Chapter 2

Literature Review

2.1. Strategies for polyphenols extraction, isolation, and purification

2.1.2. Effect of solvents and different parameters on polyphenols extraction

The extraction of polyphenols depends upon on the sample size, chemical nature, extraction time and storage conditions as well as the presence of interfering substances. Phenolics extracts are generally a combination of different classes of phenols, which are selectively soluble in the solvents. Solvent extraction is regularly used to extract polyphenols from their plant sources due to their utility, efficacy, and broad applicability. Chemical extraction is influenced by solvent polarity, type of solvent, extraction time and temperature, as well as on the chemical composition and physical characteristics of the samples (Dai and Mumper, 2010). Still, methanol extracts have established to be superior for epicatechin, catechin, and epigallocatechin extraction (Araya Barrantes, 2012). Acetone extract with some percentage of water are good solvents for polar polyphenolic compounds but the unwanted residue is found in the extracts (Grigonis et al., 2005). The low solubility of the phenolics in 100 % organic solvents is due to strong hydrogen bonds between polyphenols and protein. Addition of water to organic solvents increases the solubility of polyphenols and it may be due to solvation with water and hydrogen bonding with water. Polyphenols extraction is effected by their variation in chemical structure and their interaction with other bioactive components. Solid-liquid extraction is generally

effected by solid-liquid ratio, particle size, solvent composition, time of extraction, temperature and pH (Mandal et al., 2007).

Researchers have analyzed the impact of different types of a solvent such as methanol, hexane, and ethyl alcohol, for extraction of polyphenols from plants and its effect on antioxidant, antidiabetic, antibacterial activity. Various solvents of differing polarities must be used to extract different phenolics compounds from plants with a high degree of accuracy (Robards, 2003). Moreover, scientists have reported that methanol is one of the potential polar solvent for extraction of phenolics antioxidant constituents (Eshwarappa et al., 2014). Previous studies also discovered that acetone and N, N dimethylformamide (DMF) are also effective at extracting antioxidants, while methanol was more efficient in at a large amount of phenolics contents from walnut fruits when compared to ethanol (Jakopič et al., 2009). It has been demonstrated that higher concentrations/amount of phenolics compared to acetone, water, and methanol was found in ethanol extracts of Ivorian plants (Altemimi et al., 2017a). The comparison of polyphenol levels of extracts obtained using different solvents demonstrated that ethanol extracts contained higher levels of polyphenols compared with extracts obtained using acetone(Sultana et al., 2009). Recent literature authenticated that multiple solvents can be employed for sequential extraction process to extract bioactive compounds from a dried powder of plants and eliminate the interference of water at the same time (Altemimi et al., 2017b). Solvents have been generally chosen on the polarity of the solute of interest for the extraction of phenolics compounds. However, methanol extracts have proven to be better for catechin, epicatechin and epigallocatechin extraction (Zuo et al., 2002). Complex glycosylation and polymerization patterns resist the development of a common protocol for all polyphenols (Cheynier, 2005). Polyphenols from are generally extracted

by polar solvent because of chemical and physical properties of solute. The sequential process can be used for extraction of the desired interest of yield to limit the amount of analogous compound. The polarity of few solvents are mentioned according to polarity (Hexane < Chloroform < Ethyl acetate < Acetone < Methanol < Water) (Markom et al., 2007).

2 .1.3. Types of polyphenols extraction

Different extraction approaches have been suggested for polyphenol extraction; these include soxhlet extraction, microwave extraction, ultrasonic extraction, heat reflux extraction, and ultrahigh pressure extraction (Aspé and Fernández, 2011). Depend upon the polarity of phytochemicals like hydrophilic polyphenols including aglycones, glycosides, and oligomers, are extracted by water, polar organic solvents such as methanol, ethanol, acetonitrile, and acetone, or their mixture of water. Ethyl acetate solvent has been utilized for partitioning of liquid extract due to the solubility of target polyphenols. Stability of polyphenols is influenced by pH. Low pH helps polyphenols to maintain protonated uncharged state, thus it can be easily extracted into organic solvents (Dai and Mumper, 2010).

