COMMENTARY

Anthocyanins: Targeting of Signaling Networks in Cancer Cells

Muserref Hilal Sehitoglu[1]*, Ammad Ahmad Farooqi[2]*, Muhammad Zahid Qureshi[3], Ghazala Butt[4], Aliye Aras[5]

Abstract

It is becoming progressively more understandable that phytochemicals derived from edible plants have shown potential in modelling their interactions with their target proteins. Rapidly accumulating in-vitro and in- vivo evidence indicates that anthocyanins have anticancer activity in rodent models of cancer. More intriguingly, evaluation of bilberry anthocyanins as chemopreventive agents in twenty-five colorectal cancer patients has opened new window of opportunity in translating the findings from laboratory to clinic. Confluence of information suggests that anthocyanins treated cancer cells reveal up-regulation of tumor suppressor genes. There is a successive increase in the research-work in nutrigenomics and evidence has started to shed light on intracellular-signaling cascades as common molecular targets for anthocyanins. In this review we bring to limelight how anthocyanins induced apoptosis in cancer cells via activation of extrinsic and intrinsic pathways.

Keywords: Apoptosis - anthocyanins - signaling - cancer

Asian Pac J Cancer Prev, **15 (5)**, 2379-2381

Introduction

Research over the decades has gradually and sequentially shown that there is an organized interaction of cells with surrounding environment and cells respond to environmental clues accordingly via intracellular signal-transduction network. This well organized network is composed of hierarchy of upstream activators and downstream effectors which receive, transmit and interpret information. Due to substantial role in cellular physiology, intracellular signaling network and several of its subnetworks, have emerged as deeply studied molecular mechanisms underlying carcinogenesis. These transduction cascades are initiated by ligand-receptor interactions, and disrupt spatio-temporal behavior of the cell.

Upregulation of oncogenes and suppression of tumor suppressor genes is an essential mechanism that is regulated by intracellular signaling cascades. These cascades have downstream effectors which are activated and thus stimulate or repress the expression of oncogenes and tumor suppressors respectively. NF-κB, AP-1, STAT-1 and OCT-1 are some of the transcription factors which are activated in colorectal cancer cells. Interestingly metabolites of anthocyanins particularly, trihydroxybenzaldehyde, gallic acid and methylgallic acid have also been studied for their potential in inducing apoptosis in colon cancer cells. Evidence suggested that cancer cell treated with these metabolites had notably reduced NF-κB, AP-1, STAT-1and OCT-1 (Forester et al., 2012). Anthocyanins have recently been shown to stimulate the expression of tumor suppressor genes via demethylation of promoters of CDKN2A, and SFRP2, SFRP5, and WIF1 (Wang et al., 2013).

Tumour-associated cell cycle defects are frequently triggered by dysregulated cyclin-dependent kinase (CDK) activity. There is considerable evidence that suggests that CDKs are involved in unscheduled proliferation as well as genomic and chromosomal instability. p21 and p27 negatively regulate CDKs and are well positioned to function as both a sensors and effectors of multiple signals that can converge to induce cell cycle arrest. It is interesting to note that anthocyanins and anthocyanidins extracted from purple-shoot tea effectively target cyclin E and cyclin D1 in colorectal carcinoma cells. In addition there was a notable activation of p21 and p27 (cyclin-dependent kinase inhibitors) and caspase-3 (Hsu et al., 2012). 3-Deoxyanthocyanin isolated from red sorghum bran has been shown to stimulate the expression of p53 in breast cancer cells. Additionally, anti-apoptotic gene BCL-2 was remarkably suppressed (Suganyadevi et al., 2013). Anthocyanins have been shown to induce apoptosis in different cancer cells via a mitochondrial pathway (Banjerdpongchai et al., 2013) and human leukemia U937 cells via activation of caspase-3, -8 and -9. However, Bcl-2 overexpressing U937 cells did not show anthocyanin induced apoptosis (Lee et al, 2009).

