CHEMICAL CHARACTERISTICS OF NON-EDIBLE COCOA PLANT (Theobroma cacao L.) PARTS AND THEIR ANTIPROLIFERATIVE ACTIVITY AGAINST VARIOUS CANCER CELL LINES

ZAINAL BIN BAHARUM

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By

ZAINAL BIN BAHARUM

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfilment of the Requirements for the Degree of Doctor of Philosophy

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of requirements for the degree of Doctor of Philosophy

CHEMICAL CHARACTERISTICS OF NON-EDIBLE COCOA PLANT (*Theobroma cacao* L.) PARTS AND THEIR ANTIPROLIFERATIVE ACTIVITY AGAINST VARIOUS CANCER CELL LINES

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May 2016

Chairman : Associate Professor Abdah Md. Akim, PhD
Faculty : Medicine and Health Sciences

Plants have been a good source of therapeutic agents for thousands of years and an impressive number of modern drugs used for treating cancer diseases are derived from natural sources. Due to its antioxidant properties, which are related to potential anti-cancer effects, the *Theobroma cacao* (*T. cacao*) has recently garnered increasing attention and become the subject of research. In the present study, *in vitro* screening was performed to investigate the antiproliferative activity of fresh non-edible cocoa plant parts such as the cocoa leaf (CL), cocoa bark (CB), cocoa husk (CH), unfermented cocoa shell (UFCS), fermented coco shell (FCS), coco root (CR), cocoa cherelle (CC) and cocoa pith (CP) against several cancer cell lines. Based on the 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium (MTT) assay results, the CL extract had strong antiproliferative activity against breast cancer (MCF-7) cells and its median inhibitory concentration (IC_{50}) was 41.43 ± 3.26 µg/mL as compared to the other extracts and cell lines. While, antioxidant activity was determined using the 2,2-diphenyl-2-picrylhydrazyl (DPPH), thiobarbituric acid–reactive substances (TBARS) and Folin-Ciocalteu assays. The CR extract had the highest antioxidant activity; its median effective dose (EC_{50}) was 358.3 ± 7.0 µg/mL and total phenolic content was 22.0 ± 1.1 g gallic acid equivalents (GAE)/100 g extract as compared to the other extracts. Only CC extract demonstrated 10.4% ± 1.1 inhibitions in the lipid peroxidation assay. For the phytochemical screening, all extracts showed the presence of saponins, flavonoids, condensed tannins, triterpenes and phytosterols, but no alkaloids or hydrolysable tannins.

The CL methanolic extract was purified and identified using bioassay-guided fractionation and gas chromatography-mass spectrometry (GC-MS), respectively. Successive extraction (partitioning) of the CL extract was carried out in hexane followed by dichloromethane and methanol. The CL hexane partitioned fraction showed the highest antiproliferative activity against MCF-7 cells, with an IC_{50} of 66.7 ± 7.95 µg/mL and further purified using column chromatography, which was divided into 3 steps: fractionation, sub-fractionation and sub-sub-fractionation. The

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CL sub-fraction (II/SF7) was selected for GC-MS chemical characterisation because it had the highest anti-cancer activity, with an IC_{50} of 6.36 ± 0.71 µg/mL, and it generated 9 major active compounds with synergistic effects against MCF-7 cells. Eight compounds were known compounds and one unidentified compound was designated ZNL-UPM/MCB-1. The present study marks the first time CL sub-fraction (II/SF7) from *T. cacao* leaf extract has been investigated for its antiproliferative activity and apoptotic effect on MCF-7 cells. The MTT assay detected optimum antiproliferative activity in the cells after 48 hours treatment with CL sub-fraction (II/SF7). This sub-fraction exerted significant dose-dependent growth inhibitory effects on the cells by inducing apoptosis, demonstrated by the formation of apoptotic bodies, fragmented DNA ladder, and disruption of the mitochondrial membrane potential. The apoptosis induced by CL sub-fraction (II/SF7) was observed via caspase-3, caspase-8, and caspase-9 activation, indicating involvement of the intrinsic and extrinsic apoptotic pathways. Meanwhile, 3 genes in the MCF-7 cells, namely DDIT3, GADD45G and HRK, showed significant changes in expression after treatment with CL sub-fraction (II/SF7) at the IC_{75} (45.0 µg/mL) as compared with untreated cells and the IC_{50} (6.4 µg/mL). The post-treatment fold changes in DDIT3, GADD45G and HRK expression were 3.8, 6.73, and 2.01, respectively. However, the expression levels of some well-known apoptosis-related genes did not show changes. Further, the genes found differentially expressed in the MCF-7 cells could be closely related to apoptosis. Given the overall high IC_{50} against the normal liver WRL-68 cell line, this study showed that the active methanolic extract of *T. cacao* leaf had cytotoxic effect on cancer cells, but not on normal cells. The findings determine CL sub-fraction (II/SF7) from the *T. cacao* has significant potential for breast cancer chemoprevention and can be developed as an anti-cancer agent in the future. The unidentified active compound will be characterized in future studies.

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Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk Ijazah Doktor Falsafah

PENCIRIAN KIMIA BAGI BAHAGIAN POKOK KOKO YANG TIDAK BOLEH DIMAKAN (*Theobroma cacao* L.) DAN AKTIVITI ANTIPROLIFERATIF MEREKA MELAWAN PELBAGAI SEL-SEL KANSER

Oleh

ZAINAL BIN BAHARUM

Mei 2016

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Fakulti : Perubatan dan Sains Kesihatan

Tumbuh-tumbuhan telah menjadi satu sumber yang berguna sebagai agen-agen terapeutik selama beribu-ribu tahun dan merupakan satu jumlah yang membanggakan bagi perubatan moden yang digunakan untuk merawat penyakit-penyakit kanser yang berasal daripada sumber-sumber semula jadi. Oleh kerana ciri-ciri antioksidannya, yang berkaitan dengan potensi sebagai kesan anti-kanser, *Theobroma cacao* (*T. cacao*) baru-baru ini telah mendapat perhatian yang semakin meningkat dan menjadi subjek dalam penyelidikan. Dalam kajian ini, penyaringan secara *in vitro* telah dilakukan untuk menilai aktiviti antiproliferatif bahagian-bahagian pokok koko segar seperti daun koko (CL), kulit koko (CB), kulit buah koko (CH), kulit biji koko yang tidak difermentasi (UFCS), kulit biji koko yang difermentasi (FCS), akar koko (CR), putik koko (CC) dan empulur koko (CP) terhadap beberapa sel-sel kanser. Berdasarkan keputusan ujian 3- [4,5-dimethylthiazol-2-YL] -2,5-diphenyltetrazolium (MTT), ekstrak CL menunjukkan aktiviti antiproliferatif yang kuat terhadap sel-sel kanser payudara (MCF-7) dan kepekatan perencatan median (IC_{50}) ialah 41.43 ± 3.26 μg/mL berbanding dengan ekstrak-ekstrak dan sel-sel yang lain. Manakala, aktiviti antioksidan telah ditentukan dengan menggunakan ujian 2,2-difenil-2-picrylhydrazyl (DPPH), bahan-bahan asid-reaktif thiobarbiturik (TBARS) dan Folin-Ciocalteu. Ekstrak CR mempunyai aktiviti antioksidan yang paling tinggi; dos berkesan median (EC_{50}) ialah 358,3 ± 7.0 μg/mL dan jumlah kandungan fenolik ialah 22.0 ± 1.1 g setara Gallic asid (GAE)/100 g ekstrak berbanding dengan ekstrak-ekstrak lain. Walaubagaimanapun, hanya ekstrak CC sahaja menunjukkan 10.4% ± 1.1 perencatan dalam ujian peroksidaan lipid. Untuk penyaringan fitokimia pula, semua ekstrak telah menunjukkan kehadiran saponin, flavonoid, tanin pekat, triterpenes dan fitosterol, tetapi tiada alkaloid atau tanin terhidrolisis.

Ektrak metanol CL telah ditulenkan dan dikenalpasti menggunakan bioesei-berpandu pemeringkatan dan kromatografi gas-spektrometri massa (GC-MS). Pengekstrakan

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berturut-turut (pembahagian) bagi ekstrak CL telah dijalankan dalam heksana diikuti oleh diklorometana dan metanol. Pecahan bahagian heksana CL telah menunjukkan aktiviti antiproliferatif yang tertinggi terhadap sel-sel MCF-7, dengan IC_{50} iaitu 66.7 ± 7.95 μg/mL dan seterusnya ditulenkan menggunakan kromatografi turus, yang dibahagikan kepada 3 langkah: pemeringkatan, sub-pemeringkatan dan sub-sub-pemeringkatan. CL sub-pecahan (II/SF7) telah dipilih untuk pencirian kimia menggunakan GC-MS kerana ia mempunyai aktiviti anti-kanser yang paling tinggi, dengan IC_{50} iaitu 6.36 ± 0.71 μg/mL dan ia menjana 9 sebatian bahan aktif utama dengan kesan sinergi terhadap sel-sel MCF-7. Lapan sebatian yang dikenali dan satu sebatian yang tidak dikenali ditandakan sebagai ZNL-UPM/MCB-1. Kajian ini merupakan kali pertama bagi CL sub-pecahan (II/SF7) dari ekstrak (*T. cacao*) dinilai untuk aktiviti antiproliferatif dan kesan apoptotik pada sel-sel MCF-7. Seterusnya, ujian MTT telah mengesan aktiviti antiproliferatif yang optima dalam sel-sel tersebut selepas rawatan selama 48 jam dengan CL sub-pecahan (II/SF7). Sub-pecahan yang digunakan tersebut mempunyai kesan yg signifikasi terhadap kesan perencatan pertumbuhan bagi kebergantungan dos ke atas sel-sel dengan merangsang apoptosis yang ditunjukkan melalui pembentukan pecahan-pecahan apoptotik, pecahan penunjuk saiz DNA dan gangguan potensi membran mitokondria. Apoptosis yang dicetuskan melalui CL sub-pecahan (II/SF7) telah diperhatikan juga melalui pengaktifan caspase-3, caspase-8, dan caspase-9, dimana menunjukkan penglibatan tapak jalan apoptotik secara intrinsik dan ekstrinsik. Sementara itu, 3 gen dalam sel-sel MCF-7, iaitu DDIT3, GADD45G dan HRK telah menunjukkan perubahan ketara dalam pengekspresi selepas rawatan dengan CL sub-pecahan (II/SF7) iaitu IC_{75} (45.0 μg/mL) berbanding dengan sel-sel yang tidak dirawat dan IC_{50} (6.4 μg/mL). Pasca-rawatan bagi perubahan berganda dalam ekspresi DDIT3, GADD45G dan HRK masing-masing adalah 3.8, 6.73 dan 2.01. Walaubagaimanapun, paras ekspresi bagi beberapa gen yang dikenali berkaitan apoptosis tidak menunjukkan sebarang perubahan. Seterusnya, didapati ada gen-gen telah mengekspresi secara berbeza dalam sel-sel MCF-7 berkemungkinan berkait rapat dengan apoptosis. Memandangkan IC_{50} mempunyai nilai yang tinggi terhadap sel hati normal (WRL-68), kajian ini menunjukkan bahawa ekstrak aktif metanol bagi daun *T. cacao* mempunyai kesan sitotoksik terhadap sel-sel kanser, tetapi tidak pada sel-sel normal. Hasil kajian menunjukkan bahawa CL sub-pecahan (II/SF7) dari *T. cacao* mempunyai potensi yang signifikasi sebagai kimopreventif bagi kanser payudara dan boleh dibangunkan sebagai agen anti-kanser pada masa hadapan. Sebatian aktif yang tidak dikenali akan dijalankan penciriannya dalam kajian pada masa hadapan.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Doctor of Philosophy. The members of the Supervisory Committee were as follow:

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Declaration by Members of Supervisory Committee

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LIST OF ABBREVIATIONS

%	Percentage
>	Greater than
≥	Greater than or equal to
>	Less than
≤	Less than or equal to
ΔΔCt	Double delta Ct
°C	Celcius
±	Approximately or about
µg	Microgram
µl	Microliter
µm	Micrometer
µM	Micromolar
A549	Adenocarcinomic human alveolar basal epithelial cells
AA	Ascorbic acid
AA	Acrylamide
Abs	Absorbance
ABTS	2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid
ACF	Aberrant crypt foci
ACTB	Actin, beta
AD	Alzheimer's disease
ADP-Ribose	Protein alpha-fodrin, poly
AIF	Apoptosis-inducing factor
Akt	Protein kinase
AKT1	V-Akt murine thymoma viral oncogene homolog 1
ALS	Amyotrophic lateral sclerosis
AMPA	α-Amino-3-hydroxy-5-methyl-4-isoxazolepropionate
ANOVA	Analysis of variance
AO	Acridine orange
AP15	Apoptosis inhibitor 5
Apaf-1	Apoptotic protease activation factor-1
AR	Androgen receptor
ASTM	The American Standard Test Sieve Series
ATCC	American Type Culture Collection
ATP	Adenosine triphosphate
B2M	Beta-2-microglobulin
BAD	BCL2-associated agonist of cell death
BAG1	BCL2-associated athanogene
BAG3	BCL2-associated athanogene 3
BAG4	BCL2-associated athanogene 4
Bak	Bcl-2 antagonist killer
BAK1	BCL2-antagonist/Killer 1
BALB/C	Albino, laboratory-bred strain of the house mouse
BAX	BCL2-associated X protein
BBB	Blood-brain barrier
BBC3	BCL2-binding component 3
BC	Before Christ

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BCL10	B-cell CLL/Lymphoma 10
BCL2	B-cell CLL/Lymphoma 2
BCL2A1	BCL2-related protein A1
BCL2L1	BCL2-like 1
BCL2L11	BCL2-like 11 (apoptosis facilitator)
BCL2L14	BCL2-like 14 (apoptosis facilitator)
BCL2L2	BCL2-like 2
BCL6	B-cell CLL/Lymphoma 6
BCL-XL	B-cell lymphoma-extra large
BFAR	Bifunctional apoptosis regulator
BID	BH3 interacting domain death agonist
BIK	BCL2-interacting killer (apoptosis-inducing)
BIRC2	Baculoviral IAP repeat-containing 2
BIRC3	Baculoviral IAP repeat-containing 3
BIRC4	Baculoviral IAP repeat-containing 4
BIRC5	Baculoviral IAP repeat-containing 5
BNIP1	BCL2/adenovirus E1B 19 kDa interacting protein 1
BNIP2	BCL2/adenovirus E1B 19 kDa interacting protein 2
BNIP3	BCL2/adenovirus E1B 19 kDa interacting protein 3
BNIP3L	BCL2/adenovirus E1B 19 kDa interacting protein 3-like
BOK	BCL2-related ovarian killer
BP	Blood pressure
bp	Base pair
BRCA 1	Breast cancer type 1 susceptibility protein
BRCA 2	Breast cancer type 2 susceptibility protein
BSA	Bovine serum albumin
C	Concentration
Ca2+	Calcium ion
CAD	Caspase-activated DNase
CARD	Caspase recruitment domain
CARD10	Caspase recruitment domain family, member 10
CASP1	Caspase-1, cysteine aspartic acid-specific protease 1
CASP11	Caspase-11, cysteine aspartic acid-specific protease 11
CASP12	Caspase-12, cysteine aspartic acid-specific protease 12
CASP14	Caspase-14, cysteine aspartic acid-specific protease 14
CASP2	Caspase-2, cysteine aspartic acid-specific protease 2
CASP3	Caspase-3, cysteine aspartic acid-specific protease 3
CASP4	Caspase-4, cysteine aspartic acid-specific protease 4
CASP5	Caspase-5, cysteine aspartic acid-specific protease 5
CASP6	Caspase-6, cysteine aspartic acid-specific protease 6
CASP7	Caspase-7, cysteine aspartic acid-specific protease 7
CASP8	Caspase-8, cysteine aspartic acid-specific protease 8
CASP9	Caspase-9, cysteine aspartic acid-specific protease 9
Caspase	Cysteine aspartic acid-specific protease
CAT	Catalase
CB	Cocoa bark
CC	Cocoa cherelle
CD40	CD40 molecule, TNF receptor superfamily member 5
CD95	APO-1/Fas

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CDC	Cell division cycle
Cdc2	Cell division cycle 2
CDK	Cyclin-dependent kinase
CDKI	Cyclin-dependent kinase inhibitor
cDNA	Complementary deoxyribonucleic acid
C-erbb2	v-erb-b2 avian erythroblastic leukemia viral oncogene homolog 2
C-fos	FBJ murine osteosarcoma viral oncogene homolog
CFU	Colony-forming unit
CH	Cocoa husk
CL	Cocoa leaf
cm^2	Square centimetre
C-myc	v-myc avian myelocytomatosis viral oncogene homolog
CNS	Central nervous system
CO_2	Carbon dioxide
CP	Cocoa pith
CPE	Cocoa phenolic extract
CR	Cocoa root
CSF	Cerebrospinal fluid
CSF	Cocoa shell fermented
CSUF	Cocoa shell unfermented
Ct	Cycle threshold
Cu	Copper
Cyt C	Cytochrome C
DAD1	Defender against cell death 1
DAP1	Death-associated protein 1
DAPK1	Death-associated protein kinase 1
DAPK2	Death-associated protein kinase 2
dATP	Deoxyadenosine triphosphate
DBP	Diastolic blood pressure
DBTRG-05MG	Glioblastoma
DCF	Detannified cocoa
DCM	Dichloromethane
DDIT3	DNA damage-inducible transcript 3
DED	Death effector domain
DEN	Diethylnitrosamine
DEPC	Diethylpyrocarbonate
DEVD	Amino acid sequence Asp-Glu-Val-Asp
DIABLO	Diablo, IAP-binding mitochondrial protein
DIM	3,3′-diindolylmethane
DISC	Death-inducing signalling complex
DMEM	Dulbecco's modified eagle's medium
DMSO	Dimethyl sulfoxide
DNA	Deoxyribonucleic acid
DNase	Deoxyribonuclease
dNTP	Nucleotide triphosphates containing deoxyribose
DPF2	D4, zinc and double PHD fingers family 2
DPPH	1,1'-diphenyl-2-picrylhdrazyl
DTNB	5,5'-Dithiobis-2-nitrobenzoic acid

