Journal of
Food and Pharmaceutical Sciences

Review Article

Black Pepper's Anti-Aging and Chemoprevention Properties: A Review

Endah*
Department of Biomedical Engineering, Sumatera Institute of Technology, Lampung, Indonesia
*Corresponding author: Endah | Email: endah@bm.itera.ac.id

Received: 28 February 2023; Revised: 16 August 2023; Accepted: 20 August 2023; Published: 21 December 2023

Abstract: The possible uses of black pepper, a common spice used for cooking, have been expanded. Despite countless studies on its advantageous biological effects, it would be difficult to determine its effectiveness as an anti-aging and chemo-preventive medication. Piperine, chavicine, caryophyllene, and sabinene are a few examples of black pepper components that exhibit pharmacological characteristics. This article reviews black pepper's main ingredients and discusses how effective it is at preventing aging and cancer. Using Google Scholar, PubMed, and Medline for our electronic research, we screened pertinent papers from scientific journals. To help with the explanation in the narrative, all the data was compiled and summarized into tables and graphs. In general, the anti-aging characteristics of *Piper Nigrum L.* and its compounds are exhibited through several pathways, including senescence prevention, antioxidant protection, control of ROS levels, and the inhibition of aging-related enzymes. Additionally, cell cycle regulation, antiangiogenic and anti-metastatic action, apoptosis induction, and the suppression of carcinogenesis are some of the mechanisms utilized in cancer prevention activities. It has been shown to have synergistic effects on several cancer cells when combined with doxorubicin. According to these studies, black pepper could be considered for development as an anti-aging and chemo-preventive agent.

Keywords: Black pepper, piperine, cancer, antiaging, chemoprevention

1. INTRODUCTION

Indonesia, Bangladesh, Brazil, India, and the West Indies are all home to large populations of black pepper, which belong to the Piperaceae. This species is commonly used in the world because it is very saleable, economical, and has health benefits. Black Pepper fruits and their simplicia (Figure 1) are used frequently to improve flavour and colour in food while the utilization of black pepper essential oil is widespread in food processing, serving both as a preservative and contributing to health benefits. [1]. Black pepper has always been utilized by humans in ancient medicine and household cures in addition to their food. In the fifth-century Syriac Book of Remedies, it is suggested to use *Piper nigrum* and *Piper longum* for addressing diverse health issues. These include insect bites, gangrene, joint pain, earaches, heart problems, hernias, insomnia, , toothaches, and liver disease. [2].

(a) (b)

Figure 1. (a) Black Pepper Tree, (b) Black Pepper Seed

Various studies have explored the physiological impacts of black pepper. As reviewed by Srinivasa (2009), black pepper showed the effect on a reproductive system including decreasing fertilizing ability, as an antimutagenic and tumor inhibitory, anti-inflammatory, hepatoprotection, melanocyte stimulation, neuropharmacology, anticonvulsant, and amelioration of dysphagia [2]. Other biological effects of black pepper are a bioavailability enhancer, anti-cancer, natural antioxidant, cholesterol-lowering, immune enhancer, antipyretic, improving digestion, and promoting intestinal health [3]. Moreover, Salehi, et al (2019) summarized that black pepper exhibits antimicrobial activity, anxiolytic, antidepressant, neuroprotective, and antineuronal inflammatory effects [4].

Piperine is one of the substances in black pepper that is most prevalent. Some comprehensive studies regarding piperine biological activities are summarized, including antioxidant, anti-inflammatory, bio-enhancing ability, inhibition of drug biotransformation, and mitochondrial energetics [1], [5]. On the other hand, Piperine has antitumor effects on several malignancies, including colorectal, prostate, and breast cancer [6]. Besides piperine, black pepper also contains terpenoid substances like sabinene, limonene, and β-caryophyllene [7]. Most of them are terpenoids that have antioxidant activities.

The predominant component in black pepper essential oil is β-caryophyllene, which is 22.60% of the total 46 compounds [8]. β-caryophyllene shows better antioxidant activity than butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) [9] and is anti-inflammatory [10]. Not only acting as an antioxidant, several studies suggest that β-Caryophyllen has anticancer activity. β-Caryophyllene exhibits toxicity towards certain cancer cells, including those associated with breast cancer and colon cancer [11][10]. However, the application of black pepper essential oil in society does not suggest any harmful impact on the body. The essential oil demonstrates selectivity toward cancer cells, sparing normal cells like ovarian and fibroblast cells from its effects [8].

