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Review

Dihydromyricetin: A review on identification and quantification methods, biological activities, chemical stability, metabolism and approaches to enhance its bioavailability

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ABSTRACT

Background: Dihydromyricetin (DMY) is an important plant flavonoid, which has received great attention due to its health-benefiting activities, including antioxidant, antimicrobial, anti-inflammatory, anticancer, antidiabetic and neuroprotective activities. DMY capsules have been sold in US as a nutraceutical supplement to prevent alcoholic hangovers. The major disadvantage associated with DMY is its chemical instability and poor bioavailability caused by the combined effects of its low solubility and poor membrane permeability. This limits its practical use in the food and pharmaceutical fields.

Scope and approach: The present paper gives an overview of the current methods for the *identification* and quantification of DMY. Furthermore, recent findings regarding the main biological properties and chemical stability of DMY, the metabolism of DMY as well as different approaches to increase DMY bioavailability in both aqueous and lipid phases are discussed.

Key findings and conclusions: Current trends on identification and quantification of DMY have been focused on spectral and chromatographic techniques. Many factors such as heat, pH, metal ions, could affect the chemical stability of DMY. Despite the diverse biological effects of DMY, DMY faces with the problem of poor bioavailability. Utilization of different delivery systems including solid dispersion, nanocapsule, microemuslion, cyclodextrin inclusion complexes, co-crystallization, phospholipid complexes, and chemical or enzymatic acylation has the potential to improve both the solubility and bioavailability. DMY digested in laboratory animals undergoes reduction, dehydroxylation, methylation, glucuronidation, and sulfation. Novel DMY delivery systems and basic pharmacokinetic studies of encapsulated DMY on higher animals and humans might be required in the future.

1. Introduction

As a class of polyphenol secondary metabolites, flavonoids are frequently found in plants and foods. They possess various bioactive effects including antioxidant, antibacterial, antiviral, anti-inflammatory, anti-cancer and neuroprotective activities, etc (Wang, Li, & Bi, 2017).

Dihydromyricetin (DMY) or ampelopsin is a major bioactive flavonoid isolated from a traditional Chinese medicinal plant *Ampelopsis grossedentata* (*A. grossedentata*) (Zheng & Liu, 2006) and it is also found in various plant-based foods such as grapes and red bayberry (Gadetskaya, Tarawneh, & Zhusupova, 2015; Wu,Ma, & Li, 2015). DMY was first isolated from *Ampelopsis meliaefolia* by Kotake and Kubota in 1940, and was later reported as a major bioactive component in *A. grossedentata* (Zhang, Yang, & Xiong, 2001; Kou & Chen, 2012). The content of DMY was as high as 30–40% (w/w) in *A. grossedentata* (Tian, Zhang, Yang, Yang, & Gong, 2002; Gao, Lee, Li, & Lee, 2016). Other major sources of DMY also include medical plants such as *Hovenia dulcis* (Yoo, Mun, & Kim, 2006; Chaturvedula & Ruo, 2013) and *Cedrus deodara* (Liang, Shen, et al., 2014).

Due to its beneficial activities, there have been extensive studies on DMY structure identification, content determination, as well as pharmacological effects (Kou & Chen, 2012; Li et al., 2017). On one hand, a number of spectral (Ignat, Volf, & Popa, 2011) and chromatographic methods (Ignat et al., 2011; Naczk & Shahidi, 2006) have been

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developed for analysis and quantification of DMY. On the other hand, numerous research efforts have been devoted to the study of biological and pharmacological activities of DMY, such as antioxidative, antibacterial, anti-inflammatory, anti-cancer, anti-obesity and neroprotective effects, etc (Kou & Chen, 2012; Li et al., 2017). Nowadays, DMY prescription is specially recommended for the treatment of various diseased conditions such as alcohol use disorders and metabolic or neuro imbalance (Li et al., 2017). Presently, DMY capsules are sold in the United States as a nutraceutical supplement to prevent alcohol hangovers (DSLD, 2016).

The major disadvantages associated with DMY use is chemical instability and poor bioavailability. DMY is soluble only in hot water and ethanol and slightly soluble in water under room temperature (0.2 mg/mL at 25 °C), which is the main cause of its poor membrane permeability ($P_{eff} = (1.84 \pm 0.37) \times 10^{-6}$ cm/s) and bioavailability (Tong et al., 2015; Wang, Tong, et al., 2016). This is a determinant factor that limited the pharmacological effects and clinical application of DMY. To improve the bioavailability of DMY, researchers have tried to use DMY in new drug delivery systems such as inclusion complexes (Liu, Ma, et al., 2012; Ruan, Yu, Fu, & Zhu, 2005; Yang, Liu, Liu, & Zhang, 2011), nano-encapsule (Dalcin et al., 2017) or microemulsion (Solanki, Sarkar, & Dhanwani, 2012), co-crystals (Wang, Tong, et al., 2016), phospholipid complexes (Liu, Du, Jie, Chen, & Niu., 2009), and acylation (Cao et al., 2017; Guo, Zeng, Lu, & Shu, 2013; Li et al., 2015; Li, Zheng, & Ning., 2005) to provide higher solubility and bioavailability. In addition, the pharmacokinetic characteristics of DMY in animal models and human body are also vital to the evaluation of their *in vivo* bioavailability efficacy. However, only partial information (Fan, Tong, & Dong, 2017; Xiang, Fan, & Hou, 2018; Zhang et al., 2007) is available on basic pharmacokinetic studies of DMY such as absorption, distribution, metabolism and excretion in organisms, biotransformation processes and metabolites.

The objective of this article is to give an overview of the current methods for analysis and quantification of DMY, as well as recent findings regarding the main biological properties and chemical stability of DMY. Special attention is paid to the metabolic pathways of DMY and different approaches carried out to increase DMY bioavailability in both aqueous and lipid phases.

2. Analysis, identification and quantification of DMY

The analysis, characterization and quantification of flavonoids in natural sources present a challenge for many researchers (Marston & Hostettmann, 2006). Spectral techniques and chromatographic techniques have been developed for the determination of DMY in the plants, foods and biological samples.

2.1. Spectral methods

A number of spectral methods have been developed for quantification of total flavonoids and DMY. The complexation of flavonoids with Al(III) is the principle of spectrophotometric assays used for quantification of total flavonoids (Naczk & Shahidi, 2006). Simple flavonoids normally have maximal absorption between 220 and 300 nm (Owades, Rubin, & Brenner, 1958). The maximal absorption of DMY was observed at ~290 nm. The role of its C_2–C_3 double bond, whose presence was responsible for the absorption bands in the ultraviolet (UV) regions (Biler, Biedermann, Valentova, Kren, & Kubala, 2017). The UV absorption was also affected by the nature of solvent employed and the pH of the solution. High pH (above 11 for DMY) induced the formation of a long-wavelength peak arising from double and/or triple deprotonation (Biler et al., 2017). Moreover, there was a possibility of interference by UV-absorbing substances such as proteins, nucleic acids and amino acids. Another spectral method Fourier transform infrared spectroscopy (FT-IR) has been used for structural elucidation of complex flavonoids isolated from natural resources (Jeon, Chun, Choi, & Kwon, 2008; Wang, Xiong, & Perumalla, 2016). FT-IR has been applied to analyze the isolated compound DMY from the bark of *Salix hulteni*. In a recent paper reported by Wang, Xiong, and Perumalla (2016), FT-IR has been used for quickly distinguishing homochiral (+)DMY from racemic (+/−)DMY extracted from *A. grossedentata* leaves. The physicochemical properties of the two phases were also assessed using FT-IR. Nuclear magnetic resonance (NMR) spectroscopy and mass spectrometry (MS) are also used for elucidating the chemical structures of flavonoids. Standard ^{1H}, ^{13}C NMR spectra can give a wealth of chemical information for identifying flavonoids. MS analysis is based on ionizing chemical species and sorting the ions based on their mass-to-charge ratios (Ignat et al., 2011). By using ^{1}H and ^{13}C NMR and MS, purity and steric integrity of DMY were assessed (Outtrup, Schaumburg, & Madsen, 1985). Recently, the active compound was identified as 2R, 3R-dihydromyricetin from pine needles of *Cedrus deodara* based on MS and NMR data (Liang, Wu, Qiu, Zhong, & Gao, 2014).

2.2. Chromatographic methods

The identification and quantification of specific flavonoid compounds like DMY can be performed with the following chromatographic techniques like thin-layer chromatography (TLC), high-performance liquid chromatography (HPLC), high-speed counter current chromatography (HSCCC), and high-performance counter current chromatography (HPCCC). TLC is a rapid and simple method for the qualitative analysis of flavonoids in plant extracts (Ignat Volf, & Popa, 2011). He, Pei, and Zhou (2000) determined the content of DMY in *A. grossedentala* by using a TCL-scanner. The implementation of a modern standardized methodology led to an increasing acceptance and recognition of DMY.

HPLC is another preferred method for the identification and quantification of several flavonoids due to its simplicity and sensitivity. The HPLC conditions mainly include the use of reverse phased C18 column, a binary solvent gradient, and different detection systems such as Diode Array Detection (DAD), MS or NMR (Marston & Hostettmann, 2006). HPLC methods have been successfully applied in DMY identification and quantification in healthy food for anti-hangover and hepatoprotection (Zou, Zhou, & Sun, 2017), in the fruit-stalk extract of *H. dulcis* (Park, Kim, & Rehman, 2016), in Yeputaoteng (Jin, Ding, & Zhang, 2014) and in red wheat (Kohyama, Chono, & Nakagawa, 2017). HPLC was also used for analysis of DMY content extracted from *A. grossedentata* by different extraction methods (Li, Li, & Zhang, 2008).

Other interesting chromatographic alternatives for the analysis of flavonoid compounds include HSCCC (Du, Cai, & Xia, 2002; Gao, Ma, & Chen, 2017; Ma, Zhou, & Tong, 2017) and HPCCC (Vieira et al., 2016). HSCCC is an all-liquid chromatographic technique very suitable for preparative isolation of pure compounds (Ignat et al., 2011). Recently, HSCCC was successfully used for rapid and efficient separation of antioxidants including DMY from vine tea (*A. grossedentata*) (Gao et al., 2017; Ma et al., 2017). Furthermore, Purification of (+)-DMY from an extract of *A. grossedentata* leaves was performed using a triple-column HSCCC. With a solvent system composed of n-hexane-ethyl acetate-methanol-water (1:3:2:4, v/v) 11.3 g of (+)-DMY was obtained at a high purity of over 99% (Du et al., 2002). HPCCC is based on a support-free liquid–liquid centrifugal partition chromatography, which could be applied to the fast fractionation and recovery of flavonoid from plant extract. A developed HPCCC protocol was successfully used to isolate the main flavonoids including DMY from the crude extract of *Impatiens glandulifera Royles* (Vieira, Winterhalter, & Jerz, 2016).

Recently, the coupling of chromatographic techniques to MS or NMR has achieved growing importance for the structural elucidation of complex flavonoid compounds in natural products and biological samples. By using a HPLC-tandem electrospray ionization mass spectrometry (ESI-MS) method, the major flavonoid extracts from *A. grossedentata* leaves by traditional solvent extraction and recrystallization was identified as (+)-DMY (Gao, Liu, & Ning, 2009). A newly established ultra-high performance liquid chromatography-quadrupole mass

spectrometry method (UHPLC-MS) was successfully used for simultaneous quantification of 10 *X. sorbifolia* constituents. It was found that the wood of *X. sorbifolia* was rich in phenolic compounds with the contents of DMY being 6.76–7.89 mg/g (Zhang, Ma, & Ma, 2015). In a recent study, UPLC-MS was successfully used for analysis of the fruit metabolome DMY in *Actinidia arguta* cultivars (Li, Fang, & Qi, 2018).

Besides the natural products, there are also some reports on determining DMY in biological sample by chromatographic methods. HPLC methods have been previously reported for the determination of DMY in rat plasma after orally administrating the decoction of *A. grossedentata* (Zhang et al., 2007) Recently, a bioanalytical method with liquid chromatography (LC)–MS/MS has been developed and validated for the quantitation of DMY in rat plasma (Liu, Yin, & Wang, 2017; Tong et al., 2015; Wu, Ma, & Zhang, 2018; Yang, Yan, & Liu, 2019), for monitoring uptake and transport of DMY in human intestinal Caco-2 cells (Xiang et al., 2018) as well as for pharmacokinetic studies of DMY in organisms including absorption, distribution, metabolism and excretion (Fan et al., 2017). In contrast with HPLC, LC–MS/MS is more rapid, more sensitive for the biological analysis.

