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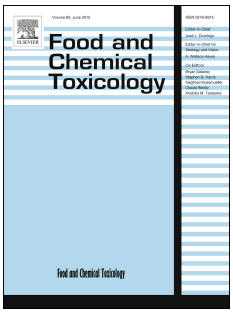

Phytol: A review of biomedical activities

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Phytol: a review of biomedical activities

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Abstract

Phytol (PYT) is a diterpene member of the long-chain unsaturated acyclic alcohols. PYT and some of its derivatives, including phytanic acid (PA), exert a wide range of biological effects. PYT is a valuable essential oil (EO) used as a fragrance and a potential candidate for a broad range of applications in the pharmaceutical and biotechnological industry. There is ample evidence that PA may play a crucial role in the development of pathophysiological states. Focusing on PYT and some of its most relevant derivatives, here we present a systematic review of reported biological activities, along with their underlying mechanism of action. Recent investigations with PYT demonstrated anxiolytic, metabolism-modulating, cytotoxic, antioxidant, autophagy- and apoptosis-inducing, antinociceptive, anti-inflammatory, immune-modulating, and antimicrobial effects. PPARs- and NF-κB-mediated activities are also discussed as mechanisms responsible for some of the bioactivities of PYT. The overall goal of this review is to discuss recent findings pertaining to PYT biological activities and its possible applications.

Keywords: phytol; phytol metabolites; phytanic acid; plant-derived compounds; biological activities.

INTRODUCTION

Phytol (PYT; 3,7,11,15-tetramethylhexadec-2-en-1-ol) is a compound found abundantly in nature. Being a part of the chlorophyll molecule, it is produced by almost all photosynthetic organisms, including algae (de Souza and Nes, 1969), plants (Ischebeck et al., 2006), and bacteria (cyanobacteria) (Proteau, 1998). It is additionally formed as an important metabolite during catabolism in ruminant animals. Consequently, PYT is considered as the most abundant acyclic isoprenoid present in the biosphere of our planet (Rontani and Volkman, 2003). In ruminants, gut digestion of consumed plant materials releases PYT, which once converted into phytanic acid (PA), is eventually stored in fat depositions (Islam et al., 2015; van den Brink and Wanders, 2006).

While PYT is primarily used as a fragrance constituent, its significant biological properties have recently drawn the attention for possible application in the pharmaceutical and biotechnological fields. Although large experimental data sets have addressed PYT metabolism, PA oxidation, and pathophysiological contributions of plasma PA levels, the exact molecular mechanisms underlying its biological actions are still insufficiently understood.

In this review, we summarize an updated and detailed overview of PYT and its derivatives based on the most recent available literature with emphasis on possible mechanisms of action of reported bioactivities, and future prospects pharmaceutical and biotechnological applications.

METABOLISM OF PYT AND PYT-DERIVATIVES

The metabolic degradation pathway of PYT is complex and has been recently described in details (Ischebeck et al., 2006; Islam et al., 2015). An overview of the overall pathway, along with the work of Ischebeck et al. on the metabolic fate of PYT in Arabidopsis, is depicted in **Figure 1** (Ischebeck et al., 2006).

PA as a primary PYT biometabolite, has been known for decades (Gloerich et al., 2007; Steinberg et al., 1966). However, the biochemical pathway of conversion between PYT and PA has not been completely characterized. Based on a comprehensive literature review, Wanders et al. (2011) have recently proposed a PYT degradation pathway: Briefly, PYT is first oxidized to phytenal by alcohol dehydrogenase (ADH), followed by oxidation to PA (Steinberg et al., 1966; Wanders et al., 2011). Based on a previous study on rat liver, van den Brink and coworkers proposed that PA is converted first to phytenoyl-CoA by acyl-CoA synthetase, followed by the action of enoyl-CoA reductase, which is located in the peroxisomal and microsomal fractions of the liver (van den Brink et al., 2005; Wanders et al., 2011). In addition, the peroxisomal enzyme *trans*-2-enoyl-CoA reductase (TER), encoded by the *PECR* gene, can convert phytenoyl-CoA to phytanoyl-CoA (Gloerich et al., 2006), which then undergoes α-oxidation in peroxisomes to give rise to pristanic acid.

α-Oxidation of PA requires the catalytic participation of different enzymes and yields pristanoyl-CoA, which is considered to be the starting product of other oxidative pathways (Wanders et al., 2011). In addition to the participating enzymes, diverse transporters play an important role in the oxidation process since the semi-permeable peroxisomal membranes allow compounds with molecular weights between 300 and 400 Da, to pass through the *PXMP2* gene

product, whereas larger molecules such as phytanoyl-CoA, require certain carrier proteins, including *PMP34* for ATP.

Upon undergoing three cycles of β-oxidation in peroxisomes, pristanoyl-CoA further produces 4,8-dimethylnonanoyl-CoA, propionyl-CoA, and acetyl-CoA. Eventually, the products of a pristanoyl-CoA β-oxidation are shuttled by distinct pathways (carnitine-dependent and carnitine-independent) to mitochondria for further oxidation.

β-Oxidation of PA is in general restricted due to the presence of a methyl (-CH_3) group at the C3-position of the structure. Higher levels of plasma PA are observed in association with a type of hereditary disorder called Refsum's disease (RD) (Gloerich et al., 2006; Gloerich et al., 2007; Islam et al., 2015). Urine of RD patients contains 3-methyladipic acid and 3,6-dimethyloctanedioic acid, which are believed to be intermediates resulting from ω-oxidation of PA. In addition, a study conducted with a mouse model, in which the gene encoding for phytanoyl-CoA 2-hydroxylase was disrupted, revealed a possible pathway for ω-oxidation of PA (Wanders et al., 2011). In brief, ω-oxidation of PA produces phytane-1,16-dioic acid, later degraded by β-oxidation from the ω-end, resulting in different acyl-CoA esters and respective free fatty acids (upon thioesterase-mediated cleavage of the CoA esters) (Wanders et al., 2011) (**Figure 2**).

ANTIMICROBIAL AND CYTOTOXIC ACTIVITIES

PYT is one of the major constituents of plant derived essential oils (EO) and much research has been done to prove that the antimicrobial or cytotoxic activity observed for those EOs is related to their phytol content. The general antimicrobial potential of EOs is well-established and well-documented (Murbach Teles Andrade et al., 2014; Prabuseenivasan et al.,

2006), but the underlying mechanisms are still often insufficiently understood. It is hypothesized that the mechanism of the antimicrobial activity might be related to the ability of lipophilic molecules to cross cell membranes and exert their inhibitory activity on multiple targets simultaneously. PYT's antimicrobial activity was reported against *Escherichia coli* (growth inhibition MIC 62.5 μg/mL) (Ghaneian et al., 2015) and *Pseudomonas aeruginosa* (growth inhibition MIC 19 μg/mL) (Pejin et al., 2015). It is well-documented that proteins and nucleic acids are important bacterial macromolecular targets of antibacterial agents. Proteins play a variety of structural and functional roles, whereas nucleic acids carry mainly genetic information (Zhang et al., 2016). Accordingly, any alteration or damage to these cellular components might enhance growth inhibition or induce cell death. According to previous works on PYT and PYT-derivatives (Ghaneian et al., 2015; Inoue et al., 2005; Saikia et al., 2010; Singh et al., 2012) the action against *Mycobacterium tuberculosis* and *Staphylococcus aureus* appears to have no specific cellular target. In addition, research findings related to eight bacterial and eight fungal strains (Pejin et al., 2014b) suggested that PYT exhibits broad-spectrum of antimicrobial effects. Pejin and co-workers investigated the *in vitro* antibacterial activity of PYT against three species of infective endocarditis (IE) bacteria, namely, *Clostridium sporogenes*, *Sarcina lutea*, and *Enterococcus faecalis* (Pejin et al., 2014a). Results revealed that PYT displays significant activity against all tested bacteria, with *Enterococcus faecalis* being the most susceptible (growth inhibition MIC < 2 μg/mL) (Pejin et al., 2014a). In addition, pivaloyl, cinnamoyl, 3,4,5-trimethoxybenzoyl, 2,3-dichlorobenzoyl, and aldehyde derivatives of PYT exhibited drug resistance reversal activity against two *E. coli* strains, potentiating up to 16-fold the efficacy of tested antibiotics (Upadhyay et al., 2014).

PYT-containing EOs were postulated to dysregulate the function of eukaryotic cells through a number of effects including disruption of membrane permeability and depolarization of the mitochondrial membrane (Bakkali et al., 2008). Consequently, ionic Ca^{2+} cycling and channels for other ions become affected, together with proton pump and the ATP pool changes. Reduction of the pH gradient and changes in the fluidity of membranes are involved in the mode of action of EOs in general, with the membranes becoming abnormally permeable. Taken all together, these processes lead to leakage of radicals, cytochrome C, Ca^{2+}, and diverse proteins, similar to conditions involving oxidative stress and failure of the cell bioenergetics. Furthermore, permeabilization of outer and inner mitochondrial membranes essentially causes cell death by apoptosis and necrosis. Additionally, subsequent processes can have a prooxidative action, causing the late onset of both apoptosis and necrosis. Such effects might be underlying the activity of PYT against eukaryotic microorganisms such as *Candida albicans* and *Aspergillus niger* (Ghaneian et al., 2015).

The difference in electric potential between the interior and exterior cellular membrane is an important parameter associated with the cytotoxic activity. Treatments that alter the membrane potential (MP) can also change cellular functions. Agents, such as carbonyl cyanide-*m*-chlorophenylhydrazone (Zhang et al., 2016), that cause the MP of bacteria to approach or become zero, can lead to irregular cellular metabolism, and finally, cell death. EOs, particularly when applied at high concentrations, are known to readily cross the cell and mitochondrial membranes. In addition to membrane disruption and loss of essential cellular materials, EOs in general, often exert oxidative effects on the cells, which might result in damage to macromolecules such as proteins, lipids, and DNA (Faix et al., 2007). In a similar fashion, a concentration-dependent action of PYT was observed with respect to the cytotoxic activity

detected in *Schistosoma mansoni* (de Moraes et al., 2014), where PYT at concentrations of 50, 75 and 100 µg/mL produces more anti- *S. mansoni* activity than that of 25 µg/mL. In a study by da Silva and colleagues, it was shown that the PYT rich hexane fraction of the leaves of *Lacistema pubescens* exerts anti-promastigote activity at both the promastigote and amastigote forms of *Leishmania amazonensis* with IC_{50} values of 44.0 and 25.8 µg/mL, respectively (da Silva et al., 2015).

