Interventional Medicine & Applied Science, Vol. 6 (4), pp. 139–146 (2014) REVIEW

Curcumin a potent cancer preventive agent: Mechanisms of cancer cell killing

MUOBARAK JABER TUORKEY*

Zoology Department, Faculty of Science, Damanhour University, Damanhour, Egypt
*E-mail: physio_mj_tuorkey@yahoo.com

(Received: June 5, 2014; Revised manuscript received: June 29, 2014; Accepted: August 1, 2014)

Abstract: There is no doubt that diet could effectively improve health and halt cancers. Dietary phytochemical compounds and their derivatives represent a cornucopia of effectively anticancer compounds. This review discusses existing data on the anticancer activities of curcumin, and then offers possible explanations for and mechanisms of its cancer-preventive action. This review also offers insights into the molecular mechanism and targets through which curcumin modulates cell cycle, apoptotic signals, anti-apoptotic proteins, miRNAs, Wnt/beta-catenin signaling, protein kinases, nuclear factor-κB, proteasome activation, epigenetic regulation including DNA methylation and histone modification. Finally, this review provides explanations for how curcumin reverses the multi-drug resistance (MDR) of cancer cells.

Keywords: cell cycle arrest, apoptotic signals, miRNAs, proteasomes, DNA methylation, histone modification

Introduction

Compounds of natural origin could lead to new, innovative therapeutic agents for cancer. Mans et al. have enlisted several examples of naturally derived anticancer compounds [1, 2], for example, vincristine, which is derived from the periwinkle plant *Vinca rosea*; etoposide, which is derived from the mandrake plant *Podophyllum peltatum*; and taxol, which is derived from the pacific yew *Taxus brevifolia*. A number of promising new agents are in clinical development based on their selective molecular targets in the field of oncology [3]. Yet, due to the depth of our understanding for the etiology of many diseases that enabled us to design and synthesis of drug molecules for specific molecular targets. Therefore, we can shift the attention from the chemically synthetic drugs to the purely natural ones [4, 5]. Curcumin, a yellow pigment obtained from the rhizomes of *Curcuma longa* (Family: *Zingiberaceae*), is a major component of turmeric and is commonly used as a spice and food coloring agent [6]. *C. longa* has been used in traditional remedy for a wide range of ailments, including wound healing, urinary tract infection, and liver ailments [7]. Various metabolites of curcumin have been reported, including dihydrocurcumin (DHC), tetrahydrocurcumin (THC), hexahydrocurcumin (HHC), octahydrocurcumin (OHC), curcumin glucuronide, and curcumin sulphate [8]. Curcumin has been speculated to have promising chemotherapeutic and preventive activities, which could approve avenues for alternative treatments for many diseases. Recently, much attention has been directed to study the medical applications of curcumin in the treatment of human cancers, since curcumin has been shown to exhibit antitumor and apoptosis activities in a wide spectrum of human cancer cell lines [6], that is aside its prospective role as a potential immunomodulatory effector both *in vivo* and *in vitro* studies [9–11]. Currently, several clinical trials have been applied curcumin for treatment of pancreatic cancer [12], multiple myeloma [13, 14], Alzheimer's [15], and colorectal cancer [16]. Curcumin is also known to activate and regulate dendritic cells and inhibit IL-1, IL-6, and TNF-α along with inhibition of NF-κB activation [17]. Curcumin could prevent production of interleukin-8 (IL-8), monocyte inflammatory protein-1 (MIP-1α), monocyte chemotactic protein-1 (MCP-1), IL-1β, tumor necrosis factor-α (TNF-α), 4-β-phorbor-12-β-myristate-13-α acetate (PMA), or lipopolysaccharide (LPS)-stimulated monocytes and macrophages [18]. The exact molecular mechanisms of curcumin-induced apoptosis in cancer cells are varied and depend on the cell type and dose used in the experiments [19, 20].

Cell Cycle Arrest

The cell cycle, arranged in the following phases, leads to cell growth and division:

- In G_1 phase, the cell grows and chromosomes prepare for replication.
- In S phase, DNA replicates and chromosomes duplicate.
- G_2 phase represents the gap between DNA synthesis and mitosis.
- In M phase (mitosis), nuclear and cytoplasmic division occurs, yielding two daughter cells.

Curcumin was found to induce cell cycle arrest at the G0/G1 phase in leukemic cells [21] and S and G2/M phases in breast cancer cells and human bladder cancer [22, 23]. The major cell-cycle proteins, cyclin-dependent kinase 1, 2, and 4 (CDK 1, 2, and 4), are considered potential molecular targets of curcumin [24]. Curcumin may inhibit the expression of cyclin D1 and CDK4 via acetylation and upregulation of p53, leading to cell cycle arrest at G1-S phase in cervical cancer [25]. Furthermore, curcumin acts as an ATP-competitive inhibitor; it downregulated the mRNA and the protein expression of cyclin D1 and suppressed transition of the cells from G1 to S phase, thus, prevents invasion of gastric cancer cells [26]. In human prostate cancer, curcumin mediates cycle arrest at G1/S phase through induction of CDK inhibitors p16(/INK4a), p21(/WAF1/CIP1), and p27(/KIP1), inhibition of cyclin E and cyclin D1 expression, and hyperphosphorylation of retinoblastoma (Rb) protein, a well-known CDK2 substrate [27]. However, in HCT116 colon cancer cells, the expression levels of CDK2 and its regulatory subunit, cyclin E, were not changed, but the phosphorylation of Rb was declined by curcumin [28]. In Ras-activated cells, curcumin enhanced Erk1/2 expression and inhibited Akt and its downstream molecules (mTOR and S6K1), thus, promotes G2/M arrest, reflecting a potential role of Ras/Erk1/2 activation in curcumin-induced G2/M arrest [29]. Treatment of Jurkat cells with different doses of curcumin enhanced JNK and p-JNK expressions without affecting the expressions of ERK1/2 and P38 MAPK or the activity of MMP-2 and MMP-9. Thus, the activation of JNK pathway but not the MMPs is the potential mechanism which might involve in cell proliferation [30].

