Curcumin in Treating Breast Cancer: A Review

Yiwei Wang^{1,2}, Jiayi Yu², Ran Cui², Jinjin Lin¹, and Xianting Ding¹

Abstract

Journal of Laboratory Automation 2016, Vol. 21(6) 723–731 © 2016 Society for Laboratory Automation and Screening DOI: 10.1177/2211068216655524 jala.sagepub.com

Breast cancer is among the most common malignant tumors. It is the second leading cause of cancer mortality among women in the United States. Curcumin, an active derivative from turmeric, has been reported to have anticancer and chemoprevention effects on breast cancer. Curcumin exerts its anticancer effect through a complicated molecular signaling network, involving proliferation, estrogen receptor (ER), and human epidermal growth factor receptor 2 (HER2) pathways. Experimental evidence has shown that curcumin also regulates apoptosis and cell phase–related genes and microRNA in breast cancer cells. Herein, we review the recent research efforts in understanding the molecular targets and anticancer mechanisms of curcumin in breast cancer.

Keywords

breast cancer, curcumin, natural compound, signaling pathways

Introduction

Breast cancer is the most commonly diagnosed malignant tumor and the second leading cause of cancer mortality among women in the United States.¹ About 70% of breast cancers are classified as estrogen receptor (ER) positive and could be treated with antiestrogens.^{2,3} Recently, growing evidence has indicated that the application of a single molecule can hardly suppress the cross talk and negative feedback loops in the complex cancer cellular networks.⁴ Therefore, drug combinations targeting different pathology signaling pathways have been considered as a major trend in drug design and discovery for killing endocrine-resistant breast cancer cells.⁵ Yet, both the research and clinical applications are still in great need of effective candidates for better treatment of breast cancer.

Curcumin is a hydrophobic polyphenol derived from turmeric, a traditional Indian spice⁶ (**Fig. 1**). Curcumin has been used as an ethnic drug for the treatment of diverse diseases. Particularly, curcumin has been recognized as an effective anticancer agent that regulates multiple intracellular signaling pathways, including transcription factors (e.g., STAT3, NF- κ B, and AP-1), receptors (e.g., IL-8, HER2, and CXCR4), kinases (e.g., EGFR, ERK, and JAK), cytokines (e.g., TNF, IL, and MIP), enzymes (e.g., MMP, iNOS, and GST), and growth factors (e.g., EGF, NGF, and HGF).⁷ Yet, the discussions on curcumin's anticancer effects have only been available in the last few decades. Sharma et al.⁸ succeeded in treating colorectal cancer with curcumin in phase I, and Cruz-Correa et al.⁹ used curcumin in the treatment of patients who were diagnosed with familial adenomatous polyposis. In an



Figure 1. Chemical structure of curcumin.

experimental investigation of mammary cancer induced by 7,12-dimethylbenzanthracene (DMBA), curcumin significantly decreased the initiation of mammary adenocarcinoma on the fourth day after DMBA administration.¹⁰ Furthermore, a large number of studies on cancer prevention at different stages have indicated curcumin as a favorable agent for cancer chemoprevention, used both alone and in combination with other drugs.^{11–13}

¹School of Biomedical Engineering, Institute for Personalized Medicine, Shanghai Jiao Tong University, Shanghai, China ²School of Medicine, Shanghai Jiao Tong University, Shanghai, China

Received March 30, 2016.

Corresponding Author:

Prof. Xianting Ding, School of Biomedical Engineering, Institute for Personalized Medicine, Shanghai Jiao Tong University, 1954 Huashan Road, Shanghai, 200030 China. Email: dingxianting@gmail.com Although healthy lifestyles, including balanced diets, regular exercise, alcohol reduction, smoking cessation, and stress management, have proved their benefits in preventing multiple kinds of cancer,^{14–18} the U.S. Food and Drug Administration (FDA) approval of tamoxifen, a selective estrogen receptor modulator (SERM), is considered to be a milestone in breast cancer chemoprevention.¹⁹ When a potential drug compound is under investigation, the utmost importance should always be

the safety issue. In this context, unlike synthetic compounds, natural products present in fruits and vegetables possess natural advantages. As such, the numerous effects of curcumin, including antioxidation and anti-inflammation, have called for extensive research efforts in recent years. In the following parts of this article, we review and summarize the recent publications in eight categories according to curcumin diversified functions on breast cancer.

Techniques for Extraction and Purification of Curcumin

Curcumin longa L. (turmeric) contains 2%-9% curcuminoids, including curcumin, demethoxycurcumin and bisdemethoxycurcumin, and cyclic curcumin. Among them, curcumin is considered the most important biomedical molecule.²⁰ Since 1815, when the first report of extraction and purification of curcumin was published, the curcumin extraction method has been continuously improving and advancing. Organic solvents, commonly reported for curcumin extraction, involve hexane, ethylacetate, acetone, and ethanol. Compared with zone-fining and dipping methods, ethanol extraction is a simpler process with a higher extraction ratio. A high extraction ratio also happens in Soxhlet extraction, ultrasonic extraction, and microwave extraction, which indicates that these techniques are suitable for industrial application.²¹⁻²⁶ Moreover, some articles showed that pulse ultrasonic and microwave-assisted extraction lead to a high curcumin ratio.²⁴ Another common method uses acid-alkali,²⁷ the principle of which is to dissolve the phenolic hydroxyl group of curcumin with sodium hydroxide at pH 7.0. This method could also separate the starch in the crude extraction product. There are also a few reports on compound enzyme extraction where curcumin is treated with enzymes such as cellulose enzyme and pectinase, which could degrade the cytoderm and intercellular substance.^{28,29} However, due to the cost and complexity of the process, the enzyme-assisted method has not been commercially adopted.

