

**Antioxidant and Antitumorigenic efficacy of methanolic
extracts of *Gloriosa superba* and Silver Nanoparticles
of methanolic extracts of *Gloriosa superba*
to DLA tumor cells**

By

Saradha Devi, K.M.

Supervisor

Dr. Tmt. S. Annapoorani

**A thesis submitted to
Avinashilingam Institute for Home Science and Higher Education
for Women, Coimbatore – 43**

**In partial fulfilment of the requirements for the degree of
Doctor of Philosophy in Biochemistry**

March, 2013

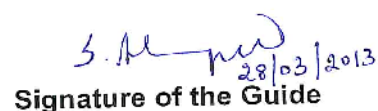
Certificate

This is to certify that the thesis entitled “**Antioxidant and Antitumorigenic efficacy of methanolic extracts of *Gloriosa superba* and Silver Nanoparticles of methanolic extracts of *Gloriosa superba* to DLA tumor cells**”, submitted to Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore, for the award of the Degree of **Doctor of Philosophy in Biochemistry**, is a record of original research work done by **Saradha Devi, K.M.** during the period of her study in the Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore, under my supervision and guidance and the thesis has not formed the basis for the award of any Degree / Diploma / Associateship / Fellowship or similar title to any candidate of any other University or Institute.



**Signature of the
Head of the Department**

Dr.R.PARVATHAM, M.Sc.,M.Phil.,Ph.D.,
Dean, Faculty of Science
Professor & Head
Dept. of Biochemistry, Biotechnology & Bioinformatics
Avinashilingam Institute for Home Science
and Higher Education for Women
Coimbatore - 641 043



Signature of the Guide

Dr. Mrs. S. ANNAPOORANI
Professor of Biochemistry
Avinashilingam University for Women
Coimbatore - 641 043.



Signature of the Dean

Dr.R.PARVATHAM, M.Sc.,M.Phil.,Ph.D.,
Dean, Faculty of Science
Professor & Head
Dept. of Biochemistry, Biotechnology & Bioinformatics
Avinashilingam Institute for Home Science
and Higher Education for Women
Coimbatore - 641 043

Declaration

I hereby declare that the matter embodied in the thesis entitled “**Antioxidant and Antitumorigenic efficacy of methanolic extracts of *Gloriosa superba* and Silver Nanoparticles of methanolic extracts of *Gloriosa superba* to DLA tumor cells**”, is the result of investigations carried out by me in the Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore, under the supervision and guidance of **Dr. Tmt. S. Annapoorani**, Professor, Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore and it has not been submitted for the award of any Degree / Diploma / Associateship / Fellowship of any other University or Institute.

S. H. [Signature]
28/03/2013

Signature of the Guide

M. S. [Signature]
28/03/2013

Signature of the Candidate

Acknowledgement

I owe my great obeisance to '**His' beatitude**, the Hosanna, the tag line of all in all.

I profusely thank our Messiah Thiru. **T.S.K.Meenakshisundaram**, Chancellor, Avinashilingam University for Women, Coimbatore for the consent to initiate my doctoral degree in the University and to mount the winning zone of my doctoral programme.

I am honoured to extend my appreciation and relentless support summoned upon me by **Dr. Tmt. Sheela Ramachandran**, Vice-Chancellor, Avinashilingam University for Women, Coimbatore for the efficient and effective utilization of infrastructure in a non-chalant way.

Former Vice-Chancellors **Thiru. K. Shanmuganandam** and **Thiru.Kulandaivel** are immensely thanked for the platform provided to pursue the research in length and breadth and courting success quotient.

Our pillar of strength **Col. Dr. Saroja Prabakaran**, Former Vice--Chancellor is deeply thanked for her benevolence and humility.

My heartfelt thanks are due to **Dr. Tmt. Gowri Ramakrishnan**, Registrar is greatly acknowledged for her delirious support, wiping away the administrative tangles and barricades to reach up trumps.