Induction of Apoptotic Signals

Apoptosis occurs through two major pathways, the extrinsic and intrinsic pathways. Intrinsic apoptosis operates by modulating of mitochondrial membrane potential ($\Delta\Psi m$) which releases cytochrome *c* and inhibits expression of antiapoptotic proteins Bcl-2 and Bcl-xL. Curcumin suppresses the Bcl-xL level, mediating imbalance in the mitochondrial membrane potential and enhancing cleavage of procaspases and poly(ADP-ribose) polymerase [31]. The extrinsic apoptosis operates through induction of TNF-related apoptosis and enhanced expression of death receptors and their downstream factors, such as DR4, DR5, tumor necrosis factor receptor apoptosis–inducing ligand (TRAIL), and Fas/FasL [32]. When the apoptotic signal is received, Fas-associated death domain (FADD) binds and recruits the death-induced signaling complex (DISC) forming initiator caspases 8 and 10 [33]. In TNF-related apoptosis inducing ligand (TRAIL)-resistant cell lines, curcumin enhanced apoptosis by upregulating the expression of DR 4 and 5 [34]. However, a recent study concluded that curcumin-induced production of reactive oxygen species did not affect total expression of DR5 but it enhanced mobilization of DR5 to the plasma membrane [35]. Curcumin induced upregulation of Fas, FasL, and DR5 expression [36] and inhibited TNF-α-induced activation of NF-κB, including NF-κB-P65, and thus mediates apoptosis [37]. Curcumin also inhibited the TNF-α-induced production of IL-6/IL-8 in HaCaT cells [37]. Curcumin could also induce apoptosis through p38-dependent upregulation of FasL in Huh7 cells [38]. The endoplasmic reticulum (ER) is pointed as a third subcellular compartment implicated in apoptotic execution [39, 40]. Gadd153, growth arrest and DNA damage-inducible protein, is activated by endoplasmic reticulum (ER) stress, resulting in the induction of oxidative stress, generation of reactive oxygen species (ROS), and disturbing iron homeostasis, which in turn mediates cellular-damage/apoptosis [41, 42]. Curcumin can mediate DNA damage whether through topoisomerase inhibition [43, 44] or via generation of ROS [45]. Like other proteasome inhibitors, curcumin upregulates the expression of the growth arrest- and DNA damage-inducible genes GADD45 and 153 [46–48].

Suppression of the Antiapoptotic Proteins

Curcumin decreases expression of the antiapoptotic Bcl-2 and Bcl-XL proteins in lung cancer and prostate cancer cell lines [49, 50]. However, an induction of expression of proapoptotic Bax protein was observed in Ehrlich's Ascites carcinoma upon curcumin treatment [51]. Bush and colleagues (2001), reported suppression of X-linked inhibitors of apoptosis (XIAP) protein, a caspase inhibitor, in human melanoma cells in response to curcumin treatment [52]. Several studies showed that curcumin is able to break the apoptosis resistance in multidrug-resistant cancer cells by modulating the expression of many resistance-associated proteins [53, 54].

Modulation of MicroRNAs (miRNAs)

miRNAs are a class of noncoding RNAs that post-transcriptionally inhibit gene expression, and their role in carcinogenesis as tumor suppressors or oncogenes has been widely reported [55]. In MCF-7 and leukemia cells, curcumin upregulating the expression of a miR-15 and miR-16-mediated downregulation of bcl-2 induced apoptosis [56–58], suggesting that the miR-15a/16 family can potentially serve as potential gene targets for bcl-2 overexpressing cancer cells. Curcumin could promote the apoptosis of A549/DDP cells through regulating the expression of miR-186 [59]. Further studies showed that curcumin altered miRNA expression, in particular, significantly downregulated the expression of miR-186 in A549/DDP [60]. miR-22, a tumor-suppressor miRNA, was one of the miRNA which was upregulated by curcumin [61]. Last but not the least, curcumin modulates miRNAs (miR-15a, miR-16, miR-21, miR-22, miR-26, miR-101, miR-146, miR-200, miR-203, and let-7) and their multiple target genes [62].

Modulation of Wnt/β-Catenin Signaling

Wnt/β-catenin signaling regulates the proliferation and differentiation of many normal and malignant cells. A number of studies have suggested that curcumin has the potential to target cancer stem cells (CSCs) through regulation of CSC self-renewal pathways including Wnt/β-catenin pathway [63]. Furthermore, curcumin treatment has found to activate GSK-3β and reduce expression of β-catenin and its downstream target cyclin D1 [64]. Curcumin as a Wnt inhibitor targets downstream β-catenin activity and effectively represses HBx-mediated regulation of c-MYC and E-cadherin [65]. Curcumin also modulates the Wnt/β-catenin signaling pathway and, thus, could inhibit LNCaP prostate cancer cells activity [66]. The expression levels of two integral proteins of Wnt signaling, GSK3β, and E-cadherin were also altered by curcumin treatment, in human breast cancer cells [67].

Modulation of Protein Kinases

It is considered that PKC, mTOR, and EGFR tyrosine kinase are the major upstream molecular targets for curcumin intervention, whereas the nuclear oncogenes such as c-jun, c-fos, c-myc, CDKs, FAS, and iNOS might act as downstream molecular targets for curcumin actions [68]. The oxidant tumor promoter TPA activates PKC by reacting with zinc thiolates present within the regulatory domain, while the oxidized form of cancer chemopreventive agent such as curcumin can inactivate PKC by oxidizing the vicinal thiols present within the catalytic domain [69]. Due to its ability to inhibit Src, JNK, and Smad3 phosphorylations, curcumin abrogates the TGFβ1-induced tissues overgrowth [70]. Curcumin directly induces a tumor-suppressive miR-203-mediated regulation of the Src-Akt axis in bladder cancer [71]. The downregulation of miR-203 may be due to DNA hypermethylation of its promoter. Another potential mechanism is that curcumin inhibits phosphorylation of Src and stat3 partly through regenerating liver-3 (PRL-3) downregulation [72]. Curcumin may play its anticancer actions partly via suppressing PI3K/Akt signal transduction pathway in several tumor models [73–75]. Curcumin significantly inhibited NF-κB and attenuated the effect of irradiation-induced prosurvival signaling through the PI3K/Akt/mTOR and NF-κB pathways in these gut-specific endothelial cells [76].