2.3. Application of capillary electrophoresis

Capillary electrophoresis (CE) is another alternative analytical tool for separation and quantification of flavonoids based on the electrophoretic migration of charged analytes (Marston & Hostettmann, 2006). This technique offers high separation efficiency and resolution power, short analysis time and low consumption of sample (Ignat et al., 2011). CE techniques have a great potential for identification of complex multicomponent mixture at the same time with good reproducibility and sensitivity (Naczk & Shahidi, 2006). Zou, Zhou, and Sun (2016) utilized CE for simultaneous determination of 7 components including DMY in functional food for anti-hangover and hepatoprotection. A baseline separation for all the target components within 8 min was achieved. The results shown that the method could meet the requirement for quality analysis.

3. Biological activities of DMY

DMY possesses diverse biological and pharmacological activities. Conventionally, DMY is used as antioxidant, antimicrobial, antiviral and anti-inflammatory agent. Current research has also shown its therapeutic benefits for the treatment of various chronic diseases such as cancer, diabetes, obesity, alcohol use disorders and metabolic or neuro imbalance (Kou & Chen, 2012; Li et al., 2017).

3.1. Antioxidant activity

Reactive oxygen species (ROS) including superoxide anion ($O2^{\bullet -}$), hydroxyl radicals ($OH^{\bullet}$) and hydrogen peroxide (H_2O_2) are produced in large amounts due to various reactions going inside human body. Oxidative damage induced by these free radicals can create deleterious effects on cells and tissues and may cause several biochemical problems including protein aggregation, DNA degradation, and oxidation of membrane lipids (Park, Chong, & Mi, 2016). Flavonoids are one of the most powerful scavengers of harmful free radicals (Burda & Oleszek, 2001). DMY has been shown to be a strong antioxidant agent both *in vitro* and *in vivo* (Table 1). DMY inhibited the increase of lipid peroxidation (LPO) in a concentration dependent manner in linoleic acid system (Zhang, Ning, Yang, & Wu, 2003). Gao, Liu, and Ning (2009) observed that antioxidant activity of flavonoid-rich extracts (DMY) from *A. grossedentata* leaves were comparable with that of tertiary butylhydroquinone (TBHQ) in a linoleic acid system. The antioxidant activity of DMY was also confirmed in lard oil (Zhao et al., 2009), soybean oil (Ye, Wang, Duncan, Eigel, & O'Keefe, 2015), sausage (Wang, Qin, Yang, Lu, & Qin, 2017) and cooked ground beef (Ye et al., 2015). The possible reason for its high antioxidant activity was due to the free hydroxyl in the C-3 position in C-ring and *o*-dihydroxy system (hydroxyl in the C-3′ and 4' position) in the B-ring, which has been shown strong effect against free radicals (Burda & Oleszek, 2001).

The antioxidant effects of DMY were also demonstrated in various cell-type assays and animal models. DMY exhibited antioxidant activity in oleic acid-induced lipid accumulation process in L02 and HepG2 cells due to a decrease in the levels of cellular triglycerides (TG), cholesterol (TC) and malondialdehyde (MDA) and an increase of superoxide dismutase (SOD) level (Xie et al., 2016).

In the case of human umbilical vein endothelial cells (HUVECs) (Hou et al., 2015a) and MG63 cell (Wang, Jiao, Zhou, & Liu, 2016), DMY demonstrated a protective effect against H_2O_2-induced oxidative stress. Similarly, DMY protected mouse kidney tissues against nephrotoxicity through attenuation of cisplatin-induced oxidative stress and inflammatory stress (Wu et al., 2016). Moreover, DMY increased the total antioxidant capacity (T-AOC) and attenuated ROS generation in Ang-II induced cardiac fibroblasts neonatal Sprague-Dawley rats model (Meng et al., 2015; Song et al., 2017) and low-density lipoprotein (LDL) receptor deficient mice (Liu, Zeng, et al., 2017).

3.2. Antimicrobial and antiviral activity

Flavonoids may serve as pharmacologically acceptable antimicrobial agents (Cushnie & Lamb, 2005). Plant flavonoid DMY from *A. grossedentata* (Liu, Pang, Ding, & Sun, 2016) and *Cedrus deodara* (Zeng, He, Sun, Zhong, & Gao, 2012) displayed strong antibacterial activities against both Gram-negative and Gram-positive bacteria. Liu et al. (2016) confirmed the inhibitory effect of DMY on Gram-negative food pathogen *V. parahaemolyticus*. The inhibitory effect increased with the increase of DMY concentration, the minimum inhibitory concentration (MIC) of DMY against *V. parahaemolyticus* was observed at 0.625 mg/mL. Membrane damage was assumed to be the main antibacterial mechanism of DMY. In addition, it was observed that DMY could bind to proline dehydrogenase (PDH), a key regulatory and rate-limiting enzyme in the metabolism of proline. DMY interacted with primary amino acid residues (Glu 292, Arg 288, Tyr285, Gly 64) located within the active hydrophobic pockets of PDH (Fig. 1a), leading to the decrease of PDH activity. The interference of normal proline metabolism by DMY was another possible reason for *V. parahaemolyticus* cell death (Ding, Xiao, Liu, & Pang, 2017).

DMY also exhibited antibacterial activity against Gram-positive bacteria *Staphylococcus aureus (S. aureus)*. The MIC of DMY against *S. aureus* was observed at 0.125 mg/mL. DMY not only changed membrane integrity, fluidity and membrane protein conformation, but also bound to intracellular DNA through the groove-binding mode in *S. aureus*. DMY achieved bactericidal activity by dual effects of cell membrane damage and DNA binding (Wu, Bai, Zhong, Huang, & Gao, 2017). However, in the study of Huang, Huang, Chen, Yang, and Huang (2015), the effect of DMY on the inhibition of *S. aureus* PriA (SaPriA), an essential helicase for DNA replication restart, was not so significant. Furthermore, Huang (2015) investigated the inhibitory effect of DMY on dihydropyrimidinase, a key member in the chain of pyrimidine catabolism and metabolism of DNA base in *Pseudomonas aeruginosa*. DMY significantly inhibited dihydropyrimidinase with IC_{50} values of 48 μM. DMY was docked in the active site pocket of dihydropyrimidinase and formed a stable complex with dihydropyrimidinase (Fig. 1b). As a competitive inhibitor of dihydropyrimidinase, DMY intervention led to inhibition of bacterial growth and promotion of cell death.

Medicinal flavonoids provide an opportunity for the discovery of human immunodeficiency virus (HIV) inhibitors with lower or no toxicity and/or side effects (Narayan, Rai, & Tewtrakul, 2013). DMY was proved to be a strong HIV inhibitor during HIV-1 absorption, incubation and acute infection by Liu et al. (2004). About 70% HIV-1 CXC-chemokine receptor 4 (CXCR4) was reduced by DMY at 1 mg/mL. The anti-HIV-1 effect of DMY was partly due to down-regulation of

Table 1
Antioxidant activity of DMY.

Study model	Method/Assay	Results	References
Linoleic acid system	DPPH and reducing power	Anti-oxidative activity of flavonoid-rich extracts (DMY) comparable with that of tertiary butylhydroquinone (TBHQ)	Gao et al. (2009)
linoleic acid	lipid peroxidation	DMY greatly inhibit the increase of lipid peroxidation (LPO) values in a concentration dependent manner	Zhang et al. (2003)
Lard oil	DPPH	Effective in quenching DPPH with IC50 of 21.48 μM. Superior to that of TBHQ	Zhao et al. (2009)
Soybean oil	Peroxide value, anisidine value, headspace volatiles	DMY was more potent than butylated hydroxyanisole (BHA) in preventing soybean oil oxidation	Ye et al. (2015)
Guizhou sausage	Peroxide value (POV) malondialdehyde (MDA)	Inhibit the oxidation of sausage and its antioxidant effect increased with the increase of the amount of DMY.	Wang et al. (2017)
Cooked ground beef	Thiobarbituric acid reactive substances	DMY showed a high antioxidant activity and comparable with that of BHA after treatment with longer time (Day 14).	Ye et al. (2015)
L02 and HepG2 cells	Oleic acid-induced lipid accumulation	DMY decreased cellular triglycerides (TG), cholesterol (TC) and MDA, increased the level of superoxide dismutase (SOD)	Xie et al. (2016)
HUVECs	SOD,MDA, ROS, nitric oxide (NO)	DMY inhibited intracellular ROS overproduction and attenuated H_2O_2-induced decrease in cell viability and apoptosis,	Hou et al. (2015)
MG63 cells	H_2O_2-induced oxidative stress	30 μM dose of DMY prevents hydrogen peroxide induced reduction in viability and apoptotic alterations	Wang et al. (2016)
Cisplatin-treated mouse model	SOD, MDA, catalase activity (CAT)	DMY decreased MDA level and increased CAT and SOD activities in mouse kidney tissues after treatment with cisplatin	Wu et al. (2016)
Neonatal rat cardiomyocytes	ROS,MDA,SOD, T-AOC	Reduced levels of MDA, increased SOD activity, T-AOC (total antioxidant capacity) and NO bioactivity by DMY	Meng et al. (2015)
Ang-II induced cardiac fibroblasts	ROS,MDA,SOD, T-AOC	DMY significantly decreased ROS production and MDA level, while increased the SOD activity and T-AOC.	Song et al. (2017)
LDL receptor deficient mice	ROS,MDA,SOD, CAT, glutathione (GSH)	DMY decreased ROS production and MDA level, increased SOD, GSH and CAT levels.	Liu, Zeng, et al. (2017)

CXCR4 on the surface of target cells. Grand, Garofalo and Neamati (2008) also confirmed CXCR4 was the major HIV co-receptors and promising targets for anti-HIV drugs. Moreover, Ren and Song (2005) observed that the combined use of myricitrin and DMY has shown great promise in resisting different kinds of virus such as *hepatitis B* virus, influenza virus and/or coronavirus.

3.3. Anti-inflammatory activity

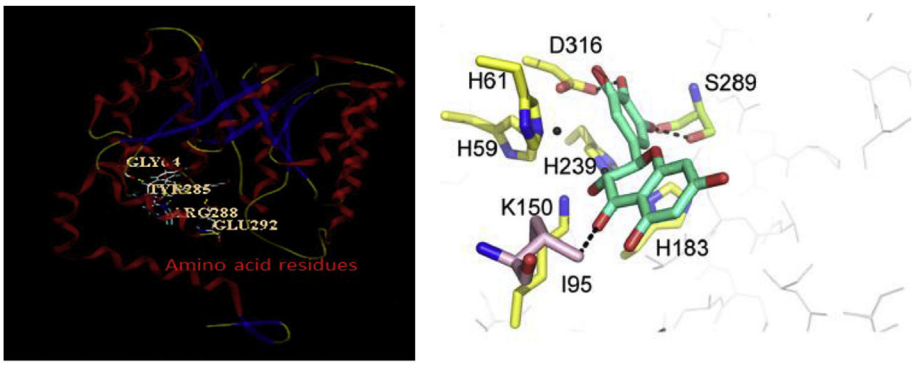
Anti-inflammatory effects of flavonoids are closely linked with its antioxidant effect (Shoham, Hadziahmetovic, Dunaief, Mydlarski, & Schipper, 2008). ROS not only existed in oxidation process, but also involved in regulating the expression of target genes related to inflammation such as nuclear factor-k-gene binding (NF-kB) (Hoffmann & Baltimore, 2006). DMY has been demonstrated to significantly down-regulate the expression of NF-kB target genes to inhibit inflammatory effect (Hou et al., 2015; Qi et al., 2012; Tang et al., 2016). Recently, Liu, Zeng, et al. (2017) observed that DMY prevented hepatic and aortic inflammation by reducing IL-6 and TNF-α-mRNA expression in high fat diet-fed LDL receptor deficient mice. Moreover, DMY could also suppress lipopolysaccharide (LPS)-induced inflammatory cytokine production in cells. For example, DMY inhibited nitric oxide secretion, inducible nitric oxide synthase (iNOS) production and p65 phosphorylation in LPS-stimulated macrophages and reduced carrageenan-induced acute inflammation *in vivo* (Wang, Pi, et al., 2016).

3.4. Anti-cancer activity

Flavonoids from natural plants demonstrate great promise as anti-cancer agents (Ren, Qiao, Wang, Zhu, & Zhang, 2010; Cragg, Kingston, & Newman, 2011). Extensive researches have been conducted to the anti-cancer activities of DMY both *in vitro* and *in vivo*. These studies indicated that DMY was cytotoxic towards a number of human cancer cell lines, including carcinoma, breast, pancreatic and lung cancer cells. DMY exerted its anti-cancer effects by modulating multiple signaling pathways, including inhibition of apoptosis, anti-proliferation and metastatic inhibition, etc (Fig. 2).