To date, there have been several studies on the cytotoxic activity of PYT, e.g., in *S. mansoni* (de Moraes et al., 2014) and lymphoid leukemia Molt 4B cells (Komiya et al., 1999). Research findings suggest that PA can disrupt redox homeostasis which results in astrogliosis in rat cerebellum. Additionally, production of reactive nitrogen species (RNS) is believed to contribute to the observed cerebellum alterations. Such a state is commonly observed in patients affected by RD and related phenomena, associated with the accumulation of PA (Borges et al., 2015). Similarly, PA at a concentration of 50 µM was shown to induce rotenone-like oxidative stress via a direct action on mitochondria (Schönfeld and Reiser, 2006). In addition, PA at a concentration of 10 µM causes cell death of neuroblastoma Neuro2a cells *via* activation of Hdac2 and Hdac3 (Nagai, 2015). Both of these studies suggested that the cytotoxic activity of PA is concentration dependent, and that different mechanisms contribute to the cell death. Moreover, PYT from *Citrullus lanatus* (watermelon) sprouts inhibited the growth of human T-cell leukemia (Jurkat) and counteracted tumor development in a xenograft model of human lung adenocarcinoma in mice (Itoh et al., 2018). PYT action was associated with S-phase cell cycle arrest and increased intracellular ROS production in the studied Jurkat cells. It also decreased the expression of cyclins A and D and the suppressed MAPK and PI3K/Akt signaling (Itoh et al., 2018). PYT also showed cytotoxic response with an IC_{50} value of 16.97 µM in human lung

carcinoma cells (A549) and caused characteristic apoptotic morphological changes and enhanced generation of ROS (Thakor et al., 2017). Furthermore, PYT caused activation of TRAIL, TNF-α receptors, and FAS, along with caspase-9 and -3 activation, and displayed a good binding affinity to glucose-6-phosphate dehydrogenase (G6PD), an enzyme which is known to promote tumor proliferation. Based on these findings, it was concluded that PYT has the potential as a drug candidate for counteracting lung carcinoma.

ANTIOXIDANT ACTION

Low concentrations of diverse EOs are well-known to possess antioxidant potentials (Bakkali et al., 2008; Islam et al., 2015). They are able to scavenge reactive oxygen and/or nitrogen species (ROS/RNS) produced by cellular stress and metabolism (Bakkali et al., 2008). Antioxidants act through three possible mechanisms: i) direct scavenging of reactive species via electron capturing and hydrogen radical transfer, ii) neutralization through redox reactions, and iii) oxidizing substrates. In this context, low concentrations of diverse EOs have proven to be beneficial, however, at high concentrations, the release of hydrogen radicals ($H^{\bullet}$) from active phenolic sites leaves phenoxy radicals ($PhO^{\bullet}$), which can eventually cause oxidation, thus leading to an antioxidant-induced oxidative effect. Therefore, phenolic antioxidants can also act as pro-oxidants causing cytogenotoxic effects. Several publications indicated that PYT has a potential for antioxidant activity (Pejin et al., 2014b; Santos et al., 2013). On the other hand, numerous studies revealed that EOs do not induce genetic mutations. They can, however, prevent the action of mutagens, by (i) inactivating them by direct scavenging, capturing radicals produced by mutagens, and (ii) activating enzymatic detoxification systems (Bakkali et al., 2008). Furthermore, antioxidants with prooxidative activity can increase the levels of

malondialdehyde (MDA), which is an important genotoxic agent (Bakkali et al., 2008). It is well known that ROS may cause DNA damage. One type of ROS is the hydroxyl radical ($^{\bullet}OH$), which has the potential to generate single strand breaks (SSB) in DNA (Sage and Harrison, 2011). Therefore, the PYT hydroxyl radical scavenging capacity may prevent such types of events of DNA damage (Pejin et al., 2014b).

The double bond in phytol is not a good candidate for acting as a free radical scavenger due to the absence of conjugation (what we usually see in case of our dietary conjugated double bond containing fatty acids). However, free radicals can be scavenged by the hydrogen of the alcohol group, and the double bond can help in producing resonance structure of the stable free radical (Islam et al., 2016a). There is one study to suggest that two different mechanisms are the base of getting twice the antioxidant effect of phytol in lipid medium than in aqueous medium in the lab experiments (Islam et al., 2016a). While in general saturated aliphatic alcohols are not good antioxidants, phytol exhibits a good antioxidant activity because of the allylic nature of the alcohol group. For the proposed one mechanism, there might be indeed some involvement of the double bond in the formation of an intermediate resonance structure where the oxygen can form a double bond with the adjacent carbon while the double bond shifts to 3,4 carbon (Islam et al., 2016a).

PA is a carboxylic acid, and therefore it may undergo oxidative decarboxylation. Enzymes involved in such reactions are known as decarboxylases. More explicitly, oxidative decarboxylations are reactions in which a carboxylate group is removed, resulting in the formation of carbon dioxide. These reactions are often found in biological systems, for example as a part of the citric acid cycle. Thus far, two variations of the free radical reactions of carboxylic acids ($RO_2^{\bullet} + R_iCOOH \rightarrow ROOH + R_iCO_2^{\bullet}$ and $RO_i^{\bullet} + R_jCOOH \rightarrow ROOH + R_i^{\bullet} +$

CO_2) have been analyzed theoretically (Denisov and Shestakov, 2013). The cytotoxic activity of PA may be linked to overproduction of free radicals under specific conditions. In this context, PA concentration between 1 and 500 μM was shown to cause oxidative damage of cerebellum and cerebral cortex of Wistar male rats (Leipnitz et al., 2010).

Some of the bioactivities of PYT which could be related to its redox properties are its reported *in vitro* anticancer effects (Chikati, 2013; Guo et al., 2014; Hibasami et al., 2002; Kim et al., 2015; Komiya et al., 1999), anti-teratogenic activity (Arnhold et al., 2002), tumor-promotor (Kagoura et al., 1999) and anti-tumor activity (Líška et al., 2011). Depicted in **Figure 3** are proposed PYT-mediated antioxidant mechanisms (Islam et al., 2016b).

INDUCTION OF APOPTOSIS AND PROTECTIVE AUTOPHAGY

PYT can induce both apoptosis and protective autophagy. However, the molecular mechanisms of the cytotoxic effects of PYT in cancer cells remain poorly understood. Song and coworkers have shown that PYT stimulates both apoptosis and autophagy in human gastric adenocarcinoma AGS cells. PYT, when incubated with cells, induced accumulation of acidic vesicle, and downregulated protein kinase B (Akt), mTOR (mechanistic target of rapamycin), and p70S6K phosphorylation. Notably, pre-incubation with chloroquine, a lysosomal inhibitor, increased PYT-stimulated apoptosis in AGS cells, which suggests that PYT might stimulate a protective autophagy. In addition, co-incubation with a ROS scavenger increased the PYT-stimulated cytotoxicity. These results may provide the foundation of novel insights into possible treatments of cancer using PYT (Song and Cho, 2015). Shown in **Figure 4** are apoptosis- and autophagy-modulating effects of PYT.

In a similar fashion, Thakor and coworkers also have demonstrated that PYT, isolated from *Abutilon indicum* leaves, decreases the viability (42%, applied at 0.37 µM) and showed different phases of ROS-mediated programmed cell death in AO-EtBr stained *Schizosaccharomyces pombe* (fission yeast) cells. These researchers concluded that this investigation could be extended to an *in vitro* cancer study using different cell lines, and could open the door to find the exact mechanism of action of phytol as a potential candidate for the treatment of malignant neoplastic disease (Thakor et al., 2016).

ANXIOLYTIC AND ANTICONVULSANT EFFECTS

Anxiety is a kind of mental condition which often results from a trauma. In some cases, anxiety can be accompanied by a depressive mood (Hunt et al., 2003; Vieweg et al., 2006). In general, anxiety disorders are a group of mental conditions which can be triggered by genetic predispositions, drugs, and withdrawal from certain drugs. Medications and change of lifestyle are common approaches in the treatment of anxiety. Benzodiazepines (BDZ) are the first-line of drugs that have been prescribed for patients with anxiety for over four decades (Starcevic, 2014). Diazepam, the most commonly prescribed BDZ used in the treatment of anxiety, is a positive allosteric modulator of γ-aminobutyric acid type A receptors ($GABA_A$). $GABA_A$ receptor is a ligand-gated chloride-selective ion channel that is activated by GABA, an inhibitory neurotransmitter in the brain (Griffin et al., 2013). Binding of BDZ to this receptor complex potentiates the binding of GABA, which ultimately enhances the total movement of chloride ions (Cl^-) across the neuronal cell membrane. This enhanced Cl^--influx hyperpolarizes the neuron's membrane and reduces the chance of action potential to occur (inhibition of neurotransmission). $GABA_A$ receptors containing the α1 subunit mediate the sedative, and in part the anticonvulsive effects of diazepam, whereas the anxiolytic action of diazepam is mediated by the $GABA_A$

receptors containing α2 subunit (Tan et al., 2014). Phytol is recognized for its wide range of pharmacological effects on the nervous system, including anxiolytic and antidepressant (Pereira Costa et al., 2014). Common neuropsychological disorders, such as anxiety, epilepsy and depression are related and coexist in some patients. The lifetime prevalence of epilepsy in association with depression was approximated to be as high as 55% (Jackson and Turkington, 2005). Yet remarkably lesser information exists on the mechanism of depression and anxiety in convulsion and its treatments.

In this context, the anticonvulsant effect of PYT has been also demonstrated (Costa et al., 2012). Recent work conducted by Costa et al. on *Swiss* albino mice (Costa et al., 2014) has found that PYT (25, 50, or 75 mg/kg by intraperitoneal injection, i.p.) also displays anti-anxiety activity. By inducing anxiolytic effects, BDZ drugs such as diazepam increase inhibitory processes in the cerebral cortex (Zakusov et al., 1977). Hoffmann-La Roche has first introduced another drug, flumazenil, a $GABA_A$ receptor antagonist, in 1987. It was primarily used to treat BDZ overdoses and to help reverse anesthesia (Rye et al., 2012). Bang and colleagues (Bang et al., 2002) suggested that PYT is able to bring the elevated levels of neurotransmitter GABA to optimal levels in the central nervous system. As previously mentioned, diazepam exerts both of its anxiolytic and anticonvulsant effects through $GABA_A$ receptor subunits α1 and α2, respectively. On the other hand, the antagonizing influence of flumazenil against both types of PYT effects, show that both flumazenil and PYT may exert their CNS effects by targeting the α1 and α2 $GABA_A$ receptor subunits (Costa et al., 2014; Costa et al., 2012). However, further research is needed to gain deeper understanding about the anxiolytic and anticonvulsant activities of PYT.