2.1.4. Purification of polyphenols

Thin-layer and chromatographic methods have been explored for isolation and purification of polyphenols from plants. Thin-layer chromatography (TLC) and column chromatography are still mostly used due to their convenience, cost-cutting measure, and accessibility in various stationary phases (Ignat et al., 2011). Bioactivity-guided solvent extraction, column chromatography, and HPLC were performed for isolation and purification of biomolecules (Resch et al., 1998). Alumina, cellulose, polyamide, and

silica show the most value for purification of polyphenols. Natural products from the plant have high amounts of complex phytochemicals, which make a good separation difficult (Tsao and Deng, 2004). Therefore, multiple mobile phases by increasing polarity of solvents may be useful for highly valued separations.

2.1.5. Mass spectrometry for chemical compounds identification

Mass spectrometry deals with bombarded with either electrons or lasers on organic molecules. and thereby converted to charged ions, that are enormously energetic. A mass spectrum is a plot of the relative abundance of a fragmented ion in opposition to the ratio of mass/charge of those ions. Relative molecular mass (molecular weight) by using mass spectrometry can be decided with high accuracy and an exact molecular system determination might be feasible with knowledge of locations where the molecule has been fragmented (Adams and Sparkman, 2007). Mass spectroscopy techniques were employed to characterize the structure of the bioactive molecule. Mass spectrometry gives abundant records for the structural elucidation of the compounds when tandem mass spectrometry is explored. Thus, the grouping of HPLC and MS allows the fast and exact identification of chemical compounds in medicinal herbs, particularly when a pure standard is unavailable (Yang et al., 2009). In recent times, HR-LC/MS has been widely exploited for the analysis of phenolic compounds. Negative and positive electrospray ionization (ESI) is an ideal source due to its more ionization efficiency for polyphenols. Literature also suggested that flavanol monomers catechin/epicatechin were identified by molecular ion $[M–H]^-$ *m/z* 289 in seed coat and embryo extracts (Bastos et al., 2007).

2.1.6. NMR spectroscopy for structural identification of natural product

NMR is the ultimate tool for polyphenol identification, but since it is quite insensitive, it cannot be used for the routine structural analysis of all the phenolics one detects in plants (Robards, 2003). For this reason other phytochemical tools are typically used first and NMR is preserved for those compounds that are new to the phytochemist and need to be thus fully identified(Macías et al., 2007). It is clear that NMR is one of the only methods that can unequivocally characterize the whole polyphenol structure, including e.g. positions of various functional groups that would be difficult or even impossible to conclude from MS or MS/MS data(Sturm and Seger, 2012).

2.1.7. Separation and quantification of polyphenols

Quantification of polyphenols is commonly done by Folin-Ciocalteu and Prussian-Blue method due to their simplicity and high sensitivity(Schofield et al., 2001). Various techniques (viz. high-performance thin layer chromatography (HPTLC), liquid chromatography coupled with mass spectroscopy(LC-MS), gas chromatography, high-performance capillary electrophoresis (HPCE) and high-performance liquid chromatography. Column chromatography (CC) over Sephadex LH-20) is widely explored for quantitative, qualitative (purification and structural analysis) and analysis of polyphenols (Fecka, 2009). For example, UV/Vis absorption spectra in parallel to the retention time can, with the use of authenticated standards, contribute to the identification of polyphenols in Lamiaceae herbs (Tzima et al., 2018). The separation of phenolics has been improved with the use of reversed-phase (RP) columns (mainly RP C18); however C8 and C12 columns have also been investigated in herbal analysis (Tzima et al., 2018).Typical C18 columns in most of the reported HPLC analysis are 100–200 mm length, internal diameters of 3.9–4.6 mm, and stationary phase particle sizes equal to 3–10 μm(Unger et al., 2008). Another spectrophotometric method widely used in the

quantification of polyphenols is the UV/Vis spectrophotometry method to determine the anthocyanin content(Benvenuti et al., 2004). Also, the evolution of separation and identification techniques of polyphenols has evolved from a simple colorimetric method to the most advanced chromatography techniques (Tsao and Yang, 2003).

2.2. Polyphenols and it classification

Polyphenols are considered to be polyhydroxy phenols. Secondary metabolites like polyphenols produced generally from two main pathways shikimate and acetate/polyketide and link with glucose metabolism (Kaur, 2016). They can be categorized into three main classes:

(a) Phenolic acids

(b) Flavonoids

(c) Stilbenoids

It may be further classified into a number of subclasses, i.e., hydroxybenzoic acids, hydroxycinnamic acids, anthocyanins, proanthocyanidins, flavonols, flavones, flavanols, flavanones, isoflavones, stilbenes, and lignans (Lattanzio, 2013).

