Biomedical Properties of a Natural Dietary Plant Metabolite, Zerumbone, in Cancer Therapy and Chemoprevention Trials

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1 Introduction

Medical herbs and plant foods such as fruits, vegetables, and spices contain many biologically active phytochemicals that have various health-promoting effects (Kwon et al., 2007). The Zingiberaceae family found in tropical and subtropical regions of the world and approximately 161 species from 18 genera of this family are found in Peninsular Malaysia (Larsen et al., 1999).

*Zingiber zerumbet* (L.) Smith tree (Figure 1A) belonging to this family, is an edible ginger, originating in South-East Asia, and has been cultivated for thousands of years as a spice and for medical purposes (Yob et al., 2011). Although this plant is known to be indigenous to India and the Malay Peninsula, it is nonetheless distributed in many other countries including Indonesia, China, Bangladesh, Vietnam, Japan, Burma, Nepal, Srilanka, Jamaica, and Nigeria, and other parts of the globe (Basak et al., 2010). This herbal plant is popularly referred to as the pinecone, wild ginger, Asian ginger, or shampoo ginger. It is also called by many other names in different countries, such as
Figure 1: *Zingiber zerumbet* tree (A) and inflorescences (B).

*Z. zerumbet* contains several types of phytochemical and is considered as one of the most widely-used traditional dietary condiments in various cuisines and beverages throughout Asia, although the essential oil is also used as perfume and in other toiletry articles (Prakash et al., 2011b). Besides its extensive use as a spice, the rhizome particularly is been used in traditional oriental medicine for many human disorders, especially in the treatment of a variety of digestive conditions (Jang et al., 2004; Prakash et al., 2011a). The rhizome and oils from the leaves of *Z. zerumbet* has been subjected to close chemical scrutiny for their medicinal value (Jang and Seo, 2005).

Ginger is generally recognized as safe and is used traditionally in local folk medicine for treatment of nausea, hangovers, asthma, morning and motion sickness, loss of appetite, dyspepsia, diarrhoea, colic, cramp, stomach upset, sprain, worm infestation in children, cough and cold, flu, sinusitis, catarrh, congestion, sore throat, migraine-headache, toothache, diabetes, bruising, carbuncles, fracture, swelling, rheumatism, arthritis, and chills and fever (Sultana et al., 2010; Butt and Sultan, 2011; Sahebkar, 2011).

Presently, rhizome’s extract has been extensively studied for its effectiveness in a broad range of biological activities including antimicrobial (Kader et al., 2011), antipyretic (Somchit et al., 2005), antispasmodic and anticonvulsant (Yob et al., 2011), antiulcer (Al-Amin et al., 2012), antioxidant (Habsah et al., 2000), antidiabetic (Tzeng et al., 2013a), antitumor (Elhassan and Syam, 2008), anticancer (Rashid and Pihie, 2005;
Rasedee et al., 2013), anti-inflammatory (Sulaiman et al., 2010; Zakaria et al., 2010), antinociceptive and analgesic (Sulaiman et al., 2009a; Somchit et al., 2012), antiallergenic (Tewtrakul and Subhadhirasakul, 2007), antiangiogenic (Rhode et al., 2007), antiproteinogenic (Tzeng and Liu, 2013), antiplatelet aggregation and anticoagulant (Jantan et al., 2008), and hepatoprotective effects (El-Sharaky et al., 2009). Other studies have shown that consuming the rhizome also exhibits hypolipidemic effect by reducing intestinal cholesterol absorption, which makes it useful for treating heart diseases (Sharma et al., 1996; Bhandari et al., 1998).

The essential oil of *Z. zerumbet* rhizome (Figure 2A) contains approximately 86% sesquiterpenoids (Srivastava et al., 2001) while the leaf and rhizome oils (Figure 2B) of this plant contain a complex mixture of 29 and 30 compounds, respectively (Bhuiyan et al., 2008). Many of these compounds are in trace amounts with great variations in their chemical compositions.

![Figure 2: Zingiber zerumbet rhizome (A) and essential oil (B)](image)

Zerumbone (Figure 3A) was first isolated from the essential volatile oil of rhizomes of *Z. zerumbet* in 1956 (Dev, 1960), while its chemical structure (Figure 3B) was determined in 1960 and later characterized by NMR and X-ray (Dev et al., 1968).

Zerumbone possesses three double bonds, two conjugated and one isolated, as well as double conjugated carbonyl group in the 11-membrane ring structure (Kitayama et al., 1999). The chemical characteristics of ZER are presented in Table 1 (Damodaran and Dev, 1965; Subba Rao et al., 1967; Chhabra et al., 1975; Hall et al., 1981; Kitayama et al., 2006).

## 2 Plant Sources of Zerumbone

Early investigations in different parts of the world showed that 12.6 to 73.1% of ZER
Figure 3: Zerumbone pure crystals (A) and Chemical structure (B).

<table>
<thead>
<tr>
<th>Characters</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Natural occurrence</td>
<td><em>Zingiber</em> species</td>
</tr>
<tr>
<td>Chemical class</td>
<td>Sesquiterpene</td>
</tr>
<tr>
<td>Chemical formula</td>
<td>(2E,6E,10E)-2,6,9,9-tetramethylcycloundeca-2,6,10-trien-1-one</td>
</tr>
<tr>
<td>Molecular formula</td>
<td>C₁₅H₂₂O</td>
</tr>
<tr>
<td>Chemical structure</td>
<td>Three double bond (two conjugated and one isolated), α,β-unsaturated carbonyl group, and a double conjugated carbonyl group in 11-membered ring structure</td>
</tr>
<tr>
<td>Molecular weight</td>
<td>218.3 dalton</td>
</tr>
<tr>
<td>Flashing point</td>
<td>272˚F</td>
</tr>
<tr>
<td>Boiling point</td>
<td>321–322˚C at 760 mmHg</td>
</tr>
<tr>
<td>Melting point</td>
<td>65.3˚C</td>
</tr>
<tr>
<td>Vapor pressure</td>
<td>0.000295mm/Hg at 25˚C</td>
</tr>
<tr>
<td>Purity</td>
<td>92–100%</td>
</tr>
<tr>
<td>Appearance</td>
<td>Solid white crystals or powder</td>
</tr>
<tr>
<td>Short term storage</td>
<td>+4˚C</td>
</tr>
<tr>
<td>Stability</td>
<td>Stable for at least 2 years when stored at –20˚C</td>
</tr>
<tr>
<td>Solubility</td>
<td>Completely soluble in ethanol, DMSO, while solubility in water is approximately 1.296 mg/L at 25˚C</td>
</tr>
<tr>
<td>Extraction</td>
<td>Mainly isolated from fresh rhizomes by hydrodistillation (steam distillation) and recrystalization methods</td>
</tr>
<tr>
<td>Usage</td>
<td>For researches and medical purposes, not for flavor or fragrance</td>
</tr>
</tbody>
</table>

Table 1: Characteristic features of zerumbone.
in Z. zerumbet are in the rhizome oils (Baby et al., 2009). The Kerala state in the South Indian accessions reported that in Z. zerumbet, 76.3 to 84.8% of its ZER content is also in the rhizome oils (Baby et al., 2009). On the other hand a Silviculture farm in India reported 1.81% ZER content was found in the rhizome, 0.16% in the root, 0.09% in the leaf and 0.03% in the flower of Z. zerumbet (Rout et al., 2009). The Penang Malaysian accession recorded the content of ZER in the plant at 68.9% (Baby et al., 2009). Another study conducted in the state of Selangor, Malaysia showed the ZER content of Z. zerumbet is 1.3 g/kg rhizome (Rasedee et al., 2013). The oils of Z. zerumbet from Tahiti Island and Vietnam were also found to be rich in ZER at 65.3 and 72.3, respectively (Duñig et al., 1993; Lechat et al., 1993). In Vietnam, ZER was also isolated from the rhizomes of the Vietnamese Curcuma zedoaria (Berg.) Roscoe (Giang et al., 2009).

Other reports on the ginger plant include that by Chane-Ming et al. (2003) and Bhuiyan et al. (2008) each showing the rhizome to contain approximately 37% of the plant ZER content. The differences in ZER content in the plant is not due to geographic or ecological variations, but instead because of differences in ZER chemotype (Yob et al., 2011).