Among all the protocols, the primary recommendation for laboratory research is supercritical CO_2 fluid extraction (SFE). Fluid under the supercritical state with supercritical temperature and pressure is regarded as high density, with characteristics of both gas and liquid. The viscosity of supercritical fluid (SF) is similar to that of gas, while its Journal of Laboratory Automation 21(6)

 CO_2 is more frequently used than ethylene and ethane hexane methyl methane.^{30,31} The normal operating condition for this process is at a pressure of 25 MPa and a temperature of 318K, which requires 4 h of static extraction and 5 h of dynamic extraction with cosolvent of 30% ethyl alcohol in 3.5 L/min CO₂ flow.³²

Curcumin could be purified from crude curcumin (a mixture of curcuminoid) by column chromatography. It is a technology that uses different organics such as activated carbon, activated clay or silica with mixtures of solvent like dichloromethane/acetic acid or methanol/chloroform/dichloromethane, and ethanol/methanol mixtures as eluents to yield fractions.33-38 Among these, macroporous resin column chromatography is widely adopted. Macroporous resin was invented in 1964, with holes of 100-1000 nm distributed on the surface area. Combined with styrol and propionate, phenylethylene forms a porous polymer with a screening function.³⁹ A recent paper introduced a novel method for curcumin preconcentration. The molecularly imprinted polymers (MIPs) based on magnetic multiwalled carbon nanotubes possessed excellent selectivity toward curcumin. Other advanced techniques based on functional comonomers, such as thermoresponsive magnetic molecularly imprinted polymers (TMMIPs), have also been developed with high reproducibility and stability for selective curcumin extraction.40-42

Meanwhile, high-performance liquid chromatography (HPLC) is widely used for the detection and estimation of curcumin. Absorption detectors with solvent containing acetonitrile/water and chloroform/methanol at the mobile phase could detect curcumin at wavelengths in the range of 350–370 nm.^{35–} ³⁸ HPLC–diode array and fluorescence detection methods are used as another simple tool for detecting curcumin, with the most sensitive region in 390–450 nm. Some researchers have developed a high-performance thin-layer chromatography method, which is assisted with a phenyl column and acetonitrile/methanol/water as the solvent for curcumin detection and separation.³⁶ Microemulsion electrokinetic chromatography, employing oil droplets and surfactants, has proved useful in the extraction and estimation of curcumin.⁴³

Proliferation

In breast cancer cells, the survival signaling molecules, such as NF- κ B, play a pivotal role in cell proliferation⁴⁴ (**Fig. 2**). Liu et al. reported that curcumin was able to inhibit NF- κ B expression⁴⁵ and toggled many downstream signaling pathways, which silenced inflammatory cytokines, such as CXCL1 and CXCL2, and mediated the expression of matrix metalloproteinase 9 (MMP-9), urokinase plasminogen activator (uPA), uPA receptor (uPAR), intercellular adhesion molecule 1 (ICAM-1), and chemokine receptor 4



Figure 2. Molecular targets of curcumin on ROS, proliferation, apoptosis, and cell phase arrest. HER2, human epidermal growth factor receptor 2; MnSOD, manganese-dependent superoxide dismutase; GGTP, gamma-glutamyltranspeptidase; Nrf-2, nuclear factor 2–related factor; Fen I, Flap structure-specific endonuclease 1; CXCR4, chemokine receptor 4; uPAR, urokinase plasminogen activator receptor; FASN, fatty acid synthase; ACC, acetyl-CoA carboxylase; ICAM-1, intercellular adhesion molecule 1; uPA, urokinase plasminogen activator; MMP-9, matrix metalloproteinase 9; hTERT, telomerase reverse transcriptase; TRAIL, TNF-related apoptosis-inducing ligand.

(CXCR4).⁴⁶ Therefore, curcumin is believed to show its impact on cell growth and invasion of breast cancer partially through the downregulation of NF-κB signaling pathways.⁴⁷

Hormone factors also contribute to the growth and proliferation of breast cancer cells. HER2 and estrogen are the typical targets belonging to this category. Yim-im et al. found that a combination of curcumin and its analogues AS-KTC006 and AS-KTC021 could inhibit one specific tyrosine kinase and work as a rival against HER2.⁴⁸ Furthermore, curcumin can enhance the doxorubicin (a chemotherapeutic drug) cytotoxicity by decreasing redundancy HER2 in breast cancer cells.⁴⁹ On the other hand, immunoliposome encapsulation is believed to enhance curcumin selectivity in breast cancer cells that overexpress HER2.⁵⁰ Recent research proved that curcumin, acting as a phytoestrogen, competitively inhibited endogenous estrogen, which also contributed to the suppression of breast cancer cell growth.⁵¹

Furthermore, curcumin suppresses breast cancer proliferation partially due to other trophic signaling pathways. Curcumin showed its influence on fatty acid synthase (FASN) and acetyl-CoA carboxylase (ACC) through AMPK activation.⁵² Curcumin also partially inhibited breast cancer cell growth by downregulating the insulin-like growth factor 1 (IGF-1) axis.⁵³ Results from Thulasiraman et al.⁵⁴ demonstrated that curcumin mediated the suppression of the fatty acid–binding protein 5 (FABP5)/peroxisome proliferator-activated receptor β/δ (PPAR β/δ) pathway by making breast cancer cells more sensitive to retinotic acid. The same situation is applicable to leptin and its receptor.⁵⁵

Apoptosis

Curcumin is reported to induce breast cancer apoptosis by regulating the expression of apoptosis related genes. Lv et al.⁵⁶ applied microarray hybridization of lone-tech apoptotic arrays with labeled first-strand probes of total RNA to analyze and characterize the genes, which were regulated by curcumin in human breast cancer cells. In MCF-7 cells, genes *HIAP1*, *CRAF1*, *GADD45*, *HPRT*, *MCL-1*, *BCL2L2*, *NIP1*, *TRAP3*, *GSTP1*, *PIG11*, *DAXX*, *PIG3*, *RBP2*, and *JNK1* were upregulated, while genes TRAIL, *AP13*, *TNFR*, *SARP3*, *TRAIL-R2*, *TNFRSF5*, *TNFb*, and *hTRIP* were downregulated.