Although various studies have demonstrated the antioxidant and anti-cancer properties of black pepper, no thorough overview of the subject has been published yet. As a result, in this article, we will analyze current original research publications that examine the effects of *Piper nigrum* and/or its constituents on anti-aging and cancer-related symptoms. This review article seeks to give a general overview of the effectiveness of black pepper as an anti-aging and chemoprevention medicine, including its extract and/or essential oil as well as its chemical ingredients. To give a deeper and more

thorough explanation of black pepper's effectiveness as an anti-aging and chemo-preventive drug, We also go through black pepper's molecular targets in cancer cells and normal cells as antiaging targets.

2. DISCUSSION

2.1. Constituent of Black Pepper

2.1.1. Black Pepper Extract Constituent

Black pepper contains alkaloids such as piperine (5.3-9.2%), chavicine (up to 1%), and methyl pyrroline. In addition, black pepper also contains essential oils (1.2-3.5%) [12]. Wongpa et al. (2007) reported that black pepper contains 1-2.5% evaporated oil, 5-9% piperine and piperetin alkaloid crystals, and resin [12]. Another study states that black pepper contains many compounds, namely protein, lignin, starch, piperidine, chavicine, cineole, carvone, pinene, schimiditin, sesquiterpenes, piperlotin, benzoic acid derivatives, volatile oils like phellandrene, terpenes, piperonaldihydrocarbeol, caryophyllene, and caryophyllene oxide [13]. The main constituent of black pepper is piperine. Gorgani et al. (2017) state that the piperine content in black pepper is above 9%, in cubeb (*Piper longum* L.*)* it is 4%, and in Javanese chili (*Piper retrofractum* Vahl) is around 4.5% [5]. The supplement of Indonesian Herbal Pharmacopoeia (2010) states that the piperine content in black pepper ethanolic extract is not less than 48.60%. The percentage of piperine content in plants depends on the environment, growing conditions, and where it is grown.

The primary component of black pepper, piperine, is identified by the IUPAC name (2E, 4E)-5-(1,3-benzodioxol-5-yl)-1-piperidin-1-ylpenta-2,4-dien-1-one. Piperine exists in four isomeric forms: piperine, isopiperine, chavicine, and isochavicine. (Figure 2) [5]. Piperine belongs to the pyridine-piperidine class of alkaloids [14].

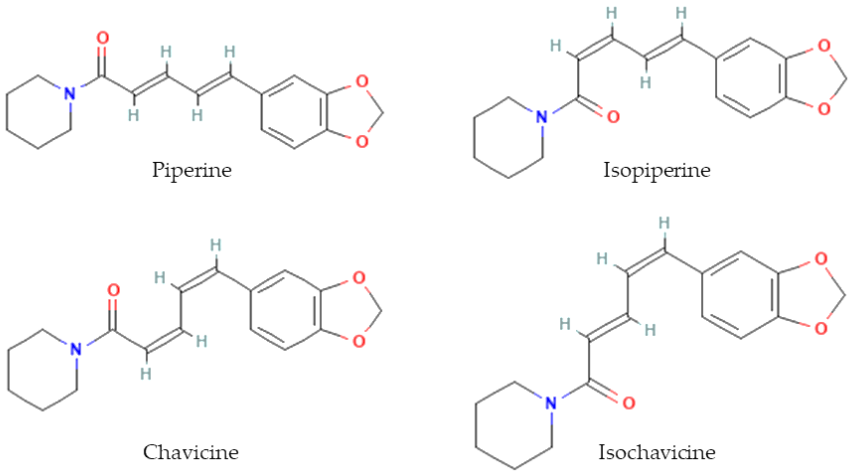

Figure 2. Chemical Structure of Piperine and Chavicine

2.1.2. Components of Black Pepper Essential Oil

Essential oil of black pepper contains caryophyllene, caryophyllene oxide, phellandrene, piperonaldihydrocarbeol, and terpenes [11]. Wongpa et al (2007) reported that Black Pepper contains 1-2.5% volatile oil [12]. Table 1 displays the primary constituents of essential oil derived from black pepper.