IMMUNE-MODULATING PROPERTIES

Several recent studies have suggested that some PYT-derivatives (phytanol, phytanyl amine, and phytanyl mannose) exert immune-stimulating activity by induction of the expression of a range of chemokines and cytokines (Aachoui et al., 2011a; Roy Chowdhury et al., 2013) and modulation of immune responses through apoptotic/necrotic (Aachoui et al., 2011b) effects on target tumor cells.

The immune system relies on production of chemically reactive molecules such as reactive oxygen and reactive nitrogen species (ROS/RNS), along with phagocytosis; these reactive intermediates can damage DNA and other molecules, and facilitates the neutralization of pathogens (Nathan and Shiloh, 2000). However, this is an example of short-term, rather than long-term oxidative stress, since the later causes cytotoxicity and genotoxicity, which may damage or kill the phagocytes as well as other host cells (Rice-Evans and Gopinathan, 1995). Nakanishi et al. (2016) have recently observed that PA, PYT, and pristanic acid (3, 10, and 30 μM) cause substantial reduction of interferon-gamma (IFN-γ), interleukin (IL)-4, and -10 production in mouse splenocytes stimulated by T-cell mitogens (Nakanishi et al., 2016). Additionally, PA and PYT inhibit production of IL-17A production, which overall suggests a potentially beneficial effect for the amelioration of T-cell mediated autoimmune diseases.

ANTINOCICEPTIVE AND ANTI-INFLAMMATORY ACTIVITY OF PYT

Nociception is defined as the process of encoding and processing of noxious stimuli by the nervous system (Loeser and Treede, 2008). Substances capable of modulating the responsible signaling pathways can be employed to control the intensity of pain which can be a vital signal for inflammation. Inflammation is the complex biological response of vascularized tissues to

harmful stimuli (e.g., pathogens, damaged cells, or irritants), involving the local formation of kinins and cytokines, that promote activation of vascular endothelial cells, followed by migration of leukocytes into the inflamed site. Production of oxidative mediators is another response to inflammation (Medzhitov, 2008; Silva et al., 2014). In this context, pain is an indispensable signal for inflammation. Degree of inflammation is a very important factor since minor localized and tightly controlled inflammation is usually beneficial for defensive purposes. However, chronic inflammation may lead to diverse diseases, such as atherosclerosis, rheumatoid arthritis, pain, and cancer, among others. Therefore, anti-inflammatory agents may be considered as preventive or therapeutic depending on the context.

The antinociceptive (Santos et al., 2013) and anti-inflammatory (Silva et al., 2014) activities of PYT may be interrelated. Recent data from *Swiss* mice treated with PYT (i.p.) showed a potent dose-dependent (applied at 25, 50, 100, and 200 mg/kg) antinociceptive response in hot-plate or chemical-induced (acetic acid, formaldehyde) nociceptive tests when compared to those treated with 10 mg/kg of the reference compounds morphine and indomethacin (i.p.). Taken together, these results demonstrate that PYT could be a potential therapeutic approach in different contexts. Additionally, anti-inflammatory tests performed by Silva et al.(2014) indicated that PYT attenuates the inflammatory response by inhibiting neutrophil migration, which is partly caused by reduction in tumor necrosis factor-alpha (TNF-α) and IL-1β levels (Silva et al., 2014). These findings suggest that the antioxidant, anti-inflammatory, and antinociception activities of PYT may be connected to each other. Additionally, these results show that PYT could be a good candidate to be further evaluated for other relevant bioactivities, such as for antipyretic, antiperiodontitis, and anti-arthritic activities.

PYT IN METABOLIC DISORDERS AND EFFECTS RELATED TO PPARs

Peroxisome proliferator-activated receptors (PPARs) are a subfamily of the nuclear receptor superfamily involved in a multitude of cellular mechanisms (Dreyer et al., 1992). PPARs can be further classified as α, β/δ, or γ, the latter being further divided into γ1, γ2, and γ3. PPARα is expressed in liver, kidney, heart, muscle, and adipose tissues (Tyagi et al., 2011), whereas PPARβ/δ is found in many tissues, specifically the brain, adipose tissues, and skin. The γ1 subunits are expressed in virtually all tissues, including the heart, muscle, colon, kidney, pancreas, and spleen, whereas the γ3 subunits are found in macrophages, the large intestine, and white adipose tissue. Finally, γ2 is mainly expressed in adipose tissue. These groups of nuclear receptor proteins are essential for the regulation of cellular differentiation, development, nutrient metabolism, and tumorigenesis (Belfiore et al., 2009) in higher organisms (Feige et al., 2006).

Activity of PPARs depends on the precise shape of the ligand-binding domain (LBD), as well as on the number and type of coactivator and the presence of corepressor proteins (Yu and Reddy, 2007). Free fatty acids and eicosanoids are believed to be endogenous ligands of the PPARs. It is important to emphasize that eicosanoid PPAR ligands are important regulators of inflammatory responses, and that overall PPAR signaling is known to modulate inflammation. In this context, there are experimental data suggesting a link between PPAR modulation and the observed anti-inflammatory activity of PYT (Silva et al., 2014).

PPARγ is an important regulator of genes that affect insulin action. A tumor necrosis factor-alpha (TNF-α), a pro-inflammatory cytokine, which is associated with insulin resistance and decreased insulin signal transduction (Berger and Moller, 2002). Therefore, the TNF-α inhibitory activity observed by Silva and coworkers may indicate not only anti-inflammatory but also a potential anti-diabetic activity of PYT. Moreover, there are recognized PPARγ ligands,

such as rosiglitazone, that exert anti-inflammatory effects by inhibiting the activation of the NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells) pathway (Zhang et al., 2016). NF-κB, a protein complex that controls DNA transcription, cytokine production, and cell survival, is found in almost all eukaryotic cell types (Brasier, 2006; Gilmore, 2006; Perkins, 2007). It is involved in cellular responses to a number of stimuli, such as stress, cytokines, free radicals, ultraviolet irradiation, oxidized low-density lipoprotein (LDL), and pathogens. It additionally plays a key role in regulating the immune response. Abnormality in its regulation has been linked to cancer, inflammatory and autoimmune diseases, viral infection, and improper immune development (Levenson et al., 2004; Perkins, 2007). In several studies, PPARγ expression has also been implied in a variety of cancer cells, including human mammary adenocarcinomas and colon cancer cells (Berger and Moller, 2002). Thus, the anticancer and immune adjuvant activities of PYT and its derivatives may be linked to potential effects on both PPARs and NF-κB. In this regard, there has also been reported evidence of PYT acting as an anti-scratching compound when applied in 5 and 10 mg/kg (dissolved in 5% cremophor; p.o.) in male ICR and BALB/*c* mice by inhibiting allergic cytokines expression through regulation of NF-κB and activator protein (AP)-1 (Ryu et al., 2011).

Research by Matsuda et al. (2013) showed that PYT, applied as PPARα ligand, increases blood NAD level by suppressing hepatic alpha-amino-beta-carboxymuconate-epsilon-semialdehyde decarboxylase (ACMSD) mRNA expression in the rat´s liver (Matsuda et al., 2013). In addition, a PPARα-mediated activity of PYT has been investigated by a number of other research groups. Mackie et al. (2009) showed that PYT (0.5 and 1.0%; p.o.) causes consistent PPARα-mediated responses such as lower body weight, higher liver weights, hepatocellular hypertrophy, peroxisome proliferation, and enhanced catalase expression in mice.

PYT additionally causes female mice-specific hepatotoxicity, which was explained by the lower hepatic expression of sterol carrier protein-x (SCP-x), linked to a lower capacity to oxidize PA. Based on the preceding discussion, it is evident that PPARα can play an important role in the induction of hepatomegaly and hepatocarcinogenesis in rodents, as well as in the expression of the fatty acid transport protein (FATP) (Berger and Moller, 2002). Thus, the hepatotoxic activity of PYT may be PPARα-dependent. However, how exactly the activity of PTY is associated with hepatotoxicity remains still not entirely clear and more research is needed to address the mechanisms underlying this bioactivity.

Hypertension is a disorder of the cardiovascular system that is often linked to insulin resistance and obesity. People with type-2-diabetes (T2D) are 1.5 to 2-fold more likely to develop hypertension than the rest of the population (Berger and Moller, 2002). PYT (250 mg/kg) showed modulatory effects in the RXR ligand and PPARγ to diabetic insulin-resistant rats. PYT docking on the RXRα/PPARγ heterodimer revealed that it has a higher binding affinity and a lesser energy score on RXRα under diabetic conditions (Elmazar et al., 2013). These findings suggest that the insulin sensitizing/anti-diabetic effect of PYT is mediated partly by activation of nuclear receptors and heterodimerization of RXR with PPARγ by phytanic acid. In addition, since PPAR responses and hypertension are related to T2D, it might be of interest to evaluate PYT as a potential antihypertensive agent. On the other hand, T2D is often associated with weight gain, high blood pressure, and increased pro-inflammatory cytokines, which are all linked to insulin resistance. In the context of metabolic disorders, research findings showed that PYT has cholesterol-lowering (Olofsson et al., 2010) potentials in mice. Furthermore, atherosclerosis has a strong association with elevated LDL-cholesterol as well as with diabetes,

therefore, PYT could be considered as a promising candidate to be further examined for possible anti-atherosclerotic action.

Adipogenesis is a key process regulating metabolic health, and PPARγ is a key regulator of adipogenesis. Wang and coworkers demonstrated that PYT administration increases adipocyte number in inguinal subcutaneous white adipose tissue and improves glucose tolerance in mice fed with high-fat and high fructose diet. This was consistent with the enhanced adipogenesis and glucose uptake in 3T3-L1 cell line, and was associated with activation of phosphatidylinositol-4,5-bisphosphate 3-kinase (PI3K)/Akt signaling pathway (Wang et al., 2017). In addition, PYT administration in mice caused increased expression of marker genes associated with adipogenesis (C/EBPα and PPARγ) in inguinal subcutaneous white adipose tissue. These data provided new insights into the regulation of adipogenesis and glucose uptake by dietary PYT and suggested the application of PYT as a potential nutritional agent to combat obesity and type 2 diabetes (Wang et al., 2017). Shown in **Table 1 and 2** is a summary of PYT bioactivity studies.

In human glioblastoma cell lines (U87MG, A172, and T98G), PYT and retinol showed dose-dependent cytotoxicity and downregulated genes involved in cholesterol or fatty acid biosynthetic pathways (Facchini et al., 2018). In particular, PYT suppressed the expression levels of sterol regulatory element-binding protein 1 (SREBP-1) together with its downstream target protein fatty acid synthase (FAS). PYT was also associated with suppression of farnesyl-diphosphate farnesyltransferase (FDFT1) which regulates cholesterol synthesis (Facchini et al., 2018). These observations suggest that PYT may have the potential to be used as an agent to regulate cholesterol biosynthesis, and thus may have possible therapeutic applications in preventing or treating hyperlipidemia conditions, as an approach to combat cardiac diseases. PYT was also shown to regulate adipocytes phenotype, which might have implications for

counteracting obesity and related cardiovascular and metabolic disorders. In this line, PYT administration suppressed body weight gain and inguinal subcutaneous white adipose tissue index, whereby stimulating the expression of brown adipocyte marker genes, including UCP1, PGC1α, PRDM16, PDH, and Cytochrome C (Zhang et al., 2018).