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E	Efficiency
E2F1	E2F transcription factor 1
E2F2	E2F Transcription factor 2
EC	(-)-epicatechin
EC_{50}	Effective concentration at 50 percent
EDTA	Ethylenediamine tetraacetic acid
EGCG	(−)-epigallocatechin-3-gallate
ELISA	Enzyme-linked immunosorbent assay
Endo G	Endonucleases G
ER	Estrogen receptor
EtOAC	Ethyl acetate
F	Fraction
FADD	Fas-associated protein with death domain
FAS	Fas cell surface death receptor
Fas/CD95	Cell death signalling receptor
FasL	Fas ligand
FASLG	Fas ligand (TNF superfamily, member 6)
FBS	Fetal bovine serum
FCC	Flash column chromatography
FCR	Folin-Ciocalteu Reagent
Fe^{2+}	Ferric (II) ion
$FeCl_3$	Ferric chloride
FLICE	FADD-like interleukin-1 beta-converting enzyme
FOXO3	Forkhead box O3
FRAP	Ferric reducing antioxidant power
FRET	Fluorescence resonance energy transfer
FRIM	Forest Research Institute of Malaysia
FTIR	Fourier transform infrared spectroscopy
g	Gram
g	Gravity
G1	Gap 1
G2	Gap 2
G2/M	Gap 2/ metaphase
GADD45G	Growth arrest and DNA damage-inducible, gamma
GAE	Gallic acid equivalent
GAPDH	Glyceraldehyde-3-phosphate dehydrogenase
GCMS	Gas chromatography mass spectroscopy
GDC	Genomic DNA control
GP	GSH peroxidase
GPR	Global pattern recognition
GPx	Glutathione peroxidase
GPX1	Glutathione peroxidase 1
GR	GSH reductase
GRx	Glutathione reductase
GSH	Glutathione
GSK3B	Glycogen synthase kinase 3 beta
GST	Glutathione S-transferase
h	Hours
H_2O	Water

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H_2O_2	Hydrogen peroxide
HCl	Hydrochloric acid
HD	Huntington's disease
HDL	High-density lipoprotein
HeLA	Helacyton gartleri
HEPES	4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid
HepG2	Hepatocellular carcinoma
Hg	Hemoglobin
HIF1A	Hypoxia-inducible factor 1, alpha subunit (basic helix-loop-helix transcription factor)
HIPK2	Homeodomain-interacting protein kinase 2
HO-	Hyrxoyl radical
HPLC	High pressure liquid chromatography
HPRT1	Hypoxanthine phosphoribosyltransferase 1
HRK	Harakiri, BCL2-interacting protein (contains only BH3 domain)
HRT	Hormone replacement theraphy
HSPA1A	Heat shock 70 kDa protein 1A
HT-29	Human colon adenocarcinoma grade II cell line
HTRA2	HtrA serine peptidase 2
i.d	Internal diameter
IAPs	Inhibitor of apoptosis proteins
IBS	Institute of Bioscience
IC_{25}	Inhibition concentration at 25 percent
IC_{50}	Inhibition concentration at 50 percent
IC_{75}	Inhibition concentration at 75 percent
ICE	IL-1β–converting enzyme
IETD	Z-IETD-fmk, caspase-8 inhibitor
IFN-γ	Interferon gamma
IGF1	Insulin-like growth factor 1 (somatomedin C)
iGluR	Ionotropic glutamate receptor
IL-1β	Interleukin-1β
IL3	Interleukin-3
IR	Infrared
IU	International unit
JC-1	Novel cationic carbocyanine dye
JNK	c-Jun N-terminal protein kinase
kbp	Kilo base pair
KCl	Potassium chloride
L	Liters
LD_{50}	Lethal dose at 50 percent of concentration
LDH	Lactate dehydrogenase
LDL	Low-density lipoprotein
LEHD	Z-LEHD-fmk, caspase-9 inhibitor
LTA	Lymphotoxin alpha
LTBR	Lymphotoxin beta receptor (TNFR superfamily, member 3)
m/z	Mass-to-charge ratio
m^2	Square meter
MAPK	Mitogen-activated protein kinase

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MBP	Myelin basic protein
MCF-7	Michigan cancer foundation – 7
MCL1	Myeloid cell leukemia sequence 1 (BCL2-related)
MCP-1	Monocyte chemoattractant protein-1
MDA	Malondialdehyde
MDA-MB-231	Mammary gland adenocarcinoma-231
MDA-MB-436	Mammary gland adenocarcinoma-436
MDA-MB-468	Mammary gland adenocarcinoma-468
MEM	Minimum essential media
MeOH	Methanol
mg	Milligram
mg/kg	Milligram per gram
mg/mL	Milligram per mililiter
$MgCl_2$	Magnesium chloride
MIC	Minimum inhibit concentration
mL	Mililiter
MLP29	Mouse liver progenitor cells
mm	Milimeter
mM	Milimolar
mmHg	Millimeter of mercury
MMP	Mitochondrial membrane potential
Mn	Manganase
MnSOD	Manganese-containing SOD
mRNA	Messenger ribonucleic acid
MS	Mass spectrometry
MST	Mean survival time
MTS	3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium
MTT	3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide
MW	Molecular weight
Na+	Sodium ion
NaCl	Sodium chloride
NADH	β-Nicotinamide adenine dinucleotide
NAIP	NLR family, apoptosis inhibitory protein
NaOH	Sodium hydroxide
NCR	National Cancer Registry
NF-κB	Nuclear factor-κB
NFKB1	Nuclear factor of kappa light polypeptide gene enhancer in B-cell 1
ng	Nanogram
ng/µl	Nanogram per microliter
NIST	National Institute Standard and Technology
nm	Nanometer
nM	Nanomolar
NMR	Nuclear magnetic resonance
NO	Nitric oxide
NOTCH	Rranslocation-associated (Drosophila)
NOX-1	NADPH oxidase-1
NRU	Neutral red uptake

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O2-	Superoxide
OD	Optical density
ONOO-	Peroxynitrite
ORAC	Oxygen radical absorbance capacity
P13K-AKT	Phosphatidylinositol-3-kinase and protein kinase B
p38-MAPK	p38 mitogen-activated-protein-kinase
p53	Tumor suppressor gene
p-AKT	Phosphate-protein kinase
PARP	Poly (ADP-ribose) polymerase
PAWR	PRKC, apoptosis, WT1, regulator
PB2	Procyanidin B2
PBS	Phosphate buffered saline
PCR	Polymerase chain reaction
PERP	PERP, TP53 apoptosis effector
pH	Potential of hydrogen
PI	Propidium iodide
p-JNK	Phosphate-c-Jun-N-terminal protein kinase
PKA	Protein Kinase
PNS	Peripheral nervous system
PRDX2	Peroxiredoxin 2
PS	Phosphatidylserine
PTEN	Phosphatase and tensin homolog
PTP	Permeability transition pore
PUFA	Polyunsaturated fatty acids
qPCR	Quantitative polymerase chain reaction
r	Correlation coefficient
RAD21	RAD21 homolog (Schizosaccharomyces pombe)
RAW264.7	Murine macrophage cell line
RES	Resazurin reduction
RF	Retention factor
RIP	Receptor-interacting protein
RIP1	Receptor-interacting protein 1
RIP3	Receptor-interacting protein 3
RIPK1	Receptor-interacting serine/threonine-protein kinase 1
RIPK3	Receptor-interacting serine/threonine-protein kinase 3
RN18S1	RNA, 18S ribosomal 1
RNA	Ribonucleic acid
RNase	Ribonuclease
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
RPL13A	Ribosomal protein L13a
rpm	Revolutions per minute
RPMI	Roswell Park Memorial Institute medium
rRNA	Ribosomal RNA
RT	RNA reverse transcription controls
RT-PCR	Real-time polymerase chain reaction
RT-qPCR	Real time-quantitative polymerase chain reaction
s	Seconds
SBP	Systolic blood pressure

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SD	Standard deviation
SDS	Sodium dodecyl sulphate
Se	Selenium
SEM	Standard error of the mean
SF	Sub-fraction
SIRT1	Sirtuin 1
SKRB-3	Retinoid receptor-positive breast cancer cell line
SOD	Superoxide dismutase
SOD1	Superoxide dismutase 1
SPSS	Statistical Package for the Social Sciences
SRB	Sulforhodamine B
SSF	Sub-sub-fraction
STAT5A	Signal transducer and activator of transcription 5A
STAT5B	Signal transducer and activator of transcription 5B
SWNCT	single-walled carbon nanotubes
SYBR Green	Asymmetrical cyanine dye
t	Time
TAE	Tris-acetate-EDTA
TBA	Thiobarbituric acid
TBARS	Thiobarbituric acid-reactive substances
TBE	Tris-borate-EDTA
TCA	Trichloroacetic acid
TCA cycle	Tricarboxylic acid cycle
TCF	Tannin rich cocoa fraction
TGs	Triacylglycerols
TLC	Thin layer chromatography
TNB	5-thionitrobenzoic acid
TNF	Tumor necrosis factor
TNF-α	Tumor necrosis factor-α
TNFAIP3	Tumor necrosis factor, alpha-induced protein 3
TNFR	Tumor necrosis factor family receptors
TNFRSF10	Tumor necrosis factor receptor superfamily
TNFRSF10A	Tumor necrosis factor receptor superfamily, member 10A
TNFRSF11B	Tumor necrosis factor receptor superfamily, member 11B
TNFRSF1A	Tumor necrosis factor receptor superfamily, member 1A
TNFRSF1B	Tumor necrosis factor receptor superfamily, member 1B
TP53	Tumor protein P53
TP53INP1	Tumor protein P53 inducible nuclear protein 1
TPC	Total phenolic contents
TPmg	Total phosphate miligram
TRADD	TNFRSF1A-associated via death domain
TRAF	TNFR-associated factor
TRAF2	TNF receptor-associated factor 2
TRAF4	TNF receptor-associated factor 4
TRAIL	TNF-related apoptosis inducing ligand
Trolox	(±)-6-Hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid
TUNEL	TdT-mediated dUTP nick-end-labeling
U	Units
UK	United Kingdom

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UNC5B	Netrin receptor Unc-5 homolog B (Caenorhabditis elegans)
UPM	University Putra Malaysia
USA	United States of America
UV	Ultraviolet
UV-VIS	Ultraviolet-visible
V	Volt
v/v	Volume per volume
w/v	Weight per volume
WHO	World Health Organization
WNT	Wingless-type MMTV integration site family
WRL-68	Hepatic human cell line
WST	Water soluble Tetrazolium salts
XIAP	X-linked inhibitor of apoptosis
XTT	Sodium 2,3,-bis(2-methoxy-4-nitro-5-sulfophenyl)-5-[(phenylamino)-carbonyl]-2H-tetrazolium inner salt
Zn	Zink
µg	Microgram
µL	Microliter
µM	Micromolar

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CHAPTER 1

INTRODUCTION

1.1 Background of study

Cancer is a complicated and multifactorial illness characterised by the unusual development of cells. The development of cancer cell formation typically occurs via a variety of genetic and epigenetic changes (Giri et al., 2006; Mbaveng et al., 2011). Cancer occurs where there is a net accumulation of aberrant abnormal cells arising from overproliferation, low apoptosis or a combination of both (Abdul et al., 2009). Hameroff, (2004) reported on the theories of cancer origin, where malignant cells divide and multiply uncontrollably. Cancer is likewise brought about by adjustments in tumour suppressor genes, oncogenes and microRNA genes (Croce, 2008). In addition, difficulty is often encountered when treating cancer due to drug resistance, toxicity, and low cancer drug specificity (de Mesquita et al., 2009). Cancer is a serious burden to the public health system and remains a challenge for scientists to solve.

As reported by Rajesh et al. (2011), cancer was expected to claim 9 million lives worldwide by 2015. In Peninsular Malaysia, the most 5 prevalent cancers such as breast, colorectal, lung, cervical and nasopharyngeal cancer were diagnosed among Malaysians in 2007 and around 21,773 cancer cases were reported (National Cancer Registry, 2011). In the past decade, natural substances have played important roles as cancer chemoprevention and therapeutic agents (Fulda, 2010). One modality of cancer treatment is chemotherapy. About 65.0% of drugs used in chemotherapy are of natural origin (Nurhanan et al., 2008). Therapeutic plants have a long history in both traditional and present day use for cancer treatment (Conforti et al., 2008; Jain & Jain, 2011) and have been used as remedies for human diseases for centuries (Nisa et al., 2011; Sharma et al., 2011; Shoeb, 2006).

Natural products confer various beneficial effects on human health and naturally occurring bioactive substances have been reported to protect against tumorigenesis and suppress the growth of established tumours. Cancer cells have progressed to various mechanisms to protect the induction of programmed cell death, or apoptosis. It has been shown that activation of the apoptosis signalling pathways by natural active compounds constitutes a key event in these anti-cancer activities. Furthermore, about 87.0% of human diseases such as cancer, bacterial infection and immunological disorder are treated using natural active products and drugs, while over 3000 plant species producing about 25.0% of prescribed drugs worldwide have been reported to have anti-cancer characteristics. In the developing countries, about 80.0% of the population still relies on traditional medicines from natural products, especially those from plants, for their primary health care needs (Khajure & Rathod, 2011; Uddin et al., 2011). Traditional medicine from plants can also be used as

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potential sources of development of new drugs that can be used as future anti-cancer agents (Caamal-Fuentes et al., 2011).

Cocoa is the seed of the cocoa tree (*T. cacao*), an evergreen plant about 6–12 m in height. It grows at suitable altitudes of 30–300 m in areas of moderate temperatures ranging 18–32°C. *T. cacao* can grow in adequate moisture and needs about 1–5 L/m^2 rainfall per year (Weisburger, 2001). Cocoa was used by the Aztec, Olmec and Maya civilisations as a divine food and they consumed cocoa products as early as 600 BC. Several documents from the Badianus Manuscript, the Florentine Codex and the Princeton Codex mention that more than 100 medicinal benefits of cocoa have been reported regarding the bioactive compounds, chemical composition and health benefits of cocoa and other cocoa-based products which have documented over 200 scientific studies (Addai, 2010; Mhd Jalil & Ismail, 2008; Ramiro et al., 2005). It has also been reported by Jalil et al. (2008) that various parts of *T. cacao*, especially the cocoa bean, are utilised as chocolate and other non-edible cocoa plant parts such as the bark, leaf, butter, pulp and flower have been used for medicinal purposes. The Mayans and Aztecs valued the cocoa bean highly as a food and they believed that cocoa was given to them directly by Gods. The name *T. cacao*, meaning 'food of the gods' was discovered by Carl Linnaeus a Swedish botanist in 1737, when cocoa came to Europe (Bennett, 2003; Rusconi & Conti, 2010).

Recently, most research on cocoa has focused on polyphenol compounds, especially flavonoid compounds that function as strong antioxidant agents (e.g. as reducing agents, free radical scavengers, metal chelators) and their potential benefits to human health (Hii et al., 2009; Osman et al., 2004; Summa et al., 2006). Numerous studies have reported that bioactive compounds in *T. cacao*, for example, phenolic compounds have potential medical advantages for anticipating several critical diseases such as cardiovascular illness, inflammation, neurodegenerative disorders and cancer (Arlorio et al., 2009; Rimbach et al., 2009; Schinella et al., 2010). The active compounds identified from cocoa bean are epicatechin, gallocatechin, anthocyanins, epigallocatechin, procyanins, catechin, flavones, flavonol glycosides (quercetin-3-*O*-arabinoside, luteolin-7-*O*-glucoside) and others (Belščak et al., 2009; Jonfia-Essien et al., 2008).

In order to develop safe and natural anti-cancer agents from cocoa plant parts, a novel approach has to be established through the screening, purification, identification and *in vivo* evaluation of *T. cacao* tissues, taking into consideration the physical and biochemical properties of the compounds and their therapeutic effects, which will not only provide the long-expected remedy and cure for cancer and other ailments but which can also be used as the basis for screening, purifying and identifying other bioactive compounds that occur naturally in hundreds and thousands of plants across the globe, as they have enormous potential in biomedical application. Therefore, the current study proposed for eavaluation the effect of non-edible cocoa plant part methanolic extracts using *in vitro* cancer studies, which hypothesized would identify novel natural anti-cancer agents with potent bioactivity.

The current study is to investigate the potential capability of the chemical constituents of the cocoa plant part extracts, which were isolated and later identified as having potential antiproliferative activities against various human cancer cell lines. The research findings will be useful for future documentation of the active constituents intended as new chemical entities. These in turn shall form the basis for developing novel drugs and for conducting pre-clinical testing and clinical phase I and II trials in the near future.

1.2 Problem statement

Currently, the health aspect of cocoa, especially in cancer studies, has focused on the seed only and not other cocoa plant parts. The intention of the present study was to discover anti-cancer compounds from cocoa bark, leaf, husk, fermented shell, unfermented shell, pith, cherelle and root, for which the active compounds and biological activities, especially against cancer, have not been established as compared to cocoa seed. A previous study reported that most plant parts contain active compounds that are useful for treating cancer. Meanwhile, a cure has not yet been found for cancer, despite cancer being a disease of worldwide importance. The synthetic therapies in use today are not only unable to cure cancer but harmful due to the side effects.

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1.3 Research objectives

1.3.1 General objective

To identify anti-cancer properties of bioactive constituents from the non-edible plant parts of *T. cacao* against several cancer cell lines in promoting apoptosis activity and to determine their cytotoxicity against normal cell line.

1.3.2 Specific objectives

1. To investigate the antioxidant activity, total phenolic content, anti-lipid peroxidation activity and phytochemical contents in methanolic leaf, bark, pod, unfermented shell, fermented shell, root, cherelle and pith extracts of *T. cacao*.

2. To screen the antiproliferative activity of methanolic leaf, bark, pod, unfermented shell, fermented shell, root, cherelle and pith extracts of *T. cacao* against several cancer cell lines.

3. To purify and identify anti-cancer agents from the most potent antiproliferative activity of methanolic plant part extract of *T. cacao* using chromatography and spectroscopy.

4. To evaluate the cytotoxic effect of the crude extract, partitioned fraction, fraction, sub-fraction and sub-sub-fraction from most potent antiproliferative activity of methanolic plant part extract of *T. cacao* against a normal WRL-68 cell line.

5. To evaluate apoptosis in selected cancer tissue following treatment with the most potent antiproliferative activity of methanolic plant part extract of *T. cacao* via morphological, mitochondrial membrane potential, DNA fragmentation, caspases activity and human cancer gene expression assay.

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1.4 Research hypothesis

The current study should provide beneficial informations about the discovery of bioactive compounds from the *T. cacao* plant for treating various critical human cancers in Malaysia, as the plant parts investigated could have effective anti-cancer properties. Cocoa is the third largest export commodity in Malaysia, can be easily found, is used widely, especially in food and beverages and contains active compounds such as flavonoids, polyphenols and alkaloids. Moreover, the active compounds and biological activities of the abovementioned non-edible cocoa plant parts have not been established. The phytochemical properties of the non-edible cocoa plant parts are expected to eliminate cancer cells by inducing apoptosis, or programmed cell death, without affecting normal cells. Furthermore, this study would also add value to the cocoa-based products already available on the market and diversify the usage of cocoa, for example, for developing anti-cancer drugs. The present study hypothesises that non-edible cocoa plant parts can play an important role as natural anti-cancer agents by preventing cancer through apoptosis.

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REFERENCES

(EBCTCG), E. B. C. T. C. G. (2011). Relevance of breast cancer hormone receptors and other factors to the efficacy of adjuvant tamoxifen: Patient-level meta-analysis of randomised trials. *The Lancet*, *378*(9793), 771–784.

Abdul, A. B., Abdelwahab, S. I., Fong, H. K., Mohan, S. M., Al-Zubairi, A. S., & Elhassan, M. M. (2009). *In vitro* response of cancer cells to the growth-inhibitory effects of dichloromethane extract of *Goniothalamus umbrosus*. *Research Journal of Pharmacology*, *3*(1), 1–6.