Recently, polyphenols from plants are exciting an increasing interest for their health-promoting potentials (Cartea et al., 2011; Dykes and Rooney, 2007). Polyphenols in faba beans are located in leaves, roots, and seeds e.g. (Baginsky et al., 2013). Distribution of phenolics in plants at the tissue, cellular, and subcellular levels is not always in similar. Soluble phenolics are present within the plant cell vacuoles, insoluble phenolics are found in cell walls but the outer layers of plants contain higher levels of phenolics than those located in their inner parts (Naczk and Shahidi, 2006). Recently, a scientist has reported a total of 104 phenolics compounds and present in faba beans (Abu-

Reidah et al., 2017). The main classes of characterized polyphenols of faba beans are shown in **Table 2.1**. Flavonoid compounds were found in faba beans that mainly include flavanol monomers (such as gallocatechin, epigallocatechin, and catechin), proanthocyanidins (prodelphinidins and procyanidins), flavonols (glycosylated derivates of myricetin, quercetin and kaempferol), flavanonols, isoflavones (genistein and daidzein) and flavanones (Abu-Reidah et al., 2017). Other phenolics compounds were also present in faba beans, including phenolic acids (caffeic acid, ferulic acid, p-coumaric acid, and synaptic acid) (Turco et al., 2016). Polyphenols have one or more aromatic rings with one or more hydroxyl group free or involved in another chemical function (Heim et al., 2002). They are categorized into four different groups based on the presence of a number of these phenol rings phenolic groups and structural elements (Ignat et al., 2011).

Table 2.1: Phenolic Compounds Characterized in *Vicia faba* L. Seeds Extract (Abu-Reidah et al., 2017).

Flavanol monomers	Gallocatechin, (epi)gallocatechin, (epi)gallocatechin, (epi)gallocatechin dihexoside, catechin, epicatechin
Prodelphynidns	(Epi)gallocatechin(epi)gallocatechinI,(epi)gallocatechin(epi)gallocatechinII,(epi)gallocatechin(epi)gallocatechinIV(epi)gallocatechin(epi)catechinII,(epi)gallocatechin(epi)catechinIII,(epi)gallocatechin(epi)gallocatechinV,(epi)catechin(epi)gallocatechinII
Procyanidin	(Epi)catechin(epi)catechin(procyanidinBII),(epi)catechin(epi)catechin II (procyanidin B III), (epi)catechin-(epi)catechin II
Phenolic acid	Salicylic acid *O*-glucoside, protocatechuic acid hexoside,(3'-*O*-methylfukiic acid), hydroxyeucomic acid, hydroxybenzyl-malic acid
Flavanone	Naringenin 7-glucoside (prunin), dihydrochrysin (pinocembrin)
Flavone	Isoschaftoside, apigenin 8-or-6-*C*-glucoside
Flavonol	Myricetin hexose I, quercetin hexose deoxyexose II

Stilbene	Resveratrol, resveratrol 3-*O*-ß-d-glucoside
Isoflavone	Genistein, formonetin, daidzein

2.2.1. Phenolic acids

Foods and naturals resources are the main source of phenolic acids and divided into two classes: one is derivatives of benzoic acid and other is derivatives of cinnamic acid(Liu, 2004). Hydroxybenzoic acid (Gallic acid) and hydroxycinnamic acids come under phenolic acid. The hydroxycinnamic acids are more universal than hydroxybenzoic acids and consist chiefly of *p*-coumaric, caffeic, ferulic and sinapic acids (Mattila et al., 2005).

2.2.2. Flavonoids and its characteristic features

Flavonoids involve the most considered group of polyphenols. Their basic structure consisting of two aromatic rings bound together by three carbon atoms that form an oxygenated heterocyclic. More than 4,000 varieties of flavonoids have been discovered. Flavonoids are accountable for the attractive colors of the flowers, fruits, and leaves (Ignat et al., 2011). flavonoids may be divided into six subclasses based on the variation in the structure of the heterocyclic ring. It contains flavonols, flavones, flavanones, flavanols, anthocyanins, and isoflavones. A variation in number and arrangement of the hydroxyl groups and their extent of alkylation and/or glycosylation are the result of individual differences within each group (Spencer et al., 2008). Quercetin, myricetin, catechins etc., are some most common flavonoids(Ghasemzadeh and Ghasemzadeh, 2011).