In addition, PYT activated the AMPKα signaling in mice and stimulated brown-adipogenic differentiation of 3T3-L1 cells by increasing the mitochondria content and stimulating oxygen consumption (Zhang et al., 2018). Obesity-regulating effects of PYT was further studied by An et al. (2018). These authors observed that a PYT-enriched diet may increase PA levels in the liver and brown adipocytes. In this way, dietary PYT may activate PPARα in liver, resulting in amelioration of obesity-induced metabolic diseases. In a similar fashion, Wang et al. (2018) reported that PYT can regulate hyperplasia/adipogenesis and glucose homeostasis in high-fat and high fructose diet (HFFD) in mice. These researchers found that PYT decreases body weight gain and inguinal subcutaneous white adipose tissue weight. Moreover, PYT improved the OGTT glucose tolerance in mice and activated marker genes associated with adipogenesis (C/EBPα and PPARγ) and glucose uptake (GLUT4 and AS160), and activated the PI3K/Akt signal transduction pathway (Wang et al., 2018). On the other hand, Mezzar and colleagues have recently investigated phytol-induced pathology in 2-hydroxyacyl-CoA lyase (HACL1) deficient mice (Mezzar et al., 2017). HACL1 is a major enzyme of the peroxisomal α-oxidation of PA. Results showed that accumulation of the PYT metabolite PA in liver tissues and serum in the KO mice was associated with a significant weight loss, absence of abdominal white adipose tissue, anatomical changes of the liver, and decreased hepatic glycogen and triglyceride content (Mezzar et al., 2017).

RECENT MISCELLANEOUS APPLICATIONS

PYT and its derivatives have also shown other interesting bioactivities. Apart from the pharmacological effects in disease management, PYT was reported to display a potential in counteracting hyperpigmentation, with a possible application as a skin whitening agent in cosmetics (Ko and Cho, 2018). Ko and Cho have recently employed B16F10 murine melanoma cells, to demonstrate that PYT can inhibit α-melanocyte-stimulating hormone-induced melanogenesis, in association with suppressed expression of tyrosinase and tyrosinase-related protein 1 (Ko and Cho, 2018).

Bioavailability of PYT could represent a barrier for its potential therapeutic applications, and the use of nanocarriers-mediated delivery systems might be a possible solution. Sathya et al. (2017) recently showed that PYT loaded on poly(lactic-co-glycolic acid) (PLGA) nanoparticles may enhance cellular uptake of the compound and thus might result in higher effectiveness when tested *in vitro* in a cellular model with a relevance for Alzheimer's disease. These authors also demonstrated that PYT is released *in vitro* in a sustained manner, and that treatment with PYT-loaded nanoparticles led to disruption of amyloid aggregates and resulted in anti-cholinesterase and anti-oxidative action, without signs of toxicity in Neuro2a cells (Sathya et al., 2018). In another work, a nanoemulsion of PYT was prepared and delivered in *Artemia salina* and *Allium cepa* for toxicity, cytotoxicity, and genotoxicity analysis (Islam et al., 2017). Results revealed that PYT-nanoemulsion has enhanced effects as compared to PYT, and that the studied PYT-nanoemulsion formulation has an improved bioavailability (Islam et al., 2017).

Finally, a very interesting aspect associated with PYT application is related to interaction with gut microbiota, which plays important roles in diverse metabolic processes and human diseases such as obesity, diabetes, fatty liver disease, and cardiovascular diseases. In this context,

food constituents such as PYT might have a variety of health and disease-related effects that might be associated with effect on the human gut microbiota (Roca-Saavedra et al., 2018). PA is, for example, known to get oxidized by the rumenal microbiota of certain marine organisms (Roca-Saavedra et al., 2017).

CONCLUSIONS

Phytol and its derivatives exhibit a wide range of bioactivities including antianxiety, cytotoxic, metabolism-modulating, antioxidant, autophagy- and apoptosis-inducing, anti-nociceptive, anti-inflammatory, immune-modulating, and antimicrobial effects. Abundance of PYT in nature and its diverse bioactivities make it a commercially important compound. Although PPARs- and NF-κB-mediated activities have been pointed out as responsible for some of the bioactivities of PYT, further studies are needed to address more thoroughly the molecular mechanisms of action of PYT and its derivatives.

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AUTHOR CONTRIBUTIONS

MTI, ESA, JAS, and AGA have written the first draft of the manuscript. All other authors have revised and improved the first draft. All authors have seen and agreed on the finally submitted version of the manuscript.

CONFLICTS OF INTEREST

Authors declare no conflict of interest.

REFERENCES

Aachoui, Y., Chowdhury, R.R., Fitch, R.W., Ghosh, S.K., 2011a. Molecular signatures of phytol-derived immunostimulants in the context of chemokine–cytokine microenvironment and enhanced immune response. Cellular Immunology 271, 227-238.

Aachoui, Y., Schulte, M.L., Fitch, R.W., Ghosh, S.K., 2011b. Synthetic adjuvants for vaccine formulations: Evaluation of new phytol derivatives in induction and persistence of specific immune response. Cellular Immunology 271, 308-318.

Arnhold, T., Elmazar, M.M.A., Nau, H., 2002. Prevention of Vitamin A Teratogenesis by Phytol or Phytanic Acid Results from Reduced Metabolism of Retinol to the Teratogenic Metabolite, All-trans-retinoic Acid. Toxicological Sciences 66, 274-282.

Bakkali, F., Averbeck, S., Averbeck, D., Idaomar, M., 2008. Biological effects of essential oils – A review. Food and Chemical Toxicology 46, 446-475.

Bang, M., Choi, S., Jang, T., Kim, S., Kwon, O., Kang, T., 2002. Phytol, SSADH inhibitory diterpenoid of Lactuca sativa. Arch Pharm Res. 25, 643-646.

Belfiore, A., Genua, M., Malaguarnera, R., 2009. PPAR-γ Agonists and Their Effects on IGF-I Receptor Signaling: Implications for Cancer. PPAR Research 2009, 830501.

Berger, J., Moller, D., 2002. The Mechanisms of Action of PPARs. Annual Review of Medicine 53, 409-435.

Borges, C.G., Canani, C.R., Fernandes, C.G., Zanatta, Â., Seminotti, B., Ribeiro, C.A.J., Leipnitz, G., Vargas, C.R., Wajner, M., 2015. Reactive nitrogen species mediate oxidative stress and astrogliosis provoked by in vivo administration of phytanic acid in cerebellum of adolescent rats: A potential contributing pathomechanism of cerebellar injury in peroxisomal disorders. Neuroscience 304, 122-132.

Brasier, A.R., 2006. The NF-kappaB regulatory network. Cardiovascular toxicology 6, 111-130.

Chikati, R., 2013. Molecular Evaluation of RNase from Aspergillus niger and Phytol of Nymphaea pubescens as Cytoskeletol targeting elements in Cancer. Epidemiology, (Aug:21-23); Holiday Inn, Ornaldo-Fl, USA.

Costa, J.P., de Oliveira, G.A.L., de Almeida, A.A.C., Islam, M.T., de Sousa, D.P., de Freitas, R.M., 2014. Anxiolytic-like effects of phytol: Possible involvement of GABAergic transmission. Brain Research 1547, 34-42.

Costa, J.P., Ferreira, P.B., De Sousa, D.P., Jordan, J., Freitas, R.M., 2012. Anticonvulsant effect of phytol in a pilocarpine model in mice. Neuroscience Letters 523, 115-118.

da Silva, J.M., Antinarelli, L.M.R., Ribeiro, A., Coimbra, E.S., Scio, E., 2015. The effect of the phytol-rich fraction from Lacistema pubescens against Leishmania amazonensis is mediated by mitochondrial dysfunction. Experimental Parasitology 159, 143-150.

de Moraes, J., de Oliveira, R.N., Costa, J.P., Junior, A.L.G., de Sousa, D.P., Freitas, R.M., Allegretti, S.M., Pinto, P.L.S., 2014. Phytol, a Diterpene Alcohol from Chlorophyll, as a Drug against Neglected Tropical Disease Schistosomiasis Mansoni. PLOS Neglected Tropical Diseases 8, e2617.

de Souza, N.J., Nes, W.R., 1969. The presence of phytol in brown and blue-green algae and its relationship to evolution. Phytochemistry 8, 819-822.

Denisov, E.T., Shestakov, A.F., 2013. Free-radical decarboxylation of carboxylic acids as a concerted abstraction and fragmentation reaction. Kinetics and Catalysis 54, 22-33.

Dreyer, C., Krey, G., Keller, H., Givel, F., Helftenbein, G., Wahli, W., 1992. Control of the peroxisomal β-oxidation pathway by a novel family of nuclear hormone receptors. Cell 68, 879-887.

Elmazar, M.M., El-Abhar, H.S., Schaalan, M.F., Farag, N.A., 2013. Phytol/Phytanic Acid and Insulin Resistance: Potential Role of Phytanic Acid Proven by Docking Simulation and Modulation of Biochemical Alterations. PLOS ONE 8, e45638.

Faix, Š., Juhas, Š., Faixová, Z., 2007. The Effect of Essential Oil Intake on Changes of Plasma Antioxidant Status in Mice. Acta Vet. Brno 76, 357-361.

Feige, J.N., Gelman, L., Michalik, L., Desvergne, B., Wahli, W., 2006. From molecular action to physiological outputs: Peroxisome proliferator-activated receptors are nuclear receptors at the crossroads of key cellular functions. Progress in Lipid Research 45, 120-159.

Ghaneian, M.T., Ehrampoush, M.H., Jebali, A., Hekmatimoghaddam, S., Mahmoudi, M., 2015. Antimicrobial activity, toxicity and stability of phytol as a novel

surface disinfectant. Environmental Health Engineering and Management Journal 2, 13-16.

Gilmore, T.D., 2006. Introduction to NF-kappaB: players, pathways, perspectives. Oncogene 25, 6680-6684.

Gloerich, J., Ruiter, J.P.N., van den Brink, D.M., Ofman, R., Ferdinandusse, S., Wanders, R.J.A., 2006. Peroxisomal trans-2-enoyl-CoA reductase is involved in phytol degradation. FEBS Letters 580, 2092-2096.

