**Introduction**

*Zingiber zerumbet* (L.) Roscoe ex Sm. is a species in the Zingiberaceae family and is commonly known as *lempoyang* in Malay and, among others, bitter ginger [1] and shampoo ginger in English [2]. It is native to tropical and subtropical Asia [3] and has spread throughout the Pacific [4] due to cultivation for ornamental and medicinal purposes, as well as naturalisation [5]. The rhizomes of *Z. zerumbet* are especially known for their medicinal properties. *Z. zerumbet* has a wide range of traditional uses, including treatments for typhoid, stomach ailments, allergies, poisoning, appetite enhancement, constipation, haemorrhoids, asthma, skin diseases, and postnatal care [6, 7].

Over the past decade, numerous narrative reviews have discussed various aspects of *Z. zerumbet*, including its botanical qualities, phytochemistry, pharmacognosy, pharmacological activities, and biological qualities, with the most recent comprehensive review dating back to 2017 [8–11]. A mini-review of *Z. zerumbet* in 2023 reported on its potential osteoinduction properties [12]. However, a consistent limitation among these works is the lack of a systematic methodological approach in evidence synthesis on the plant’s efficacy, and there is a need to elicit the current research status of this plant. This scoping review was conducted to systematically explore and collate the available scientific evidence on the efficacy of *Z. zerumbet* and its main phytoconstituents in various formulations, their biological mechanisms, and their safety. Results included 54 articles consisting of animal studies, while there were no published human studies. Only half of the included studies provided adequate reporting on the quality-related details of *Z. zerumbet* formulations. Identified pharmacological activities were analgesic, anti-inflammatory, anti-diabetic, anti-hyperlipidemic, anti-neoplastic, immunomodulatory, antioxidant, antipyretic, hepatoprotective, nephroprotective, gastroprotective, and locomotor-reducing activities. Notably, the ethanolic extract of *Z. zerumbet* was found to be well tolerated for up to 28 days. In conclusion, *Z. zerumbet* and zerumbone have various pharmacological effects, especially in analgesic and anti-inflammatory models. However, there is still a pressing need for comprehensive safety data to conduct clinical trials.
of a systematic methodological approach in evidence synthesis, with a majority focusing on in vitro studies. In view of the rising interest in the health benefits of *Z. zerumbet*, this scoping review aims to systematically explore, consolidate, and provide an overview of both animal and human studies concerning *Z. zerumbet* and its major phytoconstituents related to its pharmacological efficacy, the potential biological mechanisms involved, and their safety profile. With this information, the potential areas of its therapeutic use that remain unexplored will be uncovered.

**Results**

**Study Inclusion**

A total of 54 articles were selected from an initial pool of 2920 records. All included studies were preclinical in vivo studies, as no published clinical studies were identified. The study selection process is presented in the preferred reporting items for systematic reviews and meta-analyses (PRISMA) [13] flowchart, as shown in ▶ Fig. 1.

**Characteristics of included studies**

Overall, the studies examined the efficacy and safety of *Z. zerumbet* in the form of extracts and its primary phytoconstituent, zerumbone. These extracts and zerumbone were sourced from the rhizomes of the *Z. zerumbet* plant. Out of the included studies, 26 underwent an authentication process through the deposition of a voucher specimen of the plant. A total of 33 studies reported a qualitative analysis to identify the phytochemicals associated with *Z. zerumbet*, while 25 studies carried out a quantitative analysis to ascertain the composition of these phytochemicals in *Z. zerumbet*. Only one study utilised a standardised formulation of the ethanolic extract of *Z. zerumbet* (EEZZ). The interventions were administered via topical, oral, subcutaneous, intraperito-
neal, intraduodenally, and inhalation routes. The checklist for the qualitative, quantitative, and standardisation of the herbal interventions for all included studies can be found in Supplementary material: Table 1S.

**Risk of Bias Assessment**

The risk of bias (ROB) assessment for the studies is presented in **Fig. 2 (ROB graph)** and **Fig. 3 (ROB summary)**. Over 75% of the studies exhibited a low ROB in baseline characteristics and selective reporting. However, half of the studies showed an unclear...
ROB with regards to sequence generation, allocation concealment, random housing, blinding of trial caregivers and researchers, random outcome assessment, and blinding of outcome assessors. This suggests that many animal studies related to \textit{Z. zerumbet} show concerns regarding selection, performance, and detection bias. Nearly 25% of the studies displayed a high ROB for incomplete outcome data (attrition bias).

**Efficacy**

All 54 included studies were preclinical \textit{in vivo} studies, with 38 further supported by additional \textit{in vitro} findings that explored potential mechanisms of action. The main pharmacological activities identified from the studies encompassed analgesia, anti-inflammatory, anti-diabetic, anti-hyperlipidemic, anti-neoplastic, immunomodulatory, antioxidant, antipyretic, hepatoprotective, nephroprotective, gastroprotective, and reduced locomotor activities. The scientific evidence detailing the pharmacological properties of \textit{Z. zerumbet} and its phytoconstituent is presented in the tables and in the subsequent narrative. Only data with a statistically significant p-value of less than 0.05 were included, while results with insignificant findings were omitted.