2.2.3. Stilbenes

Stilbenes consist of two phenyl moieties associated with two-carbon methylene bridge (Pandey and Rizvi, 2009). Amount of stilbenes in the food materials(human diet) are quite low. The majority of stilbenes in plants serve as antifungal phytoalexins, compounds that are synthesized only in reaction to disease or injury. Out of total stilbenes studied, most common naturally occurring polyphenol stilbene is resveratrol (3,4′,5-trihydroxystilbene) and basically found in grapes(Pandey and Rizvi, 2009).

2.2.4. Lignans

Lignans having phenolic compounds and 2,3-dibenzylbutane structure that is formed by the dimerization of two cinnamic acid residues (Yang et al., 2001). A number of lignans, such as secoisolariciresinol from plant origin are considered to be phytoestrogens (Pandey and Rizvi, 2009).

2.3. Polyphenols and its therapeutic values

2.3.1. Antioxidant potential of polyphenols

The antioxidant prevents the free radical formation in the human body. As the name suggests that it is basically chemical substances (natural or artificial) that prevent oxidation. Observations were made by researchers that establish that antioxidants, such as vitamins C, vitamin E, carotenoids, terpenoids, and polyphenols, contribute to the antioxidant power of plant foods (Prior et al., 1998). Scientists has already reported that the antioxidant capacity of extracts from Faba bean investigated by different practical approaches such as determination of the scavenging effect on 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals, evaluation of total equivalent antioxidant capacity (TEAC),ferric-reducing antioxidant power assay (FRAP) and oxygen radical absorbance capacity (ORAC) (Ou et al., 2002). Previous literature also demonstrated that the

antioxidant properties of extracts of faba beans are directly related to total phenolics and flavonoids content(Siddhuraju and Becker, 2007).

2.3.2. Polyphenols against free-radical-mediated damage to DNA

Literature suggests that DNA single-strand breaks were done by various chemical agents (Singh, 2000). It was reported that protective effect of Faba bean extracts against peroxyl radical-induced DNA strand scission was observed using peroxyl free radicals (ROOH) produced from thermal decomposition of 2,2'-azobis (2-amidinopropane hydrochloride) (AAPH) on Bluescript-SK+ plasmid DNA. A defensive effect was also observed against hydroxyl radical-induced DNA strand scission generated by UV photolysis of hydrogen peroxide (H_2O_2) (Yang and Schaich, 1996). Therefore, it is probable to suggest that faba bean polyphenols may prevent lipid peroxidation. ROO• radicals cover one of the most important factors initiating the cascade reactions of lipid peroxidation (Birben et al., 2012). It was proposed that the anti-oxidative stress of faba bean polyphenols against hydroxyl radical-induced DNA injury may be due to their capability to check the reaction of hydroxyl radical with hydrogen atoms at C3', C4' and C5' sites of the sugar moiety of DNA (Cadet et al., 1999).

2.3.3. Protective role of polyphenols and its molecular mechanism

Polyphenols have several applications such as antioxidant, antidiabetic, antihypertensive effects and other are given in **Table 2.2** and **Table 2.3**.

Table 2.2: Effect of polyphenol as protective roles and mode of action

Protective role	Molecular mechanism
Protective effect against	Antioxidant effect

oxidative stress	Scavenging properties Inhibition of peroxyl radicals–triggered damage to DNA Inhibition of the enzyme 15-lipoxygenase Inhibition of the enzyme xanthine oxidase
Antihypertensive effect	Inhibition of angiotensin-converting enzyme (ACE)
Chemopreventive effect	Inhibition of topoisomerase I Regulation of proliferation and apoptosis
Antidiabetic properties	Inhibition of the enzyme α-glucosidase Inhibition of protein glycation (triggered by glucose or methylglyoxal) Modulation of production of receptors for advanced glycation end-products in endothelial cells

Table 2.3: Selected polyphenols and their therapeutic application

Polyphenols	Therapeutic applications	References
Caffeic acid	Antitumor, antiviral, antioxidant, anti-inflammatory	(Meyer et al., 1998) (Silva et al., 2014)
Chlorogenic acid	Antioxidant,analgesic,antipyretic, chemo-preventive activity, antitumor activity	(Pandey and Rizvi, 2009),.(Meng et al., 2013)
Gallotannin	Antioxidant, analgesic, antipyretic, chemopreventive activity, antitumor activity	(Scartezzini and Speroni, 2000)
Resveratrol	Antioxidant, anti-inflammatory, antiproliferative activity	(Liu et al., 2011)
Gallic acid	Antidepressant, antiparkinson, antidiabetic, antimalarial, diuretic cardioprotective anti-viral antifungal-wound healing anthelmintic and antioxidant	(Sarkar, 2015) (Singh et al., 2017) (Huang et al., 2017)