3.4.1. Inhibition of apoptosis

Apoptosis is a vital process of programmed cell death involved in cell turnover. Any imbalance in this process leads to abnormal pathological conditions including cancer (Elmore, 2007). During tumorigenesis, cancer cells usually evaded apoptosis. DMY has been shown to restore and promote the apoptotic mode of cell death in many types of



(a) (b)

Fig. 1. DMY-Protein Complex. a) DMY-PHD complex with labeled amino acid residues (Ding et al., 2017); b) DMY- dihydropyrimidinase complex formation by I95 (light pink), S289 (limon), and D316 (yellow) (Huang, 2015). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

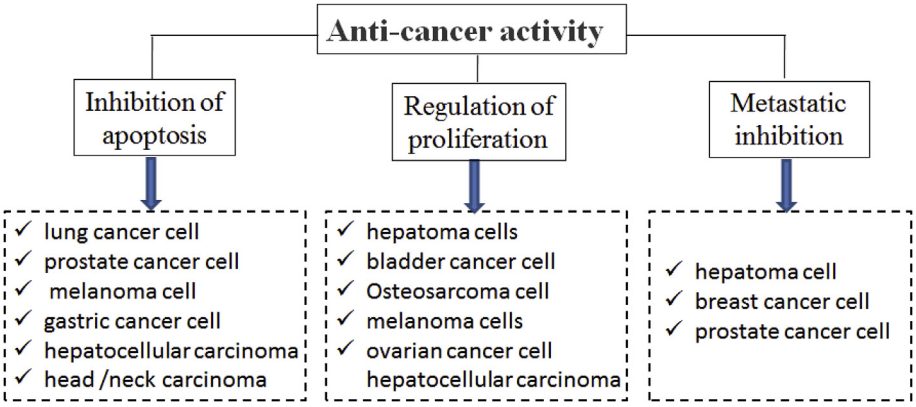


Fig. 2. Anti-cancer activity of DMY.

cancer cells. DMY was proved to promote ROS generation and activation of mitochondria-dependent apoptosis in human hepatocarcinoma HepG2 cells (Lin et al., 2014; Zhang et al., 2017) and human non-small cell lung cancer (Kao et al., 2017). Moreover, DMY induced apoptosis in human gastric cancer cells (Ji et al., 2015), mouse hepatoma Hepal-6 cell (Liu et al., 2015) and hepatocellular carcinoma hepG2 cells (Huang et al., 2017). Recently, it was found that DMY can induce not only apoptosis but also autophagy in HepG2 cell lines (Xia et al., 2014) and human melanoma cells (Zhou et al., 2017). In head and neck squamous cell carcinoma, DMY induced the upregulation of autophagic markers such as Beclin1, LC3, and p62 and promoted carcinoma cells apoptosis (Fan et al., 2016).

3.4.2. Regulation of proliferation

Control of infinite cellular proliferation and cell-cycle abnormalities also have a potential therapeutic effect in cancer treatment (Collins, Jacks, & Pavletich, 1997). Studies have confirmed that DMY played an important role in regulating cancer cell proliferation. For instance, bladder cancer was mediated by cell cycle arrest (Perez de Castro, Montoya, & Malumbres, 2008). In hepatocellular carcinoma HepG2 and Hep3B cell lines, DMY caused cell cycle arrest in G2/M phase. However, deficiency of p53 and Chk1 failed to cause DMY-induced G2/M arrest (Huang, Hu, Zhao, Li, & Li, 2013). Moreover, DMY was found to cause cell cycle arrest in G2-M phage of osteosarcoma (Zhao et al., 2014) and in G1/S phase of human melanoma SK-MEL-28 cells (Zeng et al., 2014). Recently, Xu et al. (2017) observed that DMY produced an obvious inhibition on the proliferation of A2780 and SKOV3 ovarian cancer cell lines in a dose and time-dependent manner. The data indicated that DMY was a promising cell-cycle-interfering agent in human cancer cells.

3.4.3. Metastatic inhibition

Metastasis is a life-threatening stage involving the migration of cancerous cells from their origin to other tissues (Valastyan & Weinberg, 2011). Therefore, it is important to block the migration and invasion of tumor cells in cancer treatment. Recent studies have shown that DMY could inhibit migration and invasion of a number of human cancer cell lines. DMY inhibited the expression of CXCR4, a protein associated with prostate cancer, resulting in inhibition of invasion and migration of prostate cancer cells (Ni, Gong, Li, Abdolmaleky, & Zhou, 2012). Zhou, Zhang, Zhan, and Yong (2012) observed that DMY inhibited the invasion of human breast cancer cells in a dose-dependent manner through down-regulated expression of matrix metalloproteinase (MMP-2/-9) in both the extracellular matrix and the intracellular space. Furthermore, Zhang et al. (2014) also confirmed DMY inhibition of the expression of MMP 9, which was the key factor responsible for the migration and invasion of SK-Hep-1 cells.

3.5. Other biological activities

DMY have potential for the management of metabolic diseases, such as antidiabetic and antiobesity. Shi et al. (2015) observed that DMY increased skeletal muscle insulin sensitivity, which was an important factor for management of insulin resistance for type II diabetes treatment. Recently, Liu, Wan, et al. (2017) found that DMY retarded hyperglycemia onset and ameliorated insulin resistance without weight gain in Zucker diabetic fatty rats. DMY also prevented the development of weight gain, hyperlipidemia, and atherosclerosis in ApoE knockout mouse model (Williams, Ensor, Gardner, Smith, & Lodder, 2015) and in LDL receptor knockout mice (Liu, Zeng, et al., 2017). Moreover, DMY has been demonstrated to increase irisin levels in serum, a new myokine correlated with body mass index (BMI), leading to amelioration of obesity diseases (Zhou et al., 2015). Furthermore, Zhou et al. (2017a) observed that DMY administration abrogated the adverse effects of palmitate, a major inducer of insulin resistance in obesity.

DMY may serve as a neuroprotectant in neurodegenerative conditions due to its antioxidant defense (Barnham, Masters, & Bush, 2004). DMY was found to attenuate brain aging in D-gal-induced rats (Kou et al., 2016). DMY also exhibited neuroprotective activity in Parkinson's disease (PD) (Ren, Zhao, Cao, & Zhen, 2016). Moreover, DMY demonstrated the protective effects against alcohol intoxication and alcohol tolerance (Shen et al., 2012). The molecular mechanism of anti-alcohol was possibly associated with dysfunction of GABAARs in hippocampi (Liang, Wu, et al., 2014).

DMY also demonstrated protective effect against a number of diseases and injuries such as blockage of melanogenesis in melanoma cells (Huang et al., 2016), alleviation of kidney injury (Wang, Wei, & Qiu, 2016) and liver injury (Chen et al., 2016; Xie et al., 2015) due to its antioxidant, anti-inflammatory and antiapoptotic, activities. A recent clinical study showed that DMY can improve glucose and lipid metabolism in patients with non-alcoholic fatty liver disease (Chen et al., 2015).

3.6. Toxic effects

Despite the diverse biological activities of plant flavonoids, one fundamental issue should be considered for the real application of plant flavonoids is its toxic effect. Since flavonoids are regular edible constituents of ordinary food or used in traditional medicine, examination of their toxic effects such as cytotoxicity, mutagenicity, genotoxicity and carcinogenicity have received increasing attention (Elliott Middleton, Kandaswami, & Theoharides, 2000). According to the published results, there is a controversy about the toxicity of plant flavonoids. On one hand, the use of plant flavonoids was regarded as non-toxic in some researches (Elliott Middleton et al., 2000; Middleton & Kandaswami, 1994). One the other hand, plant flavonoids was

considered to act as pro-oxidants (Sahu & Gray, 1996) and mutagenic agents in bacteria and mammalian test systems (Dzoyem, Hamamoto, Ngameni, Ngadjui, & Sekimizu, 2013; Skibola & Smith, 2000).

Toxicological effect of DMY was evaluated by several studies. Zhou, Hu, Zang, Qiu, and Liu (2001) assessed safety of *A. grossedentata* extract (Teng tea), which has high levels of DMY. Acute toxicity test, genetic toxicity tests and 90-day feeding test was conducted and found that the extract of *A. grossedentata* was toxicologically safe and the immunologic function was enhanced in mice. Furthermore, Zhong, Zhou, and Chen (2003) and Zhao et al. (2009) studied chronic toxicity of the total flavone of *A. grossedentata*. Continuous administration for a long time had no negative effect on the development and the indexes of hematology, biochemistry and pathology. They concluded that the evaluated total flavone (DMY) of *A. grossedentata* was toxicologically safe. Moreover, Xu, Yao, and Wu (2008) carried out acute toxicity test on DMY and found that the toxicity of DMY was very slight, and the greatest tolerance of oral gavage rats was 5.0 g/kg body weight.

4. Chemical stability

The phenol hydroxyl structure of DMY makes it unstable and undergoes many chemical changes such as oxidation, hydrolysis, ring fission and reduction, which resulted in metabolite formations (Xiang et al., 2017). Chemical stability of DMY is influenced by pH buffer, temperature, as well as the presence of metal ions such as Fe^{3+}, Al^{3+}, Cu^{2+}. DMY was stable in weak acid solution and was unstable under the conditions of basic solution. When pH was between 1.2 and 4.6, the solution was stable. When pH was 6.0, there was some degradation (Ruan et al., 2005). Temperature also affects the stability of DMY. However, few studies have dealt with the thermal stability of DMY due to instability and further degradations under high temperature (Chaaban et al., 2017). Only Liu, Li, et al. (2012)andLiu, Ma, et al. (2012) observed 41.47% loss of DMY in water by treatment at 60 °C for 16 days. A lot still has to be discovered about the impact of heat treatment on DMY structure and the identification of the degradation products. Such findings are essential for the biological activities of DMY.

DMY can react with metal ions to form DMY-metal complexes due to its molecular structure with upper super delocalizability, integral conjugated large π bond, strong coordinated oxygen atoms and appropriate spatial configuration (Li et al., 2007; Zhang, Brodbelt, & Wang, 2005). DMY have shown better biological activity in the presence of Zn^{2+} (Wu, Zheng, & Chen, 2011), Co^{2+} (Li, Yang, Zhai, & Chen, 2014) and Ru^{2+} (Mishra, Singh, Trigun, Singh, & Pandey, 2004). There existed three potential coordination sites in the structure of DMY which can bind with metal ions: (i) between 3-hydroxy and 4-carbonyl groups in C ring (ii) between 5-hydroxy (in A ring) and 4-carbonyl (in C ring) groups, and (iii) between 3′ and 4′-hydroxy groups in B ring (Samsonowicz & Regulska, 2017). Wu et al. (2011) reported that metal ions were more likely bound to the carbonyl oxygen and 3-OH group (in C ring) of DMY, leading to its increase of biological activity.

5. *Absorption, metabolism and elimination of DMY*

Understanding the mechanism of DMY absorption, metabolism and elimination is essential for evaluating its bioavailability efficacy and level of drug intake (Manach, Scalbert, Morand, Remesy, & Jimenez, 2004). However, little information is available on DMY's absorption profiles, distribution, metabolism, and excretion in animal models and in human body. In a recent study (Xiang et al., 2018), a human intestinal Caco-2 cell model was used to investigate the uptake and transport mechanism of DMY. The effect of time, concentration, pH, temperature and efflux transporters on its uptake and transport were systematically evaluated. The results showed that DMY was poorly absorbed by a passive diffusion mechanism. The uptake and transport of DMY were time and concentration dependent. Decreasing the pH from 8.0 to 6.0 markedly enhanced the DMY uptake, but didn't significantly affect its bidirectional transport. DMY's poor absorptions into blood and instability under the intestinal environment were also observed in a pharmacokinetic study conducted by Tong et al. (2015), which suggesting that DMY might be metabolized and eliminated in the intestinal tract. In addition, the gastrointestinal stability of DMY *in vitro* was investigated as well (Xiang et al., 2017). DMY was stable in simulated gastric fluids and buffer solutions (pH 1.2), but encountered a pseudo-first-order kinetic degradation in simulated intestinal fluids and buffer solutions (pH 6.8), which indicated gastrointestinal pH is an important factor that strongly influenced the stability, absorption and bioavailability of DMY (Abuhelwa, Foster, & Upton, 2016). The efflux transporters present in the human intestinal tract are assumed to be another factor that influenc the processes of drug absorption and distribution (Couture, Nash, & Turgeon, 2006). Multidrug resistance protein 2 (MRP2) and breast cancer resistance protein (BCRP) were proved to be involved in the uptake and transport of DMY, which hindered absorption of DMY in the intestinal tract (Xiang et al., 2018). Previous reports revealed that the transport of flavonoids such as kaempferol (Zheng et al., 2016) and baicalein (Kalapos-Kovacs et al., 2015) was also modulated by these 2 efflux transporters in Caco-2 cells.