Table 1. Compound Ingredient derived from Black Pepper Oil

Ingredients	The abundance of literature (%)					
	[9]	**[15]**	**[16]**	**[17]**	**[18]**	**[7]**
Caryophyllene	-	18.393	-	-	19.12	26.2
β-caryophyllene	29.9	-	18.6	20.225	-	-
Limonene	13.2	-	15.0	16.924	9.74	-
D-limonene	-	16.168	-	-	-	-
Pinene	-	-	-	-	-	5.5
α-Pinene	-	16.685	-	-	-	-
β-pinene	7.9	13.204	-	-	-	-
Sabinene	5.9	-	13.2	18.054	-	-
3-Carene	-	-	8.6	-	-	-
Camphene	-	-	-	-	8.44	-
P-cymene	-	-	-	-	-	5.8

β-Caryophyllene, also known as caryophyllene or (-) - β-Caryophyllene, is a sesquiterpene class of compounds commonly found in essential oils in cloves *Syzygium aromaticum*, *cannabis sativa*, rosemary, and hops. These compounds are often found in mixtures with isocaryophyllene and α-humulene (called α-Caryophyllene) (Figure-3). β-Caryophyllene has the IUPAC name (1R, 4E, 9S) - 4,11,11-trimethyl8-methylidenbisiklo [7.2.0] undec-4-ene (PubChem., nd).

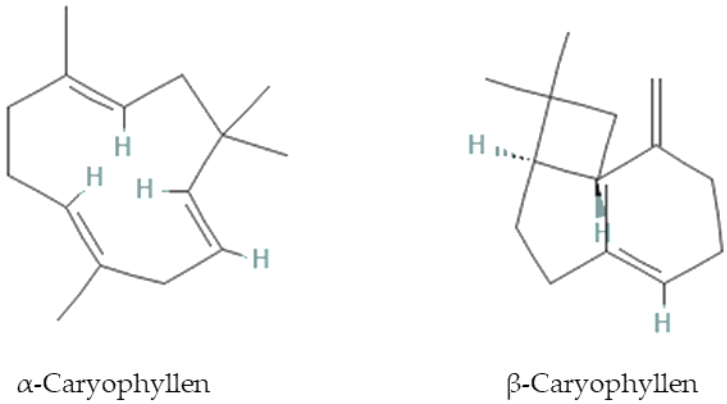

Figure 3. Chemical structure of α-Caryophyllene and β-Caryophyllene

2.2. Anti-Aging Activities of Black Pepper

Black Pepper is utilized as an anti-fever and diarrhoea remedy in the fifth century. [2]. The ancient book of Ayurvedic Materia Medica states that Black Pepper and Ginger are effective in balancing the body's physiology so that it can prevent disease [19]. The Arabs discovered essential oils from many plants, including black pepper, throughout the Middle Ages. So far, Essential oils are frequently used as a local anaesthetic, sedative, anti-inflammatory, and food preservative [20]. Furthermore, joint pains, antipyretic, muscular pains, and respiratory infections have all been treated with black pepper essential oil. In addition, it is also utilized to make more saliva., increase appetite, and in improving digestion [21]. Table 2 provides an overview of earlier studies on the anti-aging impact of extract from Black Pepper.

Table 2. The aging effects slowed down by Black Pepper.

Anti-aging Effect	Material	Activity	Reference
Anti-oxidant	Black pepper oil	Reduces the level of free radicals better than BHA and BHT	[22], [9]
		lowers peroxide and thiobarbituric acid compared to BHA and BHT	[23]
	Black pepper extract	Reduces the level of free radicals and increases superoxide dismutase and GST in the liver of female balb/c mice	[24], [25]
		Prevents LDL oxidation which can increase free radicals	[26]
	Piperine	modulates the peroxidation level of lipids in a rat increasing antioxidant enzymes activity	[27]
Modulation of reactive oxygen species (ROS) Level	Black pepper extract	Inhibits ROS and lipogeneses on Male Wistar rats Reduce the activity of catalase, superoxide dismutase, glutathione S-transferases (GST), and glutathione (GSH) in the heart, ren, liver, aorta, and intestine	[28]
		Lowers the level of ROS on normal cells (CHO-K1 cell lines)	[29]
		Increases ROS on cancer cells (HeLa, ZR-75-1, and MCF-7 cell lines)	[30], [31]
	β-Caryophyllene	Decreases GSH on lymphocyte cells	[32]
Anti-senescence	Black pepper oil	Lowers senescence cells on normal cells (CHO-K1 and NIH-3T3 cell lines)	[8]
	Piperine	Reduces senescence cells and increases in protection of hippocampal and hippocampal volume neurons in Wistar rats	[33]
		Increases senescence cells on cancer cells (4T1 cell lines),	[34]