Inhibition of Nuclear Factor-κB (NF-κB)

NF-κB is synthesized in the cytoplasm and complexed with its inhibitor I-κB; thus, NF-κB is released in an activate form. In order to activate, I-κB must be phosphorylated followed by a proteasomal degradation of the NF-κB/p-κB complex. The free p-NF-κB then translocates to the nucleus to transcribe and activate genes to synthesize progrowth and antiapoptosis [77]. Treatment with curcumin and resveratrol suppressed NF-κB-regulated gene products involved in inflammation (cyclooxygenase-2, matrix metalloproteinase (MMP)-3, MMP-9, vascular endothelial growth factor), inhibited apoptosis (Bcl-2, Bcl-xL, and TNF-α receptor-associated factor 1), and prevented activation of caspase-3. Curcumin inhibits the activation of NF-κB and the expressions of oncogenes including c-jun, c-fos, c-myc, NIK, MAPKs, ERK, ELK, PI3K, Akt, CDKs, and iNOS [69]. Curcumin prevents the entry of NF-κB into the nucleus thereby decreasing the expression of cell cycle regulatory proteins and survival factors such as Bcl-2 and surviving [24]. The long-term effect of curcumin may contribute to attenuate cancer progression via the downregulation of TNF-α and IL-6 modulated by E26 transformation-specific protein (ETS) and nuclear factor-κB (NF-κB). That is may through inhibition binding of nuclear protein with ETS and NF-κB binding elements of TNF-α and IL-6 promoters, respectively [78]. TNF-induced NF-κB-regulated gene products involved in cellular proliferation COX-2, cyclin D1, and c-myc, antiapoptosis including inhibitor of apoptosis protein (IAP)1, IAP2, X-chromosome-linked IAP, Bcl-2, Bcl-x(L), and metastasis (vascular endothelial growth factor, matrix metalloproteinase-9, and intercellular adhesion molecule-1) were also downregulated by curcumin [79].

Modulation of Proteasome Activation

Curcumin-induced apoptosis is mediated through the impairment of ubiquitin proteasome system (UPS) [46]. Curcumin can inhibit activation of the proinflammatory transcription factor NF-κB by inhibiting the 26S proteasomal degradation of IκBα, an inhibitor of NF-κB [80]. Furthermore, the inhibition of the 26S proteasomal activity by curcumin is mediated through α,β-unsaturated ketone and two sterically accessible β-carbons. On the other hand, curcumin, a potent inhibitor of the JNK-AP-1 pathway, abrogated the induction of monocyte chemoattractant protein 1 (MCP-1) by MG132, a proteasome inhibitor. The transcriptional activation by proteasome inhibitors was observed not only in MCP-1, but also in other AP-1-dependent genes, including stromelysin and mitogen-activated protein kinase phosphatase 1 [81]. These data revealed that curcumin-mediated proteasome inhibition triggered the expression of MCP-1 and other genes via the multisteps induction of the JNK-c-Jun/AP-1 pathway.

Epigenetic Regulation

In recent years, researchers have extensively documented that epigenetic mechanisms such as DNA methylation and histone modifications regulate many cancer cells activities, and thus, epigenetic regulation has been postulated as an attractive target for the cancer therapeutics [82]. In fact, the human genome has four DNA methyltransferase (*DNMT*) genes, which encoded proteins with distinct functions [83]. The new advantage of epigenetic modifications is that they can be modulated by treatment with HDAC (histone deacetylase) and DNMT (DNA methyltransferase) inhibitors, some of which have already been approved by the FDA for the treatment of myelodysplastic syndromes and acute myeloid leukemia [84]. The U.S. Food and Drug Administration has already approved some HDAC and DNMT inhibitors, such as azanucleoside drugs to treat myelodysplastic syndromes and acute myeloid leukemia [85]. By taking advantage of epigenetic modifications, we can use HDAC and DNMT inhibitors to control various cancer cell activities. Histone tails and their modifications regulate diverse biological processes such as transcription, DNA repair, recombination, cell division and differentiation etc. [86–88]. It is also possible that curcumin may disrupt some cellular processes that function parallel with histone modification. One possible mechanism by which curcumin might exert its numerous effects is through epigenetic modulation by targeting various epigenetic factors, such as HDAC, HAT, DNMTs, and miRNAs [89, 90] which regulates various cellular pathways. A recent study provided that curcumin mediates histone H3, H4, H2A N-terminal tail modification leading to a significant growth inhibition, which suggests that curcumin mediates its action through the N-terminal tail regions of histones [91]. Curcumin is found to reduce the expression of positive regulators of DNA methyltransferase 1 (DNMT1), p65 and Sp1, which correlated with a reduction in binding of these transcription factors to the DNMT1 promoter in acute myeloid leukemia cell lines [85]. DNMT1 catalyzes the transfer of methyl groups to DNA, which represents a crucial mediator of DNA methylation. Exposure of MCF7 cells to curcumin resulted in increased global levels of acetylated H3K18 and H4K16 and was less effective in inducing DNA damage markers accompanied by upregulation of DNA damage signalling markers such as γH2A.X and H3K56Ac [92].

Reverse the Multidrug Resistance of Cancer Cells

Multidrug resistance (MDR) is an obstacle in cancer treatment, often because less drug accumulates in patient's tumor cells owing to enhanced drug efflux [93]. P-glycoprotein (P-gp) is an ATP-dependent drug efflux pump and major player in the development of resistance in cancer cells. Curcumin is found to reverse the MDR of the human gastric carcinoma SGC7901/vincristine cell line [94]. That was associated with decreased P-gp function and expression, and the promotion of caspase-3 activation in MDR cells. Similarly, treatment of drug-resistant KB-V1 cells with curcumin increased their sensitivity to vinblastine that was associated with a decreased amount of P-glycoprotein on the cell plasma membrane [93]. Curcumin can partially reverse the paclitaxel-resistance of SKOV3-TR30 cells through a downregulation of glycogen synthase kinase-3 (GSK-3) [95]. In a short cohort open-labeled study, curcumin decreased multidrug resistance 1 (MDR1) mRNA level in patient leukemic cells, especially in high level of MDR1 gene groups [96]. Thus, curcumin treatment may provide a lead for clinical treatment of leukemia patients in the future.