Ellagic acid	Antioxidant, anti diabetic	(Vattem and Shetty, 2005)

2.3.4. Inhibition of α-amylase by dietary polyphenols

Dietary polyphenols associated with functional food recommend the opportunity to alter the absorption of sugars, leading to benefits for diabetics (metabolic syndrome). Enzymes α-amylase takes participate in the hydrolysis of starch breakdown and it is directly linked to one of the main sources of postprandial glucose in the blood. Pancreatic α-amylases and salivary α-amylases hydrolyze starch to produce maltose and other oligosaccharides by breaking the α-1,4 glycosidic bonds (Nyambe-Silavwe et al., 2015). Acarbose as a drug is explored in the management of type 2 diabetes and perform by inhibiting α-amylase and α-glucosidases. Edible polyphenols like molecular similar structure-activity relationship might have acarbose-like effects (Nyambe-Silavwe et al., 2015) and so could offer an appropriate approach to control type 2 diabetes. Acarbose shows many side effects including nausea, flatulence, diarrhea. Therefore, dietary polyphenols like faba bean may be a strategy for inhibiting α-amylase, an acarbose-like action but without the side effects.

2.3.5. Inhibition of α-glucosidase and lipase by polyphenols

Enzymes involved in breaking down carbohydrates and fat are α- glucosidase. and lipase By inhibiting this α-glucosidase enzyme, carbohydrates are not broken down as efficiently and glucose absorption is delayed and lipase inhibitors may affect the amount of fat absorbed. Consequently, the α-glucosidase (brush-border surface membrane of intestinal cells) is accountable for oligosaccharides into glucose, which is then transported into the blood by glucose transporter type 2 (GLUT2; SLC2A2), transporters sodium-dependent glucose transporter type 1 (SGLT1; SLC5A1) (Scheepers et al., 2004).

Literature also suggested that polyphenols inhibit the enzymes α-glucosidase and lipase which are important in the digestive tract, being responsible for carbohydrate and lipid digestion. α-glucosidase inhibitors were accountable for diminishing the starch digestion and sugar absorption, contributing to a lower postprandial glycemic response (KWON et al., 2008; Siah et al., 2012). Then, α-glucosidase has been accepted as a therapeutic agent for modulation of postprandial hyperglycemia and similarly, lipase inhibitors play an important role in decreasing the fat uptake (Moreno et al., 2003). It was hypothesized that proanthocyanidin enzyme complexes were formed during the interaction of enzymes and polyphenols (Pourcel et al., 2007). Polyphenols structure such as a ring, hydroxyl group are accountable for the interaction of enzyme-like alpha-glucosidase and lipase.

2.3.6. Inhibition of xanthine oxidase by polyphenols

Xanthine oxidase (XO) involve in purine degradation and its major role has seen as a free radicals formation during exercise (Glantzounis et al., 2005). Biochemical pathway informs that uric acid is a major product from xanthine and hypoxanthine. Molecular mechanism suggests that molecular oxygen acts like as an electron acceptor, thereby resulting in the production of superoxide radical and hydrogen peroxide (McCord, 2000). During this process, the hydroxylation reaction takes place at the molybdopterin center of xanthine oxidase and the electrons are quickly transferred to the other linearly aligned redox centers. Xanthine oxidase (XO) is also concerned about the pathogenesis of numerous diseases such as cancer, gout, and diabetes (Puddu et al., 2012). It was also discovered that polyphenols were acting like potent inhibitors of XO (Turco et al., 2016). Crucial flavonoids (quercetin and kaempferol glycosides compound) were reported as xanthine oxidase inhibitors and structure-related inhibitory activity was suggested(Tung and Chang, 2010). It has been reported that the important inhibitor of

xanthine oxidase in faba bean extracts was kaempferol 3-O-(5-O-acetyl-β-d-apiofuranosyl)-7-O-α-rhamnopyranose(Wang et al., 2015).

