

CONTENTS

	<i>Pages</i>
CHAPTER I: Introduction	1–58
I.1 Introduction	1
I.2 Importance of fruits for human health and disease prevention	4
I.3 Minerals	5–11
I.3.1 Macro-elements	8
I.3.2 Micro-elements	9
I.4 Vitamin C (Ascorbic acid)	11
I.5 Phytochemicals	11
I.6 Antioxidants	13
I.7 Antimicrobials	16
I.8 Amino acid	17
I.9 Anti-nutritional factors	20
I.10 Review of literature	23–37
I.11 Objectives of present study	38
References	39
CHAPTER II: Determination of proximate composition of wild edible fruits	59–79
II.1 Introduction	59
II.2 Materials and Methods	66–69
II.2.1 Chemicals	66
II.2.2 Collection and authentication of plant materials	66
II.2.3 Sample preparation	67
II.2.4 Determination of moisture content	67
II.2.5 Determination of ash content	68
II.2.6 Determination of crude fat	68
II.2.7 Determination of crude protein	68
II.2.8 Determination of crude fibre	69
II.2.9 Determination of total carbohydrate	69
II.2.10 Calorific value of fruits	69
II.2.11 Statistical analysis	69

II.3 Results and Discussion	71–74
II.4 Conclusion	74
References	75
CHAPTER III: Determination of metal contents of wild edible fruits	80–89
III.1 Materials and Methods	80–81
III.1.1 Sample preparation	80
III.1.2 Determination of metal contents	81
III.1.3 Statistical analysis	81
III.2 Results and Discussion	81–86
III.3 Conclusion	86
References	87
CHAPTER IV: Study of phytochemicals and antioxidant properties of wild fruits	90–128
IV.1 Materials and Methods	91–97
IV.1.1 Chemicals	91
IV.1.2 Sample preparation	91
IV.1.3 Preliminary phytochemical screening	91–94
IV.1.4 Determination of antioxidant properties	94–96
IV.1.4.1 DPPH free radical scavenging assay	94
IV.1.4.2 ABTS free radical scavenging assay	95
IV.1.4.3 H ₂ O ₂ radical scavenging assay	95
IV.1.4.4 FRAP antioxidant assay	96
IV.1.5 Determination of total phenolic content (TPC)	96
IV.1.6 Determination of total flavonoid content (TFC)	96
IV.1.7 Determination of vitamin C	97
IV.1.8 Statistical analysis	97
IV.2 Results and Discussion	97–119
IV.2.1 Phytochemical screening	97–99
IV.2.2 Antioxidant property	106–113
IV.2.2.1 DPPH free radical scavenging activity	106
IV.2.2.2 ABTS free radical scavenging assay	108

IV.2.2.3 H ₂ O ₂ scavenging assay	111
IV.2.2.4 Ferric reducing antioxidant power (FRAP) assay	113
IV.2.3 Total phenolic and flavonoid contents	114–117
IV.2.4 Evaluation of vitamin C contents	117-118
IV.3 Pearson's correlation study	119
IV.4 Conclusion	119
References	121
CHAPTER V: Study of antimicrobial property of wild fruits	129–142
V.1 Materials and Methods	129–131
V.1.1 Materials	129
V.1.2 Preparation of fruit extracts	130
V.1.3 Bacterial strains	130
V.1.4 Inoculums preparation	130
V.1.5 Determination of antibacterial activity of fruit extracts	130
V.1.6 Determination of minimum inhibitory concentration (MIC)	131
V.1.7 Determination of minimum bactericidal concentration (MBC)	131
V.2 Results and Discussion	136–140
V.3 Conclusion	140
References	141
CHAPTER VI: Amino acid analysis of wild fruits	143–153
VI.1 Materials and Methods	143–144
VI.1.1 Sample preparation	143
VI.1.2 Amino acid analysis	143
VI.2 Results and Discussion	144–151
VI.3 Conclusion	151
References	152
CHAPTER VII: Study of anti-nutritional factors of wild fruits	154–164
VII.1 Materials and Methods	154–157
VII.1.1 Materials	154
VII.1.2 Sample preparation	155