2.2.1. Antioxidant Activity

Antioxidants are chemicals that limit or stop cell oxidation caused by free radicals. [35]. To keep the body functioning at its best, antioxidant supplementation is a growingly common practice [36]. Atoms or molecules with one or more unpaired electrons are known as free radicals. Due to their high reactivity, these electrons combine with other molecules very fast to produce free radicals. Free radicals can harm cells, resulting in early aging and several disorders. One effort to capture free radicals is antioxidants such as vitamins C and E ([35].

Black pepper has been studied in vitro for its antioxidant activity. In vitro, the antioxidant test can be done using cell culture, namely through the DPPH test and using cell culture through DCFDA staining. Meanwhile, in vivo black pepper was done with experimental animals such as Balb/C mice. Black pepper oil and extract reduced free radical levels through the DPPH assay [9]; [25]. Another mechanism, black pepper extract increases superoxide dismutase and GST in the liver [25] and antioxidant enzymes activity by modulating the peroxidation level of lipids in a rat [27]. Hence, black pepper may be able to slow down aging due to its antioxidant properties.

2.2.2. ROS Level Modulation

Reactive Oxygen Species (ROS) are reported to interact with macromolecules in cells such as lipid peroxidation, DNA, RNA, and protein oxidative damage [37]. Free radicals can attack both

purine and pyrimidine bases.ROS causes Genotoxicity by cleaving single-stranded and double-stranded DNA, altering bases, changing deoxyribose sugars, and cross-linking DNA [37]. Intracellular ROS levels play a role in the redox balance in cells so ROS levels must be maintained. The antioxidant activity of Black pepper is useful for suppressing the level of ROS in cells so that oxidative stress does not occur which results in carcinogenesis and cell death.

Black pepper has been studied for its effect on modulating ROS levels. Black pepper extract restrains ROS and lipogenesis in Wistar rats, diminishing the function of catalase, GST, superoxide dismutase, and GSH in the heart, liver, intestine, aorta, and kidneys of male Wistar rats [28]. Besides, black pepper extract lowers the level of ROS in normal ovary cells (CHO-K1) while it is increased in various cancer cells (HeLa, MCF-7, and ZR-75-1) [29]–[31]. Intracellular ROS levels that increase above the threshold in cancer cells become lethal to some cancer cells through senescence and apoptosis mechanisms. Conversely, decreasing intracellular ROS levels in normal cells conduces these cells to avoid the process of senescence [38]. Senescence is a factor that leads to aging [39]. Thus, black pepper can reduce ROS levels in normal cells to delay aging and reverse the effect on cancer cells.

2.2.3. Anti-senescence

As cells permanently lose their ability to proliferate, they enter senescence, a complex stress response [40]. Stimuli triggering aging can be done utilizing genotoxic drugs, radiation, metabolic disorders, and tissue remodelling [41]. Senescence may result from cells entering a state of cell cycle arrest, DNA damage responses, and oxidative stress due to increased ROS accumulation. Cellular senescence refers to cell arrest during the phase G1 of the cellular cycle that continues to proliferate, in response to stress [42]. Normal cells can experience natural or premature aging. Premature aging of normal cells can occur due to cellular damage which is generally triggered by oxidative stress. Premature aging that occurs in normal cells is allowed to trigger a decline in fungal physiology which results in degenerative diseases like cardiovascular, diabetes, and cancer [43]. Furthermore, the aging of fibroblast cells can interfere with an appearance that can cause wrinkles on the skin. Therefore, inhibition of aging in reproductive cells is important to maintain human survival.