Abu, M. N., Ashraf, M., Salleh, M., Hani, N., & Radzman, M. (2013). Insulin sensitivity enhancement of the mixture of *Tinospora crispa* and gelam (*Melaleuca cajuputi*) honey and its antiproliferative activity on hepatocellular carcinoma , HepG2 : A preliminary study. *Journal of Medical Research and Development*, *2*(3), 48–54.

Addai, F. K. (2010). Natural cocoa as diet-mediated antimalarial prophylaxis. *Medical Hypotheses*, *74*, 825–830.

Adibhatla, R. M., & Hatcher, J. F. (2008). Phospholipase A(2), reactive oxygen species, and lipid peroxidation in CNS pathologies. *BMB Reports*, *41*(8), 560–567.

Adi-Dako, O., Ofori-Kwakye, K., Frimpong Manso, S., Boakye-Gyasi, M. EL, Sasu, C., & Pobee, M. (2016). Physicochemical and antimicrobial properties of cocoa pod husk pectin intended as a versatile pharmaceutical excipient and nutraceutical. *Journal of Pharmaceutics*, *2016*, 1–12.

Adler, V., Schaffer, A., Kim, J., Dolan, L., & Ronai, Z. (1995). UV irradiation and heat shock mediate JNK activation via alternate pathways. *Journal of Biological Chemistry*, *270*(44), 26071–26077.

Ahmad, R., Srivastava, S. P., Maurya, R., Rajendran, S. M., Arya, K. R., & Srivastava, A. K. (2008). Mild antihyperglycaemic activity in *Eclipta alba*, *Berberis aristata*, *Betula utilis*, *Cedrus deodara*, *Myristica fragrans* and *Terminalia chebula*. *Indian Journal of Science and Technology*, *1*(5), 1–6.

Aiyegoro, O. a, & Okoh, A. I. (2010). Preliminary phytochemical screening and *in vitro* antioxidant activities of the aqueous extract of *Helichrysum longifolium* DC. *BMC Complementary and Alternative Medicine*, *10*(1), 21.

Aiyegoro, O. A., & Okoh, A. I. (2009). Phytochemical screening and polyphenolic antioxidant activity of aqueous crude leaf extract of *Helichrysum pedunculatum*. *International Journal of Molecular Sciences*, *10*(11), 4990–5001.

© COPYRIGHT UPM

Ajiboye, T. O. (2015). Standardized extract of *Vitex doniana* sweet stalls protein oxidation, lipid peroxidation and DNA fragmention in acetaminophen-induced hepatotoxicity. *Journal of Ethnopharmacology*, *164*, 273–282.

Akim, A., Ling, L. C., Rahmat, A., & Zakaria, Z. A. (2011). Antioxidant and antiproliferative activities of Roselle juice on Caov-3 , MCF-7 , MDA-MB-231 and HeLa cancer cell lines. *African Journal of Pharmacy and Pharmacology*, *5*, 957–965.

Alabsi, A. M., Ali, R., Ali, A. M., Al-Dubai, S. A. R., Harun, H., Kasim, N. H. A., & Alsalahi, A. (2012). Apoptosis induction, cell cycle arrest and *in vitro* anticancer activity of gonothalamin in a cancer cell lines. *Asian Pacific Journal of Cancer Prevention*, *13*(10), 5131–5136.

Albishi, T., John, J. a., Al-Khalifa, A. S., & Shahidi, F. (2013). Phenolic content and antioxidant activities of selected potato varieties and their processing by-products. *Journal of Functional Foods*, *5*(2), 590–600.

Allan, L. A., & Clarke, P. R. (2007). Phosphorylation of caspase-9 by CDK1/Cyclin B1 protects mitotic cells against apoptosis. *Molecular Cell*, *26*(2), 301–310.

Alon, T., & Amirav, A. (2006). Isotope abundance analysis methods and software for improved sample identification with supersonic gas chromatography/mass spectrometry. *Rapid Communications in Mass Spectrometry*, *20*(17), 2579–2588.

Al-Rashidi, W., Mat Supri, N. N., & Manshoor, N. (2011). Cytotoxic activity of crude extract from *Costus malortieanus* (Costaceae). *American-Eurasian Journal of Toxicological Sciences*, *3*(2), 63–66.

Alteri, R., Barnes, C., Burke, A., Gansler, T., Gapstur, S., Gaudet, M., … Simpson, S. (2014). Breast Cancer Facts & Figures 2013-2014. Retrieved from http://www.cancer.org/acs/groups/content/@research/documents/document/acspc-040951.pdf

Althunibat, O. Y., Hashim, R. Bin, Taher, M., Mohd. Daud, J., Ikeda, M.-A., & Zali, B. I. (2009). *In vitro* antioxidant and antiproliferative activities of three Malaysian sea cucumber species. *European Journal of Scientific Research*, *37*, 376–387.

Amaravadi, R. K., & Thompson, C. B. (2007). The roles of therapy-induced autophagy and necrosis in cancer treatment. *Clinical Cancer Research*, *13*(24), 7271–7279.

Anand, P., Kunnumakara, A. B., Sundaram, C., Harikumar, K. B., Tharakan, S. T., Lai, O. S., … Aggarwal, B. B. (2008). Cancer is a preventable disease that requires major lifestyle changes. *Pharmaceutical Research*, *25*(9), 2097–2116.

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Andres-Lacueva, C., Monagas, M., Khan, N., Izquierdo-Pulido, M., Urpi-Sarda, M., Permanyer, J., & Lamuela-Raventós, R. M. (2008). Flavanol and flavonol contents of cocoa powder products: Influence of the manufacturing process. *Journal of Agricultural and Food Chemistry*, *56*(9), 3111–3117.

Anwar, F., Kalsoom, U., Sultana, B., Mushtaq, M., Mehmood, T., & Arshad, H. A. (2013). Effect of drying method and extraction solvent on the total phenolics and antioxidant activity of cauliflower (*Brassica oleracea* L.) extracts. *International Food Research Journal*, *20*(2), 653–659.

Arbab, I. A., Abdul, A. B., Sukari, M. A., Abdullah, R., Syam, S., Kamalidehghan, B., … Mohan, S. (2013). Dentatin isolated from *Clausena excavata* induces apoptosis in MCF-7 cells through the intrinsic pathway with involvement of NF-κB signalling and G0/G1 cell cycle arrest: A bioassay-guided approach. *Journal of Ethnopharmacology*, *145*(1), 343–54.

Arikawa, E., Sun, Y., Wang, J., Zhou, Q., Ning, B., Dial, S. L., … Yang, J. (2008). Cross-platform comparison of SYBR Green real-time PCR with TaqMan PCR, microarrays and other gene expression measurement technologies evaluated in the MicroArray Quality Control (MAQC) study. *BMC Genomics*, *9*(1), 328.

Arlorio, M., Bottini, C., Travaglia, F., Locatelli, M., Bordiga, M., Coïsson, J. D., … Tessitore, L. (2009). Protective activity of *Theobroma cacao* L. phenolic extract on AML12 and MLP29 liver cells by preventing apoptosis and inducing autophagy. *Journal of Agricultural and Food Chemistry*, *57*(22), 10612–10618.

Arulvasu, C., Prabhu, D., Manikandan, R., Srinivasan, P., Dinesh, D., Babu, G., & Sellamuthu, S. (2010). Induction of apoptosis by the aqueous and ethanolic leaf extract of *Vitex negundo* L. in MCF-7 human breast cancer cells. *International Journal of Drug Discovery*, *2*, 1–7.

Arya, M., Shergill, I. S., Williamson, M., Gommersall, L., Arya, N., & Patel, H. R. H. (2005). Basic principles of real-time quantitative PCR. *Expert Review of Molecular Diagnostics*, *5*(2), 209–219.

Asadzadeh Vostakolaei, F., Broeders, M. J. M., Mousavi, S. M., Kiemeney, L. A. L. M., & Verbeek, A. L. M. (2013). The effect of demographic and lifestyle changes on the burden of breast cancer in Iranian women: A projection to 2030. *Breast*, *22*, 277–281.

Atjanasuppat, K., Wongkham, W., Meepowpan, P., Kittakoop, P., Sobhon, P., Bartlett, A., & Whitfield, P. J. (2009). *In vitro* screening for anthelmintic and antitumour activity of ethnomedicinal plants from Thailand. *Journal of Ethnopharmacology*, *123*(3), 475–82.

© COPYRIGHT UPM

Bagnardi, V., Blangiardo, M., La Vecchia, C., & Corrao, G. (2001). A meta-analysis of alcohol drinking and cancer risk. *British Journal of Cancer*, *85*(11), 1700–1705.

Bajpai, V. K., Sharma, A., Kang, S. C., & Baek, K.-H. (2014). Antioxidant, lipid peroxidation inhibition and free radical scavenging efficacy of a diterpenoid compound sugiol isolated from *Metasequoia glyptostroboides*. *Asian Pacific Journal of Tropical Medicine*, *7*(1), 9–15.

Balis, J. U., Bumgarner, S. D., Paciga, J. E., Paterson, J. F., & Shelley, S. A. (1984). Synthesis of lung surfactant-associated glycoproteins by A549 cells: Description of an *in vitro* model for human type II cell dysfunction. *Experimental Lung Research*, *6*(3-4), 197–213.

Balunas, M. J., & Kinghorn, A. D. (2005). Drug discovery from medicinal plants. In *Life Sciences*, *78*,431–441.

Barnes, M. A., McMullen, M. R., Roychowdhury, S., Pisano, S. G., Liu, X., Stavitsky, A. B., … Nagy, L. E. (2013). Macrophage migration inhibitory factor contributes to ethanol-induced liver injury by mediating cell injury, steatohepatitis, and steatosis. *Hepatology*, *57*(5), 1980–1991.

Barnhart, B. C., Alappat, E. C., & Peter, M. E. (2003). The CD95 Type I/Type II model. *Seminars in Immunology*, *15*(3), 185–193.

Bartley, B. G. D. (2005). *The genetic diversity of cacao and its utilization*. In B. G. D. Bartley (Ed.), *Production*. Wallingford: CABI.

Bates, S. E., Valverius, E. M., Ennis, B. W., Bronzert, D. A., Sheridan, J. P., Stampfer, M. R., … Dickson, R. B. (1990). Expression of the transforming growth factor-α/epidermal growth factor receptor pathway in normal human breast epithelial Cells. *Endocrinology*, *126*(1), 596–607.

Becker, E. M., Nissen, L. R., & Skibsted, L. H. (2004). Antioxidant evaluation protocols: Food quality or health effects. *European Food Research and Technology*, *219*, 561-571.

Becker, K., Geisler, S., Ueberall, F., Fuchs, D., & Gostner, J. M. (2013). Immunomodulatory properties of cacao extracts - potential consequences for medical applications. *Frontiers in Pharmacology*, *4*(154), 1-9.

Beckett, S. (2000). The History of Chocolate. *The Science of Chocolate*, 1–7.

Beesoo, R., Neergheen-Bhujun, V., Bhagooli, R., & Bahorun, T. (2014). Apoptosis inducing lead compounds isolated from marine organisms of potential relevance in cancer treatment. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis*, *768*, 84–97.

© COPYRIGHT UPM

Belpomme, D., Irigaray, P., Hardell, L., Clapp, R., Montagnier, L., Epstein, S., & Sasco, A. J. (2007). The multitude and diversity of environmental carcinogens. *Environmental Research*, *105*(3), 414–429.

Belščak, A., Komes, D., Horžić, D., Ganić, K. K., & Karlović, D. (2009). Comparative study of commercially available cocoa products in terms of their bioactive composition. *Food Research International*, *42*, 707–716.

Benarba, B., Meddah, B., & Aoues, A. (2012). *Bryonia dioica* aqueous extract induces apoptosis through mitochondrial intrinsic pathway in BL41 Burkitt's lymphoma cells. *Journal of Ethnopharmacology*, *141*(1), 510–6.

Bennett, A. B. (2003). Out of the Amazon: *Theobroma cacao* enters the genomic era. *Trends in Plant Science*, *8*(12), 561–563.

Bennett, L. L., Rojas, S., & Seefeldt, T. (2012). Role of antioxidants in the prevention of cancer. *Journal of Experimental and Clinical Medicine*, *4*, 215–222.

Bento, C., Andersson, M. K., & Åman, P. (2009). DDIT3/CHOP and the sarcoma fusion oncoprotein FUS-DDIT3/TLS-CHOP bind cyclin-dependent kinase 2. *BMC Cell Biology*, *10*(1), 89.

Bernabeu, A., Guillén, J., Pérez-Berná, A. J., Moreno, M. R., & Villalaín, J. (2007). Structure of the C-terminal domain of the pro-apoptotic protein Hrk and its interaction with model membranes. *Biochimica et Biophysica Acta (BBA) - Biomembranes*, *1768*(6), 1659–1670.

Bhattacharjee, R., & Kumar, P. L. (2007). Cacao. *Genome Mapping and Molecular Breeding in Plants*, 6, 127–142.

Blanchard, H., Kodandapani, L., Mittl, P. R., Marco, S. Di, Krebs, J. F., Wu, J. C., … Grütter, M. G. (1999). The three-dimensional structure of caspase-8: An initiator enzyme in apoptosis. *Structure*, *7*(9), 1125–1133.

Boffetta, P., Hashibe, M., La Vecchia, C., Zatonski, W., & Rehm, J. (2006). The burden of cancer attributable to alcohol drinking. *International Journal of Cancer*, *119*(4), 884–887.

Borchers, A. T., Keen, C. L., Hannum, S. M., & Gershwin, M. E. (2000). Cocoa and chocolate: Composition, bioavailability, and health implications. *Journal of Medicinal Food*, *3*(2), 77–105.

Borrás-Linares, I., Pérez-Sánchez, A., Lozano-Sánchez, J., Barrajón-Catalán, E., Arráez-Román, D., Cifuentes, A., … Carretero, A. S. (2015). A bioguided identification of the active compounds that contribute to the antiproliferative/cytotoxic effects of rosemary extract on colon cancer cells. *Food and Chemical Toxicology*, *80*(0), 215–222.

© COPYRIGHT UPM

Bossis, G., Sarry, J.-E., Kifagi, C., Ristic, M., Saland, E., Vergez, F., … Piechaczyk, M. (2014). The ROS/SUMO axis contributes to the response of acute myeloid leukemia cells to chemotherapeutic drugs. *Cell Reports*, *7*(6), 1815–1823.

Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*, *72*(1-2), 248–254.

Bratton, S. B., & Salvesen, G. S. (2010). Regulation of the Apaf-1-caspase-9 apoptosome. *Journal of Cell Science*, *123*(19), 3209–3214.

Brentnall, M., Rodriguez-Menocal, L., De Guevara, R., Cepero, E., & Boise, L. H. (2013). Caspase-9, caspase-3 and caspase-7 have distinct roles during intrinsic apoptosis. *BMC Cell Biology*, *14*(1), 32.

Brücher, B. L., & Jamall, I. S. (2014). Epistemology of the origin of cancer: A new paradigm. *BMC Cancer*, *14*, 331.

Bruna, C., Eichholz, I., Rohn, S., Kroh, L. W., & Huyskens-Keil, S. (2009). Bioactive compounds and antioxidant activity of cocoa hulls (*Theobroma cacao* L.) from different origins. *Journal of Applied Botany and Food Quality-Angewandte Botanik*, *83*, 9–13.

Brusotti, G., Cesari, I., Dentamaro, A., Caccialanza, G., & Massolini, G. (2014). Isolation and characterization of bioactive compounds from plant resources: The role of analysis in the ethnopharmacological approach. *Journal of Pharmaceutical and Biomedical Analysis*, *87*, 218–228.

Busch, S. J., Barnhart, R. L., Martin, G. A., Flanagan, M. A., & Jackson, R. L. (1990). Differential regulation of hepatic triglyceride lipase and 3-hydroxy-3-methylglutaryl-CoA reductase gene expression in a human hepatoma cell line, HepG2. *The Journal of Biological Chemistry*, *265*(36), 22474–22479.

Bustin, S. a. (2005). Quantitative real-time RT-PCR - a perspective. *Journal of Molecular Endocrinology*, *34*(3), 597–601.

Bustin, S. A. (2000). Absolute quantification of mRNA using real-time reverse transcription polymerase chain reaction assays. *Journal of Molecular Endocrinology*, *25*(2), 169–93.

Caamal-Fuentes, E., Torres-Tapia, L. W., Simá-Polanco, P., Peraza-Sánchez, S. R., & Moo-Puc, R. (2011). Screening of plants used in Mayan traditional medicine to treat cancer-like symptoms. *Journal of Ethnopharmacology*, *135*, 719–724.

Cádiz-Gurrea, M. de la L., Lozano-Sánchez, J., Contreras-Gámez, M. del M., Legeai-Mallet, L., Fernández-Arroyo, S., & Segura-Carretero, A. (2014). Isolation, comprehensive characterization and antioxidant activities of *Theobroma cacao* extract. *Journal of Functional Foods*, *10*, 485–498.

© COPYRIGHT UPM

Cai, Y., Luo, Q., Sun, M., & Corke, H. (2004). Antioxidant activity and phenolic compounds of 112 traditional Chinese medicinal plants associated with anticancer. *Life Sciences*, *74*(17), 2157–2184.

Cailleau, R., Olivé, M., & Cruciger, Q. V. J. (1978). Long-term human breast carcinoma cell lines of metastatic origin: Preliminary characterization. *In Vitro*, *14*(11), 911–915.

Calatayud, M., López-De-Dicastillo, C., López-Carballo, G., Vélez, D., Hernández Muñoz, P., & Gavara, R. (2013). Active films based on cocoa extract with antioxidant, antimicrobial and biological applications. *Food Chemistry*, *139*, 51–58.

Camargo, M. S., Oliveira, M. T., Santoni, M. M., Resende, F. A., Oliveira-Höhne, A. P., Espanha, L. G., … Varanda, E. A. (2015). Effects of nemorosone, isolated from the plant *Clusia rosea*, on the cell cycle and gene expression in MCF-7 BUS breast cancer cell lines. *Phytomedicine*, *22*(1), 153–157.

Campbell, R. A., Bhat-Nakshatri, P., Patel, N. M., Constantinidou, D., Ali, S., & Nakshatri, H. (2001). Phosphatidylinositol 3-Kinase/AKT-mediated activation of estrogen receptor : A new model for anti-estrogen resistance. *Journal of Biological Chemistry*, *276*(13), 9817–9824.

Campeau, P. M., Foulkes, W. D., & Tischkowitz, M. D. (2008). Hereditary breast cancer: New genetic developments, new therapeutic avenues. *Human Genetics*, *124*(1), 31–42.

Carnesecchi, S., Schneider, Y., Lazarus, S. A., Coehlo, D., Gosse, F., & Raul, F. (2002). Flavanols and procyanidins of cocoa and chocolate inhibit growth and polyamine biosynthesis of human colonic cancer cells. *Cancer Lett*, *175*, 147–155.

Carrington, P. E., Sandu, C., Wei, Y., Hill, J. M., Morisawa, G., Huang, T., … Werner, M. H. (2006). The structure of FADD and its mode of interaction with procaspase-8. *Molecular Cell*, *22*(5), 599–610.

