Studies on Cell Lines	67
17. Angiogenic activity	68
18. Gel Formulation	68
19. Animal Studies	69
20. Statistical analysis	71

CHAPTER 4: RESULTS	72
A. Quantification and Characterization of Bioactive Compounds	72
B. Antimicrobial activity of Extracts	78
C. Antioxidant Analysis of Extract	78
D. Isolation and Characterization of Proteins	78
E. Antimicrobial and Antioxidant Activity of Protein Mixture	80
F. Synthesis of Silver Nanoparticles from Bioactive Compounds Extracts and Its Fraction	80
G. Antioxidant activity	91
H. Protein Nanoconjugate	92
I. Antimicrobial activity of Protein and Protein Nanoconjugate	94
J. Antioxidant activity	100
K. <i>In vitro</i> Studies on Cell Lines	100
L. Angiogenic activity	108
M. <i>In vivo</i> studies on Wistar Rat Models	110
CHAPTER 5: DISCUSSION	121
CHAPTER 6: CONCLUSIONS	126
REFERENCES	127
LIST OF CREDENTIALS	149
LIST OF CONFERENCES	150

LIST OF TABLES

<i>Tables</i>	<i>Title</i>	<i>Page No.</i>
Table 1	Phytochemical screening of ethanolic extract of stem of <i>N. tabacum</i>	73
Table 2	Minimum inhibitory concentration of ethanolic extract of defatted stem of <i>Nicotiana tabacum</i> against bacterial cells.	80
Table 3	Antimicrobial activity of Protein Nanoconjugate against bacterial cells.	95
Table 4	Antimicrobial activity of Silver as control against bacterial cells	95
Table 5	Physical properties of formulated gel from nanoparticles of stem of <i>N. tabacum</i> .	113

LIST OF FIGURES

Figure	Title	Page No.
Fig. 1	HPLC Flow Diagram indicating the components and working.	50
Fig. 2	HPLC Separation	51
Fig. 3	Chromatogram and Retention Time.	53
Fig. 4	A Stream Lined diagram of a gas chromatograph-mass spectrometer displaying the following components: (1) carrier gas (2) autosampler (3) inlet (4) analytical column (5) interface (6) vacuum (7) ion source (8) mass analyzer (7) ion detector (8) PC (10).	56
Fig. 5	Thin Layer chromatography of (Q) standard quercetin 1mg/ml, (E) ethanolic extract and (M) methanolic extract of stem of <i>Nicotiana tabacum</i> .	74
Fig. 6	Thin layer chromatogram of (Q) quercetin, (T) tannic acid, (P) piperine and ethanolic extract of stem of <i>N. tabacum</i> (TSE).	75
Fig. 7	HPLC Chromatogram of standard quercetin, tannic acid, piperine and ethanolic extract of stem of <i>N. tabacum</i> (TSE).	76
Fig. 8	HPLC Chromatogram of standard Quercetin and B fraction of ethanolic extract of defatted stem of <i>Nicotiana tabacum</i>	77
Fig. 9	Antimicrobial activity of ethanolic and methanolic extract of defatted stem of <i>Nicotiana tabacum</i> against bacterial cells.	79
Fig. 10	UV –Spectra Scan of synthesized silver nanoparticle from ethanolic extract of stem of <i>Nicotiana tabacum</i>	81
Fig. 11	UV –Spectra Scan of synthesized silver nanoparticle from B fraction of ethanolic extract of stem of <i>Nicotiana tabacum</i>	82
Fig. 12	DLS of synthesized silver nanoparticle from ethanolic extract of stem of <i>Nicotiana tabacum</i> .	82