Gloerich, J., van den Brink, D.M., Ruiter, J.P., van Vlies, N., Vaz, F.M., Wanders, R.J., Ferdinandusse, S., 2007. Metabolism of phytol to phytanic acid in the mouse, and the role of PPARalpha in its regulation. Journal of lipid research 48, 77-85.

Griffin, C.E., 3rd, Kaye, A.M., Bueno, F.R., Kaye, A.D., 2013. Benzodiazepine pharmacology and central nervous system-mediated effects. Ochsner J 13, 214-223.

Guo, J., Yuan, Y., Lu, D., Du, B., Xiong, L., Shi, J., Yang, L., Liu, W., Yuan, X., Zhang, G., Wang, F., 2014. Two natural products, trans-phytol and (22E)-ergosta-6,9,22-triene-3β,5α,8α-triol, inhibit the biosynthesis of estrogen in human ovarian granulosa cells by aromatase (CYP19). Toxicology and Applied Pharmacology 279, 23-32.

Hibasami, H., Kyohkon, M., Owaki, S., Katsuzaki, H., Imai, K., Ohnishi, K., 2002. Diol- and triol-types of phytol induce apoptosis in lymphoid leukemia Molt 4B cells. International Journal of Molecular Medicine 10, 555-559.

Hunt, M., Auriemma, J., Cashaw, A.C.A., 2003. Self-Report Bias and Underreporting of Depression on the BDI-II. Journal of Personality Assessment 80, 26-30.

Inoue, Y., Hada, T., Shiraishi, A., Hirose, K., Hamashima, H., Kobayashi, S., 2005. Biphasic Effects of Geranylgeraniol, Teprenone, and Phytol on the Growth of Staphylococcus aureus. Antimicrobial Agents and Chemotherapy 49, 1770-1774.

Ischebeck, T., Zbierzak, A.M., Kanwischer, M., Dörmann, P., 2006. A Salvage Pathway for Phytol Metabolism in Arabidopsis. Journal of Biological Chemistry 281, 2470-2477.

Islam, M.T., de Alencar, M.V.O.B., da Conceição Machado, K., da Conceição Machado, K., de Carvalho Melo-Cavalcante, A.A., de Sousa, D.P., de Freitas, R.M., 2015. Phytol in a pharma-medico-stance. Chemico-Biological Interactions 240, 60-73.

Islam, M.T., Streck, L., Correia Jardim Paz, M.F., de Castro e Sousa, J.M., Alencar, M.V.O.B.d., Oliveira Ferreira da Mata, A.M., Melo de Carvalho, R., de Oliveira Santos, J.V., da Silva-Junior, A.A., Pinheiro Ferreira, P.M., de Carvalho Melo-Cavalcante, A.A., 2016a. Preparation of Phytol-Loaded Nanoemulsion and Screening for Antioxidant Capacity. International Archives of Medicine 9.

Islam, M.T., Streck, L., Paz, M.F.C.J., e Sousa, J.M.d.C., de Alencar, M.V.O.B., Maria, A., da Mata, O.F., da Silva, M.B.S., de Carvalho, R.M., 2016b. Preparation of phytol-loaded nanoemulsion and screening for antioxidant capacity: data-in-brief. International Archives Of Medicine 9.

Jackson, M.J., Turkington, D., 2005. Depression and anxiety in epilepsy. J Neurol Neurosurg Psychiatry 76 Suppl 1, i45-47.

Kagoura, M., Matsui, C., Morohashi, M., 1999. Phytol Is a Novel Tumor Promoter on ICR Mouse Skin. Japanese Journal of Cancer Research 90, 377-384.

Kim, C.-W., Lee, H.J., Jung, J.H., Kim, Y.H., Jung, D.-B., Sohn, E.J., Lee, J.H., Woo, H.J., Baek, N.-I., Kim, Y.C., Kim, S.-H., 2015. Activation of Caspase-9/3 and Inhibition of Epithelial Mesenchymal Transition are Critically Involved in Antitumor Effect of Phytol in Hepatocellular Carcinoma Cells. Phytotherapy Research 29, 1026-1031.

Komiya, T., Kyohkon, M., Ohwaki, S., Eto, J., Katsuzaki, H., Imai, K., 1999. Phytol induces programmed cell death in human lymphoid leukemia Molt 4B cells. International Journal of Molecular Medicine 4, 377-380.

Leipnitz, G., Amaral, A.U., Zanatta, Â., Seminotti, B., Fernandes, C.G., Knebel, L.A., Vargas, C.R., Wajner, M., 2010. Neurochemical evidence that phytanic acid induces oxidative damage and reduces the antioxidant defenses in cerebellum and cerebral cortex of rats. Life Sciences 87, 275-280.

Levenson, J.M., Choi, S., Lee, S.-Y., Cao, Y.A., Ahn, H.J., Worley, K.C., Pizzi, M., Liou, H.-C., Sweatt, J.D., 2004. A Bioinformatics Analysis of Memory Consolidation Reveals Involvement of the Transcription Factor c-Rel. The Journal of Neuroscience 24, 3933.

Líška, J., Ondková, S., Macejová, D., Brtko, J., 2011. Histological evaluation of rat mammary tumours after treatment with retinoic acid analogues — phytol, TTNPB and vitamin D3 analogue seocalcitol. Biologia 66, 365-369.

Loeser, J.D., Treede, R.-D., 2008. The Kyoto protocol of IASP Basic Pain Terminology☆. PAIN 137, 473-477.

Mackie, J.T., Atshaves, B.P., Payne, H.R., McIntosh, A.L., Schroeder, F., Kier, A.B., 2009. Phytol induced hepatotoxicity in mice. Toxicologic pathology 37, 201-208.

Matsuda, H., Gomi, R.-t., Hirai, S., Egashira, Y., 2013. Effect of Dietary Phytol on the Expression of α-Amino-β-carboxymuconate-ε-semialdehyde Decarboxylase, a Key Enzyme of Tryptophan-niacin Metabolism, in Rats. Bioscience, Biotechnology, and Biochemistry 77, 1416-1419.

Medzhitov, R., 2008. Origin and physiological roles of inflammation. Nature 454, 428-435.

Murbach Teles Andrade, B.F., Nunes Barbosa, L., da Silva Probst, I., Fernandes Júnior, A., 2014. Antimicrobial activity of essential oils. Journal of Essential Oil Research 26, 34-40.

Nagai, K., 2015. Phytanic acid induces Neuro2a cell death via histone deacetylase activation and mitochondrial dysfunction. Neurotoxicology and Teratology 48, 33-39.

Nakanishi, T., Anraku, M., Suzuki, R., Kono, T., Erickson, L., Kawahara, S., 2016. Novel immunomodulatory effects of phytanic acid and its related substances in mice. Journal of Functional Foods 21, 283-289.

Nathan, C., Shiloh, M., 2000. Reactive oxygen and nitrogen intermediates in the relationship between mammalian hosts and microbial pathogens. Proc Natl Acad Sci. USA 97, 8841-8848.

Olofsson, P., Hultqvist, M., Holmdahl, R., 2010. Phytol as a cholesterol lowering agent. US Patents, 2011,0015,278,.

Pejin, B., Ciric, A., Glamoclija, J., Nikolic, M., Sokovic, M., 2015. In vitro anti-quorum sensing activity of phytol. Natural Product Research 29, 374-377.

Pejin, B., Kartali, T., Stanimirovic, B., Karaman, M., 2014a. Phytol May Inspire New Medicinal Foods for the Treatment of Heart Disease. Asian. J. Chem. 26, 8261-8262.

Pejin, B., Savic, A., Sokovic, M., Glamoclija, J., Ciric, A., Nikolic, M., Radotic, K., Mojovic, M., 2014b. Further in vitro evaluation of antiradical and antimicrobial activities of phytol. Natural Product Research 28, 372-376.

Pereira Costa, J., da Silva Oliveira, J., Mario Rezende Junior, L., de Freitas, R.M., 2014. Phytol a Natural Diterpenoid with Pharmacological Applications on Central Nervous System: A Review. Recent Pat Biotechnol 8, 194-205.

Perkins, N.D., 2007. Integrating cell-signalling pathways with NF-[kappa]B and IKK function. Nat Rev Mol Cell Biol 8, 49-62.

Prabuseenivasan, S., Jayakumar, M., Ignacimuthu, S., 2006. In vitro antibacterial activity of some plant essential oils. BMC Complementary and Alternative Medicine 6, 39-39.

Proteau, P.J., 1998. Biosynthesis of Phytol in the Cyanobacterium Synechocystis sp. UTEX 2470: Utilization of the Non-Mevalonate Pathway. Journal of Natural Products 61, 841-843.

Rice-Evans, C., Gopinathan, V., 1995. Oxygen toxicity, free radicals and antioxidants in human disease: biochemical implications in atherosclerosis and the problems of premature neonates. Essays Biochem. 29, 39-63.

Rontani, J.-F., Volkman, J.K., 2003. Phytol degradation products as biogeochemical tracers in aquatic environments. Organic Geochemistry 34, 1-35.

Roy Chowdhury, R., Fitch, R.W., Ghosh, S.K., 2013. Efficacy of phytol-derived diterpenoid immunoadjuvants over alum in shaping the murine host's immune response to Staphylococcus aureus. Vaccine 31, 1178-1186.

Rye, D.B., Bliwise, D.L., Parker, K., Trotti, L.M., Saini, P., Fairley, J., Freeman, A., Garcia, P.S., Owens, M.J., Ritchie, J.C., Jenkins, A., 2012. Modulation of Vigilance in the Primary Hypersomnias by Endogenous Enhancement of GABA_A Receptors. Science Translational Medicine 4, 161ra151.

Ryu, K.-R., Choi, J.-Y., Chung, S., Kim, D.-H., 2011. Anti-scratching Behavioral Effect of the Essential Oil and Phytol Isolated from Artemisia princeps Pamp. in Mice. Planta Medica 77, 22-26.

Sage, E., Harrison, L., 2011. Clustered DNA lesion repair in eukaryotes: Relevance to mutagenesis and cell survival. Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis 711, 123-133.

Saikia, D., Parihar, S., Chanda, D., Ojha, S., Kumar, J.K., Chanotiya, C.S., Shanker, K., Negi, A.S., 2010. Antitubercular potential of some semisynthetic analogues of phytol. Bioorganic & Medicinal Chemistry Letters 20, 508-512.

Santos, C.C.d.M.P., Salvadori, M.S., Mota, V.G., Costa, L.M., de Almeida, A.A.C., de Oliveira, G.A.L., Costa, J.P., de Sousa, D.P., de Freitas, R.M., de Almeida, R.N., 2013. Antinociceptive and Antioxidant Activities of Phytol In Vivo and In Vitro Models. Neuroscience Journal 2013, 9.