The perennial source of knowledge **Dr. Tmt. R.Parvatham**, Dean, Faculty of Science, for her incessant patronage rendered throughout the research endeavors and without lampooning for my nagging help.

I overcame the blockades in the academic arena, through the warmth and concern shown by **Dr. Santhana Krishnan**, Research Director, Avinashilingam University for Women, Coimbatore.

I extend a deep sense of gratitude to my 'Guardian Angel', **Dr. Tmt. S. Annapoorni**, M.Sc,Dip.Ed.,Mphil.,PhD., Professor of Biochemistry, for the innocuous remarks, impeccable ideas, invaluable suggestions, fertile thought, broad intellectual acumen and assiduous guidance for fine tuning and tailor making my dissertation to shape into a fool-proof blue print and also maneuvering the rough edges of my work.

I whole heartedly thank a battalion of apostles **Dr. Tmt. G.P.Jeyanthi** Controller of Examination.

I am indebted to **Mrs. M.Lakshmi** laboratory stalwart for her whole hearted, rough and ready support for speedy execution of the analysis.

I deeply thank my beloved **family** members viz., loveband **Dr. R. Rajasekaran**, (Asst. Professor), Tamil Nadu Agricultural University, Coimbatore, Clubbing thanks with him are my lovable son master **R. Devanand**, dotting father **Thiru. K.Muthukrishnan**, Caring mother **Mrs.K.Vijayalakshmi**, affectionate brothers **Dr. K. M.Parivelan** and **Mr.K.M.Elango** who were always in toes, withstanding my fluctuating Psyche at times of crises and mood swing and while taking the rough with the smooth.

I am immensely appeased to extend my thankfulness to the blush of intellects **Dr. J.Venkatapirabhu** (Professor), TNAU, Coimbatore, **Dr. S. Ramesh**, Associate Professor & Head, Central Instrumentation Laboratory, TANUVAS, Chennai, sincere thanks to, **Mr.Srinivasan**, SAIF, IIT- Madras, **Dr.Parimelazhagan** (Asso. Prof.), Dept., of Botany, **Dr. Prabakaran** (Asst. Prof.), **Dr.Girija** (Asst. Prof.) and **Ms.Ramya** (Research scholar), Dept., of Biotechnology, Bharathiar University, Coimbatore, **Mr.Harihara Subaramanian**, (Asst.Prof.), Dr.N.G.P. College of Pharmacy, Coimbatore, **Dr.Amutha Venkatapirabhu** (Asst. Prof.), Dept of Physics, PSG College of Tech. Consummate patronage rendered upon me by **K.M.Sridhar**, Section Officer and **Ms.TamilMani** (Research scholar), CLRI, Chennai are duly acknowledged, **Dr.K.Pandian**,(Asst. Prof.), **Mr.Alexander**, Research Scholar, Madras University, Chennai for their auxiliary help in time.I express my sincere thanks to **Ms.S.V.Soundariya** for her constant support for my research work.

My sincere thanks to **Ms.Gomathi**, **Mrs. R. Amutha priya**, **Mrs. D. Jeba Sweetly**, **Mrs.Smithi Rao**, **Mrs.Sumaya**, **Mrs.Krithika**, **Ms.A.Poongothai**, **Mrs.K.P.Sreena**, **Mrs.M.Saroja**, **Dr.Mrs.R.Santhi**, **Ms. P.S. Sayana** and **Dr.Mrs.D.Chandraprabha** for their help and constant support throughout my research period.

I owe my sincere thanks to all the other **Staff members and Lab assistants** of the Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam University for Women, Coimbatore, for their constant support and help to finish my Ph.D., successfully.

I express my sincere thanks to **Mr. K. Selvakumar**, Euro Informatics for his faultless editing and flawless typing of the thesis.