Recently, Fan et al. (2017) estimated tissue distribution, excretion, and metabolic profile of DMY after oral administration in rats. The results showed that unconverted DMY could be distributed rapidly in various tissues especially in the gastrointestinal tract and was able to cross the blood−brain barrier. The elimination of DMY was rapid as well, which could almost be completed within 12 h. Most of eliminated DMY (unconverted) were found in feces rather than urine. DMY metabolites were detected and identified in urine and feces. This is possibility due to the different excretion pathways. Normally, metabolites of flavonoids may follow 2 pathways of excretion, i.e., via the biliary or the urinary route. Large conjugated metabolites are more likely to be eliminated in the bile, whereas small conjugates are preferentially excreted in urine (Manach et al., 2004). The relative magnitude of urinary and biliary excretion varies from one flavonoid to another in animals (Crespy et al., 2003). Besides, metabolite structures and metabolic pathways were also identified by Fan et al. (2017). A total of eight metabolites were detected and five metabolic pathways consisting of reduction, dehydroxylation, methylation, glucuronidation, and sulfation were proposed, which was agree with the previously reported results (Zhang et al., 2007).

6. Approaches to enhance solubility and bioavailability of DMY

The biological indications mentioned above clearly indicate the potential of DMY as a natural functional modulator, but the problem with DMY use lies in its poor bioavailability and low intestine permeability. DMY is sparingly soluble in water and therefore not fully absorbed from the intestine. In fact, its aqueous solubility was only 0.2 mg/mL at 25 °C (Ruan et al., 2005), which led to an absolute bioavailability of less than 10% in rats (Levet-Trafit, Gruyer, Marjanovic, & Chou, 1996; Ruan et al., 2006; Liu, Yin, Wang, & Li, 2017). A better understanding of biopharmaceutical properties of DMY would be of great help for developing strategies for bioavailability improvements. Biopharmaceutics classification system (BCS) is a useful tool for decision-making in formulation development (Amidon, Lennernas, Shah, & Crison, 1995). Based on BCS, drugs can be classified into four categories: (1) high solubility/high cell membrane permeability (class I); (2) low solubility/high cell membrane permeability (class II); and (3) high solubility/low cell membrane permeability (class III); (4) low solubility/low cell membrane permeability (class IV) (Table 2) (Kawabata, Wada, Nakatani, Yamada, & Onoue, 2011). Currently, only 4.5% of new drug candidates have both high solubility and permeability, however, an estimated 36% of marketed drugs, and nearly 90% of drugs in the developmental pipeline, are poorly water soluble (classes II and IV) (Lipp, 2013). DMY belongs to BCS class IV,

Table 2
Biopharmaceutical drug classification (Kawabata et al., 2011).

BCS class	Solubility	Permeability	% Drug on market	% Drug on pipeline
I	High	High	42	4.5
II	Low	High	30	70
III	High	Low	22	5.5
IV	Low	Low	6	20

which requires the design of different formulations (Fig. 3) to increasing its aqueous and lipid solubility and bioavailability. Each formulation strategy has its own features (Table 3) and is still at an early investigational stage.

6.1. Enhanced aqueous solubility

6.1.1. Nanoparticulate systems

The decrease in the particle size leads to an increase in the saturation solubility, an enlarged surface area and wettability and a higher dissolution velocity (Vasconcelos, Sarmento, & Costa, 2007). Solid dispersion is an effective strategy to improve the bioavailability of poorly water soluble drugs (Fig. 3a). Hydrophilic polymers polyethylene glycol (PEG) or polyvinylpyrrolidone (PVP) were used as polymeric carriers to form solid dispersions by Ruan et al. (2005) to increase DMY solubility and dissolution rate. DMY concentration in water increased as a function of PEG and PVP concentration at 25 °C and 37 °C. The improvement of the solubility might be attributed to improved wettability and dispersibility of DMY by forming intermolecular hydrogen bonding between DMY and PVP or PEG. DMY was dissolved or suspended in the carrier (PVP or PEG), resulting in a really true solution (Leuner & Dressman, 2000). Solid dispersions, although effective for improving the biopharmaceutical performance of poorly soluble compounds, are faced with the problems of physical instability, recrystallization tendency and poor scale-up (Vasconcelos et al., 2007).

Nanoencapsulation allows a sustained release of encapsulated drugs maintaining plasma concentrations at therapeutic levels during certain periods of time, and is very important to control drug release (Frank, Contri, Beck, Pohlmann, & Guterres, 2015; Mora-Huertas, Fessi, & Elaissari, 2010). Nanocapsules are polymeric nanoparticles composed of an oily core surrounded by a polymeric wall stabilized by surfactants at the particle/water interface (Fig. 3b). Dalcin et al. (2017) prepared DMY-loaded nanocapsules by insertion of DMY in polymeric nanoparticles. DMY-loaded nanocapsules not only demonstrated improved physicochemical properties (bioavailability) but also effective antimicrobial and anti-biofilm activity on urinary catheters infected by *Pseudomonas aeruginosa*. DMY-loaded nanocapsules reduced 67% of the biofilm population in urinary catheters in 96 h of treatment, while free DMY only eliminated 41%. Furthermore, a sustained release of DMY-loaded nanocapsule was observed as compared to free DMY. However, poor scalability, poor stability against aggregation and use of organic solvents are the major problems encountered during nanoencapsulation.

Microemulsion system is another popular formulation approach for solving the problems of low bioavailability. Microemulsion is a single optically isotropic and thermodynamically stable solution composed of drug, oil/lipid, surfactant, and/or co-surfactant with droplet sizes in the submicron range (Fig. 3c). (Lawrence & Rees, 2012). Solanki, Sarkar, and Dhanwani (2012) formulated a DMY microemulsion containing Capmul MCM (oil phase), Transcutol P (cosurfactant) and Cremophor EL (surfactant) in a ratio of 1:1.5:4.5, which attained maximal DMY content of 98.11%. DMY microemulsion showed higher drug release (72.34%) as compared to plain drug suspension (36.28%) and the commercially available tablet (46.91%), due to solubility-enhancing component of surfactant and cosurfactant. Despite the improvement in drug solubility and enhancement of bioavailability, micromulsions are not so effective in improving absorption because they have lower membrane permeability (Lawrence & Rees, 2012).

6.1.2. Cyclodextrin inclusion complex

Inclusion complexes prepared by kneading and co-evaporation methods exhibited higher dissolution efficiencies than their corresponding physical mixtures (Xu, 2015). Cyclodextrins as

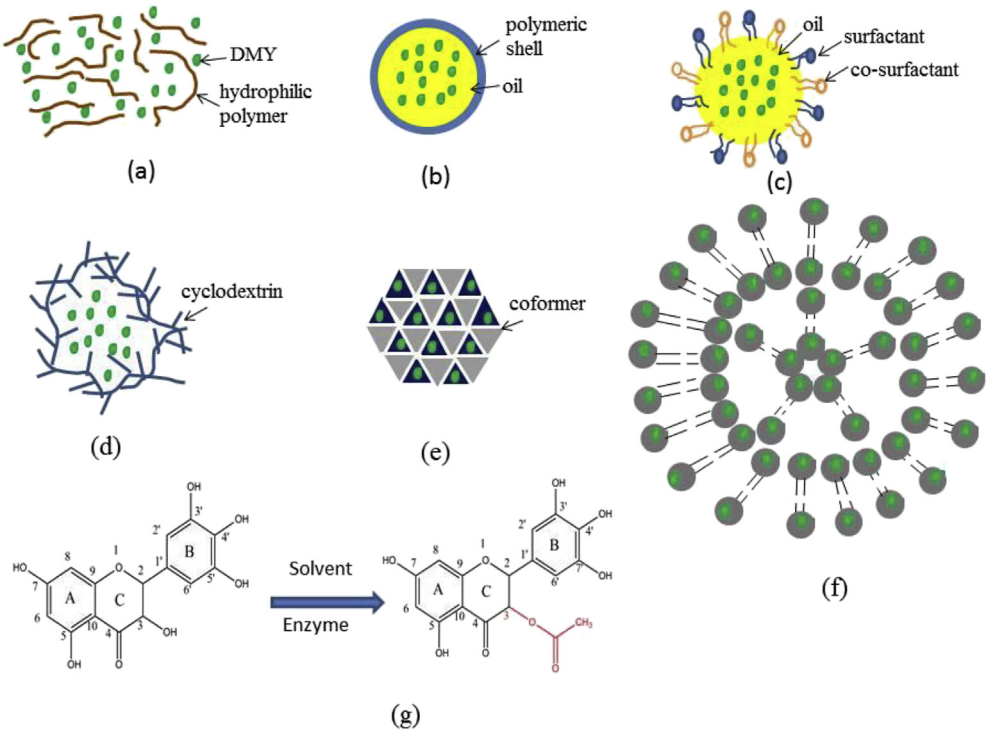


Fig. 3. Schematic representation of different formulation systems. a) solid dispersion; b) nano-encapsule; c) microemulsion; d) cyclodextrin inclusion complex; e)co-crystals; f) phospholipid complex; g) acylation.

Table 3
Formulation strategies for enhancing bioavailability of DMY.

Formulations	Main features	References
Solid dispersion	✓ reduced particle size, improved wettability and dispersibility by hydrophilic polymeric carriers faced with storage instability, recrystallization tendency and poor scale-up for manufacturing	Leuner et al., 2000; Vasconcelos et al., 2007;
Nanoencapsulation	✓ simple and inexpensive technology, Small size and higher interface area, controlled release of encapsulated material Poor scalability, poor stability against aggregation and use of organic solvents.	Frank et al., 2015; Vincekovic et al., 2017
Microemulsion	✓ Thermodynamic and colloidal stability, high encapsulation efficiency, eases of manufacturing. Rapid release, limited permeable capacity, high amount of surfactants or surface active agents	Lawrence et al., 2012; Solanki et al., 2012
Cyclodextrin inclusion complexes	✓ Controlled release, Increased solubility, dissolution rates, and improved physicochemical properties Not stable in the presence of competitive compounds and in polar solvent, potential for disruption, lower apparent permeability	Liu, Ma, et al., 2012; Suvarna et al., 2017;
Co-crystallization	✓ Improve d physical and pharmacological activities Complex preparation methods, high manufacturing costs, premature precipitation of poorly soluble drug	Wang, Tong, et al., 2016; Dalpiaz et al., 2017
Phospholipid complexes	✓ Carriers for both lipophilic and hydrophilic molecules Low stability at acidic pH, High cost of raw materials	Khan, Alexander, AjazuddinSaraf, & Saraf, 2013; Bei et al., 2014;
Chemical acylation	✓ Improved lipid solubility and radical scavenging abilities unsatisfactory yields, low regioselectivities, harsh reaction conditions, time consuming and arduous purification processes	Li et al., 2005; Guo et al. (2013)
Enzymatic acylation	✓ Improved lipid solubility, excellent regioselectivities, wide substrate specificity, green process, and mild reaction conditions Problem with stability, reusability, and catalytic performances of enzyme	Li et al., 2015; Cao et al., 2017

pharmaceutical excipients are mainly used as solubilizing and stabilizing agents for lipophilic substances in aqueous preparations (Astray, Gonzalez-Barreiro, Mejuto, Rial-Otero, & Simal-Gándara, 2009; Suvarna, Gujar, & Murahari, 2017). DMY has been encapsulated in different substituents of cyclodextrin to increase the solubility and dissolution rate (Fig. 3d). Solubility enhancement for DMY was 14.1-fold at 25 °C and 10.7-fold at 37 °C by forming inclusion complexes with β-cyclodextrin (β-CD) (Ruan et al., 2005). In another study of Liu, Li, et al. (2012) and Liu, Ma, et al. (2012), DMY was completely dispersed in the hydroxypropyl-β-cyclodextrin (HP-β-CD) matrix, the solubility of DMY in water increased from 0.74 to 53.64 mg/mL. The stability and antioxidant activity of DMY was greatly enhanced. Furthermore, Yang et al. (2011) also confirmed the solubility and stability enhancement of DMY in water by HP-β-CD; the inclusion complex not only affected the progress of the Human Hep G2 cell cycle but also induced cells to enter into apoptosis. Recently, a systemic investigation of the inclusion complexation between different HP–CDs and β-CD with DMY was conducted by Liu, Li, Nguyen, and Zhao (2012). The stability of their inclusion complexes formed with different CDs followed the rank order: HP-β-CD (MW 1540) > HP-β-CD (MW 1460) > HP-β-CD (MW 1380) > β-CD > HP-γ-CD > HP-α-CD. Steric effect and hydrophobicity of the DMY was the major cause for the stability of the formed inclusion complex. The B ring of the DMY was most likely involved in hydrogen bonding with the side groups in the cavity of the CDs, through which the inclusion complex was stabilized. Although being as a good solubilizing and stabilizing agents, cyclodextrins are prone to disruption and are coupled with lower apparent permeability.