Black pepper oil reduces the percentage of senescence cells due to doxorubicin induction in CHO-K1 normal cells and NIH-3T3 cells [8]. While in cancer cells, piperine increases cell senescence in 4T1 breast cancer cells where the effect is further enhanced after being combined with the compound curcumin analog Pentagamavunon-1 [34]. Thus, black pepper has effects related to aging that are opposite to normal cells and cancer cells. Therefore, the encouraging potential of black pepper in demonstrating both anti-aging and anti-cancer properties is noteworthy.

Based on the previous explanation, it can be inferred that black pepper functions as an antioxidant, decreases the levels of intracellular ROS, and possesses anti-senescence properties. Senescence can be postponed by regulating intracellular ROS levels to remain within normal limits. Antioxidants, on the other hand, contribute to the maintenance of intracellular ROS levels by restoring redox balance. In simpler terms, antioxidants reduce intracellular ROS levels, consequently slowing down the senescence process. Thus, delayed senescence serves as an anti-aging mechanism, as previously discussed.

2.3. Chemo-preventive characteristics of Black Pepper against cancer

Research has been conducted on the pharmacological impact of black pepper extract and essential oil against cancer cells. The potential of inducing cancer cell death has been explored in the study of both black pepper extract and its constituent treatment. The chemo-preventive effect of piperine can modify the expression and activity of various enzymes, proteins, gene products, and transcription factors related to cell viability and proliferation [6]. A summary of previous research could be seen in Table 3.

Table 3. Target of Chemo-preventive effects of Black pepper

Chemo-preventive effect	Material	Molecular/Cellular Effect	Reference
Anti-oxidant	Piperine	Increases ROS on HeLa, MCF-7, and ZR-75-1 cell lines	[30], [31]
		Decreases GSH on lymphocyte cells	[32]
	β-caryophyllene	Increases GST on cancer cells	[44]
	Trans anethole	Its anti-oxidant activity is better than α-tocopherol	[45]
	α-pinene, trans-anethole, thymol	Increases the activity SOD, CAT, POX, GST, APOX, and GPDH *Ephestia kuehniella*	[46]
Carcinogenesis inhibition	Piperine	Decreases ALDH on breast cancer stem cells	[47]
		Suppresses the expression of HER2 on SKBR3, BT-474, MCF7/HER2, MDA-MB231.	
		Decreases NADPH-C reductase, cyt-p450, cyt-b5 and increase GPx, GR G6PDH on lung cancer-induced mice	[27]
Control of the cell cycle	Piperine	Causes G2/M arrest on HeLa cells	[30]
		Causes cell cycle accumulation in the G1/G0 phase in Caco2, L-MDR1, and LLC-Pk1 cell lines	[48]
		Induces cell cycle arrest Supresses cell division	[31]
		Increases the accumulation of cells in the G2/M phase of the cell cycle. and downregulated the expression of cyclin B1	[49]
		Induces mitotic catastrophe on 4T1 cells	[34]
	β-caryophyllene	Shows cytotoxic effect on A-549 and DLD-1 cell lines	[11]
Apoptosis	Piperine	Activates caspase-3 on HeLa Cells	[30]
		Activates caspase-3 on 4T1 Cells	[49]
		Increases PARP on SKBR3, BT-474, MCF7/HER 2, MDA-MB231	[47]
		Down-regulates p53 genes on lymphocyte cells and up-regulates it on ZR-75-1 and MCF-7 cells	[32], [31]
Anti-angiogenesis and anti-metastasis	Piperine	Down-regulates MMP-9 on HT-1080	[50]