Catalá, A. (2009). Lipid peroxidation of membrane phospholipids generates hydroxy-alkenals and oxidized phospholipids active in physiological and/or pathological conditions. *Chemistry and Physics of Lipids*, *157*(1), 1–11.

Cerretti, D. P., Kozlosky, C. J., Mosley, B., Nelson, N., Van Ness, K., Greenstreet, T. A., … Cannizzaro, L. A. (1992). Molecular cloning of the interleukin-1 beta converting enzyme. *Science*, *256*(5053), 97–100.

Cerveira, N., Bizarro, S., & Teixeira, M. R. (2012). Cancer cell cycle. *canalBQ*, *9*, 40–47.

© COPYRIGHT UPM

Chan, G. K. Y., Kleinheinz, T. L., Peterson, D., & Moffat, J. G. (2013). A simple high-content cell cycle assay reveals frequent discrepancies between cell number and ATP and MTS proliferation assays. *PLoS ONE*, *8*(5), e63583.

Chandranayagam, C., Veeraraghavan, G., Subash, A., & Vasanthi, H. R. (2013). Restoration of arsenite induced hepato-toxicity by crude tannin rich fraction of *Theobroma cacao* in Sprague Dawley rats. *Food Research International*, *50*, 46–54.

Chang-Claude, J. (2001). Inherited genetic susceptibility to breast cancer. *IARC Scientific Publications*, *154*, 177–190.

Chauhan, A. (2014). GC-MS Technique and its analytical applications in science and technology. *Journal of Analytical & Bioanalytical Techniques*, *5*(6), 1–5.

Chen, S.-T., Pan, T.-L., Tsai, Y.-C., & Huang, C.-M. (2002). Proteomics reveals protein profile changes in doxorubicin--treated MCF-7 human breast cancer cells. *Cancer Letters*, *181*(1), 95–107.

Chew, H. K. (2001). Genetic evaluation of cancer: The importance of family history. *Texas Medicine*, *97*(2), 40–45.

Cho, Y., Challa, S., Moquin, D., Genga, R., Ray, T. D., Guildford, M., & Chan, F. K.-M. (2009). Phosphorylation-driven assembly of the RIP1-RIP3 complex regulates programmed necrosis and virus-induced inflammation. *Cell*, *137*(6), 1112–1123.

Chowdhury, I., Tharakan, B., & Bhat, G. K. (2008). Caspases-An update. comparative biochemistry and physiology Part B. *Biochemistry and Molecular Biology*, *151*(1), 10–27.

Cieckiewicz, E., Angenot, L., Gras, T., Kiss, R., & Frédérich, M. (2012). Potential anticancer activity of young *Carpinus betulus* leaves. *Phytomedicine*, *19*(3-4), 278–83.

Cienfuegos-Jovellanos, E., Del Mar Quiñones, M., Muguerza, B., Moulay, L., Miguel, M., & Aleixandre, A. (2009). Antihypertensive effect of a polyphenol-rich cocoa powder industrially processed to preserve the original flavonoids of the cocoa beans. *Journal of Agricultural and Food Chemistry*, *57*, 6156–6162.

Colvin, T. A., Gabai, V. L., Gong, J., Calderwood, S. K., Li, H., Gummuluru, S., … Sherman, M. Y. (2014). Hsp70-Bag3 interactions regulate cancer-related signaling networks. *Cancer Research*, *74*(17), 4731–4740.

Conforti, F., Ioele, G., Statti, G. A., Marrelli, M., Ragno, G., & Menichini, F. (2008). Antiproliferative activity against human tumor cell lines and toxicity test on Mediterranean dietary plants. *Food and Chemical Toxicology*, *46*, 3325–3332.

© COPYRIGHT UPM

Cooper, K. A., Campos-Giménez, E., Alvarez, D. J., Nagy, K., Donovan, J. L., & Williamson, G. (2007). Rapid reversed phase ultra-performance liquid chromatography analysis of the major cocoa polyphenols and inter-relationships of their concentrations in chocolate. *Journal of Agricultural and Food Chemistry*, *55*(8), 2841–2847.

Cordero-Herrera, I., Martín, M. Á., Goya, L., & Ramos, S. (2014). Cocoa flavonoids attenuate high glucose-induced insulin signalling blockade and modulate glucose uptake and production in human HepG2 cells. *Food and Chemical Toxicology*, *64*, 10–19.

Corti, R., Flammer, A. J., Hollenberg, N. K., & Luscher, T. F. (2009). Cocoa and cardiovascular health. *Circulation*, *119*(10), 1433–1441.

Counet, C., Callemien, D., & Collin, S. (2006). Chocolate and cocoa: New sources of trans-resveratrol and trans-piceid. *Food Chemistry*, *98*(4), 649–657.

Cragg, G. M., & Newman, D. J. (2013). Natural products: A continuing source of novel drug leads. *Biochimica et Biophysica Acta*, *1830*(6), 3670–95.

Croce, C. M. (2008). Oncogenes and cancer. supplementary appendix. *The New England Journal of Medicine*, *358*(5), 502–11.

Cui, Y., Deming-Halverson, S. L., Shrubsole, M. J., Beeghly-Fadiel, A., Fair, A. M., Sanderson, M., … Zheng, W. (2014). Associations of hormone-related factors with breast cancer risk according to hormone receptor status among white and African American women. *Clinical Breast Cancer*, *14*(6), 417–425.

Dai, J., & Mumper, R. J. (2010a). Plant phenolics: Extraction, analysis and their antioxidant and anticancer properties. *Molecules*, *15*(10), 7313–7352.

Dakin, K., & Wichmann, S. (2000). Cacao and chocolate: A Uto-Azteca Perspective. *Ancient Mesoamerica*, *11*(1), 55-75.

Danial, N. N., & Korsmeyer, S. J. (2004). Cell death. *Cell*, *116*(2), 205–219.

Darbre, P. D. (2006). Environmental oestrogens, cosmetics and breast cancer. *Best Practice & Research Clinical Endocrinology & Metabolism*, *20*(1), 121–143.

Darlington, G. J., Kelly, J. H., & Buffone, G. J. (1987). Growth and hepatospecific gene expression of human hepatoma cells in a defined medium. *In vitro Cellular & Developmental Biology*, *23*(5), 349–354.

de Mesquita, M. L., de Paula, J. E., Pessoa, C., de Moraes, M. O., Costa-Lotufo, L. V., Grougnet, R., … Espindola, L. S. (2009). Cytotoxic activity of Brazilian Cerrado plants used in traditional medicine against cancer cell lines. *Journal of Ethnopharmacology*, *123*, 439–445.

© COPYRIGHT UPM

de Oliveira, T. B., Rogero, M. M., & Genovese, M. I. (2015). Poliphenolic-rich extracts from cocoa (*Theobroma cacao* L.) and cupuassu (*Theobroma grandiflorum* Willd. Ex Spreng. K. Shum) liquors: A comparison of metabolic effects in high-fat fed rats. *PharmaNutrition*, *3*(2), 20–28.

Degterev, A., Huang, Z., Boyce, M., Li, Y., Jagtap, P., Mizushima, N., … Yuan, J. (2005). Chemical inhibitor of nonapoptotic cell death with therapeutic potential for ischemic brain injury. *Nature Chemical Biology*, *1*(2), 112–119.

Dellai, A., Laajili, S., Le Morvan, V., Robert, J., & Bouraoui, A. (2013). Antiproliferative activity and phenolics of the Mediterranean seaweed *Laurencia obusta*. *Industrial Crops and Products*, *47*, 252–255.

De-qiang, L., Zhao, J., Wu, D., & Shao-ping, L. (2016). Discovery of active components in herbs using chromatographic separation coupled with online bioassay. *Journal of Chromatography B*, *1021*, 81–90.

Desch, S., Schmidt, J., Kobler, D., Sonnabend, M., Eitel, I., Sareban, M., … Thiele, H. (2010). Effect of cocoa products on blood pressure: Systematic review and meta-analysis. *American Journal of Hypertension*, *23*, 97–103.

Devedjian, J. C., Fargues, M., Denis-Pouxviel, C., Daviaud, D., Prats, H., & Paris, H. (1991). Regulation of the alpha 2A-adrenergic receptor in the HT29 cell line. Effects of insulin and growth factors. *The Journal of Biological Chemistry*, *266*(22), 14359–14366.

Dias, D. a, Urban, S., & Roessner, U. (2012). A historical overview of natural products in drug discovery. *Metabolites*, *2*(2), 303–36.

Diaz-Padilla, I., Siu, L. L., & Duran, I. (2009). Cyclin-dependent kinase inhibitors as potential targeted anticancer agents. *Investigational New Drugs*, *27*(6), 586–594.

Dillinger, T. L., Barriga, P., Escárcega, S., Jimenez, M., Salazar Lowe, D., & Grivetti, L. E. (2000). Food of the gods: Cure for humanity? A cultural history of the medicinal and ritual use of chocolate. *The Journal of Nutrition*, *130*(8S Suppl), 2057S–72S.

Ding, D., Shinichi, S., Hai–yan, J., Wei–dong, Q., Dong–zhen, Y., Tanokura, M., & Salvi, R. (2011). Detection of apoptosis by RT–PCR array in mefloquine–induced cochlear damage. *Journal of Otology*, *6*(1), 1–9.

Dinh, C. T., Goncalves, S., Bas, E., Van De Water, T. R., & Zine, A. (2015). Molecular regulation of auditory hair cell death and approaches to protect sensory receptor cells and/or stimulate repair following acoustic trauma. *Frontiers in Cellular Neuroscience*, *9*, 1–15.

Dittmer, J., & Leyh, B. (2015). The impact of tumor stroma on drug response in breast cancer. *Seminars in Cancer Biology*, *31*, 3–15.

© COPYRIGHT UPM

Dixon, R. A. (1995). Stress-Induced Phenylpropanoid Metabolism. *The Plant Cell Online*, *7*(7), 1085–1097.

Doonan, F., & Cotter, T. G. (2008). Morphological assessment of apoptosis. *Methods (San Diego, Calif.)*, *44*(3), 200–4.

Duarte, K., Rocha-Santos, T. A. P., Freitas, A. C., & Duarte, A. C. (2012). Analytical techniques for discovery of bioactive compounds from marine fungi. *TrAC Trends in Analytical Chemistry*, *34*, 97–110.

Dutt, R., Garg, V., & Madan, A. (2014). Can plants growing in diverse hostile environments provide a vital source of anticancer drugs? *Cancer Therapy*, *10*, 13–37.

Eberl, M. M., Sunga, A. Y., Farrell, C. D., & Mahoney, M. C. (2005). Patients with a family history of cancer: Identification and management. *The Journal of the American Board of Family Medicine*, *18*(3), 211–217.

Edelmann, B., Bertsch, U., Tchikov, V., Winoto-Morbach, S., Perrotta, C., Jakob, M., … Schütze, S. (2011). Caspase-8 and caspase-7 sequentially mediate proteolytic activation of acid sphingomyelinase in TNF-R1 receptosomes. *The EMBO Journal*, *30*(2), 379–394.

Edinger, A. L., & Thompson, C. B. (2004). Death by design: Apoptosis, necrosis and autophagy. *Current Opinion in Cell Biology*, *16*(6), 663–669.

El-Beltagi, H., & Mohamed, H. (2013). Reactive oxygen species, lipid peroxidation and antioxidative defense mechanism. *Notulae botanicae Horti Agrobotanici Cluj-Napoca*, *41*(1), 44–57.

Elezabeth, V. D., & Arumugam, S. (2014). GC – MS Analysis of ethanol extract of *Cyperus rotundus* leaves. *International Journal of Current Biotechnology*, *2*(1), 19–23.

El-Khattouti, A., Selimovic, D., Haikel, Y., & Hassan, M. (2013). Crosstalk between apoptosis and autophagy: Molecular mechanisms and therapeutic strategies in cancer. *Journal of Cell Death*, *6*, 37–55.

Elmore, S. (2007). Apoptosis: A review of programmed cell death. *Toxicologic Pathology*, *35*(4), 495–516.

Elumalai, P., Gunadharini, D. N., Senthilkumar, K., Banudevi, S., Arunkumar, R., Benson, C. S., … Arunakaran, J. (2012). Ethanolic neem (*Azadirachta indica* A. Juss) leaf extract induces apoptosis and inhibits the IGF signaling pathway in breast cancer cell lines. *Biomedicine & Preventive Nutrition*, *2*(1), 59–68.

Exner, V. (2010). Quantitative real time PCR in plant developmental biology. In L. Hennig & C. Köhler (Ed.), *Plant Developmental Biology* (pp. 275–291). Totowa, NJ: Humana Press.

© COPYRIGHT UPM

Fadeel, B., & Orrenius, S. (2005). Apoptosis: A basic biological phenomenon with wide-ranging implications in human disease. *Journal of Internal Medicine*, *258*(6), 479–517.

Fang, X. (1997). Lysophosphatidylcholine stimulates activator protein 1 and the c-Jun N-terminal kinase activity. *Journal of Biological Chemistry*, *272*(21), 13683–13689.

Fang, X., Shao, L., Zhang, H., & Wang, S. (2005). CHMIS-C: A comprehensive herbal medicine information system for cancer. *Journal of Medicinal Chemistry*, *48*(5), 1481–1488.

Fantini, J., Yahi, N., Baghdiguian, S., & Chermann, J. C. (1992). Human colon epithelial cells productively infected with human immunodeficiency virus show impaired differentiation and altered secretion. *Journal of Virology*, *66*(1), 580–585.

Fass, L. (2008). Imaging and cancer: A review. *Molecular Oncology*, *2*(2), 115–152.

Fauzi, A. N., Norazmi, M. N., & Yaacob, N. S. (2011). Tualang honey induces apoptosis and disrupts the mitochondrial membrane potential of human breast and cervical cancer cell lines. *Food and Chemical Toxicology*, *49*(4), 871–8.

Ferlay, J., Soerjomataram, I., Ervik, M., Dikshit, R., Eser, S., Mathers, C., … Bray, F., *Cancer Incidence and Mortality Worldwide*. International Agency for Research on Cancer: Lyon, France. 2013.

Fernandes-Alnemri, T., Litwack, G., & Alnemri, E. S. (1994). CPP32, a novel human apoptotic protein with homology to Caenorhabditis elegans cell death protein Ced-3 and mammalian interleukin-1β-converting enzyme. *Journal of Biological Chemistry*, *269*(49), 30761–30764.

Ferrazzano, G. F., Amato, I., Ingenito, A., De Natale, A., & Pollio, A. (2009). Anti-cariogenic effects of polyphenols from plant stimulant beverages (cocoa, coffee, tea). *Fitoterapia*, *80*(5), 255–262.

Fink, S. L., & Cookson, B. T. (2005). Apoptosis, pyroptosis, and necrosis: Mechanistic description of dead and dying eukaryotic cells. *Infection and Immunity*, *73*(4), 1907–1916.

Fogh, J., Wright, W. C., & Loveless, J. D. (1977). Absence of HeLa cell contamination in 169 cell lines derived from human tumors. *Journal of the National Cancer Institute*, *58*(2), 209–214.

Forgo, P., Zupkó, I., Molnár, J., Vasas, A., Dombi, G., & Hohmann, J. (2012). Bioactivity-guided isolation of antiproliferative compounds from *Centaurea jacea* L. *Fitoterapia*, *83*(5), 921–925.

© COPYRIGHT UPM

Forouzanfar, M. H., Foreman, K. J., Delossantos, A. M., Lozano, R., Lopez, A. D., Murray, C. J. L., & Naghavi, M. (2011). Breast and cervical cancer in 187 countries between 1980 and 2010: A systematic analysis. *The Lancet*, *378*, 1461–1484.

Francken, A. B., Schouten, P. C., Bleiker, E. M. a, Linn, S. C., & Rutgers, E. J. T. (2013). Breast cancer in women at high risk: The role of rapid genetic testing for BRCA1 and -2 mutations and the consequences for treatment strategies. *Breast*. Elsevier Ltd.

Freshney, R. I. (2010). Culture of Animal Cells. Culture of Animal Cells: A Manual of Basic Technique and Specialized Applications. Hoboken, NJ, USA: John Wiley & Sons, Inc.

Friedman, A. D. (1996). GADD153/CHOP, a DNA damage-inducible protein, reduced CAAT/enhancer binding protein activities and increased apoptosis in 32D c13 myeloid cells. *Cancer Research*, *56*(14), 3250–6.

Fu, W., Chen, J., Cai, Y., Lei, Y., Chen, L., Pei, L., … Ruan, J. (2010). Antioxidant, free radical scavenging, anti-inflammatory and hepatoprotective potential of the extract from *Parathelypteris nipponica* (Franch. et Sav.) Ching. *Journal of Ethnopharmacology*, *130*, 521–528.

Fuentes-Prior, P., & Salvesen, G. S. (2004). The protein structures that shape caspase activity, specificity, activation and inhibition. *The Biochemical Journal*, *384*, 201–232.

Fulda, S. (2009). Tumor resistance to apoptosis. *International Journal of Cancer*, *124*(3), 511–515.

Fulda, S. (2010). Modulation of apoptosis by natural products for cancer therapy. *Planta Medica*, 76(11), 1075-9.

Fulda, S. (2015a). Targeting apoptosis for anticancer therapy. *Seminars in Cancer Biology*, *31*, 84–88.

Fulda, S. (2015b). Targeting extrinsic apoptosis in cancer: Challenges and opportunities. *Seminars in Cell & Developmental Biology*, *39*, 20–25.

Fulda, S., & Debatin, K.-M. (2006). Extrinsic versus intrinsic apoptosis pathways in anticancer chemotherapy. *Oncogene*, *25*(34), 4798–4811.

Fuster, V., Moreno, P. R., Fayad, Z. A., Corti, R., & Badimon, J. J. (2005). Atherothrombosis and high-risk plaque: Part I: Evolving concepts. *Journal of the American College of Cardiology*, *46*(6), 937-54.

Galleano, M., Oteiza, P. I., & Fraga, C. G. (2009). Cocoa, chocolate, and cardiovascular disease. *Journal of Cardiovascular Pharmacology*, *54*, 483–490.

© COPYRIGHT UPM

Galluzzi, L., De Santi, M., Crinelli, R., De Marco, C., Zaffaroni, N., Duranti, A., … Magnani, M. (2012). Induction of endoplasmic reticulum stress response by the indole-3-carbinol cyclic tetrameric derivative CTet in human breast cancer cell lines. *PLoS ONE*, *7*(8), e43249.

Gammon, M. D., Eng, S. M., Teitelbaum, S. L., Britton, J. A., Kabat, G. C., Hatch, M., … Santella, R. M. (2004). Environmental tobacco smoke and breast cancer incidence. *Environmental Research*, *96*, 176–185.

Garrido, C., Galluzzi, L., Brunet, M., Puig, P. E., Didelot, C., & Kroemer, G. (2006). Mechanisms of cytochrome c release from mitochondria. *Cell Death and Differentiation*, *13*(9), 1423–1433.

Gentilella, A., & Khalili, K. (2011). BAG3 Expression in glioblastoma cells promotes accumulation of ubiquitinated clients in an Hsp70-dependent manner. *Journal of Biological Chemistry*, *286*(11), 9205–9215.