VII.1.3 Determination of oxalate	155
VII.1.4 Determination of tannin	155
VII.1.5 Determination of phytate	155
VII.1.6. Determination of saponin	156
VII.1.7 Determination of alkaloid	156
VII.1.8 Statistical analysis	157
VII.2 Results and Discussion	157–160
VII.3 Conclusion	160
References	162
Conclusion	165-167
Publications	168

Abbreviations

L	=	litre
d	=	day
mg	=	milligram
mg/day	=	milligram per day
mg/kg/day	=	milligram per kilogram per day
g	=	gram
kg	=	kilogram
μg	=	microgram
μL	=	microlitre
mL	=	millilitre
nm	=	nanometre
kcal	=	kilocalorie
mM	=	millimolar
mmol	=	millimole
μmol	=	micromole
μM	=	micromolar
μm	=	micrometre
mm	=	millimetre
N	=	normality
M	=	molarity
viz.	=	namely
h	=	hour
min	=	minute
sec	=	second
$^{\circ}\text{C}$	=	degree Celsius
%	=	percentage
K	=	potassium
Ca	=	calcium
Mg	=	magnesium
Na	=	sodium
Mg	=	magnesium

Mn	=	manganese
Cu	=	copper
Zn	=	zinc
Fe	=	iron
Al	=	aluminium
Ba	=	barium
Li	=	lithium
Ni	=	nickel
Ti	=	titanium
Sr	=	strontium
Cr	=	chromium
Cd	=	cadmium
Pb	=	lead
DNA	=	deoxyribonucleic acid
RNA	=	ribonucleic acid
ICP-MS	=	inductively coupled plasma mass spectrometry
HCN	=	hydrogen cyanide
USDA	=	United States Department of Agriculture
CHD	=	coronary heart disease
LDL	=	low density lipoprotein
ATP	=	adenosine triphosphate
DPPH	=	2,2-diphenyl-1-picrylhydrazyl
ABTS	=	2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid)
FRAP	=	ferric reducing anti-oxidant power
TE	=	trolox equivalent
DE	=	dry extract
DW	=	Dry weight
FW	=	fresh weight
GAE	=	gallic acid equivalent
AEAC	=	ascorbic acid equivalent antioxidant content
TEAC	=	Trolox equivalent antioxidant capacity
EAA	=	Essential amino acid

FAO	=	Food and Agriculture Organization
WHO	=	World Health Organization
UNU	=	United Nations University
AOAC	=	Association of Official Analytical Chemists
MTCC	=	Microbial type culture collection
PITC	=	Phenyl isothiocyanate
HPLC	=	High-performance liquid chromatography

List of Tables	Pages
Table I.1 : Recommended dietary allowances for total water and macronutrients	3
Table I.2 : Recommended dietary allowances of elements	7
Table I.3 : Important classes of phenolic compounds in plants	13
Table I.4 : Essential amino acid requirements for adult (FAO/WHO/UNU 2007)	18
Table I.5 : Reported wild fruits with biological properties	35
Table II.1 : Wild edible fruits collected for the study	67
Table II.2 : Proximate composition of wild edible fruits per 100 g of DW	70
Table III.1 : Macro-element analysis of wild fruits (mg/100 g DW)	82
Table III.2 : Micro-element analysis of wild fruits (mg/100 g DW)	82
Table IV.1 : Phytochemical screening of freeze dried fruit extracts of <i>G. sapida</i>	99
Table IV.2 : Phytochemical screening of freeze dried fruit extracts of <i>E. operculata</i>	100
Table IV.3 : Phytochemical screening of freeze dried fruit extracts of <i>A. dioica</i>	101
Table IV.4 : Phytochemical screening of freeze dried fruit extracts of <i>A. bunius</i>	102
Table IV.5 : Phytochemical screening of freeze dried fruit extracts of <i>O. alismoides</i>	103
Table IV.6 : DPPH free radical scavenging activity of methanolic extract of wild fruits	105
Table IV.7 : ABTS radical scavenging activity of methanolic extract of wild fruits	107
Table IV.8 : Hydrogen peroxide scavenging activity of methanolic extract of wild fruits	110
Table IV.9 : FRAP value, TPC, TFC and vitamin C content of wild fruits	114
Table IV.10 : Pearson's correlation coefficients of antioxidant activities (DPPH, ABTS, H ₂ O ₂ , FRAP), TPC, TFC and vitamin C content in the wild fruits	119