Other ginger plant species with ZER among their constituents include the Zingiber amaricans (Riyanto, 2003), Zingiber ottensii Valeton (Sirat and Nordin, 1994), Zingiber aromaticum (17.72%) (S Muhammad, 2009), Zingiber cassumunar Roxb (1%) (Kishore and Dwivedi, 1992), Zingiber ottensii (Thubthimthed et al., 2003) and Zingiber montanum (Al-Amin et al., 2012). Various other plants also contain ZER; among them are and Curcuma amada Robx (Srivastava et al., 2001) from India, Alpinia galangal from Sri Lanka (Arambewela et al., 2007), Xylopia aethiopica from Ibadan, Southwest Nigeria (Ogunwande et al., 2005).

3 Anticancer Properties of Zerumbone

Several researchers have reported that ZER has both in vitro (Table 2) and in vivo (Table 3) anticancer properties at different concentrations and doses (Kapoor, 2012). Zerumbone possesses antiproliferative properties towards several cancer cell lines with minimal effect on normal cells (Sadhu et al., 2007; Prasannan et al., 2012; Rahman, H.S. et al., 2013). Among the effects of ZER is induction of high intracellular redox potential that can inhibit proliferation of cancer cells (Hoffman et al., 2002). The cytotoxic effect of ZER on the cancer cells appear to be attributed to the versatile α,β-unsaturated carbonyl group in its structure, which plays an important role in the interaction of the compound with the most biologically active molecules.