Ghavami, S., Hashemi, M., Ande, S. R., Yeganeh, B., Xiao, W., Eshraghi, M., … Los, M. (2009). Apoptosis and cancer: Mutations within caspase genes. *Journal of Medical Genetics*, *46*(8), 497–510.

Gianni, L., Norton, L., Wolmark, N., Suter, T. M., Bonadonna, G., & Hortobagyi, G. N. (2009). Role of anthracyclines in the treatment of early breast cancer. *Journal of Clinical Oncology*, *27*(28), 4798–4808.

Giard, D. J., Aaronson, S. A., Todaro, G. J., Arnstein, P., Kersey, J. H., Dosik, H., & Parks, W. P. (1973). *In vitro* cultivation of human tumors: Establishment of cell lines derived from a series of solid tumors. *Journal of the National Cancer Institute*, *51*(5), 1417–1423.

Gibbs, W. W. (2008). Untangling the roots of cancer. *Scientific American Sp*, *18*(3), 30–39.

Giri, B., Gomes, A., Debnath, A., Saha, A., Biswas, A. K., Dasgupta, S. C., & Gomes, A. (2006). Antiproliferative, cytotoxic and apoptogenic activity of Indian toad (*Bufo melanostictus*, Schneider) skin extract on U937 and K562 cells. *Toxicon*, *48*, 388–400.

Golias, C. H., Charalabopoulos, A., & Charalabopoulos, K. (2004). Cell proliferation and cell cycle control: A mini review. *International Journal of Clinical Practice*, *58*(12), 1134–1141.

Gonzalez, C. A., & Riboli, E. (2006). Diet and cancer prevention: Where we are, where we are going. *Nutrition and Cancer*, *56*(2), 225–231.

Gopinath, P., D, S. V., Kamatchiammal, S., & Saroja, V. (2013). Anti-cancerous activity of *Albizia Amara* (Roxb .) boivin using human reast cancer cells (MCF-7) by *in vitro* methods. *International Journal of Pharma Research & Review*, *2*, 23–32.

© COPYRIGHT UPM

Granado-Serrano, A. B., Martín, M. A., Bravo, L., Goya, L., & Ramos, S. (2009). A diet rich in cocoa attenuates N-nitrosodiethylamine-induced liver injury in rats. *Food and Chemical Toxicology*, *47*, 2499–2506.

Grassi, D., Desideri, G., & Ferri, C. (2010). Blood pressure and cardiovascular risk: What about cocoa and chocolate? *Archives of Biochemistry and Biophysics*, *501*(1), 112-5.

Grassi, D., Necozione, S., Lippi, C., Croce, G., Valeri, L., Pasqualetti, P., … Ferri, C. (2005). Cocoa reduces blood pressure and insulin resistance and improves endothelium-dependent vasodilation in hypertensives. *Hypertension*, *46*, 398–405.

Gruss, H. J., Duyster, J., & Herrmann, F. (1996). Structural and biological features of the TNF receptor and TNF ligand superfamilies: Interactive signals in the pathobiology of Hodgkin's disease. *Annals of Oncology*, *7*(4), 19–26.

Häcker, H.-G., Sisay, M. T., & Gütschow, M. (2011). Allosteric modulation of caspases. *Pharmacology & Therapeutics*, *132*(2), 180–195.

Halliwell, B., & Gutteridge, J. M. C. (2007). Free radicals in biology and medicine. Clarendon: Oxford.

Hameroff, S. R. (2004). A new theory of the origin of cancer: Quantum coherent entanglement, centrioles, mitosis, and differentiation. *Biosystems*, *77*(1-3), 119–136.

Hamid, R., Rotshteyn, Y., Rabadi, L., Parikh, R., & Bullock, P. (2004). Comparison of alamar blue and MTT assays for high through-put screening. *Toxicology In Vitro*, *18*, 703–710.

Hanahan, D., & Weinberg, R. A. (2000). The hallmarks of cancer. *Cell*, *100*, 57–70.

Hanahan, D., & Weinberg, R. A. (2011). Hallmarks of cancer: The next generation. *Cell*, *144*(5), 646–674.

Hansen, M. B., Nielsen, S. E., & Berg, K. (1989). Re-examination and further development of a precise and rapid dye method for measuring cell growth/cell kill. *Journal of Immunological Methods*, *119*, 203–210.

Hanski, C., Stolze, B., & Riecken, E. O. (1992). Tumorigenicity, mucin production and AM-3 epitope expression in clones selected from the HT-29 colon carcinoma cell line. *International Journal of Cancer*, *50*(6), 924–929.

Hazirah, A., Zainal, B., & Abdah, M. (2013). Total phenolic content, antioxidant and cytotoxic activity of cocoa (*Theobroma cacao* L.) polyphenols extracts on cancer cell lines. *Malaysian Journal of Nutrition*, *19*(2), 223 – 232.

© COPYRIGHT UPM

Heid, C. a, Stevens, J., Livak, K. J., & Williams, P. M. (1996). Real time quantitative PCR. *Genome Research*, *6*(10), 986–994.

Hema, R., Kumaravel, S., Gomathi, S., Sivasubramaniam, C., & Sciences, H. (2010). Gas Chromatography – Mass spectroscopic analysis of *Lawsonia inermis* leaves. *New York Science Journal*, *3*(11), 141–143.

Heptinstall, S., May, J., Fox, S., Kwik-Uribe, C., & Zhao, L. (2006). Cocoa flavanols and platelet and leukocyte function: Recent *in vitro* and *ex vivo* studies in healthy adults. *Journal of Cardiovascular Pharmacology*, *47*(2), S197–S205.

Hii, C. L., Law, C. L., Suzannah, S., Misnawi, & Cloke, M. (2009). Polyphenols in cocoa (*Theobroma cacao* L.). *Asian Journal of Food and Agro-Industry*, *2*(04), 702–722.

Hirao, C., Nishimura, E., Kamei, M., Ohshima, T., & Maeda, N. (2010). Antibacterial effects of cocoa on periodontal pathogenic bacteria. *Journal of Oral Biosciences*, *52*(3), 283–291.

Hollman, P. C., & Arts, I. C. (2000). Flavonols, flavones and flavanols - nature, occurrence and dietary burden. *Journal of the Science of Food and Agriculture*, *80*(7), 1081–1093.

Hossain, M. A., Shah, M. D., Gnanaraj, C., & Iqbal, M. (2011). *In vitro* total phenolics, flavonoids contents and antioxidant activity of essential oil, various organic extracts from the leaves of tropical medicinal plant Tetrastigma from Sabah. *Asian Pacific Journal of Tropical Medicine*, *4*(9), 717–21.

Hseu, Y.-C., Chen, S.-C., Chen, H.-C., Liao, J.-W., & Yang, H.-L. (2008). Antrodia camphorata inhibits proliferation of human breast cancer cells *in vitro* and *in vivo*. *Food and Chemical Toxicology*, *46*, 2680–2688.

Hu, Q., Wu, D., Chen, W., Yan, Z., Yan, C., He, T., … Shi, Y. (2014). Molecular determinants of caspase-9 activation by the Apaf-1 apoptosome. *Proceedings of the National Academy of Sciences*, *111*(46), 16254–16261.

Hu, W., Yu, L., & Wang, M.-H. (2011). Antioxidant and antiproliferative properties of water extract from *Mahonia bealei* (Fort.) Carr. leaves. *Food and Chemical Toxicology*, *49*(4), 799–806.

Huang, T.-Y., Hsu, C.-W., Chang, W.-C., Wang, M.-Y., Wu, J.-F., & Hsu, Y.-C. (2012). Demethoxycurcumin retards cell growth and induces apoptosis in human brain malignant glioma GBM 8401 Cells. *Evidence-Based Complementary and Alternative Medicine*, *2012*, 396573.

© COPYRIGHT UPM

Huguet, E. L., McMahon, J. A., McMahon, A. P., Bicknell, R., & Harris, A. L. (1994). Differential expression of human Wnt genes 2, 3, 4, and 7B in human breast cell lines and normal and disease states of human breast tissue. *Cancer Research*, *54*(10), 2615–2621.

Ignat, I., Volf, I., & Popa, V. I. (2011). A critical review of methods for characterisation of polyphenolic compounds in fruits and vegetables. *Food Chemistry*, *126*(4), 1821–1835.

Indran, I. R., Tufo, G., Pervaiz, S., & Brenner, C. (2011). Recent advances in apoptosis, mitochondria and drug resistance in cancer cells. *Biochimica et Biophysica Acta (BBA) - Bioenergetics*, *1807*(6), 735–745.

Ishak, D. H. A., Ooi, K. K., Ang, K.-P., Akim, A. M., Cheah, Y.-K., Nordin, N., … Tiekink, E. R. T. (2014). A bismuth diethyldithiocarbamate compound promotes apoptosis in HepG2 carcinoma, cell cycle arrest and inhibits cell invasion through modulation of the NF-κB activation pathway. *Journal of Inorganic Biochemistry*, *130*, 38–51.

Ismail, A., Marjan, Z., & Foong, C. (2004). Total antioxidant activity and phenolic content in selected vegetables. *Food Chemistry*, *87*(4), 581–586.

Jain, R., & Jain, S. K. (2011). Screening of *in vitro* cytotoxic activity of some medicinal plants used traditionally to treat cancer in Chhattisgarh state, India. *Asian Pacific Journal of Tropical Biomedicine*, *1*(2), S147–S150.

Jalil, A. M. M., & Ismail, A. (2008). Polyphenols in cocoa and cocoa products: Is there a link between antioxidant properties and health? *Molecules*, *13*(9), 2190-2219.

Jalil, A. M. M., Ismail, A., Pei, C. P., Hamid, M., & Kamaruddin, S. H. S. (2008). Effects of cocoa extract on glucometabolism, oxidative stress, and antioxidant enzymes in obese-diabetic (Ob-db) rats. *Journal of Agricultural and Food Chemistry*, *56*(17), 7877–7884.

Jones, H. W. J., Mc Kusick, V. A., Harper, P. S., & Wuu, K.-D. (1971). George Otto Gey (1899-1970): The heLa cell and a reappraisal of its origin. *Obstetrics & Gynecology*, *38*(6), 945–949.

Jonfia-Essien, W. A., West, G., Alderson, P. G., & Tucker, G. (2008). Phenolic content and antioxidant capacity of hybrid variety cocoa beans. *Food Chemistry*, *108*, 1155–1159.

Judd, W. S., Campbell, C. S., Kellogg, E. A., Stevens, P. F., & Donoghue, M. J. (2002). Plant systematics: A phylogenetic approach. Sunderland, MA: Sinauer Associates.

Kalaisezhiyen, P., & Sasikumar, V. (2012). GC- MS evaluation of chemical constituents from methanolic leaf extract of *Kedrostis foetidissima* (Jacq.) Cogn. *Asian Journal of Pharmaceutical and Clinical Research*, *5*(3), 0–4.

© COPYRIGHT UPM

Kalinowska, M., Bielawska, A., Lewandowska-Siwkiewicz, H., Priebe, W., & Lewandowski, W. (2014). Apples: Content of phenolic compounds vs. variety, part of apple and cultivation model, extraction of phenolic compounds, biological properties. *Plant Physiology and Biochemistry, 84*, 169–188.

Kampinga, H. H., Hageman, J., Vos, M. J., Kubota, H., Tanguay, R. M., Bruford, E. a., … Hightower, L. E. (2009). Guidelines for the nomenclature of the human heat shock proteins. *Cell Stress and Chaperones*, *14*(1), 105–111.

Kantari, C., & Walczak, H. (2011). Caspase-8 and Bid: Caught in the act between death receptors and mitochondria. *Biochimica et Biophysica Acta (BBA) - Molecular Cell Research*, *1813*(4), 558–563.

Karagöz, A., Turgut-Kara, N., Çakir, Ö., Demirgan, R., & Ar, Ş. (2007). Cytotoxic activity of crude extracts from *Astragalus chrysochlorus* (Leguminosae). *Biotechnol. & Biotechnol. EQ.*, *21*(2), 2–4.

Karp, G. (2008). Cell and molecular biology: Concepts and experiments. John New Jersey: Wiley and Sons.

Keane, M. M., Ettenberg, S. A., Nau, M. M., Russell, E. K., & Lipkowitz, S. (1999). Chemotherapy augments TRAIL-induced apoptosis in breast cell lines. *Cancer Research*, *59*(3), 734–741.

Keen, C. L. (2001). Chocolate: Food as medicine/medicine as food. *Journal of the American College of Nutrition*, *20*, 436S–439S.

Keen, C. L., Holt, R. R., Oteiza, P. I., Fraga, C. G., & Schmitz, H. H. (2005). Cocoa antioxidants and cardiovascular health. *The American Journal of Clinical Nutrition*, *101*(10), S75-S86.

Kerr, J. F. R., Wyllie, A. H., & Currie, A. R. (1972). Apoptosis: A basic biological phenomenon with wideranging implications in tissue kinetics. *British Journal of Cancer*, *26*(4), 239–257.

Khajure, P. V, & Rathod, J. L. (2011). Potential anticancer activity of achantus ilicifolius extracted from the mangroves forest of Karwar , West Coast of India. *World Journal of Science and Technology*, *1*(1), 1–6.

Khalil, H., Bertrand, M. J. M., Vandenabeele, P., & Widmann, C. (2014). Caspase-3 and RasGAP: A stress-sensing survival/demise switch. *Trends in Cell Biology*, *24*(2), 83–89.

Khan, N., Afaq, F., & Mukhtar, H. (2006). Apoptosis by dietary factors: The suicide solution for delaying cancer growth. *Carcinogenesis*, *28*(2), 233–239.

Khoddami, A., Wilkes, M., & Roberts, T. (2013). Techniques for analysis of plant phenolic compounds. *Molecules*, *18*(3), 2328–2375.

© COPYRIGHT UPM

Kim, H., Choi, H.-K., Moon, J. Y., Kim, Y. S., Mosaddik, A., & Cho, S. K. (2011). Comparative antioxidant and antiproliferative activities of red and white pitayas and their correlation with flavonoid and polyphenol content. *Journal of Food Science*, *76*(1), C38–C45.

Kim, J., Kim, J., Shim, J., Lee, C. Y., Lee, K. W., & Lee, H. J. (2014). Cocoa Phytochemicals: Recent advances in molecular mechanisms on health. *Critical Reviews in Food Science and Nutrition*, *54*(11), 1458–1472.

Kim, J., Lee, K. W., & Lee, H. J. (2011). Cocoa (*Theobroma cacao*) Seeds and Phytochemicals in Human Health. In V. Preedy, E. E. Vatson, & V. B. Patel (Ed.), *Nuts and Seeds in Health and Disease Prevention* (pp. 351–360). London, United Kingdom: Academic Press.

Kinghorn, A. D., Chin, Y.-W., & Swanson, S. M. (2009). Discovery of natural product anticancer agents from biodiverse organisms. *Current Opinion in Drug Discovery & Development*, *12*, 189–196.

Kinghorn, A. D., Su, B.-N., Jang, D. S., Chang, L. C., Lee, D., Gu, J.-Q., … Pezzuto, J. M. (2004). Natural inhibitors of carcinogenesis. *Planta Medica*, *70*(8), 691–705.

Kjer, J., Debbab, A., Aly, A. H., & Proksch, P. (2010). Methods for isolation of marine-derived endophytic fungi and their bioactive secondary products. *Nature Protocols*, *5*(3), 479–490.

Klener, P., Anděra, L., Klener, P., Nečas, E., & Živný, J. (2006). Cell death signalling pathways in the pathogenesis and therapy of haematologic malignancies: Overview of therapeutic approaches. *Folia Biologica*, *52*(4), 119–136.

Knowles, B., & Aden, D. (1983). Human hepatoma derived cell line, process for preparation thereof, and uses therefor. *U.S. Paten 4393133*.

Koff, J., Ramachandiran, S., & Bernal-Mizrachi, L. (2015). A time to kill: Targeting apoptosis in cancer. *International Journal of Molecular Sciences*, *16*(2), 2942–2955.

Kominami, K., Nakabayashi, J., Nagai, T., Tsujimura, Y., Chiba, K., Kimura, H., … Sakamaki, K. (2012). The molecular mechanism of apoptosis upon caspase-8 activation: Quantitative experimental validation of a mathematical model. *Biochimica et Biophysica Acta (BBA) - Molecular Cell Research*, *1823*(10), 1825–1840.

Kruidering, M., & Evan, G. (2000). Caspase-8 in apoptosis: The beginning of "the end"? *IUBMB Life*, *50*(2), 85–90.

Kuida, K. (2000). Caspase-9. The International Journal of Biochemistry & Cell Biology, *32*(2), 121–124.

© COPYRIGHT UPM

Kumar, V., Abbas, A. K., Aster, J. C., & Robbins, S. L. (2013). Neoplasia. In V. Kumar, A. K. Abbas, J. C. Aster & S. L. Robbins (Ed.), *Robbins basic pathology* (161-214). Canada: Elsevier Saunders.

Kuno, T., Tsukamoto, T., Hara, A., & Tanaka, T. (2012). Cancer chemoprevention through the induction of apoptosis by natural compounds. *Journal of Biophysical Chemistry*, *03*(02), 156–173.

Kuppusamy, P., Yusoff, M. M., Maniam, G. P., & Govindan, N. (2013). A case study – Regulation and functional mechanisms of cancer cells and control its activity using plants and their derivatives. *Journal of Pharmacy Research*, *6*(8), 884–892.

Kurzawa, M., Filipiak-Szok, A., Kłodzińska, E., & Szłyk, E. (2015). Determination of phytochemicals, antioxidant activity and total phenolic content in *Andrographis* paniculata using chromatographic methods. *Journal of Chromatography B*, *995-996*, 101–106.

Kusuma, I. W., Kuspradini, H., Arung, E. T., Aryani, F., Min, Y.-H., Kim, J.-S., & Kim, Y. (2011). Biological activity and phytochemical analysis of three Indonesian medicinal plants, *Murraya koenigii*, *Syzygium polyanthum* and *Zingiber purpurea*. *Journal of Acupuncture and Meridian Studies*, *4*(1), 75–9.

Laguerre, M., Lecomte, J., & Villeneuve, P. (2007). Evaluation of the ability of antioxidants to counteract lipid oxidation: Existing methods, new trends and challenges. *Progress in Lipid Research*, *46*(5), 244–282.

Lai, C.-S., Mas, R. H. M. H., Nair, N. K., Majid, M. I. A., Mansor, S. M., & Navaratnam, V. (2008). *Typhonium flagelliforme* inhibits cancer cell growth *in vitro* and induces apoptosis: An evaluation by the bioactivity guided approach. *Journal of Ethnopharmacology*, *118*(1), 14–20.

Lamina, S., Ezema, C., Theresa, A., & Anthonia, E. (2013). Effects of free radicals and antioxidants on exercise performance. *Oxidants and Antioxidants in Medical Science*, *2*(2), 83–91.

Lampe, J. W. (2003). Spicing up a vegetarian diet: Chemopreventive effects of phytochemicals. *The American Journal of Clinical Nutrition*, *78*(3 Suppl), 579S–583S.