Contents

Chapter No.	Title	Page No
	List of tables	
	List of figures	
	List of plates	
	List of appendices	
	List of abbreviations	
	List of publications	
1	Introduction	1
2	Review of literature	6
3	Experimental procedure	66
4	Results and discussion	82
5	Summary and conclusion	175
	Bibliography	185
	Appendices	229

List of Tables

Table No.	Title	Page No.
1.	<i>In vitro</i> cytotoxicity studies of AgNPs on different cell lines	13
2.	Phytochemical constituents of MGsSTL	87
3.	HPTLC - Peak table for alkaloid profile of MGsS	90
4.	HPTLC - Peak table for alkaloid profile of MGsT	91
5.	HPTLC - Peak table for alkaloid profile of MGsL	91
6.	X-RAY Diffraction pattern of AgMGsSTL	115
7.	Measurement of the crystalline size of AgMGsS using Debye-Scherrer's equation	115
8.	Measurement of the crystalline size of AgMGsT using Debye-Scherrer's equation	116
9.	Measurement of the crystalline size of AgMGsL using Debye-Scherrer's equation	116
10.	Activities of hepatic enzymic antioxidants in controls and experimental Swiss albino mice	143
11.	Levels of hepatic non enzymic antioxidants in controls and experimental Swiss albino mice	151
12.	Levels of hepatic MDA in controls and experimental Swiss albino mice	155
13.	The average life span of DLA tumor induced mice treated with MGsSTL and AgMGsSTL	168

List of Figures

Figure No.	Title	Page No.
1.	Green route of nanoparticle synthesis	12
2.	The AgNP induced apoptotic pathway	13
3.	Capping and stabilization of AgNPs	14
4.	Characterization and detection of AgNPs	15
5.	Biosynthesis of AgNPs by plant leaf extract	16
6.	Localized Surface Plasmon Resonance	17
7.	FTIR absorption spectra of the biomolecules of aqueous ethanolic carom seed extract	17
8.	DLS apparatus	19
9.	Zeta potential	20
10.	Oxidative stress in cellular signaling	22
11.	Oxidative stress in health disorders	23
12.	Formation of Reactive Oxygen Species	25
13.	Generation of reactive and damaging hydroxyl radicals (OH•)	28
14.	Redox status in Cancer	33
15.	Antioxidants in ROS balance	34
16.	Defense against free radicals	35
17.	Mechanism of action of SOD	37
18.	Glutathione Oxidation Reduction (Redox) Cycle	40
19.	Non enzymatic antioxidants in the management of oxidative stress	41
20.	Oxidative stress and glutathione	45
21.	Normal angiogenesis and tumor angiogenesis	49

Figure No.	Title	Page No.
22.	Gamma scintigraphic image of Etoposide loaded tripalmitin NPs after 48 h of intratumoral injection	50
23.	Principle of MTT assay	51
24.	Nanocarriers for cancer therapy	53
25.	Nanoparticle localisation in solid tumours	54
26.	Targeted delivery of AgNPs in cancer therapy	55
27.	EEPS/NEEPS-induced apoptosis of cancer cell (A549)	61
28.	Stages of Apoptosis	62
29.	Phospholipid flipping and binding of Annexin V during apoptosis	63
30.	Detection of Apoptotic by Propidium iodide and Annexin V Staining	63
31.	Track A –MGsS Baseline display and Peak densitogram display	92
32.	Track B –MGsT Baseline display and Peak densitogram display	93
33.	Track C –MGsL Baseline display and Peak densitogram display	93
34.	Track D – Colchicine standard Baseline display and Peak densitogram display	93
35.	3D display of HPTLC chromatogram of MGsSTL and colchicine	94
36.	UV-Visible spectra of MGsS and AgMGsS	97
37.	UV-Visible spectra of MGsT and AgMGsT	97
38.	UV-Visible spectra of MGsL and AgMGsL	98
39.	FT-IR spectra of MGsS and AgMGsS	101
40.	FT-IR spectra of MGsT and AgMGsT	102