Clearly the carbonyl group is important for biological activity because α-humulene, also found in ginger, lacking in this functional group is virtually and consistently pharmacological inactive (Murakami et al., 2002). The α,β-unsaturated carbonyl group in ZER effectively removes the intracellular glutathione (GSH) through the formation of Michael adducts, thus enhancing the potential of intracellular redox (E), resulting in the inhibition of spread of cancerous cells. However, the average intracellular
<table>
<thead>
<tr>
<th>Organ</th>
<th>Cell line</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>Human acute lymphocytic leukemia (CEM-ss) (Abdelwahab et al., 2011)</td>
<td>Induces apoptosis and DNA internucleosomal degradation&lt;br&gt;Activate caspase-3</td>
</tr>
<tr>
<td></td>
<td>Human acute lymphoblastic leukemia (Jurkat) (Rahman, H.S. et al., 2013b)</td>
<td>Induces G2/M cell cycle arrest&lt;br&gt;Induces intrinsic apoptotic pathway via activation of caspase -3 and -9, cytochrome c release from mitochondria, PARP cleavage</td>
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<td></td>
<td>Human chronic myeloid leukemia (KBM-5) (Takada et al., 2005)</td>
<td>Induces cytotoxicity</td>
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<td></td>
<td>Human acute promyelocytic leukemia (HL-60) (Murakami et al., 2002; Xian et al., 2007)</td>
<td>Suppresses TPA-induced superoxide anion generation from NADPH oxidase&lt;br&gt;Induces G2/M cell cycle arrest in time- and concentration-dependent manner&lt;br&gt;Decreases cyclin B1/CDK1 protein level</td>
</tr>
<tr>
<td></td>
<td>Human acute promyelocytic leukemia (NB4) (Xian et al., 2007)</td>
<td>Induces G2/M cell cycle arrest associated with decline of cyclin B1 protein, and phosphorylation of ATM/Chk1, induced apoptosis via expression of Fas (CD95)/Fas ligand (CD95L), with the activation of caspase-8</td>
</tr>
<tr>
<td></td>
<td>Human acute myelocytic leukaemia (U937) (Xian et al., 2007)</td>
<td>Antagonizes action of DDT and TCDD by up-regulating the expressions of COX-2 and VEGF mRNA</td>
</tr>
<tr>
<td></td>
<td>Human acute lymphoblastic leukaemia (MOLT4), human acute lymphocytic leukaemia (OKM-2T), human chronic myelocytic leukaemia (K562 and KT-1) (Xian et al., 2007)</td>
<td>No cytotoxicity at concentration of 10 µM</td>
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<td></td>
<td>Human peripheral blood multiple myeloma (U266) (Sung, B. et al., 2009)</td>
<td>Suppresses CXCR4 expression</td>
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<td></td>
<td>Murine lymphoid neoplastic (P-388D1) (Huang et al., 2005)</td>
<td>Causes DNA fragmentation and growth inhibition</td>
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<td></td>
<td>Murine acute myelocytic leukemia (WEHI-3B) (Rahman et al., 2013a)</td>
<td>Induces G2/M cell cycle arrest and apoptosis</td>
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<tr>
<td>Organ</td>
<td>Cell line</td>
<td>Biological Effect of ZER</td>
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<tr>
<td>Blood</td>
<td>Normal human umbilical vein endothelial cell (HUVEC) (Xian et al., 2007)</td>
<td>Does not inhibit proliferation at concentration of 10 µM.</td>
</tr>
<tr>
<td></td>
<td>Normal human primary mono nuclear cells (PBMCs) (Al-Zubairi et al., 2010a; Rahman et al., 2014)</td>
<td>No cytotoxicity (1-100 µg/mL) Cytotoxic at high doses (40-80 µM)</td>
</tr>
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<td></td>
<td>Mice thymocytes and splenocytes Human PBMC (Keong et al., 2010)</td>
<td>Stimulates time- and dose-dependent proliferation of mice cells and human PBMC Up-regulates human cytokine (Interleukin IL-2 and IL-12) Immunomodulatory effect</td>
</tr>
<tr>
<td></td>
<td>Human peripheral blood lymphocytes (PBL) Al-(Al-Zubairi et al., 2010b)</td>
<td>Cytotoxic but not clastogenic at 40 and 80 µM Does not induce chromosomal aberration and micronuclei formation</td>
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<td></td>
<td>Lymphoblastoid (Raji) cells</td>
<td>Suppresses tumor promoter 12-O-tetradecanoylphorbol 13-acetate (TPA)-induced activation of Epstein–Barr virus</td>
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<td></td>
<td>Human monocyte-like cells (THP-1) (Eguchi et al., 2007)</td>
<td>Suppresses TPA-induced LOX-1 mRNA expression Attenuates expression of SR-A, SR-PSOX and CD-36 and led to block DiI-AcLDL uptake Inhibits AP-1 and NF-kB transcriptional activity</td>
</tr>
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<td></td>
<td>Normal murine macrophages 9RAW264.7) (Sung, B. et al., 2009)</td>
<td>Markedly diminishes inducible nitric oxide synthase (iNOS) and cyclooxygenase (COX)-2 expression. Suppresses free radical generation, and inhibits tumor necrosis factor (TNF)-α release. Induces phase II drug metabolizing enzymes GSTP1 and NQO1 mRNA expressions</td>
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<td></td>
<td>Immortalized mouse embryonic fibroblasts (SV40) (Sehrawat et al., 2012)</td>
<td>Not cytotoxic</td>
</tr>
<tr>
<td></td>
<td>Human whole blood (Jantan et al., 2008)</td>
<td>Inhibits platelet aggregation induced by arachidonic acid (AA), collagen and ADP</td>
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<tr>
<td>Organ</td>
<td>Cell line</td>
<td>Biological Effect of ZER</td>
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<tr>
<td>Skin</td>
<td>Human melanoma (WM1552C) (Ni, 2013)</td>
<td>Induces apoptosis and autophagy</td>
</tr>
<tr>
<td></td>
<td>Murine melanoma (B16-F0) (Ni, 2013)</td>
<td>Induces apoptosis and autophagy</td>
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<td>Normal human dermal fibroblast (2F0-C25) (Ni,</td>
<td>Not cytotoxic at a concentration of 13 µM</td>
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<tr>
<td></td>
<td>2013)</td>
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<td></td>
<td>Murine epidermal cells (JB6 Cl41) (Shin et al.,</td>
<td>Induces heme oxygenase-1 expression through activation of Nrf2</td>
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<td></td>
<td>2011)</td>
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<tr>
<td>Liver</td>
<td>Human liver adenocarcinoma (HepG2) (Sakinah et</td>
<td>Induces apoptosis via up- and down-regulation of Bax/Bcl-2 proteins independent of functional p53 activity</td>
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<td></td>
<td>al., 2007)</td>
<td>Induces DNA fragmentation</td>
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<td></td>
<td>Human hepatoma (HTC) (Matthes et al., 1980; Mat</td>
<td>Cytotoxic</td>
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<td></td>
<td>thes et al., 1982)</td>
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<tr>
<td></td>
<td>Murine hepatoma cells (Hepa1c1c7) (Ohnishi et</td>
<td>Marked up-regulation of multiple HSPs, such as HSP40 and HSP70HSPs</td>
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<td></td>
<td>al., 2013a; Ohnishi et al., 2013b; Ohnishi et</td>
<td>Increases proteasome activity with up-regulation of β5, a major proteasome functional protein</td>
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<td>al., 2013c)</td>
<td>Up-regulates expressions of several pro-autophagic markers, including p62 and microtubule-associated protein 1 light-chain 3 (LC3)-II</td>
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<td>Suppresses cellular protein modifications by 4-hydroxy-2-nonenal (HNE)</td>
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<td></td>
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<td>Confers resistance to toxicity of HNE via p62 induction</td>
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<td></td>
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<td>Induces ubiquitination and aggregation of cellular proteins</td>
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<td></td>
<td></td>
<td>Activates ubiquitin–proteasome system and autophagy</td>
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<td></td>
<td>Normal human liver cells (Chang) (Sakinah et</td>
<td>Inhibits cell growth with an IC₅₀ value of 10.96 ± 0.059 µg/ml</td>
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<td>al., 2007)</td>
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<td></td>
<td>Normal rat liver epithelial cells (RL34) (Nakamura et al.,</td>
<td>Activates phase II drug metabolizing enzymes, such as GST (Glutathione S-transferase), epoxide hydrolase and hemeoxygenase via the transcription factor Nrf2 dependent pathway</td>
</tr>
<tr>
<td></td>
<td>2004)</td>
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<td></td>
<td>Normal human liver cells (WRL-68) (Nozlena et</td>
<td>Not cytotoxic</td>
</tr>
<tr>
<td></td>
<td>al., 2014)</td>
<td></td>
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<tr>
<td>Organ</td>
<td>Cell line</td>
<td>Biological Effect of ZER</td>
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<td>------------------------------------------------------------------------------------------</td>
</tr>
</tbody>
</table>
| Cervical   | Human cervical cancer (HeLa) (Devi Tailan, 2007; Abdelwahab et al., 2010; Abdelwahab et al., 2012) | Causes growth inhibition and induces apoptosis  
Decreased level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest |
|            |                                                                           | Decreased level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest |
| Colon      | Human colonic adenocarcinoma (Caco-2, Colo320DM, and HT-29) (Murakami et al., 2002) | Markedly induces expressions of interleukin (IL)-1α, IL-1β, IL-6, and tumor necrosis factor (TNF)-α |
|            |                                                                           | Inhibits cell proliferation in dose-dependant manner |
|            | Normal human colon fibroblast (CCD-18Co) (Murakami et al., 2002)          | Not cytotoxic at a concentration of 13 µM |
| Colorectal | Human colorectal carcinoma (HCT116) (Yodkeeree et al., 2009; Deorukhkar et al., 2010) | Enhances TRAIL-induced apoptosis  
Causes activations of caspase-8, -9, -3 and PARP in combination with TRAIL  
Induces expression of TRAIL receptors DR4 and DR5  
Down-regulates expression of anti-apoptotic protein c-FLIP  
Causes activation of ERK in time-dependent manner |
|            | Human colon carcinoma (HCT-116) (Sehrawat et al., 2012)                   | Induces apoptosis |
| Bile duct  | Poorly-differentiated adenocarcinoma (KKU-100), squamous cell carcinoma (KKU-M139), moderately-differentiated adenocarcinoma (KKU-M156), adenosquamous carcinoma (KKUM213) and moderately differentiated adenocarcinoma (KKU-M214) (Songsiang et al., 2010) | ZER derivatives (5, 10, 14 and 20) showed antiproliferative activity |
| Breast     | Human breast adenocarcinoma cell lines (MCF-7 and MDA-MB 231) (Sung, B. et al., 2009; Yodkeeree et al., 2009)  
Human breast benign cell line (MCF-10A) (Sehrawat et al., 2012) | G2/M phase cell cycle arrest  
Down-regulates cyclin B1, cyclin-dependent kinase 1, Cdc25C, and Cdc25B and Bax/Bak-mediated apoptosis  
Induces significant expression of DR4  
Activation of Bax and Bak |
<p>|            |                                                                           | Not cytotoxic |</p>
<table>
<thead>
<tr>
<th>Organ</th>
<th>Cell line</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
</table>
| Ovarian   | Human ovarian cancer (Caov-3) (Prasannan et al., 2012) | Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest |
|           | Normal Chinese hamster ovarian cells (AS52) (Murakami et al., 2002) | Suppresses tumor promoter 12-O-tetradecanoylphorbol-13-acetate (TPA)-induced superoxide anion (O₂⁻) generation from xanthine oxidase (XO)  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
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Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
|           | Normal Chinese hamster ovary cells (CHO) (Al-Zubairi, 2012) | High concentrations produce genotoxic and cytotoxic effects (40-80 µM)  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
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Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
| Pancreatic | Human pancreatic carcinoma (Pa-Ca) (Chakraborty et al., 2013) | Novel inhibitor of Jak2/Stat3, which inhibits promigratory gene expression, growth, and migration of pancreatic cancer cells  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
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Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
|           | Human pancreatic cancer (PANC-28, MIA PaCa-2, and AsPC-1) (Sung, B. et al., 2008) | Inhibits CXCL12-induced invasion of pancreatic tumor cells  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
|           | Human pancreatic carcinoma (PANC-1 and SW1990)(Zhang et al., 2012) | Time-dependent inhibition of cell viability  
Induces apoptosis  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
|           | Human pancreatic carcinoma (Pa-Ca)(Shamoto et al., 2014) | Inhibits PaCa-associated angiogenesis through the inhibition of NF-κB and NF-κB-dependent proangiogenic gene products  
Induces apoptosis  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
| Lung      | Human non-small cell lung carcinoma (H1299 cells) (Takada et al., 2005; Yodkeeree et al., 2009) | Enhances TNF-induced cytotoxicity and potentiates apoptosis  
Inhibits TNF-induced IκBα protein degradation and phosphorylation  
Inhibits TNF-induced phosphorylation of p65 protein  
Suppresses TNF-induced invasion activity  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
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Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
|           | Human small cell lung carcinoma (NCI-H187) (Pitchuanchom et al., 2011) | Inhibits monomeric form of the HSP 27 protein  
ZER derivative (parent alcohol 8) induces strong cytotoxicity  
Induces apoptosis  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
| Kidney    | Human embryonic kidney carcinoma cell line (A293 cells) (Takada et al., 2005) | Inhibits cell growth  
Induces apoptosis  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
|           | Bovine normal kidney cell line (MDBK) (Sakinah et al., 2007) | Inhibits cell growth with an IC₅₀ value of 10.02 ± 0.03 µg/ml  
Induces apoptosis  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
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Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  
Causes growth inhibition and induces apoptosis  
Decreases level of IL-6 secretion and membrane bound IL-6 receptor  
Induces G2/M cell cycle arrest  |
<p>| | | |
|           |                                                     |                                                                                                           |</p>
<table>
<thead>
<tr>
<th>Organ</th>
<th>Cell line</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kidney</td>
<td>Human kidney embryonic cells (HEK 293) (Tang et al., 2011)</td>
<td>ZER derivative (parent alcohol 8) could protects irradiation-induces cell apoptosis and DNA damage, at least partly, via activation of Keap1/Nrf2/ARE pathway. Non-significant cytotoxicity with IC50 of 30 µM.</td>
</tr>
<tr>
<td></td>
<td>Normal African green monkey kidney cells (Vero) (Pitchuanchom et al., 2011)</td>
<td></td>
</tr>
<tr>
<td>Brain</td>
<td>Human brain malignant glioma (GBM8401) (Weng et al., 2012)</td>
<td>Induces human glioblastoma multiforme cell apoptosis via inhibition of the IKKα-Akt FOXO1 cascade and activation of caspase-3.</td>
</tr>
<tr>
<td></td>
<td>Human brain malignant glioma (U87MG) (Weng et al., 2012)</td>
<td>Significantly decreases cell viability at the concentration of 30 and 50 µM.</td>
</tr>
<tr>
<td>Prostate</td>
<td>Human adenocarcinoma (DU145) (Yodkeeree et al., 2009)</td>
<td>Induces cytotoxicity and significant PARP cleavage. Effectively blocks Jak2/STAT3-mediated signaling pathways. Induces non significant expression of DR4.</td>
</tr>
<tr>
<td></td>
<td>Human adenocarcinoma (PC3) (Yodkeeree et al., 2009)</td>
<td>Induces non-significant expression of DR4.</td>
</tr>
<tr>
<td>Stomach</td>
<td>Human gastric adenocarcinoma (AGS) (Tsuboi et al., 2014)</td>
<td>Inhibits tumor angiogenesis via reduction of VEGF production and NF-κB activity.</td>
</tr>
<tr>
<td>Oral</td>
<td>Human oral cancer (KB) (Pitchuanchom et al., 2011)</td>
<td>ZER derivative (parent alcohol 8) induces strong cytotoxicity.</td>
</tr>
<tr>
<td>Head &amp; neck</td>
<td>Human squamous cell carcinomas (SCC4) (Sung, B. et al., 2008)</td>
<td>Suppressed CXCR4 expression and cancer invasion and metastasis.</td>
</tr>
<tr>
<td></td>
<td>Human squamous cell carcinoma (FaDu) (Takada et al., 2005)</td>
<td>Inhibits NF-κB and IκBα kinase activation. Suppresses antiapoptotic and metastatic gene expression, Up-regulates apoptosis and down-regulates cancer invasion.</td>
</tr>
</tbody>
</table>
**Table 2:** In Vitro biological effects of zerumbone.

