Ginsenosides from American ginseng: Chemical and pharmacological diversity

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Abstract
Ginseng occupies a prominent position in the list of best-selling natural products in the world. Compared to the long history of use and widespread research on Asian ginseng, the study of American ginseng is relatively limited. In the past decade, some promising advances have been achieved in understanding the chemistry, pharmacology and structure-function relationship of American ginseng. To date, there is no systematic review of American ginseng. In this review, we present the different structures of the ginsenosides in American ginseng, including naturally occurring compounds and those resulting from steaming or biotransformation. Preclinical and clinical studies published in the past decade will also be discussed. We highlight the chemical and pharmacological diversity and potential structural-activity relationship of ginsenosides. Our hope is that this article is a useful reference to chemists and biologists researching American ginseng, and will open the door to novel agents in drug discovery.

Keywords
American ginseng; Panax quinquefolius; Asian ginseng; ginsenosides; diversity; structural-activity relationship

1. Introduction
Ginseng root has been used for thousands of years in the traditional medical system in oriental countries (Ang-Lee et al., 2001; Attele et al., 1999; Wang and Yuan, 2008). It occupies a prominent position on the list of the best-selling medicinal plants in the world (Yun, 2001). Asian ginseng (Panax ginseng C. A. Meer) and American ginseng (Panax quinquefolius L.) are the two most recognized ginseng botanicals around the world (Ang-Lee et al., 2001; Jia and Zhao, 2009). Compared to the long history of use and the copious amounts of research on Asian ginseng (Ang-Lee et al., 2001; Yun, 2001), the study of American ginseng and its constituents is much less extensive (Yuan et al., 2004).

Table 1 and Fig. 1 show the chemical differences of American ginseng and Asian ginseng. As one of the best selling herbs in the U.S., American ginseng is grown in the eastern temperate forest areas of North America, from southern Quebec, Minnesota, and Wisconsin in the north, to Oklahoma, the Ozark Plateau, and Georgia in the south (Assinewe et al., 2003). With the widespread popularity of herbal medicines in the West, the past few decades have witnessed some promising advances in research on American ginseng and its constituents (Li et al., 2010a; Sengupta et al., 2004; Wang and Yuan, 2008). The triterpenoid

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saponins, called ginseng saponins or ginsenosides, are the major active constituents in American ginseng (Attele et al., 1999; Jia and Zhao, 2009). As shown in Fig. 1 and Table 1, however, American ginseng has a ginsenoside profile different from that of Asian ginseng in terms of total ginsenosides, the ratio of propanaxadiol (PPD) to propanaxatriol (PPT), and other marker ginsenosides. In addition, although low Rg1/high Re was reported in most populations of American ginseng, the high Rg1/low Re chemotype was also observed (Schlag and McIntosh, 2006).

Like Asian ginseng, American ginseng has been reported to have a wide range of pharmacological effects, including effects on the central nervous system, cardiovascular system, endocrine system, immune system and cancer (Court, 2000; Jin et al., 2010; Li et al., 2010a; Yuan and Dey, 2001). The pharmacological activities of American ginseng may be different from those of Asian ginseng (Sievenpiper et al., 2004a). Some structure-activity relationship (SAR) comparisons of homologs have generated interesting evidence supporting the role of specific structural components as requisites in ginsenosides for their activities (Kang et al., 2006; Li et al., 2009c; Popovich and Kitts, 2002; Qi et al., 2010b; Wang et al., 2007c). However, to date, there has been no systematic summary of the chemistry, pharmacology, and structure-function relationships of American ginseng.

In this review, we present the different structures of ginsenosides in American ginseng, including naturally occurring compounds and those resulting from steaming or biotransformation. Preclinical and clinical studies of American ginseng and ginsenosides from the past decade also will be discussed and compared with those of Asian ginseng. We highlight the chemical and pharmacological diversity of ginsenosides and their structure-function relationships. Furthermore, we will attempt to offer perspectives and predict future research trends for this herb.