Fig. 13	DLS of synthesized silver nanoparticle from B fraction of ethanolic extract of stem of <i>Nicotiana tabacum</i> .	83
Fig. 14	AFM results of Silver nanoparticles synthesized from the ethanolic extract of stem of <i>Nicotiana tabacum</i> .	83
Fig. 15	AFM results of Silver nanoparticles synthesized from the B Fraction of ethanolic extract of stem of <i>Nicotiana tabacum</i>	84
Fig. 16	SEM of synthesized silver nanoparticle from ethanolic extract of stem of <i>Nicotiana tabacum</i>	85
Fig. 17	SEM of synthesized silver nanoparticle from B fraction of ethanolic extract of stem of <i>Nicotiana tabacum</i>	86
Fig. 18	Antimicrobial activity of Silver Nanoparticles synthesized from B fractions of ethanolic extract of stem of <i>Nicotiana tabacum</i> .by Broth Dilution Method.	87
Fig. 19	Protein leakage assay in treated bacterial cell with synthesize silver nanoparticles from ethanolic extract of stem of <i>Nicotiana tabacum</i> .	88
Fig. 20	Nucleic acid leakage assay of Synthesized silver nanoparticles from ethanolic extract of stem of <i>Nicotiana tabacum</i> .	89
Fig. 21	Antimicrobial activity of Silver Nanoparticles synthesized from protein of stem of <i>Nicotiana tabacum</i> .by Broth Dilution Method.	90
Fig. 22	Protein leakage assay in treated bacterial cell with synthesized silver nanoparticles from B fraction of ethanolic extract of stem of <i>Nicotiana tabacum</i>	90
Fig. 23	Nucleic acid leakage assay in treated bacterial cell with synthesize silver nanoparticles from B fraction of ethanolic extract of stem of <i>Nicotiana tabacum</i> .	91
Fig. 24	UV –Spectra of synthesized silver nanoparticles from protein of stem of <i>Nicotiana tabacum</i> .	92
Fig. 25	DLS of synthesized silver nanoparticle from protein of stem of <i>Nicotiana tabacum</i> .	93
Fig. 26	AFM results of Silver nanoparticles synthesized from protein of stem of <i>Nicotiana tabacum</i>	93

Fig. 27	SEM of synthesized silver nanoparticle from protein of stem of <i>Nicotiana tabacum</i>	94
Fig. 28	Antimicrobial activity of silver as control and protein nanoconjugate against bacterial cells.	96
Fig. 29	Antibacterial activity of Silver control(Ag), Protein Nanoconjugate (Ag P) and Precipitate protein (P) against <i>Acinetobacter baumannii</i> (A), <i>Pseudomonas aeruginosa</i> (B), <i>Bacillus mycoides</i> (C) and <i>Staphylococcus aureus</i> (D).	97
Fig. 30	Antimicrobial activity of Silver Nanoparticles synthesized from protein of stem of <i>Nicotiana tabacum</i> by Broth Dilution Method.	98
Fig. 31	Protein leakage assay in treated bacterial cell with synthesize silver nanoparticles from Protein of stem of <i>Nicotiana tabacum</i> .	99
Fig. 32	Nucleic acid leakage assay in treated bacterial cell with synthesize silver nanoparticles from Protein of stem of <i>Nicotiana tabacum</i> .	100
Fig. 33	Representative pictures illustrate the proliferative activity of ethanolic, B fraction, Ethanolic extract Ag-NPs, and B Fraction Ag-NPs on DF-1 cells	102
Fig. 34	The effect of TSE on DF-1 cell line viability was determined by MTT assay method. Each bar graph represents % viability of DF-1cells against 15 to 480 µg/mL concentrations	103
Fig. 35	The effect of B Fraction on DF-1 cell line viability was determined by MTT assay method. Each bar graph represents the % viability of DF-1cells against 15 to 480 µg/mL concentrations.	104
Fig. 36	The effect of Ethanolic extract Ag-NPs on DF-1 cell line viability was determined by the MTT assay method. Each bar graph represents the % viability of DF-1cells against 15 to 480 µg/mL concentrations.	105
Fig. 37	The effect of B-Fraction Ag-NPs on DF-1 cell line viability was determined by the MTT assay method. Each bar graph represents the % viability of DF-1cells against 15 to 480 µg/mL concentrations.	106
Fig. 38	Representative pictures illustrate the neuroprotective efficacy of ethanolic, B fraction, Ethanolic extract Ag-NPs & B Fraction Ag-NPs on Rat PC-12 cells.	107

Fig. 39	Antioxidant activity of ethanolic, B fraction, Ethanolic extract Ag-NPs & B Fraction Ag-NPs on Rat PC-12 cells against Neurotoxic shock.	108
Fig. 40	Representative pictures illustrate the angiogenic activity of extracts and Ag-NPs on chick embryo chorioallantoic membrane (CAM). The pictures show the angiogenesis in the presence of 50 µl/egg concentration of extracts and Ag-NPs in which: (A) Control, (B) ethanolic extract, (C) B Fraction, (D) Ethanolic extract Ag-NPs and (E) B Fraction Ag-NPs.	109
Fig. 41	The graph depicts the wound healing effect on excision wounds in Wistar rats with the percentage of wound contraction per day. Data is presented as the mean standard error of the mean ($n = 3$). Statistical significance was assessed using a student t-test as compared to disease control. * $p < 0.05$, ** $p < 0.005$, *** $p < 0.00005$	111
Fig. 42	Pictorial representation of wound healing contraction of Ethanolic Extract (Treatment 1), B Fraction (Treatment 2), Ethanolic Extract-AgNPs (Treatment 3), B Fraction (Treatment 4) and Soframycin (Standard Drug).	112
Fig. 43	The graph depicts the wound healing effect of the formulated gel on excision wounds in Wistar rats with the percentage of wound contraction per day. Data is presented as the mean standard error of the mean ($n = 3$). Statistical significance was assessed using a student t-test as compared to disease control. * $p < 0.05$, ** $p < 0.005$, *** $p < 0.00005$	114
Fig. 44	Pictorial representation of wound healing contraction of formulated Gel.	115
Fig. 45	Histology studies of wound healing efficacies of extracts, AgNPs, and formulated gel showing the epithelialization, granulation tissue, and blood vessels.	117

