

Phenolic Amides (Avenanthramides) in Oats – A review

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Abstract

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Whole grain cereals such as oats are important sources of phenolic compounds. Oats contain phenolic amides, also named avenanthramides (AVAs), which have beneficial health properties because of their antioxidant, anti-inflammatory, and antiproliferative effects. The most common avenanthramides are esters of 5-hydroxyanthranilic acid with *p*-coumaric (AVA-A), ferulic (AVA-B), or caffeic (AVA-C) acids. The studies related to the stability of AVAs showed that AVA-B is sensitive to alkaline and neutral conditions and this sensitivity increases with higher temperatures. However, has been reported that AVA-A and AVA -C are more stable under the same conditions (alkaline and neutral conditions), and in addition that AVAs content of oats is increasing significantly with the germination process. AVAs help in preventing free radicals from damaging LDL cholesterol while AVAs-enriched extract of oats combined with vitamin C synergistically inhibited LDL oxidation *in vitro*. Both animal studies and human clinical trials confirmed that oats antioxidants have the potential of reducing cardiovascular risks by lowering serum cholesterol and inhibiting LDL oxidation and peroxidation. Therefore, the consumption of oats and products thereof is extremely important in reducing the risk of cardiovascular diseases.

Keywords: bioactive compounds; antioxidant; antiproliferative

Oats (*Avena sativa* L.) is a cereal consumed at lower rates than wheat and rice all over the World. However the dietary fiber content and nutritional value of oats are high. Oats contain many essential amino acids (methionine, cysteine, threonine, isoleucine, tryptophan, valine, leucine, histidine, methionine, phenylalanine, and tyrosine) necessary for human body (BIEL 2009), and high antioxidant activity components such as tocopherols, tocotrienols, and flavanoids (PETERSON 2001; LIU *et al.* 2011; KOENIG *et al.* 2014).

In recent years, there has been considerable interest in the influence of phenolic amides from plant-based foods on human health. These phenolic amides have antioxidant properties and potential therapeutic benefits including antiinflammatory, antiproliferative, and antigenotoxic effects (EÜDES *et al.* 2011). AVAs (anthranilic acid amides) are a group of naturally occurring phenolic amides in oats. Oats contain a unique group of approximately 40 different types of AVAs that consist of an anthranilic acid derivatives and hydroxycinnamic acid derivatives (COLLINS

1989; BRATT *et al.* 2003; DOKUYUCU *et al.* 2003; PETERSON & DIMBERG 2008; WISE 2011).

AVAs are low molecular weight soluble phenolic compounds. Among cereals, only oats contain AVAs (KOVÁČOVÁ & MALINOVÁ 2007; MOGLIA *et al.* 2010; SHI *et al.* 2014). These compounds are antipathogens (phytoalexins), which are produced by the plant in response to the exposure to pathogens such as fungus (COLLINS & MULLIN 1988; COLLINS 1989; OKAZAKI *et al.* 2004). The three most abundant AVAs in oats are AVA-A, AVA-B, and AVA-C, which contain hydroxyanthranilic acid and a kind of hydroxycinnamic acids like *p*-coumaric, caffeic, or ferulic acids, respectively (PETERSON *et al.* 2002; OKAZAKI *et al.* 2004; GANI *et al.* 2012).

Antioxidant properties

Oxidative stress and inflammation play a critical roles in the development of many health problems such as cancer, obesity, and cardiovascular diseases

(DE HEREDIA *et al.* 2012; SESTI *et al.* 2012; CHU *et al.* 2013). Beneficial effects on human health of the phenolic compounds obtained from herbal products have been reported in many studies. Epidemiological studies suggest that a relationship exists between whole grain consumption and chronic diseases. Phytochemicals with antioxidant properties found in cereals have been shown to protect cells against oxidative damage (OKARTER & LIU 2010; CHU *et al.* 2013).

