

Chemical Properties and Anti-Cancer Effects of Texas Wines

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Introduction

Wine and specifically red wine has widely reported health benefits including the prevention of chronic and degenerative diseases, e.g. cardiovascular disease and cancer {Grenett, 2007 #211;He, 2008 #215;Lopez-Velez, 2003 #243;Stockley, 2005 #309;Sun, 2008 #312;Sun, 2008 #312;Oak, 2005 #263}. The chemopreventive properties of red wine have at least in part been ascribed to polyphenolic compounds (German & Walzem 2000), including flavonoids and non-flavonoids. Flavonoids include flavan-3-ols (catechins, epicatechins, procyanidins), anthocyanins (delphinidin, cyanidin, peonidin), flavonols (quercetin, myricetin, rutin). Non-flavonoid group includes stilbenes (resveratrol), hydroxycinnamic acids (caffeic, coumaric, ferulic), and hydroxybenzoic acids (gallic, protocatechuric) {Lopez-Velez, 2003 #243;He, 2008 #215}.

Red wine has shown cytotoxic effects in different cancer cell lines (Kampa et al 2000; Romero et al 2002; Sun et al 2008). The potential anti-cancer mechanisms of red wine include the induction of apoptosis, cell cycle arrest in one or more control points, modulation of p53 gene expression, inhibition of DNA synthesis, and interfering with signal transduction pathways (He et al 2008; Mertens-Talcott et al 2008; Soleas et al 2001; Soleas et al 2006).

Micro RNAs have recently been discovered (Sassen et al 2008), and are small non-coding RNA molecules not longer which play an important role in cancer. MicroRNAs can regulate gene expression of target gene mRNAs (Fabbri et al 2007; Mertens-Talcott et al 2007). MiRNAs downregulate gene expression either by degradation of mRNA or by inhibiting protein translation (Sassen et al 2008; Williams 2008). MiRNA-mediated oncogenic activity may result from either downregulation of tumor suppressor genes or upregulation of oncogenic genes (Lu et al 2008; Williams 2008).

Specifically, micro-RNA27a (miR-27a) is widely expressed in cancer cells (Mertens-Talcott et al 2007). Previous studies have shown that miR-27a inhibits the expression of zinc-finger protein ZBTB10/RINZF, a known suppressor of the oncogenic specificity protein Sp1 (Scott et al 2006). Several reports have demonstrated the overexpression and oncogenic activity of Sp1 protein in several tumor tissues (Jiang et al 2008; Lou et al 2005; Mertens-Talcott et al 2007; Shi et al 2001; Wang et al 2003). As a result of this connection of pathways, Sp1 protein enhances the expression of angiogenic and antiapoptotic genes such as vascular endothelial growth factor (VEGF) and survivin (Mertens-Talcott et al 2007).

The state of Texas is among the leading wine producers in the United States. Little research has been conducted that relates processing techniques of Texas wines to chemical composition, and subsequently potentially health-promoting benefits.

The aim of this study was to assess wines produced in Texas in an effort to provide insight on the effects of common processing techniques used in the industry on the chemical content of the wines. Moreover, we investigated the anti-cancer effect of a red wine from Black Spanish grapes (*Vitis labrusca*) in colorectal adenocarcinoma cells (HT-29) and assessed whether miR-27a may play a role as underlying mechanism.

Materials and Methods

Chemical Analysis of submitted Texas wines: Fifty-one Texas wines were kindly donated by their respective wineries to Texas A&M University for participation in this commercial wine chemistry trial. Chemical traits assessed include antioxidant capacity, total anthocyanins, total soluble phenolics, color density, hue/tint, monomeric-polymeric anthocyanins, ammonium, potassium, glucose, lactate, pH, soluble solids, titratable acidity, turbidity, and individual phenolics (gallic acid, (+)-catechin, (-)-epicatechin, and resveratrol). Additionally, each participating winery was asked to provide certain processing information.

Chemical Analysis of Black Spanish Grape Wine: A red wine, Port Barrel Reserve made of Black Spanish grapes (*Vitis labrusca*), was provided by Messina Hof (Bryan, TX). Wine polyphenolics were extracted as previously described. Extracts were dissolved in dimethyl sulfoxide (DMSO) for use in cell culture.

Total soluble phenolics were determined by Folin-Ciocalteu assay (Singleton & Rossi 1965). Total anthocyanin contents were determined spectrophotometrically by pH differential method. The Percentage of polymeric anthocyanins was determined based on color retention in the presence of sodium sulfite (Rodriguez-Saona et al 1999). The antioxidant capacity was determined using the oxygen radical absorbance capacity assay (ORAC) (Talcott & Lee 2002; Wang et al 1996). The wine extract was analyzed by HPLC-PDA; the compounds were identified by retention time and PDA spectrum with original standards as described by Talcott and Lee (Lee & Talcott 2002).

Cell Culture: HT-29 human adenocarcinoma cells were obtained from American Type Culture Collection (ATCC, Manassas, VA), and cultured according to manufacturer's guidelines. The generation of intracellular reactive oxygen species (ROS) was determined as previously performed (Meng et al 2008). Real time PCR, cell cycle kinetics, western blotting, transfections, and statistical analysis were performed as previously described.

Results and Discussion

The chemical analysis of Texas wines demonstrated that as expected, red wines had an overall higher antioxidant capacity and total soluble phenolics compared to white wines. Based on a small sample size and inhomogeneous sample characteristics there were no statistically significant effects of processing on any of the investigated wine characteristics. Several trends were observed including a positive correlation between wine aged in oak barrels, the addition of oak-based products, fining of the wine and antioxidant capacity or total soluble phenolics.

Results from the cell culture study show that the proliferation and generation of reactive oxygen species (ROS) in colon cancer cells was decreased and mRNA of p53 was increased compared to the control indicating pro-apoptotic activity of the wine extract. It was previously shown that the expression of the oncogenic specificity protein Sp1 is increased in several types of cancer and that the oncogenic microRNA27a (miR-27a), which has been found to suppress zinc-finger protein ZBTB10 a transcription factor a known inhibitor of Sp1. In this study Sp1 expression was significantly decreased by the wine extract while ZBTB10 was increased. MiR-27a was significantly decreased in a concentration-dependent manner, indicating the involvement of this microRNA (miRNA) as underlying mechanism. Vascular endothelial growth factor (VEGF) and survivin protein and mRNA, both genes regulated by Sp1 were significantly decreased by the wine extract, indicating its anti-angiogenic and pro-apoptotic effects. The involvement of miR-27a was confirmed by transient transfection with the mimic of miR-27a, where effects partially were reversed by the addition of wine extract. Overall, a polyphenolic extract from red wine had cytotoxic effects in colon cancer cells where a decrease in miR-27a was involved in the underlying anti-cancer mechanisms.

Overall, the gained information demonstrates the chemical properties of the submitted Texas wines and indicated the anti-cancer effects of Texas wines in colon-cancer cells.

Applications of Research

- Texas wines have not been specifically investigated regarding their chemical properties and the influence of processing on these
- Gained data may provide a scientific basis for the improvement of processing techniques
- The demonstration of health benefits of specifically Texas wines may increase the consumption of Texas wines by Texans and beyond.

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