Curcumin was also reported to regulate the apoptosisrelated proteins. Exerting the antiapoptotic action by blocking pro-apoptotic counterparts, Bcl-2 inhibited the extrinsic apoptosis pathway.⁵⁷ Induced by curcumin, the antiapoptotic protein Bcl-2 increased, while the pro-apoptotic protein Bax decreased, leading to an elevated Bax/Bcl-2 ratio.^{58,59} Sun et al.⁶⁰ studied the curcumin treatment on triple-negative breast cancer (TNBC) and found that these cells significantly inhibited the phosphorylation levels of endothelial growth factor receptor (EGFR) and downstream signaling molecules, such as ERK1/2. Recent studies reported that curcumin enhanced the TNF-related apoptosis-inducing ligand (TRAIL)–induced apoptosis even in TRAIL-resistant breast cancer cells.^{61,62} Moreover, telomerase activity was inhibited by curcumin through downregulating the expression of hTERT (telomerase reverse transcriptase).⁶³

Cell Phase Arrest

Curcumin could also affect human breast cancer cells through inducing cell cycle arrest at the G2M phase and late S phase in MCF-7 cells. Curcumin led to an obvious increase of the G2M phase fraction.⁶⁴ Furthermore, Ke et al.⁶⁵ indicated that the percentage of cells arrested in the G2 phase and late S phase increased along with an increase of curcumin dose. Curcumin also acted similarly to Aurora-A SiRNA (small interfering RNA), leading to monopolar spindle formation, S and G2/M arrest, and cell division reduction in MCF-7 cells.⁶⁶

Curcumin induces cancer cell phase arrest through regulating spindle-related signaling pathways. A profound effect on mitotic spindle organization was exerted by curcumin directly, and the monopolar spindles were formed. Consequently, chromosomes were not able to be segregated normally and cells were arrested in the M phase.⁶⁷ Alternatively, curcumin also affected the expression levels of spindle motor protein indirectly, including the *Xenopus laevis*/Aurora/Ip11p-Related Kinase pEg2. Phosphorylation of Eg5 regulated the activity of the spindle motor protein, and downregulation of pEg2 resulted in monopolar spindles.⁶⁸

Modulation of MicroRNA

Curcumin suppresses breast cancer oncogenesis partially due to modulation of microRNA (miR), DNA, histone, and mitochrondria. The expression of miR-15a, miR-16, and miR-34a was upregulated in curcumin-treated cell models, and that of Bcl-2 and Bmi-1 was downregulated.69,70 Besides, recent evidence demonstrated that curcumin upregulated miR-22, which was conjectured to target estrogen receptor α (ER α) and the transcription factor Sp1, while miR-196 was dramatically downregulated.⁷¹ Also, curcumin mediated the downexpression of two cytokines, CXCL-1 and CXCL-2, by steering miR181b.72 Several papers revealed that DNA methyltransferase 1 (DNMT1), which epigenetically silenced multiple genes, decreased with curcumin treatment singly or in combination with other nature compounds.73,74 The same modulation happened upon acetylation of different histones, including H3K18, H4K16, and p53K120.75

Immune and Metastasis Factors

Curcumin suppresses the growth of breast cancer partially by mediating the immune system. Recent research has indicated that curcumin could prevent the loss of T cells and inhibit immune suppressive cytokines, including transforming growth factor beta (TGF- β) and interleukin 10 (IL-10) in carcinogenesis.^{76,77} Particularly, tumor exosome-mediated inhibition of nature killer cell augmentation was partially reversed by curcumin through the impairment of a ubiquitin–proteasome system.⁷⁸ Moreover, curcumin acted as an inhibitor of the MEK/ERK signaling pathway, thus preventing tumor-dependent TGF- β -induced T–regulatory cell proliferation.⁷⁹

Curcumin also suppresses the growth of breast cancer by affecting metastasis factors in two ways (Fig. 3). First, curcumin inhibited angiogenesis factors, such as vascular endothelial growth factor (VEGF) and basic fibroblast growth factor (bFGF), in ER-negative breast cancer cells.^{80,81} Meanwhile, Carroll et al.⁸² reported that curcumin could suppress the secretion of angiogenesis factors, as one complication of medroxyprogesterone acetate. Second, curcumin also affects metastasis factors through invasion. Curcumin not only limited the expression of adhesion molecules (such as integrin $\alpha 6\beta 4$) and the cytoskeleton protein (such as matrix metalloproteinase 9 and intercellular adhesion molecule 1), but also blocked recepteur d'origine nantais (RON) tyrosine kinase-mediated invasion.⁸²⁻⁸⁵ Moreover, it is certified that curcumin prevents hematogenous metastases in immunodeficient models.86

Reactive Oxidative Stress

Reactive oxidative stress (ROS) expresses double-sided effects on carcinogenesis. On the one hand, ROS triggers cancer by DNA mutations. On the other hand, ROS has also been reported in some literature to kill tumor cells. Minimizing DNA damage resulting by ROS was always a vital consideration in the treatment of neoplasm. Curcumin has shown its effect on downregulating Flap endonuclease 1 (Fen1) expression, the overexpression of which was involved in the development of breast cancer, by interfering with the level of nuclear factor 2–related factor (Nrf-2), thus preventing the growth of breast cancer.^{87–89} Furthermore, curcumin could potentially increase the scavenger elements, such as manganese superoxide dismutase (MnSOD) and catalase.⁹⁰

Curcumin could also induce breast cancer cell apoptosis by promoting ROS. Curcumin-induced ROS accumulation leads to P53/p21- and p16/Rb-mediated breast cancer inhibition.⁹¹ Moreover, Quiroga et al. detected a lower level of gamma-glutamyltranspeptidase (GGTP) activity even in an oxidation-resistant cell line treated with curcumin.⁹²



Figure 3. Molecular targets of curcumin on metastasis factors. VEGF, vascular endothelial growth factor; bFGF, basic fibroblast growth factor; ANG, angiopoietin; integrin $\alpha 6\beta 4$, adhesion molecules; MMP-9, matrix metalloproteinase 9; RON, recepteur d'origine nantais.

Oncogene and Tumor Suppressor Genes

In certain circumstances, oncogenes are activated and reprogram normal cells to cancer cells. Recently, Hong et al.⁹³ reported that the *erbB2/neu* (*HER2*) gene–encoded p185*neu* tyrosine kinase, the overexpression of which was assured to cause breast cancer, could be inhibited by curcumin. On the other hand, tumor suppressor genes prevent normal cells from being designated into cancer cells. Treatment of curcumin resulted in a high level of tumor suppressor genes, such as maspin and p16INK4A in invasive ductal carcinoma and cancer-associated myofibroblasts, respectively.^{93,94}

Current Trend in Curcumin Pharmacy

In spite of the anticancer and antiantigenic properties of curcumin, its low potency and poor absorption limit the clinical application.⁹⁵ Analogues are designed and synthesized to improve the water solution.

