

Euro Biotechnology 2018: Exogenous application of dsRNA for fruit developmental improvement- Cedillo Jimenez- Autonomous University of Queretaro

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Abstract

Fruit analysis provides insights about the potential for crop improvement strategies and consequently different perspectives reviews have been published. Some works have focused on the global molecular interactions as an overview of the phenomenon for fruit development and ripening. In the other hand, particular features or processes during fruit development has been reviewed including the primary and secondary metabolism, pigment biosynthesis, softening process, regulation of size and shape. On the whole, fruit development is under regulation at physiological, biochemical and molecular level and it explains the dynamics that alters the color, firmness, taste and flavor which are characteristics intimately related to fruit quality.

A few years ago, the non-coding RNAs (ncRNAs) have gained weight as development regulators in plants. Moreover, the microRNAs (miRNAs) are the most studied class of ncRNA and they are small molecules of 21-24 nucleotides that regulate gene expression at posttranscriptional level through the interaction of miRNA and its mRNA target. Furthermore, it has been shown that miRNAs have an important role during fruit development in *Solanum lycopersicum*, supporting the idea that its regulation is essential for a better understanding on plant biology and such knowledge and later technology important even for economics traits.

Owing to the rapid increase on miRNA discovery rate, databases development is a critical issue and miRBase is the largest database. However, it was out of maintenance for four years but recently it was updated and it might result in new trends for miRNA research.

The aim of this work was to update the role of miRNAs in tomato fruits regarding their development and ripening and highlight the potential MTI that some researchers have pointed in order to enhance climacteric crops. Functional reports have shown that MTI misregulation result in fruit development alteration.

Biochemical and molecular changes during tomato fruit ripening A: fresh weight, sugar content (starch and sugar accumulation, data taken from Tricon and Ge, 2009) and color, arbitrary units. B: ET biosynthesis genes ACO1,3; ACS2,4; MADSRIN, SPL-CNR. Gene expression was taken from TomExpress for Alisa Craig fruits and data was ordered preventing overlapping areas. C: Tomato fruit development and ripening representation at 17 days postanthesis (dpa), 20 dpa, mature green (MG), breaker (BR), red ripe (RR).

Tomato: the climacteric and fleshy fruit - Tomato is the model system for fleshy and climacteric fruits so sources of information as well as germplasm collection are available. Moreover, its genome has been sequenced and the fruit development has been characterized in terms of its nutritional value and molecular expression. Furthermore, researchers have developed bioinformatic tools as well as omics databases including for miRNAs analysis.

miRNAs regulate key genes for fruit development and ripening - Eventhough that ncRNAs have moved forward from "junk" to functional molecules that have role in gene silencing, chromatin structure alteration (Wang et al., 2018a) and DNA-methylation.

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They are key regulators in developmental programs in animals, and also in plants. MiRNA is the most studied class of ncRNA. MIR genes are transcribed to produce a primary transcript that folds in a stem-loop structure, which is processed and it results in a duplex molecule that contains two mature miRNA sequences: the miRNA-3p and -5p, corresponding to their arm location in the primary transcript. Any of the mature miRNA sequences might be loaded into Argonaute (AGO) protein to form the RNA-induced gene silencing complex, which regulates gene expression by near-perfect base pairing. Finally, the miRNAs expression is a complex mechanism that depends on plant internal and external cues, including phenological status and environmental factors.

Functional fruit biology of miRNAs: understanding the phenotype. The ncRNA annotation has increased dramatically, however, Hüttenhofer et al. have criticized reports for putative role based, perhaps, on speculation but not on evidence for its functional activity. In such way, here are presented the latest advances in miRNA validation and the functional analysis as strong evidence for the regulatory role during fruit development and ripening in tomato.

The miR156 family has five members and it is highly conserved in plants, however three genes were previously known as miR157. In sly-MIR156a-c it has been detected only the arm product, whereas in sly-MIR156d/e both products were found and they are expressed under biotic and abiotic stress. Silva et al. detected the expression of sly-miR156a-c in placenta and ovules in both, pre and post-anthesis tomato flowers. They found that the expression of SQUAMOSA promoters binding protein-like have negative correlation to miR156 in the same tissue. Moreover, tomato plants overexpressing miR156a-c (OE-156) resulted in the enhancement of vegetative development, delay of flowering time, and a smaller number of fruits which presented ectopic leaf-like structures.

Besides, OE-156 altered the expression of miR164:GOB, genes related to organ identity (LeT6/TKn2), as well as carotenoid biosynthesis.

It has been reported that the regulation of miR156-SBPs and SBP-LIKE (SPL) genes is important for plant development in *A. thaliana*, somatic embryogenesis *in vitro* citrus callus and its alteration might lead to morphology changes including meristem identity, cell division, organ abscission and polarity. In another study, Qin et al. performed VIGS to deliver premiR156a-c and their mature sequences (V156a). They found that fruit ripening and LeSPL-CNR decreased until 50% in the VIGS-affected zone, which is consistent with climacteric fruit behavior. Moreover, V156a accelerated fruit softening only after BR stage, suggesting regulation between fruit development but it might not be detected in the overall ripening time which is consistent to previous reports on tomato. These data suggest miR156 regulate genes related to transition phase during fruit development.

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