DISCOVERY, TARGET PREDICTION AND EXPRESSION OF MICRORNAS IN PINEAPPLE (Ananas comosus var. comosus)

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ABSTRACT

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Pineapple (Ananas comosusvar. comosus) is a non-climacteric fruit, of which the underlying mechanism of fruit ripening is still unknown. The non-ethylene induced ripening of non-climacteric fruitis very much different from climacteric ripening as revealed by the model organism of flesh-fruit i.e. tomato. Recently discovered gene regulators, i.e. miRNAs, are known to silence the translation of mRNA transcripts, leading to a silencing effect on proteins and affecting various phenotype changes in plants throughout plant development. Thus, this study aims to determine the mechanism of non-climacteric ripening through the differential expression of miRNA genes. Orthologous miRNAs were first amplified through stem-loop RT-PCR to prove for the existence of thepresence of miRNAs in pineapples. Large scale mining of miRNA from smallRNA libraries (fruit and leaf) constructed through the use of high-throughput Solexa technology was then carried out and the target mRNA transcripts were subsequently predicted. For this purpose, the miRNA registry (miRBase) and EST database (NCBI) were referred to anda bioinformatics pipeline was developed and utilized. The regulation of miRNAs in regulation of fruit ripening was postulated through correlating the target protein predicted with the pattern of expression of miRNAs during pineapple ripening obtained through stem-loop RT-qPCR. As a result, 12 orthologous miRNAs were amplified and showed the presence of miRNAs in pineapple, while finding of 153 miRNAs, 41 miRNA families, 20 miRNA*s, and 20 target transcripts obtained showed the conservation of miRNAs in pineapple. The miRNAs were then characterized. Finally, differential regulation was postulated where 7 miRNAs (miR165, miR166, miR164, miR171, miR444, miR1088, and miR396) are believed to be involved in fruit development and 4 miRNAs (miR172, miR156, miR535, miR319) in phase transition from flowering to senescence. Besides that, throughout pineapple development, 5 miRNAs (miR159, miR167, miR390, miR393 and miR394) control hormone signaling, 4 miRNAs (miR395, miR397, miR399, and miR893) control nutrient uptake and homeostasis, and 2 miRNAs (miR162 and miR168) control the overall homeostasis of miRNAs. Thus, the preliminary putative of pineapple regulated by miRNAs is postulated and revealed.