**Analgesia**

The analgesic effects of \textit{Z. zerumbet} methanol extract, \textit{Z. zerumbet} essential oil, and zerumbone were reported via intraperitoneal, oral, and subcutaneous routes. Detailed findings on the analgesic effects of \textit{Z. zerumbet} and zerumbone are presented in ▶ Table 1.

**Anti-inflammatory**

The anti-inflammatory properties of \textit{Z. zerumbet} were reported in the form of essential oil via the intraperitoneal route and zerumbone through topical, intraperitoneal, and oral administration. Detailed findings on the anti-inflammatory properties of \textit{Z. zerumbet} and zerumbone are presented in ▶ Table 2.

**Anti-diabetic**

Ethanol extract of \textit{Z. zerumbet} and zerumbone was reported to have anti-diabetic properties. Detailed findings of the anti-diabetic effects of \textit{Z. zerumbet} and zerumbone are presented in ▶ Table 3.

**Anti-hyperlipidaemia**

EEZZ and zerumbone administered orally showed anti-hyperlipidaemic properties. Detailed findings on the anti-hyperlipidaemic properties of \textit{Z. zerumbet} and zerumbone are presented in ▶ Table 4.

**Anti-neoplastic**

\textit{Z. zerumbet} was shown to have anti-angiogenetic and anti-tumour properties. Detailed findings on the anti-neoplastic properties of \textit{Z. zerumbet} extracts and zerumbone are presented in ▶ Table 5.
Three studies reported the immunomodulatory properties of *Z. zerumbet* and zerumbone. In male BALB/c mice, zerumbone was observed to suppress macrophage phagocytosis (part of the innate immune system) and inhibit nitrous oxide production in a concentration-dependent manner at dosages ranging from 25 to 100 mg/kg when administered orally, once daily for 14 days [14]. In female BALB/c mice with ovalbumin (OVA)-induced T helper 2 (Th2)-mediated asthma, zerumbone improved airway hyperresponsiveness and reduced airway inflammation. This was noted at dosages of 0.1 to 10 mg/kg, administered orally three times daily for 17 days [15]. Studies on male Wistar rats revealed that an 80% ethanol extract of *Z. zerumbet* has mild immunosuppressive effects by reducing the phagocytic activity of neutrophils (another component of the innate immune system). Additionally, the ethanol extract of *Z. zerumbet* influenced the adaptive immune system by inhibiting neutrophil migration, CD11β/CD18 integrin expression, and production of reactive oxygen species (ROS) in a dose-dependent manner, at dosages ranging from 100 to 400 mg/kg when given orally daily for 15 days [16].

### Table 1

The mechanisms by which *Z. zerumbet* formulations can contribute to analgesic and antinociceptive effects.

<table>
<thead>
<tr>
<th>Animal</th>
<th>Intervention</th>
<th>Disease model</th>
<th>Administration details</th>
<th>Mechanism</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat</td>
<td>Zerumbone</td>
<td>Osteoarthritis</td>
<td>10–50 mg/kg, single dose, i.p</td>
<td>Suppress NO, PGE₂, and MMP production</td>
<td>Chien, 2016 [45]</td>
</tr>
<tr>
<td>Mice</td>
<td>80% methanol extract of <em>Z. zerumbet</em></td>
<td>Inflammation and nociception</td>
<td>25–100 mg/kg, single dose, s.c</td>
<td>Inhibit opioid receptors, bradykinin, prostaglandin, and histamine-mediated actions</td>
<td>Zakaria, 2010 [46]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Chronic constriction injury-induced</td>
<td>10 mg/kg, single dose, i.p</td>
<td>Stimulate serotonin inhibitory pathway (5-HT receptor subtypes 1A, 1B, 2A, 3, 6, and 7)</td>
<td>Chia, 2016 [47]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Chronic constriction injury-induced</td>
<td>10 mg/kg, single dose, i.p</td>
<td>Agonist of potassium channels (voltage-dependent K⁺, ATP-sensitive K⁺ and Ca²⁺-K⁺ channels) Agonist of the non-selective opioid receptors and selective opioid receptors (μ-opioid receptors, δ-opioid and κ-opioid)</td>
<td>Gopalsamy, 2020 [36]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Neuropathic pain</td>
<td>5–50 mg/kg, once daily, 14 days, p.o</td>
<td>Agonist of CB-1 receptor</td>
<td>Chia, 2021 [48]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Neuropathic pain</td>
<td>5–50 mg/kg, once daily, 14 days, p.o</td>
<td>Inhibit production of IL-1β, IL-6 and TNF-α in blood plasma and spinal cord tissues</td>
<td>Gopalsamy, 2017 [37]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Neuropathic pain</td>
<td>5–100 mg/kg, once daily, 7 days, i.p</td>
<td>Inhibit mechanical allodynia, thermal allodynia, and hyperalgesia. The mechanism of action was not reported</td>
<td>Zulazmi, 2015 [49]</td>
</tr>
<tr>
<td>Mice</td>
<td><em>Z. zerumbet</em> essential oil</td>
<td>General antinociception</td>
<td>50–300 mg/kg, single dose i.p and p.o</td>
<td>Activate L arginine/NO/cGMP/ATP-sensitive K⁺ channel pathway Inhibit glutamatergic system and TRPV1 receptors Activate opioidergic system by acting as an agonist to the non-selective opioid receptors Inhibit the inflammatory mediators, prostaglandin, histamine, serotonin, and bradykinin</td>
<td>Khalid, 2011 [50] Sulaiman, 2010b [30]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>General antinociception</td>
<td>10–100 mg/kg, single dose, i.p</td>
<td>Agonist of the non-selective opioid receptors</td>
<td>Sulaiman, 2009 [38]</td>
</tr>
</tbody>
</table>