Figure No.	Title	Page No.
41.	FT-IR spectra of MGsL and AgMGsL	103
42.	SEM micrograph of AgMGsS	106
43.	EDAX spectrum of AgMGsS	106
44.	SEM micrograph of AgMGsT	106
45.	EDAX spectrum of AgMGsT	106
46.	TEM micrograph of AgMGsS	108
47.	TEM micrograph of AgMGsT	108
48.	Particle size distribution histogram of AgMGsS	110
49.	Particle size distribution histogram of AgMGsT	110
50.	Particle size distribution histogram of AgMGsL	111
51.	Zeta potential of AgMGsS	112
52.	Zeta potential of AgMGsT	113
53.	Zeta potential of AgMGsL	113
54.	XRD of AgMGsS	117
55.	XRD of AgMGsT	117
56.	XRD of AgMGsL	118
57.	Percentage DPPH radical scavenging activity of MGsSTL, AgMGsSTL and Vitamin C	121
58.	Percentage hydroxyl radical scavenging activity of MGsSTL, AgMGsSTL and Vitamin C	122
59.	Percentage superoxide radical scavenging activity of MGsSTL, AgMGsSTL and Vitamin C	123
60.	Percentage hydrogen peroxide scavenging activity of MGsSTL, AgMGsSTL and Vitamin C	124
61.	Percentage nitric oxide scavenging activity of MGsSTL, AgMGsSTL and Vitamin C	125

Figure No.	Title	Page No.
62.	IC ₅₀ values of MGsSTL and Vitamin C	129
63.	IC ₅₀ values of AgMGsSTL and Vitamin C	130
64.	<i>In vitro</i> cytotoxic effect of MGsSTL and AgMGsSTL to DLA tumor cells by MTT cell proliferation assay	132
65.	ED ₅₀ values of MGsSTL and AgMGsSTL to DLA tumor cells by MTT cell proliferation assay	132
66.	<i>In vitro</i> cytotoxic effect of MGsSTL and AgMGsSTL to DLA tumor cells by trypan blue exclusion assay	135
67.	ED ₅₀ values of MGsSTL and AgMGsSTL to DLA tumor cells by trypan blue exclusion assay	135
68.	Apoptotic effect of AgMGsS to DLA cells by fluorescent-activated cell sorting analysis	139
69.	Apoptotic effect of AgMGsT and AgMGsL to DLA cells by fluorescent-activated cell sorting analysis	140
70.	Effect of MGsSTL and AgMGsSTL on the activities of CAT in normal and DLA induced Swiss albino mice	145
71.	Effect of MGsSTL and AgMGsSTL on the activities of SOD in normal and DLA induced Swiss albino mice	146
72.	Effect of MGsSTL and AgMGsSTL on the activities of GST in normal and DLA induced Swiss albino mice	147
73.	Effect of MGsSTL and AgMGsSTL on the activity of GPx in normal and DLA induced Swiss albino mice	148
74.	Effect of MGsSTL and AgMGsSTL on the levels of GR in normal and DLA induced Swiss albino mice	149
75.	Effect of MGsSTL and AgMGsSTL on the levels of Vitamin A in normal and DLA induced Swiss albino mice	152
76.	Effect of MGsSTL and AgMGsSTL on the levels of Vitamin E in normal and DLA induced Swiss albino mice	152
77.	Effect of MGsSTL and AgMGsSTL on the levels of Vitamin C in normal and DLA induced Swiss albino mice	153

Figure No.	Title	Page No.
78.	Effect of MGsSTL and AgMGsSTL on the levels of GSH in normal and DLA induced Swiss albino mice	154
79.	Effect of MGsSTL and AgMGsSTL on the levels of MDA in normal and DLA induced Swiss albino mice	156