Lanoue, L., Green, K. K., Kwik-Uribe, C., & Keen, C. L. (2010). Dietary factors and the risk for acute infant leukemia: Evaluating the effects of cocoa-derived flavanols on DNA topoisomerase activity. *Experimental Biology and Medicine*, *235*, 77–89.

Lawal, T. O., Olorunnipa, T. a., & Adeniyi, B. a. (2014). Susceptibility testing and bactericidal activities of *Theobroma cacao* Linn. (cocoa) on *Helicobacter pylori* in an *in vitro* study. *Journal of Herbal Medicine*, *4*(4), 201–207.

© COPYRIGHT UPM

Lee, I.-K., Han, M.-S., Kim, D.-W., & Yun, B.-S. (2014). Phenylpropanoid acid esters from Korean propolis and their antioxidant activities. *Bioorganic & Medicinal Chemistry Letters*, *24*(15), 3503–3505.

Lee, J.-H. (2012). *Lethariella zahlbruckneri* acetone extract-induced apoptosis of MCF-7 human breast cancer cells involves caspase cascade and mitochondria- mediated death signaling. *Journal of Nutrition & Food Sciences*, *01*(S2), 2–7.

Levenson, A. S., & Jordan, V. C. (1997). MCF-7: The first hormone-responsive breast cancer cell line. *Cancer Research*.

Li, C., Schuetz, J. D., & Naren, A. P. (2010). Tobacco carcinogen NNK transporter MRP2 regulates CFTR function in lung epithelia: Implications for lung cancer. *Cancer Letters*, *292*(2), 246–253.

Li, H.-B., Wong, C.-C., Cheng, K.-W., & Chen, F. (2008). Antioxidant properties *in vitro* and total phenolic contents in methanol extracts from medicinal plants. *LWT - Food Science and Technology*, *41*(3), 385–390.

Li, P., Nijhawan, D., Budihardjo, I., Srinivasula, S. M., Ahmad, M., Alnemri, E. S., & Wang, X. (1997). Cytochrome c and dATP-dependent formation of Apaf-1/caspase-9 complex initiates an apoptotic protease cascade. *Cell*, *91*(4), 479–489.

Lichtenstein, A. H., Appel, L. J., Brands, M., Carnethon, M., Daniels, S., Franch, H. A., … Wylie-Rosett, J. (2006). Summary of american heart association diet and lifestyle recommendations revision 2006. *Arteriosclerosis, Thrombosis, and Vascular Biology*, *26*, 2186–2191.

Liebermann, D., Tront, J. S., Sha, X., Mukherjee, K., Mohamed-Hadley, A., & Hoffman, B. (2011). Gadd45 Stress Sensors in Malignancy and Leukemia. *Critical Reviews™ in Oncogenesis*, *16*(1-2), 129–140.

Lim, T. K. (2012). Edible Medicinal And Non Medicinal Plants. New YorK: Springer Science & Business Media.

Lima, L. J. R., Almeida, M. H., Nout, M. J. R., & Zwietering, M. H. (2011). *Theobroma cacao* L., "The food of the Gods": Quality determinants of commercial cocoa beans, with particular reference to the impact of fermentation. *Critical Reviews in Food Science and Nutrition*, *51*, 731–761.

Lin, H.-H., Chan, K.-C., Sheu, J.-Y., Hsuan, S.-W., Wang, C.-J., & Chen, J.-H. (2012). *Hibiscus sabdariffa* leaf induces apoptosis of human prostate cancer cells *in vitro* and *in vivo*. *Food Chemistry*, *132*(2), 880–891.

Lin, Y., Zhang, M., & Barnes, P. F. (1998). Chemokine production by a human alveolar epithelial cell line in response to *Mycobacterium tuberculosis*. *Infection and Immunity*, *66*(3), 1121–1126.

© COPYRIGHT UPM

Liu, H., Zang, C., Fenner, M. H., Possinger, K., & Elstner, E. (2003). PPARgamma ligands and ATRA inhibit the invasion of human breast cancer cells *in vitro*. *Breast Cancer Research and Treatment*, *79*(1), 63–74.

Livak, K. J., & Schmittgen, T. D. (2001). Analysis of relative gene expression data using real-time quantitative PCR and the 2−ΔΔCT method. *Methods*, *25*(4), 402–408.

Llaverias, G., Escolà-Gil, J. C., Lerma, E., Julve, J., Pons, C., Cabré, A., … Blanco-Vaca, F. (2013). Phytosterols inhibit the tumor growth and lipoprotein oxidizability induced by a high-fat diet in mice with inherited breast cancer. *The Journal of Nutritional Biochemistry*, *24*(1), 39–48.

Ludwiczuk, a, Saha, a, Kuzuhara, T., & Asakawa, Y. (2011). Bioactivity guided isolation of anticancer constituents from leaves of *Alnus sieboldiana* (Betulaceae). *Phytomedicine*, *18*(6), 491–8.

Luo, X., Budihardjo, I., Zou, H., Slaughter, C., & Wang, X. (1998). Bid, a Bcl2 Interacting protein, mediates cytochrome c release from mitochondria in response to activation of cell surface death receptors. *Cell*, *94*(4), 481–490.

Ly, J. D., Grubb, D. R., & Lawen, A. (2003). The mitochondrial membrane potential (deltapsi(m)) in apoptosis; an update. *Apoptosis*, *8*(2), 115–28.

Ma, W.-D., Zou, Y.-P., Wang, P., Yao, X.-H., Sun, Y., Duan, M.-H., … Yu, B. (2014). Chimaphilin induces apoptosis in human breast cancer MCF-7 cells through a ROS-mediated mitochondrial pathway. *Food and Chemical Toxicology*, *70*, 1–8.

Ma, X., & Wang, Z. (2009). Anticancer drug discovery in the future: An evolutionary perspective. *Drug Discovery Today*, *14*(23-24), 1136–42.

Majors, B. S., Betenbaugh, M. J., & Chiang, G. G. (2007). Links between metabolism and apoptosis in mammalian cells: Applications for anti-apoptosis engineering. *Metabolic Engineering*, *9*(4), 317–326.

Maltseva, D. V, Khaustova, N. a, Fedotov, N. N., Matveeva, E. O., Lebedev, A. E., Shkurnikov, M. U., … Tonevitsky, A. G. (2013). High-throughput identification of reference genes for research and clinical RT-qPCR analysis of breast cancer samples. *Journal of Clinical Bioinformatics*, *3*(1), 13.

Mandal, S., Yadav, S., Yadav, S., & Nema, R. K. (2009). Antioxidants : A review. *Journal of Chemical and Pharmaceutical Research*, *1*(1), 102–104.

Mann, J. (2002). Natural products in cancer chemotherapy: Past, present and future. *Nature Reviews. Cancer*, *2*(2), 143–8.

© COPYRIGHT UPM

Manorenjitha, M. S., Norita, A. K., Norhisham, S., & Asmawi, M. Z. (2013). GC-MS analysis of bioactive components of Ficus religiosa (Linn.) stem. *International Journal of Pharma and Bio Sciences*, *4*(2), 99–103.

Marcotullio, M. C., Loizzo, M. R., Messina, F., Temperini, A., Tundis, R., Menichini, F., & Curini, M. (2015). Bioassay-guided fractionation of *Euphrasia pectinata* Ten. and isolation of iridoids with antiproliferative activity. *Phytochemistry Letters*, *12*, 252–256.

Marshall, N. J., Goodwin, C. J., & Holt, S. J. (1995). A critical assessment of the use of microculture tetrazolium assays to measure cell growth and function. *Growth Regulation*, *5*, 69–84.

Martin, M. A., Goya, L., & Ramos, S. (2013). Potential for preventive effects of cocoa and cocoa polyphenols in cancer. *Food and Chemical Toxicology*, *56*, 336-51.

Martin, M. C., Allan, L. A., Lickrish, M., Sampson, C., Morrice, N., & Clarke, P. R. (2005). Protein kinase A regulates caspase-9 activation by Apaf-1 downstream of cytochrome c. *Journal of Biological Chemistry*, *280*(15), 15449–15455.

Martínez, R., Torres, P., Meneses, M. A., Figueroa, J. G., Pérez-Álvarez, J. A., & Viuda-Martos, M. (2012). Chemical, technological and *in vitro* antioxidant properties of cocoa (*Theobroma cacao* L.) co-products. *Food Research International*, *49*(1), 39–45.

Mascotti, K., McCullough, J., & Burger, S. R. (2000). HPC viability measurement: Trypan blue versus acridine orange and propidium iodide. *Transfusion*, *40*(6), 693–696.

Mavri, J. (2013). Can the chemical reactivity of an ultimate carcinogen be related to its carcinogenicity? An application to propylene oxide. *Toxicology In Vitro*, *27*(1), 479–485.

Mbaveng, A. T., Kuete, V., Mapunya, B. M., Beng, V. P., Nkengfack, A. E., Meyer, J. J. M., & Lall, N. (2011). Evaluation of four Cameroonian medicinal plants for anticancer, antigonorrheal and antireverse transcriptase activities. *Environmental Toxicology and Pharmacology*, *32*, 162–167.

McCullough, K. D., Martindale, J. L., Klotz, L.-O., Aw, T.-Y., & Holbrook, N. J. (2001). Gadd153 sensitizes cells to endoplasmic reticulum stress by down-regulating Bcl2 and perturbing the cellular redox state. *Molecular and Cellular Biology*, *21*(4), 1249–1259.

McIlwain, D. R., Berger, T., & Mak, T. W. (2013). Caspase functions in cell death and disease. *Cold Spring Harbor Perspectives in Biology*, *5*(4), a008656–a008656.

© COPYRIGHT UPM

Mielgo, A., Torres, V. A., Schmid, M. C., Graf, R., Zeitlin, S. G., Lee, P., … Stupack, D. G. (2009). The death effector domains of caspase-8 induce terminal differentiation. *PLoS ONE*, *4*(11), e7879.

Mignotte, B., & Vayssiere, J.-L. (1998). Mitochondria and apoptosis. *European Journal of Biochemistry*, *252*(1), 1–15.

Miki, Y., Swensen, J., Shattuck-Eidens, D., Futreal, P. A., Harshman, K., Tavtigian, S., … Ding, W. (1994). A strong candidate for the breast and ovarian cancer susceptibility gene BRCA1. *Science (New York, N.Y.)*, *266*(5182), 66–71.

Mita, H., Tsutsui, J., Takekawa, M., Witten, E. A., & Saito, H. (2002). Regulation of MTK1/MEKK4 kinase activity by its N-terminal autoinhibitory domain and GADD45 binding. *Molecular and Cellular Biology*, *22*(13), 4544–55.

Mithraja, M. J., @ Antonisamy, J. M., Mahesh, M., Paul, Z. M., & Jeeva, S. (2011). Phytochemical studies on *Azolla pinnata* R. Br., *Marsilea minuta* L. and *Salvinia molesta* Mitch. *Asian Pacific Journal of Tropical Biomedicine*, *1*(1), S26–S29.

Mokgotho, M. P., Gololo, S. S., Masoko, P., Mdee, L. K., Mbazima, V., Shai, L. J., … Mampuru, L. (2013). Isolation and chemical structural characterisation of a compound with antioxidant activity from the roots of *Senna italica*. *Evidence-Based Complementary and Alternative Medicine*, *2013*, 1–6.

Molinski, T. F., Dalisay, D. S., Lievens, S. L., & Saludes, J. P. (2009). Drug development from marine natural products. *Nature Reviews. Drug Discovery*, *8*(1), 69–85.

Mondal, S., Bandyopadhyay, S., K. Ghosh, M., Mukhopadhyay, S., Roy, S., & Mandal, C. (2012). Natural products: Promising resources for cancer drug discovery. *Anti-Cancer Agents in Medicinal Chemistry*, *12*(1), 49–75.

Moreno-Montoro, M., Olalla-Herrera, M., Gimenez-Martinez, R., Navarro-Alarcon, M., & Rufián-Henares, J. a. (2015). Phenolic compounds and antioxidant activity of Spanish commercial grape juices. *Journal of Food Composition and Analysis*, *38*, 19–26.

Mosmann, T. (1983). Rapid colorimetric assay for cellular growth and survival: Application to proliferation and cytotoxicity assays. *Journal of Immunological Methods*, *65*(1-2), 55–63.

Muchuweti, M., Kativu, E., Mupure, C. H., Chidewe, C., Ndhlala, A. R., & Benhura, M. A. N. (2007). Phenolic composition and antioxidant properties of some spices. *American Journal of Food Technology*, *2*(5), 414–420.

Murray, A. W. (2004). Recycling the cell cycle. *Cell*, *116*(2), 221–234.

© COPYRIGHT UPM

Nagmoti, D. M., Khatri, D. K., Juvekar, P. R., & Juvekar, A. R. (2012). Antioxidant activity and free radical-scavenging potential of *Pithecellobium dulce* Benth seed extracts. *Free Radicals and Antioxidants*, *2*(2), 37–43.

National Cancer Registry. (2011). Malaysia Cancer Statistics-Data and Figure 2007. In Z. A. Omar & N. S. I. Tamin (Eds.), *National Cancer Registry Report* (pp. 1–126). Putrajaya: Ministry of Health.

Nazni, P., & Dharmaligam, R. (2014). Isolation and separation of phenolic compound from coriander flowers. *International Journal of Agricultural and Food Science*, *4*(1), 13–21.

Newman, D. J., Cragg, G. M., & Snader, K. M. (2003). Natural products as sources of new drugs over the period 1981-2002. *Journal of Natural Products*, *66*(7), 1022–37.

Ng, K.-B., Bustamam, A., Sukari, M. A., Abdelwahab, S. I., Mohan, S., Buckle, M. J. C., … Rahman, H. S. (2013). Induction of selective cytotoxicity and apoptosis in human T4-lymphoblastoid cell line (CEMss) by boesenbergin a isolated from *Boesenbergia rotunda* rhizomes involves mitochondrial pathway, activation of caspase 3 and G2/M phase cell cycle arrest. *BMC Complementary and Alternative Medicine*, *13*, 41.

Nguta, J. M., Appiah-Opong, R., Nyarko, A. K., Yeboah-Manu, D., & Addo, P. G. a. (2015). Current perspectives in drug discovery against tuberculosis from natural products. *International Journal of Mycobacteriology*, *4*(3), 165–183.

Nicole Cotelle, B. S. P. (2001). Role of flavonoids in oxidative stress. *Current Topics in Medicinal Chemistry*, *1*(6), 569–590.

Nikoletopoulou, V., Markaki, M., Palikaras, K., & Tavernarakis, N. (2013). Crosstalk between apoptosis, necrosis and autophagy. *Biochimica et Biophysica Acta (BBA) - Molecular Cell Research*, *1833*(12), 3448–3459.

Ningsih, S., Fachrudin, F., Rismana, E., & Purwaningsih, E. H. (2014). Evaluation of antilipid peroxidation activity ff gambir extract on liver homogenat *in vitro*. *International Journal of PharmTech Research*, *6*(3), 982–989.

Nisa, S., Bibi, Y., Waheed, A., Zia, M., Sarwar, S., Ahmed, S., & Chaudhary, M. F. (2011). Evaluation of anticancer activity of *Debregeasia salicifolia* extract against estrogen receptor positive cell line. *African Journal of Biotechnology*, *10*(6), 990–995.

Nsor-Atindana, J., Zhong, F., Mothibe, K. J., Bangoura, M. L., & Lagnika, C. (2012). Quantification of total polyphenolic content and antimicrobial activity of cocoa (*Theobroma cacao* L.) Bean shells. *Pakistan Journal of Nutrition*, *11*(7), 574–579.

© COPYRIGHT UPM

Nurhanan, M. Y., Asiah, O., Ilham, M. A. M., Syarifah, M. M. S., Norhayati, I., & Sahira, H. L. (2008). Antiproliferative activities of 32 Malaysian plant species in breast cancer cell lines. *Journal of Tropical Forest Science*, *20*, 77–81.

O'Donovan, N., Crown, J., Stunell, H., Hill, A. D. K., McDermott, E., O'Higgins, N., & Duffy, M. J. (2003). Caspase 3 in breast cancer. *Clinical Cancer Research*, *9*(2), 738–742.

Ode, O. J., Asuzu, I. U., & Ajayi, I. E. (2011). Research bioassay-guided fractionation of the crude methanol extract of *Cassia singueana* leaves. *J Adv Scient Res Journal of Advanced Scientific Research*, *2*(24), 81–86.

Ogbourne, S. M., & Parsons, P. G. (2014). The value of nature's natural product library for the discovery of new chemical entities: The discovery of ingenol mebutate. *Fitoterapia*, *98C*, 36–44.

Oktay, M., Gülçin, İ., & Küfrevioğlu, Ö. İ. (2003). Determination of *in vitro* antioxidant activity of fennel (*Foeniculum vulgare*) seed extracts. *LWT - Food Science and Technology*, *36*(2), 263–271.

Olguín-Martínez, M., Hernández-Espinosa, D. R., & Hernández-Muñoz, R. (2013). α-Tocopherol administration blocks adaptive changes in cell NADH/NAD+ redox state and mitochondrial function leading to inhibition of gastric mucosa cell proliferation in rats. *Free Radical Biology & Medicine*, *65*, 1090–100.

On, R., To, I., Docking, M., In, T., & Chemistry, M. (2012). International Journal of Drug Research and Technology. *International Journal of Drug Research and Technology*, *2*(2), 189–197.

Osman, H., Nasarudin, R., & Lee, S. L. (2004). Extracts of cocoa (*Theobroma cacao* L.) leaves and their antioxidation potential. *Food Chemistry*, *86*, 41–46.

Othman, A., Ismail, A., Abdul Ghani, N., & Adenan, I. (2007). Antioxidant capacity and phenolic content of cocoa beans. *Food Chemistry*, *100*(4), 1523–1530.

Ovadje, P., Chatterjee, S., Griffin, C., Tran, C., Hamm, C., & Pandey, S. (2011). Selective induction of apoptosis through activation of caspase-8 in human leukemia cells (Jurkat) by dandelion root extract. *Journal of Ethnopharmacology*, *133*(1), 86–91.

Ozsoy, N., Can, A., Yanardag, R., & Akev, N. (2008). Antioxidant activity of *Smilax excelsa* L. leaf extracts. *Food Chemistry*, *110*, 571–583.

Pan, L., Lantvit, D. D., Riswan, S., Kardono, L. B. S., Chai, H.-B., Carcache de Blanco, E. J., … Kinghorn, A. D. (2010). Bioactivity-guided isolation of cytotoxic sesquiterpenes of *Rolandra fruticosa*. *Phytochemistry*, *71*(5-6), 635–640.

© COPYRIGHT UPM

Parker, G., Parker, I., & Brotchie, H. (2006). Mood state effects of chocolate. *Journal of Affective Disorders*, *92*(2-3), 149–159.

Peltomäki, P. (2012). Mutations and epimutations in the origin of cancer. *Experimental Cell Research*, *318*(4), 299–310.