*[1]Department of Medical Biochemistry, Canakkale Onsekiz Mart University, Canakkale, [5]Department of Biology, Faculty of Science, Istanbul University, Istanbul, Turkey [2]Laboratory for Translational Oncology and Personalized Medicine, Rashid Latif Medical College, [3]Biochemistry Lab, Department of Chemistry, [4]Phycology Lab, Department of Botany, GC University, Lahore, Pakistan, *For correspondence: hilals@comu.edu.tr ammadahmad638@yahoo.com*

Targeting of Pro-survival Signaling

Pro-survival signaling in cancer cells is an essential mechanism to overcome apoptosis inducing signals. NFKB inhibition is necessary to induce apoptosis. It is noteworthy that anthocyanins isolated from Vitis coignetiae Pulliat induced apoptosis in colon cancer HCT-116 cells. The results revealed that anthocyanins inhibited NFKB HCT-116 cells. Moreover, p38-MAPK was activated in HCT-116 cells treated with anthocyanins (Shin et al., 2009). Certain reports have provided evidence that metabolites of anthocyanins are more effective as anticancer agents. Metabolites of anthocyanins efficiently reduced proliferation of Caco-2 cells (Forester and Waterhouse, 2010). Anthocyanin rich-fraction considerably reduced B16-F10 melanoma murine cells proliferation (Bunea et al., 2013). Peonidin-3-glucoside and cyaniding-3-glucoside are anthocyanin pigments and remarkably effective against HER positive cancer cells. Results revealed that there was a reduced HER2 phosphorylation in anthocyanin pigments treated cancer cells. Moreover pAkt levels were also repressed suggesting that pigments substantially inhibited HER2 signaling axis (Liu et al., 2013). Anthocyanins induced apoptosis was noted in MCF-7 cells as evidenced by increase in amounts of oligonuceosomal-sized fragments (Devi et al., 2011).

Animal Model Studies

Anthocyanins have shown potential via exerting their inhibitory effects in prostatic hyperplasia-induced rat model. Mice were given anthocyanin for 4 weeks and it was noted that prostate weights were significantly lower in the group receiving oral doses of anthocyanin (Jang et al., 2010). Preneoplastic hepatic nodules in diethylnitrosamine-induced hepatocellular carcinogenesis in rats have also been noted to be reduced considerably. Additionally it was found that there was an increase in protein expression of Bax (Bishayee et al., 2011). On a similar note efficacy of anthocyanins has been tested in BALB/c nude mice inoculated with MDA-MB-453 cells and results revealed substantial regression of tumor growth (Hui et al., 2010). Colon cancer cells have also been studied to underscore regulation of mTOR signaling by anthocyanins and it was shown that cancer cells pretreated with anthocyanins displayed marked decrease in phosphorylated mTOR levels. Furthermore, mice xenografted with HT-29 colon cancer cells indicated decrease in tumor volumes (Lee et al., 2010). There was a notable reduction in intestinal adenoma development in mice fed with Red grape pomace extract (Cai et al., 2010). It is noteworthy that dietary anthocyanin suppressed azoxymethane-induced formation of aberrant crypt foci in the colons of CF-1 mice (Lim et al., 2013). Peonidin 3-glucoside has been shown to inhibit metastasizing potential of Lewis lung carcinoma cells in cancer bearing mice (Ho et al., 2010). Prostate cancer has also been noted to be inhibited in athymic nude mice xenografted with prostate cancer cells. This inhibitory effect was exerted through delphinidin via inhibition of NFKB and its DNA binding activity Hafeez et al. (2008). Anthocyanins are also found in sweet potato greens extract (SPGE) and effectively inhibited cancer progression in nude mice inoculated with prostate cancer cells (Karna et al., 2008). Metastasizing potential of B16-F1 cells in C57BL/6 mice was considerably reduced by mulberry anthocyanins mediated inhibitory effects on PI3K, Ras and NFKB (Huang et al., 2008). Pharmacokinetics of Cyanidin-3-glucoside (C3G) have been studied in C57BL6J and results revealed that after oral administration C3G accumulated in gastrointestinal mucosal tissues and liver (Marczylo et al., 2009). It has been experimentally shown that 12-O-tetradecanoylphorbol-13-acetate (TPA) induced epidermal hyperplasia in mice. Mechanistically it was noted that TPA induced activation of NFKB, IKK and inhibition of IKB. Moreover, TPA-induced phosphorylation of ERK1/2, p38 and JNK1/2. However, anthocyanins inhibited TPA induced hyperplasia via inhibition of NFKB, ERK, p38 MAPK and JNK (Afaq et al., 2005). TPA-induced neoplastic transformation in JB6 P+ cells and H-Ras-transformed JB6 P+ mouse epidermal cells. *in vitro* analysis revealed that Raf/MEK/ERK signaling axis was activated in TPA treated cells. Delphinidin substantially inhibited NFKB activation and Raf/MEK/ERK signaling axis (Kang et al., 2008). Nitrosomethylbenzylamine (NMBA)-induced tumors in the rat esophagus were also considerably suppressed in anthocyanins fed rats. It was observed that anthocyanins induced apoptosis in preneoplastic esophageal tissues (Wang et al., 2009).