List of Plates

Plate No.	Title	Page No.
I	Synthesis of AgMGsSTL from MGsSTL	69
II.	Medicinal plant selected for the study	76
III.	Swiss albino mice (<i>Mus musculus</i>)	77
IV.	HPTLC Chromatogram pattern of MGsSTL	92
V.	DLA cells before and after treatment with extracts in trypan blue exclusion method	136
VI.	Histological view of the hepatocytes of controls, MGsSTL and AgMGsSTL treated Swiss albino mice	172
VII.	Histological view of the hepatocytes of controls, MGsSTL and AgMGsSTL in DLA induced Swiss albino mice	173

List of Appendices

Appendix No.	Title	Page No.
I	Preliminary phytochemical analysis	229
II	DPPH Radical Scavenging assay	230
III	Hydroxyl Radical scavenging assay	230
IV	Superoxide Radical scavenging assay	231
V	Hydrogen Peroxide scavenging assay	232
VI	Nitric oxide Scavenging assay	232
VII	MTT assay	233
VIII	Trypan blue exclusion assay	234
IX	Estimation of Catalase	235
X	Estimation of Superoxide Dismutase	235
XI	Estimation of Glutathione S- Transferase	236
XII	Estimation of Glutathione Peroxidase	237
XIII	Estimation of Glutathione Reductase	237
XIV	Estimation of Protein	238
XV	Estimation of Vitamin A	238
XVI	Estimation of Vitamin E	239
XVII	Estimation of Vitamin C	240
XVIII	Estimation of Reduced Glutathione	240
XIX	Estimation of the level of MDA	241
XX	Histological analysis of liver	242

List of Abbreviations

$\cdot\text{O}_2^-$	Superoxide anion
μm	micrometer
ACA	1'-(S)-1'-Acetoxy Chavicol Acetate
ADP	Adenosine diphosphate
Ag	Silver
AgMGsL	Silver nanoparticles of methanolic extract of <i>Gloriosa superba</i> leaves
AgMGsS	Silver nanoparticles of methanolic extract of <i>Gloriosa superba</i> seed
AgMGsT	Silver nanoparticles of methanolic extract of <i>Gloriosa superba</i> tuber
AgNO_3	Silver nitrate
AgNPs	Silver Nanoparticles
AIDS	Acquired Immune Deficiency Syndrome
ANOVA	Analysis Of Variance
ANSA	1-Amino-2-Naphthol-6-Sulphonic acid
ARDS	Acute Respiratory Distress Syndrome
ATP	Adenosine triphosphate
Au	Gold
AUC	Area under the curve
Bad	Bcl-2-associated death promoter
Bak	BCL2-antagonist/killer
Bax	protein of the Bcl-2_gene family
Bcl2	B-cell lymphoma 2
BclXL	B-cell lymphoma-extra large
BRL	Buffalo rat liver cells
BSA	Bovine serum albumin
CAT	Catalase
CCl_4	Carbon tetrachloride
CDNB	1-(4-chlorophenyl)-3-(3-dimethylamino)phenyl-4-methyl-5-pyrazolone
cm	Centimeter

Cmyc	myelocytomatosis cancer oncogene
CO	Carbon monoxide
cP	centipoise
CSCs	cancer stem-like cells
CSSs	Colloidal silver solutions
Cu K α	Copper X-unit
CV	– Central Vein
CVC	Central venous catheters
DLA	Dalton's Lymphoma Ascite
DLS	Dynamic Light Scattering
DMBA	7,12-dimethylbenz[α]anthracene
DMF	DiMethyl Formamide
DMSO	Dimethyl sulphoxide
DNA	Deoxyribonucleic acid
DNPH	2,4,Dinitro phenyl hydrazine
DPPH	1,1-diphenyl-2-picryl hydrazyl radical
DTNB	5,5'-dithio-2-nitro benzoic acid
EAC	Ehrlich Ascite Carcinoma
EC ₅₀	Fifty per cent Effective Concentration
EDTA	Ethylene Diamine Tetra Acetic acid
EDX	Energy Dispersive Spectroscopy
EGCG–pNPAu	(–)-Epigallocatechin-3-gallate NanoGold particles
FAD	Flavin adenine Dinucleotide
FBS	Fetal bovine serum
fCC	face-centered cubic
Fe ²⁺	Ferrous ion
Fe ³⁺	Ferric ion
FITC	Fluorescein IsoThioCyanate
FT IR	Fourier Transform Infrared spectroscopy
FWHM	<i>Full Width Half Maximum</i>
GPx	Glutathione Peroxidase
GR	Glutathione Reductase
Grx	Glutaredoxins