Peng, M., Aryal, U., Cooper, B., & Biswas, D. (2015). Metabolites produced during the growth of probiotics in cocoa supplementation and the limited role of cocoa in host-enteric bacterial pathogen interactions. *Food Control*, *53*, 124–133.

Perez-Berezo, T., Ramirez-Santana, C., Franch, A., Ramos-Romero, S., Castellote, C., Perez-Cano, F. J., & Castell, M. (2012). Effects of a cocoa diet on an intestinal inflammation model in rats. *Experimental Biology and Medicine*. *237*(10), 1181-8.

Pérez-Ríos, M., Ruano-Ravina, A., Etminan, M., & Takkouche, B. (2010). A meta-analysis on wood dust exposure and risk of asthma. *Allergy*, *65*(4), 467–473.

Petti, S., & Scully, C. (2005). Oral cancer: The association between nation-based alcohol-drinking profiles and oral cancer mortality. *Oral Oncology*, *41*(8), 828–834.

Peyressatre, M., Prével, C., Pellerano, M., & Morris, M. (2015). Targeting cyclin-dependent kinases in human cancers: From small molecules to peptide inhibitors. *Cancers*, *7*(1), 179–237.

Pham-Huy, L. A., He, H., & Pham-Huy, C. (2008). Free radicals, antioxidants in disease and health. *International Journal of Biomedical Science*, *4*(2), 89–96.

Pietta, P.-G. (2000). Flavonoids as antioxidants. *Journal of Natural Products*, *63*(7), 1035–1042.

Pingitore, a., Pereira Lima, G. P., Mastorci, F., Quinones, A., Iervasi, G., & Vassalle, C. (2015). Exercise and oxidative stress: Potential effects of antioxidant dietary strategies in sports. *Nutrition*, *31*(7-8), 916–922.

Planas-Silva, M. D., & Weinberg, R. A. (1997). The restriction point and control of cell proliferation. *Current Opinion in Cell Biology*, *9*(6), 768–772.

Pollack, M. S., Heagney, S. D., Livingston, P. O., & Fogh, J. (1981). HLA-A, B, C and DR alloantigen expression on forty-six cultured human tumor cell lines. *Journal of the National Cancer Institute*, *66*(6), 1003–1012.

Polyak, K. (2007). Breast cancer: Origins and evolution. *Journal of Clinical Investigation*, *117*(11), 3155–3163.

© COPYRIGHT UPM

Prakash, H., Doherty, D. O., & Bittenbender, H. C. (2011). Farm and forestry production and marketing profile for cacao. *Permanent Agriculture Resouces*, 1-23.

Prasad, K. N., Hao, J., Shi, J., Liu, T., Li, J., Wei, X., … Jiang, Y. (2009). Antioxidant and anticancer activities of high pressure-assisted extract of longan (*Dimocarpus longan* Lour.) fruit pericarp. *Innovative Food Science & Emerging Technologies*, *10*(4), 413–419.

Prasanna, R., Chinnakonda Chandramoorthy, H., Ramaiyapillai, P., & Sakthisekaran, D. (2011). *In vitro* evaluation of anticancer effect of *Cassia auriculata* leaf extract and curcumin through induction of apoptosis in human breast and larynx cancer cell lines. *Biomedicine & Preventive Nutrition*, *1*(2), 153–160.

Preza, A. M., Jaramillo, M. E., Puebla, A. M., Mateos, J. C., Hernández, R., & Lugo, E. (2010). Antitumor activity against murine lymphoma L5178Y model of proteins from cacao (*Theobroma cacao* L.) seeds in relation with *in vitro* antioxidant activity. *BMC Complementary and Alternative Medicine*, *10*, 61.

Prudnikova, T. Y., Mostovich, L. A., Domanitskaya, N. V, Pavlova, T. V, Kashuba, V. I., Zabarovsky, E. R., & Grigorieva, E. V. (2010). Antiproliferative effect of D-glucuronyl C5-epimerase in human breast cancer cells. *Cancer Cell International*, *10*(1), 27.

Pucciarelli, D. L., & Grivetti, L. E. (2008). The medicinal use of chocolate in early North America. *Molecular Nutrition and Food Research*, *52*(10), 1215-1227.

Puthalakath, H., O'Reilly, L. a., Gunn, P., Lee, L., Kelly, P. N., Huntington, N. D., … Strasser, A. (2007). ER stress triggers apoptosis by activating BH3-only protein Bim. *Cell*, *129*(7), 1337–1349.

Ramasamy, S., Abdul Wahab, N., Zainal Abidin, N., & Manickam, S. (2013). Effect of extracts from *Phyllanthus watsonii* airy shaw on cell apoptosis in cultured human breast cancer MCF-7 cells. *Experimental and Toxicologic Pathology, 65*(3), 341–9.

Ramiro, E., Franch, À., Castellote, C., Andrés-Lacueva, C., Izquierdo-Pulido, M., & Castell, M. (2005). Effect of *Theobroma cacao* flavonoids on immune activation of a lymphoid cell line. *British Journal of Nutrition*, *93*(06), 859.

Ramiro, E., Franch, À., Castellote, C., Pérez-Cano, F., Permanyer, J., Izquierdo-Pulido, M., & Castell, M. (2005). Flavonoids from *Theobroma cacao* down-regulate inflammatory mediators. *Journal of Agricultural and Food Chemistry*, *53*(22), 8506–8511.

Ramljak, D., Romanczyk, L. J., Metheny-Barlow, L. J., Thompson, N., Knezevic, V., Galperin, M., … Dickson, R. B. (2005). Pentameric procyanidin from *Theobroma cacao* selectively inhibits growth of human breast cancer cells. *Molecular Cancer Therapeutics*, *4*, 537–546.

© COPYRIGHT UPM

Ramos, S. (2008). Cancer chemoprevention and chemotherapy: Dietary polyphenols and signalling pathways. *Molecular Nutrition & Food Research*, *52*(5), 507–526.

Ranneh, Y., Ali, F., & Fadel, A. (2014). The antioxidant activity of cocoa polyphenolic extract-treated 3T3 fibroblast cells. *International Journal of Toxicological and Pharmacological Research*, *6*(1), 14–19.

Reddy, A. S., Abd Malek, S. N., Ibrahim, H., & Sim, K. S. (2013). Cytotoxic effect of *Alpinia scabra* (Blume) Náves extracts on human breast and ovarian cancer cells. *BMC Complementary and Alternative Medicine*, *13*, 314.

Reed, J. C. (1997). Bcl-2 family proteins: Regulators of apoptosis and chemoresistance in hematologic malignancies. *Seminars in Hematology*, *34*(4 Suppl 5), 9–19.

Reed, J. C., & Tomaselli, K. J. (2000). Drug discovery opportunities from apoptosis research. *Current Opinion in Biotechnology*, *11*(6), 586-592.

Reidy, J., McHugh, E., & Stassen, L. F. A. (2011). A review of the relationship between alcohol and oral cancer. *The Surgeon*, *9*(5), 278–283.

Reiter, L. S., Kruithof, E. K., Cajot, J. F., & Sordat, B. (1993). The role of the urokinase receptor in extracellular matrix degradation by HT29 human colon carcinoma cells. *International Journal of Cancer*, *53*(3), 444–450.

Rice-Evans, C. A., Miller, N. J., & Paganga, G. (1996). Structure-antioxidant activity relationships of flavonoids and phenolic acids. *Free Radical Biology and Medicine*, *20*(7), 933–956.

Riddle, J. M. (2002). History as a tool in identifying "new" old drugs. *Advances in Experimental Medicine and Biology*, *505*, 89–94.

Rieder, C. L. (2011). Mitosis in vertebrates: The G2/M and M/A transitions and their associated checkpoints. *Chromosome Research*, *19*(3), 291–306.

Riley, B. D., Culver, J. O., Skrzynia, C., Senter, L. A., Peters, J. A., Costalas, J. W., … Trepanier, A. M. (2012). Essential elements of genetic cancer risk assessment, counseling, and testing: Updated recommendations of the National Society of Genetic Counselors. *Journal of Genetic Counseling*, *21*(2), 151–161.

Rimbach, G., Melchin, M., Moehring, J., & Wagner, A. E. (2009). Polyphenols from cocoa and vascular health - A critical review. *International Journal of Molecular Sciences*, *10*(10), 4290-4309.

Riss, T. L., & Moravec, R. A. (2004). Use of multiple assay endpoints to investigate the effects of incubation time, dose of toxin, and plating density in cell-based cytotoxicity assays. *Assay and Drug Development Technologies*, *2*, 51–62.

© COPYRIGHT UPM

Rodríguez-Ramiro, I., Ramos, S., Bravo, L., Goya, L., & Martín, M. Á. (2011). Procyanidin B2 and a cocoa polyphenolic extract inhibit acrylamide-induced apoptosis in human Caco-2 cells by preventing oxidative stress and activation of JNK pathway. *Journal of Nutritional Biochemistry*, *22*, 1186–1194.

Rodríguez-Ramiro, I., Ramos, S., López-Oliva, E., Agis-Torres, A., Gómez-Juaristi, M., Mateos, R., … Martín, M. Á. (2011). Cocoa-rich diet prevents azoxymethane-induced colonic preneoplastic lesions in rats by restraining oxidative stress and cell proliferation and inducing apoptosis. *Molecular Nutrition & Food Research*, *55*(12), 1895–1899.

Rohloff, J. (2015). Analysis of phenolic and cyclic compounds in plants using derivatization techniques in combination with GC-MS-based metabolite profiling. *Molecules*, *20*(2), 3431–3462.

Roleira, F. M. F., Tavares-da-Silva, E. J., Varela, C. L., Costa, S. C., Silva, T., Garrido, J., & Borges, F. (2015). Plant derived and dietary phenolic antioxidants: Anticancer properties. *Food Chemistry*, *183*, 235–258.

Ronco, A. L., Stoll, M., De Stéfani, E., Maisonneuve, J. E., Mendoza, B. A., & Deneo-Pellegrini, H. (2009). Rh factor, family history and risk of breast cancer: A case-control study in Uruguay. *Cancer Detection and Prevention*, *32*, 277–285.

Rosati, A., Graziano, V., De Laurenzi, V., Pascale, M., & Turco, M. C. (2011). BAG3: A multifaceted protein that regulates major cell pathways. *Cell Death & Disease*, *2*(4), e141.

Rubio-Moscardo, F. (2005). Characterization of 8p21.3 chromosomal deletions in B-cell lymphoma: TRAIL-R1 and TRAIL-R2 as candidate dosage-dependent tumor suppressor genes. *Blood*, *106*(9), 3214–3222.

Rufini, A., & Melino, G. (2011). Cell death pathology: The war against cancer. *Biochemical and Biophysical Research Communications*, *414*(3), 445–450.

Rusconi, M., & Conti, A. (2010). Theobroma cacao L., the Food of the Gods: A scientific approach beyond myths and claims. *Pharmacological Research*, *61*(1), 5-13.

Russo, P., Cardinale, A., Margaritora, S., & Cesario, A. (2012). Nicotinic receptor and tobacco-related cancer. *Life Sciences*, *91*, 1087–1092.

Ruzaidi, A., Amin, I., Nawalyah, A. G., Hamid, M., & Faizul, H. A. (2005). The effect of Malaysian cocoa extract on glucose levels and lipid profiles in diabetic rats. *Journal of Ethnopharmacology*, *98*, 55–60.

© COPYRIGHT UPM

Ruzaidi, A. M. M., Abbe, M. M. J., Amin, I., Nawalyah, A. G., & Muhajir, H. (2008). Protective effect of polyphenol-rich extract prepared from Malaysian cocoa (*Theobroma cacao*) on glucose levels and lipid profiles in streptozotocin-induced diabetic rats. *Journal of the Science of Food and Agriculture*, *88*, 1442–1447.

Saeki, H., & Sugimachi, K. (2001). Carcinogenic risk factors. *Journal of the Japan Medical Association*, *125*(3), 297–300.

Saelens, X., Festjens, N., Walle, L. Vande, Gurp, M. van, Loo, G. van, & Vandenabeele, P. (2004). Toxic proteins released from mitochondria in cell death. *Oncogene*, *23*(16), 2861–2874.

Sahil, K., Prashant, B., Akanksha, M., Premjeet, S., & Devashish, R. (2011). Gas chromatography-mass spectrometry : Applications. *International Journal of Pharmaceutical & Biological Archives*, *2*(6), 1544–1560.

Sahin Yaglioglu, A., Demirtas, I., & Goren, N. (2014). Bioactivity-guided isolation of antiproliferative compounds from *Centaurea carduiformis* DC. *Phytochemistry Letters*, *8*(1), 213–219.

Sakahira, H., Enari, M., & Nagata, S. (1998). Cleavage of CAD inhibitor in CAD activation and DNA degradation during apoptosis. *Nature*, *391*(6662), 96–99.

Salem, M. S. Z. (2014). Biological imprinting: Some genetic considerations. *The Egyptian Journal of Medical Human Genetics*. *15*, 219-226.

Sareen, D., van Ginkel, P. R., Takach, J. C., Mohiuddin, A., Darjatmoko, S. R., Albert, D. M., & Polans, A. S. (2006). Mitochondria as the primary target of resveratrol-induced apoptosis in human retinoblastoma cells. *Investigative Ophthalmology & Visual Science*, *47*(9), 3708–16.

Sarkar, F. H., Li, Y., Wang, Z., & Kong, D. (2009). Cellular signaling perturbation by natural products. *Cellular Signalling*, *21*(11), 1541–7.

Scapagnini, G., Davinelli, S., Di Renzo, L., De Lorenzo, A., Olarte, H., Micali, G., … Gonzalez, S. (2014). Cocoa bioactive compounds: Significance and potential for the maintenance of skin health. *Nutrients*, *6*(8), 3202–3213.

Scherer, W. F. (1953). Studies on the propagation *in vitro* of poliomyelitis viruses: IV. Viral multiplication in a stable strain of human malignant epithelial cells (strain HeLa) derived from an epidermoid carcinoma of the cervix. *Journal of Experimental Medicine*, *97*(5), 695–710.

Schewe, T., Sadik, C., Klotz, L. O., Yoshimoto, T., Kühn, H., & Sies, H. (2001). Polyphenols of cocoa: Inhibition of mammalian 15-lipoxygenase. *Biological Chemistry*, *382*, 1687–1696.

© COPYRIGHT UPM

Schinella, G., Mosca, S., Cienfuegos-Jovellanos, E., Pasamar, M. Á., Muguerza, B., Ramón, D., & Ríos, J. L. (2010). Antioxidant properties of polyphenol-rich cocoa products industrially processed. *Food Research International*, *43*, 1614–1623.

Schröterová, L., Králová, V., Voráčová, A., Hašková, P., Rudolf, E., & Červinka, M. (2009). Antiproliferative effects of selenium compounds in colon cancer cells: Comparison of different cytotoxicity assays. *Toxicology In Vitro*, *23*, 1406–1411.

Schwarz, E., Freese, U. K., Gissmann, L., Mayer, W., Roggenbuck, B., Stremlau, A., & zur Hausen, H. (1985). Structure and transcription of human papillomavirus sequences in cervical carcinoma cells. *Nature*, *314*(6006), 111–114.

Seitz, H. K., Stickel, F., & Homann, N. (2004). Pathogenetic mechanisms of upper aerodigestive tract cancer in alcoholics. *International Journal of Cancer*, *108*(4), 483–487.

Selmi, C., Cocchi, C. A., Lanfredini, M., Keen, C. L., & Gershwin, M. E. (2008). Chocolate at heart: The anti-inflammatory impact of cocoa flavanols. *Molecular Nutrition and Food Research*, *52*(11), 1340-1348.

Serrano, F. a, Matsuo, A. L., Monteforte, P. T., Bechara, A., Smaili, S. S., Santana, D. P., … Rodrigues, E. G. (2011). A cyclopalladated complex interacts with mitochondrial membrane thiol-groups and induces the apoptotic intrinsic pathway in murine and cisplatin-resistant human tumor cells. *BMC Cancer*, *11*(1), 296.

Shabbir, M., Khan, M., & Saeed, N. (2013). Assessment of phytochemicals, antioxidant, anti-lipid peroxidation and anti-hemolytic activity of extract and various fractions of *Maytenus royleanus* leaves. *BMC Complementary and Alternative Medicine*, *13*(1), 143.

Sharma, J. V. C., Chandra, B., Chakraborty, R., & Chanda, H. (2011). Anticancer activity of aqueous extract of roots of *Glochidion zeylanicum* (Gaertn.). *Journal of Pharmaceutical and Biomedical Sciences Anticancer*, *06*(06), 1–4.

Sharma, S. K., Singh, L., & Singh, S. (2013). A review on medicinal plants having antioxidant potential. *Indian Journal of Research in Pharmacy and Biotechnology*, *1*, 404–409.

Sheela, D., & Uthayakumari, F. (2013). GC-MS analysis of bioactive constituents from Coastal Sand Dune Taxon- *Sesuvium portulacastrum* (L.). *Bioscience Discovery*, *4*(1), 47–53.

Shinde, A., Ganu, J., & Naik, P. (2012). Effect of free radicals & antioxidants on oxidative stress : A review. *Journal of Dental & Allied Sciences*, *1*(2), 63–66.

© COPYRIGHT UPM

Shoeb, M. (2006). Minireview Anticancer agents from medicinal plants. *Bangladesh Journal of Pharmacology*, *1*, 35–41.

Sims, A., Howell, A., Howell, S., & Clarke, R. (2007). Origins of breast cancer subtypes and therapeutic implications. *Nature Clinical Practice Oncology*, *4*, 516–25.

Sindhi, V., Gupta, V., Sharma, K., Bhatnagar, S., Kumari, R., & Dhaka, N. (2013). Potential applications of antioxidants-A review. *Journal of Pharmacy Research*, *7*, 828–835.

Singh, S., Chitkara, D., Mehrazin, R., Behrman, S. W., Wake, R. W., & Mahato, R. I. (2012). Chemoresistance in prostate cancer cells is regulated by miRNAs and Hedgehog pathway. *PLoS ONE*, *7*(6), e40021.

Singh, S. R. (2013). Cancer stem cells: Recent developments and future prospects. *Cancer Letters*, *338*(1), 1–2.

Sinicropi, D., Cronin, M., & Liu, M.-L. (2007). Gene expression pofiling utilizing microarray technology and RT-PCR. In M. Ferrari, M. Ozkan, & M. Heller (Eds.), *BioMEMS and Biomedical Nanotechnology* (pp. 23–46). Heidelberg, Germany: Springer-Verlag.

Sisein, E. A. (2014). Review article biochemistry of free radicals and antioxidants. *Scholars Academic Journal of Biosciences*, *2*(2), 110–118.

Skaar, J. R., & Pagano, M. (2009). Control of cell growth by the SCF and APC/C ubiquitin ligases. *Current Opinion in Cell Biology*, *21*(6), 816–824.

Sneddon, J., Masuram, S., & Richert, J. C. (2007). Gas chromatography-mass spectrometry-basic principles, instrumentation and selected applications for detection of organic compounds. *Analytical Letters*, *40*(6), 1003–1012.