2. Structural diversity

2.1. Ginsenosides isolated from American ginseng

Ginsenosides share a dammarane-type triterpenoid saponin structure (Fuzzati, 2004). Most ginsenosides belong to a family of steroids with a four trans-ring rigid steroid skeleton (Attele et al., 1999; Wang et al., 2005). More than 60 ginsenosides have been isolated from different parts of *Panax quinquefolius* (referred to as *Panax quinquefolium* in some publications), such as the roots, leaves, stems, flower buds and berries (Christensen, 2009; Jia and Zhao, 2009; Jiang et al., 2008; Nakamura et al., 2007; Qu et al., 2009; Yoshikawa et al., 1998). As chemical purification and structural identification techniques are developed, novel ginsenosides continue to be reported (Chen et al., 2009a; Jia et al., 2008; Li et al., 2009a; Nakamura et al., 2007; Wang et al., 2001; Yoshikawa et al., 1998). Differences in sugar types, quantities and attachment positions provide diversity in ginsenoside structures (Fuzzati, 2004; Jia and Zhao, 2009). The changeable C-20 side chain and stereoisomerism further enrich the structural diversity of ginsenosides (Christensen, 2009; Nakamura et al., 2007).

As shown in Fig. 2, ginsenosides isolated from *P. quinquefolius* can be divided into several groups. PPD and PPT are the two main groups of ginsenosides (Qu et al., 2009). In the PPD group, sugar residues are attached to the β-OH at C-3 and/or C-20. PPD compounds include compounds 1–17 (Chen et al., 2009a; Jiang et al., 2008; Li et al., 2009a; Nakamura et al., 2007; Wang et al., 2001; Yoshikawa et al., 1998). Four malonyl derivatives (18–21, also called “acidic” ginsenosides) have been characterized (Du et al., 2004). In the PPT group, sugar moieties are attached to the α-OH at carbon-6 and/or β-OH at C-20. PPT constituents include compounds 22–32 (Jia et al., 2008; Nakamura et al., 2007; Yoshikawa et al., 1998).
Minor ginsenosides isolated from *P. quinquefolius* include ocottillol-type (compounds 33–36), oleane-type ginsenosides (compounds 37–38), and dammarane saponins with a modified steroid skeleton (compounds 39–41) (Nakamura et al., 2007; Yoshikawa et al., 1998). Other isolated compounds can be classified as modified C-20 side chain ginsenosides (compounds 42–65). According to C-20 side chain differences, these compounds are subdivided into nine groups (Jiang et al., 2008; Nakamura et al., 2007; Qiu et al., 2009).

### 2.2. Ginsenosides characterized from steamed American ginseng

The steaming or heating process changed the ginsenoside profile of ginseng products (Chang and Ng, 2009; Wang et al., 2007a). During the steaming process, the notable structural changes are the elimination of sugar moieties and subsequent dehydration at C-20 in ginsenosides (Lau et al., 2004; Ren and Chen, 1999; Wang et al., 2007a). After steaming, the number of original polar ginsenosides decreased, and less polar ginsenosides increased in number correspondingly (Ren and Chen, 1999; Wang et al., 2007a). After steaming, structural changes are the elimination of sugar moieties and subsequent dehydration at C-20 (Chang and Ng, 2009; Wang et al., 2007a). During the steaming process, the notable structural changes are the elimination of sugar moieties and subsequent dehydration at C-20. Rg3 may be further transformed to 20(20-dehydr)-Rh3 (75), and subsequently become the aglycone 20(S)-PPD (76)/20(R)-PPD (77) or even 20-dehydr-PPD (78/79) through chemical degradation. Rk3 and Rh5 may be again transformed to their degradation products like Rk2 (80) and Rh3 (81) (Shin et al., 2006b). Rh1 can be changed to the aglycone 20(S)-PPT (82)/20(R)-PPT (83) or even 20-dehydr-PPT (84/85).