Fig. 46	<i>In vivo</i> antioxidant activity of extracts and nanoextracts on excision wound in Wistar rat model. Data is presented as the mean standard error of the mean ($n = 3$). Statistical significance was assessed using a student t-test as compared to disease control and standard drugs. * $p < 0.05$, ** $p < 0.005$, *** $p < 0.00005$	119
Fig. 47	<i>In vivo</i> hydroxyproline content of extracts and nanoextracts on excision wound in Wistar rat model. Data is presented as the mean standard error of the mean ($n = 3$). Statistical significance was assessed using a student t-test as compared to disease control and standard drugs. * $p < 0.05$, ** $p < 0.005$, *** $p < 0.00005$	120

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PREFACE

In recent years, the exploration of natural bioactive compounds for therapeutic purposes has gained momentum. The potential of bioactive compounds derived from plant sources for promoting wound healing has emerged as a promising area of research. This project, entitled "Formulation of Tobacco Stem Bioactive Compound Loaded Nanoparticle for Wound Healing," represents a step forward in this exciting field, combining traditional knowledge with cutting-edge nanotechnology to develop innovative healthcare solutions.

The journey of this study is both challenging and rewarding. It began with the identification and extraction of bioactive compounds from tobacco stems, a process requiring meticulous attention and a deep understanding of plant biochemistry. Following this, we embarked on the complex task of formulating these compounds into nanoparticles by leveraging advanced nanotechnology techniques to enhance their stability, bioavailability, and therapeutic efficacy.

Our research was driven by the recognition of a significant clinical need for the development of more effective and efficient wound-healing treatments. Chronic wounds, including those associated with diabetes and other underlying conditions, pose serious health challenges, often leading to prolonged patient suffering and increased healthcare costs. By focusing on the bioactive compounds from tobacco stems, we aimed to harness their natural healing properties in a novel nanoparticle-based formulation.

This project would not have been possible without the support and collaboration of many individuals and institutions. Our heartfelt gratitude goes to the Department of Science and Technology, Science and Engineering Research Board (DST SERB), whose generous funding made this study feasible. We also acknowledge the significant contributions of our research team, including Ph.D. scholars and M.Tech and B.Tech trainees, whose dedication and hard work have been instrumental in advancing this project.

In addition, the expertise and guidance provided by Dr. Sunita Garg from CSIR-NISCAIR, support from the Amity Institute of Pharmacy, infrastructure provided by the Amity Institute of Biotechnology, and visionary leadership of Dr. Ashok K. Chauhan, Founder President of Amity University, have been crucial to our progress. Collaboration with the National Centre for Cell Sciences (NCCS) in Pune for providing cell lines and analytical support from the Advance Research Analytical Service in Ghaziabad has further strengthened our research.

This preface is a testament to the collaborative spirit underpinning scientific research. This reflects the collective efforts of numerous individuals and institutions working towards a common goal. As we presented the findings of our study, we do so with a deep sense of gratitude and acknowledgment of the contributions that have made this work possible.

We hope that the insights gained from this research will pave the way for future studies and ultimately lead to the development of new and effective treatments for wound healing. Our journey, marked by perseverance and innovation, serves as an example of what can be achieved when diverse expertise and resources come together in the pursuit of scientific advancement.

Thank you.

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"Gratitude is the fairest blossom which springs from the soul."

—Henry Ward Beecher

We deeply appreciate the invaluable support provided by various individuals and institutions throughout this research project. First and foremost, we extend our sincere gratitude to the Department of Science and Technology, Science and Engineering Research Board (DST SERB), for granting the project entitled "Formulation of Tobacco Stem Bioactive Compound Loaded Nanoparticle for Wound Healing." Their generous funding was pivotal in driving this research.