Soto-Vaca, A., Gutierrez, A., Losso, J. N., Xu, Z., & Finley, J. W. (2012). Evolution of phenolic compounds from color and flavor problems to health benefits. *Journal of Agricultural and Food Chemistry*, *60*(27), 6658–6677.

Spencer, C. M., & Faulds, D. (1994). Paclitaxel. A review of its pharmacodynamic and pharmacokinetic properties and therapeutic potential in the treatment of cancer. *Drugs*, *48*(5), 794–847.

Sreelatha, S., Jeyachitra, A., & Padma, P. R. (2011). Antiproliferation and induction of apoptosis by *Moringa oleifera* leaf extract on human cancer cells. *Food and Chemical Toxicology*, *49*(6), 1270–5.

Sreelekshmi, R., Latha, P. G., Arafat, M. M., Shyamal, S., Shine, V. J., Anuja, G. I., … Rajasekharan, S. (2007). Anti-inflammatory, analgesic and anti-lipid peroxidation studies on stem bark of *Ficus religiosa* Linn. *Natural Product Radiance*, *6*(5), 377–381.

© COPYRIGHT UPM

Stennicke, H. R., & Salvesen, G. S. (1998). Properties of the caspases. *Biochimica et Biophysica Acta (BBA) - Protein Structure and Molecular Enzymology*, *1387*(1-2), 17–31.

Sugimura, T. (1998). Cancer prevention : Past , present , future. *Mutation Research*, *402*, 7–14.

Suliman, A., Lam, A., Datta, R., & Srivastava, R. K. (2001). Intracellular mechanisms of TRAIL: Apoptosis through mitochondrial-dependent and - independent pathways. *Oncogene*, *20*(17), 2122–2133.

Summa, C., Raposo, F. C., McCourt, J., Scalzo, R. Lo, Wagner, K. H., Elmadfa, I., & Anklam, E. (2006). Effect of roasting on the radical scavenging activity of cocoa beans. *European Food Research and Technology*, *222*, 368–375.

Sun, L., Wang, H., Wang, Z., He, S., Chen, S., Liao, D., … Wang, X. (2012). Mixed lineage kinase domain-like protein mediates necrosis signaling downstream of RIP3 kinase. *Cell*, *148*(1-2), 213–227.

Sunayama, J., Ando, Y., Itoh, N., Tomiyama, A., Sakurada, K., Sugiyama, A., … Kitanaka, C. (2004). Physical and functional interaction between BH3-only protein Hrk and mitochondrial pore-forming protein p32. *Cell Death and Differentiation*, *11*(7), 771–781.

Syam, S., Bustamam, A., Abdullah, R., Sukari, M. A., Hashim, N. M., Ghaderian, M., … Ali, H. M. (2014). β-Mangostin induces p53-dependent G2/M cell cycle arrest and apoptosis through ROS mediated mitochondrial pathway and NfkB suppression in MCF-7 cells. *Journal of Functional Foods*, *6*(1), 290–304.

Takahashi, T., Taguchi, H., Yamaguchi, H., Osaki, T., Sato, S., Kamei, M., … Kamiya, S. (1999). Antibacterial effects of cacao mass on enterohemorrhagic *Escherichia coli* (O157:H7). *Kansenshogaku Zasshi*, *73*, 694–701.

Talhaoui, N., Gómez-Caravaca, A. M., León, L., De la Rosa, R., Segura-Carretero, A., & Fernández-Gutiérrez, A. (2014). Determination of phenolic compounds of "Sikitita" olive leaves by HPLC-DAD-TOF-MS. Comparison with its parents "Arbequina" and "Picual" olive leaves. *Food Science and Technology*, *58*(1), 28–34.

Tambellini, N., Zaremberg, V., Turner, R., & Weljie, A. (2013). Evaluation of extraction protocols for simultaneous polar and non-polar yeast metabolite analysis using multivariate projection methods. *Metabolites*, *3*(3), 592–605.

Tamura, R. E., de Vasconcellos, J. F., Sarkar, D., Libermann, T. A., Fisher, P. B., & Zerbini, L. F. (2012). GADD45 proteins: Central players in tumorigenesis. *Current Molecular Medicine*, *12*(5), 634–51.

© COPYRIGHT UPM

Tan, M. L., Sulaiman, S. F., Najimuddin, N., Samian, M. R., & Muhammad, T. S. T. (2005). Methanolic extract of P*ereskia bleo* (Kunth) DC. (Cactaceae) induces apoptosis in breast carcinoma, T47-D cell line. *Journal of Ethnopharmacology*, *96*(1-2), 287–294.

Tang, E. L. H., Rajarajeswaran, J., Fung, S. Y., & Kanthimathi, M. S. (2013). Antioxidant activity of *Coriandrum sativum* and protection against DNA damage and cancer cell migration. *BMC Complementary and Alternative Medicine*, *13*, 347.

Taylor, R. C., Cullen, S. P., & Martin, S. J. (2008). Apoptosis: Controlled demolition at the cellular level. *Nature Reviews Molecular Cell Biology*, *9*(3), 231–241.

Thorburn, A. (2004). Death receptor-induced cell killing. *Cellular Signalling*, *16*(2), 139–144.

Trainer, D. L., Kline, T., McCabe, F. L., Faucette, L. F., Feild, J., Chaikin, M., … Li, D. J. (1988). Biological characterization and oncogene expression in human colorectal carcinoma cell lines. *International Journal of Cancer. Journal International Du Cancer*, *41*(2), 287–296.

Tyagi, S. N., Saxena, A., & Patel, B. D. (2010). *In vitro* antioxidant activity of methanolic and aqueous extract of *Flacourtia indica* Merr. *American-Eurasian Journal of Scientific Research*, *5*(3), 201–206.

Uddin, G., Rauf, A., Siddiqui, B. S., & Shah, S. Q. (2011). Cytotoxic effects of Bangladeshi medicinal plant extracts. *Evidence-Based Complementary and Alternative Medicine*, *2011*, 578092.

Uibel, F., Mühleisen, A., Köhle, C., Weimer, M., Stummann, T. C., Bremer, S., & Schwarz, M. (2010). ReProGlo: A new stem cell-based reporter assay aimed to predict embryotoxic potential of drugs and chemicals. *Reproductive Toxicology*, *30*, 103–112.

Ujang, Z. B., Subramaniam, T., Diah, M. M., Wahid, H. B., Abdullah, B. B., Rashid, A. H. B. A., & Appleton, D. (2013). Bioassay guided fractionation and purification of natural bioactives obtained from *Alpinia conchigera* water extract with melanin inhibition activity. *Journal of Biomaterials and Nanobiotechnology*, *04*(03), 265–272.

Umthong, S., Phuwapraisirisan, P., Puthong, S., & Chanchao, C. (2011). *In vitro* antiproliferative activity of partially purified *Trigona laeviceps* propolis from Thailand on human cancer cell lines. *BMC Complementary and Alternative Medicine*, *11*(1), 37.

Unterthiner, T., Mayr, A., Klambauer, G., Steijaert, M., Wegner, J. K., & Ceulemans, H. (2014). Deep learning as an opportunity in virtual screening. *Deep Learning and Representation Learning Workshop: NIPS 2014*, 1–9.

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Urrego, D., Tomczak, A. P., Zahed, F., Stühmer, W., & Pardo, L. a. (2014). Potassium channels in cell cycle and cell proliferation. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences*, *369*(1638), 20130094.

Van de Craen, M., Declercq, W., Van den brande, I., Fiers, W., & Vandenabeele, P. (1999). The proteolytic procaspase activation network: An *in vitro* analysis. *Cell Death and Differentiation*, *6*(11), 1117–1124.

Van Meerloo, J., Kaspers, G. J. L., & Cloos, J. (2011). Cell sensitivity assays: The MTT assay. *Methods in Molecular Biology*, *731*, 237–245.

Vandenabeele, P., Galluzzi, L., Vanden Berghe, T., & Kroemer, G. (2010). Molecular mechanisms of necroptosis: An ordered cellular explosion. *Nature Reviews Molecular Cell Biology*, *11*(10), 700–714.

Vardiman, J. W., Harris, N. L., & Brunning, R. D. (2002). The World Health Organization (WHO) classification of the myeloid neoplasms. *Blood*, *100*(7), 2292-302.

Vermeulen, K., Van Bockstaele, D. R., & Berneman, Z. N. (2003). The cell cycle: A review of regulation, deregulation and therapeutic targets in cancer. *Cell Proliferation*, *36*(3), 131–149.

Vijayarathna, S., & Sasidharan, S. (2012). Cytotoxicity of methanol extracts of *Elaeis guineensis* on MCF-7 and Vero cell lines. *Asian Pacific Journal of Tropical Biomedicine*, *2*(10), 826–9.

Vistica, D. T., Skehan, P., Scudiero, D., Monks, A., Pittman, A., & Boyd, M. R. (1991). Tetrazolium-based assays for cellular viability: A critical examination of selected parameters affecting formazan production. *Cancer Research*, *51*, 2515–2520.

Wadood, A. (2013). Phytochemical analysis of medicinal plants occurring in local area of Mardan. *Biochemistry & Analytical Biochemistry*, *02*(04), 2–5.

Wajant, H., Pfizenmaier, K., & Scheurich, P. (2002). TNF-related apoptosis inducing ligand (TRAIL) and its receptors in tumor surveillance and cancer therapy. *Apoptosis*, *7*(5), 449-459.

Wang, Y., Qian, J., Lin, J., Yao, D., Qian, Z., Zhu, Z., & Li, J. (2010). Methylation status of DDIT3 gene in chronic myeloid leukemia. *Journal of Experimental & Clinical Cancer Research*, *29*(1), 54.

Wangcharoen, W., & Morasuk, W. (2007). Antioxidant capacity and phenolic content of chilies. *Kasetsart Journal (Nat. Sci)*, *41*, 561–569.

© COPYRIGHT UPM

Warner, D. S., Sheng, H., & Batinić-Haberle, I. (2004). Oxidants, antioxidants and the ischemic brain. *The Journal of Experimental Biology*, *207*(18), 3221–3231.

Watt, W., Koeplinger, K. A., Mildner, A. M., Heinrikson, R. L., Tomasselli, A. G., & Watenpaugh, K. D. (1999). The atomic-resolution structure of human caspase-8, a key activator of apoptosis. *Structure*, *7*(9), 1135–1143.

Weinberg, F., & Chandel, N. S. (2009). Mitochondrial metabolism and cancer. *Annals of the New York Academy of Sciences*, *1177*, 66–73.

Weinberg, R. (2013). *The Biology of Cancer, Second Edition*. New York: Garland Science.

Weisburger, J. H. (2001). Chemopreventive effects of cocoa polyphenols on chronic diseases. *Experimental Biology and Medicine*, *226*, 891–897.

Wilcken, N., Zdenkowski, N., White, M., Snyder, R., Pittman, K., Mainwaring, P., … Bell, R. (2014). Systemic treatment of HER2-positive metastatic breast cancer: A systematic review. *Asia-Pacific Journal of Clinical Oncology*, *10*, 1–14.

Wiley, J. (2008). Chocolate in Mesoamerica: A cultural history of cacao (review). *Journal of Latin American Geography*, *7*(1), 229–231.

Willis, S. N., & Adams, J. M. (2005). Life in the balance: How BH3-only proteins induce apoptosis. *Current Opinion in Cell Biology*, *17*(6), 617–625.

Wollenweber, E., & H. Dietz, V. (1981). Occurrence and distribution of free flavonoid aglycones in plants. *Phytochemistry*, *20*(5), 869–932.

Wollgast, J., & Anklam, E. (2000a). Polyphenols in chocolate: Is there a contribution to human health? *Food Research International*, *33*, 449–459.

Wollgast, J., & Anklam, E. (2000b). Review on polyphenols in *Theobroma cacao*: Changes in composition during the manufacture of chocolate and methodology for identification and quantification. *Food Research International*, *33*, 423–447.

Won, Y.-S., Lee, J.-H., Kwon, S.-J., Kim, J.-Y., Park, K.-H., Lee, M.-K., & Seo, K.-I. (2014). α-Mangostin-induced apoptosis is mediated by estrogen receptor α in human breast cancer cells. *Food and Chemical Toxicology*, *66*, 158–65.

Wong, M., & Medrano, J. (2005). Real-time PCR for mRNA quantitation. *BioTechniques*, *39*(1), 75–85.

Wong, R. S. (2011). Apoptosis in cancer: From pathogenesis to treatment. *Journal of Experimental & Clinical Cancer Research*, *30*(1), 87.

© COPYRIGHT UPM

World Health Organization. Definition, diagnosis and classification of diabetes mellitus and its complication. Part 1: Diagnosis and classification of ciabetes cellitus. Geneva, 1999.

Würstle, M. L., Laussmann, M. A., & Rehm, M. (2012). The central role of initiator caspase-9 in apoptosis signal transduction and the regulation of its activation and activity on the apoptosome. *Experimental Cell Research*, *318*(11), 1213–1220.

Xu, M., Chen, X., Chen, N., Nie, L., Li, X., Li, Q., … Zhou, Q. (2013). Synergistic silencing by promoter methylation and reduced AP-2α transactivation of the proapoptotic HRK gene confers apoptosis resistance and enhanced tumor growth. *The American Journal of Pathology*, *182*(1), 84–95.

Yan-Fang, T., Dong, W., Li, P., Wen-Li, Z., Jun, L., Na, W., … Jian, P. (2012). Analyzing the gene expression profile of pediatric acute myeloid leukemia with real-time PCR arrays. *Cancer Cell International*, *12*(1), 40.

Yang, X., Tian, Z., Gou, W., Takahashi, H., Yu, M., Xing, Y., … Zheng, H. (2013). Bag-3 expression is involved in pathogenesis and progression of colorectal carcinomas. *Histology and Histopathology*, *28*(9), 1147–56.

Yazdani, D., Tan, Y. H., Zainal Abidin, M. A., & Jaganath, I. B. (2011). A review on bioactive compounds isolated from plants against plant pathogenic fungi. *Journal of Medicinal Plants Research*, *5*(30), 6584–6589.

Yeo, C. I., Ooi, K. K., Akim, A. M., Ang, K. P., Fairuz, Z. A., Halim, S. N. B. A., … Tiekink, E. R. T. (2013). The influence of R substituents in triphenylphosphinegold(I) carbonimidothioates, Ph3PAu[SC(OR)=NPh] (R=Me, Et and iPr), upon *in vitro* cytotoxicity against the HT-29 colon cancer cell line and upon apoptotic pathways. *Journal of Inorganic Biochemistry*, *127*, 24–38.

Ying, J., Srivastava, G., Hsieh, W.-S., Gao, Z., Murray, P., Liao, S.-K., … Tao, Q. (2005). The stress-responsive gene GADD45G is a functional tumor suppressor, with its response to environmental stresses frequently disrupted epigenetically in multiple tumors. *Clinical Cancer Research*, *11*(18), 6442–9.

Yoon, J. H., & Baek, S. J. (2005). Molecular targets of dietary polyphenols with anti-inflammatory properties. *Yonsei Medical Journal*, *46*(5), 585.

Yoshikawa, N., Yamada, J., Tsuno, N. H., Okaji, Y., Kawai, K., Tsuchiya, T., … Takahashi, K. (2009). Plaunotol and geranylgeraniol induce caspase-mediated apoptosis in colon cancer. *The Journal of Surgical Research*, *153*(2), 246–53.

Yu, B., Zhang, B., Zhou, W., Wang, Q., Yuan, P., Yang, Y., & Kong, J. (2015). Assessing apoptosis gene expression profiling with a PCR array in the Hippocampus of Ts65Dn Mice. *BioMed Research International*, *2015*, 1–8.

© COPYRIGHT UPM

Zaid, H., Rayan, A., Said, O., & Saad, B. (2010). Cancer Treatment by Greco-Arab and Islamic Herbal Medicine. *The Open Nutraceuticals Journal*, *3*(1), 203-212.

Zakaria, Z. A., Rofiee, M. S., Mohamed, A. M., Teh, L. K., & Salleh, M. Z. (2011). *In vitro* antiproliferative and antioxidant activities and total phenolic contents of the extracts of *Melastoma malabathricum* leaves. *Journal of Acupuncture and Meridian Studies*, *4*(4), 248–56.

Zhang, D.-W., Shao, J., Lin, J., Zhang, N., Lu, B.-J., Lin, S.-C., … Han, J. (2009). RIP3, an energy metabolism regulator that switches TNF-induced cell death from apoptosis to necrosis. *Science*, *325*(5938), 332–336.

Zhang, L., Ravipati, A. S., Koyyalamudi, S. R., Jeong, S. C., Reddy, N., Smith, P. T., … Wu, M. J. (2011). Antioxidant and anti-inflammatory activities of selected medicinal plants containing phenolic and flavonoid compounds. *Journal of Agricultural and Food Chemistry*, *59*, 12361–12367.

Zheng, Y., Le, V., Cheng, Z., Xie, S., Li, H., Tian, J., & Liu, J. (2013). Development of rapid and highly sensitive HSPA1A promoter-driven luciferase reporter system for assessing oxidative stress associated with low-dose photodynamic therapy. *Cell Stress and Chaperones*, *18*(2), 203–213.

Zou, H., Henzel, W. J., Liu, X., Lutschg, A., & Wang, X. (1997). Apaf-1, a human protein homologous to C. elegans CED-4, participates in cytochrome c–dependent activation of caspase-3. *Cell*, *90*(3), 405–413.

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LIST OF PUBLICATIONS

Zainal, B., Abdah, M. A., Taufiq-Yap, Y. H., Roslida, A. H., & Rosmin, K. (2014). *In vitro* antioxidant and antiproliferative activities of methanolic plant part extracts of *Theobroma cacao*. *Molecules*, *19*(11), 18317–31.

Zainal, B., Abdah, M. A., Taufiq-Yap, Y. H., & Rosmin, K. (2014). Anti-cancer agents from non-edible parts of *Theobroma cacao*. *Natural Products Chemistry & Research*, *2*(4), 1–8.

Zainal, B., Abdah, M. A., Taufiq-Yap, Y. H., & Rosmin, K. (2014). Anti-cancer agents from non-edible parts of *Theobroma cacao*. Oral presented at the Pharmacognosy, Phytochemistry & Natural Products International Conference 2014 from August 25-27, 2014 in Beijing, China.

Hazirah, A. R., Zainal, B, & Abdah, M. A. (2013). Total phenolic content, antioxidant and cytotoxic activity of cocoa (*Theobroma cacao* L.) polyphenols extracts on cancer cell lines. *Malaysian Journal of Nutrition 19*(2), 223-232.

Zainal, B., Abdah, M. A., Taufiq-Yap, Y. H., & Rosmin, K. (2016). *Theobroma cacao*: Review of the extraction, isolation, and bioassay of its potential anti-cancer compounds. *Tropical Life Sciences Research 27*(1), 21-42.

Zainal, B., Abdah, M. A., Taufiq-Yap, Y. H., Rosmin, K. & Mohd Redzuan, S. (2016). *In vitro* antioxidant and antiproliferative activities of non-edible plant tissue extracts from *Theobroma cacao*. Poster presented at the International Conference On Natural Products 2016 from March 14-17, 2016 in Terengganu, Malaysia.

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