GSH	Reduced Glutathione
GSSG	Oxidised Glutathione
GST	Glutathione S-transferase
H ₂ O ₂	Hydrogen peroxide
H ₂ SO ₄	Sulfuric acid
H ₃ PO ₄	Phosphoric acid
HCl	HydroChloric acid
HEp -2	Human epidermoid larynx carcinoma-2
HGF	Hepatocyte growth factor
HPD	hematoporphyrin
HPTLC	High Performance Thin Layer liquid Chromatography
i.p	intra peritoneal
IC ₅₀	Fifty percent Inhibitory Concentration
IMR	Human Neuroblastoma cell line
KCl	Potassium Chloride
KH ₂ PO ₄	Potassium Dihydrogen Phosphate
kV	Kilovolts
LO ₂ ^o	Lipid peroxide
LPO	Lipid Peroxidation
LSPR	Localized Surface Plasmon Resonance
MBT-2	Murine Bladder Tumor-2
MDL	Malondi Aldehyde
MDR	multiple-drug resistance
mg	milligram
mg/g b wt	Milligram per gram body weight
MGsL	Methanolic extract of Gloriosa superba leaves
MGsS	Methanolic extract of Gloriosa superba seed
MGsT	Methanolic extract of Gloriosa superba tuber
ml	millilitre
mm	millimetre
mM	millimolar
MTP	microtiter plate

MTT	2-(4, 4-diMethyl-2-Tetrazoyl)-2, 5-diphenyl-2, 4 Tetrazolium salt
N ₂ O ₄	Dinitrogen tetroxide
N ₃ O ₄	TriNitrogen TetaOxide
Na ₂ CO ₃	Sodium carbonate
Na ₂ HPO ₄	Disodium Hydrogen Phosphate
NaCl	Sodium Chloride
NaCN	Sodium Cyanide
NADH	Nicotinamide Adenine Dinucleotide
NADPH	Nicotinamide Adenine Dinucleotide Phosphate
NaOH	Sodium Hydroxide
NBT	Nitroblue Tetrazolium
NO	Nitric oxide
NO ₂	Nitrogen dioxide
NPs	Nanoparticles
O ₂ .	Singlet oxygen
OD	Optical Density
OH.	Hydroxyl radical
ONOO.	Peroxy nitrite anion
PAHS	Polycyclic Aromatic Hydro compounds
PARP	Poly (ADP-ribose) polymerase
Pd	Palladium
PE	phycoerythrin
PEG	Polyethylene glycol
PI	Propidium iodide
PKB	protein kinase B
PLA	Poly Lactic Acid
Pt	Platinum
PUFA	Polyunsaturated fatty acids
RBC	Red Blood Cells
RGD	arginine-glycine aspartate
RI	Refractive Index
RNS	Reactive Nitrogen Species

ROS	Reactive Oxygen Species
RSS	Reactive sulfur species
SD	Standard Deviation
SEM	Scanning Electron Microscope
SO	Superoxide
SOD	Super Oxide Dismutase
Srx	Sulfiredoxin
TBA	Thio Barbituric Acid
TBARS	Thio Barbituric Acid Reactive Substances
TCA	TriChloro Acetic acid
TEM	Transmission Electron Microscope
TLC	Thin Layer Chromatography
Tris HCl	Tris Hydrochloric acid
UHMWPE	Ultra High Molecular Weight Polyethylene
w/v	Weight per volume
WHO	World Health Organization
XO	xanthine oxidase
XRD	X-RAY Diffraction analysis