Literature showed that analogues of curcumin have an anticarcinogenic effect on breast cancer due to mechanisms similar to those of curcumin. Hutzen et al.⁹⁶ compared the effect of GO-Y030 and curcumin, and their data showed that GO-Y030 had a more potent effect on inhibiting STAT3, especially in HER2-positive breast cancer cells. Another two analogues, FLLL11 and FLLL12, suppressed the growth of breast carcinoma cells by activating transcription 3 phosphorylation. Lin et al.'s results⁹⁷ showed that FLLL11 and FLLL12 were stronger inhibitors of STAT3 and AKT compared to curcumin. Moreover, analogues EF24 and EF31 blocked the nuclear translocation of NF- κ B with a lower dosage, and PAC had a stronger effect on delaying the G2M phase.^{98,99} Furthermore, analogues had more cytotoxic effects through stimulating the cell phase 1 and phase 2 dox system, thus inducing stronger apoptosis of breast cancer cells.¹⁰⁰

Curcumin as Potent Chemosensitizer

Curcumin could serve as a chemosensitizer for other anticancer agents in treating human breast cancer cells, especially multi-drug-resistant (MDR) breast cancer cells. Limtrakul et al.¹⁰¹⁻¹⁰⁴ investigated the effects of tetrahydrocurcumin on three ATP-binding cassette (ABC) drug transporters, including P-glycoprotein (ABCB1/P-gp), multidrug resistance protein 1 (ABCC1), and mitoxantrone resistance protein (ABCG2/MXR). The results illustrated that in a concentration-dependent manner, P-gp ATPase activity is stimulated and [(125)I]-iodoarylazidoprazosin (IAAP) is inhibited. Thus, the binding of MDR with IAAP leads to the efflux of mitoxantrone and the inhibition of pheophorbide. Curcumin also exerts early modification of relevant gene transcription, such as downregulation of Bcl-2 and inhibition of apoptosis proteins (IAPs) in MCF-7R cells with overexpression of P-gp.¹⁰⁵ In 5-FU-resistant cells, curcumin acts as a chemosensitizer for 5-FU by silencing the enzyme thymidylate synthase (TS) in breast cancer cells.¹⁰⁶ TS, an upstream molecule of NF-kB, can be inhibited by 5-FU. However, prolonged exposure of 5-FU reversely increases the expression of TS and activates TS-dependent NF-kB, which causes 5-FU resistance in breast cancer cells. Curcumin can silence TS and suppress NF-kB, and thus chemosensitize the 5-FU anticancer capability. A similar mechanism can be found in doxorubicin-resistant breast cancer cells. Meivanto et al.'s study¹⁰⁷ suggested that curcumin inactivated NF- κ B and reduced the expression of HER2 in doxorubicin-resistant breast cancer cells.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The authors disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This paper has been supported by the National Natural Science Foundation of China (81301293) and National Science and Technology Major Projects for "Major New Drugs Innovation and Development" (2014ZX09507008).

References

- Siegel, R.; Naishadham, D.; Jemal, A. Cancer Statistics. CA Cancer J. Clin. 2013, 63 (1), 11–30.
- Sørlie, T.; Charles, M. P.; Robert, T.; et al. Gene Expression Patterns of Breast Carcinomas Distinguish Tumor Subclasses with Clinical Implications. *Proc. Natl. Acad. Sci. U.S.A.* 2001, *98* (19), 10869–10874.
- Anderson, W. F.; Chatterjee, N.; Ershler, W. B.; et al. Estrogen Receptor Breast Cancer Phenotypes in the Surveillance, Epidemiology, and End Results Database. *Breast Cancer Res. Treat.* 2002, *76* (1), 27–36.
- 4. Espinoza-Fonseca, L. M. Theoretical Biology and Medical Modelling. *Theor. Biol. Med. Model.* **2005**, *2*, 38.
- Giordano, S.; Petrelli, A. From Single- to Multi-Target Drugs in Cancer Therapy: When Aspecificity Becomes an Advantage. *Curr. Med. Chem.* 2008, 15 (5), 422–432.
- Singletary, K.; MacDonald, C.; Wallig, M.; et al. Inhibition of 7, 12-Dimethylbenzanthracene (DMBA)-Induced Mammary Tumorigenesis and DMBA-DNA Adduct Formation by Curcumin. *Cancer Lett.* **1996**, *103* (2), 137–140.
- Goel, A.; Kunnumakkara, A. B.; Aggarwal, B. B. Curcumin as "Curecumin": From Kitchen to Clinic. *Biochem. Pharmacol.* 2008, 75 (4), 787–809.
- Sharma, R. A.; Euden, S. A.; Platton, S. L.; et al. Phase I Clinical Trial of Oral Curcumin Biomarkers of Systemic Activity and Compliance. *Clin. Cancer Res.* 2004, *10* (20), 6847–6854.
- Cruz-Correa, M.; Shoskes, D. A.; Sanchez, P.; et al. Combination Treatment with Curcumin and Quercetin of Adenomas in Familial Adenomatous Polyposis. *Clin. Gastroenterol. Hepatol.* 2006, *4* (8), 1035–1038.
- Zang, S.; Liu, T.; Shi, J.; et al. Curcumin: A Promising Agent Targeting Cancer Stem Cells. *Anti-Cancer Agents Med. Chem.* 2014, 14 (6), 787–792.
- Shpaisman, N.; Sheihet, L.; Bushman, J.; et al. One-Step Synthesis of Biodegradable Curcumin-Derived Hydrogels as Potential Soft Tissue Fillers after Breast Cancer Surgery. *Biomacromolecules* 2012, *13* (8), 2279–2286.
- Asatiani, M. D.; Wasser, S. P.; Nevo, E.; et al. The Shaggy Inc Cap Medicinal Mushroom, *Coprinus comatus* Pers. (Agaricomycetideae) Substances Interfere with H₂O₂ Induction of the NF-κB Pathway through Inhibition of IκBα Phosphorylation

in MCF7 Breast Cancer Cells. *Int. J. Med. Mushrooms* **2011**, *13* (1), 19–25.