Oat (*Avena sativa* L.) has been recognised as healthy, due to its containing antioxidants, such as vitamin E and many kinds of phenolic compounds, which have been proved to have strong antioxidant activities *in vitro* and *in vivo* (CAI *et al.* 2011). Oat phenolics may serve as potent antioxidants by scavenging reactive oxygen and nitrogen species and/or by chelating transition minerals (CHEN *et al.* 2004). AVAs are important phenolic compounds found in oats. They are constitutively expressed in the kernels, appearing in almost all milling fractions, but occur in the highest concentration in the bran and outer layers of the kernel (PETERSON *et al.* 2002; JASTREBOVA *et al.* 2006; MEYDANI 2009). Their concentrations are affected by the genotype and growing environment, in one location the combined concentration of A, B and C having exceeded 300 mg/kg (EMMONS & PETERSON 2001; PETERSON *et al.* 2002). They have been reported to contribute to the fresh taste of products made from oats (MOLTEBERG *et al.* 1996). Oat flakes (whole grain) contain more AVAs (26–27 µg/g) than oat bran (13 µg/g) (MATTILA *et al.* 2005; DYKES & RONEY 2007).

The antioxidant activity of AVAs has been found to be 10–30 times higher than those of the typical cereal components ferulic acid, gentisic acid, phydroxybenzoic acid, protocatechuic acid, syringic acid, vanillic acid, and vanillin (DIMBERG *et al.* 2002; MEYDANI 2009; VERARDO *et al.* 2011; OROZCO-MENA *et al.* 2014; YANG *et al.* 2014). The AVAs differ in the antioxidant activity, AVA-C having the highest activity, followed by AVA-B and AVA-A (PETERSON *et al.* 2002; JI *et al.* 2003). They are reported to be stopping the oxidation of lipid and linoleic acid in foods (FAGERLUND *et al.* 2009; SINGH *et al.* 2013). FAGERLUND *et al.* (2009) found that, compared with α-tocopherol, AVAs protected linoleic acid from oxidation initially to a smaller extent initially. Also, to prevent the oxidative reaction of unsaturated fatty acids, AVAs can easily be used in the food industry (ISHIHARA *et al.* 2014) due to the radical scavenging effect (SINGH *et al.* 2013). Similarly, LIU *et al.* (2011) studied the antioxidant ability of capsules contain-

ing oats AVAs on human body and found that oats extract containing AVAs possessed a high antioxidant activity on humans (LIU *et al.* 2011).

Effects of avenanthramides on health

Whole grain cereals such as oats are important sources of phenolic compounds. Phenolic compounds are of interest because of their high antioxidant capacity and potential health benefits. Especially in recent years, there has been interest in oats and oat products as bioactive high-value sources for human health in industries such as food, pharmaceutical, and cosmetic (CHU *et al.* 2013; OROZCO-MENA *et al.* 2014).

Oats and oats products are generally considered healthy and the consumption of oat bran is believed to lower LDL cholesterol (BROWN *et al.* 1999; LIU *et al.* 2004; SINGH *et al.* 2013). AVAs help in preventing free radicals from damaging LDL cholesterol (SINGH *et al.* 2013) and AVAs-enriched extract of oats combined with vitamin C synergistically inhibits LDL oxidation *in vitro* (MEYDANI 2009). Both animal studies and human clinical trials confirmed that oats antioxidants have the potential of reducing cardiovascular risks by lowering serum cholesterol, inhibiting LDL cholesterol oxidation and peroxidation (COOK & SAMMAN 1996; JI *et al.* 2003; INGLETT & CHEN 2012). Therefore, it is emphasised that the consumption of oats and oats products is extremely important to reduce the risk of cardiovascular disease (BAZZANO *et al.* 2003; CHEN *et al.* 2004, 2007; SINGH *et al.* 2013). Another study has indicated that the consumption of oats and oats bran may reduce the risk of colon cancer not only because of their high fiber contents but also due to AVAs (GUO *et al.* 2010).