<table>
<thead>
<tr>
<th>Organ</th>
<th>Cell line</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
</table>
| Bone   | Mouse macrophage (RAW 264.7) (Sung, B. et al., 2009) | Inhibits RANKL-induced NF-κB activation through inhibition of activation of IKBA kinase, IKBA phosphorylation, and IKBA degradation  
Suppresses RANKL-induced differentiation of an osteoclast precursor cells to osteoclasts  
Inhibits osteoclastogenesis induced by RANKL and tumor (RAW264.7) cells after incubation in the presence of MDA-MB-231 cells or U266 cells for 24 h, then exposed to ZER for 5 days, and finally stained for TRAP expression  
Potential therapeutic agent for osteoporosis and cancer-associated bone loss |

<table>
<thead>
<tr>
<th>Organ</th>
<th>Animal Model</th>
<th>ZER Route</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
</table>
| Cervix | Female BALB/c mice (Devi Tailan, 2007; Abdelwahab et al., 2010) | Intraperitoneal injection | Suppresses cervical intraepithelial neoplasia in female Balb/c mice prenatally exposed to Diethylstilbestrol (DES)  
Reduces the expression of cell proliferation marker PCNA in dose dependent manner  
Causes over-expression of pro-apoptotic protein Bax  
Suppresses Bcl-2 specific mRNA expression  
Inhibits progression of cervical dysplasia from becoming more severe dysplasia (CIN 3) and suppresses level of serum IL-6. |
<p>| Colon  | Male Sprague-Dawley rats (Kirana et al., 2003) | Oral dose                | Suppresses azoxymethane (AOM)-induced colon cancer using aberrant crypt foci (ACFs) as a preneoplastic marker                                                                                                           |</p>
<table>
<thead>
<tr>
<th>Organ</th>
<th>Animal Model</th>
<th>ZER Route</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
</table>
| Colon | Male ICR mice (Kim, M. et al., 2009) | Oral dose     | Inhibits multiplicity of colonic adenocarcinomas induced by azoxymethane (AOM)  
Suppresses colonic inflammation in dose-dependent manner  
Inhibits cancer proliferation, potentiates apoptosis, suppresses NF-kB and HO-1 expressions                                                                                                      |
|       | Female ICR mice (Murakami et al., 2003) | Oral dose     | Suppresses acute ulcerative colitis (UC) induced by dextran sodium sulphate (DSS)  
Significantly lowers levels of inflammatory biomarkers IL-1β, TNF-α, and PGE2 in colonic mucosa  
Suppresses expression of inflammatory cytokines, TNF and IL-1β in LPS/IFN-γ                                                                                                     |
|       | Male F344 rats (Tanaka et al., 2001)  | Oral dose     | Reduces development AOM-induced colonic aberrant crypt foci  
Reduces expression of COX-2 and prostaglandins in colonic mucosa  
Reduces number of AgNORs in colonic crypt cell nuclei                                                                                                                                  |
| Liver | Male Sprague Dawley rats (Taha et al., 2010) | Intraperitoneal injection | Protects rat liver from carcinogenic effects of DEN and AAF  
Lowers serum ALT, AST, AP and AFP concentrations  
Lowers concentration of GSH in hepatic tissue  
Lowers expression of PCNA in the rat liver  
Increases Bax and decreases Bcl-2 protein expression in the liver                                                                                                                     |
|       | Male Sprague Dawley rats (Fakurazi et al., 2008; Fakurazi et al., 2009) | Oral dose     | Suppresses fatty liver formation induced by overdosage of ethanol  
Prevents necrosis of liver tissues after administration of overdosage of paracetamol  
Reduces levels of liver ALT, AST and ALP at 24 h after administration of overdosage of paracetamol                                                                                   |
<table>
<thead>
<tr>
<th>Organ</th>
<th>Animal Model</th>
<th>ZER Route</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
</table>
| Liver      | Male Golden Syrian hamsters (Tzeng et al., 2013b)                            | Oral dose     | Attenuates nonalcoholic fatty liver disease  
Implements insulin sensitivity, decreases lipogenesis, and increases lipid oxidation                                                                                                                                                                                                                     |
|            | Male Sprague Dawley rats (Ohnishi et al., 2013a)                             | Oral dose     | Up-regulates heat shock protein expressions in the liver  
Confers thermoresistant phenotype                                                                                                                                                                                                                                                                               |
| Lung       | Female A/J mice (Kim, M. et al., 2009)                                       | Oral dose     | Significantly inhibits multiplicity of lung adenomas induced by 4-(Nmethyl-N-nitrosamino)-1-(3-pyridyl)-1-butanone (NNK)  
Inhibits cancer proliferation, potentiates apoptosis, suppresses NF-kB and HO-1 expressions                                                                                                                                                           |
| Breast     | Female Sprague Dawley rats (Safa, 2013)                                      | Intraperitoneal injection | Inhibits tumor growth via Wint pathway in LA-7 bearing rats                                                                                                                                                                                                                                                                                                                     |
|            | Female severe combined immune deficient (SCID) mice (Sahrawat et al., 2012)  | Intraperitoneal injection | Retards growth of orthotopic MDA-MB-231 xenografts in association with apoptosis induction and suppression of cell proliferation (Ki-67 expression)                                                                                                                                                                                                                     |
|            | Female BALB/c nu/nu mice (Sung, B. et al., 2009)                             | Intraperitoneal injection | Decreases osteolytic bone metastasis in MDA-MB-231 bearing athymic nude mice dose-dependently                                                                                                                                                                                                                   |
| Blood      | WEHI-3B bearing male BALB/c mice (Rahman, H.S. et al., 2013a)                | Oral dose     | Induces apoptosis via the mitochondrial intrinsic pathway  
Increases expression of Bax, Cyt-c, and PARP and decreases the expression of Bcl-2                                                                                                                                                                                                                              |
<p>|            | CDF mice (Huang et al., 2005)                                                | Intraperitoneal injection | Significantly prolongs life of P-388D1-bearing CDF mice                                                                                                                                                                                                                                                                                                                      |</p>
<table>
<thead>
<tr>
<th>Organ</th>
<th>Animal Model</th>
<th>ZER Route</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
<tbody>
<tr>
<td>Skin</td>
<td>C57 BL/6 male mice (Ni, 2013)</td>
<td>Intraperitoneal injection</td>
<td>Significantly reduces tumor mass and lung metastasis in B16-F0 bearing mice through the activation of Akt and MAPK and inhibition of NF-κB activity</td>
</tr>
<tr>
<td></td>
<td>ICR mice (Murakami et al., 2004a)</td>
<td>Topical application</td>
<td>Suppresses 7,12-dimethylbenz[Allensworth et al.]anthracene (DMBA) and TPA-induced initiation and promotion of skin tumor formation. Enhances expression of anti-oxidative and phase II xenobiotics metabolizing enzymes manganese superoxide dismutase (MnSOD), glutathione peroxidise-1 (GPx-1), glutathione S-transferase-P1 (GST-P1) and NAD (P) H quinine oxido-reductase (NQO1) mRNA in the epidermis. Suppresses TPA-induced COX-2 expression and phosphorylation of ERK1/2. Suppresses TPA-induced leukocyte maturation and dermal infiltration as well as activation stages of skin tumors</td>
</tr>
<tr>
<td></td>
<td>Female HR-1 hairless mice (Shin et al., 2011)</td>
<td>Topical application</td>
<td>Induces HO-1 expression through activation of Nrf2</td>
</tr>
<tr>
<td>Paw</td>
<td>Mice (Sulaiman et al., 2010)</td>
<td>Intraperitoneal injection</td>
<td>Inhibits carrageenan-induced paw edema dose-dependently. Suppresses granulomatous tissue formation in cotton pellet-induced granuloma test</td>
</tr>
<tr>
<td>Organ</td>
<td>Animal Model</td>
<td>ZER Route</td>
<td>Biological Effect of ZER</td>