We would like to express our heartfelt thanks to the dedicated Ph.D. scholars in our lab, Mr. Yash Sharma, Mr. Manish Kumar, and Ms. Mohini. Their relentless efforts and commitment to mastering the intricate techniques required for this project are commendable. Their perseverance and enthusiasm have greatly contributed to the successful progress of this research. Additionally, we extend our appreciation to M. Tech and B. Tech trainees who have actively participated and contributed to various aspects of this project.

Special acknowledgment goes to Dr. Sunita Garg, Emeritus Scientist at CSIR-NISCAIR, Raw Material Herbarium and Museum, Delhi, for her expertise in identifying and authenticating our research materials. Her guidance and support were instrumental in ensuring the scientific integrity of our project. The reference number NISCAIR/RHMD/Consult/2020/3697-98-3 stands a testament to her invaluable contribution.

We are deeply grateful to the Amity Institute of Pharmacy for providing access to the animal house, which was essential for the in vivo experiments conducted as part of this research. The support and cooperation of lab attendants deserve special mention; their assistance has been crucial in maintaining the smooth operation of our research activities.

The infrastructure and facilities provided by the Amity Institute of Biotechnology were fundamental to the success of this project. We are profoundly thankful to Dr. Ashok K. Chauhan, Founder and President of Amity University, for his visionary leadership and unwavering support in establishing our laboratory. His commitment to fostering a robust research environment is a significant enabler of our work.

Furthermore, we acknowledge the National Center for Cell Sciences (NCCS), Pune, India, for providing the cell lines necessary for our cellular studies. Their contribution is vital in advancing our understanding of the cellular mechanisms involved in wound healing. We also extend our gratitude to the Advanced Research Analytical Service, Ghaziabad, for their expertise in High-Performance Liquid Chromatography (HPLC) analysis, which was critical for the accurate quantification and characterization of bioactive compounds in our study.

In conclusion, this research endeavor has been a collective effort, and we are immensely grateful to everyone who has been part of this journey. Each individual's contribution has been the cornerstone of the realization of this project. The collaborative spirit and unwavering support from all involved not only facilitated the successful completion of this research, but also enriched the learning experiences of all team members. We sincerely thank all of them for their dedication, support, and encouragement.

Thank you.

ABBREVIATIONS

°C	:	Degree Celsius
µm	:	Micrometre
µL	:	Microliter
µg	:	Microgram
<i>N. tabacum</i>	:	<i>Nicotiana tabacum</i>
TLC	:	Thin Layer Chromatography
HPLC	:	High Pressure Liquid Chromatography
DPPH	:	2,2-Diphenyl-1-picrylhydrazyl
TPTZ	:	2,4,6-Tris(2-pyridyl)-s-triazine
MTT	:	3-(4,5-Dimethylthiazol-2-yl)-2,5 Diphenyltetrazolium Bromide
SOD	:	Superoxide dismutase
Cat	:	Catalase
GST	:	Glutathione S- transferase
GSH	:	Glutathione
MDA	:	Malondialdehyde
TGF-β	:	Transforming Growth Factor
Hr	:	Hour
M	:	Meter
NF-κB	:	Nuclear factor kappa B
MAPK	:	Mitogen-activated proteins
HSP90	:	Heat Shock Protein 90
Cdc37	:	Cochaperone of HSP90
PBMC	:	Primary peripheral blood mononuclear Cells
DMBA	:	2,4-Dimethoxybenzaldehyde
MNNG	:	Methylnitronitrosoguanidine
DFF45	:	DNA Fragmentation Factor 45
PARP	:	Poly (ADP-ribose) polymerase
MAP30	:	Momordica anti-HIV protein
Mg	:	Milligram
LC	:	Liquid Chromatography
GC	:	Gas Chromatography
TOF	:	Time of Flight