- Sen, G. S.; Mohanty, S.; Hossain, D. M. S.; et al. Curcumin Enhances the Efficacy of Chemotherapy by Tailoring p65NFκB-p300 Cross-Talk in Favor of p53-p300 in Breast Cancer. J. Biol. Chem. 2011, 286 (49), 42232–42247.
- Morabia, A.; Bemstein, M.; Héritier, S.; et al. Relation of Breast Cancer with Passive and Active Exposure to Tobacco Smoke. *Am. J. Epidemiol.* **1996**, *143* (9), 918–928.
- Schreinemachers, D. M.; Everson, R. B. Aspirin Use and Lung, Colon, and Breast Cancer Incidence in a Prospective Study. *Epidemiology* 1994, 5 (2), 138–146.
- Antoni, M. H.; Lehman, J. M.; Kilbourn, K. M.; et al. Cognitive-Behavioral Stress Management Intervention Decreases the Prevalence of Depression and Enhances Benefit Finding among Women under Treatment for Early-Stage Breast Cancer. *Health Psychol.* 2001, 20 (1), 20.
- Holmes, M. D.; Chen, W. Y.; Feskanich, D.; et al. Physical Activity and Survival after Breast Cancer Diagnosis. *JAMA* 2005, 293 (20), 2479–2486.
- Calle, E. E.; Rodriguez, C.; Walker-Thurmond, K.; et al. Overweight, Obesity, and Mortality from Cancer in a Prospectively Studied Cohort of US Adults. *N. Engl. J. Med.* 2003, *348* (17), 1625–1638.
- Amin, A. R. M. R.; Kucuk, O.; Khuri, F. R.; et al. Perspectives for Cancer Prevention with Natural Compounds. J. Clin. Oncol. 2009, 27 (16), 2712–2725.
- Priyadarsin, K. I. The Chemistry of Curcumin: From Extraction to Therapeutic Agent. *Molecules* 2014, 19 (12), 20091–20112.
- Paulucci, V. P.; Couto, R. O.; Teixeira, C. C. C.; et al. Optimization of the Extraction of Curcumin from *Curcuma longa* Rhizomes. *Braz. J. Farmacogn.* 2013, 23, 94–100.
- Lee, K. J.; Yang, H. J.; Jeong, S. W.; et al. Solid-Phase Extraction of Curcuminoid from Turmeric Using Physical Process Method. *Korean J. Farmacogn.* 2012, *43*, 250–256.
- Lee, K. J.; Ma, J. Y.; Kim, Y. S.; et al. High Purity Extraction and Simultaneous High-Performance Liquid Chromatography Analysis of Curcuminoids in Turmeric. *J. Appl. Biochem.* 2012, 55, 61–65.
- Li, M.; Ngadi, M. O.; Ma, Y. Optimisation of Pulsed Ultrasonic and Microwave-Assisted Extraction for Curcuminoids by Response Surface Methodology and Kinetic Study. *Food Chem.* 2014, 165, 29–34.
- Patel, K.; Krishna, G.; Sokoloski, E.; et al. Preparative Separation of Curcuminoids from Crude Curcumin and Turmeric Powder by pH-Zone-Refining Counter Current Chromatography. J. Lig. Chromatogr. 2000, 23, 2209–2218.
- Kim, Y. J.; Lee, H. J.; Shin, Y. Optimization and Validation of High-Performance Liquid Chromatography Method for Individual Curcuminoids in Turmeric by Heat-Refluxed Extraction. J. Agric. Food Chem. 2013, 61, 10911–10918.
- Song, C. S.; Wu, B. P.; Wang, H. Y. The Research on Extract Curcumin from Tumeric through Basic Solution Method. *Adv. Fine Petrochem.* 2006, 7 (4), 39–40.
- Kurmudle, N.; Kagliwal, L. D.; Bankar, S. B.; et al. Enzyme-Assisted Extraction for Enhanced Yields of Turmeric Oleoresin and Its Constituents. *Food Biosci.* 2013, *3*, 36–51.

- Price, L. C.; Buescher, R. W. Kinetics of Alkaline Degradation of the Food Pigments Curcumin and Curcuminoids. *J. Food Sci.* 1997, 62 (2), 267.
- Durante, M.; Lenucci, M. S.; Mita, G. Supercritical Carbon Dioxide Extraction of Carotenoids from Pumpkin (*Cucurbita* spp.): A Review. *Int. J. Mol. Sci.* 2014, *15* (4), 6725–6740.
- Díaz-Reinoso, B.; Moure, A.; Domínguez, H.; et al. Supercritical CO2 Extraction and Purification of Compounds with Antioxidant Activity. *J. Agric. Food Chem.* 2006, 54 (7), 2441–2469.
- Xiu, S. L.; Wu, Q. N.; Ouyang, Z.; et al. Study on the SFE Condition for Curcumin in *Curcuma longa*. *Zhongguo Zhong Yao Za Zhi* 2004, *29* (9), 857–860.
- Grykiewicz, G.; Silfirski, P. Curucmin and Curcuminoids in Quest for Medicinal Status. *Acta Biochim. Pol.* 2012, 59, 201–212.
- Esatbeyoglu, T.; Huebbe, P.; Ernst, I.; et al. Curcumin—From Molecule to Biological Function. *Angew. Chem. Int. Ed.* 2012, 51, 5308–5332.
- Pramasivam, M.; Poi, R.; Banerjee, H.; et al. High Performance Thin Layer Chromatography Method for Quantitative Determination of Curcuminoids in *Curcuma longa. Food Chem.* 2009, *113*, 639–644.
- Ali, I.; Haque, A.; Saleem, K. Separation and Identification of Curcuminoids in Turmeric Powder by HPLC Using Phenyl Column. *Anal. Methods* 2014, *6*, 2526–2536.
- 37. Lee, K.; Kim, Y. S.; Jung, P. M.; et al. Optimization of the Conditions for the Analysis of Curcumin and a Related Compound in *Curcuma longa* with Mobile-Phase Composition and Column Temperature via RP-HPLC. *Asian J. Chem.* 2013, 25, 6306–6310.
- Lee, K. J.; Kim, Y. S.; Ma, J. Y. Separation and Identification of Curcuminoids from Asian Turmeric (*Curcuma longa* L.) Using RP-HPLC and LC-MS. *Asian J. Chem.* 2013, 25, 909– 912.
- Wang, M. J.; Zhang, L.; Qiao, X. T.; et al. Optimization of Purification Process of Curcumin with Macroporous Resins. *China Condiment* 2015, 39 (5), 39–45.
- You, Q.; Zhang, Y.; Zhang, Q.; et al. High-Capacity Thermo-Responsive Magnetic Molecularly Imprinted Polymers for Selective Extraction of Curcuminoids. J. Chromatogr. A 2014, 1354, 1–8
- Wulandari, M.; Urraca, J. L.; Descalzo, A. B.; et al. Molecularly Imprinted Polymers for Cleanup and Selective Extraction of Curcuminoids in Medicinal Herbal Extracts. *Anal. Bional. Chem.* 2015, *397* (3), 803–812.
- Zhang, Z.; Chen, X.; Rao, W.; et al. Preparation of Novel Curcumin-Imprinted Polymers Based on Magnetic Multi-Walled Carbon Nanotubes for the Rapid Extraction of Curcumin from Ginger Powder and Kiwi Fruit Root. J. Sep. Sci. 2015, 38 (1), 108–114.
- Nhujak, T.; Saisuwan, W.; Srisa-art, M.; et al. Microemulsion Electrokinetic Chromatography for Separation Analysis of Curcumionids in Turmeric Samples. J. Sep. Sci. 2006, 29, 666–676.
- Lin, M. T.; Chang, C. C.; Chen, S. T.; et al. Cyr61 Expression Confers Resistance to Apoptosis in Breast Cancer MCF-7 Cells by a Mechanism of NF-κB-Dependent XIAP Up-Regulation. *J. Biol. Chem.* 2004, 279 (23), 23915–23923.