In a study on laboratory animals, the supplementation of the diet of rats with AVAs-enriched extract of oats at 100 mg/kg diet has been reported to increase superoxide dismutase activity in skeletal muscle, liver, and kidneys, and to enhance glutathione peroxidase activity in heart and skeletal muscles (JI *et al.* 2003; MEYDANI 2009). LIU *et al.* (2004) indicated that oats AVAs provide another potential protective mechanism by which the consumption of oats may contribute to the reduction of the risk of atherosclerosis through inhibiting vascular smooth muscle cells proliferation. AVAs enriched oat extracts and synthetic dihydroavenanthramide-D and AVA-C methyl ester (CH₃-AVA-C) have been shown to inhibit the

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activation of the NF- κ B transcription factor, which is the master regulator of infection and inflammation (EUDES *et al.* 2014). NF- κ -B inhibitory and other functional properties of AVAs make it a candidate for supplementation in the cause of decreasing inflammation and muscle damage in post-menopausal women (KOENIG *et al.* 2014).

It is interesting that AVAs are very similar in their chemical structure to tranilast (*N*-3,4-dimethoxycinnamoylanthranilic acid), which is an antiallergic drug used to treat asthma and autoimmune diseases (AZUMA *et al.* 1976; ISHIHARA *et al.* 2014). AVAs, having a similar structure to that of tranilast, can be used in the treatment of allergic reactions (SUR *et al.* 2008). Also, it has been stated that they could contribute to lowering blood pressure by expanding the blood vessels due to the increase of the production of nitric oxide (NIE *et al.* 2006).

Chemical structures of avenanthramides

On the basis of their chemical structures, AVAs represent amides of different hydroxycinnamic acids with different anthranilic acids (Figure 1) (MATTILA *et al.* 2005; SINGH *et al.* 2013; ORTIZ-ROBLEDO *et al.* 2013). All three contain 5-hydroxyanthranilic acid while hydroxycinnamic acids involved are *p*-coumaric acid for AVA-A, ferulic acid for AVA-B, and caffeic acid for AVA-C (KOENIG *et al.* 2011; KOENIG 2012). There is a small fraction of anionic, nitrogen-containing, covalently linked hydroxycinnamic acid compounds in their structures (JI *et al.* 2003). It has been stated that they have a structure decorated with pharmaceutically antioxidant tranilast (SUR *et al.* 2008; LEE-MANION *et al.* 2009). Predominant AVAs in oats are esters of

5-hydroxyanthranilic acid with *p*-coumaric, caffeic, or ferulic acids (COLLINS & MULLIN 1988; COLLINS *et al.* 1991; SINGH *et al.* 2014).

The most commonly used solvents for the extraction of AVAs are methanol, ethanol, acetonitrile, formic acid, and their combinations. AVAs are generally determined by various chromatographic methods. For the determination of the AVAs amounts, high-performance liquid chromatography (HPLC) (BRYNGELSSON *et al.* 2002; PETERSON *et al.* 2002; CHEN *et al.* 2004; MATTILA *et al.* 2005; JASTREBOVA *et al.* 2006; PETERSON & DIMBERG 2008; SKOGLUND *et al.* 2008; WISE *et al.* 2011; ORTIZ-ROBLEDO *et al.* 2013; ISHIHARA *et al.* 2014; KOENIG *et al.* 2014), liquid chromatography-mass spectrometry (LC-MS) (OKAZAKI *et al.* 2004), liquid chromatography-mass/mass spectrometry (LC-MS/MS) (ISHIHARA *et al.* 2014), and ion-exchange chromatography (COLLINS 1989) have been used. HPLC currently represents the most popular technique for the analysis of AVAs.