Publications

- ✚ Saradha, D.M. and Annapoorani, S.(2012) Phytochemical Constituents of *Gloriosa superba* Seed, Tuber and Leaves, Research Journal of Pharmaceutical, Biological and Chemical Sciences (RJPBCS) 3(3): 111-117.
- ✚ Saradha, D.M. and Annapoorani, S. (2012) *Invitro* and *invivo* antitumourigenic activity of Methanolic extract of *Gloriosa superba* leaves and its Silver nanoparticles, Journal of Medicinal Plants Research, Ms Number : JMPR-12-1149 (Accepted for Publication).
- ✚ Saradha, D.M. and Annapoorani, S. (2013) Antioxidative activity of methanolic extract of *Gloriosa superba* Seed, Tuber and Leaves, International Journal of Pharmaceutical Research and Development (IJPRD) 5(6):102-108.

Paper Presented at Conferences

- ✚ Saradha, D.M. and Annapoorani, S.(2010) Phytochemical analysis and *invitro* free radical scavenging efficacy of *Gloriosa superba seed*”, Indian Chemical Engineering Congress (CHEMCON- 2010) on “Emergence of Biotechnology in process industry- A sustainable growth & clean world” 27th - 29th, December, 2010, Annamalai University, Chidambaram. 75 (FPT 14).
- ✚ Saradha, D.M. and Annapoorani, S. (2011) *Invitro* free radical scavenging efficacy of *Gloriosa superba tuber*”, National conference on “Recent Trends in Organic Sythesis-2011 (RTOS 11)” 24th – 26th February, 2011, Bharathidasan University, Tiruchirappalli. 152, PP-101.
- ✚ Saradha, D.M. Annapoorani,S., Poongothai,A. and K.P.Sreena., (2011) Screening of Phytochemical constituents of *Gloriosa superba tuber*, National conference on “Emerging trends in Biological Research (NCEBR,11)” 21st and 22nd February, 2011, University of Madras, Chennai. 73 (04-16).

- ✚ Saradha, D.M. Annapoorani,S., Poongothai,A. and K.P.Sreena., (2011) *In vitro* free radical scavenging efficacy of *Gloriosa superba* seed”, National Conference on “Biorevolution- A Promising Strategy, (NCBPS’11)”, Sri Krishna Arts And Science College, Coimbatore-08, 11th February,2011.
- ✚ Saradha, D.M. and Annapoorani, S. (2012) Antitumorigenic activity of methanolic extract of *Gloriosa superba* leaves, National conference on Biorevolution “A Promising Strategy,(NCBPS’12)” organized by Department of Biosciences, Sri Krishna Arts and Science College, Coimbatore-08, 8th Feb, 2012.
- ✚ Saradha, D.M. and Annapoorani, S. (2012) *In vitro* Antioxidative activity of Methanolic extract of *Gloriosa superba* leaves and its Phytochemical constituents in GC-MS, 4th Indo Korean Conference on Integrative Bioscience Research Opportunities and Challenges, organized by Faculty of Science and Home Science, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore-43, 10th and 11th Feb, 2012.
- ✚ Saradha, D.M. and Annapoorani, S. (2012) *In vitro* Antitumorigenic efficacy of silver nanoparticles of methanolic extracts of *Gloriosa superba* to DLA tumor cells”, Regional Science Congress on Science for Shaping the Future of India, organized by Indian Science Congress Association and Kongunadu Arts and Science College (Autonomous), Coimbatore-29, 15th and 16th Dec, 2012.
- ✚ Saradha, D.M. and Annapoorani, S. (2013) “Free radical scavenging efficacy and Antitumorigenic effect of silver nanoparticles of methanolic extracts of *Gloriosa superba* to DLA tumor cells”, *International Symposium on Free Radical Damage and Herbal Antioxidants in Health and Disease*, organized by Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore-43, 4th and 5th Feb, 2013.