- 45. Liu, Q.; Loo, W. T. Y.; Sze, S. C. W.; et al. Curcumin Inhibits Cell Proliferation of MDA-MB-231 and BT-483 Breast Cancer Cells Mediated by Down-Regulation of NFκB, CyclinD and MMP-1 Transcription. *Phytomedicine* **2009**, *16* (10), 916-922.
- Bachmeier, B. E.; Mohrenz, I. V.; Mirisola, V.; et al. Curcumin Downregulates the Inflammatory Cytokines CXCL1 and -2 in Breast Cancer Cells via NFκB. *Carcinogenesis* 2008, 29 (4), 779–789.
- 47. Yodkeeree, S.; Ampasavate, C.; Sung, B.; et al. Demethoxycurcumin Suppresses Migration and Invasion of MDA-MB-231 Human Breast Cancer Cell Line. *Eur. J. Pharmacol.* 2010, 627 (1), 8–15.
- Yim-im, W.; Sawatdichaikul, O.; Semsri, S.; et al. Computational Analyses of Curcuminoid Analogs against Kinase Domain of HER2. *BMC Bioinformatics* 2014, 15 (1), 1–13.
- Meiyanto, E.; Putri, D. D. P.; Susidarti, R. A.; et al. Curcumin and Its Analogues (PGV-0 and PGV-1) Enhance Sensitivity of Resistant MCF-7 Cells to Doxorubicin through Inhibition of HER2 and NF-κB Activation. *Asian Pac. J. Cancer Prev.* 2014, *15* (1), 179–184.
- Catania, A.; Barrajón-Catalán, E.; Nicolosi, S.; et al. Immunoliposome Encapsulation Increases Cytotoxic Activity and Selectivity of Curcumin and Resveratrol against HER2 Over-Expressing Human Breast Cancer Cells. *Breast Cancer Res.* 2013, 140 (1), 55–65.
- Bachmeier, B. E.; Mirisola, V.; Romeo, F.; et al. Reference Profile Correlation Reveals Estrogen-Like Transcriptional Activity of Curcumin. *Cell Physiol. Biochem.* 2010, 26 (3), 471–482.
- 52. Shieh, J. M.; Chen, Y. C.; Lin, Y. C.; et al. Demethoxycurcumin Inhibits Energy Metabolic and Oncogenic Signaling Pathways through AMPK Activation in Triple-Negative Breast Cancer Cells. J. Agric. Food Chem. 2013, 61 (26), 6366–6375.
- Xia, Y.; Jin, L.; Zhang, B.; et al. The Potentiation of Curcumin on Insulin-Like Growth Factor-1 Action in MCF-7 Human Breast Carcinoma Cells. *Life Sci.* 2007, 80 (23), 2161–2169.
- Thulasiraman, P.; McAndrews, D. J.; Mohiudddin, I. Q. Curcumin Restores Sensitivity to Retinoic Acid in Triple Negative Breast Cancer Cells. *BMC Cancer* 2014, 14 (1), 724.
- Nejati-Koshki, K.; Akbarzadeh, A.; Pourhasan-Moghadam, M.; et al. Inhibition of Leptin and Leptin Receptor Gene Expression by Silibinin-Curcumin Combination. *Asian Pac. J. Cancer Prev.* 2013, 14 (11), 6595–6599.
- Lv, Z. D.; Liu, X. P.; Zhao, W. J.; et al. Curcumin Induces Apoptosis in Breast Cancer Cells and Inhibits Tumor Growth In Vitro and In Vivo. *Int. J. Clin. Exp. Pathol.* 2014, 7 (6), 2818.
- Narasimhan, S. R.; Yang, L.; Gerwin, B. I.; et al. Resistance of Pleural Mesothelioma Cell Lines to Apoptosis: Relation to Expression of Bcl-2 and Bax. *Am. J. Physiol. Lung Cell. Mol. Physiol.* **1998**, 275 (1), L165–L171.
- Ramachandran, C.; Rodrigue, Z.; Ramachandran, R.; et al. Expression Profiles of Apoptotic Genes Induced by Curcumin in Human Breast Cancer and Mammary Epithelial Cell Lines. *Anticancer Res.* 2005, 25 (5), 3293–3302.
- Fang, C. C.; Yen, C. J.; Tsai, T. J.; et al. Antibiotics Induce Apoptosis of Human Peritoneal Mesothelial Cells. *Nephrology* 2003, 8 (3), 142–149.