### 2.3. Ginsenosides identified after biotransformation

As a dietary supplement, American ginseng is usually taken orally. Therefore, the metabolism of ginsenosides has been investigated (Cui et al., 1997). Many experiments tested the degradation of ginsenosides using microbes, enzymes and intestinal bacteria, or animals and humans (Hasegawa, 2004; Tawab et al., 2003). A number of novel ginsenoside structures have been identified after biotransformation. PPD groups like Rb1 and Rd are metabolized to IH-901 (86) (also known as compound K or M1) (Cho et al., 2009; Lee et al., 2000). Rg3 and Rg5, the principal components in steamed American ginseng, are transformed to Rh2 and Rh3, respectively (Shin et al., 2006a). PPT groups like Rg1 and Re are mainly converted to Rh1, F1, and the aglycone PPT (Hasegawa, 2004; Tawab et al., 2003).

As can be seen in Fig. 2, using 20(S)-PPT as the substrate, four new metabolites (87–90) resulted from microbial biotransformation via the fungus *Mucor spinosus* (Tian et al., 2005). Biotransformation of 20(S)-PPD by the fungus *M. spinosus* yielded eight metabolites (91–98) (Li et al., 2009b). The results suggest that *M. spinosus* selectively catalyzed the specific C-12 dehydrogenation of ginsenosides and could catalyze hydroxylation at different positions. Additionally, *Mycobacterium sp.* selectively catalyzed the specific C-3 dehydrogenation of ginsenosides (Wang et al., 1997). These regiospecific hydroxylation reactions may be important to increase bioactivity (Li et al., 2009b; Tian et al., 2005). *Abisidia coerulea* selectively transformed the PPD group rather than the PPT group of ginsenosides, and produced a series of C-20 side-chain-modified metabolites (Chen et al., 2007).
3. Most studied pharmacological activities

Like Asian ginseng, American ginseng has multiple pharmacological actions. In general, antioxidant, anti-inflammatory, and immunostimulatory activities seem to be related to the possible mechanisms of ginseng. In this section, we report the recent preclinical and clinical advancements in the study of American ginseng and the effects of its ginsenosides on the central nervous system, cardiovascular system, and antidiabetic and anticancer activities (Fig. 3). American ginseng’s immunomodulatory effects and prophylactic effect on acute respiratory illness are not discussed here (McElhaney et al., 2004; Predy et al., 2006; Vohra et al., 2008).

3.1. Effects on the central nervous system

Ginseng exerted beneficial effects on aging, central nervous system (CNS) disorders, and neurodegenerative diseases (Christensen et al., 2009). Ginsenosides played a major role in these effects (Rausch et al., 2006). The protective effects of ginsenosides Rb1 (Chen et al., 2008d; Yuan et al., 2007), Rg1 (Liu et al., 2010), Rg3 (Tian and Fu, 2006), Rd (Ye et al., 2009), and Re (Chen et al., 2008c) on neurodegeneration have been well studied in animals and in neuronal cell cultures.

American ginseng and ginsenosides enhanced cognitive performance and mood (Bao et al., 2005; Wang et al., 2009b; Zhang et al., 2008; Zhao and Li, 2004). Long-term ginsenoside administration to mice prevented memory loss or impairment (Zhao et al., 2009a; Zhao et al., 2009b). Corsi block and calmness were enhanced after administration of American ginseng to healthy young adults (Scholey et al., 2010).

Ginseng and ginsenosides can rescue neuronal cells by increasing cell survival, extending neurite growth, and rescuing neurons from death either in vivo or in vitro (Radad et al., 2004b; Radad et al., 2006; Rausch et al., 2006). The beneficial effect of ginseng and ginsenosides were shown on neurodegenerative disease models of Parkinson’s and Alzheimer’s diseases (Xu et al., 2005; Xu et al., 2009). Possible mechanisms were inhibition of uptake of MPTP and its active metabolite MPP⁺ in dopaminergic neurons (Van Kampen et al., 2003), suppression of oxidative stress (Chen et al., 2003), attenuation of MPP⁺-induced apoptosis (Xu et al., 2005), potentiation of nerve growth factor (Radad et al., 2004a), and activation of the insulin-like growth factor-I receptor signaling pathway (Xu et al., 2009).