6.1.3. Co-crystallization

Co-crystallization is a new and incipient approach to improve the solubility and permeability of a pharmaceutical product. Cocrystals are structurally homogeneous crystalline materials containing two or more components with one component as active pharmaceutical ingredient (API), the other component as coformers (Bavishi & Borkhataria, 2016) (Fig. 3e). In cocrystals, the molecular components are held together essentially via 'nonbinding' interactions (H-bonds, van der Waals forces, hydrophobic interactions, etc.) without altering the covalent bond structure of the APIs (Dalpiaz, Pavan, & Ferretti, 2017), thus retaining their pharmacological activity properties. The physical properties, such as solubility, dissolution rate, permeability, stability, and compaction behavior can be improved by co-crystallization process. Wang, Tong, et al. (2016) prepared two novel soluble cocrystals of DMY with caffeine and urea and evaluated their physicochemical properties. The maximum solution concentrations were reached within 20 min for two cocrystals (1.061 and 0.625 mg/mL for DMY-caffeine and DMY-urea, respectively). Both cocrystal powders could generate solutions of higher concentrations than DMY and maintained some degree of supersaturation for at least 3 days. The enhanced solubility of DMY in co-crystals was achieved mainly due to a decrease in lattice energy and an increase in solvent affinity. The co-crystallization process faces with the problems of complex preparation and the premature precipitation of poorly soluble drug.

6.2. Enhanced lipid solubility

6.2.1. Phospholipid complexes

The technique of complexing bioactive molecules with dietary phospholipids has been developed for improving the bioavailability of plant extracts/actives with poor absorption (Alexander, Ajazuddin, Patel, Saraf, & Saraf, 2016; Hu, Liu, Zhang, & Zeng, 2017). Phospholipids act as an amphipathic molecules showing considerable solubility in both aqueous and lipid media (Fig. 3f). Besides, phospholipids are one of the major components of the cell membrane, which facilities the permeation of the drug across the lipid-rich membrane without disturbing the cellular lipid bilayer, hence increasing its bioavailability (Khan, Alexander, Ajazuddin, Saraf, & Saraf, 2013; Zhang et al., 2016). Liu, Du, Jie, Chen, and Niu (2009) developed DMY-lecithin complex in order to improve the hydrophobicity of DMY.The solubility of DMY in n-octanol rose from 9.63 to 22.38 mg/mL. The lipophilic property of DMY was significantly improved due to DMY combination with lecithin by a non-covalent bond. Furthermore, the antioxidant performance of DMY was improved after being complexed with the lecithin. Phospholipid complex may be considered as a promising drug delivery system for improving the overall absorption and bioavailability of the plant flavonoid. However, low stability at acidic pH and high cost of raw materials limit its applications.

6.2.2. Acylation

In order to increase the solubility and stability of DMY in fatty phase, acylation of DMY (Fig. 3g) makes them more hydrophobic by fatty acid linkage, as reported in several studies (Guo et al., 2013; Li, Zheng, & Ning, 2005; Matsumoto & Tahara, 2001). Li et al. (2005) esterified DMY with lauroyl chloride, and the antioxidant activity of the obtained product (DMY-laurate) in lard oil was superior to that of DMY. Furthermore, the solubility and antioxidant activity of synthesized single-and multi-acylated DMY in peanut oil phase increased compared with DMY (Guo et al., 2013). However, chemical acylation is not regioselective and leads to an unwanted functionalization of phenolic hydroxyl groups. On the contrary, the enzymatic acylation of DMY has gained increasing attention due to its high efficiency, green process, mild reaction conditions and high regioselectivity. As for instance, enzymatic acylation of DMY was achieved by several lipases with fast reaction rate and high conversion yield (Li et al., 2015). Furthermore, Deng et al. (2016) designed a polydopamine-coated magnetic iron oxide nanoparticle (PD-MNPs) to immobilize *Aspergillus niger* lipase (ANL) to increase stability, reusability and catalytic performances of the lipase. The ANL@PD-MNPs was applied as a biocatalyst for the regioselective acylation of DMY in DMSO and gave a conversion of 79.3%, which was higher than that of previous reports (Li et al., 2015). Recently, Cao et al. (2017) used a novel deep eutectic solvent (DES) − DMSO cosolvent system as the reaction medium for enzymatic acylation of DMY catalyzed by ANL@PD-MNPs. The conversion of DMY was 91.6%. The lipid-solubility of DMY-16-acetate was 10 times higher than that of DMY. The lipase-catalyzed derivatives exhibited relatively strong radical scavenging abilities.

7. Conclusions and future trends

DMY is a bioactive flavonoid present in plant and various plant-based foods such as grapes and red bayberry. Both conventional and innovative methods including spectral and chromatographic methods have been reported for the identification and quantification of DMY in natural sources and biological samples. Numerous studies have reported diverse pharmacological activities of DMY, including antioxidant, antimicrobial anti-inflammatory, anti-cancer, antidiabetic, and neuroprotective effects, etc. This indicates that DMY has a potential to be used as a nutritional supplement or the treatment of various diseases.

The phenol hydroxyl groups of DMY make it chemically unstable. Oxidation was reported as the main cause of changes in DMY, Avoiding metal ions, high temperature and alkaline conditions during processing and storage is important to minimize the degradation of DMY. DMY also faces with the problem of poor bioavailability, which limited its pharmacological effects and clinical application. To overcome this barrier, researchers have tried to use different strategies to enhance DMY solubility and bioavailability in both aqueous or lipid phase. Formulations based on solid dispersion, nanocapsule, microemuslion, cyclodextrin inclusion complexes, co-crystallization, phospholipid complexes, and chemical or enzymatic acylation have been proposed to increase lipid solubility. Understanding the mechanism of DMY absorption, metabolism and elimination is essential for evaluating its *in vivo* bioavailability efficacy. DMY digested in laboratory animals undergoes reduction, dehydroxylation, methylation, glucuronidation, and sulfation. On the basis of the current review, the future research needs for DMY should focus on the following three aspects: (i) development of new delivery systems for DMY to obtain higher encapsulation, overall absorption and bioavailability efficiencies; (ii) pharmacokinetic studies of encapsulated DMY such as absorption, distribution, metabolism and excretion in an *in vivo* setting on higher animals and humans; (iii) transformation of these encapsulated DMY into safe products providing health benefits for the consumer.

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A list of abbreviations

A. grossedentata	Ampelopsis grossedentata
ANL	*Aspergillus niger* lipase
API	active pharmaceutical ingredient
β-CD	β-cyclodextrin
BCRP	breast cancer resistance protein
BCS	biopharmaceutics classification system
BHA	hydroxyanisole
BMI	body mass index
CAT	catalase activity
CE	capillary electrophoresis
C NMR	carbon-nuclear magnetic resonance
CXCR4	C-X-C chemokine receptor type 4
DAD	diode array detection
DES	deep eutectic solvent
DMSO	dimethyl sulfoxide
DMY	dihydromyricetin
DPPH	1,1-Diphenyl-2-picrylhydrazyl radical 2,2-Diphenyl- 1-(2,4,6-trinitrophenyl) hydrazyl
ESI-MS	electrospray ionization mass spectrometry
FT-IR	fourier transform infrared spectroscopy
GSH	glutathione
HIV	human immunodeficiency virus
H NMR	Hydrogen-nuclear magnetic resonance
H_2O_2	hydrogen peroxide
HP-β-CD	hydroxypropyl-β-cyclodextrin
HPCCC	high-performance counter current chromatography
HPLC	high performance liquid chromatography
HSCCC	high-speed counter current chromatography
HUVECs	human umbilical vein endothelial cells
iNOS	inducible nitric oxide synthase
LC	liquid chromatography
LDL	low-density lipoprotein
LPO	lipid peroxidation
LPS	lipopolysaccharide
MCM	microemulsion containing Capmul
MDA	malondialdehyde
MIC	minimum inhibitory concentration
MMP	matrix metalloproteinase
MRP2	multidrug resistance protein 2
MS	mass spectrometry
NF-kB	nuclear factor-k-gene binding
NMR	nuclear magnetic resonance
$O_2^{\bullet -}$	superoxide anion
OH•	hydroxyl radicals
P_{eff}	effective permeability
PDH	proline dehydrogenase
PD-MNPs	polydopamine-coated magnetic iron oxide nanoparticle
PEG	polymers polyethylene glycol
POV	Peroxide value
PVP	or polyvinylpyrrolidone
ROS	reactive oxygen species
S. aureus	*Staphylococcus aureus*
SOD	superoxide dismutase
T-AOC	total antioxidant capacity
TBHQ	tertiary butylhydroquinone
TC	cholesterol
TG	triglycerides

TLC	thin-layer chromatography
UHPLC	ultra-high performance liquid chromatography
UV	ultraviolet

References

Abuhelwa, A. Y., Foster, D. J., & Upton, R. N. (2016). A quantitative review and meta-models of the variability and factors affecting oral drug absorption-Part I: Gastrointestinal pH. *The AAPS Journal, 18*, 1309–1321.

Alexander, A., Ajazuddin Patel, R. J., Saraf, S., & Saraf, S. (2016). Recent expansion of pharmaceutical nanotechnologies and targeting strategies in the field of phyto-pharmaceuticals for the delivery of herbal extracts and bioactives. *Journal of Controlled Release, 241*, 110–124.

Amidon, G. L., Lennernas, H., Shah, V. P., & Crison, J. R. (1995). A theoretical basis for a biopharmaceutic drug classification: The correlation of *in vitro* drug product dissolution and *in vivo* bioavailability. *Pharmaceutical Research, 12*, 413–420.

Astray, G., Gonzalez-Barreiro, C., Mejuto, J. C., Rial-Otero, R., & Simal-Gándara, J. (2009). A review on the use of cyclodextrins in foods. *Food Hydrocolloids, 23*(7), 1631–1640.

Barnham, K. J., Masters, C. L., & Bush, A. I. (2004). Neurodegenerative diseases and oxidative stress. *Nature Reviews Drug Discovery, 3*(3), 205–214.

Bavishi, D. D., & Borkhataria, C. H. (2016). Spring and parachute: How cocrystals enhance solubility. *Progress in Crystal Growth and Characterization of Materials, 62*(3), 1–8.

Biler, M., Biedermann, D., Valentova, K., Kren, V., & Kubala, M. (2017). Quercetin and its analogues: Optical and acido-basic properties. *Physical Chemistry Chemical Physics, 19*(39), 26870–26879.

Burda, S., & Oleszek, W. (2001). Antioxidant and antiradical activity of flavonoids. *Journal of Agricultural and Food Chemistry, 49*(6), 2774–2779.

Cao, S. L., Deng, X., Xu, P., Huang, Z. X., Zhou, J., Li, X. H., et al. (2017). Highly efficient enzymatic acylation of dihydromyricetin by the immobilized lipase with deep eutectic solvents as cosolvent. *Journal of Agricultural and Food Chemistry, 65*(10), 2084–2088.

Chaaban, H., Ioannou, I., Chebil, L., Slimane, M., Gérardin, C., Paris, C., et al. (2017). Effect of heat processing on thermal stability and antioxidant activity of six flavonoids. *Journal of Food Processing and Preservation, 41*(5), e13203.

Chaturvedula, V. S. P., & Ruo, H. (2013). Isolation and NMR spectral studies of dihydromyricetin. *Journal of Pharmacognosy and Phytochemistry, 2*(4), 113–115.

Chen, Y., Lv, L., Pi, H., Qin, W., Chen, J., Guo, D., et al. (2016). Dihydromyricetin protects against liver ischemia/reperfusion induced apoptosis via activation of FOXO3a-mediated autophagy. *Oncotarget, 7*(47), 76508–76522.

Chen, S., Zhao, X., Wan, J., Ran, L., Qin, Y., Wang, X., et al. (2015). Dihydromyricetin improves glucose and lipid metabolism and exerts anti-inflammatory effects in nonalcoholic fatty liver disease: A randomized controlled trial. *Pharmacological Research, 99*, 74–81.

Collins, K., Jacks, T., & Pavletich, N. P. (1997). The cell cycle and cancer. *Proceedings of the National Academy of Sciences, 94*, 2776–2778.

