Summary

Avenanthramides are a unique group of polyphenolics mainly found in oat with strong antioxidant and anti-inflammatory properties. However, information on the type and content, health-promoting properties and biosynthetic mechanisms of oat avenanthramides is limited. This research started with analysis of three most abundant avenanthramides, (Avn-A, B and C), in Canadian oat cultivars. Subsequently, in vitro antioxidant activities of these avenanthramides were evaluated. To investigate the cytoprotective activity of avenanthramides, human skin fibroblast cells were treated with Avn C followed by exposure to extracellular stress and its ability to reduce cellular damage was determined. Pre-treatment of cells with Avn-C reduced hydrogen peroxide-induced oxidative stress and decreased the levels of gene transcripts encoding pro-inflammatory cytokines. Furthermore, we identified and characterized three different types of genes from oat encoding 4-coumarate-CoA ligase (4CL), hydroxycinnamoly-CoA:hydroxyanthranilate N-hydroxycinnamoly transferanse (HHT) and caffeoyl-CoA O-methyltransferase (CCoAOMT)-like enzyme involved in the complete biosynthetic process of the avenanthramides. In vitro enzymatic assays using the proteins expressed in Escherichia coli as enzyme sources showed that oat 4CL could activate 4-coumaric acid, caffeic acid and ferulic acid to their CoA thioesters, respectively. Oat HHTs were only responsible for the biosynthesis of Avn-A and Avn-C using hydroxyanthranilic acid as an acyl acceptor and 4-coumaroyl-CoA and caffeoyl-CoA as an acyl donor. Avn-B was synthesized by a CCoAOMT-like enzyme through the methylation of Avn-C. These results have elucidated the molecular mechanisms for the complete biosynthesis of three major avenanthramides in oat and paved ways for genetic improvement of the nutritional trait through marker-assisting breeding in oat, and metabolic engineering of the biosynthetic pathway in heterologous systems to produce the nutritionally important compounds.