TOFMS	:	Time-of-flight mass spectrometry
HRMS	:	High-resolution mass spectrometry
ROS	:	Reactive Oxygen Species
HPV	:	Human Papillomavirus
BM	:	Basement Membrane
TNM	:	Classification of Malignant Tumors
ALDH	:	Aldehyde dehydrogenase
ADH	:	Alcohol dehydrogenase
HSV	:	Herpes Complex Virus
HIV	:	Human Immunodeficiency Virus
IARC	:	International Agency for Research on Cancer
CDK	:	Cyclin dependent kinase
PUFAs	:	Polyunsaturated fats
MnSOD	:	Mitochondrial manganese SOD
PEDXs	:	Peroxiredoxin
GPX	:	Glutathione Peroxidases
HO [·]	:	Hydroxyl radical
NBT	:	Nitroblue tetrazolium
ER	:	Endoplasmic reticulum
TNF	:	Tumor Necrosis factor
FADD	:	Fas associated via death domain
DISC	:	Death Inducing Signalling Complex
MPT	:	Mitochondrial Permeability Pore
Apaf-1	:	Apoptotic peptidase activating factor 1
QE	:	Quercetin Equivalent
RMSD	:	Root mean square deviation
ADME	:	Absorption, Distribution, Metabolism and Excretion
DMSO	:	Dimethyl sulfoxide
dNTP	:	Deoxynucleoside triphosphate
Et	:	Ethanol
Ac	:	Acetone
Aq	:	Aqueous
Rf	:	Retardation Factor
NIST	:	National Institute of Standards and Technology
mM	:	Millimolar

ABSTRACT

The skin is the largest organ in the surface area of the human body. It has a critical structure that shields internal tissues from mechanical damage, microbial infection, ultraviolet radiation, and extreme temperature. This makes it highly susceptible to injury and significantly impacts individual patients and the healthcare economy. Wound healing has been studied for decades, but the underlying molecular mechanism remains unclear. Present books include the nano-formulation of tobacco stems to observe wound healing in Wistar rat models. Chromatographic analysis showed that flavonoids were separated from the extracts and contained quercetin, rutin, and tannic acid. The antimicrobial and antioxidant activities of the extracts were found to be present in the extracts and fractions of *N. tabacum* stems. Extracted flavonoids from the stem have revealed angiogenic activity and wound healing efficacy against excision wounds in Wistar rat models. Flavonoids have been used for the synthesis of AgNPs using the bio-reduction method. Characterization of the synthesized silver nanoparticles, such as SEM, DLS, and AFM, revealed the formation of nanoparticles smaller than 100 nm. Antimicrobial assays using the broth dilution method and mode of action showed maximum antimicrobial activity against gram-positive and gram-negative bacteria. To observe the antioxidant capacity, electron transfer assay, enzymatic activity, and non-enzyme content have been determined and have revealed maximum capacity in the fractions of tobacco stem and AgNPs. The angiogenic activity of extracts and nanoparticles has revealed blood vessel formation in CAM models. The wound healing efficacy of Ethanolic Extract-AgNPS and B Fraction-AgNPs showed early wound contraction before 14 days in the excised wound of Wistar rat models. Histopathological studies demonstrated epithelialization, granulation tissue, and blood vessels in treated wounds. The formulated nanogels also induced early wound contraction with re-epithelialization in the treated wounds. It can be concluded from the study that formulated nano gels prepared from extracted flavonoids of the tobacco stem that have shown potent antimicrobial, antioxidant, and angiogenic agents can be utilized as herbal gels for wound healing purposes.

CHAPTER 1

INTRODUCTION

The skin is the largest organ in the surface area of the human body and has a critical structure that shields internal tissues from mechanical damage, microbial infection, ultraviolet radiation, and extreme temperature. This makes it highly susceptible to injury and has a significant impact on both individual patients and the healthcare economy. Skin repair requires intricate synchronization of different cell types in sequential steps. The epidermis is the outer layer that withstands the harsh external environment [1]. The dermis is rich in extracellular matrix (ECM), vasculature, and mechanoreceptors and provides the skin with strength, nutrients, and immunity [2]. Subcutaneous adipose tissue underlies the dermis and functions as an energy reserve. When the skin is wounded, multiple steps are involved in processes that deal with the three layers of healing [1]. Wound healing has been studied for decades, but the underlying molecular mechanism remains unclear. Skin wounds are typically divided into acute and chronic types. The skin serves as a protective barrier against physical and chemical threats, such as exposure to radiation or thermal stress, and pathogen entry, which radically compromises the functionality of the barrier. The public health sector is concerned about skin wounds and poor wound healing. Treatments that are complex and time-consuming add to the cost of healthcare. Even in most uncomplicated cases, burns, chronic wounds, and other difficult-to-treat wounds necessitate surgery and prolonged hospitalization [3]. Silver products (e.g., silver nitrate and silver sulfadiazine) are commonly used in infected chronic wounds and burn dressings because of the release of silver ions, which exhibit potent antibacterial effects. Silver ions bind to the thiol groups of peptidoglycans, causing bacterial cell lysis [4]. In addition, microbial DNA is altered by blockage of respiratory enzyme pathways. Moreover, silver compounds are effective against multidrug-resistant bacteria and bacterial biofilms. However, silver-derived products may cause tissue toxicity [5]. Nanomaterials