Couture, L., Nash, J. A., & Turgeon, J. (2006). The ATP-binding cassette transporters and their implication in drug disposition: A special look at the heart. *Pharmacological Reviews, 58*, 244–258.

Cragg, G. M., Kingston, D. G., & Newman, D. J. (2011). *Anticancer agents from natural products.* CRC Press.

Crespy, V., Morand, C., Besson, C., Cotelle, N., Vezin, H., Demigné, C., et al. (2003). The splanchnic metabolism of flavonoids highly differed according to the nature of the compound. *AJP Gastrointestinal and Liver Physiology, 284*(6), G980–G988.

Cushnie, T. P., & Lamb, A. J. (2005). Antimicrobial activity of flavonoids. *International Journal of Antimicrobial Agents, 26*(5), 343–356.

Dalcin, A., Santos, C. G., Gündel, S. S., Roggia, I., Raffin, R. P., Ourique, A. F., et al. (2017). Anti biofilm effect of dihydromyricetin-loaded nanocapsules on urinary catheter infected by *pseudomonas aeruginosa. Colloids Surface B Biointerfaces, 156*, 282–291.

Dalpiaz, A., Pavan, B., & Ferretti, V. (2017). Can pharmaceutical co-crystals provide an opportunity to modify the biological properties of drugs? *Drug Discovery Today, 22*, 1134–1138.

Deng, X., Cao, S., Li, N., Wu, H., Smith, T. J., Zong, M., et al. (2016). A magnetic biocatalyst based on mussel-inspired polydopamine and its acylation of dihydromyricetin. *Chinese Journal of Catalysis, 37*(4), 584–595.

Ding, L., Xiao, S., Liu, D., & Pang, W. (2017). Effect of dihydromyricetin on proline metabolism of *vibrio parahaemolyticus*: Inhibitory mechanism and interaction with molecular docking simulation. *Journal of Food Biochemistry*. https://doi.org/10.1111/jfbc.12463.

DSLD (2016). Dietary supplement label database. Retrieved 25/10/2016 from https://dsld.nlm.nih.gov/dsld/ .

Du, Q. Z., Cai, W. J., & Xia, M. (2002). Purification of (+)-dihydromyricetin from leaves extract of *Ampelopsis grossedentata* using high-speed countercurrent chromatograph with scale-up triple columns. *Journal of Chromatography A, 973*(1–2), 217–220.

Dzoyem, J. P., Hamamoto, H., Ngameni, B., Ngadjui, B. T., & Sekimizu, K. (2013). Antimicrobial action mechanism of flavonoids from *dorstenia* species. *Drug Discoveries & Therapeutics, 7*(2), 66–72.

Elliott Middleton, J. R., Kandaswami, C., & Theoharides, T. C. (2000). The effects of plant flavonoids on mammalian cells: Implications for inflammation, heart disease, and cancer. *Pharmacological Reviews, 52*(4), 673.

Elmore, S. (2007). Apoptosis: A review of programmed cell death. *Toxicologic Pathology, 35*, 495–516.

Fan, L., Tong, Q., & Dong, W. W. (2017). Tissue distribution, excretion, and metabolic profile of dihydromyricetin, a flavonoid from vine tea (*Ampelopsis grossedentata*) after oral administration in rats. *Journal of Agricultural and Food Chemistry, 65*(23), 4597–4604.

Fan, T. F., Wu, T. F., Bu, L. L., Ma, S. R., Li, Y. C., Mao, L., et al. (2016). Dihydromyricetin promotes autophagy and apoptosis through ROS-STAT3 signaling in head and neck squamous cell carcinoma. *Oncotarget, 7*(37), 59691–59703.

Frank, L. A., Contri, R. V., Beck, R. C., Pohlmann, A. R., & Guterres, S. S. (2015). Improving drug biological effects by encapsulation into polymeric nanocapsules. *Wiley Interdisciplinary Reviews Nanomedicine & Nanobiotechnology, 7*(5), 623.

Gadetskaya, A. V., Tarawneh, A. H., Zhusupova, G. E., Gemejiyeva, N. G., Cantrell, C. L., Cutler, S. J., et al. (2015). Sulfated phenolic compounds from *Limonium caspium*: Isolation, structural elucidation, and biological evaluation. *Fitoterapia, 104*, 80–85.

Gao, W., Lee, S. U., Li, J., & Lee, J. W. (2016). Development of improved process with treatment of cellulase for isolation of ampelopsin from dried fruits of *ampelopsis grossedentata*. *Bioresources, 11*(1), 2712–2722.

Gao, J. H., Liu, B. G., & Ning, Z. X. (2009). Characterization and antioxidant activity of flavonoid-rich extracts from leaves of *ampelopsis grossedentata*. *Journal of Food Biochemistry, 33*(6), 808–820.

Gao, Q. P., Ma, R. Y., & Chen, L. (2017). Antioxidant profiling of vine tea (*Ampelopsis grossedentata*): Off-line coupling heart-cutting HSCCC with HPLC-DAD-QTOF-MS/MS. *Food Chemistry, 225*, 55–61.

Grande, F., Garofalo, A., & Neamati, N. (2008). Small molecules anti-HIV therapeutics targeting CXCR4. *Current Pharmaceutical Design, 14*(4), 385–404.

Guo, Q. Q., Zeng, J. H., Lu, Y., & Shu, X. G. (2013). Effects of solubility, thermal stability and antioxidant properties of acylating dihydromyricetin. *Advanced Materials Research, 791–793*, 101–105.

He, G. X., Pei, G., & Zhou, T. D. (2000). Determination of total flavonoids and dihydromyricetin in Ampelopsis grossedentala (Hand-Mazz) W. T. Wang. *Zhongguo Zhongyao Zazhi, 25*(7), 423–425.

Hoffmann, A., & Baltimore, D. (2006). Circuitry of nuclear factor kappaB signaling. *Immunological Reviews, 210*(1), 171–186.

Hou, X. L., Tong, Q., Wang, W. Q., Shi, C. Y., Xiong, W., Chen, J., et al. (2015a). Suppression of inflammatory responses by dihydromyricetin, a flavonoid from *ampelopsis grossedentata*, via inhibiting the activation of NF-kB and MAPK signaling pathways. *Journal of Natural Products, 78*(7), 1689–1696.

Hou, X., Tong, Q., Wang, W., Xiong, W., Shi, C., & Fang, J. (2015). Dihydromyricetin protects endothelial cells from hydrogen peroxide-induced oxidative stress damage by regulating mitochondrial pathways. *Life Sciences, 130*, 38–46.

Huang, C. Y. (2015). Inhibition of a putative dihydropyrimidinase from *Pseudomonas aeruginosa* PAO1 by flavonoids and substrates of cyclic amidohydrolases. *PLoS One, 10*(5), e0127634. https://doi.org/10.1371/journal.pone.0127634.

Huang, Y. H., Huang, C. C., Chen, C. C., Yang, K. J., & Huang, C. Y. (2015). Inhibition of *staphylococcus aureus*, pria helicase by flavonol kaempferol. *The Protein Journal, 34*(3), 169–172.

Huang, H., Hu, M., Zhao, R., Li, P., & Li, M. (2013). Dihydromyricetin suppresses the proliferation of hepatocellular carcinoma cells by inducing G2/M arrest through the Chk1/Chk2/Cdc25C pathway. *Oncology Reports, 30*(5), 2467–2475.

Huang, X., Lian, T., Guan, X., Liu, B., Song, Z., Zhang, J., et al. (2017). Dihydromyricetin reduces TGF-β via P53 activation-dependent mechanism in hepatocellular carcinoma hepG2 cells. *Protein and Peptide Letters, 24*(999), 419–424.

Huang, H. C., Liao, C. C., Peng, C. C., Lim, J. M., Siao, J. H., Wei, C. M., et al. (2016). Dihydromyricetin from *ampelopsis grossedentata* inhibits melanogenesis through down-regulation of MAPK, PKA and PKC signaling pathways. *Chemico-Biological Interactions, 258*, 166–174.

Hu, B., Liu, X., Zhang, C., & Zeng, X. (2017). Food macromolecule based nanodelivery systems for enhancing the bioavailability of polyphenols. *Journal of Food and Drug Analysis, 25*(1), 3–15.

Ignat, I., Volf, I., & Popa, V. I. (2011). A critical review of methods for characterization of polyphenolic compounds in fruits and vegetables. *Food Chemistry, 126*, 1821–1835.

Jeon, S. H., Chun, W., Choi, Y. J., & Kwon, Y. S. (2008). Cytotoxic constituents from the bark of *Salix hulteni*. *Archives of Pharmacal Research, 31*(8), 978–982.

Jin, M.-Y., Ding, Y., & Zhang, T. (2014). Simultaneous determination of dihydromyricetin and resveratrol in *Ampelopsis sinica (Miq.)* W.T. Wang by high-performance liquid chromatography coupled with a diode array detection method. *Journal of Chromatographic Science, 52*(4), 339–343.

Ji, F. J., Tian, X. F., Liu, X. W., Fu, L. B., Wu, Y.,Y., Fang, X. D., et al. (2015). Dihydromyricetin induces cell apoptosis via a p53-related pathway in AGS human gastric cancer cells. *Genetics & Molecular Research Gmr, 14*(4), 15564–15571.

Kalapos-Kovacs, B., Magda, B., Jani, M., Fekete, Z., Szabo, P. T., Antal, I., et al. (2015). Multiple ABC transporters efflux baicalin. *Phytotherapy Research, 29*, 1987–1990.

Kao, S. J., Lee, W. J., Chang, J. H., Chow, J. M., Chung, C. L., Hung, W. Y., et al. (2017). Suppression of reactive oxygen species-mediated ERK and JNK activation sensitizes dihydromyricetin-induced mitochondrial apoptosis in human non-small cell lung cancer. *Environmental Toxicology, 32*(4), 1426–1438.

Kawabata, Y., Wada, K., Nakatani, M., Yamada, S., & Onoue, S. (2011). Formulation design for poorly water-soluble drugs based on biopharmaceutics classification system: Basic approaches and practical applications. *International Journal of Pharmaceutics, 420*(1), 1–10.

Khan, J., Alexander, A., Ajazuddin, Saraf, S., & Saraf, S. (2013). Recent advances and future prospects of phyto-phospholipid complexation technique for improving pharmacokinetic profile of plant actives. *Journal of Controlled Release Official Journal of the Controlled Release Society, 168*(1), 50–60.

Kohyama, N., Chono, M., & Nakagawa, H. (2017). Flavonoid compounds related to seed coat color of wheat. *Bioscience Biotechnology and Biochemistry, 81*(11), 2112–2118.

Kou, X., & Chen, N. (2012). Pharmacological potential of ampelopsin in rattan tea. *Food*