- Sun, X. D.; Liu, X. E.; Huang, D. S. Curcumin Induces Apoptosis of Triple-Negative Breast Cancer Cells by Inhibition of EGFR Expression. *Mol. Med. Rep.* 2012, 6 (6), 1267–1270.
- Park, S.; Cho, D. H.; Andera, L.; et al. Curcumin Enhances TRAIL-Induced Apoptosis of Breast Cancer Cells by Regulating Apoptosis-Related Proteins. *Mol. Cell. Biochem.* 2013, 383 (1–2), 39–48.
- Mahalingam, D.; Szegezdi, E.; Keane, M.; et al. TRAIL Receptor Signalling and Modulation: Are We on the Right TRAIL? *Cancer Treat. Rev.* 2009, *35* (3), 280–288.
- 63. Jiang, M.; Huang, O.; Zhang, X.; et al. Curcumin Induces Cell Death and Restores Tamoxifen Sensitivity in the Antiestrogen-Resistant Breast Cancer Cell Lines MCF-7/ LCC2 and MCF-7/LCC9. *Molecules* **2013**, *18* (1), 701–720.
- Ramachandran, C.; Fonseca, H. B.; Jhabvala, P.; et al. Curcumin Inhibits Telomerase Activity through Human Telomerase Reverse Transcriptase in MCF-7 Breast Cancer Cell Line. *Cancer Lett.* 2002, 184 (1), 1–6.
- 65. Ke, C. S.; Liu, H. S.; Yen, C. H.; et al. Curcumin-Induced Aurora-A Suppression Not Only Causes Mitotic Defect and Cell Cycle Arrest but Also Alters Chemosensitivity to Anticancer Drugs. J. Nutr. Biochem. 2014, 25 (5), 526–539.
- Mehta, K.; Pantazis, P.; McQueen, T.; et al. Antiproliferative Effect of Curcumin (Diferuloylmethane) against Human Breast Tumor Cell Lines. *Anti-Cancer Drug* 1997, 8 (5), 470– 481.
- Shao, Z. M.; Shen, Z. Z.; Liu, C. H.; et al. Curcumin Exerts Multiple Suppressive Effects on Human Breast Carcinoma Cells. *Int. J. Cancer* 2002, *98* (2), 234–239.
- Giet, R.; Prigent, C. The *Xenopus laevis*/Aurora/Ip11p-Related Kinase pEg2 Participates in the Stability of the Bipolar Mitotic Spindle. *Exp. Cell. Res.* 2000, 258 (1), 145– 151.
- Guo, J.; Li, W.; Shi, H.; et al. Synergistic Effects of Curcumin with Emodin against the Proliferation and Invasion of Breast Cancer Cells through Upregulation of miR-34a. *Mol. Cell. Biochem.* 2013, 382 (1–2), 103–111.
- Yang, J.; Cao, Y.; Sun, J.; et al. Curcumin Reduces the Expression of Bcl-2 by Upregulating miR-15a and miR-16 in MCF-7 Cells. *Med. Oncol.* 2010, 27 (4), 1114–1118.
- Sun, M.; Estrov, Z.; Ji, Y.; et al. Curcumin (Diferuloylmethane) Alters the Expression Profiles of MicroRNAs in Human Pancreatic Cancer Cells. *Mol. Cancer Ther.* 2008, 7 (3), 464– 473.
- Kronski, E.; Fiori, M. E.; Barbieri, O.; et al. miR181b Is Induced by the Chemopreventive Polyphenol Curcumin and Inhibits Breast Cancer Metastasis via Down-Regulation of the Inflammatory Cytokines CXCL1 and -2. *Mol. Oncol.* 2014, 8 (3), 581–595.
- 73. Mirza, S.; Sharma, G.; Parshad, R.; et al. 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* **2013**, *16* (1), 23– 31.
- Du, L.; Xie, Z.; Wu, L.; et al. Reactivation of RASSF1A in Breast Cancer Cells by Curcumin. *Nutr. Cancer* 2012, 64 (8), 1228–1235.

- Collins, H. M.; Abdelghany, M. K.; Messmer, M.; et al. Differential Effects of Garcinol and Curcumin on Histone and p53 Modifications in Tumour Cells. *BMC Cancer* 2013, *13* (1), 37.
- Bhattacharyya, S.; Hossain, D. M. S.; Mohanty, S.; et al. Curcumin Reverses T Cell-Mediated Adaptive Immune Dysfunctions in Tumor-Bearing Hosts. *Cell. Mol. Immunol.* 2010, 7 (4), 306–315.
- Luo, F.; Song, X.; Zhang, Y.; et al. Low-Dose Curcumin Leads to the Inhibition of Tumor Growth via Enhancing CTL-Mediated Antitumor Immunity. *Int. Immunopharmacol.* 2011, *11* (9), 1234–1239.
- Zhang, H. G.; Kim, H.; Liu, C.; et al. Curcumin Reverses Breast Tumor Exosomes Mediated Immune Suppression of NK Cell Tumor Cytotoxicity. *BBA Mol. Cell. Res.* 2007, *1773* (7), 1116–1123.
- Hossain, D. M. S.; Panda, A. K.; Chakrabarty, S.; et al. MEK Inhibition Prevents Tumor-Shed TGFβ-Induced T-Regulatory Cell Augmentation in Tumor Milieu. *Immunology* 2015, *144* (4), 561–573.
- Di, G. H.; Li, H. C.; Shen, Z. Z.; et al. Analysis of Anti-Proliferation of Curcumin on Human Breast Cancer Cells and Its Mechanism. *Chin. Med. J. Pekin.* 2003, *83* (20), 1764– 1768.
- Shao, Z. M.; Shen, Z. Z.; Liu, C. H.; et al. Curcumin Exerts Multiple Suppressive Effects on Human Breast Carcinoma Cells. *Int. J. Cancer* 2002, *98* (2), 234–239.
- Carroll, C. E.; Ellersieck, M. R.; Hyder, S. M. Curcumin Inhibits MPA-Induced Secretion of VEGF from T47-D Human Breast Cancer Cells. *Menopause* 2008, *15* (3), 570– 574.
- Palange, A. L.; Di Mascolo, D.; Singh, J.; et al. Modulating the Vascular Behavior of Metastatic Breast Cancer Cells by Curcumin Treatment. *Front. Oncol.* 2012, 2, 161.
- Aggarwal, B. B.; Shishodia, S.; Takada, Y.; et al. Curcumin Suppresses the Paclitaxel-Induced Nuclear Factor-κB Pathway in Breast Cancer Cells and Inhibits Lung Metastasis of Human Breast Cancer in Nude Mice. *Clin. Cancer Res.* 2005, *11* (20), 7490–7498.
- Im Kim, H.; Huang, H.; Cheepala, S.; et al. Curcumin Inhibition of Integrin (a6b4)-Dependent Breast Cancer Cell Motility and Invasion. *Cancer Prev. Res.* 2008, 1 (5), 385– 391.
- Narasimhan, M.; Ammanamanchi, S. Curcumin Blocks RON Tyrosine Kinase–Mediated Invasion of Breast Carcinoma Cells. *Cancer Res.* 2008, 68 (13), 5185–5192.
- Bachmeier, B. E.; Nerlich, A. G.; Iancu, C. M.; et al. The Chemopreventive Polyphenol Curcumin Prevents Hematogenous Breast Cancer Metastases in Immunodeficient Mice. *Cell. Physiol. Biochem.* 2007, *19* (1–4), 137–152.
- Chen, B.; Zhang, Y.; Wang, Y.; et al. Curcumin Inhibits Proliferation of Breast Cancer Cells through Nrf2-Mediated Down-Regulation of Fen1 Expression. *J. Steroid Biochem.* 2014, 143, 11–18.
- DeNicola, G. M.; Karreth, F. A.; Humpton, T. J.; et al. Oncogene-Induced Nrf2 Transcription Promotes ROS Detoxification and Tumorigenesis. *Nature* 2011, 475 (7354), 106–109.