continued table 3

		Down-regulates MMP-2, E-cad, and c-Myc on SKBR3, BT-474, MCF7/HER 2, and MDA-MB231 cell lines	[47]
		Down-regulates VEGF on MCF-7 cell lines	[31]
		Inhibits the Wnt/ β-catenin pathway and suppresses β-catenin nuclear localization on HCT116, SW480, and DLD-1 cell lines	[51]
		Decreases MMP-9 and MMP-13 expression and prevented 4T1 cell movement.	[49]
Co-chemotherapy/ anti-resistance	Piperine	Inhibits ABC transporter genes (P-gp, MRP1, and BCRP dependent multidrug resistant cancer cell lines, MCF-7 and A-549	[52]
		Increases AB digoxin transport and inhibits digoxin transport in the BA crosswise the L-MDR1 and Caco-2 cells	[53]
		Increases P-gp protein expression and up-regulation of MDR1 mRNA levels on Caco2	
		Modulates doxorubicin tissue distribution, induces drug interactions, and facilitates the conveyance of drugs classified as P-glycoprotein substrates to target tissues and tumors.	[54]
		Combination with tamoxifen decreases the expression of P-gp mRNA in MCF-7	[55]
		Enhances the cytotoxicity of doxorubicin in Caco-2 and CEM/ADR 5000 cells.	
		Increases the intracellular retention of the fluorescent P-glycoprotein substrates rhodamine and calcein and restrains their removal from the MDR cell lines.	[56]
		Inhibits the transcription of ABC genes on MCF-7 and A-549 cell lines	[57]
		Increases the cytotoxicity of PGV-1 in 4T1 cells	[34]
	β-caryophyllene	Increases the cytotoxicity of α-humulene and paclitaxel on MCF-7 and DLD-1 cell lines	[10]

2.3.1. Antioxidant Activity

Antioxidants could interact with free radicals and stabilize them, which in turn prevents damage to cells. As a result, they are believed to be capable of stopping or preventing further damage during cancer development, as free radical-induced DNA damage is a potential cause of cancer. Even though the connection between DNA oxidation and carcinogenesis is not yet definitively established, some studies have suggested that certain DNA oxidation products can be used as biomarkers to assess antioxidant levels and cancer risk [58].

The activity of piperine in antioxidant activity in cancer cells can be seen in Table 3. Based on the explanation above, piperine Increases ROS levels in ZR-75-1, HeLa, and MCF-7 cell lines [30], [31]. Elevating the levels of intracellular ROS results in the suppression of cancer cell proliferation [38]. As cancer cells undergo accelerated proliferation and metabolic changes, their basal levels of ROS approach a threshold [43], [59], and continued escalation in intracellular ROS levels, that normal cells can endure, proves fatal for the majority of cancer cells. [60], [43]. Thus, black pepper's antioxidant properties offer the possibility of developing it into an anti-cancer agent.

2.3.2. Carcinogenesis Inhibition

Carcinogenesis refers to the process of triggering and advancement of cancer. The intricate processes at play are influenced by a variety of variables, including age, genetics, and environmental exposure. Cancer cells must be able to promote their growth, accumulate the necessary ingredients, resist elimination processes, and proliferate to start. Chemopreventive drugs are substances that can stop, slow down, or prevent the development of cancer [61].

Since piperine has antiinflammatory, antioxidant, and antitumoral effects, various cancer models have been studied to investigate its potential as a chemopreventive agent. It is used for therapy in clinical oncology. Several studies have shown that piperine can suppress carcinogenesis. Piperine regulates various enzymes that have a role in the mitochondrial Krebs cycle and increases the enzymes GPx, GR, and G6PDH [27]. This enzyme is a glutathione metabolizing enzyme that plays a role in cancer formation. In addition, piperine treatment alone or when combined with curcumin, it had the ability to decrease ALDH in breast cancer stem cells [57]. ALDH is a marker for breast cancer cells. ALDH plays a role in cancer cell resistance to chemotherapy agents [62]. Other reports reveal that piperine has been found to inhibit genes such as BCRP, P-gp, and MRP1 that plays a role in the emergence of cancer cells' resistance to medications, specifically in A-549 and MCF-7 cell lines [52]. In various breast cancer cell lines such as SKBR3, BT-474, MCF7/HER2, and MDA-MB231, piperine has demonstrated the ability to suppress HER2 expression [47]. As a result, black pepper holds promise as a potential anti-breast cancer agent.