for tissue regeneration can be developed under different structures: nanoparticles, nanospheres, nanocapsules, nanoemulsions, nanocarriers, and nanocolloids [4]. Nanoparticles are being used for diverse purposes, from medical treatments, in various branches of industry production such as solar and oxide fuel batteries for energy storage, to wide incorporation into diverse materials of everyday use such as cosmetics or clothes. Metallic and metal oxide nanoparticles have been extensively studied, with silver, gold, and zinc compounds being the most researched due to their unique properties such as antibacterial activity and reduced skin penetration. The effectiveness and toxicity of these nanoparticles depend on specific features such as size, structure (smaller particles are more biologically active), surface functionalization, zeta potential, and polydispersity index. Previous reports have indicated that higher concentrations of silver nanoparticles (AgNPs) can decrease keratinocyte viability, metabolism, migration, and differentiation, leading to cell death through the activation of caspase 3 and 7 (proteases involved in programmed cell death) and dose-dependent DNA damage. To minimize side effects, silver nanoparticles can be used in low doses in combination with antimicrobial drugs to achieve increased efficiency [6].

The current book includes the nano-formulation of tobacco stem to observe wound healing on Wistar rat models.

CHAPTER 2

REVIEW OF LITERATURE

1. Medicinal Plants

For millennia, medicinal plants have played a pivotal role in human healing practices, maintaining their significance in the realm of wound care. Various traditional healing systems across the globe have harnessed the therapeutic potential of diverse plants to address wounds, injuries, and skin issues. These plants harbor bioactive compounds with distinct pharmacological actions that actively participate in the wound-healing process [7]. In recent years, scientific research has endeavored to uncover the mechanisms of action of these compounds and validate their effectiveness in controlled clinical settings. Before delving into the specifics of medicinal plants and their bioactive compounds, it is crucial to understand the fundamental stages of wound healing. This intricate process unfolds in a series of well-coordinated events, typically categorized into inflammation, proliferation, and tissue remodeling.

Aloe vera, a succulent plant revered for its medicinal properties across various cultures, holds a special place in wound care. The gel extracted from its leaves encompasses a rich array of bioactive compounds with demonstrated potential in wound healing. Aloe vera polysaccharides play a pivotal role by stimulating fibroblast proliferation and collagen synthesis, essential for tissue repair [8]. Anthraquinones found in aloe vera exhibit anti-inflammatory and antimicrobial properties, contributing to the creation of an optimal environment for wound healing [9]. Turmeric, derived from the *Curcuma longa* plant, has a longstanding history of medicinal use in traditional systems like Ayurveda. The primary bioactive compound in turmeric, curcumin, has garnered significant attention for its therapeutic properties. Curcumin modulates inflammatory responses, thereby reducing inflammation and promoting tissue repair [10]. Its antioxidant effects protect against oxidative stress, fostering a conducive environment for

wound healing. Furthermore, curcumin's antimicrobial properties play a role in preventing infections, bolstering the overall healing process. Calendula, commonly known as marigold, has a traditional history of use in addressing skin conditions and promoting wound healing. The flowers of this plant contain bioactive compounds with anti-inflammatory, antioxidant, and antimicrobial properties. Collectively, these compounds create an optimal environment for supporting the wound-healing process. While clinical trials provide substantiation for the efficacy of these medicinal plants, challenges such as standardization and integration into mainstream medicine warrant attention. This marks a promising yet evolving frontier in wound care, where traditional wisdom meets modern scientific validation [11].

2. *Nicotiana tabacum*

Nicotiana tabacum (Tobacco) belongs to a family of Solanaceae. It's a perennial herbaceous plant that is found only in cultivation, it grows up to 2 meters in height. It is native to tropical and subtropical America but today it is cultivated throughout the world. All the parts are sticky and are covered with short viscid-glandular hairs which exude a yellow secretion containing nicotine. Its synonyms are tobacco, tamak, and siah (marma). 20% of tobacco resources are discarded as processing waste, which pollutes the environment and causes a large amount of waste [12]. In India, the leaves of tobacco plants have been used as sedative, antispasmodic, vermifuge, antiseptic, emetic, and narcotic. The decoction of leaves is also applied for muscle relaxation and relieving pain [13]. Discarded tobacco leaves are valuable because of the presence of bioactive compounds. However, tobacco leaf is rich in polyphenols which possess various bioactive that affect the quality of tobacco leaf [14]. Nicotine which is isolated from leaves of tobacco in associated with zinc has shown antibacterial activity against ten different strains of Gram-positive and Gram-negative bacterial strains [15]. The antinociceptive activities of methanolic leaf extract of tobacco using tail immersion, hot plate, and acetic acid have revealed abdominal constrictions in albino Wistar mice [16]. Tobacco has also given its antifungal activity against *Fusarium solani* [17]. As a traditional medicine, for the treatment of tuberculosis coughs were also screened for activity against *Mycobacterium tuberculosis* [18]. Tobacco contains 30 -40% of