Ginsenosides regulated various types of ion channels by interacting with ligand-binding sites or channel pore sites in neuronal and heterologously expressed cells (Nah, et al., 2007). They inhibited voltage-dependent Ca²⁺, K⁺, and Na⁺ channel activities in a stereospecific manner (Liu et al., 2010). Ginsenosides also inhibited ligand-gated ion channels such as N-methyl-D-aspartate, some subtypes of nicotinic acetylcholine, and 5-hydroxytryptamine type 3 receptors (Chen et al., 2010). Ginsenosides also modulated neurotransmission in the brain (Liu et al., 2010; Xue et al., 2006).

When the effects of Rg1 and Rb1 were compared (Joo et al., 2005), both enhanced CNS activities, but there were some differences in pharmacology and mechanism (Cheng et al., 2005b; Liao et al., 2002). For example, Ginsenoside Rb1 promoted neurotransmitter release by a cAMP-dependent protein kinase pathway (Xue et al., 2006); Rg1 produced this effect through a protein kinase II-dependent signaling pathway (Liu et al., 2010). Compared with Rg1, Rb1’s effects were weaker and in some cases even produced an inhibitory effect on the CNS (Chen et al., 2008a). The neuroprotective differences between American and Asian ginseng have not been reported. Since American ginseng has a lower ratio of Rg1/Rb1, it seems to calm the CNS. In contrast, Asian ginseng appears to stimulate the CNS.
3.2. Cardiovascular activities

In the U.S. American ginseng is a popular herbal supplement for patients suffering from cardiovascular disease (Xie et al., 2005a; Wang et al., 2007b). Several anti-ischemic, anti-arrhythmic and anti-hypertensive effects have been observed after the use of American ginseng (Wang et al., 2007b). The pharmacological effects may be produced by the antioxidant properties of the herb (Xie et al., 2005a). The antioxidant activities and the relationship between chemical structure and cardiovascular-protecting functions have been reviewed (Prior and Cao, 2000; Wang et al., 2007b). American ginseng extract had a stronger antioxidant activity than Asian ginseng root (Shao et al., 2004). American ginseng root or berry extract showed antioxidant and protective effects in cultured cardiomyocytes by up-regulating peroxide detoxifying mechanisms (Mehendale et al., 2006; Shao et al., 2004) and activating the Nrf2 pathway (Li et al., 2010b). Ginsenoside Re was one major antioxidant agent that protected cardiomyocytes by scavenging H$_2$O$_2$ and hydroxyl radicals (Xie et al., 2006).

In a randomized, double-blind, placebo-controlled trial of 16 hypertensive individuals given 3 g of American ginseng powder or placebo, American ginseng exerted a neutral acute effect on blood pressure (Stavro et al., 2005). In another clinical trial in 52 hypertensive individuals who took American ginseng 12 weeks, there was no effect on 24-hour blood pressure and renal function (Stavro et al., 2006). Taking American ginseng for 4 weeks (1.6 g/day) before subjects on a treadmill reduced the leakage of creatine kinase during exercise but did not enhance aerobic work capacity (Hsu et al., 2005).

3.3. Antidiabetic effects

Type 2 diabetes, accounting for over 90% of diabetic cases, is a syndrome with disordered metabolism of carbohydrates and lipids because of resistance to insulin action and impaired insulin secretion (Qi et al., 2010a). Both Asian ginseng and American ginseng root showed hypoglycemic effects in diabetic mice models (Attele et al., 2002; Dey et al., 2003; Xie et al., 2002). Using the ob/ob mouse model, we demonstrated that a 12-day treatment of American ginseng leaf and berry extracts decreased fasting blood glucose, improved glucose disposal, and reduced body weight (Attele et al., 2002; Xie et al., 2004a; Xie et al., 2004b). Heated American ginseng had stronger effects than unprocessed ginseng in inhibiting advanced accumulation of glycation endproducts in the diabetic rat kidney (Kim et al., 2007a).

Antidiabetic effects of ginsenosides have been demonstrated in animal models by Rb1 (Shang et al., 2007), Re (Xie et al., 2005b), transformed compounds such as Rb2 (Yokozawa et al., 1993), Rh2 (Lee et al., 2006), compound K (Yoon et al., 2007), and the aglycone 20(3)-PPT (Han et al. 2006). They decreased oxidative stress (Lin et al., 2005; Lin et al., 2008), activated peroxisome proliferator-activated receptor γ, increased GLUT expression, and enhanced PKA-dependent pathways (Park et al., 2008; Shang et al., 2008).