Effect of germination

Generally, the germination process increases the nutritional value of cereal grains (WU 1983; TIAN *et al.* 2010). Especially in oats, germination has been reported to increase the bioavailability of the proteins due to the increase in the free amino acid content (TIAN *et al.* 2010). Several studies have shown an increase of approximately 20% in AVA content in oat grains following the germination process (BRYNGELSSON *et al.* 2002; KAUKOVIRTA-NORJA *et al.* 2004; SKOGLUND *et al.* 2008; HÜBNER & ARENDT 2013). This increase primarily occurred due to the activation of hydroxyanthranilate *N*-hydroxycinnamoyl transferase

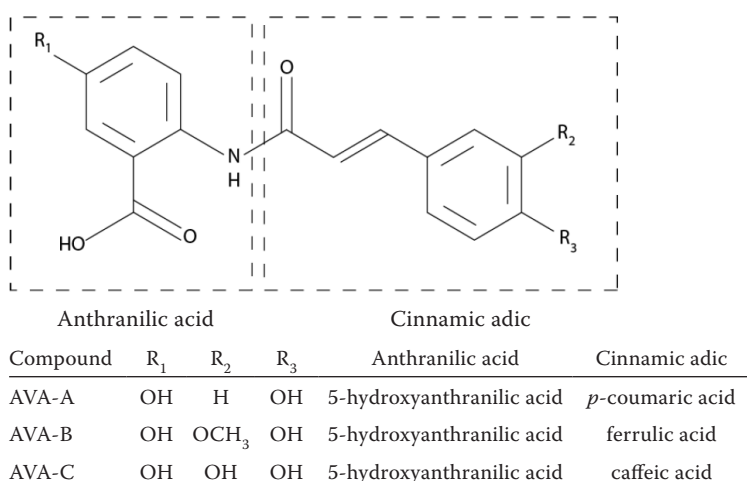


Figure 1. AVA structures (KOENIG 2012)

enzyme involved in AVA synthesis during germination. Also, SKOGLUND *et al.* (2008) have indicated that germination can be a valuable method to increase the contents of AVAs in oats. On the other hand, MATTILA *et al.* (2005) have stated that the contents of AVAs can be increased in oats products by simple processes such as steeping, germination, or malting.

Stability of avenanthramides

In general, it has been stated that phenolic compounds possessing antioxidant properties are resistant to the food processing conditions, but as the most important factors affecting their activities are reported high temperature and pH (DIMBERG *et al.* 2001). Furthermore, the enzymatic activity and temperature are reported as the cause of the phenolic compounds degradation (ROSSI *et al.* 2003; NAYAK *et al.* 2014). Studies related to the stability of AVAs have shown that AVA-B is sensitive to alkaline and neutral conditions, this sensitivity increasing with higher temperature. However, it has been reported that AV-A and AVA-C are more stable under the same conditions (alkaline and neutral conditions). Another study has emphasised that AVAs are more resistant to ultraviolet light and pH than phenolic acids (DIMBERG *et al.* 2001). Drum-drying (8 bar steam pressure) of whole meal or rolled oats decreases all tocopherols and phenolic compounds while AVAs are unaffected (BRYNGELSSON *et al.* 2002; NAYAK *et al.* 2014).

Steaming and flaking oat groats has been reported to decrease some AVAs. Whereas autoclaving oats decreased AVAs but increased the contents of tocopherols, tocotrienols, and phenolic acids (BRYNGELSSON *et al.* 2002; STEVENSON *et al.* 2008).

CONCLUSION

Avenanthramides are a group of naturally occurring phenolic amides found only in oats. The antioxidant activity of avenanthramides was 10–30 times higher than those of the typical cereal components such as phenolic acids. Both animal studies and human clinical trials confirmed that oats antioxidants have the potential of reducing cardiovascular risks by lowering serum cholesterol, inhibiting LDL cholesterol oxidation and peroxidation. AVAs, having a similar structure to tranilast, can be used in the treatment

of allergic reactions. Therefore, the consumption of oats and products thereof is extremely important for the reduction of the risk of cardiovascular disease.

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