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</tbody>
</table>
| Eye   | ICR mice (Chen, B et al., 2011; Chen, BY et al., 2011) | Oral dose     | Protects mouse cornea from ultraviolet B (UVB)-induced inflammatory photokeratitis  
Inhibits NF-kB, iNOS and TNF-α expressions  
Abrogates nuclear translocation of NF-kB  
Reduces malonyldialdehyde (MDA) accumulation and increases GSH and glutathione reductase levels  
Protects mice cornea from UVB-induced cataractogenesis |
| Pancreas | Male Wistar rats (Szabolcs et al., 2007) | Oral dose     | Suppresses cholecystokinin octapeptide (CCK-8)-induced acute pancreatitis  
Significantly reduces serum amylase and lipase activities  
Reduces cytosolic IL-6 and TNF-α and increases cytosolic IκBα concentration  
Reduces iNOS and Mn- and Cu/Zn-superoxide dismutase activities  
Significantly reduces pancreatic weight to body weight ratio |
|        | Male SPF Wistar rats (Wenhong et al., 2012) | Intravenous injection | Attenuates severity of acute necrotizing pancreatitis induced by sodium taurocholate and pancreatitis-induced hepatic injury, via inhibition of NF-κB activity and down-regulation of ICAM-1 and IL-1β expressions |
| Bone  | Male Sprague Dawley rats (Ganabadi et al., 2009) | Oral dose     | Reduces inflammatory process in collagen-induced osteoarthritis (OA)  
Significantly reduces number of major histocompatibility complex type II cells (MHC) expression in the affected synovial membrane  
Reduces the number of antigen presenting type A cells presented during arthritis |
<table>
<thead>
<tr>
<th>Organ</th>
<th>Animal Model</th>
<th>ZER Route</th>
<th>Biological Effect of ZER</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bone</td>
<td>Male Sprague Dawley rats (Al-Saffar et al., 2010; Al-Saffar et al., 2011)</td>
<td>Oral dose</td>
<td>Produces chondroprotective effects in MIA-induced knee osteoarthritis</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Improved immunoreactivity of neuropeptides</td>
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<td></td>
<td>Improves density of protein gene products (PGP); calcitonin gene-related peptide (CGRP) and neuropeptides-Y (NPY) immunoreactive nerve fibers</td>
</tr>
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<td></td>
<td>Reduces the level of PGE₂</td>
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<td></td>
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<td></td>
<td>Produces induction of cytochrome P450 and cytosolic GST</td>
</tr>
<tr>
<td>Miscellaneous</td>
<td>Male ICR mice (Perimal et al., 2011)</td>
<td>Intraperitoneal</td>
<td>Produces pronounced antinociception against chemical models of nociception through L-arginine-nitric oxide-cGMP-PKC-K+ ATP channel pathways, the TRPV1 and kinin B2 receptors.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>injection</td>
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</tr>
<tr>
<td></td>
<td>Male BALB/c mice (Sulaiman et al., 2009b)</td>
<td>Intraperitoneal</td>
<td>Produces significant peripheral and central antinociceptive effects when assessed in acetic acid-induced abdominal writhing and hot-plate test models</td>
</tr>
<tr>
<td></td>
<td></td>
<td>injection</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Female and male BALB/c mice (Rahman, H.S. et al., 2014)</td>
<td>Oral dose</td>
<td>Not toxic effects to liver and renal tissues</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Does not cause significant change in haematological and serum biochemical parameters</td>
</tr>
<tr>
<td></td>
<td>Female and male ICR mice (Jin et al., 2013)</td>
<td>Intraperitoneal</td>
<td>Does not cause mortality or change in the general condition, growth, organ weights, hematology, serum biochemistry, or histopathological after a single dosage of 500 mg/kg or multiple dosage of 5, 25 and 50 mg/kg for a period of 28 days</td>
</tr>
<tr>
<td></td>
<td></td>
<td>injection</td>
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<tr>
<td><strong>Organ</strong></td>
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<td><strong>ZER Route</strong></td>
<td><strong>Biological Effect of ZER</strong></td>
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</tr>
<tr>
<td>Miscellaneous</td>
<td>Female Sprague Dawley rats (Ibrahim et al., 2010)</td>
<td>Single intraperitoneal injection</td>
<td>Not toxic to liver and renal tissues at dose of 100–200 mg/kg</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Produces severe renal and hepatic damage at a dose of 500 mg/kg with</td>
</tr>
<tr>
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<td></td>
<td></td>
<td>Increases serum creatinine, BUN, liver enzymes (ALT, ALP, and GGT) and MDA concentrations</td>
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<tr>
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<td></td>
<td></td>
<td>Does not cause mortality at 100, 200, 500 and 1000 mg/kg</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>Causes 20 and 40% death for animals receiving 1500 and 2000 mg/kg, respectively</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Causes 100% death in animals receiving 2500 and 3000 mg/kg</td>
</tr>
<tr>
<td></td>
<td>Male Sprague Dawley rats (Al-Zubairi et al., 2010a; Al-Zubairi et al., 2010b)</td>
<td>Intraperitoneal injection</td>
<td>Induces significant increase in the frequency of micronuclei in polychromatic erythrocytes (PCEs) at dose 1000 mg/kg after 24 h injection</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Inhibits cell proliferation and causes cytotoxicity in the rat bone marrow</td>
</tr>
<tr>
<td></td>
<td>Female Sprague Dawley rats (Ibrahim et al., 2012)</td>
<td>Intraperitoneal injection</td>
<td>Beneficial in cisplatin-induced renal dysfunction, toxicity and organ damage in via preservation of antioxidant glutathione and prevention of lipid peroxidation</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Attenuates cisplatin, decreases renal GSH and increased MDA levels</td>
</tr>
<tr>
<td></td>
<td>Male New Zealand white rabbits (Hemn, H.O. et al., 2012; Hemn, H.O. et al., 2012)</td>
<td>Oral dose</td>
<td>Significantly averts and decreases early atheroma plague formation and development via reduction in monocytes and/or macrophages migration, aggregation and smooth muscle cells proliferation in rabbits fed on cholesterol-rich diet</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Repairs endothelial dysfunction resulting from hyperlipidemia in rabbit atherosclerosis model</td>
</tr>
</tbody>
</table>
redox potential of normal cells differ from that of cancerous cells; this difference could be the reason for ZER not inducing proliferation of normal cells (Hoffman et al., 2002; Murakami et al., 2002). Because there is a close link between tumor promotion, inflammation and oxidative stress, the anti-inflammatory and/or antioxidant compounds could also act as an anti-carcinogenic agent (Cerutti and Trump, 1991). Although the stimulation of neoplastic cell death by ZER was reported to be through the mitochondrial pathway of apoptosis (Giang et al., 2009), it also exhibits antiproliferative and anti-inflammatory activities through the modulation of NF-κB activity. Zerumbone inhibits NF-κB in association with the sequential suppressions of IκBα kinase activity, phosphorylation and degradation. This compound also inhibits NF-κB-dependent reporter gene expression activated by TNF, TNFRI, TRADD, TRAF2, NIK, and IKK but not by the p65 subunit of NF-κB. Zerumbone also down-regulate NF-κB-regulated gene products, including cyclin D1, COX-2, MMP-9, ICAM-1, c-myc, survivin, IAP1, IAP2, XIAP, Bcl-2, Bcl-xL, Bfl-1/A1, TRAF1 and FLIP. These effects lead to the potentiation of apoptosis induced by cytokines and chemotherapeutic agents. The inhibition of these NF-κB-regulated genes expression is in association with the suppression of TNF-induced cancer invasiveness. Thus, it is hypothesized that inhibition of NF-κB and NF-κB-regulated gene expression induced by carcinogens may also represent the molecular basis for cancer prevention and treatment by ZER (Takada et al., 2005). Furthermore, it was shown that ZER is a novel inhibitor of CXC chemokine receptor-4 (CXCR4) expression, which mediates homing of tumor cells to specific organs during metastasis, suggesting the potential of the compound in the suppression of metastasis (Sung, B. et al., 2008). This re-

<table>
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</tr>
</thead>
<tbody>
<tr>
<td>Miscellaneous</td>
<td>Male Golden Syrian hamsters (Tzeng et al., 2014)</td>
<td>Oral dose</td>
<td>Improves dyslipidemia by modulating the genes expression involving in the lipolytic and lipogenic pathways of lipids metabolism. Decreases hepatic mRNA levels of fatty acid synthase, malic enzyme, sterol-regulatory element binding protein and 3-hydroxy-3-methyl-glutaryl-CoA reductase.</td>
</tr>
<tr>
<td></td>
<td>Male Wistar rats (Tzeng et al., 2013a)</td>
<td>Oral dose</td>
<td>Ameliorates streptozotocin-induced diabetic nephropathy (DN) by reducing the hyperglycemia-induced inflammatory response. Decreases infiltration of macrophages, IL-1, IL-6 and TNF-α produced by p38 mitogen-activated protein kinase activation.</td>
</tr>
</tbody>
</table>

Table 3: In Vivo biological effects of zerumbone.
ceptor has been identified in various tumors including those in the breast, ovary, prostate, gastrointestinal tract, head, neck, bladder, brain, and skin.