Abbreviations: i.p: intraperitoneal; s.c: subcutaneous; p.o: per oral; NO: nitric oxide; PGE₂: prostaglandin E₂; MMP: matrix metalloproteinase; 5-HT: 5-hydroxytryptamine; CB: cannabinoid; IL-1β: interleukin-1 beta; IL-6: interleukin-6; TNF-α: tumour necrosis factor alpha; cGMP: cyclic guanosine monophosphate; ATP: adenosine triphosphate; TRPV1: transient receptor potenzial vanilloid 1.
ported in animal models of brain, lung, and skin damages. In male Wistar rats with induced brain damage, the treatment of ethylacetate Z. zerumbet extract significantly reduced the level of oxidative stress markers such as malondialdehyde (MDA) and protein carbonyl in the brain homogenate. This treatment given at dosages of 200 to 400 mg/kg, once daily by oral gavage 30 minutes before ethanol exposure via intraperitoneal route for 14 consecutive days, also enhanced the activities of serum superoxide dismutase (SOD), catalase (CAT), and glutathione (GSH) levels in a dose-dependent manner [17]. In adult male pathogen-free Institute of Cancer Research mice with lipopolysaccharide (LPS)-induced acute lung injury (ALI), zerumbone pretreatment ameliorated histopathological lung changes, such as neutrophil infiltration, increased alveolar wall thickness, haemorrhage, and hyaline membrane formation. Zerumbone at dosages from 1 to 10 µmol/kg suppressed LPS-induced activation of myeloperoxidase (MPO), metalloproteinase-9 (MMP-9), and lipid peroxidation in the lungs, reversed the LPS-induced reduction in antioxidative enzyme (superoxide dismutase, catalase, and glutathione peroxidase) activities in a concentration-dependent manner, and reduced LPS-induced oxidative stress through the mechanism of nuclear factor erythroid 2-related factor (Nrf2) and heme oxygenase (HO-1) [18]. In a separate study on athymic female nude mice (BALB/c-nu) exploring skin damage from UVA radiation, topical zerumbone pretreatment significantly countered the damage. Applied at 55 or 110 µg/day for 14 days, the treatment upregulated Nrf2- and Nrf2-dependent antioxidative genes, particularly HO-1 and γ-glutamyl cysteine ligase (γ-GCLC). This protective action functioned in a dose-dependent manner, further involving the downregulation of the Bax/Bcl-2 ratio in keratinocytes and the prevention of DNA fragmentation [19].

Abbreviations. w/w: weight for weight; COX-2: cyclooxygenase-2; EOZZ: essential oil of Z. zerumbet; VEGF: vascular endothelial growth factor; TGF-β1: transforming growth factor beta 1; IL-10: interleukin 10; iNOS: inducible nitric oxide synthase; NFXB: nuclear factor kappa-light-chain-enhancer of activated B cells; IκB: IκBα; LPS: lipopolysaccharide; ICAM-1: intercellular adhesion molecule-1; IL-1β: interleukin 1 beta; MIP-2: macrophage inflammatory protein 2; ETBF: enterotoxigenic B. fragilis; NR: not reported

Table 2: The mechanisms by which Z. zerumbet formulations can contribute to anti-inflammation.