2.3.3. Control of The Cell Cycle

Cell cycle dysregulation is believed to be the underlying cause of cancer. While damage or mutations would typically lead to cell death, in cancer cells, this process is disrupted, and the cell continues to divide through the cell cycle, despite accumulating mutations. A characteristic feature of cancer cells is their capability to undergo rapid proliferation without the need for external stimuli, thanks to alterations in intracellular signaling pathways that allow them to enter the cell cycle without requiring any favorable or unfavorable external factors [62]. The cell's release of growth factors signaled the start of the stimulation of growth. The release of the following proteins into the cell nucleus was triggered by signals that crossed the membrane and entered the cytoplasm and transcription factors. Cells are prompted to begin the cell cycle by it. The initial gap phase, or G1, is followed by the S phase for DNA synthesis in cells that will multiply. Following the S phase, the cell enters the G2 phase where it prepares to duplicate its machinery for the M phase (mitosis) [63].

Another study reported that piperine regulates the cell cycle in some cancer cells. Piperine has been shown to induce halting of the cell cycle at G1/G0 stage on Caco2, L-MDR1, and LLC-Pk1 cell lines [48] and G2/M phase in HeLa and 4T1 cell lines [30], [49]. Moreover, the halting of cell

progression is distinguished by the increased expression of p53 and cyclin B1, tightly linked to the control of the cell cycle, resulting in a reduction in tumor size [30], [31], [49]. Relevant research reveals that piperine induces mitotic catastrophe in 4T1 cells [34] that refers to an event where the cell cycle fails during the initiation phase of mitosis due to disturbances in cell cycle regulatory proteins such as CDK and PLK-1, ultimately resulting in triggering of senescence and apoptosis [34].

2.3.4. Effect on Apoptosis

Apoptosis is distinguished by various factors, such as cellular shrinkage, condensation of chromatin, the creation of apoptotic bodies, fragmentation of DNA, and blebbing of the plasma membrane [64]. This mechanism necessitates the production of ATP and certain proteins without triggering an inflammatory reaction [64]. Extrinsic and intrinsic routes are the two basic mechanisms through which apoptosis can occur. The intrinsic mechanism happens through the mitochondrial system, whereas the extrinsic pathway of apoptosis necessitates the initiation of death receptors by binding to ligands, leading to caspase 8 triggering [64].

In inducing apoptosis, piperine can increase caspase-3 expression in the 4T1 and HeLa [30], [49]. The caspase-dependent pathway is the primary mechanism for cellular apoptosis, where it contrubutes a crucial function in the programmed cell death process by cleaving the nuclear DNA of apoptotic cells [65]. Another mecanism, piperine increases p53 expression in MCF-7 and ZR-75-1 line cells [31], while it is decreased on lymphocyte cells [47]. Elevated levels of p53 have been investigated to act a significant function in promoting apoptosis in cancer cells. P53 actively participates in extrinsic and intrinsic pathways of apoptosis by upregulating the expression of death receptors and apoptotic regulator including Bax and Bid [66]. In addition, PARP (poly-ADPribose polymeras) is marker of apoptosis. piperine increases PARP on SKBR3, BT-474, MCF7/HER 2, MDA-MB231 [47].

2.3.5. Anti-angiogenesis Effectiveness

To give cancerous tissue oxygen and nutrition, cancer cells could promote angiogenesis, which results in the formation of new blood vessels around the diseased tissue. Angiogenesis is surprisingly triggered early in the multistage development of invasive cancers, observed in both animal models and humans. Cancer cells can readily infect different tissues if the conditions are fulfilled with healthy cells. The bodily tissues are susceptible to invasion and spread by cancer cells [67]. Certain angiogenic regulators consist of signaling proteins that engage with receptors on the surface of vascular endothelial cells, which can either stimulate or inhibit cellular responses. Vascular endothelial growth factor-A (VEGF-A) and thrombospondin-1 (TSP-1) are widely acknowledged as representative examples of angiogenesis inducers and inhibitors, respectively [67].

Piperine has been demonstrated to have an impact on the angiogenesis. Doucette et al. (2013) presented findings that indicate piperine's interference with Akt activation in human umbilical vein endothelial cells (HUVECs) that endured to growth factors that promote angiogenesis, like VEGF. The study suggests that piperine achieves this inhibitory effect by preventing Akt phosphorylation at specific sites, namely Ser 473 and Thr 308 residues [68]. As Akt is a crucial signalling molecule during angiogenesis, this inhibition is likely responsible for piperine's ability to suppress the process [69]. Concurrently, a separate study demonstrated that piperine reduced the expression of VEGF in the MCF-7 cell line [31]. As a consequence of this down-regulation, breast cancer angiogenesis was inhibited. In the normal angiogenic process, VEGF is released in epithelial cells, prompting tumor

cells to liberate MMPs (matrix metalloproteinases) that fragment the extracellular matrix (ECM) [70]. Subsequently, tumor cells undergo invasion and angiogenesis. However, piperine's action on reducing VEGF levels disrupted this process, leading to the inhibition of breast cancer angiogenesis.