Conclusion

It is encouraging to note that evaluation of bilberry anthocyanins as chemopreventive agents in twenty-five colorectal cancer patients has opened anew window of opportunity in translating the findings from laboratory to clinic (Gu et al., 2013).

References

Afaq F, Saleem M, Krueger CG, Reed JD, Mukhtar H (2005). Anthocyanin- and hydrolyzable tannin-rich pomegranate fruit extract modulates MAPK and NF-kappaB pathways and inhibits skin tumorigenesis in CD-1 mice. *Int J Cancer*, **113**, 423-33.

Banjerdpongchai R, Wudtiwai B, Sringarm K (2013). Cytotoxic and apoptotic-inducing effects of purple rice extracts and chemotherapeutic drugs on human cancer cell lines. *Asian Pac J Cancer Prev*, **14**, 6541-8

Bishayee A, Mbimba T, Thoppil RJ, et al (2011). Anthocyanin-rich black currant (Ribes nigrum L.) extract affords chemoprevention against diethylnitrosamine-induced hepatocellular carcinogenesis in rats. *J Nutr Biochem*, **22**, 1035-46.

Bunea A, Rugina D, Sconta Z, et al (2013). Anthocyanin determination in blueberry extracts from various cultivars and their antiproliferative and apoptotic properties in B16-F10 metastatic murine melanoma cells. *Phytochemistry*, **95**, 436-44.

Cai H, Marczylo TH, Teller N, et al (2010). Anthocyanin-rich red grape extract impedes adenoma development in the Apc(Min) mouse: pharmacodynamic changes and anthocyanin levels in the murine biophase. *Eur J Cancer*,

46, 811-7.

Devi PS, Kumar MS, Das SM (2011). Evaluation of antiproliferative activity of red sorghum bran anthocyanin on a human breast cancer cell line (mcf-7). *Int J Breast Cancer*, **2011**, 891481.

Forester SC, Choy YY, Waterhouse AL, Oteiza PI (2012). The anthocyanin metabolites gallic acid, 3-O-methylgallic acid, and 2,4,6-trihydroxybenzaldehyde decrease human colon cancer cell viability by regulating pro-oncogenic signals. Mol Carcinog.

Forester SC, Waterhouse AL (2010). Gut metabolites of anthocyanins, gallic acid, 3-O-methylgallic acid, and 2,4,6-trihydroxybenzaldehyde, inhibit cell proliferation of Caco-2 cells. *J Agric Food Chem*, **58**, 5320-7.

Gu J, Ahn-Jarvis JH, Riedl KM, et al (2013). Characterization of black raspberry functional food products for cancer prevention human clinical trials. J Agric Food Chem.

Hafeez BB, Siddiqui IA, Asim M, et al (2008). A dietary anthocyanidin delphinidin induces apoptosis of human prostate cancer PC3 cells *in vitro* and *in vivo*: involvement of nuclear factor-kappaB signaling. *Cancer Res*, **68**, 8564-72.