<table>
<thead>
<tr>
<th>Animal</th>
<th>Intervention</th>
<th>Disease model</th>
<th>Administration details</th>
<th>Mechanism</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>2A. Acute</td>
<td>Rat</td>
<td>Zerumbone</td>
<td>Excisional wound (for wound-healing effects)</td>
<td>0.5 mg/mL, once daily, 15 days, topical</td>
<td>Downregulate IL-6, TNF-α, and COX-2 gene, while increasing IL-10 expression in wound tissues</td>
</tr>
<tr>
<td></td>
<td>Mice</td>
<td>Zerumbone</td>
<td>Excisional wound (for wound-healing effects)</td>
<td>0.01 or 1% (w/w), once daily, 15 days, topical</td>
<td>Increase VEGF, TGF-β1, and collagen IV expressions which correlates with increase fibroblast proliferation and collagen synthesis</td>
</tr>
<tr>
<td></td>
<td>Mice</td>
<td>Zerumbone</td>
<td>Acute lung injury</td>
<td>0–10 mmol/kg, single dose, i.p</td>
<td>Inhibit expression of TNF-α, IL-6, iNOS, and COX-2</td>
</tr>
<tr>
<td></td>
<td>Mice</td>
<td>Zerumbone</td>
<td>Acute lung injury</td>
<td>0–2183.4 µg/kg, single dose, i.p</td>
<td>Reduce neutrophil infiltration by decreasing expression of ICAM-1</td>
</tr>
<tr>
<td>2B. Chronic</td>
<td>Mice</td>
<td>Zerumbone</td>
<td>Enterotoxigenic Bacteroides fragilis (ETBF) infection</td>
<td>30–60 mg/kg/day, 7 days, p.o</td>
<td>Inhibit NFXB signalling that decreases ETBF-induced colitis</td>
</tr>
<tr>
<td>2C. Mixed</td>
<td>Mice</td>
<td>Zerumbone</td>
<td>Ulcerative colitis</td>
<td>0.1%, ad libitum, 14 days, p.o</td>
<td>Reduce PGE2 formation in colonic mucus membrane</td>
</tr>
<tr>
<td></td>
<td>Mice</td>
<td>Zerumbone</td>
<td>Acute and chronic inflammation</td>
<td>5–100 mg/kg, single dose, i.p</td>
<td>Inhibit fibroblasts activity and synthesis of collagen with mucopolysaccharide, in granulation tissue formation</td>
</tr>
<tr>
<td></td>
<td>Rat</td>
<td>EOZZ</td>
<td>General anti-inflammatory activity</td>
<td>Acute inflammation: 30–300 mg/kg, single dose, i.p; Chronic inflammation: 30–300 mg/kg, once daily, 7 days, i.p</td>
<td>Reduce oedema, acute inflammation, chronic inflammation, and inflammatory- and noninflammatory-mediated pain. Mechanism of action was not reported</td>
</tr>
</tbody>
</table>

Z. zerumbet; VEGF: vascular endothelial growth factor; TGF-β1: transforming growth factor beta 1; IL-10: interleukin 10; iNOS: inducible nitric oxide synthase; NFXB: nuclear factor kappa-light-chain-enhancer of activated B cells; IκB: IκBα; LPS: lipopolysaccharide; ICAM-1: intercellular adhesion molecule-1; IL-1β: interleukin 1 beta; MIP-2: macrophage inflammatory protein 2; ETBF: enterotoxigenic B. fragilis; NR: not reported.
As a result, there might be a decrease in the conversion of fatty acids, ultimately contributing to an increase in body weight [21].

The inhalation of zerumbone were found to reduce the rectal temperature in rats by about 1.3 °C within 2 hours. However, this reduction was not as pronounced as that produced by paracetamol, which lowered the rate of gluconeogenesis in the liver

The inhalation of Z. zerumbet essential oil did not have any effect on the toxicity and oxidative stress in male Sprague-Dawley rats. When the Z. zerumbet extract was administered intraperitoneally at doses of 200 and 400 mg/kg for 7 days, there were marked reductions in creatinine elevations and oxidative stress indicators. Specifically, there were decreased levels of renal homogenate, plasma

**Table 3** The mechanisms by which Z. zerumbet formulations can affect diabetic-related diseases.

<table>
<thead>
<tr>
<th>Animal</th>
<th>Intervention</th>
<th>Disease model</th>
<th>Administration details</th>
<th>Mechanism</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat</td>
<td>EEZZ</td>
<td>Diabetic retinopathy</td>
<td>200–300 mg/kg, once daily, 3 months, p.o</td>
<td>Stabilise tight junction proteins, leading to decreasing blood-retinal-barrier permeability</td>
<td>Tzeng, 2015 [57]</td>
</tr>
<tr>
<td>Rat</td>
<td>EEZZ</td>
<td>Diabetic retinopathy</td>
<td>200–300 mg/kg, once daily, 3 months, p.o</td>
<td>Prevent activation of ERK1/2 phosphorylation and NFκB, downregulating pro-inflammatory mediators</td>
<td>Hong, 2016 [58]</td>
</tr>
<tr>
<td>Rat</td>
<td>Zerumbone</td>
<td>Diabetic retinopathy</td>
<td>40 mg/kg, once daily, 8 weeks, p.o</td>
<td>Inhibit NF-κB activation and reduce VEGF expression in retinal tissue, thereby inhibiting retinal inflammation</td>
<td>Tzeng, 2016 [59]</td>
</tr>
<tr>
<td>Rat</td>
<td>EEZZ</td>
<td>Diabetic nephropathy</td>
<td>200–300 mg/kg, once daily, 8 weeks, p.o</td>
<td>Inhibit AMPK dephosphorylation in the kidneys</td>
<td>Tzeng, 2013 [60]</td>
</tr>
<tr>
<td>Rat</td>
<td>Zerumbone</td>
<td>Diabetic nephropathy</td>
<td>20–40 mg/kg, once daily, 8 weeks, p.o</td>
<td>Reduce upregulation of protein expression of TNF-α, IL-1β and IL-6 in the kidneys</td>
<td>Tzeng, 2013 [61]</td>
</tr>
</tbody>
</table>

**Antipyretic**

One study involving albino rats reported on the antipyretic properties of the EEZZ at doses of 1 to 4 g/kg and zerumbone at 0.75 g/kg of body weight, administered orally. Both EEZZ and zerumbone were found to reduce the rectal temperature in rats by about 1.3 °C within 2 hours. However, this reduction was not as pronounced as that produced by paracetamol, which lowered the temperature by 1.7 °C within 3 hours [20].