3.1 Blood Cancer (Leukemia)

It has been shown that ZER effectively suppresses the tumor promoter 12-O-tetradecanoylphorbol-13-acetate (TPA)-induced superoxide anion (O$_2^-$) generation from NADP oxidase in dimethyl sulphoxide (DMSO)-differentiated human acute promyelocytic leukemia (HL-60) cells (Hoffman et al., 2002). One study determined the effect of diethyl ether extract of *Zingiber zerumbet* fresh rhizome on cultured P-388D1 cells and in P-388D-bearing CDF mice. This study showed that the extract could induce DNA fragmentation in P-388D1 cells in vitro, and significantly prolonged the life of P-388D1-bearing CDF mice. The same result was obtained when the activity of ZER isolated from the same extract was examined in vitro and in vivo (Huang et al., 2005). The study further found that ZER inhibited the growth of HL-60 cells, in time- and concentration-dependent manner. HL-60 cell cycle analysis after treatment with ZER showed induction of G2/M arrest and decreased cyclin B1/CDK1 protein level. Using CEM-ss cells as targets, it was shown that ZER increased the number of TUNEL-positive cells and cellular caspase-3 level; the hallmarks of apoptosis (Abdelwahab et al., 2011). The anticancer effects of ZER seems boundless when it was shown that it inhibits the proliferation of NB4 cell line, derived from acute promyelocytic leukemia cells, through the induction of G2/M phase cell cycle arrest associated with a decline of cyclinB1 protein, and phosphorylation of ATM/Chk1. The study indicated that ZER induction of NB4 cell apoptosis was initiated by the expression of Fas (CD95)/Fas ligand (CD95L), concomitant with the activation of caspase-8. At the same time, they found that ZER induced cleavage of Bid, Bax and Mcl-1 proteins, phosphorylation of Cdc25C and Cdc2 at the Thr48 and Thr14/Tyr15 residues respectively, degradation of the proteolytic poly-(ADP-ribose) polymerase (PARP), and triggering of cytochrome c release into the cytoplasm (Huang et al., 2005). On leukemic cells ZER is cytotoxic to human myeloid (KBM-5) (Xian et al., 2007), mouse myelomonocytic (WEHI-3B) (Alitheen et al., 2012) and human acute lymphoblastic leukemic (Jurkat) cell lines (Rahman, H.S. et al., 2013b). Zerumbone also regulate expression of apoptotic biomarkers in BALB/c mice model of acute myelocytic leukemia via the mitochondrial intrinsic pathway (Rahman, H.S. et al., 2013a).

3.2 Skin Cancer

Zerumbone suppressed 7,12-dimethylbenz (Allensworth et al.) anthracene (DMBA)- and TPA-induced initiation and promotion of skin tumors in female ICR mice. Using RT-PCR, it was shown that ZER enhance expression of manganese superoxide dismutase (MnSOD), glutathione peroxidase-1 (GPx-1), glutathione S-transferase-P1 and NAD (P) H quinine oxido-reductase (NQO1) mRNA in the epidermis while diminishing TPA-induced COX-2 protein expression and phosphorylation of extracellular signal-regulated kinase 1 and 2 (ERK1/2) (Murakami et al., 2004b). The phorbol ester-induced papilloma formation in mouse skin can also be inhibited by ZER (Murakami et al.,
2004a). Recently, it was found that ZER induces heme oxygenase-1 expression in female HR-1 hairless mouse skin and cultured murine epidermal (JB6 Cl4) cells, through the activation of Nrf2 (Shin et al., 2011). More recently, ZER was found to induce apoptosis and autophagy in human (WM1552C) and murine (B16-F0) melanoma cell lines (Liu, 2011). Zerumbone also significantly reduced tumor mass and lung metastasis in B16-F0 bearing C57 BL/6 male mice through the activation of Akt and MAPK and suppression of NF-κB activation (Ni, 2013).

3.3 Liver Cancer

Zerumbone was also found to inhibit the proliferation of non-malignant Chang liver cell line (Alwi et al., 2007), while being innocuous to the normal human liver (WRL-68) cells (Nozlena et al., 2014). DNA fragmentation and apoptosis induced by ZER is by way of up- and down-regulation of Bax/Bcl-2 proteins independent of functional p53 activity in the liver adenocarcinoma (HepG2) cell lines. In vivo, ZER inhibits diethyl nitrosamine (DEN) and dietary 2-acetylaminofluorene (AAF)-induced Sprague Dawley rat hepatocarcinogenesis. This effect was suggested to be through the reduction of oxidative stress, inhibition of cancer cell proliferation, and induction of mitochondria-regulated apoptosis of liver cancers (Taha et al., 2010).

3.4 Cervical Cancer

Zerumbone is known to exhibit an antiproliferative effect on human cervical cancer (HeLa) cell line (Abdelwahab et al., 2012). In diethylstilboestrol (DES)-induced mice cervical inter-epithelial neoplasia (CIN), ZER caused over-expression of pro-apoptotic protein, Bax (Al-Zubairi and Syam, 2008; Abdelwahab et al., 2010).

When ZER and cisplatin was used in combination, the cervical cancer in BALB/c mice was suppressed through the modulation of serum interleukin-6 (Abdul et al., 2009). One experiment was conducted on pregnant BALB/c rats treated with DES to develop cervical intra-epithelial neoplasia. When the progenies were treated with different doses of ZER, histological examination revealed that ZER had inhibited the cervical dysplasia from developing into more severe dysplasia (Devi Tailan, 2007).

3.5 Colon Cancer

Zerumbone was shown to inhibit the proliferation of human colonic adenocarcinoma (LS174T, LS180, COLO205, and COLO320DM) cell lines in a dose-dependent manner, while the growth of normal human colon (CCD-18Co) fibroblasts and normal human dermal (2F0-C25) cells was less affected (Murakami et al., 2004a; Yodkeeree et al., 2009).

The effect of ZER on human colorectal cancer (HCT116) cells, was via potentiation of TRAIL–induced apoptosis (Yodkeeree et al., 2009; Deorukhkar et al., 2010) as indicated by the expression of TRAIL death receptor (DR) 4 and 5. The subsequent effects were activations of caspase-8, -9 and -3 and PARP and down-regulation anti-apoptotic protein c-FLIP expression and activation of ERK in a time-dependent manner. The RT-PCR
assay showed that ZER markedly induced the expressions of IL-1α, IL-1β, IL-6, and TNF-α in human colon adenocarcinoma (Caco-2, Colo320DM, and HT-29) cell lines, in concentration- and time-dependent manners (Murakami et al., 2004a). Developing azoxymethane (AOM)-induced rat colonic aberrant crypt foci (ACF) in male F344 rat can be significantly inhibited by ZER treatment through suppression of COX-2 expression, cell spreading activity of colonic mucosa, and induction of phase II detoxification enzymes (Tanaka et al., 2001). Similarly, using ACF as a preneoplastic marker, ZER was shown to suppress AOM-induced colon cancer in male Sprague-Dawley rats (Kirana et al., 2003). Zerumbone inhibited the multiplicity of colonic adenocarcinoma induced by AOM, potentiated apoptosis, and suppressed NF-κB and HO-1 expressions in male ICR mice (Kim, M. et al., 2009).

3.6 Bile Duct Cancer

Amine 5 derived from ZER showed potent antiproliferative activity against cholangiocarcinoma (CCA) cell line and poorly-differentiated adenocarcinoma (KKU-100). However, amine 5 and other ZER derivatives (10, 14 and 20) (Figure 4) showed lesser cytotoxicity toward other CCA cell lines including squamous (KKU-M139) cell carcinoma, moderately-differentiated adenocarcinoma (KKU-M156), adenosquamous carcinoma (KKUM213) and moderately differentiated adenocarcinoma (KKU-M214) (Songsiang et al., 2010).

