March 31, 2021

[AgroScience Today]

Popular Article

e-ISSN: 2583-0147

Volume 2 Issue 3
Page: 0101 – 0103

Citrus Genomes: Enigma Code Breaker

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A genome is the complete set of instructions needed to build an organism and allow it to grow and develop. The genome sequence of any organism helps to understand its evolution and lays a foundation for the functional characterization means for understanding the genetic basis of differences between plants and other eukaryotes, and provides the foundation for detailed functional characterization of plant genes. The discovery of DNA as a genetic material has revolutionized the science and researchers striving to decode the genetic information. The arrival of next generation sequencing technologies has reduced the time and cost required to generate draft genomes. In citrus group, 9 species were sequenced, several assemblies were available in public domain and several were in pipeline. The analysis of genomes unraveled the origin, evolution of citrus species and identification genes involved in the characters typical to citrus like apomixis, vitamin C synthesis.

INTRODUCTION

A genome harbours the information required for the functioning of an organisms. The scanning of genome can reveal information leading to a particular phenotype. The quest for sequencing plant genomes started in the year 1996. The advanced technology with reduced cost of sequencing led to the rise in sequence data in exponential proportions. Citrus with its predominant presence in India as well as world has spurred the researchers to sequence their genomes. Citrus contains several species like sweet orange, mandarin, citron, pummelo, grapefruit, lime, lemons and several probable hybrids. It has several culinary, therapeutic, cosmetic and medicinal values. In some countries it is imbued in their culture and symbol of spiritualism. The citrus species has travelled across the world through travelers, explorers and birds. The analysis of citrus genomes may help to unravel several underlying molecular mechanisms and biochemical pathways responsible for its unique botanical features.

GENOME

A genome has several definitions, among them, one of is "a genome is an organism's complete set of genetic instructions". Each living organisms contain unique genome. In 1952, Hershey and Chase confirmed that DNA is the genetic material. In the following year *i.e.,* 1953, Watson and Crick have revealed the structure of the DNA. These discoveries have changed the landscape of the research and its direction.

SEQUENCING

In the second half of 20th century, several methods were identified for sequencing of DNA. Among them, Sanger method of sequencing has stood the test of the times and used in sequencing many prokaryotic genomes. *Haemophilus influenzae* is the first organism has its entire genome sequenced in the year 1995. Initially, due to fund crunch as well as limited expertise availability, model organisms were selected and sequenced accordingly. Later on with the advent of next generation sequencing technologies, reduced cost and increased expertise has led the sequencing of several organisms.

PLANT GENOMES

The first plant to be sequenced was *Arabidopsis thaliana*, a wild member of the mustard family with a genome size of 125 Mb in the year 2000. The genomes of plants vary greatly with respect to their chromosome number, ploidy level and size. Despite its variation, more than 230 plant genomes were sequenced. This has enormous amount of genome data and genes are identified for several characters like flowering, cell division, nodulation, etc.

CITRUS GENOMES

Citrus species are diploid with a basic chromosome number x = 9. Citrus have small genomes with a size ranging between 280-400 Mb. The International Citrus Genome Consortium (ICGC) was organized in 2003 composed of researchers from Australia, Brazil, China, France, Israel, Italy, Japan, Spain, and USA to sequence the genome of sweet orange. In their quest, initially a haploid plant derived from 'Clementine' mandarin, was sequenced to serve as the reference genome for citrus. A second genome assembly was produced from the sweet orange clone 'Ridge Pineapple'. Since then, a total of 9 species were sequenced (Table 1).

Table 1. Details of the citrus genome available in the public domain

Sl.No	Species	Cultivar	Size (Mbp)	Assemblies	Year
1	*Citrus sinensis*	Sweet orange	327.669	3	2012
2	*Citrus clementina*	Clementine	301.365	2	2013
3	*Citrus unshiu*	Satsuma mandarin	359.652	2	2017
4	*Citrus x paradisi x Citrus trifoliata*	Citrumelo	265.534	1	2017
5	*Citrus maxima*	Buntan	345.757	1	2017
6	*Citrus cavaleriei*	Ichang papeda	357.621	1	2018
7	*Citrus medica*	Buddha's hand	406.058	1	2018
8	*Citrus reticulata*	Mandarin orange	344.273	1	2018
9	*Citrus hindsii*	Hong Kong kumquat	373.170	1	2019

DECODING CITRUS GENOMES

Citrus species contains several crops like sweet orange, mandarin, pummelo, citron, grapefruit, lime,

Figure 1. True citrus species

lemon and several probable hybrids. It was hypothesized all these are the derivatives of three major/true species *i.e., Citrus medica* L. (citron), *Citrus reticulata* Blanco (mandarin), *Citrus maxima* (Burm.) Merrill (pummelo) (Figure 1). Through genome sequence data analysis, their status as the major/true species has been confirmed. Citrus possesses several peculiar characters like, apomixis/ polyembryony. The genome sequencing of 130 citrus accessions resulted in fine mapping of a locus responsible for polyembryony. It was also speculated that sweet orange is the cross between pummelo and mandarin. The genome sequence revealed it was indeed a cross but it is (pummelo x mandarin) x mandarin. It contains 25% of pummelo genome and 75% of mandarin genome. Citrus species were constant companion for the sea travelers to avoid scurvy result of vitamin C deficiency. Sequencing has helped to identify the regulatory enzymes involved in the vitamin C production/synthesis. It was also found, the mandarin which are now cultivated are entirely different from the wild mandarins. The taxonomy of citrus is chaotic one because of its interspecific and intergeneric hybridization events. The genome data analysis has solved some the evolutionary aspects of citrus. There was also suggestion that kumquat (*Fortunella hindsii)* should be considered as a species of citrus group. Identification of SSR and SNP sets will be crucial in the studies of gene and allelic content, comparisons of gene content and genome structure. This genome data is valuable for understanding evolutionary genomics and facilitates the discovery of genes involved in citrus fruit biology and nutritionally important traits.

CONCLUSION

Citrus is the third major crop in India. The production and consumption of citrus fruits like mandarin, sweet orange and lime has increased over the years in India. In improving citrus, breeders are facing several impediments like juvenility, heterozygosity, apomixis. The availability of genomes provides various means and tools to expedite the citrus improvement. These discoveries from citrus genomes could have far reaching implications not only on the citrus improvement but in the hindsight positive impact on the health and economy of any society or country.