**Weight gain**

In male Sprague-Dawley rats on a high-fat diet, the inhalation of Z. zerumbet essential oil and zerumbone was observed to further increase body weight. While the inhalation of zerumbone decreased brown adipose tissue (BAT) sympathetic nerve activity, inhalation of Z. zerumbet essential oil did not have any effect on the BAT activity. It has been suggested that this decrease in BAT sympathetic nerve activity could lead to diminished thermogenesis. As a result, there might be a decrease in the conversion of fatty acids, ultimately contributing to an increase in body weight [21].

**Hepatoprotective effects**

Two studies investigated the hepatoprotective properties of zerumbone in male Sprague-Dawley rats and C57BL/6 mice. In both studies, zerumbone was found to restore neutrophil levels, to reduce ALT and AST levels, and to maintain normal hepatic tissue histology [22, 23]. At high doses of 50 mg/kg, zerumbone was observed to downregulate the expression levels of IL-1β and TNFα. It also reduces the terminal deoxynucleotidyl transferase dUTP nick end labelling (TUNEL)-positive area in male C57BL/6 mice subjected to hepatotoxin-mediated acute and chronic liver injuries [23].

**Nephroprotective effects**

A study highlighted the nephroprotective effect of the ethyl acetate extract of Z. zerumbet against paracetamol-induced nephrotoxicity and oxidative stress in male Sprague-Dawley rats. When the Z. zerumbet extract was administered intraperitoneally at doses of 200 and 400 mg/kg for 7 days, there were marked reductions in creatinine elevations and oxidative stress indicators.
malondialdehyde (MDA), plasma protein carbonyl, and renal advanced oxidation protein product (AOPP). Additionally, the histological evaluation indicated better protection of the kidneys, especially in the appearance of glomeruli and tubules, when compared to the untreated group. This protection was observed to be dose-dependent [24].

**Gastroprotective effects**

One study reported on the gastroprotective property of zerumbone in an ethanol-induced gastric ulcer model using male Sprague-Dawley rats. When zerumbone was administered intraduodenally at doses of 5 and 10 mg/kg, there was a significant reduction in the acidity of gastric juice compared to the control group subjected to pylorus ligation. This effect was comparable to that of omeprazole at 30 mg/kg. Rats pretreated with zerumbone demonstrated a decrease in ulcer area formation, an increase in mucus production, and a reduction in both oedema and leukocyte infiltration. There was also a noticeable flattening of the mucosal fold and preservation of the gastric mucosa layer. Additionally, there was an overexpression of heat shock protein 70 (HSP-70) in the gastric tissue, suggesting enhanced protection of the gastric mucosa, since HSP-70 combats stress-induced protein denaturation. Following zerumbone treatment, there was a restoration in the levels of prostaglandin E2 (PGE2), glutathione (GSH), and lipid peroxidation in comparison to the ulcer control group [25].
Two studies investigated the locomotor-reducing effects of the phytoc constituents of *Z. zerumbet* rhizomes. Ogawa et al. reported a decrease in total spontaneous locomotor activity in mice after a 60-minute inhalation of zerumbone and its derivatives, with a concentration of $4.5 \times 10^{-3}$ mg being the most significant [26]. Another study by the same primary author focused on inhaled hexahydrozerumbone derivatives and zerumbol. Hexahydrozerumbone significantly reduced the total spontaneous locomotor activity in mice at a dose of $4.5 \times 10^{-3}$ mg, whereas zerumbol did not show any significant effects [27]. The mechanism behind this reduced locomotion was not determined in either of the studies.

**Safety**

General toxicity studies for the ethanol extract of *Z. zerumbet* and zerumbone were conducted in seven studies [14–16, 28–31], with results presented in Table 6. Overall, no deaths or severe abnormalities were observed for most of the investigated doses. In addition to these toxicity studies, four studies reported no adverse events from the use of *Z. zerumbet* extracts and zerumbone during efficacy studies [32–35], while three other studies indicated that zerumbone did not exert sedative effects [36–38]. The ethanolic extract of *Z. zerumbet* demonstrated no genotoxic effects in mice based on their bone marrow studies [31]. A summary of the

**Locomotor-reducing activity**

Two studies investigated the locomotor-reducing effects of the phytoc constituents of *Z. zerumbet* rhizomes. Ogawa et al. reported a decrease in total spontaneous locomotor activity in mice after a 60-minute inhalation of zerumbone and its derivatives, with a concentration of $4.5 \times 10^{-3}$ mg being the most significant [26]. Another study by the same primary author focused on inhaled hexahydrozerumbone derivatives and zerumbol. Hexahydrozerumbone significantly reduced the total spontaneous locomotor activity in mice at a dose of $4.5 \times 10^{-3}$ mg, whereas zerumbol did not show any significant effects [27]. The mechanism behind this reduced locomotion was not determined in either of the studies.