The Flip Side of Curcumin

Despite its utilization and effectiveness in clinical trials for cancer as shown above, the genotoxicity, poor water solubility, and rapid intestinal and hepatic metabolism of curcumin are the major challenges limiting its clinical utilizes. These challenges represent that the dark side of curcumin is not demonstrated here in detail because of space limitations, and thus, readers are referred to the excellent review of Burgos-Moron et al. (2010) [97]. However, to summarize briefly, unlike the common belief in the scientific literature that there is no major toxicity that has been found in humans, curcumin

is not approved as a safe agent, since such belief was established based on short-term studies. Additionally, numerous analogues of curcumin are synthesized and tested for their bioavailability, selectivity, and stability and it showed encouraging results in cancer treatment [98–100]. A mass of accumulated studies confirmed that curcumin possesses DNA damage and chromosomal alterations both *in vitro* and *in vivo* at concentrations similar to those reported to exert beneficial effect [101, 102]. The increased incidence of carcinomas of the small intestine was observed in mice taking average daily doses of curcumin of about 0.2 mg kg^{-1} body weight [103, 104]. Moreover, a recent study has also shown that curcumin can mediate lung cancer in mice [105]. Several approaches were established to enhance curcumin's bioavailability, the plasma concentration, and the cellular permeability. The black pepper alkaloid piperine (bioperine) was used in order to increase the bioavailability of curcumin [106]. However, such strategy should be used cautiously, as piperine is a potent inhibitor of drug metabolism and may cause toxicity in people taking specific drugs [107, 108]. Nanoparticles as a new approach for targeting drug delivery have tested and proven for providing curcumin with longer circulation, better permeability, and stronger resistance to metabolic processes. It is important to take into account that any strategy that increases the levels of curcumin in tissues will not only increase the effectiveness of curcumin, but also its toxicity. Overcoming these problems represents a validated and effective therapeutic tool in the battle against cancer.

Conclusion

Curcumin is a potent cancer fighter; through several mechanisms, it can kill a wide range of tumor cell types including MDR cancer cells. Notably, due to the diversity of cell death mechanisms mediated by curcumin, few cancer cells can develop resistance to curcumin-induced cell death. Urgently, clinical trials are required to place this fascinating molecule at the fore front of novel therapeutics and translate this laboratory concept into clinical benefits.

* * *

Funding sources: Author declares no grant has received, no a commercial interest or other relationship with manufacturers of pharmaceuticals, laboratory supplies, and/or medical devices or with commercial providers of related medical services.

Conflict of interest: The author declares no conflict of interest.

References

1. Rice MC, Bockman J: Anticancer drug discovery and development – SRI's seventh annual summit. IDrugs 8(10), 805–808 (2005)
2. Mans DR, da Rocha AB, Schwartsmann G: Anti-cancer drug discovery and development in Brazil: targeted plant collection as a rational strategy to acquire candidate anti-cancer compounds. Oncologist 5(3), 185–198 (2000)
3. Rengarajan T, Nandakumar N, Rajendran P, Haribabu L, Nishigaki I, Balasubramanian MP: D-pinitol promotes apoptosis in MCF-7 cells via induction of p53 and Bax and inhibition of Bcl-2 and NF-kappaB. Asian Pac J Cancer Prev 15(4), 1757–1762 (2014)
4. Montaser R, Luesch H: Marine natural products: a new wave of drugs? Future Med Chem 3(12), 1475–1489 (2011)
5. Ortholand JY, Ganesan A: Natural products and combinatorial chemistry: back to the future. Curr Opin Chem Biol 8(3), 271–280 (2004)
6. Huang AC, Lin SY, Su CC, Lin SS, Ho CC, Hsia TC, Yu CS, Ip SW, Lin TP, Chung JG: Effects of curcumin on N-bis(2-hydroxypropyl) nitrosamine (DHPN)-induced lung and liver tumorigenesis in BALB/c mice *in vivo*. In Vivo 22(6), 781–785 (2008)
7. Kim YS, Ahn Y, Hong MH, Joo SY, Kim KH, Sohn IS, Park HW, Hong YJ, Kim JH, Kim W, Jeong MH, Cho JG, Park JC, Kang JC: Curcumin attenuates inflammatory responses of TNF-alpha-stimulated human endothelial cells. J Cardiovasc Pharmacol 50(1), 41–49 (2007)
8. Anand P, Thomas SG, Kunnumakkara AB, Sundaram C, Harikumar KB, Sung B, Tharakan ST, Misra K, Priyadarsini IK, Rajasekharan KN, Aggarwal BB: Biological activities of curcumin and its analogues (Congeners) made by man and Mother Nature. Biochem Pharmacol 76(11), 1590–1611 (2008)
9. Churchill M, Chadburn A, Bilinski RT, Bertagnolli MM: Inhibition of intestinal tumors by curcumin is associated with changes in the intestinal immune cell profile. J Surg Res 89(2), 169–175 (2000)
10. Kurup VP, Barrios CS, Raju R, Johnson BD, Levy MB, Fink JN: Immune response modulation by curcumin in a latex allergy model. Clin Mol Allergy 5, 1 (2007)
11. Varalakshmi C, Ali AM, Pardhasaradhi BV, Srivastava RM, Singh S, Khar A: Immunomodulatory effects of curcumin: *in-vivo*. Int Immunopharmacol 8(5), 688–700 (2008)
12. Swamy MV, Citineni B, Patlolla JM, Mohammed A, Zhang Y, Rao CV: Prevention and treatment of pancreatic cancer by curcumin in combination with omega-3 fatty acids. Nutr Cancer 60(Suppl 1), 81–89 (2008)
13. Milacic V, Banerjee S, Landis-Piwowar KR, Sarkar FH, Majumdar AP, Dou QP: Curcumin inhibits the proteasome activity in human colon cancer cells *in vitro* and *in vivo*. Cancer Res 68(18), 7283–7292 (2008)
14. Jiao Y, Wilkinson Jt, Di X, Wang W, Hatcher H, Kock ND, D'Agostino R Jr, Knovich MA, Torti FM, Torti SV: Curcumin, a cancer chemopreventive and chemotherapeutic agent, is a biologically active iron chelator. Blood 113(2), 462–469 (2009)
15. Wang Q, Sun AY, Simonyi A, Jensen MD, Shelat PB, Rottinghaus GE, MacDonald RS, Miller DK, Lubahn DE, Weisman GA, Sun GY: Neuroprotective mechanisms of curcumin against cerebral ischemia-induced neuronal apoptosis and behavioral deficits. J Neurosci Res 82(1), 138–148 (2005)
16. Half E, Arber N: Colon cancer: preventive agents and the present status of chemoprevention. Expert Opin Pharmacother 10(2), 211–219 (2009)
17. Vojdani A, Erde J: Regulatory T cells, a potent immunoregulatory target for CAM researchers: modulating tumor immunity, autoimmunity and alloreactive immunity (III). Evid Based Complement Alternat Med 3(3), 309–316 (2006)
18. Abe Y, Hashimoto S, Horie T: Curcumin inhibition of inflammatory cytokine production by human peripheral blood monocytes and alveolar macrophages. Pharmacol Res 39(1), 41–47 (1999)
19. Karunagaran D, Rashmi R, Kumar TR: Induction of apoptosis by curcumin and its implications for cancer therapy. Curr Cancer Drug Targets 5(2), 117–129 (2005)

