

Antioxidants from Medicinal Plants and of Synthetic Origin

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Ph.D. Dissertation

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Pakistan.



2005

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

ALLAH in the name of the most Affectionate, the Merciful

His (ALLAH) Dignity, when He intends anything, is only to say to it, "Be", so it is (Quran: Sura 36; Verse 82).

He is ALLAH, besides whom there is no god, the Knower of the unseen and the seen; He is the Beneficent, the Merciful. He is ALLAH, besides whom there is no god, the King, the Holy, the Giver of peace, the Granter of security, Guardian over all, the Mighty, the Supreme, the Possessor of every greatness, Glory be to ALLAH from what they set up (with him). He is ALLAH, the Creator, the Maker, the Fashioner, His are the most excellent names; whatever is in the heavens and the earth declares His Glory and He is the Mighty and Wise (Quran: Sura 60; Verse 22-24).

To My Parents

*Without their love, care and prayers
I would never succeed.*

Acknowledgements

First of all I am grateful to Almighty ALLAH who in every moment of my life always with me and blessed me beyond my imagination, who helped me in every difficult time, who loved me, who give me strength and perseverance, whatever I am today is just because Him (ALLAH Subhana-Wu-Tala).

I am grateful to my supervisor, Prof. Dr. Atta-ur-Rahman N. I. H. I., S. I., T. I., Director of International Center for Chemical Sciences, University of Karachi, Federal Minister/ Chairman, Higher Education Commission of Pakistan, for giving me the opportunity to work in his research group and for experienced guidance into the scientific way of thinking.

I would like to thank my advisor, Prof. Dr. Muhammad Iqbal Choudhary S. I., T. I., Acting Director of International Center for Chemical Sciences, University of Karachi for providing me an interesting research project, his constant support, constant encouragement, guidance and providing me enough independence to decide the things throughout the study. He helped me to grow in both my competence and in confidence as a researcher.

I want to acknowledge all the research collaborators who have provided me a large number of interesting samples to make this study successful and comprehensive. I want to express my sincere gratitude to Dr. Khalid M. Khan T. I. and his students Dr. Maimona Rasheed, Ms. Shagufta Rahat, Mr. M. Zarrar Khan and Mr. Sajid Iqbal for fruitful collaboration by providing a large number of synthetic compounds for antioxidant studies.

I am grateful to Ms. Suad Naheed for helping me during the hepatoprotective studies of chemical constituents. For her help in every aspect, for introducing me to basics of in vivo study, for many discussions on the results, for always sharing her knowledge and ideas throughout this study. My special thanks to Dr. Junaid M. Alam for providing the working facilities at the Department of Biochemistry and Chemical Pathology, Liaquat National Post Graduate Medical Center.

I am especially thankful to Ms. Saima Jalil who conducted a part of the cytotoxicity studies presented in this thesis. I also enjoyed the discussions with her. My special thanks go to Mr. Imran Hameed for encouragement.

I especially thankful my dear friends, Ms. Humera Naz, Ms. Seema Zareen, Ms. Shahida Shujant, Ms. Juveria Siddiqui, Ms. Suad Naheed, Ms. Naheed Fatima, Ms. Shehnaz Parveen, Ms. Aniq Naz, Ms. Nurgul Sultanova, Ms. Salma Shahnaz and Ms. Rasheeda Swaleh for being so encouraging, patience and friendly.

I want to express special thanks to my senior colleagues, Dr. Zareen Amtul and Dr. Usman Ghani for their genuine concern and many discussions on my research project. I also thank to my colleagues, Dr. Shakeel Ahmad, Mr. Sajjad Hussain, Mr. Arif Lodhi, Mr. Sarfaraz A. Nawaz, Dr. Asad Khalid and Dr. Mohammad Ahmad for their co-operation.

I express my thanks to all the faculty members at the H. E. J. Research Institute of Chemistry who taught me chemistry in a very nice way and gave me good concepts of organic chemistry. I am also thankful to the staff of H. E. J. R. I. C. for their help and cooperation.

I am very grateful to my dear parents, grand mother and brothers, Taqi, Naqi, Kashif and Saad for their endless love, prayers, support and continuous encouragement. Without their faithful love, care and understanding it would be difficult for me to find the passion to succeed.

In the end, I want to express my sincere gratitude to everyone who, in their own way, has helped me to complete this thesis.