Schönfeld, P., Reiser, G., 2006. Rotenone-like Action of the Branched-chain Phytanic Acid Induces Oxidative Stress in Mitochondria. Journal of Biological Chemistry 281, 7136-7142.

Silva, R.O., Sousa, F.B.M., Damasceno, S.R.B., Carvalho, N.S., Silva, V.G., Oliveira, F.R.M.A., Sousa, D.P., Aragão, K.S., Barbosa, A.L.R., Freitas, R.M., Medeiros, J.V.R., 2014. Phytol, a diterpene alcohol, inhibits the inflammatory response by reducing cytokine production and oxidative stress. Fundamental & Clinical Pharmacology 28, 455-464.

Singh, R., Meena, A., Negi, A., Shanker, K., 2012. Quantitative relationships between molecular descriptors, chromatographic retention behavior, and in vitro antituberculosis activity of phytol derivatives. JPC - Journal of Planar Chromatography - Modern TLC 25, 10-18.

Song, Y., Cho, S.K., 2015. Phytol Induces Apoptosis and ROS-Mediated Protective Autophagy in Human Gastric Adenocarcinoma AGS Cells. Biochem. Anal. Biochem. 4, 4-11.

Starcevic, V., 2014. The reappraisal of benzodiazepines in the treatment of anxiety and related disorders. Expert Rev Neurother 14, 1275-1286.

Steinberg, D., Avigan, J., Mize, C.E., Baxter, J.H., Cammermeyer, J., Fales, H.M., Highet, P.F., 1966. Effects of dietary phytol and phytanic acid in animals. Journal of lipid research 7, 684-691.

Tan, K., Rudolp, U., Lüscher, C., 2014. Hooked on benzodiazepines: GABAA receptor subtypes and addiction. University of Geneva, Retrieved December 12,.

Thakor, P., Mehta, J.B., Patel, R.R., Patel, D.D., Subramanian, R.B., Thakkar, V.R., 2016. Extraction and purification of phytol from Abutilon indicum: cytotoxic and apoptotic activity. RSC Advances 6, 48336-48345.

Tyagi, S., Gupta, P., Saini, A.S., Kaushal, C., Sharma, S., 2011. The peroxisome proliferator-activated receptor: A family of nuclear receptors role in various diseases. Journal of Advanced Pharmaceutical Technology & Research 2, 236-240.

Upadhyay, H.C., Dwivedi, G.R., Roy, S., Sharma, A., Darokar, M.P., Srivastava, S.K., 2014. Phytol Derivatives as Drug Resistance Reversal Agents. ChemMedChem 9, 1860-1868.

van den Brink, D.M., van Miert, J.N., Dacremont, G., Rontani, J.F., Wanders, R.J., 2005. Characterization of the final step in the conversion of phytol into phytanic acid. The Journal of biological chemistry 280, 26838-26844.

van den Brink, D.M., Wanders, R.J., 2006. Phytanic acid: production from phytol, its breakdown and role in human disease. Cellular and molecular life sciences : CMLS 63, 1752-1765.

Vieweg, W.V.R., Julius, D.A., Fernandez, A., Beatty-Brooks, M., Hettema, J.M., Pandurangi, A.K., 2006. Posttraumatic Stress Disorder: Clinical Features, Pathophysiology, and Treatment. The American Journal of Medicine 119, 383-390.

Wanders, R.J.A., Komen, J., Ferdinandusse, S., 2011. Phytanic acid metabolism in health and disease. Biochimica et Biophysica Acta (BBA) - Molecular and Cell Biology of Lipids 1811, 498-507.

Wang, J., Hu, X., Ai, W., Zhang, F., Yang, K., Wang, L., Zhu, X., Gao, P., Shu, G., Jiang, Q., Wang, S., 2017. Phytol increases adipocyte number and glucose tolerance through activation of PI3K/Akt signaling pathway in mice fed high-fat and high-fructose diet. Biochemical and biophysical research communications 489, 432-438.

Yu, S., Reddy, J.K., 2007. Transcription coactivators for peroxisome proliferator-activated receptors. Biochimica et Biophysica Acta (BBA) - Molecular and Cell Biology of Lipids 1771, 936-951.

Zakusov, V., Ostrovskaya, R., Kozhechkin, S., Markovich, V., Molodavkin, G., Voronina, T., 1977. Further evidence for GABA-ergic mechanisms in the action of benzodiazepines. Arch Int Pharmacodyn Thérap 229, 313-326.

Zhang, Y., Liu, X., Wang, Y., Jiang, P., Quek, S., 2016. Antibacterial activity and mechanism of cinnamon essential oil against Escherichia coli and Staphylococcus aureus. Food Control 59, 282-289.

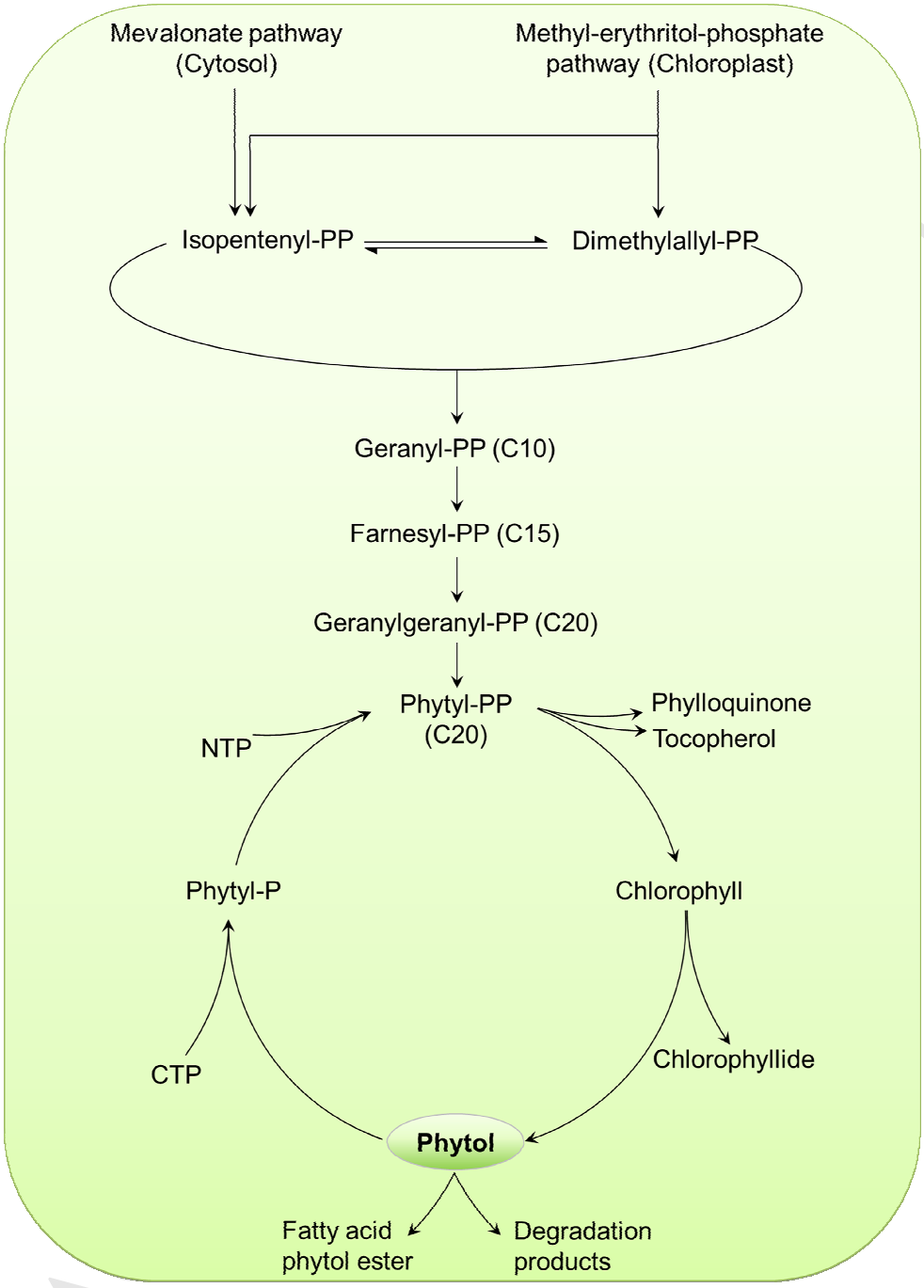

FIGURE 1. **A salvage pathway of PYT metabolism**. Phylloquinone, tocopherol, PYT-fatty acid ester, and other degradation products are known to be of phytyl-PP origin, which can be synthesized through the mevalonate or methyl-erythritol-phosphate pathway (Ischebeck et al., 2006; Islam et al., 2015).