Ho ML, Chen PN, Chu SC, et al (2010). Peonidin 3-glucoside inhibits lung cancer metastasis by downregulation of proteinases activities and MAPK pathway. *Nutr Cancer*, **62**, 505-16.

Hsu CP, Shih YT, Lin BR, Chiu CF, Lin CC (2012). Inhibitory effect and mechanisms of an anthocyanins- and anthocyanidins-rich extract from purple-shoot tea on colorectal carcinoma cell proliferation. *J Agric Food Chem*. **60**, 3686-92.

Huang HP, Shih YW, Chang YC, Hung CN, Wang CJ (2008). Chemoinhibitory effect of mulberry anthocyanins on melanoma metastasis involved in the Ras/PI3K pathway. *J Agric Food Chem*, **56**, 9286-93.

Hui C, Bin Y, Xiaoping Y, et al (2010). Anticancer activities of an anthocyanin-rich extract from black rice against breast cancer cells *in vitro* and *in vivo*. *Nutr Cancer*, **62**, 1128-36.

Jang H, Ha US, Kim SJ, et al (2010). Anthocyanin extracted from black soybean reduces prostate weight and promotes apoptosis in the prostatic hyperplasia-induced rat model. *J Agric Food Chem*, **58**, 12686-91.

Kang NJ, Lee KW, Kwon JY, et al (2008). Delphinidin attenuates neoplastic transformation in JB6 Cl41 mouse epidermal cells by blocking Raf/mitogen-activated protein kinase kinase/extracellular signal-regulated kinase signaling. *Cancer Prev Res*, **1**, 522-31.

Karna P, Gundala SR, Gupta MV, et al (2011). Polyphenol-rich sweet potato greens extract inhibits proliferation and induces apoptosis in prostate cancer cells *in vitro* and *in vivo*. *Carcinogenesis*, **32**, 1872-80.

Lee SH, Park SM, Park SM, et al (2009). Induction of apoptosis in human leukemia U937 cells by anthocyanins through down-regulation of Bcl-2 and activation of caspases. *Int J Oncol*, **34**, 1077-83.

Lee YK, Lee WS, Kim GS, Park OJ (2010). Anthocyanins are novel AMPKα1 stimulators that suppress tumor growth by inhibiting mTOR phosphorylation. *Oncol Rep*, **24**, 1471-7.

Lim S, Xu J, Kim J, et al (2013). Role of anthocyanin-enriched purple-fleshed sweet potato p40 in colorectal cancer prevention. *Mol Nutr Food Res*, **57**, 1908-17.

Liu W, Xu J, Wu S, et al (2013). Selective anti-proliferation of HER2-positive breast cancer cells by anthocyanins identified by high-throughput screening. *PLoS One*, **8**, 81586.

Marczylo TH, Cooke D, Brown K, Steward WP, Gescher AJ (2009). Pharmacokinetics and metabolism of the putative cancer chemopreventive agent cyanidin-3-glucoside in mice. *Cancer Chemother Pharmacol*, **64**, 1261-8.

Shin DY, Lee WS, Lu JN, et al (2009). Induction of apoptosis in human colon cancer HCT-116 cells by anthocyanins through suppression of Akt and activation of p38-MAPK. *Int J Oncol*, **35**, 1499-504.

Suganyadevi P, Saravanakumar KM, Mohandas S (2013). The antiproliferative activity of 3-deoxyanthocyanins extracted from red sorghum (Sorghum bicolor) bran through P(53)-dependent and Bcl-2 gene expression in breast cancer cell line. *Life Sci*, **92**, 379-82.

Wang LS, Hecht SS, Carmella SG, et al (2009). Anthocyanins in black raspberries prevent esophageal tumors in rats. *Cancer Prev Res*, 84-93.

Wang LS, Kuo CT, Cho SJ, et al (2013). Black raspberry-derived anthocyanins demethylate tumor suppressor genes through the inhibition of DNMT1 and DNMT3B in colon cancer cells. *Nutr Cancer*, **65**, 18-25.