3.7 Breast Cancer

In breast cancers, ZER caused G2/M phase cell cycle arrest associated with down-regulation of cyclin B1, Ddk1, Cdc25C, and Cdc25B and Bax/Bak-mediated apoptosis in human breast cancer (MDA-MB-231 and MCF-7) cells and retarded growth of MDA-MB-231 xenografts in vivo (Sehrawat et al., 2012). In addition, its derivative, parent alcohol 8 (2E,6Z,10E)-13-Hydroxy-2,9,9-trimethylcycloundeca-2,6,10-trienone (Figure 5A) significantly displayed antiproliferative effect towards human breast cancer (MCF-7) cell line (Pitchuanchom et al., 2011). The inhibition of mammary tumor growth in LA7-bearing Sprague Dawley rats was via Wnt/β-catenin signaling pathway (Safa, 2013).

3.8 Ovarian Cancer

The anti-proliferative effect of ZER towards human ovarian cancer (Caov-3) cell line is dose-and time-dependent. Zerumbone also effectively suppressed tumor promoter TPA-induced superoxide anion (O₂⁻) generation from xanthine oxidase (XO) in Chinese hamster ovary (AS52) cells (CHO) (Murakamii et al., 1999), while even at high concentrations it does not adversely affect normal cultured CHO (Al-Zubairi, 2012).

3.9 Pancreatic Cancer

Zerumbone is a novel inhibitor of Jak2/Stat3, which inhibits promigratory gene expres
Figure 4: Zerumbone derivatives.

sion, growth, and migration of human pancreatic carcinoma (PaCa) (Chakraborty et al., 2013). It also inhibit CXCL12-induced spread of pancreatic (PANC-28, MIA PaCa-2, and AsPC-1) tumors (Sung, B. et al., 2008). The antipancreatic cancer effect of ZER is facilitated by the inhibition of cancer angiogenesis through the inhibition of NF-κB and NF-κB-dependent proangiogenic gene products (Shamoto et al., 2014). The inhibition and apoptosis of human pancreatic carcinoma cell lines (PANC-1 and SW1990) were via p53 signaling pathway (Zhang et al., 2012).

3.10 Lung Cancer

The non-small lung adenocarcinoma (H1299) cell can be suppressed by ZER, while its derivative, the parent alcohol 8 (2E,6Z,10E)-13-Hydroxy-2,9,9-trimethylcloundeca-2,6,10-trienone, is one of the most potent cytotoxic compound against human small cell lung carcinoma (NCI-H187) (Pitchuanchom et al., 2011). Zerumbone also effectively inhibited proliferation, multiplicity of lung adenomas induced by NNK, potentiated
apoptosis, and suppressed NF-κB and HO-1 expressions in female A/J mice (Kim, Mihye et al., 2009).

3.11 Renal Cancer

Human embryonic kidney carcinoma (A293) cell (Sung, B. et al., 2008) and kidney epithelial (MDBK) cell line (Alwi et al., 2007) proliferation was found to be inhibited by ZER treatment. Zerumbone could also protect irradiation-induced cell apoptosis and DNA damage, partly through the activation of the Keap1/Nrf2/ARE pathway in human kidney embryonic (HEK 293) cells (Tang et al., 2011). The ZER derivative, parent alcohol 8 (2E,6Z,10E)-13-Hydroxy-2,9,9-trimethylcycloundeca-2,6,10-trienone, showed non-significant cytotoxicity toward normal monkey kidney (Vero) cell line (Pitchuanchom et al., 2011).

3.12 Brain Cancer

Zerumbone can induce human glioblastoma multiforme (GBM8401) cell apoptosis via inhibition of the IKKα-Akt FOXO1 cascade (Weng et al., 2012).
3.13 Prostate Cancer

Zerumbone induced cytotoxicity and significant PARP cleavage in human prostate cancer (DU145) cell line through the inhibition of Jak2/STAT3-mediated signaling pathways (Chakraborty and Jorvig, 2011).

3.14 Gastric Cancer

Zerumbone inhibit tumor angiogenesis in human gastric adenocarcinoma (AGS) cells of via reduction of VEGF production and NF-κB activity (Tsuboi et al., 2012).

3.15 Oral Cancer

Parent alcohol 8 (2E,6Z,10E)-13-Hydroxy-2,9,9-trimethylcycloundeca-2,6,10-trienone is one of the most powerful compound inducing cytotoxicity of human oral cancer (KB) cells (Pitchuanchom et al., 2011).

3.16 Head and Neck Cancer

Expression of CXCR4, invasion and metastasis of human tongue squamous (SCC4) cell carcinoma can occur with ZER treatment (Sung, B. et al., 2008). Similarly, ZER inhibited the NF-κB- and NF-κB-regulated gene expression induced by various carcinogens and inflammatory stimuli, such as TNF, okadaic acid, cigarette smoke condensate, phorbol myristate acetate, and H₂O₂. It also suppressed IκBα kinase activity, phosphorylation, and degradation and p65 phosphorylation, nuclear translocation, and acylation in human squamous (LICR-LONHN5) cell carcinoma line (Takada et al., 2005).

3.17 Pharyngeal Cancer

Zerumbone inhibited NF-κB and IκBα kinase, suppressed antiapoptotic and metastatic gene expression, up-regulated apoptosis, and inhibit proliferation of human hypopharyngeal carcinoma (FaDu) cells (Takada et al., 2005).

4 Anti-Inflammatory Activity

Zerumbone been shown to possess anti-inflammatory properties (Zakaria et al., 2010; Somchit et al., 2012). Oral ZER treatment suppressed dextran sodium sulphate (DSS)-induced acute ulcerative colitis (AUC) in female ICR mice. The anti-inflammatory effect of ZER was reflected by the significant lowering of the inflammatory biomarkers, IL-1β, TNF-α, and PGE2 (Murakami et al., 2003). In a female ICR mouse ultraviolet B (UVB) photokeratitis and cataractogenesis model, dietary ZER prevented corneal damage by inhibiting NF-κB, iNOS and TNF-α expression with concomitant reduction of malondialdehyde (MDA) and increase of glutathione (GSH) and GSH reductase (GR) levels (Chen, B et al., 2011; Chen, BY et al., 2011). Moreover, ZER inhibited iNOS and
COX-2 expression and release of TNF-α in a mouse macrophage (RAW264.7) cell line treated with lipopolysaccharide (LPS) and IFN-γ. Zerumbone also inhibited the NO/O₂⁻ generation in inflammatory leukocytes (Murakami et al., 2002; Murakami et al., 2003). Oral feeding of ZER compound reduced the inflammatory process in collagen-induced osteoarthritis (OA) in Sprague Dawley rats. The treatment caused a significant reduction in the number of major histocompatibility complex (MHC) type II cells expression in the affected synovial membrane and thus reducing accumulation of antigen presenting type A cells in arthritis (Ganabadi et al., 2009). In a rat knee osteoarthritis model, induced with monosodium iodoacetate (MIA), oral administration of ZER improved the densities of protein gene products (PGP), calcitonin gene-related peptide (CGRP) and neuropeptides-Y (NPY) immunoreactive nerve (Al-Saffar et al., 2010; Al-Saffar et al., 2011).

In male Wistar rats, ZER suppressed cholecystokinin octapeptide (CCK-8)-induced acute pancreatitis with significant reduction in serum amylase and lipase, cytosolic IL-6, iNOS, Mn- and Cu/Zn-SOD activities and TNF-α concentration (Szabolcs et al., 2007). In these rats ZER treatment attenuates the severity of acute necrotizing pancreatitis and pancreatitis-induced hepatic injury via the inhibition of NF-κB and down-regulation of ICAM-1 and IL-1β expressions (Wenhong et al., 2012).

5 Antioxidant Activity

The antioxidant activity of ZER has been reported to occur through the attenuation of reactive oxygen (RO) and generation of nitrogen species (Murakami and Ohigashi, 2006). Thus, it is plausible that the potential of ZER as an agent against cancer-related inflammation may be mediated through its antioxidant activity. The ability of ZER to stimulate phase II detoxification enzymes was determined in the RL34 cells, a normal rat liver epithelial cell line. Induction of phase II enzymes is known to protect cells and tissues against toxicity and chemical carcinogenesis, particularly in the early phase. The effect of ZER on the stimulation of glutathione S-transferase is dose- and time-dependent and causes considerable increase in the level of the GSTP1-1 protein. Zerumbone also elicited significant induction in the nuclear localization of Nrf2, a transcription factor that binds to the antioxidant response element (ARE) of phase II enzyme genes, activating expression of phase II enzyme genes. Among the phase II enzyme involved in the activation are γ-glutamylcysteine synthetase (GCS), glutathione peroxidase (GPx) and HO-1. These enzyme systems, through their conjugation reactions, play important roles in the metabolic inactivation of pharmacologically active substances, thus minimizing cell damage (Nakamura et al., 2004).