Talat Maksimoor

Summary

This dissertation describes studies on antioxidant activities of a number of chemical compounds, natural in origin or synthesized in the laboratory. As a result of this study, a total of eighty four compounds were identified as antioxidants. Out of these eighteen new and twelve known phytochemicals were isolated from different medicinally important plants and fungal cultures, while fifty four synthetic chemical constituents, belonging to six different classes of compounds, were identified as antioxidants. Modification of the structures of parent chemical constituents of each class of synthetic compounds resulted in enhancement of antioxidant activity. The antioxidant activities of these compounds were determined through their radical scavenging and xanthine oxidase inhibition potential using three assays. First, all the compounds were checked for radical scavenging activity in an *in vitro* method, i.e. DPPH radical scavenging assay. Compounds which showed radical scavenging activity in DPPH radical scavenging assay were further studied for their superoxide anion scavenging potential using NADH/PMS/O₂^{•-} assay. The compounds which were checked for radical scavenging activity were also assayed for xanthine oxidase inhibition. Based on the bioassay results, structure-activity relationship (SAR) studies on different classes of antiradical and xanthine oxidase inhibitors were conducted. The six most active xanthine oxidase inhibitors were also studied for their kinetic behavior. *In vivo* studies using CCl₄-induced liver toxicity assay in rat model on the selected twenty eight compounds of synthetic and natural origins, were also carried out. From these different classes of compounds, the most active compounds were subjected to *in vivo* studies. Subsequently, these compounds were

also checked for their cytotoxicity using a sensitive *in vitro* cytotoxicity assay on human neutrophils.

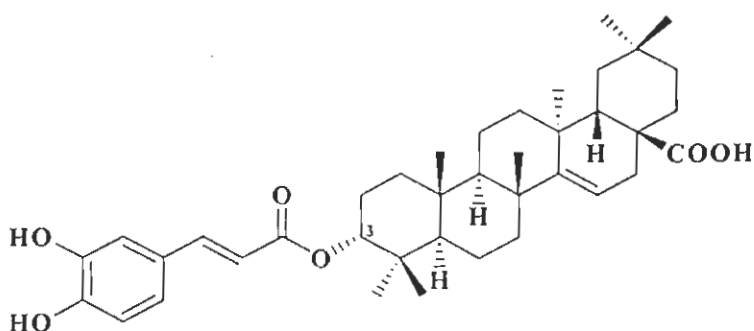
ANTIOXIDANTS FROM MEDICINAL PLANTS

New Compound

Radical Scavenger

Hepatoprotective

Cytotoxicity: $ED_{50} = 90.35 \mu\text{g/mL}$



3 α -[3',4'-Dihydroxy *trans*-cinnamoyl]-oxy-D-friedoolean-14-en-28-oic acid (2)

Isolated from aerial parts of *Tamarix hispida* Willd. (Tamaricaceae)

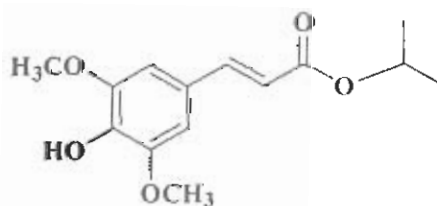
New Compound

Radical Scavenger

Hepatoprotective

Xanthine oxidase inhibitor

Cytotoxicity: $ED_{50} = 63.24 \mu\text{g/mL}$

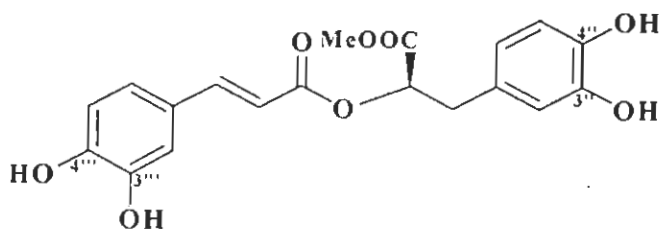


Iso-propyl-3,5-dimethoxy-4-hydroxycinnamic acid (8)

Isolated from the brown algae, *Spatoglossum variable* Figari et De Notar (Dictyoaceae)

Known Compound

Radical Scavenger
Hepatoprotective
Xanthine oxidase inhibitor
Cytotoxicity: $ED_{50} = <12.50 \mu\text{g/mL}$

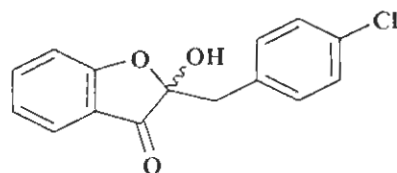


1-(3'',4''-Dihydroxybenzyl)-2-methoxy-2-oxoethyl-(E)-3'-(3'',4''-dihydroxyphenyl)-2-propenoate (11)

Isolated from stem bark of *Cussonia bancocensis* Arev. & Pellegr.
(Araliaceae)

New Compound

Radical Scavenger
Hepatoprotective
Xanthine oxidase inhibitor
Cytotoxicity: $ED_{50} = 200 \mu\text{g/mL}$



4'-Chloro-2-hydroxyaurone (33)

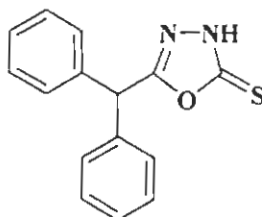
Isolated from the brown algae, *Spatoglossum variable* Figari et De Notar
(Dictyoaceae)

Oxadiazole Thione Derivative

Radical Scavenger

Hepatoprotective

Cytotoxicity: $ED_{50} = 69.92 \mu\text{g}/\text{mL}$



5-Diphenylmethyl-1,3,4-oxadiazole-2-(3H) thione (66)