**Table 5** The mechanisms by which *Z. zerumbet* formulations contribute to anti-neoplastic effects.

<table>
<thead>
<tr>
<th>Animal</th>
<th>Intervention</th>
<th>Disease model</th>
<th>Administration details</th>
<th>Mechanism</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Angiogenesis</td>
<td>10–200 µM, single dose, s.c</td>
<td>Inhibit proliferation, migration and blood capillary formation</td>
<td>Park, 2015 [69]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone and ELEZZ</td>
<td>Colon and lung cancer</td>
<td>0.5–2.0 mg/kg, once daily, 8 days, i.p (in vivo antitumor P388D1; assay) ELEZZ: 1.25–10.0 mg/kg, once daily, 8 days, i.p (in vivo antitumor P388D1; assay)</td>
<td>Prolong survival days in lymphoma animal model (mechanism unclear)</td>
<td>Huang, 2005 [70]</td>
</tr>
<tr>
<td>Mice</td>
<td>MEZZR</td>
<td>Ehrlich ascites carcinoma</td>
<td>10–20 mg/kg/day, 5 days, i.p</td>
<td>Cancer cell apoptosis in the presence of caspase-3, -8, and -9 inhibitors</td>
<td>Hanif, 2022 [71]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Colon carcinogenesis</td>
<td>Colon carcinogenesis 100–500 ppm, ad libitum, 17 weeks, p.o Lung carcinogenesis 100–500 ppm, ad libitum, 21 weeks, p.o</td>
<td>Reduce NFκB and HO-1 expression in tumours. Suppress cell proliferation Induce apoptosis</td>
<td>Kim, 2009 [33]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Skin cancer</td>
<td>1–10 µmol, topical on dorsal skin, 24 hours</td>
<td>Increase HO-1 mRNA expression through transcriptional activation of Hmox1, mediated through the activation of Nrf2 signalling.</td>
<td>Shin, 2011 [72]</td>
</tr>
<tr>
<td>Mice</td>
<td>Zerumbone</td>
<td>Non-small-cell lung cancer</td>
<td>Mice treated 5 times (route, dose and duration of zerumbone not stated)</td>
<td>Inhibit the binding activity between HSP27 and PKCδ or cytochrome C in tumour tissue lysates, improving the effects of chemo- or radiation treatment</td>
<td>Choi, 2011 [73]</td>
</tr>
<tr>
<td>Rat</td>
<td>Zerumbone</td>
<td>Liver cancer</td>
<td>15–60 mg/kg, twice per week, 11 weeks, i.p</td>
<td>Induce apoptosis via increasing Bax gene while decreasing Bcl-2 protein expression</td>
<td>Taha, 2010 [34]</td>
</tr>
<tr>
<td>Rat</td>
<td>Zerumbone</td>
<td>Colon cancer</td>
<td>0.01–0.05 %, ad libitum, 5 weeks, p.o</td>
<td>Reduce expression of COX-2, PGE2 and PGD2 in colonic mucosa Reduce cell proliferation activity (seen by decreased AgNORs number) in colonic cryptal cell nuclei</td>
<td>Tanaka, 2001 [35]</td>
</tr>
</tbody>
</table>

**Abbreviations:** VEGFR2: vascular endothelial growth factor receptor 2; FGFR1: fibroblast growth factor receptor 1; MEZZR: Methanol extract of *Z. zerumbet* rhizome; P388D1: murine lymphoid neoplasms cell line; HL-60: human promyelocytic leukaemia cell; G2/M: Gap 2 phase mitosis; ppm: parts per million; HO-1: heme oxygenase-1; ADC: Adenocarcinoma; AD: Adenoma; Hmox1: heme oxygenase 1 gene; Nrf2: nuclear factor-erythroid 2-related factor 2; HSP27: heat shock protein 27; PKCδ: protein kinase C delta; Bax: B-cell lymphoma protein 2- associated X; Bcl-2 protein: B-cell lymphoma protein 2; AgNORs: silver-stained nucleolar organiser regions protein; PGE2: prostaglandin E2; PGD2: prostaglandin D2
preclinical in vivo safety studies done for Z. zerumbet and zerumbone can also be found in Table 6.

**Discussion**

The bulk of the evidence focused on the analgesic, anti-inflammatory, anti-diabetic, anti-hyperlipidemia, and anti-neoplastic properties of Z. zerumbet and zerumbone. A small number of studies reported their antioxidant, antipyretic, hepatoprotective, nephroprotective, and gastroprotective properties, as well as their locomotor-reducing activities. Among these pharmacological effects, the most researched areas were analgesia and anti-inflammation. In terms of formulations and dosages, three were commonly utilised: the methanolic extract of Z. zerumbet at dosages of 25–100 mg/kg administered via the intraperitoneal route; the essential oil of Z. zerumbet at dosages of 30–300 mg/kg given orally or intraperitoneally; and zerumbone derived from Z. zerumbet at dosages 5–100 mg/kg administered either orally or intraperitoneally. Z. zerumbet may exert its various pharmacological effects through the phytochemicals contained in the plant such as triterpenes, saponins, tannins, and other volatile oils, particularly the zerumbone compound, which is a sesquiterpenoid [11].

