Proceedings of the LXIII SIGA Annual Congress
Napoli, Italy – 10/13 September, 2019
ISBN **978-88-904570-9-8**

Poster Communication Abstract – 6.37

METABOLIC ENGINEERING FOR THE ENRICHMENT OF DIETARY POLYPHENOLS AND THE NUTRITIONAL IMPROVEMENT IN TOMATO

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polyphenols, tomato, nutritional value, IBD, genome editing

Tomato is one of the most consumed crop worldwide, being versatile in many uses, from the daily consuming as fresh to the industrial or handmade processing in different ways. Metabolic engineering in tomato has been successfully applied for its nutritional improvement, since tomato is an excellent candidate for the genetic engineering/manipulation, both by using conventional transgnesis, or by applying the novel approach of genome editing (GE).

Following the transgenic approach, three regulatory genes (*AmDel/AmRos1/AtMYB12*) and one structural gene (*VvStSy*) have been over-expressed in a tomato line, named Bronze. As a result, Bronze tomato accumulates high levels of polyphenols, such as anthocyanins, flavonoids and stilbenoids. The daily administration of Bronze tomato-based diets in healthy and IBD (inflammatory bowel disease) chemically induced mice models has been showed promote health benefits for the gut microbiota and ameliorating the disease symptoms.

In this study, we tested Bronze and WT tomatoes as dietary supplement and carried out a metagenomic analysis on gut microbiota in Winnie mice, which is a model with spontaneously arising inflammation. Our results indicated that the Bronze diet was effective in this model and provide more information on the presence of beneficial microbial groups whose growth is promoted after polyphenol administration. These findings underline how the metabolic engineering approach in tomato is a powerful tool to understand the polyphenol metabolism and how it is possible to exploit novel functional foods in a context of nutritional prevention against inflammatory diseases.

To further explore the possibility of nutritional improvement by metabolic engineering, we also used the GE approach in a hairy root transient assay to test the possible event of HDR (homologous dependent repair)-mediated insertion of a fruit-specific promoter, upstream the *SlMYB12* gene in tomato. So far, the most used GE strategies are based on endonucleases inducing double strand breaks (DSBs), which are repaired by the NHEJ (non-homologous end joining) mechanism of repair. Although very challenging, the HDR-mediated replacement/insertion of new desired traits for the nutritional value, is a novel and alternative way to provide new insights in the knowledge of the GE application on the secondary metabolism in tomato plant.