A series of randomized, placebo-controlled acute clinical studies were conducted to evaluate the efficacy of American ginseng in lowering postprandial glycemia in subjects with and without diabetes (Vuksan et al., 2000a; Vuksan et al., 2001a; Vuksan et al., 2000b; Vuksan et al., 2000c). American ginseng demonstrated a good acute safety profile. Escalation of dose and time of administration offered no added benefit in people with diabetes. A time, but not dose-dependent effect was observed in healthy individuals, suggesting that people without diabetes are sensitive to the time of ginseng administration. The effects of 1 g of ginseng extract on glycemic control were tested by a placebo-controlled, crossover trial in subjects with type 2 diabetes (Vuksan and Sievenpiper, 2005; Vuksan et al., 2001b). Fasting
glucose and HbA1c were decreased in the extract group compared with the placebo group after 8 weeks. The trials were small, however, longer-term studies are needed.

Evidence indicates that the glycemia-lowering effect of ginseng root may be species dependent. In healthy humans, American ginseng lowered postprandial glycemia, red Asian had no effects, and Asian and wild American ginseng raised glycemia (Sievenpiper et al., 2004b). The part of American ginseng that produced the hypoglycemic effects remains unclear. Some clinical evidence suggested that the ratio of protopanaxadiols to protopanaxatriols is inversely correlated with the glycemia-lowering efficacy of ginseng root. American ginseng with a relatively high ratio has a better effect on acute postprandial glycemic indices in healthy humans than does Asian ginseng (Sievenpiper et al., 2004b).

3.4. Cancer chemoprevention

Another pharmacological activity of American ginseng and its constituents is cancer chemoprevention and inhibition of tumor growth (Qi et al., 2010b; Wang et al., 2007a; Wang et al., 2009a). American ginseng extract enhanced the chemopreventive effect of 5-fluorouracil (Li et al., 2009d) in human colon cells, suppressed the chromosomal aberration induced by mitomycin C in mice (Pawar et al., 2007), improved cancer-related fatigue in clinic (Barton et al., 2010), and produced radioprotective potential in the lymphocytes of healthy individuals (Lee et al., 2008b; Lee et al., 2010).

Steamed American ginseng has more potent activity than white ginseng on human cancer cells (Wang et al., 2007a; Wang et al., 2006). Steamed ginseng berry extract inhibited colorectal cancer growth both in vitro and in vivo (Xie et al., 2009). Enhanced anticancer potential results from chemical degradation and conversion of the original saponins to new compounds during the steaming process (Wang et al., 2007a; Wang et al., 2009a). Because of higher total ginsenoside concentration, American ginseng had stronger anticancer potential than Asian ginseng (Sun et al., 2010).

The mechanism and cellular/molecular targets of American ginseng against cancer have been studied. Several molecular mechanisms exist and collectively converge on various signaling pathways. These pathways include the regulation of the cell cycle (Wang et al., 2007a), induction of apoptosis (Wang et al., 2006; Wang et al., 2009a), inhibition of angiogenesis (Sengupta et al., 2004; Yue et al., 2006), preventing invasion (Kim et al., 2007b), and reduction of inflammatory response (Jin et al., 2008; Jin et al., 2010). A series of cell cycle proteins, apoptosis-related proteins, growth factors, protein kinases and transcription factors are affected by American ginseng and ginsenosides (King and Murphy, 2010; Kim et al., 2007b; Lee et al., 2000; Peralta et al., 2009; Sengupta et al., 2004; Yue et al., 2006). For example, American ginseng extract can selectively inhibit the expression of the inducible nitric oxide synthase via suppression of signal transducer and activator of transcription cascade in inflamed macrophages (Ichikawa et al., 2009). A lyophilized aqueous extract of American ginseng inhibited induced cyclooxygenase-2 and NF-kappa B activation in breast cancer cells (Peralta et al., 2009). The anticancer effect of steamed American ginseng was enhanced by antioxidants or inhibitors of the NF-kappa B pathway (Li et al., 2010a).