Based on the included studies, the ethanolic extract of Z. zerumbet appears safe in short-term animal toxicity studies for up to 28 days, with no evident safety concerns. The essential oil of Z. zerumbet, when administered intraperitoneally in up to doses of 5000 mg/kg, showed neither mortality nor adverse effects. Zerumbone, however, presented a more mixed picture. One study reported adverse effects at a dose of 200 mg/kg, but other studies using even higher doses of up to 1000 mg/kg did not confirm these findings. These adverse effects encompassed appetite loss, lowered body temperature, changes in general behavioural activities; and colour of skin, hairs, teeth, and eyes in 200 mg/kg group. Based on the included studies, the ethanolic extract of Z. zerumbet appears safe in short-term animal toxicity studies for up to 28 days, with no evident safety concerns. The essential oil of Z. zerumbet, when administered intraperitoneally in up to doses of 5000 mg/kg, showed neither mortality nor adverse effects. Zerumbone, however, presented a more mixed picture. One study reported adverse effects at a dose of 200 mg/kg, but other studies using even higher doses of up to 1000 mg/kg did not confirm these findings. These adverse effects encompassed appetite loss, lowered body temperature, changes in general behavioural activities; and colour of skin, hairs, teeth, and eyes in 200 mg/kg group.
Documented traditional uses of *Z. zerumbet* that we have access to include its use as an appetiser and as treatment for stomach aches [7], pain relief, toothaches, alleviation of a cough related to cavities, asthma, deworming, and various unspecified skin diseases [39]. Based on our findings, the most substantiated therapeutic claims of *Z. zerumbet* are analgesic and anti-inflammatory. This can be linked, both directly and indirectly, to toothaches, cough, asthma, and skin diseases—primarily through its anti-inflammatory attributes. Modern research has identified claims for *Z. zerumbet* that are not documented in its traditional uses. These claims include anti-diabetic, anti-hyperlipidemic, anti-neoplastic, hepatoprotective, and nephroprotective effects, as well as the reduction of locomotor activity.

We found that approximately half of the studies reported, in detail, the qualitative and quantitative phytochemical analyses of the herbal interventions. A significant gap in the herbal medicine literature on safety and efficacy is the lack of comprehensive reporting on the quality details of the formulations under investigation [40, 41]. Given that the phytoconstituents of medicinal plants can vary based on agroclimatic conditions and processing methods [41], it is vital to provide detailed reports on the quality-related components of a formulation being studied. Despite the substantial amount of preclinical evidence, we could not find any published clinical trial. The availability of such data will facilitate a more insightful interpretation of the dose-response relationship and enable extrapolation to similar formulations of the same plant, further bridging the gap towards successful clinical studies. Currently, based on the preclinical in vivo efficacy data, most of the research focuses on the anti-inflammatory and analgesic effects of *Z. zerumbet*, indicating a promising direction for future clinical trials.

One limitation of this review is the inclusion of only articles written in English. Due to the limited availability of human literature, a meaningful appraisal could not be conducted. Our safety data are derived primarily from animal toxicity studies and from the extraction of safety-related data within efficacy studies, given the design of our search strategy. This approach might not capture all safety-related data. Furthermore, our institution might not have access to all traditional medicinal claims related to *Z. zerumbet*, especially those from non-English sources or global traditional practices, potentially leading to certain oversights.

In conclusion, the outcomes of the studies demonstrate that *Z. zerumbet* holds promise in the field of natural products with therapeutic claims, particularly in addressing pain and anti-inflammatory conditions. The combined effects of this plant could potentially offer comprehensive symptom relief for various diseases. However, the future prospects of this review suggest the need for further research. This includes standardising *Z. zerumbet* formulations, extending the safety studies based on its duration of use, and investigating its pharmacokinetic properties. A specialised review centred on the safety and potential herb–drug interactions of *Z. zerumbet* would further enrich the field. Furthermore, it is imperative to establish rigorous herbal quality standards to enhance the interpretation of results and pave the way for successful clinical trials in the future.

### Methodology

We conducted a scoping review according to the York framework of scoping studies by Arskey and O’Malley [42]. This framework was appropriate for the broad range of preclinical evidence comprising of the efficacy and safety of *Z. zerumbet*. This scoping review has been registered with the National Medical Research Register (NMRR) under the research ID 21–526–59312 with an *a priori* protocol prepared. To ensure the transparency and comprehensiveness of our scoping review, we followed the preferred reporting items for systematic reviews and meta-analyses (PRISMA) guidelines, which involved using the PRISMA flowchart to document the screening and selection process, as well as the PRISMA scoping review checklist (Supplementary material: Table 2S), to ensure relevant items were included in the review [13].