20. Duvoix A, Blasius R, Delhalle S, Schnekenburger M, Morceau F, Henry E, Dicato M, Diederich M: Chemopreventive and therapeutic effects of curcumin. Cancer Lett 223(2), 181–190 (2005)
21. Tima S, Ichikawa H, Ampasavate C, Okonogi S, Anuchapreeda S: Inhibitory effect of turmeric curcuminoids on FLT3 expression and cell cycle arrest in the FLT3-overexpressing EoL-1 leukemic cell line. J Nat Prod 77(4), 948–954 (2014)
22. Liu HS, Ke CS, Cheng HC, Huang CY, Su CL: Curcumin-induced mitotic spindle defect and cell cycle arrest in human bladder cancer cells occurs partly through inhibition of aurora A. Mol Pharmacol 80(4), 638–646 (2011)
23. Ke CS, Liu HS, Yen CH, Huang GC, Cheng HC, Huang CY, Su CL: Curcumin-induced Aurora-A suppression not only causes mitotic defect and cell cycle arrest but also alters chemosensitivity to anticancer drugs. J Nutr Biochem 25(5), 526–539 (2014)
24. Kuttan G, Kumar KB, Guruvayoorappan C, Kuttan R: Antitumor, anti-invasion, and antimetastatic effects of curcumin. Adv Exp Med Biol 595, 173–184 (2007)
25. Roy M, Mukherjee S: Reversal of resistance towards cisplatin by curcumin in cervical cancer cells. Asian Pac J Cancer Prev 15(3), 1403–1410 (2014)
26. Cai XZ, Wang J, Li XD, Wang GL, Liu FN, Cheng MS, Li F: Curcumin suppresses proliferation and invasion in human gastric cancer cells by downregulation of PAK1 activity and cyclin D1 expression. Cancer Biol Ther 8(14), 1360–1368 (2009)
27. Srivastava RK, Chen Q, Siddiqui I, Sarva K, Shankar S: Linkage of curcumin-induced cell cycle arrest and apoptosis by cyclin-dependent kinase inhibitor p21(/WAF1/CIP1). Cell Cycle 6(23), 2953–2961 (2007)
28. Lim TG, Lee SY, Huang Z, Lim do Y, Chen H, Jung SK, Bode AM, Lee KW, Dong Z: Curcumin suppresses proliferation of colon cancer cells by targeting CDK2. Cancer Prev Res (Phila) 7(4), 466–474 (2014)
29. Ono M, Higuchi T, Takeshima M, Chen C, Nakano S: Differential anti-tumor activities of curcumin against Ras- and Src-activated human adenocarcinoma cells. Biochem Biophys Res Commun 436(2), 186–191 (2013)
30. Zhu G, Zhang Q, Dai H, Shen Q: Effect of curcumin on expressions of mitogen-activated protein kinases and matrix metalloproteinases in Jurkat cells. Nan Fang Yi Ke Da Xue Xue Bao 33(12), 1792–1795 (2013)
31. Balasubramanian S, Eckert RL: Curcumin suppresses AP1 transcription factor-dependent differentiation and activates apoptosis in human epidermal keratinocytes. J Biol Chem 282(9), 6707–6715 (2007)
32. Ashour AA, Abdel-Aziz AA, Mansour AM, Alpay SN, Huo L, Ozpolat B: Targeting elongation factor-2 kinase (eEF-2K) induces apoptosis in human pancreatic cancer cells. Apoptosis 19(1), 241–258 (2014)
33. Park W, Amin AR, Chen ZG, Shin DM: New perspectives of curcumin in cancer prevention. Cancer Prev Res (Phila) 6(5), 387–400 (2013)
34. Moragoda L, Jaszewski R, Majumdar AP: Curcumin induced modulation of cell cycle and apoptosis in gastric and colon cancer cells. Anticancer Res 21(2A), 873–378 (2001)
35. Park S, Cho DH, Andera L, Suh N, Kim I: Curcumin enhances TRAIL-induced apoptosis of breast cancer cells by regulating apoptosis-related proteins. Mol Cell Biochem 383(1–2), 39–48 (2013)
36. Lee HP, Li TM, Tsao JY, Fong YC, Tang CH: Curcumin induces cell apoptosis in human chondrosarcoma through extrinsic death receptor pathway. Int Immunopharmacol 13(2), 163–169 (2012)
37. Sun J, Han J, Zhao Y, Zhu Q, Hu J: Curcumin induces apoptosis in tumor necrosis factor-alpha-treated HaCaT cells. Int Immunopharmacol 13(2), 170–174 (2012)
38. Wang WZ, Li L, Liu MY, Jin XB, Mao JW, Pu QH, Meng MJ, Chen XG, Zhu JY: Curcumin induces FasL-related apoptosis through p38 activation in human hepatocellular carcinoma Huh7 cells. Life Sci 92(6–7), 352–358 (2013)