6 Immunomodulatory Activity

Zerumbone has effect on the proliferation, cell cycle progression, and induction of cytokine (IL-2 and IL-12) of immune cells in vitro. This was shown by the proliferation of
ICF mice thymocytes and splenocytes and human peripheral blood mononuclear cells (PBMC). Using flow cytometry, ZER treatment was shown to cause the highest population of PBMC to enter G2/M phase (Keong et al., 2010). This study showed prominent up-regulation of IL-2 and IL-12 in activated lymphocytes after ZER treatment.

7 Other Biomedical Properties of Zerumbone

7.1 Hepatoprotective Activity

Zerumbone was shown to have hepatoprotective properties in ethanol-induced liver injury in male Sprague Dawley rats, while ZER pretreatment extensively reduced fatty liver development in these rats (Fakurazi et al., 2009). Similar ZER has healing effects in paracetamol-induced hepatotoxicity in male Sprague Dawley rats as indicated by the corresponding reductions of alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) blood concentrations in the treated rats (Abdelwahab et al., 2012).

7.2 Anti-atherosclerotic Activity

Zerumbone is a phytochemical with potential for the regulation of atherosclerosis because it suppresses TPA-induced oxidized low density lipoprotein (LDL) receptor-1 (LOX-1) mRNA expression in THP-1 human monocyte-like cells and in differentiated colonic adenocarcinoma (Caco-2) cells. A key event in the development of atherosclerosis is the unregulated uptake of oxidized LDL via scavenger receptors (SR), which are integral membrane proteins. Zerumbone reduces the expression of several subclasses of the macrophage SR such as SR-A, SR-PSOX and CD36, leading to the inhibition of uptake of DiI-acLDL, a modified LDL. Down-regulation in the expression of SR by ZER was postulated to be partly attributed to the inhibition of transcriptional activities of activator protein-1 and NF-κB (Eguchi et al., 2007). In rabbits fed cholesterol-rich diet, oral ZER treatment significantly decreased or averted early atheroma plaque formation and development via reduction in monocytes and/or macrophages migration, aggregation and smooth muscle cells proliferation. In a rabbit atherosclerosis model, ZER was also shown to repair endothelial dysfunction (Hemn, H.O. et al., 2012; Hassan et al., 2015).

7.3 Antinociceptive Activity

Significant antinociceptive effects of intraperitoneal ZER was observed in adult male BALB/c mice. The results of this study indicated that ZER possesses considerable marginal and central antinociceptive effects at various dosages (Sulaiman et al., 2009a). The production of antinociception in the mice model suggest significant involvement of L-arginine-nitric oxide-cGMP-PKC-K+ ATP channel pathways, the TRPV1 and kinin B2 receptors (Perimal et al., 2011).
### 7.4 Antimicrobial Activity

Zerumbone and its derivatives such as 4 (10E/10Z = 3/2) and NH0891 (Figure 5B and 5C) were found to be selective inhibitors of Gram-positive bacteria, *Bacillus subtilis* 168 growth. It was suggested that the new halo-olefinic acids synthesized by the cleavage of the C1-C2 bone of ZER inhibits growth of Gram-positive bacteria by inhibiting YycG histidine kinase (Yamamoto et al., 2001; Kitayama et al., 2004). Zerumbone also inhibits *Salmonella cholerasuis*, a Gram-positive bacteria while not affecting the viability of *Escherichia coli* (Abdul et al., 2008). Similarly, ZER and its synthetic analogues (azazerumbone 1 and azazerumbone 2) (Figure 6) exhibited strong protection against sodium azide-induced mutagenicity of *Salmonella typhimurium* (TA 98 and TA 1531) strains. Among the bacteria tested, *Bacillus cereus* was most sensitive to these analogues (Santosh Kumar et al., 2013).

![Figure 6: Zerumbone derivatives.](image)

Other antipathogen effect of ZER include inhibition of human immunodeficiency virus (HIV) activity (Sharma et al., 1996) and antifungal activity towards *Rhizoctonia solani*, the damping-off pathogen (Kishore and Dwivedi, 1992).

Zerumbone was reported to have antimalarial activities by inhibiting propogation of *Plasmodium falciparum* (Sriphanaa et al., 2013). Exposure of the nematode *Caenorhabditis elegans* to ZER increased expression of HSP16.41 mRNA, suggesting that ZER can increase the survival of nematodes after heat-shock treatment.

In lipid metabolism, ZER improved dyslipidemia by modulating expression of genes involved in the lipolytic and lipogenic pathways of a diet-induced hyperlipidemic animal model (Tzeng et al., 2014). This study suggests that ZER is beneficial to patients with hypercholesterolemia and hypertriglyceridemia. Another study showed that ZER attenuated nonalcoholic fatty liver disease, improved insulin sensitivity, decreased lipogenesis, and increased lipid oxidation in male golden Syrian hamster (Tzeng et al., 2013b). Zerumbone also seems to be beneficial in alleviating symptoms of renal dysfunction. Treatment of Female Sprague Dawley rats with cisplatin-induced renal disease with ZER had reduced toxicity and organ damage via the preservation of antioxidant glutathione and prevention of lipid peroxidation (Ibrahim et al., 2012).

Zerumbone induces genotoxic and cytotoxic effects on cultured human peripher-
al blood lymphocytes (Al-Zubairi et al., 2010a), CHO cells and rat bone marrow polychromatic erythrocytes (PCEs) (Al-Zubairi et al., 2010b; Chang et al., 2012). In fact highly concentrated ZER could cause substantial increase in the frequency of micronuclei in these cells. This study suggests that there are safety issues in the development of ZER as a potential therapeutic compound, because very high doses of ZER may produce adverse effects.

Finally, there is evidence that ZER may be useful in the treatment of Alzheimer’s disease. This was suggest by a recent study that showed ZER inhibits acetylcholinesterase (Bustamam et al., 2008). The enzymolytic effect of ZER towards AChE (acetylcholinesterase) could be the basis for the development of ZER in the treatment of Alzheimer’s disease.

8 Discussion

Many natural compounds possess various and significant biological activities. Thus traditionally these compounds are included in the diet of many Asian societies because they are not only non-toxic but also beneficial to health (Jagtap et al., 2009). However, there a dearth of scientific and clinical evidence supporting effectiveness, usefulness and safety of herbal compound used in traditional medicine. Because of lack evaluation of the toxicity and negative reactions of medicinal herbs, the use of natural compounds may prove unsafe.

Malaysia, with its tropical rainforests, is blessed with high biodiversity. The Malaysian forest is an enormous potential source of chemicals and metabolites that can be developed into new agents or novel drugs for treatment of chronic diseases (Indubala and Ng, 2000). The jungles of South East Asia has provided more than 6,500 different plants that have been used in the treatment of various illnesses particularly cancers (Burkill et al., 1966). The South East Asians seemed to have lower risks for development cancers including colon, gastrointestinal, prostate and breast compared to Westerners (Lim, 2002). It is probably the practice of regular consumption of natural plant products that contributes to the lower incidence of these debilitating diseases in the South East Asians.

Recently, in our laboratory, ZER was made soluble by incorporating in the cyclodextrin complex. The production of the ZER-cyclodextrin complex enabled ZER to be formulated as an encapsulated natural compound ready for use, either as an injectable solution or delivered orally as an anticancer product (Eid et al., 2011; Nabilah et al., 2013). The usefulness of encapsulated ZER complex as potential anticancer is worth future exploration through pre-clinical and human clinical trials to determine efficacy and safety of the product for human use. More recently we also encapsulated ZER into a nanostructured lipid carrier (NLC) using the high pressure homogenization (HPH) technique. The physiochemical properties, entrapment efficiency, storage stability, in vitro release and cytotoxic effect of this formulation against human acute lymphocytic leukemia (Jurkat) cell line was studied and showed promising results. Our study also showed the ZER-loaded NLC can be further developed as a drug delivery system for
cancer therapy (Rahman, H.S. et al., 2013b; Rasedee et al., 2013). This new approach to using a natural metabolite in innovative delivery systems would seemingly be an alternative and new approach in the treatment of cancers (Rahman, H.S. et al., 2014).

This review has clearly indicated that ZER from Z. zerumbet Smith possesses various beneficial in vitro and in vivo biological activities. The findings from all the researches reviewed in this article are conclusive evidences that ZER is a strong potential candidate for anti-cancer compound. There is need to conduct animal studies and human clinical trials to ascertain the efficacy, usefulness and safety of this compound as an intended pharmaceutical drug.

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