### Research Questions

This scoping review was based on the research question “What is the current scientific evidence on *Z. zerumbet* as a natural product?” and was further subdivided to categorise the types of evidence, which include the following:

1. What is the pharmacological scientific evidence of *Z. zerumbet*?
2. What is the safety profile of *Z. zerumbet* in animal toxicity studies and its potential harm to humans?

The population, intervention, comparison, and outcomes (PICO) framework shown in Table 7 was used to approach the research study questions.

### Search Strategy

A systematic search was conducted by two independent investigators on electronic databases including MEDLINE, CENTRAL, Lilacs, and Google Scholar from the period since commencement to 31st March 2023. A predetermined combination of keywords that include “Zingiber zerumbet” and its synonyms, “medicinal”, “therapeutic”, “benefit”, “effect”, “properties”, and “bioactive” were used. An example of the keyword search used in the databases is presented in the Supplementary material: Table 3S.
The abstracts of the searched results were extracted with duplicates removed using the bibliographical software EndNote 20.

**Article Inclusion and Data Extraction**

The search result was transferred to a Microsoft Excel sheet. Title, abstract, and full-text article screening was performed by two independent investigators, with disagreements resolved by a third investigator. This review accounted for *Z. zerumbet* as a whole plant used in any formulation (crude, extract, and essential oil) and its major compound studied, zerumbone. Only English-language articles were included. The inclusion criteria comprised all published primary literature of animal and clinical studies on the efficacy and safety of *Z. zerumbet*, of animal studies that incorporate *in vitro* studies to elicit the mechanism of action, of any plant part, and of any formulations with *Z. zerumbet* as a sole active ingredient and its representative compound isolated from the plant (i.e., zerumbone). The exclusion criteria comprised review papers, book sections, combination products and formulation, and *in silico* and purely *in vitro* studies. A data extraction table of included studies (the table layout provided in Supplementary material: Table 7S) was created to record all the relevant data upon full-text screening.

**Data analysis**

**Full-Text Analysis**

Descriptive numerical analysis on the efficacy and safety of *Z. zerumbet* was performed. For efficacy, we focused on data related to its intended pharmacological effects, the underlying cellular and molecular mechanisms, and the range of doses shown to be effective. In terms of safety, the primary data was sourced from animal toxicity studies. This encompassed information about the dose range tested, any resulting morbidity or mortality, and other pertinent findings from clinical evaluations, histopathological examinations, and laboratory tests.

**Risk of Bias Assessment**

The risk of bias for each included study was assessed independently by two authors, TYCT and JSWC. For this assessment, we used the systematic review Centre for Laboratory Animal Experimentation risk of bias tool (SYRCLE’s RoB) [43]. This tool has 10 domains:

1. Sequence generation;
2. Baseline characteristics;
3. Allocation concealment;
4. Random housing;
5. Blinding of trial caregivers;
6. Random outcome assessment;
7. Blinding of outcome assessors;
8. Incomplete outcome data;
9. Selective reporting;
10. Other biases.

For each criterion, the study was judged as having a ‘low’, ‘unclear’, or ‘high’ risk of bias. Justifications for each judgment were provided in a risk-of-bias table. Additionally, we visualised the overall results using the review manager application by Cochrane (RevMan 5.4.1) to generate the risk-of-bias graph and summary [44].

**Supporting Information**

The herbal intervention qualitative, quantitative, and standardisation checklist, the PRISMA scoping review checklist, the keyword search strategy, and the data extraction table layout are provided in the Supporting Information.

**Contributors’ Statement**

All the authors were involved in the abstract and full-text screening of the included studies, crosschecked among pairs, and tabulated data from the included studies into the data extraction sheet. JSWC prepared the data extraction table for full text analysis, analysed and interpreted the results of the included studies, drafted the manuscript, designed the research framework, critically revised the manuscript, and discussed the results. XYL analysed, critically reviewed the interpreted data in the drafted manuscript, provided inputs on tabulating the interpreted data and discussion and interpreted the safety aspect section of the results. NJ and TYCT descriptively analysed and interpreted the data on several pharmacological efficacy aspects in the results section. TYCT provided input on the overall discussion. IFA contributed to the manuscript literature review, introduction, and proofreading. All authors have read and agreed to the published version of the manuscript.

**Acknowledgements**

The authors gratefully acknowledge the Director General of Health Malaysia, Deputy Director General of Health Malaysia (Research and Technical Support), Director of Institute for Medical Research, and Head of Herbal Medicine Research Centre for their authorisation and permission for the publication of this article. The authors have no relevant financial or non-financial interests to disclose. No funding was received to assist with the preparation of this manuscript.

**Conflict of Interest**

The authors declare that they have no conflict of interest.

**References**


Taha MM, Abdul AB, Abdullah R, Ibrahim TA, Abdulwahab Si, Mohan S. Potential chemoprevention of diethylnitrosamine-initiated and 2-acetylaminofluorene-promoted hepatocarcinogenesis by zerumbone from...


[58] Tseng TF, Liou SS, Chang CJ, Liu IM. Zerumbone, a natural cyclic sesquiterpene of Zingiber zerumbet Smith, in...