Synthetic compound

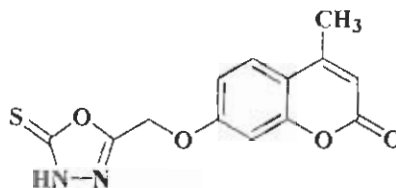
Coumarin Derivative

Radical Scavenger

Hepatoprotective

Xanthine oxidase inhibitor

Cytotoxicity: $ED_{50} = 112.72 \mu\text{g}/\text{mL}$



4-Methyl-7-[(5-thio-4,5-dihydro-1,3-oxazol-2-yl)methoxy]-2H-chromen-2-one (87)

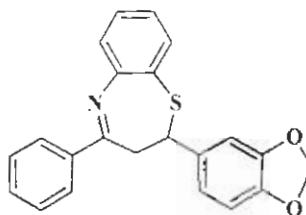
Synthetic compound

Benzothiazepine Derivative

Radical Scavenger

Hepatoprotective

Cytotoxicity: $ED_{50} = >200 \mu\text{g}/\text{mL}$



2-(1,3-Benzodioxol-5-yl)-4-(phenyl)-2,3-dihydro-1,5-benzothiazepine (102)

Synthetic compound

خلاصہ

بیشمار تعداد میں کیمیائی مرکبات جو کہ قدرتی ماخذ اور تجربہ گاہ میں تیار شدہ تھے، ان کی ضد تکیدی عاملیت کی جانچ کی گئی۔ اس مطالعے کے نتیجے میں چوراسی (84) مرکبات بطور ضد تکید کے شناخت ہوئے۔ جن میں سے اٹھارہ (18) نئے اور بارہ (12) پہلے سے دریافت ہوئے۔ یہ مرکبات جو کہ طبی لحاظ سے اہم پودوں اور فنجائی کے کلچرز سے علیحدہ کیئے گئے تھے، بطور ضد تکید خصوصیت دریافت کئے جبکہ پھون (5) کیمیائی اجزاء جو کہ چھ (6) مختلف جماعتوں سے تعلق رکھتے تھے تجربہ گاہ میں تالیف کیئے گئے۔ ان مرکبات کی ضد تکیدی عاملیت بذریعہ ضد ریڈیکل اور زینتھین آکسیڈیز خامرے کے خلاف عملیت کی صلاحیت کے معلوم کیا گیا جو کہ تین (3) طریقہء کار کے لیے کیا گیا۔ پہلا، تمام مرکبات کی ضد ریڈیکل عاملیت کو ایک خارجی طریقہ کار یعنی DPPH ریڈیکل خاکروبی صلاحیت کے لیے جانچا گیا۔ دوئم، جن مرکبات نے اس طریقے سے بہترین ضد ریڈیکل عاملیت کا اظہار کیا ان کو مزید NADH/PMS/C نظام کا استعمال کرتے ہوئے سپر آکسائیڈ این آئن خاکروبی صلاحیت کے لیے جانچا گیا۔ سوئم، ان ہی بات جو کہ DPPH خاکروبی صلاحیت کے لیے جانچے گئے تھے، کو زینتھین آکسیڈیز خامرے کے خلاف بطور مانع بھی جانچا گیا۔ نیاتی مطالعے کے نتائج کی بنیاد پر ساخت عاملیت تعلق کا مطالعہ کیا گیا جو کہ مرکبات کی مختلف جماعتوں کی ضد ریڈیکل اور زینتھین آکسیڈیز کے بہترین مانع مرکبات پر کیا گیا۔ چھ (6) سب سے زیادہ عامل مانع کو زینتھین آکسیڈیز کے خلاف ان کے مرکبی روئے لیے بھی مطالعہ کیا گیا۔ داخلی مطالعے کے لیے چوہوں کو بطور نمونہ استعمال کرتے ہوئے CCl₄ سے جگر میں سمیاتی اثرات پیدا کیئے اور پہلے سے منتخب شدہ اٹھائیس (28) مرکبات جو کہ قدرتی اخز شدہ اور تالیفات کو اس سمیاتی اثر کے خلاف جانچا گیا۔ علاوہ ازیں، مرکبات کو ان کی خلوی سمیاتی اثرات کے مطالعے کے لیے بھی جانچا گیا، جو کہ ایک حساس خارجی طریقہء کار سے انسانی خون کے خلیوں کو استعمال کر کے کیا گیا۔

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1. Atta-ur-Rahman, F. N. Ngounou, M. Iqbal Choudhary, Shahid Malik, Talat Makhmoor, M. Nur-e-Alam, Seema Zareen, D. Lontsi, J. F. Ayafor and B. L. Sondengam, New Antioxidant and Antimicrobial Ellagic acid Derivatives from *Pteleopsis hylodendron*, *Planta Medica*, 2001, **67**, 335-39.
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