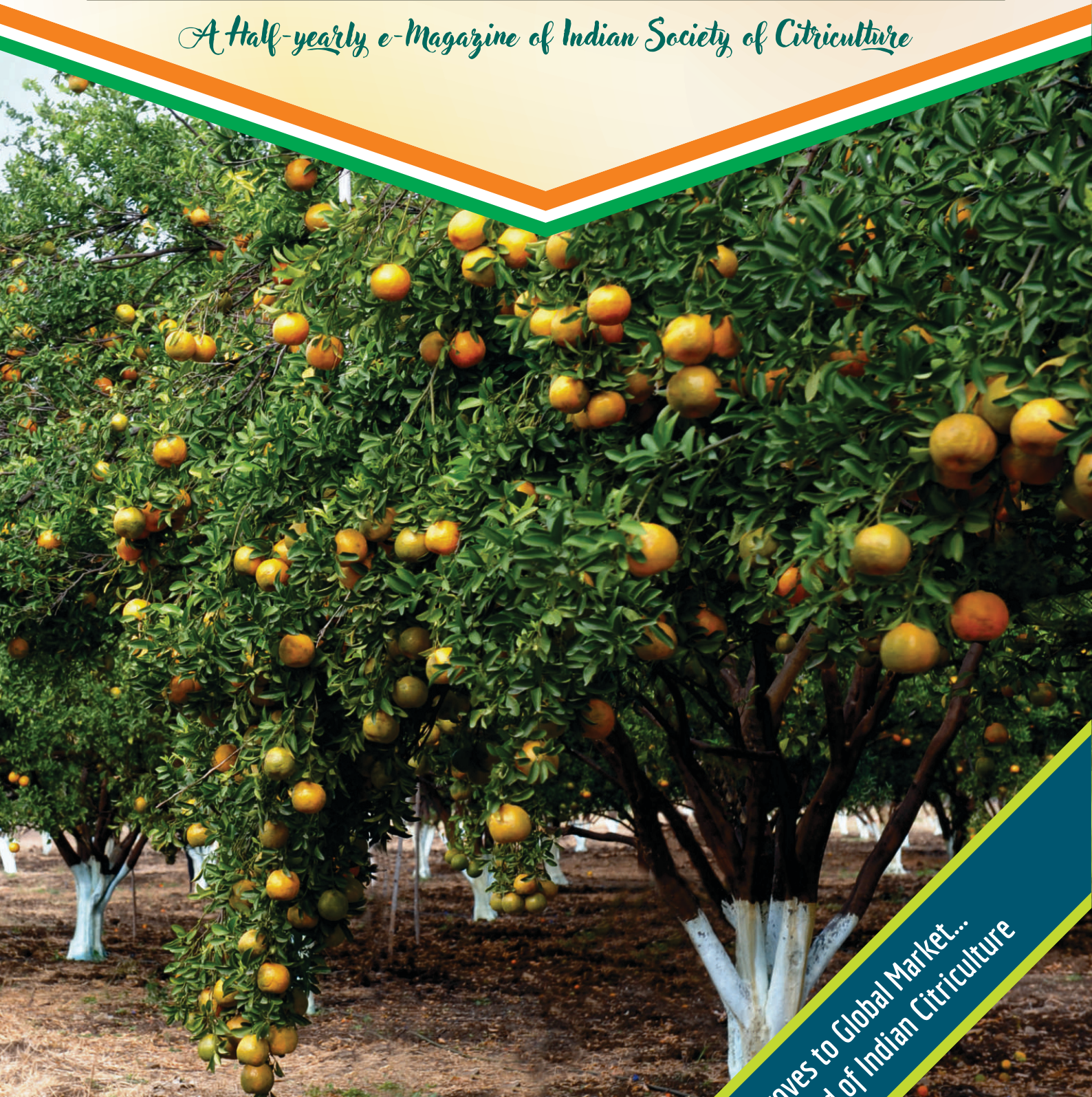


Vol. 1, Issue 1 (2023)