Because tumor malignancy is a complex interaction among genes, cells, and tissues (Aggarwal et al., 2009), there are probably many unknowns in the anticancer mechanisms of ginseng. Because of complex chemical composition and difficulty in reproducibility, most studies focus on individual ginsenosides but not American ginseng extract. Therefore, more scientific clinical trials are needed to test the effects of American ginseng and steamed ginseng against cancer.
4. Potential structural-activity relationship

4.1. Positive relationship of sugar moieties in ginsenosides with antioxidant activity

Oxidative stress contributes to the development of a wide range of diseases: neurodegenerative disorders, cardiovascular diseases, diabetes, cancer, and chronic fatigue syndrome (Giustarini et al., 2009; Heistad et al., 2009). Ameliorating oxidative stress with antioxidants might be an effective strategy for treating various diseases (Giustarini et al., 2009). American ginseng extract exhibited antioxidant activity in lipid and aqueous mediums by both chelation of metal ions and scavenging of free radicals (Kitts et al., 2000). In clinical surveys American ginseng supplementation reduced oxidative stress markers in healthy volunteers (Lee et al., 2008a). Most of the beneficial effects of American ginseng and ginsenosides are partly attributable to their antioxidant and chelating abilities (Liu et al., 2003; Zhao et al., 2009b). The radical scavenging, chelation and oxidant activity of ginsenosides depends upon their sugar moieties and linkage positions, the types of aglycone, and total number of hydroxyl groups. In contrast to flavonoids, ginsenosides are more potent antioxidants than their corresponding glycosides, and sugar moieties are positively correlated to their activities.

Liu et al. (2003) observed that ginsenoside aglycones (i.e., protopanaxadiol and protopanaxatriol), ginsenosides Rg2, Rg3 and Rh2 were prooxidative; ginsenosides Rb1, Rc, R1, Rd, Re, Rb3, Rg1, and Rh1 functioned as antioxidants, and Rc protected human erythrocytes mostly against hemin-induced hemolysis (Li and Liu, 2008). A recent study evaluated the antioxidant activities of ginsenosides on the intracellular reactive oxygen species (ROS) and the radical scavenging activity by a 2’,7’-dichlorodihydrofluorescin diacetate (DCF-DA) method (Chae et al., 2010). Results showed that ginsenosides Rb2 and Rc effectively inhibited intracellular ROS better than ginsenosides Rb1, Rd, Re, Rf, Rg1, Rg2, Rg3, Rh1 and Rh2. The presence of arabinose linked at the glucopyranosyl group may have enhanced the antioxidant activity.

For identification of the active part or point of interaction, some sugar moieties were selected to test hydroxyl scavenging activity. Sugars showed no hydroxyl scavenging activity (Kang et al., 2007). Any sugar substituent that occupies free hydroxyl groups is capable of increasing hydrophilicity and altering access to lipid peroxyl and alkoxyl radicals in membranes (Heim et al., 2002). For antioxidant activity, there might be complicated interactions within one molecule of ginsenoside between sugar moieties and the triterpene dammarane.

4.2. Negative correlation of sugar molecules in ginsenosides to cancer chemoprevention

Sugar molecules within a ginsenoside impact tumor cells. The structure-function relationship of sugar molecules in ginsenosides to anticancer activity has been reviewed (Qi et al., 2010b). In general, anticancer activity is inversely correlated to the number of sugars, i.e., anticancer activities increase with the decrease of sugar number. Ginsenosides Rg3, Rh2, IH-901 or compound K, and ginsenoside aglycones (i.e., PPD and PPT) have remarkable effects on inhibition of various cancer cell growth (Musende et al., 2009). They induce apoptosis (Cheng et al., 2005a), perturb cell cycle events (Choi et al., 2009), prevent invasion and metastasis (Hasegawa et al., 2002), block angiogenesis (Chen et al., 2008b), reverse P-glycoprotein-mediated multidrug resistance (Kim et al., 2003), and produce synergistic effects with conventional chemotherapy agents (Yu et al., 2007). The presence of sugar moieties may reduce the hydrophobic character of the compounds and decrease their permeability to cell membranes. These properties are required to interact with specific membrane proteins or to pass into the nucleus (Ha et al. 2010; Li et al., 2009c).
Sugar linkage positions also affect anticancer activities. The anticancer activity of ginsenosides with a sugar substitute at C-6 is attenuated compared to activity of ginsenosides with sugar linkages at C-3 or C-20 (Li et al., 2009c; Popovich and Kitts, 2002). Any sugar moiety at C-6 may increase steric hindrance and block compounds from extracellular binding to their targets, thus significantly reducing the anticancer activities of ginsenosides (Chen et al., 2009b).