2.3.6. Effect on Metastasis

Metastasis is the process of spreading cancer cells to other tissues or organs and results in the formation of new tumors in other organs. Metastasis begins with the infiltration and movement of cancer cells to leave the primary organ [67]. extracellular matrix (ECM) acts as a vehicle for interaction between cells and contains various growth factors [76]. ECM degradation causing metastasis is mediated by matrix metalloproteinase (MMP), which is a zinc-dependent endopeptidase [77]. Cancer cells synthesize MMP in small amounts, but through the secretion of interleukins, interferons, and growth factors trigger host cells to produce MMPs, which in turn bind to the surface of cancer cells [78]. MMP-2 and MMP-9 are gelatinases that can degrade gelatine and ECM components including aggrecan core protein, collagen types IV, V, XI, and laminin [79]. Degraded ECM provides a pathway for cancer cells to migrate using pseudopodia [80].

Piperine has been demonstrated to have an impact on the metastasis of cancer cells, as indicated in table 3 above. Piperine down regulates MMP-9 on HT-1080 [50], MMP-2 on SKBR3, BT-474, MCF7/HER 2, MDA-MB231 [47], and MMP-13 on 4T1 cell lines [49]. Increasing tumor-derived MMP-13 expression has been proven to independently predict poor prognoses [71], and it has been proven that more aggressive breast cancer cells express MMP-13 [72]. In cases of breast cancer, MMP-9 expression is observed in cancer tissue, and its activity is higher in malignant breast cancers compared to benign tumors [73]. Therefore, piperine could be developed as a chemoprevention agent by inhibiting metastasis.

3.1. Black pepper as Potential Cancer Co-Chemotherapeutic Agent

Cancer research is focused on developing companion drugs that can be used in combination with chemotherapy because breast cancer cells can develop resistance to chemotherapeutic agents, which limits their effectiveness. An alternated strategy to combat cancer cells' resistance to chemotherapeutic drugs and boost their efficacy is to combine them with a chemo-preventive agent [74]. The idea behind combining drugs is to improve their effectiveness in fighting cancer cells while minimizing their toxicity to healthy tissues [75].

It is recognized that piperine can enhance the potency of other anti-cancer drugs and act as a co-chemotherapy agent. Several earlier investigations have examined the advantages of piperine as a co-chemotherapy agent, both in laboratory experiments and in live organisms. Piperine has the potential to enhance the cytotoxic impact of doxorubicin on Caco-2 cells by enhancing the buildup of fluorescent substances that are substrates for P-glycoprotein (P-gp) within the cell [56] and increase the effectiveness of tamoxifen on MCF-1 cells by reducing P-gp mRNA expression [55] and modulating the distribution of doxorubicin in tissues and induce drug interactions in mice [54]. Recent studies have revealed that piperine enhances the effectiveness of Pentagamavunon-1 through mitotic catastrophe and cell senescence [34]. Thus, piperine has the potential a co-therapy with other anti-cancer agents.

3. CONCLUSION

Black pepper and its components demonstrate anti-aging effects via various mechanisms, which encompass anti-senescence, antioxidant activity, and modulation of ROS (reactive oxygen species) levels. The mechanisms involved in cancer prevention activities from Black pepper and its major constituents are anti-oxidant, suppression of carcinogenesis, cell cycle regulation and apoptosis, and antiangiogenic and anti-metastasis effect. Furthermore, black pepper is great in increasing the effectiveness of chemotherapeutic agents.

Funding: No external funding was received for this research.
Acknowledgments: The author thanks Prof. Dr. apt. Edy Meiyanto, M.Si. in Gadjah Mada University for advice in the writing of this manuscript.
Conflicts of interest: The authors affirm the absence of any conflicts of interest.

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