Science & Human Wellness, 1(1), 14–18.
Kou, X., Liu, X., Chen, X., Jie, L., Yang, X., Fan, J., et al. (2016). Ampelopsin attenuates brain aging of D-gal-induced rats through mir-34a-mediated SIRT1/mTOR signal pathway. *Oncotarget, 7*(46), 74484–74495.
Lawrence, M. J., & Rees, G. D. (2012). Microemulsion-based media as novel drug delivery systems. *Advanced Drug Delivery Reviews, 64*(1), 175–193.
Leuner, C., & Dressman, J. (2000). Improving drug solubility for oral delivery using solid dispersions. *European Journal of Pharmaceutics and Biopharmaceutics, 50*, 47–60.
Levet-Trafit, B., Gruyer, M. S., Marjanovic, M., & Chou, R. C. (1996). Estimation of oral drug absorption in man based on intestine permeability in rats. *Life Sciences, 58*(24), 359–363.
Liang, J., Shen, Y., Shao, X. M., Scott, M. B., Ly, E., Wong, S., et al. (2014a). Dihydromyricetin prevents fetal alcohol exposure-induced behavioral and physiological deficits: The roles of GABBA receptors in adolescence. *Neurochemical Research, 39*(6), 1147–1161.
Liang, X., Wu, Y. P., Qiu, J. H., Zhong, K., & Gao, H. (2014b). A potent antibrowning agent from pine needles of *cedrus deodara*: 2R,3R-dihydromyricetin. *Journal of Food Science, 79*(9), C1643–C1648.
Li, Y. K., Fang, J. B., & Qi, X. J. (2018). Combined analysis of the fruit metabolome and transcriptome reveals candidate genes involved in flavonoid biosynthesis in *Actinidia argute*. *International Journal of Molecular Sciences, 19*(5), 147.
Li, H., Li, Q., Liu, Z., Yang, K., Chen, Z., Cheng, Q., et al. (2017). The versatile effects of dihydromyricetin in health. *Evidence-based Complementary and Alternative Medicine, 6*, 1–10.
Li, H., Li, Y. N., & Zhang, Y. K. (2008). Comparison of refluxing, ultrasonic- and Microwave-assisted extraction of dihydromyricetin from *Ampelopsis grossedentata*. *Journal of AOAC International, 91*(6), 1278–1283.
Lin, B., Tan, X., Liang, J., Wu, S., Liu, J., Zhang, Q., et al. (2014). A reduction in reactive oxygen species contributes to dihydromyricetin-induced apoptosis in human hepatocellular carcinoma cells. *Scientific Reports, 4*, 7041.
Lipp, R. (2013). The innovator pipeline: Bioavailability challenges and advanced oral drug delivery opportunities. *American Pharmaceutical Review, 16*, 10–16.
Liu, B., Du, J., Jie, Z., Chen, C., & Niu, S. (2009). Characterization and antioxidant activity of dihydromyricetin–lecithin complex. *European Food Research and Technology, 230*(2), 325–331.
Liu, D. Y., Jian-Tao, Y. E., Yang, W. H., Yan, J., Zeng, C. H., & Zeng, S. A. (2004). Ampelopsin, a small molecule inhibitor of HIV-1 infection targeting HIV entry. *Biomedical and Environmental Sciences, 17*(2), 153–164.
Liu, B., Li, W., Nguyen, T. A., & Zhao, J. (2012). Empirical, thermodynamic and quantum-chemical investigations of inclusion complexation between flavanones and (2-hydroxypropyl)-cyclodextrins. *Food Chemistry, 134*(2), 926–932.
Liu, B., Ma, Y., Yuan, C., Su, C., Hu, L., & Wang, J. (2012). Characterization, stability and antioxidant activity of the inclusion complex of dihydromyricetin with hydroxypropyl-β-cyclodextrin. *Journal of Food Biochemistry, 36*(5), 634–641.
Liu, D., Pang, W., Ding, L., & Sun, J. (2016). An insight into the inhibitory activity of dihydromyricetin against *vibrio parahaemolyticus*. *Food Control, 67*, 25–30.
Liu, L., Wan, J., Lang, H., Si, M., Zhu, J., Yong, Z., et al. (2017a). Dihydromyricetin delays the onset of hyperglycemia and ameliorates insulin resistance without excessive weight gain in Zucker diabetic fatty rats. *Molecular and Cellular Endocrinology, 439*(C), 105–115.
Liu, L., Yin, X. L., Wang, X., & Li, X. H. (2017b). Determination of dihydromyricetin in rat plasma by LC-MS/MS and its application to a pharmacokinetic study. *Pharmaceutical Biology, 55*, 657–662.
Liu, T. T., Zeng, Y., Tang, K., Chen, X., Zhang, W., & Xu, X. L. (2017c). Dihydromyricetin ameliorates atherosclerosis in LDL receptor deficient mice. *Atherosclerosis, 262*, 39.
Liu, B., Zhou, W., Chen, X., Xu, F., Chen, Y., Liu, J., et al. (2015). Dihydromyricetin induces mouse hepatoma Hepal-6 cell apoptosis via the transforming growth factor-β pathway. *Molecular Medicine Reports, 11*(3), 1609–1614.
Li, W., Wang, J., Shao, Y., Gao, Y., Ning, Z., Jiang, Y., et al. (2007). Microwave multistage countercurrent extraction of dihydromyricetin from *ampelopsis grossedentata*. *Food Technology and Biotechnology, 45*(4), 374–380.
Li, H. X., Yang, W., Zhai, R. R., Chen, L. Z., et al. (2014). Synthesis and antibacterial activity of dihydromyricetin-Co (II). *Asian Journal of Chemistry, 26*(19), 6309–6312.
Li, W., Wu, H., Liu, B., Hou, X., Wan, D., Lou, W., et al. (2015). Highly efficient and regioselective synthesis of dihydromyricetin esters by immobilized lipase. *Journal of Biotechnology, 199*, 31–37.
Li, W., Zheng, C., & Ning, Z. X. (2005). The antioxidation activity of DMYL in lard system. *Food Science, 26*(9), 73–76.
Manach, C., Scalbert, A., Morand, C., Remesy, C., & Jimenez, L. (2004). Polyphenols: Food sources and bioavailability. *American Journal of Clinical Nutrition, 79*, 727–747.
Marston, A., & Hostettmann, K. (2006). In M. M. Andersen, & K. R. Markham (Eds.). *Flavonoids Chemistry, biochemistry and applications* (pp. 1e36). Florida: Taylor & Francis Group.
Matsumoto, T., & Tahara, S. (2001). Ampelopsin, a major antifungla constitutent from *salix sachalinensis*, and its methyl ethers. *Journal of the Agricultural Chemical Society of Japan, 75*(6), 659–667.
Ma, R. Y., Zhou, R. R., & Tong, R. N. (2017). At-line hyphenation of high-speed countercurrent chromatography with Sephadex LH-20 column chromatography for bioassay-guided separation of antioxidants from vine tea (*Ampelopsis grossedentata*). *Journal of Chromatography B-Analytical Technologies in the Biomedical and Life Sciences, 1040*, 112–117.
Meng, G., Yang, S., Chen, Y., Yao, W., Zhu, H., & Zhang, W. (2015). Attenuating effects of dihydromyricetin on angiotensin II-induced rat cardiomyocyte hypertrophy related to antioxidative activity in a no-dependent manner. *Pharmaceutical Biology, 53*(6), 904–912.
Middleton, J. E., & Kandaswami, C. (1994). The impact of plant flavonoids on mammalian biology: Implications for immunity, inflammation and cancer. *The flavonoids–Advances in research since,* 1619–1652 1986.
Mishra, L., Singh, A. K., Trigun, S. K., Singh, S. K., & Pandey, S. M. (2004). Anti-hiv and cytotoxic ruthenium (II) complexes containing flavones: Biochemical evaluation in mice. *Indian Journal of Experimental Biology, 42*(7), 660–666.
Mora-Huertas, C. E., Fessi, H., & Elaissari, A. (2010). Polymer-based nanocapsules for drug delivery. *International Journal of Pharmaceutics, 385*(1), 113–142.
Naczk, M., & Shahidi, F. (2006). Phenolics in cereals, fruits and vegetables: Occurrence, extraction and analysis. *Journal of Pharmaceutical and Biomedical Analysis, 41*, 1523–1542.
NaNarayan, L. C., Rai, V. R., & Tewtrakul, S. (2013). Emerging need to use phyto-pharmaceuticals in the treatment of HIV. *Journal of Pharmacy Research, 6*(1), 218–223.
Ni, F., Gong, Y., Li, L., Abdolmaleky, H. M., & Zhou, J. R. (2012). Flavonoid ampelopsin inhibits the growth and metastasis of prostate cancer *in vitro* and in mice. *PLoS One, 7*(6), e38802.
Outtrup, H., Schaumburg, K., & Madsen, J.Ø. (1985). Isolation of dihydromyricetin and dihydroquercetin from bark of *pinus contorta*. *Carlsberg Research Communications, 50*, 369.
Owades, J. L., Rubin, G., & Brenner, M. W. (1958). Food tannins Measurement, determination of food tannins by ultraviolet spectrophotometry. *Journal of Agricultural and Food Chemistry, 6*, 44–46.
Park, K. S., Chong, Y., & Mi, K. K. (2016a). Myricetin: Biological activity related to human health. *Applied Biological Chemistry, 59*(2), 259–269.
Park, J. S., Kim, I. S., & Rehman, S. U. (2016b). HPLC determination of bioactive flavonoids in hovenia dulcis fruit extracts. *Journal of Chromatographic Science, 54*(2), 130–135.
Pérez, d. C. I., De, C. G., Montoya, G., & Malumbres, M. (2008). Emerging cancer therapeutic opportunities by inhibiting mitotic kinases. *Current Opinion in Pharmacology, 8*(4), 375–383.
Qi, S., Xin, Y., Guo, Y., Diao, Y., Kou, X., Luo, L., et al. (2012). Ampelopsin reduces endotoxic inflammation via repressing ros-mediated activation of PI3K/Akt/NF-kB signaling pathways. *International Immunopharmacology, 12*(1), 278–287.
Ren, W., Qiao, Z., Wang, H., Zhu, L., & Zhang, L. (2010). Flavonoids: Promising anticancer agents. *Medicinal Research Reviews, 23*(4), 519–534.
Ren, Q., & Song, X. (2005). *Antivirus dihydromyricetin and myricetin containing pharmaceutical composition.* CN 1605335 A.
Ren, Z. X., Zhao, Y. F., Cao, T., Zhen, X. C., et al. (2016). Dihydromyricetin protects neurons in an MPTP induced model of Parkinson's disease by suppressing glycogen synthase kinase-3 beta activity. *Acta Pharmacologica Sinica, 37*(10), 1315–1324.
Ruan, L. P., Chen, S., Yu, B. Y., Zhu, D. N., Cordell, G. A., & Qiu, S. X. (2006). Prediction of human absorption of natural compounds by the non-everted rat intestinal sac model. *European Journal of Medicinal Chemistry, 41*(5), 605–610.
Ruan, L. P., Yu, B. Y., Fu, G. M., & Zhu, D. N. (2005). Improving the solubility of ampelopsin by solid dispersions and inclusion complexes. *Journal of Pharmaceutical and Biomedical Analysis, 38*(3), 457–464.
Sahu, S. C., & Gray, G. C. (1996). Pro-oxidant activity of flavonoids: Effects on glutathione and glutathione s -transferase in isolated rat liver nuclei. *Cancer Letters, 104*(2), 193–196.
Samsonowicz, M., & Regulska, E. (2017). Spectroscopic study of molecular structure, antioxidant activity and biological effects of metal hydroxyflavonol complexes. *Spectrochimica Acta Part A Molecular & Biomolecular Spectroscopy, 173*, 757–771.
Shen, Y., Lindemeyer, A. K., Gonzalez, C., Shao, X. M., Spigelman, I., Olsen, R. W., et al. (2012). Dihydromyricetin as a novel anti-alcohol intoxication medication. *Journal of Neuroscience the Official Journal of the Society for Neuroscience, 32*(1), 390–401.
Shi, L., Zhang, T., Zhou, Y., Zeng, X., Ran, L., Zhang, Q., et al. (2015). Dihydromyricetin improves skeletal muscle insulin sensitivity by inducing autophagy via the AMPK-PGC-1α-Sirt3 signaling pathway. *Endocrine, 50*(2), 378–389.
Shoham, A., Hadziahmetovic, M., Dunaief, J. L., Mydlarski, M. B., & Schipper, H. M. (2008). Oxidative stress in diseases of the human cornea. *Free Radical Biology & Medicine, 45*(8), 1047–1055.
Skibola, C. F., & Smith, M. T. (2000). Potential health impacts of excessive flavonoid intake. *Free Radical Biology & Medicine, 29*(3), 375–383.
Solanki, S. S., Sarkar, B., & Dhanwani, R. K. (2012). Microemulsion drug delivery system: For bioavailability enhancement of ampelopsin. *Isrn Pharmaceutics, 2012*(2), 108164.
Song, Q., Liu, L., Yu, J., Zhang, J., Xu, M., Sun, L., et al. (2017). Dihydromyricetin attenuated Ang II induced cardiac fibroblasts proliferation related to inhibitory of oxidative stress. *European Journal of Pharmacology, 807*, 159–167.
Suvarna, V., Gujar, P., & Murahari, M. (2017). Complexation of phytochemicals with cyclodextrin derivatives - an insight. *Biomedicine & Pharmacotherapy, 88*, 1122–1144.
Tang, N., Ma, J., Wang, K. S., Mi, C., Lv, Y., Piao, L. X., et al. (2016). Dihydromyricetin suppresses TNF-α-induced NF-kB activation and target gene expression. *Molecular and Cellular Biochemistry, 422*(1–2), 1–10.
Tian, S., Zhang, Y., Yang, Y., Yang, W., & Gong, Y. (2002). Dihydromyricetin in *ampelopsis grossedentata* by reversed-phase high performance liquid chromatograpohy. *Journal of Hunan Agricultural University, 28*(1), 32–34.
Tong, Q., Hou, X., Fang, J., Wang, W., Wei, X., Xu, L., et al. (2015). Determination of dihydromyricetin in rat plasma by LC–MS/MS and its application to a pharmacokinetic study. *Journal of Pharmaceutical and Biomedical Analysis, 114*, 455–461.
Valastyan, S., & Weinberg, R. A. (2011). Tumor metastasis: Molecular insights and evolving paradigms. *Cell, 147*, 275–292.
Vasconcelos, T., Sarmento, B., & Costa, P. (2007). Solid dispersions as strategy to improve oral bioavailability of poor water soluble drugs. *Drug Discovery Today, 12*(23), 1068–1075.
Vieira, M. N., Winterhalter, P., & Jerz, G. (2016). Flavonoids from the flowers of *Impatiens glandulifera Royle* isolated by high performance countercurrent chromatography.