39. Rao RV, Ellerby HM, Bredesen DE: Coupling endoplasmic reticulum stress to the cell death program. Cell Death Differ 11(4), 372–380 (2004)
40. Nakagawa T, Zhu H, Morishima N, Li E, Xu J, Yankner BA, Yuan J: Caspase-12 mediates endoplasmic-reticulum-specific apoptosis and cytotoxicity by amyloid-beta. Nature 403(6765), 98–103 (2000)
41. McCullough KD, Martindale JL, Klotz LO, Aw TY, Holbrook NJ: Gadd153 sensitizes cells to endoplasmic reticulum stress by down-regulating Bcl2 and perturbing the cellular redox state. Mol Cell Biol 21(4), 1249–1259 (2001)
42. Vecchi C, Montosi G, Zhang K, Lamberti I, Duncan SA, Kaufman RJ, Pietrangelo A: ER stress controls iron metabolism through induction of hepcidin. Science 325(5942), 877–880 (2009)
43. Snyder RD, Arnone MR: Putative identification of functional interactions between DNA intercalating agents and topoisomerase II using the V79 *in vitro* micronucleus assay. Mutat Res 503(1–2), 21–35 (2002)
44. Martin-Cordero C, Lopez-Lazaro M, Galvez M, Ayuso MJ: Curcumin as a DNA topoisomerase II poison. J Enzyme Inhib Med Chem 18(6), 505–509 (2003)
45. Yoshino M, Haneda M, Naruse M, Htay HH, Tsubouchi R, Qiao SL, Li WH, Murakami K, Yokochi T: Prooxidant activity of curcumin: copper-dependent formation of 8-hydroxy-2′-deoxyguanosine in DNA and induction of apoptotic cell death. Toxicol In Vitro 18(6), 783–789 (2004)
46. Dikshit P, Goswami A, Mishra A, Chatterjee M, Jana NR: Curcumin induces stress response, neurite outgrowth and prevent NF-kappaB activation by inhibiting the proteasome function. Neurotox Res 9(1), 29–37 (2006)
47. Saha A, Kuzuhara T, Echigo N, Fujii A, Suganuma M, Fujiki H: Apoptosis of human lung cancer cells by curcumin mediated through up-regulation of "growth arrest and DNA damage inducible genes 45 and 153". Biol Pharm Bull 33(8), 1291–1299 (2010)
48. Wu SH, Hang LW, Yang JS, Chen HY, Lin HY, Chiang JH, Lu CC, Yang JL, Lai TY, Ko YC, Chung JG: Curcumin induces apoptosis in human non-small cell lung cancer NCI-H460 cells through ER stress and caspase cascade- and mitochondria-dependent pathways. Anticancer Res 30(6), 2125–2133 (2010)
49. Mukhopadhyay A, Bueso-Ramos C, Chatterjee D, Pantazis P, Aggarwal BB: Curcumin downregulates cell survival mechanisms in human prostate cancer cell lines. Oncogene 20(52), 7597–7609 (2001)
50. Radhakrishna Pillai G, Srivastava AS, Hassanein TI, Chauhan DP, Carrier E: Induction of apoptosis in human lung cancer cells by curcumin. Cancer Lett 208(2), 163–170 (2004)
51. Pal S, Choudhuri T, Chattopadhyay S, Bhattacharya A, Datta GK, Das T, Sa G: Mechanisms of curcumin-induced apoptosis of Ehrlich's ascites carcinoma cells. Biochem Biophys Res Commun 288(3), 658–665 (2001)
52. Bush JA, Cheung KJ, Jr., Li G: Curcumin induces apoptosis in human melanoma cells through a Fas receptor/caspase-8 pathway independent of p53. Exp Cell Res 271(2), 305–314 (2001)
53. Bielak-Mijewska A, Piwocka K, Magalska A, Sikora E: P-glycoprotein expression does not change the apoptotic pathway induced by curcumin in HL-60 cells. Cancer Chemother Pharmacol 53(2), 179–185 (2004)
54. Piwocka K, Bielak-Mijewska A, Sikora E: Curcumin induces caspase-3-independent apoptosis in human multidrug-resistant cells. Ann N Y Acad Sci 973, 250–254 (2002)
55. Seol HS, Akiyama Y, Shimada S, Lee HJ, Kim TI, Chun SM, Singh SR, Jang SJ: Epigenetic silencing of microRNA-373 to epithelial-mesenchymal transition in non-small cell lung cancer through IRAK2 and LAMP1 axes. Cancer Lett 353, 232–241 (2014)