- Dickinson, D. A.; Iles, K. E.; Zhang, H.; et al. Curcumin Alters EpRE and AP-1 Binding Complexes and Elevates Glutamate-Cysteine Ligase Gene Expression. *FASEB J.* 2003, *17* (3), 473–475.
- Calaf, G. M.; Echiburiú-Chau, C.; Roy, D.; et al. Protective Role of Curcumin in Oxidative Stress of Breast Cells. *Oncol. Rep.* 2011, *26* (4), 1029.
- Quiroga, A.; Quiroga, P. L.; Martínez, E.; et al. Anti-Breast Cancer Activity of Curcumin on the Human Oxidation-Resistant Cells ZR-75-1 with Gamma-Glutamyltranspeptidase Inhibition. J. Pharmacol. Exp. Ther. 2009, 8 (3), 261–266.
- Hong, R. L.; Spohn, W. H.; Hung, M. C. Curcumin Inhibits Tyrosine Kinase Activity of p185neu and Also Depletes p185neu. *Clin. Cancer Res.* **1999**, *5* (7), 1884–1891.
- 94. Prasad, C. P.; Rath, G.; Mathur, S.; et al. Expression Analysis of Maspin in Invasive Ductal Carcinoma of Breast and Modulation of Its Expression by Curcumin in Breast Cancer Cell Lines. *Chem. Biol. Interact.* **2010**, *183* (3), 455–461.
- 95. Hendrayani, S. F.; Al-Khalaf, H. H.; Aboussekhra, A. Curcumin Triggers p16-Dependent Senescence in Active Breast Cancer-Associated Fibroblasts and Suppresses Their Paracrine Procarcinogenic Effects. *Neoplasia* 2013, 15 (6), 631-IN11.
- Hutzen, B.; Friedman, L.; Sobo, M.; et al. Curcumin Analogue GO-Y030 Inhibits STAT3 Activity and Cell Growth in Breast and Pancreatic Carcinomas. *Int. J. Oncol.* 2009, *35* (4), 867–872.
- Lin, L.; Hutzen, B.; Ball, S.; et al. New Curcumin Analogues Exhibit Enhanced Growth-Suppressive Activity and Inhibit AKT and Signal Transducer and Activator of Transcription 3 Phosphorylation in Breast and Prostate Cancer Cells. *Cancer Sci.* 2009, 100 (9), 1719–1727.
- Shoba, G.; Joy, D.; Joseph, T.; et al. Influence of Piperine on the Pharmacokinetics of Curcumin in Animals and Human Volunteers. *Planta Med.* **1998**, *64* (04), 353–356.
- Fadda, A. A.; Badria, F. A.; El-Attar, K. M. Synthesis and Evaluation of Curcumin Analogues as Cytotoxic Agents. *Med. Chem. Res.* 2010, *19* (5), 403–430.

- 100. Sun, A.; Lu, Y. J.; Hu, H.; et al. Curcumin Analog Cytotoxicity against Breast Cancer Cells: Exploitation of a Redox-Dependent Mechanism. *Bioorg. Med. Chem. Lett.* 2009, 19 (23), 6627–6631.
- 101. Fadda, A. A.; Badria, F. A.; El-Attar, K. M. Synthesis and Evaluation of Curcumin Analogues as Cytotoxic Agents. *Med. Chem. Res.* 2010, 19 (5), 403–430.
- 102. Sun, A.; Lu, Y. J.; Hu, H.; et al. Curcumin Analog Cytotoxicity against Breast Cancer Cells: Exploitation of a Redox-Dependent Mechanism. *Bioorg. Med. Chem. Lett.* 2009, 19 (23), 6627–6631.
- 103. Al-Hujaily, E. M.; Mohamed, A. G.; Al-Sharif, I.; et al. PAC, a Novel Curcumin Analogue, Has Anti-Breast Cancer Properties with Higher Efficiency on ER-Negative Cells. *Breast Cancer Res. Treat.* 2011, *128* (1), 97–107.
- 104. Limtrakul, P.; Chearwae, W.; Shukla, S.; et al. Modulation of Function of Three ABC Drug Transporters, P-Glycoprotein (ABCB1), Mitoxantrone Resistance Protein (ABCG2) and Multidrug Resistance Protein 1 (ABCC1) by Tetrahydrocurcumin, a Major Metabolite of Curcumin. *Mol. Cell. Biochem.* 2007, 296 (1–2), 85–95.
- 105. Poma, P.; Notarbartolo, M.; Labbozzetta, M.; et al. The Antitumor Activities of Curcumin and of Its Isoxazole Analogue Are Not Affected by Multiple Gene Expression Changes in an MDR Model of the MCF-7 Breast Cancer Cell Line: Analysis of the Possible Molecular Basis. *Int. J. Mol. Med.* 2007, 20 (3), 329.
- 106. Vinod, B. S.; Antony, J.; Nair, H. H.; et al. Mechanistic Evaluation of the Signaling Events Regulating Curcumin-Mediated Chemosensitization of Breast Cancer Cells to 5-Fluorouracil [J]. *Cell Death Dis.* **2013**, *4* (2), e505.
- 107. Meiyanto, E.; Putri, D. D. P.; Susidarti, R. A.; et al. Curcumin and Its Analogues (PGV-0 and PGV-1) Enhance Sensitivity of Resistant MCF-7 Cells to Doxorubicin through Inhibition of HER2 and NF-κB Activation. *Asian Pac. J. Cancer Prev.* 2014, *15* (1), 179–184.