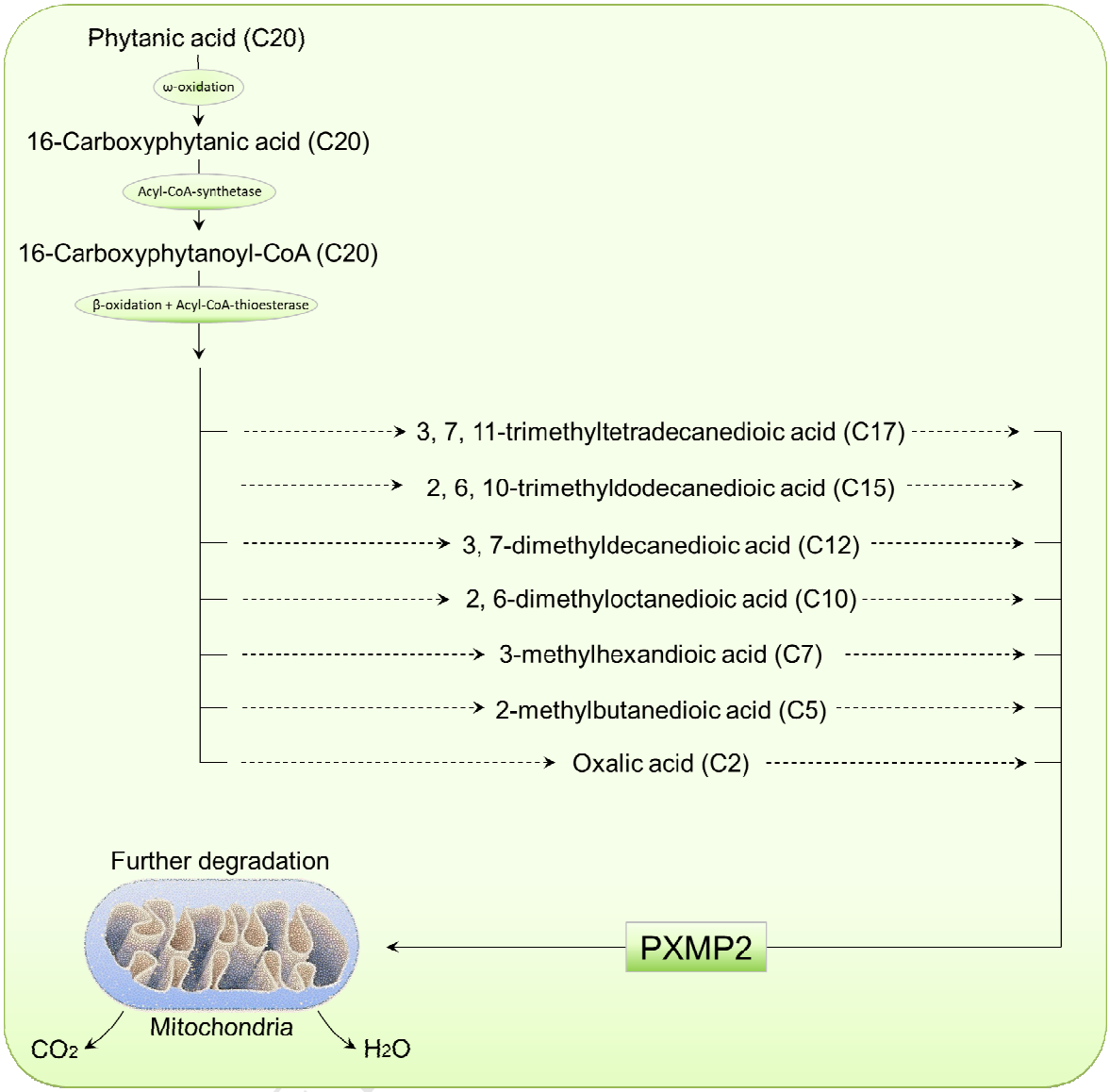

FIGURE 2. ω-Oxidation products of phytanic acid. Production of small and medium chain fatty acids with subsequent degradation to CO_2 and H_2O in mitochondria (Islam et al., 2015; Wanders et al., 2011).

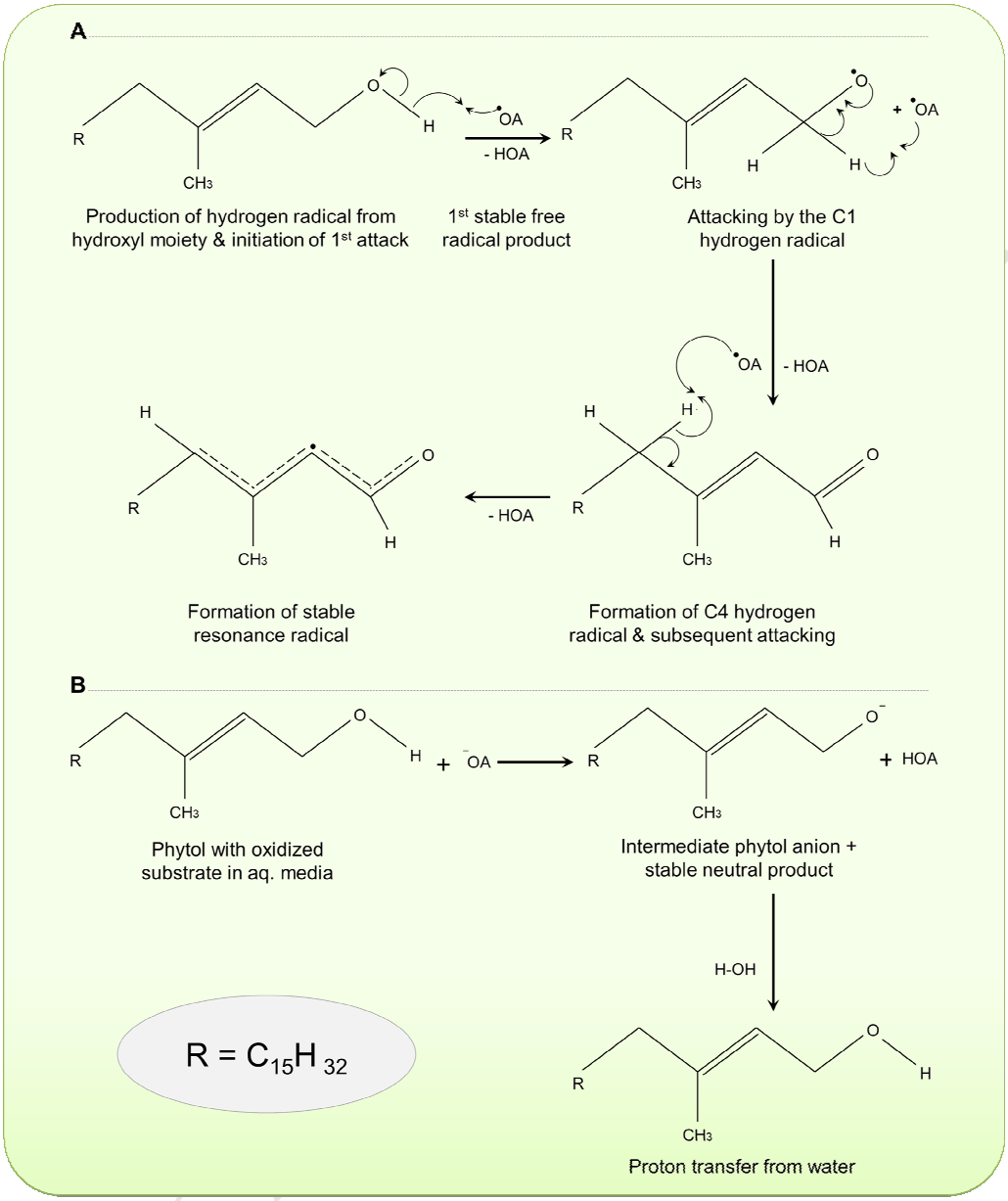

FIGURE 3. **Phytol mediated antioxidant mechanisms.** A. Free radical pathway: neutralization of oxidisable radicals by hydrogen radicals formation from different positions; **B.** Ionization pathway: neutralization of oxidisable anion through the liberated proton from alcoholic (OH) group.

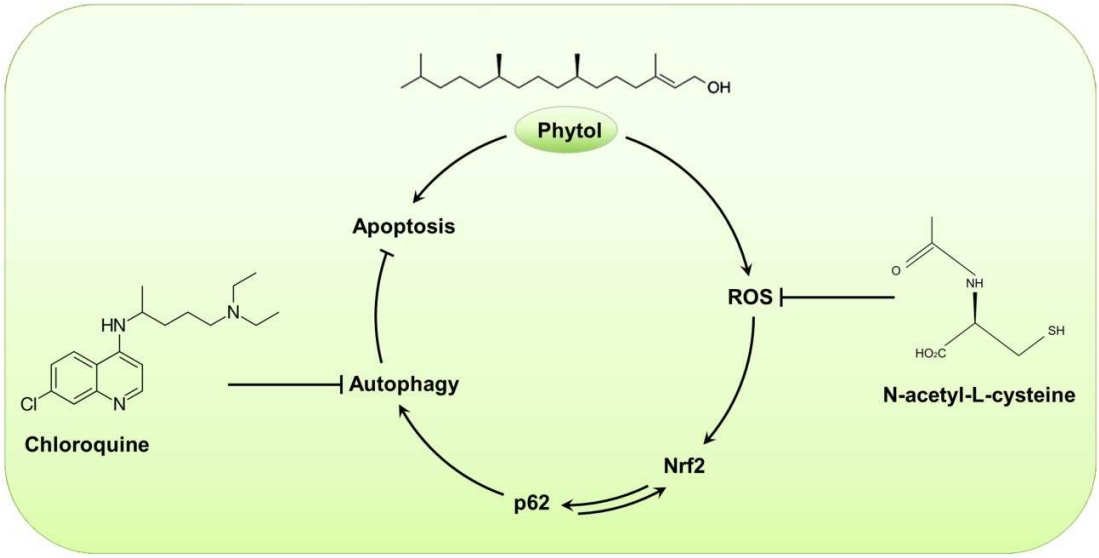

FIGURE 4. **Mechanism of apoptosis and autophagic effects of phytol**.

1 **Table 1. Summary of bioactivity studies of phytol and its derivatives**
2

Tested compound	Dose	Tests	Results	References
Phytol	62.5, 125, 250, 500 and 1000 µg/mL	Antimicrobial test against *Escherichia coli (*ATCC-25922*), Staphylococcus aureus (*ATCC-25923*), Aspergillus niger (*ATCC-16888*),* and *Candida albicans (*ATCC-10231*).*	MIC: 62.5 µg/mL (*E. coli, C. albicans*, and *A. niger*) MIC: >1000 µg/mL (*S. aureus*)	(Ghaneian et al., 2015)
	62.5, 125, 250, 500 and 1000 µg/mL	Toxicity on mouse skin cell suspension	Time and dose-dependent toxicity. Decrease of incubation time led to an increase of cell viability	
Phytol	0.5, 0.25 and 0.125 µg/mL of MIC (MIC was 19 µg/mL)	Anti-quorum sensing activity against *Pseudomonas aeruginosa* (*in vitro*)	Ability to reduce *P. aeruginosa* biofilm formation as well as twitching and flagella motility	(Pejin et al., 2015)
Phytol	0.15 to 160 µg/mL	Antibacterial activity against *S. aureus* (FDA209P)	Inhibitory activity on the growth within the range of 1.25 to 40 µg/mL	(Inoue et al., 2005)
Phytol	0 to 200 µM (24 h)	Cytotoxic activity in human gastric adenocarcinoma AGS cells	Anti-proliferative activity associated with autophagy induction with increase in acidic vesicular organelles formation	(Song and Cho, 2015)
Phytol	25, 50, 100, and 200 mg/kg (i.p.)	Antinociceptive (mice) and antioxidant tests	Antinociceptive and antioxidant activity in a dose-dependent manner	(Santos et al., 2013)
Phytol	10-100 µM	Anticancer test in lymphoid leukemia Molt 4B cells	Growth inhibition of Molt 4B cells due to the induction of apoptosis	(Komiya et al., 1999)
Phytol	0.5 µg/kg (per week) (i.p.)	Histological evaluation of Sprague-Dawley rats (female) mammary tumours	Inhibition of progression of MNU-induced tumours in mammary gland, along with decreased tumour volume	(Líška et al., 2011)
Phytol	50 - 200 mg; topically (16 w)	Tumor promoter test in Female ICR mice skin	Weak tumor promoter activity	(Kagoura et al., 1999)
Phytol	20, 40, 80 and 100 µM (24 h)	Antitumor mechanism of PYT in Huh7 and HepG2 cells	Antitumor activity *via* apoptosis induction through activation of caspase-9/3 and inhibition of EMT in hepatocellular carcinoma cells	(Kim et al., 2015)
Phytol	25, 50 and 75 mg/kg (i.p.)	Anticonvulsant effect in *Swiss* mice	Anticonvulsant activity in a dose-dependent way by modulating of neurotransmitter systems	(Costa et al., 2012)
Phytol	1 and 2 µM	Anticancer test in MDA-MB-231breast cancer cell line	Inhibition of the MDA-MB-231 cell line invasiveness	(Chikati, 2013)