56. Cimmino A, Calin GA, Fabbri M, Iorio MV, Ferracin M, Shimizu M, Wojcik SE, Aqeilan RI, Zupo S, Dono M, Rassenti L, Alder H, Volinia S, Liu CG, Kipps TJ, Negrini M, Croce CM.: miR-15 and miR-16 induce apoptosis by targeting BCL2. Proc Natl Acad Sci U S A 102(39), 13944–13949 (2005)
57. Yang J, Cao Y, Sun J, Zhang Y: Curcumin reduces the expression of Bcl-2 by upregulating miR-15a and miR-16 in MCF-7 cells. Med Oncol 27(4), 1114–1118 (2010)
58. Gao SM, Yang JJ, Chen CQ, Chen JJ, Ye LP, Wang LY, Wu JB, Xing CY, Yu K: Pure curcumin decreases the expression of WT1 by upregulation of miR-15a and miR-16-1 in leukemic cells. J Exp Clin Cancer Res 31, 27 (2012)
59. Tang N, Zhang J, Du Y: Curcumin promoted the apoptosis of cisplain-resistant human lung carcinoma cells A549/DDP through down-regulating miR-186*. Zhongguo Fei Ai Za Zhi 13(4), 301–306 (2010)
60. Zhang J, Du Y, Wu C, Ren X, Ti X, Shi J, Zhao F, Yin H: Curcumin promotes apoptosis in human lung adenocarcinoma cells through miR-186* signaling pathway. Oncol Rep 24(5), 1217–1223 (2010)
61. Sreenivasan S, Thirumalai K, Danda R, Krishnakumar S: Effect of curcumin on miRNA expression in human Y79 retinoblastoma cells. Curr Eye Res 37(5), 421–428 (2012)
62. Teiten MH, Dicato M, Diederich M: Curcumin as a regulator of epigenetic events. Mol Nutr Food Res 57(9), 1619–1629 (2013)
63. Li Y, Zhang T: Targeting cancer stem cells by curcumin and clinical applications. Cancer Lett 346(2), 197–205 (2014)
64. He M, Li Y, Zhang L, Li L, Shen Y, Lin L, Zheng W, Chen L, Bian X, Ng HK, Tang L: Curcumin suppresses cell proliferation through inhibition of the Wnt/beta-catenin signaling pathway in medulloblastoma. Oncol Rep 32(1), 173–180 (2014)
65. Hsieh A, Kim HS, Lim SO, Yu DY, Jung G: Hepatitis B viral X protein interacts with tumor suppressor adenomatous polyposis coli to activate Wnt/beta-catenin signaling. Cancer Lett 300(2), 162–172 (2011)
66. Choi HY, Lim JE, Hong JH: Curcumin interrupts the interaction between the androgen receptor and Wnt/beta-catenin signaling pathway in LNCaP prostate cancer cells. Prostate Cancer Prostatic Dis 13(4), 343–349 (2010)
67. Prasad CP, Rath G, Mathur S, Bhatnagar D, Ralhan R: Potent growth suppressive activity of curcumin in human breast cancer cells: modulation of Wnt/beta-catenin signaling. Chem Biol Interact 181(2), 263–271 (2009)
68. Lin JK: Molecular targets of curcumin. Adv Exp Med Biol 595, 227–243 (2007)
69. Lin JK: Suppression of protein kinase C and nuclear oncogene expression as possible action mechanisms of cancer chemoprevention by curcumin. Arch Pharm Res 27(7), 683–692 (2004)
70. Yang WH, Kuo MY, Liu CM, Deng YT, Chang HH, Chang JZ: Curcumin inhibits TGFbeta1-induced CCN2 via Src, JNK, and Smad3 in gingiva. J Dent Res 92(7), 629–634 (2013)
71. Saini S, Arora S, Majid S, Shahryari V, Chen Y, Deng G, Yamamura S, Ueno K, Dahiya R: Curcumin modulates microRNA-203-mediated regulation of the Src-Akt axis in bladder cancer. Cancer Prev Res (Phila) 4(10), 1698–1709 (2011)
72. Wang L, Shen Y, Song R, Sun Y, Xu J, Xu Q: An anticancer effect of curcumin mediated by down-regulating phosphatase of regenerating liver-3 expression on highly metastatic melanoma cells. Mol Pharmacol 76(6), 1238–1245 (2009)
73. Jiang QG, Li TY, Liu DN, Zhang HT: PI3K/Akt pathway involving into apoptosis and invasion in human colon cancer cells LoVo. Mol Biol Rep 41(5), 3359–3367 (2014)
74. Qiao Q, Jiang Y, Li G: Inhibition of the PI3K/AKT-NF-kappaB pathway with curcumin enhanced radiation-induced apoptosis in human Burkitt's lymphoma. J Pharmacol Sci 121(4), 247–256 (2013)
75. Wang S, Yu S, Shi W, Ge L, Yu X, Fan J, Zhang J: Curcumin inhibits the migration and invasion of mouse hepatoma Hca-F cells through down-regulating caveolin-1 expression and epidermal growth factor receptor signaling. IUBMB Life 63(9), 775–782 (2011)
76. Rafiee P, Binion DG, Wellner M, Behmaram B, Floer M, Mitton E, Nie L, Zhang Z, Otterson MF: Modulatory effect of curcumin on survival of irradiated human intestinal microvascular endothelial cells: role of Akt/mTOR and NF-{kappa}B. Am J Physiol Gastrointest Liver Physiol 298(6), G865–877 (2010)
77. Pannaccione A, Secondo A, Scorziello A, Cali G, Taglialatela M, Annunziato L: Nuclear factor-kappaB activation by reactive oxygen species mediates voltage-gated K+ current enhancement by neurotoxic beta-amyloid peptides in nerve growth factor-differentiated PC-12 cells and hippocampal neurones. J Neurochem 94(3), 572–586 (2005)
78. Das L, Vinayak M: Long-term effect of curcumin down-regulates expression of tumor necrosis factor-alpha and interleukin-6 via modulation of E26 transformation-specific protein and nuclear factor-kappaB transcription factors in livers of lymphoma bearing mice. Leuk Lymphoma 55, 2627–2636 (2014)
79. Aggarwal S, Ichikawa H, Takada Y, Sandur SK, Shishodia S, Aggarwal BB: Curcumin (diferuloylmethane) down-regulates expression of cell proliferation and antiapoptotic and metastatic gene products through suppression of IkappaBalpha kinase and Akt activation. Mol Pharmacol 69(1), 195–206 (2006)
80. Hasima N, Aggarwal BB: Targeting proteasomal pathways by dietary curcumin for cancer prevention and treatment. Curr Med Chem 21(14), 1583–1594 (2014)
81. Nakayama K, Furusu A, Xu Q, Konta T, Kitamura M: Unexpected transcriptional induction of monocyte chemoattractant protein 1 by proteasome inhibition: involvement of the c-Jun N-terminal kinase-activator protein 1 pathway. J Immunol 167(3), 1145–1150 (2001)
82. Ptak C, Petronis A: Epigenetics and complex disease: from etiology to new therapeutics. Annu Rev Pharmacol Toxicol 48, 257–276 (2008)
83. Mirza S, Sharma G, Parshad R, Gupta SD, Pandya P, Ralhan R: Expression of DNA methyltransferases in breast cancer patients and to analyze the effect of natural compounds on DNA methyltransferases and associated proteins. J Breast Cancer 16(1), 23–31 (2013)
84. Garcia-Manero G, Fenaux P: Hypomethylating agents and other novel strategies in myelodysplastic syndromes. J Clin Oncol 29(5), 516–523 (2011)
85. Yu J, Peng Y, Wu LC, Xie Z, Deng Y, Hughes T, He S, Mo X, Chiu M, Wang QE, He X, Liu S, Grever MR, Chan KK, Liu Z: Curcumin down-regulates DNA methyltransferase 1 and plays an anti-leukemic role in acute myeloid leukemia. PLoS One 8(2), e55934 (2013)
86. Strahl BD, Allis CD: The language of covalent histone modifications. Nature 403(6765), 41–45 (2000)
87. Duncan EM, Muratore-Schroeder TL, Cook RG, Garcia BA, Shabanowitz J, Hunt DF, Allis CD: Cathepsin L proteolytically processes histone H3 during mouse embryonic stem cell differentiation. Cell 135(2), 284–294 (2008)
88. Scully R: A histone code for DNA repair. Nat Rev Mol Cell Biol 11(3), 164 (2010)
89. Reuter S, Gupta SC, Park B, Goel A, Aggarwal BB: Epigenetic changes induced by curcumin and other natural compounds. Genes Nutr 6(2), 93–108 (2011)
90. Balasubramanyam K, Varier RA, Altaf M, Swaminathan V, Siddappa NB, Ranga U, et al.: Curcumin, a novel p300/CREB-binding protein-specific inhibitor of acetyltransferase, represses the acetylation of histone/nonhistone proteins and histone acetyltransferase-dependent chromatin transcription. J Biol Chem 279(49), 51163–51171 (2004)