Table V.1	: Antibacterial activity of methanol extracts of fruits against <i>B. cereus</i>	137
Table V.2	: Antibacterial activity of methanol extracts of fruits against <i>S. aureus</i>	137
Table V.3	: Antibacterial activity of methanol extracts of fruits against <i>E. coli</i>	138
Table V.4	: Antibacterial activity of methanol extracts of fruits against <i>P. vulgaris</i>	138
Table VI.1	: Amino acid profiles of amino acid standard (mixture)	145
Table VI.2	: Amino acid profiles of five wild edible fruits in % of total amino acids	148
Table VI.3	: Amino acid score of five wild edible fruits based on FAO/WHO/UNU (2007) consultation pattern	149
Table VII.1	: Anti-nutritional parameters of wild fruits in mg/g dried sample	158

<i>List of Figures</i>	<i>Pages</i>
Fig. II.1a : <i>Grewia sapida</i> plant	61
Fig. II.1b : <i>Grewia sapida</i> fruit	61
Fig. II.2a : <i>Eugenia operculata</i> plant	62
Fig. II.2b : <i>Eugenia operculata</i> fruit	62
Fig. II.3a : <i>Aporosa dioica</i> plant	63
Fig. II.3b : <i>Aporosa dioica</i> fruit	63
Fig. II.4a : <i>Antidesma bunius</i> plant	64
Fig. II.4b : <i>Antidesma bunius</i> fruit	64
Fig. II.5a : <i>Ottelia alismoides</i> plant	65
Fig. II.5b : <i>Ottelia alismoides</i> fruit	65
Fig. II.6 : Proximate composition in five wild edible fruits per 100 g of DW	71
Fig. II.7 : Total carbohydrate content in five wild edible fruits per 100 g of DW	72
Fig. III.1 : Variation of macro-elements in wild edible fruits	83
Fig. III.2 : Variation of micro-elements in wild edible fruits	83
Fig. IV.1 : Plot of inhibition (%) against extract concentration for calculation ($\mu\text{g/mL}$) of IC_{50} value of DPPH	104
Fig. IV.2 : Plot of inhibition (%) against extract concentration and standard for ABTS assay	109
Fig. IV.3 : Plot of inhibition (%) against extract concentration and standard for H_2O_2 assay	112
Fig. IV.4 : Trolox standard curve for FRAP assay	113
Fig. IV.5 : Gallic acid standard curve for determination of total phenolic content	114
Fig. IV.6 : Quercetin standard curve for determination of total flavonoid content	115
Fig. IV.7 : Bar diagram showing the total phenolic and flavonoid contents in the fruits	115
Fig. IV.8 : Variation of vitamin C contents of fresh sample of five wild edible fruits	118

Fig. V.1	: Antibacterial activity of methanol extracts of fruits against <i>B. cereus</i>	132
Fig. V.2	: Antibacterial activity of methanol extracts of fruits against <i>S. aureus</i>	133
Fig. V.3	: Antibacterial activity of methanol extracts of fruits against <i>E. coli</i>	134
Fig. V.4	: Antibacterial activity of methanol extracts of fruits against <i>P. vulgaris</i>	135
Fig. VI.1	: HPLC chromatogram of acidic and basic amino acid mixture standard	144
Fig. VI.2	: HPLC chromatogram of <i>G. sapida</i> fruit	145
Fig. VI.3	: HPLC chromatogram of <i>O. alismoides</i> fruit	146
Fig. VI.4	: HPLC chromatogram of <i>A. dioica</i> fruit	146
Fig. VI.5	: HPLC chromatogram of <i>A. bunius</i> fruit	147
Fig. VI.6	: HPLC chromatogram of <i>E. operculata</i> fruit	147
Fig. VII.1	: Standard curve of catechin for determination of tannin	158
Fig. VII.2	: Standard curve of phytic acid for determination of phytate	158
Fig. VII.3	: Variation of anti-nutritional contents in mg/g of dried sample	159