4.3. Stereoselectivity of 20(S) but not 20(R) of ginsenosides in bioactivities

20(S) and 20(R) are stereoisomers of each other that depend on the position of the C-20 hydroxyl in ginsenosides. The different stereochemistries of the 20(S)- and 20(R)-ginsenosides produce different pharmacological effects. 20(S)-Rg3 is more soluble in water than 20(R)-Rg3 (Kang et al., 2007). The hydroxyl radical scavenging activity of 20(S)-Rg3 is higher than that of 20(R)-Rg3 (Lee et al., 2008c). 20(S)-Rg3 is a more efficient regulator of voltage-dependent Ca\(^{2+}\), K\(^{+}\) or Na\(^{+}\) channels (Kang et al., 2005). Rg3 provided neuroprotection against ischemia-induced injury in rat brain by reducing lipid peroxides, scavenging free radicals and improving the energy metabolism (Tian and Fu, 2005). 20(S)-ginsenosides had stronger cytotoxicity effects than their 20(R)-stereoisomers (Popovich and Kitts, 2002; Qi et al., 2010b). In some exceptional examples, 20(R)-Rh2 had a stronger inhibitory effect on osteoclast formation than did 20(S)-Rh2 (Liu et al., 2009). 20(R)-Rg3 inhibited cancer cell invasion and metastasis through angiosuppression (Chen et al., 2008b; Yue et al., 2006).

20(S)-OH is geometrically close to the C-12 hydroxyl of ginsenosides; 20(R)-OH is far from the C-12 hydroxyl (Jeong et al., 2004). 20(S) ginsenosides tend to process the geometrical arrangement of the hydroxyl groups at carbon-12 and -20 (Jeong et al., 2004; Kang et al., 2006). The alkene chain connected to carbon-20 in 20(S)-ginsenosides has a stable, fixed orientation and is packed tightly near the terpenoid. The chain in 20(R)-ginsenosides protrudes further outside and has a flexible structure (Kang et al., 2005; Kang et al., 2007). 20(S)-ginsenosides are thus inaccessible to water molecules because of the alkene chain, which may stabilize hydrogen bonding between these hydroxyl groups and receptors (Kang et al., 2005; Kim et al., 2005). Compact packing around the chiral center of 20(S)-Rg3 aids in hydrophobic interactions with the hydrophobic pocket of the receptor (Kang et al., 2005; Kang et al., 2006). Therefore, the geometrical arrangement of hydroxyl groups at the chiral centers, inaccessibility to water, hydrophobic interactions, and compact structure may be the crucial factors accounting for the stereospecific action of 20(S) ginsenosides.

5. Summary and future perspectives

To date, correlation of various structures to specific pharmacological activity is somewhat limited. The structural heterogeneity of ginsenosides, their multiple mechanisms of action, and the diverse experimental methods used to evaluate their activities pose challenges in assembling a collective hierarchy of SAR. In this article we summarized some views regarding sugar moieties with antioxidant activity, sugar molecules with cancer chemoprevention, and stereoselectivity. SAR comparisons of homologs that differ in a single structural attribute are still needed to generate consistent lines of evidence to support the role of specific structural components as requisites in ginsenosides for their activities. A better understanding of the structure-activity relationships is required for helpful modifications to produce novel agents in medical oncology.