Phytol	5 and 10 mg/kg (dissolved in 5% cremophor) (p.o.)	Anti-scratching behavioral test in ICR and BALB/*c* mice	Improving scratching behavior by inhibiting the allergic cytokines expression through NF-κB regulation in skin	(Ryu et al., 2011)
Phytol	0.5%, 1%, or 2% (p.o.) (7 d)	NAD synthesis and ACMSD expression in male Sprague-Dawley rats	Decrease of ACMSD activity and its mRNA expression in a dose-dependent manner in liver and in primary rat hepatocytes	(Matsuda et al., 2013)
Phytol	7.5, 25, 50, and 75 mg/kg (i.p.)	Anti-inflammatory test in mice (paw edema induced by diverse inflammatory agents)	Decrease of inflammatory response by inhibiting neutrophil migration partly caused by reduction in IL-1-beta and TNF-α levels	(Silva et al., 2014)
Phytol	25, 50 and 75 mg/kg (i.p.)	Anxiolytic-*like* effects in *Swiss* mice	Sedative and anxiolytic-*like* effects probably due to interaction with $GABA_A$ receptor	(Costa et al., 2014)
Phytol	12.5 to 100 µg/mL	Activity against *Schistosoma mansoni* worms (*in vitro*)	Death and reduced motor activity of the worms (*in vitro*), and worm burden reduction in the infected mice (*in vivo*)	(de Moraes et al., 2014)
	40 mg/kg (p.o.)	Test in Balb/*c* mice infected with *S. mansoni*		
Pivaloyl, 3,4,5-trimethoxybenzoyl, 2,3-dichlorobenzoy, cinnamoyl derivatives of phytol	Serial dilutions	Drug resistance reversal test against *E. coli* (CA8000 and DH5 alpha)	Drug resistance reversal activity (up to 16-fold activity potentiation)	(Upadhyay et al., 2014)
Phytol rich hexane fraction of *Lacistema pubescens*	5 to 111 µg/mL	Anti-promastigote and anti-amastigote assays with *Leishmania amazonensis*	Anti-promastigote and anti-amastigote activity in *L. amazonensis* with IC_{50} values of 44.0 and 25.8 µg/mL, respectively	(da Silva et al., 2015)
Phytanic acid	50 µM	Mitochondria from brain and heart of adult rats	Rotenone-like oxidative stress via a direct action on mitochondria	(Schönfeld and Reiser, 2006)
Phytanic acid	10 µM	Effects of PA on mouse neuroblastoma Neuro2a cells	Cell death *via* activation of Hdac2, 3	(Nagai, 2015)
Phytanic acid	1-500 µM	Effects of PA in Wistar male rats cerebellum and cerebral cortex	Increase of TBA-RS levels in both cerebellum and cerebral cortex significantly diminished GSH concentration	(Leipnitz et al., 2010)

Phytol and phytanic acid	Phytol: 500 mg/kg (p.o.) and phytanic acid 100 mg/kg (p.o.)	Anti-teratogenic activity in mice (female)	Reduction of teratogenesis caused by retinol	(Arnhold et al., 2002)
Phytanol and phytanyl chloride	44 mg per mice (i.p.)	Immunoadjuvant activity in mice exposed to *Staphylococcus aureus*	Strong induction of the protective cytokines IL-17 and IL-1β and enhancement of the anti-*S. aureus* immune response	(Roy Chowdhury et al., 2013)
Phytanol, phytanyl amine	Phytanol: 40 mg (i.p.); Phytanyl amine: 5 mg (i.p.)	Immunoadjuvant effects in BALB/*c* mice	Activation of neutrophils and monocytes, release of B-lymphocyte chemo-attractant (BLC), and enhanced expression of NLRP genes including NLRP3	(Aachoui et al., 2011a)
Phytanol, phytanyl amine	Phytanol: 40 mg (i.p.); Phytanyl amine: 2.5 mg (i.p.)	Immunoadjuvant activity in C57Bl/6 and BALB/*c* mice	Increase of immunogen-specific IgG1 and IgG2a antibody responses, modulatory immune response through apoptotic/necrotic effects on target tumor cells	(Aachoui et al., 2011b)
Phytol, phytanic acid and pristanic acid	3, 10 and 30 µM	Immunomodulatory activity on mouse splenocytes stimulated by T-cell mitogens	Reduction of IFN-γ, IL-4, and IL-10 by all the test substances and IL-17A by PYT and PA	(Nakanishi et al., 2016)
Phytol	250 mg/kg (p.o.)	Anti-diabetic test in adult male Wistar albino rats	Insulin-sensitizing/anti-diabetic effect of PYT *via* nuclear receptors activation and heterodimerization of RXR with PPARγ by PA	(Elmazar et al., 2013)

Table 2. Summary of test dose/conc., test system(s) and active dose/conc. of phytol and its derivatives

Phytol and/or its derivatives	Tested dose/Conc.	Test system(s)	Active dose/conc.	References
Phytol	62.5, 125, 250, 500 and 1000 µg/mL	*Escherichia coli*, *Staphylococcus aureus*, *Aspergillus niger*, and *Candida albicans*	62.5 & >1000 µg/mL	(Ghaneian et al., 2015)
	62.5, 125, 250, 500 and 1000 µg/mL	Mouse skin suspension	62.5 - 1000 µg/mL	
Phytol	0.5, 0.25 and 0.125 µg/mL of MIC	*Pseudomonas aeruginosa*	19 µg/mL	(Pejin et al., 2015)

Phytol	0.15, 0.30, 0.60, 1.25, 2.50, 5.0, 10, 20, 40, 80 160 µg/mL	*S. aureus*	0.15 µg/mL	(Inoue et al., 2005)
Phytol	12.5, 25, 50, 100 and 200 µM	Human gastric adenocarcinoma AGS cells	12.5 - 200 µM	(Song and Cho, 2015)
Phytol	*In vivo*: 25, 50, 100, and 200 mg/kg (i.p.); *In vitro*: 0.9, 1.8, 3.6, 5.4 and 7.2 ng/mL	*In vivo*: Swiss albino mice (n = 8); *In vitro*: TBARS, NO and OH scavenging assay	*In vivo*: 50 – 200 mg/kg; *In vitro*: 0.9 – 5.4 ng/mL	(Santos et al., 2013)
Phytol	10, 20, 40, 80 and 100 µM	Lymphoid leukemia Molt 4B cells	20 – 100 µM	(Komiya et al., 1999)
Phytol	0.5 µg/kg (per week); i.p.	Sprague-Dawley female rats	0.5 µg/kg	(Líška et al., 2011)
Phytol	50, 100, 150 and 200 mg	Female ICR mouse skin	50 - 200 mg	(Kagoura et al., 1999)
Phytol	20, 40, 80 and 100 µM	Huh7 and HepG2 cells	20 – 100 µM	(Kim et al., 2015)
Phytol	25, 50 and 75 mg/kg (i.p.)	*Swiss* mice (n = 24)	25 – 75 mg/kg	(Costa et al., 2012)
Phytol	1 and 2 µM	MDA-MB-231breast cancer cells	1 -2 µM	(Chikati, 2013)
Phytol	5 or 10 mg/kg (in 5% cremophor; p.o.)	ICR and BALB/*c* mice (n = 6)	5 and 10 mg/kg	(Ryu et al., 2011)
Phytol	0.5, 1, or 2%; p.o. (7 d)	Male Sprague-Dawley rats (n = 6)	0.5 – 2%	(Matsuda et al., 2013)
Phytol	7.5, 25, 50 and 75 mg/kg (i.p.)	Swiss albino mice (n = 5)	7.5 – 75 mg/kg	(Silva et al., 2014)
Phytol	25, 50 and 75 mg/kg (i.p.)	*Swiss* albino mice (n =7)	25 – 75 mg/kg	(Costa et al., 2014)
Phytol	12.5, 25, 50, 75 and 100 µg/mL	*Schistosoma mansoni*	50 - 100 µg/mL & 40 mg/kg	(de Moraes et al., 2014)

	40 mg/kg (p.o.)	Balb/*c* mice infected with *S. mansoni*		
Phytol rich hexane fraction of *Lacistema pubescens*	5 to 111 µg/mL	*Leishmania amazonensis*	25.8 – 44.0 µg/mL	(da Silva et al., 2015)
Phytanic acid	50 µM	Rat mitochondria from brain and heart	50 µM	(Schönfeld and Reiser, 2006)
Phytanic acid	10 µM	Mouse neuroblastoma Neuro2a cells	10 µM	(Nagai, 2015)
Phytanic acid	1, 10, 100 and 500 µM	Wistar male rats cerebellum and cerebral cortex	100 & 500 µM	(Leipnitz et al., 2010)
Phytol and phytanic acid	Phytol: 500 mg/kg (p.o.) and phytanic acid 100 mg/kg (p.o.)	Anti-teratogenic activity in female mice	Phytol at 500 mg/kg; Phytanic acid at 100 mg/kg	(Arnhold et al., 2002)
Phytanol and phytanyl chloride	44 mg per mice (i.p.)	Mice exposed to *S. aureus*	44 mg	(Roy Chowdhury et al., 2013)
Phytanol, phytanyl amine	Phytanol: 40 mg (i.p.) Phytanyl amine: 5 mg (i.p.)	BALB/*c* mice (n = 3)	Phytanol: 40 mg; Phytanyl amine: 5 mg	(Aachoui et al., 2011a)
Phytanol, phytanyl amine	Phytanol: 40 mg (i.p.), Phytanyl amine: 2.5 mg (i.p.)	C57Bl/6 and BALB/*c* mice (n = 6)	Phytanol: 40 mg; Phytanyl amine: 2.5 mg	(Aachoui et al., 2011b)
Phytol, phytanic acid and pristanic acid	3, 10 and 30 µM	Mouse splenocytes stimulated by T-cell mitogens	Phytol: 30 µM; Pristanic acid: 10 and 30 µM	(Nakanishi et al., 2016)
Phytol	250 mg/kg (p.o.)	Male Wistar albino rats (n = 20/80)	250 mg/kg	(Elmazar et al., 2013)