2.3.7. Protective role of polyphenols against diabetes mellitus in relation to phenolics content, antioxidant and oxidative stress

Diabetes mellitus is generally happened by an imbalance in radicals or oxidative stress due to hyperglycemia and hyperlipidemia (Bullon et al., 2014). It was also confirmed that, in the path of diabetes and its complications, levels of plasma antioxidants as well as α- and γ-tocopherol, α- and β-carotene, lycopene, β-cryptoxanthin, lutein, zeaxanthin, retinol and ascorbic was significantly decreased(Dietrich et al., 2002). Whole grains may be beneficial for metabolic homeostasis and the development of type 2 diabetes (Ye et al., 2012). Another work reported by a researcher showed that antioxidant properties of *Ascophyllum nodosum* were linked with the phenolic contents, while the α-glucosidase inhibitory activity was enhanced due to greater phenol content (Apostolidis and Lee, 2010). Antioxidant effect of polyphenols prevents oxidative stress and diabetes-related complications (Rahimi et al., 2005). It has been reported that antidiabetic and antilipolytic activity of lime and berry plant (e.g., α-glucosidase and lipase inhibition) were also related to flavones contents and may offer dietary and convenient alternatives to control hyperglycemia in diabetes (Zhang et al., 2015). Major polyphenolic constituents of natural fruits (fruit juice) were associated with antidiabetic activity antioxidant potential in rat and mice (Chuang and McIntosh, 2011; Zhang et al., 2015). Polyphenols from plants are called as antioxidant and it acts as blocking endotoxin-mediated kinases and transcription factors to exert its antidiabetic activity(Chuang and McIntosh, 2011). Polyphenols can prevent could prevent diabetes, anti-inflammatory

activity, improving pancreatic function by antioxidant activity and synergistic action of all the bioactive constituents.

2.3.8. Chemopreventive effect, regulation of cell proliferation and apoptosis by polyphenols

Previous studies suggested that polyphenols also modulate cell signaling and could behave as potential anticancer agents (Aggarwal and Shishodia, 2006; Bullon et al., 2014).In the past few years, polyphenol antioxidant capacity has been taken into account as one of the outstanding mechanisms of action to inhibit mutagenesis and cancer initiation, by means of their capacity to scavenge reactive oxygen species (ROS), activate antioxidant enzymes, prevent carcinogen-induced DNA adduct formation, enhance DNA repair and reduce overall oxidative DNA injury (Valko et al., 2007). Oxidative stress, peroxyl radicals and lipid peroxidation products can independently cause mutations in DNA, which are known to be crucial for the initiation of the carcinogenic process (Valko et al., 2007). The ability of Faba bean extracts to inhibit al cell growth, inhibitors of its activity are considered to be promising anticancer agents (Dai and Mumper, 2010; Jordinson et al., 1999; Siah et al., 2012) Flow cytometric analyses showed that faba bean extracts successfully induced apoptosis of HL-60 (acute promyelocytic leukemia) cells (Byczkowska et al., 2013; Siah et al., 2012) .

2.3.9. Effects of polyphenols on oxidative stress

Free radicals like reactive oxide species are the central point of normal metabolism and take part in cell signaling transduction. Increase in excess reactive oxygen species results in oxidative stress that damage to cells (DNA, protein, RNA) and also cause in a decrease of ATP level in cells, elevation of cytosolic Ca^{2+}, dysfunction of

biological function in the lipid bilayer and so on. Oxidative stress is also linked with an unbalanced production of adipokines that will further encourage the development of metabolic syndrome. Polyphenols are a powerful antioxidant and minimize the oxidative stress by scavenging ROS and generating more stable phenolic radicals (Forester and Lambert, 2011). These effects are achieved by the talent of polyphenols to transform the expression and activity of antioxidant enzymes and several signaling pathways involved in cells survival (Rahman et al., 2006). In addition to their well-known antioxidant effects, polyphenols also have insulin-potentiating, anti-inflammatory, anti-carcinogenic, anti-viral, anti-ulcer, and anti-apoptotic properties (Panickar and Anderson, 2011).

2.3.10. Effect of polyphenols on modulation of protein glycation and formation of advanced glycation end-products

Advanced glycation end-products (AGE) is accountable for the molecular mechanisms of metabolic disorder (Diabetes, cancer e.g.) (Brownlee, 2001; Goh and Cooper, 2008). It was demonstrated that AGE inhibitors would recommend a possible beneficial approach for the prevention of diabetic complications (Vlassara and Palace, 2002). The previous study authenticated that polyphenols inhibit glycation of bovine serum albumin (BSA) triggered by methylglyoxal (MGO) or glucose and formation of AGE(Yao et al., 2011). Inhibition of glycation of BSA is directly or indirectly related to with total phenols content in seed extract(Yao et al., 2011).