A total of 98 ginsenosides have been identified from American ginseng, including naturally occurring compounds and those resulting from steaming or biotransformation. With the development of chemical and analytical techniques and the characterization of novel compounds, the diversity of ginseng saponins is constantly revealed. The ginsenoside family
can also be expanded through the characterization of novel compounds from a closely related genus like *Oplopanax* (Huang et al., 2010; Li et al., 2010c). Chemical modification further produces a series of novel compounds and expands the targets for the pharmacological activities of ginsenosides. Although many ginsenosides have been characterized from American ginseng, their potential effects have not been quantitatively compared under standard conditions.

Multiple pharmacological actions of American ginseng have been observed on the central nervous, cardiovascular, endocrine, and immune systems. Their neuroprotective, cardioprotective, antidiabetic, antioxidant and anticancer properties have been reviewed above. Reports of the effectiveness of ginseng are sometimes contradictory, perhaps because the chemical content of ginseng root or root extract differs, depending on the method of extraction, subsequent handling, or even the season of its collection. The high variability in ginsenoside composition of ginseng among different species and batches may contribute to equally high variability in efficacy (Vuksan and Sievenpiper, 2005). For example, five batches representative of Ontario-grown American ginseng root produced comparable reductions of postprandial glyceremia in healthy individuals; yet 40% of batches may not exert antihyperglycemic activity (Dascalu et al., 2007; Sievenpiper et al., 2004a). American ginseng with a similar profile could have similar efficacy. Unmeasured components such as different peptidoglycans (quinquefolans for American ginseng), various ginsenans, peptides, polysaccharides, fatty acids, and other organic compounds may be active.

Obviously, we must know more to answer the questions about the observed effects of ginseng in complementary and alternative medicine. In the future, widespread interest in American ginseng seems certain to ensure continued research with this herb. With the trend of interdisciplinary research and the development of modern combinatorial techniques, the possibility of gaining novel agents from ginseng seems promising.

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Fig. 1.
Typical chromatograms of Asian ginseng and American ginseng extract by HPLC-UV. Pseudoginsenoside F11 has the same molecular weight and retention times similar to those of ginsenoside Rf. It cannot be detected by UV because there are no chromospheres. Chromatographic and analytical conditions were shown in Sun et al. (2011).
Fig. 2.
Ginsenosides characterized from American ginseng. PPD, protopanaxadiol; PPT, protopanaxatriol; G, ginsenoside; Q, quinquenoside; F, floralquinquenoside; NG, notoginsenoside; QF, quinquefolioside.
Fig. 3.
Biological and pharmacological activities of American ginseng and ginsenosides.
Comparison of typical ginsenoside composition of American ginseng (*Panax quinquefolius* L.) and Asian ginseng (*Panax ginseng* C. A. Meer).

<table>
<thead>
<tr>
<th>Chemical composition</th>
<th>American ginseng</th>
<th>Asian ginseng</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total ginsenosides</td>
<td>40–60 gram/kilogram</td>
<td>20–40 gram/kilogram</td>
</tr>
<tr>
<td>Major ginsenosides</td>
<td>Rb1, Re, Rd</td>
<td>Rb1, Rg1, Rb2</td>
</tr>
<tr>
<td>Pseudoginsenoside F11</td>
<td>1.0–2.0 gram/kilogram</td>
<td>0</td>
</tr>
<tr>
<td>Ginsenoside Rf</td>
<td>0</td>
<td>1.0–2.0 gram/kilogram</td>
</tr>
<tr>
<td>PPD-group to PPT-group</td>
<td>&gt; 2.0</td>
<td>&lt; 2.0</td>
</tr>
<tr>
<td>Rb1: Rg1</td>
<td>&gt; 5.0</td>
<td>&lt; 5.0</td>
</tr>
<tr>
<td>Rg1: Re&lt;sup&gt;a&lt;/sup&gt;</td>
<td>&lt; 1.0</td>
<td>&gt; 1.0</td>
</tr>
<tr>
<td>Rb2: Rc</td>
<td>&lt; 0.4</td>
<td>&gt; 0.4</td>
</tr>
</tbody>
</table>

<sup>a</sup> Limited American ginseng population with another phytotype was not involved in this table (Schlag and McIntosh, 2006).