91. Azad GK, Singh V, Golla U, Tomar RS: Depletion of cellular iron by curcumin leads to alteration in histone acetylation and degradation of Sml1p in Saccharomyces cerevisiae. PLoS One 8(3), e59003 (2013)
92. Collins HM, Abdelghany MK, Messmer M, Yue B, Deeves SE, Kindle KB, Mantelingu K, Aslam A, Winkler GS, Kundu TK, Heery DM: Differential effects of garcinol and curcumin on histone and p53 modifications in tumour cells. BMC Cancer 13, 37 (2013)
93. Limtrakul P, Anuchapreeda S, Buddhasukh D: Modulation of human multidrug-resistance MDR-1 gene by natural curcuminoids. BMC Cancer 4, 13 (2004)
94. Tang XQ, Bi H, Feng JQ, Cao JG: Effect of curcumin on multidrug resistance in resistant human gastric carcinoma cell line SGC7901/VCR. Acta Pharmacol Sin 26(8), 1009–1016 (2005)
95. Qiu J, Fu YF, Cheng Q, Cheng XD, Xie X, Lu WG: Reversing paclitaxel-resistance of SKOV3-TR30 cell line by curcumin. Zhonghua Yi Xue Za Zhi 92(27), 1926–1928 (2012)
96. Anuchapreeda S, Thanarattanakorn P, Sittipreechacharn S, Tima S, Chanarat P, Limtrakul P: Inhibitory effect of curcumin on MDR1 gene expression in patient leukemic cells. Arch Pharm Res 29(10), 866–873 (2006)
97. Burgos-Moron E, Calderon-Montano JM, Salvador J, Robles A, Lopez-Lazaro M: The dark side of curcumin. Int J Cancer 126(7), 1771–1775 (2010)
98. Zhou DY, Ding N, Du ZY, Cui XX, Wang H, Wei XC, Conney AH, Zhang K, Zheng X: Curcumin analogues with high activity for inhibiting human prostate cancer cell growth and androgen receptor activation. Mol Med Rep 10(3), 1315–1322 (2014)
99. Chuprajob T, Changtam C, Chokchaisiri R, Chunglok W, Sornkaew N, Suksamrarn A: Synthesis, cytotoxicity against human oral cancer KB cells and structure-activity relationship studies of trienone analogues of curcuminoids. Bioorg Med Chem Lett 24(13), 2839–2844 (2014)
100. Ahsan MJ, Khalilullah H, Yasmin S, Jadav SS, Govindasamy J: Synthesis, characterisation, and *in vitro* anticancer activity of curcumin analogues bearing pyrazole/pyrimidine ring targeting EGFR tyrosine kinase. Biomed Res Int 2013, 239354 (2013)
101. Verschoyle RD, Steward WP, Gescher AJ: Putative cancer chemopreventive agents of dietary origin-how safe are they? Nutr Cancer 59(2), 152–162 (2007)
102. Cao J, Jia L, Zhou HM, Liu Y, Zhong LF: Mitochondrial and nuclear DNA damage induced by curcumin in human hepatoma G2 cells. Toxicol Sci 91(2), 476–483 (2006)
103. Dunnick JK, Nyska A: The toxicity and pathology of selected dietary herbal medicines. Toxicol Pathol 41(2), 374–386 (2013)
104. National Toxicology Program: NTP toxicology and carcinogenesis studies of turmeric oleoresin (CAS no. 8024-37-1) (major component 79%–85% curcumin, CAS no. 458-37-7) in F344/N rats and B6C3F1 mice (feed studies). Natl Toxicol Program Tech Rep Ser 427, 1–275 (1993)
105. Dance-Barnes ST, Kock ND, Moore JE, Lin EY, Mosley LJ, D'Agostino RB, Jr., McCoy TP, Townsend AJ, Miller MS: Lung tumor promotion by curcumin. Carcinogenesis 30(6), 1016–1023 (2009)
106. Shoba G, Joy D, Joseph T, Majeed M, Rajendran R, Srinivas PS: Influence of piperine on the pharmacokinetics of curcumin in animals and human volunteers. Planta Med 64(4), 353–356 (1998)
107. Mancuso C, Barone E: Curcumin in clinical practice: myth or reality? Trends Pharmacol Sci 30(7), 333–334 (2009)
108. Bhardwaj RK, Glaeser H, Becquemont L, Klotz U, Gupta SK, Fromm MF: Piperine, a major constituent of black pepper, inhibits human P-glycoprotein and CYP3A4. J Pharmacol Exp Ther 302(2), 645–650 (2002)