vegetal oil and can produce oil and biodiesel. Tobacco also consists of citric acid that can be used for the production of dyes and varnishes. As far as the stem of tobacco is concerned, the production of briquettes has values of tobacco stem [19]. It's been reported that extracts of seeds have shown antibacterial activity on *Staphylococcus* [20]. The antioxidant properties of flavonoids and polysaccharides from tobacco (*Nicotiana tabacum* L.) leaves were evaluated in *vitro* systems, e.g., scavenging activities on hydroxyl, superoxide anion, 1,1-diphenyl-2-picrylhydrazyl (DPPH) and 2,2'-casinobis (3-ethylbenzthiazoline-6-sulphonic acid) (ABTS) radicals, and reducing power. Flavonoids showed much better activity than polysaccharides in scavenging activities on free radicals. When compared to the positive control, ascorbic acid, both showed weaker antioxidant potential. However, flavonoids possessed comparable superoxide anion, DPPH, and ABTS radical scavenging abilities to ascorbic acid at high concentrations (600 µg/mL). Meanwhile, it was found that flavonoids had prominent effects on the reducing power, which was equivalent to ascorbic acid, and was significantly higher than polysaccharides [35]. Different parts of tobacco plants were dried at 40 and 70 °C. Some of them were also dried at room temperature. Dried plant material was extracted by sonication to obtain hydro-alcoholic extracts (70%). Total phenol and total flavonoids were determined as well as antioxidant activities which were evaluated through different methods (capacity for scavenging DPPH, ABTS, superoxide, and hydroxyl radicals; capacity for preventing lipid peroxidation using egg yolk as substrate; and reducing power). In young and adult plants, leaves generally had higher amounts of phenols (14.46-23.05 mg g⁻¹) than the remaining parts of the plant, independent of the temperature used. Generally, roots had lower amounts of phenols (1.56-4.63 mg g⁻¹). Leaves and flowers had significantly higher concentrations of flavonoids (3.08-4.17 mg QE g⁻¹ and 1.17-2.12 mg QE g⁻¹, respectively) than the remaining parts. The antioxidant activity was generally higher in leaf extracts, although stalk ones had also a good capacity for scavenging hydroxyl radicals [21]. Different isoforms of chitinases and @- 1,3-glucanases of tobacco (*Nicotiana tabacum* cv Samsun NN) were tested for their antifungal activities. The class I, vacuolar chitinase, and @-1,3-glucanase isoforms were the most active against *Fusarium solani* germings, resulting in lysis of the hyphal tips and growth inhibition. In addition, we observed that the

class I chitinase and α -1,3 -glucanase acted synergistically [22]. The antibacterial activity of extracts of twelve Nigerian medicinal plant species and a “wonder cure” concoction (Epa – Ijebu) used in traditional medicine for the treatment of tuberculosis and cough were screened for activity against *Mycobacterium tuberculosis* isolated from tuberculosis patient sputum and the control strains of *M. tuberculosis* (H37RV). Both ethanolic and aqueous solutions of the extract of *Allium ascalonicum*, *Terminalia glaucescent*, *Allium cepa*, and *Securidaca long pedunculate* (ethanolic extract only) at 0.05 g/ml as well as aqueous solution of “wonder cure” concoction at same concentration inhibited the growth of *M. tuberculosis*. Aqueous and ethanol extract of *Nicotiana tobacco* is used as a medicinal plant extract in the treatment of tuberculosis [23].