Phytochemical Analysis, 27(2), 116–125.
Wang, J. T., Jiao, P., Zhou, Y., & Liu, Q. (2016a). Protective effect of dihydromyricetinagainst lipopolysaccharide-induced acute kidney injury in a rat model. *Medical Science Monitor International Medical Journal of Experimental & Clinical Research, 22*, 454–459.
Wang, R., Pi, J., Su, X., Liu, J., Zeng, X., Wong, L., et al. (2016b). Dihydromyricetin suppresses inflammatory responses *in vitro* and *in vivo* through inhibition of IKKβ activity in macrophages. *Scanning, 33*(2), 119–124.
Wang, E. H., Qin, Z. H., Yang, L. S., Lu, F. X., & Qin, T. (2017). Antioxidant activity evaluation of dihydromyricetin from ampelopsis grossedentata on Guizhou traditional sausage. *Food Science and Technology, 42*, 128–132.
Wang, C. G., Tong, Q., Hou, X., Hu, S., Fang, J., & Sun, C. C. (2016c). Enhancing bioavailability of dihydromyricetin through inhibiting precipitation of soluble cocrystals by a crystallization inhibitor. *Crystal Growth & Design, 16*(9), 5030–5039.
Wang, Y., Wei, W., & Qiu, E. (2016d). Protection of oxidative stress induced apoptosis in osteosarcoma cells by dihydromyricetin through down-regulation of caspase activation and up-regulation of BcL-2. *Saudi Journal of Biological Sciences, 24*(4), 837.
Wang, C. G., Xiong, W., & Perumalla, S. R. (2016e). Solid-state characterization of optically pure (+)Dihydromyricetin extracted from *Ampelopsis grossedentata* leaves. *International Journal of Pharmaceutics, 511*(1), 245–252.
Williams, J., Ensor, C., Gardner, S., Smith, R., & Lodder, R. (2015). BSN723T prevents atherosclerosis and weight gain in APOE knockout mice fed a western diet. *Webmedcentral, 6*(12).
Wu, Y., Bai, J., Zhong, K., Huang, Y., & Gao, H. (2017). A dual antibacterial mechanism involved in membrane disruption and DNA binding of 2R,3R-dihydromyricetin from pine needles of *cedrus deodara* against staphylococcus aureus. *Food Chemistry, 218*, 463–470.
Wu, F., Li, Y., Song, H., Zhang, Y., Zhang, Y., Jiang, M., et al. (2016). Preventive effect of dihydromyricetin against cisplatin-induced nephrotoxicity *in vitro* and *in vivo*. *Evidence-based Complementary and Alternative Medicine, 4*, 1–9.
Wu, P., Ma, G., Li, N., Deng, Q., Yin, Y., & Huang, R. (2015). Investigation of in vitro and in vivo antioxidant activities of flavonoids rich extract from the berries of *Rhodomyrtus tomentosa* (Ait.) Hassk. *Food Chemistry, 173*, 194–202.
Wu, D.-D., Ma, Y., & Zhang, Y. (2018). Pharmacokinetic study of the major chemical constituents in Xanthoceras sorbifolia wood after oral administration of methanol extract, wood powder, and single constituents. *Journal of Liquid Chromatography & Related Technologies, 41*(3), 135–142.
Wu, C., Zheng, X. P., & Chen, L. L. (2011). Study on antioxidant activity of dihydromyricetin-zinc(II) complex. *Advanced Materials Research, 183–185*, 863–867.
Xia, J., Guo, S., Fang, T., Feng, D., Zhang, X., Zhang, Q., et al. (2014). Dihydromyricetin induces autophagy in hepG2 cells involved in inhibition of mTOR and regulating its upstream pathways. *Food and Chemical Toxicology, 66*(4), 7–13.
Xiang, D., Fan, L., & Hou, X. L. (2018). Uptake and transport mechanism of dihydromyricetin across human intestinal Caco-2 cells. *Journal of Food Science, 83*(7), 1941–1947.
Xiang, D., Wang, C., Wang, W., Shi, C., Xiong, W., Wang, M., et al. (2017). Gastrointestinal stability of dihydromyricetin, myricetin, and myricitrin: An *in vitro* investigation. *International Journal of Food Sciences & Nutrition,* 1–11.
Xie, C., Chen, Z., Zhang, C., Xu, X., Jin, J., Zhan, W., et al. (2016). Dihydromyricetin ameliorates oleic acid-induced lipid accumulation in L02 and hepG2 cells by inhibiting lipogenesis and oxidative stress. *Life Sciences, 157*, 131–139.
Xie, J., Liu, J., Chen, T. M., Lan, Q., Zhang, Q. Y., Liu, B., et al. (2015). Dihydromyricetin alleviates carbon tetrachloride-induced acute liver injury via JNK-dependent mechanism in mice. *World Journal of Gastroenterology, 21*(18), 5473–5481.
Xu, W. (2015). *Dihydromyricetin cyclodextrin inclusion compound and preparation method there of CN.* 104666293 A.
Xu, Y., Wang, S., Chan, H. F., Lu, H., Lin, Z., He, C., et al. (2017). Dihydromyricetin induces apoptosis and reverses drug resistance in ovarian cancer cells by p53-mediated downregulation of survivin. *Scientific Reports, 7*, 46060.
Xu, J. J., Yao, M. J., & Wu, M. C. (2008). Study on biological efficacy of dihydromyricetin. *Food Science, 29*(11), 622–625.
Yang, J., Liu, B., Liu, F., & Zhang, Y. (2011). Apoptosis induced by the inclusion complex of dihydromyricetin with hydroxypropyl-β-cyclodextrin in human hepG2 cells. *Journal of Medicinal Plants Research, 5*(1), 114–118.
Yang, X. Y., Yan, L. Y., & Liu, T. (2019). Simultaneous determination of bioactive flavonoids of Hoveniae Semen in rat plasma by LC-MS/MS: Application to a comparative pharmacokinetic study. *Journal of Chromatography B-Analytical Technologies in the Biomedical and Life Sciences, 1104*, 73–80.
Ye, L., Wang, H., Duncan, S. E., Eigel, W. N., & O'Keefe, S. F. (2015). Antioxidant activities of vine tea (*ampelopsis grossedentata*) extract and its major component dihydromyricetin in soybean oil and cooked ground beef. *Food Chemistry, 172*, 416–422.
Yoo, S. M., Mun, S., & Kim, J. H. (2006). Recovery and pre-purification of (+)-dihydromyricetin from *hovenia dulcis*. *Process Biochemistry, 41*(3), 567–570.
Zeng, W. C., He, Q., Sun, Q., Zhong, K., & Gao, H. (2012). Antibacterial activity of water-soluble extract from pine needles of *cedrus deodara*. *International Journal of Food Microbiology, 153*(1), 78–84.
Zeng, G., Liu, J., Chen, H., Liu, B., Zhang, Q., Li, M., et al. (2014). Dihydromyricetin induces cell cycle arrest and apoptosis in melanoma SK-MEL-28 cells. *Oncology Reports, 31*(6), 2713–2719.
Zhang, J., Brodbelt, J. S., & Wang, J. (2005). Threshold dissociation and molecular modeling of transition metal complexes of flavonoids. *Journal of the American Society for Mass Spectrometry, 16*(2), 139–151.
Zhang, Q. Y., Li, R., Zeng, G. F., Liu, B., Liu, J., Shu, Y., et al. (2014). Dihydromyricetin inhibits migration and invasion of hepatoma cells through regulation of MMP-9 expression. *World Journal of Gastroenterology, 20*(29), 10082–10093.
Zhang, Y., Ma, J. N., & Ma, C.-L. (2015). Simultaneous quantification of ten constituents of *Xanthoceras sorbifolia* Bunge using UHPLC-MS methods and evaluation of their radical scavenging, DNA scission protective, and alpha-glucosidase inhibitory activities. *Chinese Journal of Natural Medicines, 13*(11), 873–880.
Zhang, Y. S., Ning, Z. X., Yang, S. Z., & Wu, H. (2003). Antioxidation properties and mechanism of action of dihydromyricetin from *ampelopsis grossedentata*. *Acta Pharmaceutica Sinica, 38*(4), 241–244.
Zhang, Y., Que, S., Yang, X., Wang, B., Qiao, L., & Zhao, Y. (2007). Isolation and identification of metabolites from dihydromyricetin. *Magnetic Resonance in Chemistry, 45*, 909–916.
Zhang, Y., Yang, W., & Xiong, H. (2001). Basic constituent of *ampelopsis grossedentata*. *Natural Product Research and Development, 13*(5), 46–48.
Zhang, Z., Zhang, H., Chen, S., Xu, Y., Yao, A., Liao, Q., et al. (2017). Dihydromyricetin induces mitochondria-mediated apoptosis in hepG2 cells through down-regulation of the Akt/Bad pathway. *Nutrition Research, 38*, 27–33.
Zhang, K., Zhang, Y., Zhang, M., Gu, L., Liu, Z., Jia, J., et al. (2016). Effects of phospholipid complexes of total flavonoids from persimmon (*Diospyros kaki L.)* leaves on experimental atherosclerosis rats. *Journal of Ethnopharmacology, 191*, 245–253.
Zhao, L., Wang, A., Liu, B., Li, G., Zhang, Z., & Chen, S. (2009). Antioxidant and cytotoxic activity of dihydromyricetin from: *Ampelopsis grossedentata* leaves. *Agro Food Industry Hi-Tech, 20*(3), 14–17.
Zhao, Z., Yin, J. Q., Wu, M. S., Song, G., Xie, X. B., Zou, C., et al. (2014). Dihydromyricetin activates AMP-activated protein kinase and p38 (MAPK) exerting antitumor potential in osteosarcoma. *Cancer Prevention Research, 7*(9), 927–938.
Zheng, D., & Liu, G. (2006). Research and development of Chinese teng cha resources. *Agriculture Network Information, 6*, 136–142.
Zheng, L., Zhu, L., Zhao, M., Shi, J., Li, Y., Yu, J., et al. (2016). *In vivo* exposure of kaempferol is driven by phase II metabolic enzymes and efflux transporters. *The AAPS Journal, 18*, 1289–1299.
Zhong, Z., Zhou, G., & Chen, X. (2003). The rat chronic toxicity test of total flavone of *ampelopsis grossedentata* from guangxi. *Lishizhen Medicine & Materia Medical Research, 14*(4), 193–195.
Zhou, Q., Chen, K., Liu, P., Gao, Y., Zou, D., Deng, H., et al. (2015). Dihydromyricetin stimulates irisin secretion partially via the PGC -1α pathway. *Molecular and Cellular Endocrinology, 412*(C), 349–357.
Zhou, Q., Gu, Y., Lang, H., Wang, X., Chen, K., Gong, X., et al. (2017a). Dihydromyricetin prevents obesity-induced slow-twitch-fiber reduction partially via FLCN/FNIP1/AMPK pathway. *Biochimica et Biophysica Acta, 1863*(6), 1282–1291.
Zhou, Y., Hu, Y., Zang, B., Qiu, F., & Liu, X. (2001). Toxicological assessment on *ampelopsis grossedentata* and its immune regulation study. *Practical Preventive Medicine, 8*, 412–414.
Zhou, D. Z., Sun, H. Y., Yue, J. Q., Peng, Y., Chen, Y. M., & Zhong, Z. J. (2017). Dihydromyricetin induces apoptosis and cytoprotective autophagy through ROS-NF-κB signalling in human melanoma cells. *Free Radical Research, 51*(5), 517–528.
Zhou, F. Z., Zhang, X. Y., Zhan, Y. J., & Yong, G. (2012). Dihydromyricetin inhibits cell invasion and down-regulates MMP-2/-9 protein expression levels in human breast cancer cells. *Progress In Biochemistry and Biophysics, 39*(4), 352–358.
Zou, H. M., Zhou, C., & Sun, C. J. (2016). Simultaneous determination of 7 components in functional food for anti-hangover and hepatoprotection by capillary electrophoresis. *Chemical Journal of Chinese Universities-Chinese, 37*(7), 1276–1281.
Zou, H. M., Zhou, C., & Sun, C. J. (2017). Determination of 8 components in healthy food for anti-hangover and hepatoprotection by high performance liquid chromatography. *Journal of Hygiene Research, 46*(4), 633–639.