3. Role of Secondary Metabolite in Wound Healing

Secondary metabolites refer to organic compounds synthesized by plants, fungi, and microorganisms, which don't directly contribute to the organism's growth, development, or reproduction, unlike primary metabolites essential for basic life processes such as photosynthesis and cell division. These secondary metabolites, in contrast, play diverse roles, often enhancing the organism's survival in its environment [24]. A significant function is their role in defense against herbivores, pathogens, and competitors, acting as chemical deterrents, toxins, or attractants. Plants, for instance, produce alkaloids, terpenoids, and phenolic compounds to deter herbivores and protect against infections. Beyond defense and competition, secondary metabolites function as signaling molecules, facilitating intercellular communication in response to environmental changes. In microbial communities, quorum sensing involves the production and detection of specific secondary metabolites, allowing bacteria to coordinate behaviors like forming biofilms. Additionally, these compounds have pharmaceutical importance, contributing to drugs like penicillin, paclitaxel, and artemisinin. In the context of wound healing, secondary metabolites play a crucial role through various mechanisms. They aid in tissue repair, reduce inflammation, and prevent infections. Compounds like flavonoids, terpenoids, and alkaloids, known for their anti-inflammatory properties, help manage inflammation during wound healing. Further, metabolites with antimicrobial properties, including tannins and alkaloids, contribute to preventing infections,

creating an optimal wound-healing environment [25]. Promoting cell proliferation and tissue regeneration is vital in wound healing. Secondary metabolites like growth factors, peptides, and polyphenols stimulate cell proliferation, angiogenesis, and tissue repair. Certain polyphenols, such as epigallocatechin gallate (EGCG) in green tea, show the potential to enhance collagen synthesis, crucial for wound closure and tissue strength. Traditional medicine has long utilized plants rich in secondary metabolites for wound healing, such as aloe vera with anti-inflammatory properties or honey with antimicrobial and anti-inflammatory components [26]. While the potential of secondary metabolites in wound healing is promising, further research is needed to understand their specific mechanisms, optimal formulations, and potential side effects. Standardization of plant extracts or isolated compounds is crucial to ensure consistent therapeutic effects. The multifaceted contributions of secondary metabolites underline their significance in both ecological interactions and medical applications [27].

4. Flavonoids as a wound healer

Flavonoids, a category of polyphenolic compounds present in a variety of plants, fruits, vegetables, and beverages, have garnered attention for their potential to promote wound healing. These compounds showcase a diverse array of biological activities, including antioxidant, anti-inflammatory, antimicrobial, and angiogenic effects, all of which can contribute to different stages of the wound healing process. Several flavonoids have undergone scrutiny for their potential impact on wound healing, and some noteworthy ones include:

1. Quercetin: Widely distributed in fruits, vegetables, tea, and red wine, quercetin boasts anti-inflammatory and antioxidant properties. Research indicates its ability to foster wound healing by encouraging fibroblast proliferation and collagen synthesis, crucial for effective tissue repair [28].
2. Kaempferol: Found in various fruits and vegetables, kaempferol possesses anti-inflammatory and antioxidant properties. Studies suggest that kaempferol may expedite wound closure by promoting cell migration and proliferation, along with angiogenic effects that contribute to the formation of new blood vessels in the wounded area [29].

3. Epigallocatechin gallate (EGCG): Derived from green tea, EGCG is a flavonoid with potent antioxidant and anti-inflammatory activities. Research has demonstrated its potential in promoting wound healing by augmenting collagen synthesis, hastening tissue regeneration, and mitigating inflammation [29].

4. Hesperidin: Abundant in citrus fruits, hesperidin has been explored for its wound-healing attributes. It exhibits anti-inflammatory effects and has been shown to promote angiogenesis, a critical process for the development of new blood vessels in the wound bed [30].

5. Rutin: Commonly found in buckwheat, citrus fruits, and tea, rutin possesses antioxidant and anti-inflammatory properties. Research has delved into its potential to enhance wound healing by promoting collagen deposition and angiogenesis [31].

The exploration of flavonoids and wound healing involves a comprehensive range of both *in vitro* and *in vivo* studies, shedding light on their mechanisms of action. These studies often focus on various facets of the wound-healing process, including inflammation, cell proliferation, collagen synthesis, angiogenesis, and tissue regeneration. While the potential advantages of flavonoids in wound healing are promising, it's crucial to acknowledge that the efficacy of these compounds can be influenced by factors such as concentration, formulation, and the specific type of wound. Furthermore, additional research is imperative to establish standardized protocols for the application of flavonoids in wound care.

5. Polyphenols as a wound-healing agent

Polyphenols, a diverse group of naturally occurring compounds found in plants, have garnered significant attention for their potential involvement in wound healing. These compounds, known for their antioxidant and anti-inflammatory properties, play a contributory role in various stages of the wound healing process. In this comprehensive exploration, we delve into the mechanisms through which polyphenols impact inflammation, cell proliferation, collagen synthesis, angiogenesis, and tissue regeneration. Polyphenols, with their anti-inflammatory properties, can modulate this phase [32]. As per the previous studies, a study investigated the effects of