Indian Citriculture

A Half-yearly e-Magazine of Indian Society of Citriculture



Indian Society of Citriculture

Hqs: ICAR-Central Citrus Research Institute
Amravati Road, Nagpur, Maharashtra - 440033

*From Local Groves to Global Market...
Explore the World of Indian Citriculture*



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Asian Citrus Congress - 2023

Advancing Citriculture for Agro-economic Prosperity

28-30 October, 2023
Nagpur, Maharashtra, India

In Association with

Organized by



Indian Society of Citriculture (ISC)

Headquarters: ICAR-CCRI, Nagpur, Maharashtra, India



ICAR - Central Citrus Research Institute
Nagpur, Maharashtra, India



**Asia-Pacific Association of
Agricultural Research Institutions**
Bangkok, Thailand



**Korean Society for Citrus and
Subtropical Climate Fruits**
Jeju City, South Korea

Invitation

Dear friends,

On behalf of the Indian Society of Citriculture, we are delighted to extend our invitation to the scientists, technical, extension personnel, policy makers and students from public and private research institutions as well as other stakeholders from the production, processing and commercial sector of citrus industry to attend the Asian Citrus Congress-2023 (ACC-2023) that will be held in Nagpur, India from 28-30 October, 2023, under the theme "*Advancing Citriculture for Agro-economic Prosperity*". Indian Society of Citriculture (ISC) is organizing the congress, in association with ICAR-Central Citrus Research Institute (ICAR-CCRI), India; Asia-Pacific Association of Agricultural Research Institutions (APAARI), Bangkok, Thailand and Korean Society for Citrus and Subtropical Climate Fruits (KSCSCF), Jeju City, South Korea. The citrus sector in Asian countries will be represented at the Congress by all relevant stakeholders, and the event will feature a variety of technical sessions comprising of plenary lecture, lead/invited talks, contributory oral presentations and poster session by the experts from various countries. There will also be brainstorming session, panel discussion, business meet and field trip among other events.

The ACC-2023, a unique event dedicated to advancing the citrus industry in Asia, will bring together experts from across the globe to share their knowledge, experiences and innovations. This unique event aims to establish new linkages, collaborations and networking among the delegates to transform the citrus industry in Asia.

On behalf of the organizing committee, I appeal to you to be a part of this mega event by participating in ACC-2023. We are excited to have you join us for this congress and look forward to work together to make the citrus sector in Asia vibrant and brighter in the years to come.

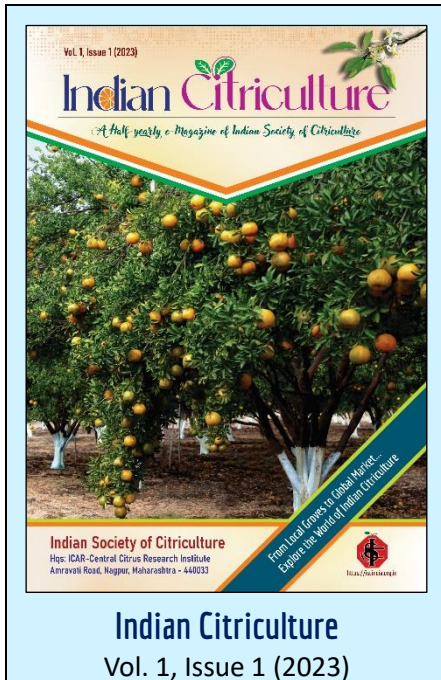
Eagerly waiting to meet you in Nagpur, India.

(Dilip Ghosh)

Convenor, Asian Citrus Congress - 2023
President, Indian Society of Citriculture
Director, ICAR- Central Citrus Research Institute



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Indian Citriculture
Vol. 1, Issue 1 (2023)

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Dr HIMANSHU PATHAK

SECRETARY (DARE) & DIRECTOR GENERAL (ICAR)



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Message

It brings me great excitement to learn that the Indian Society of Citriculture is bringing out an e-magazine titled "Indian Citriculture." The citrus industry has long been a vital contributor to the agricultural sector in India, providing livelihoods for millions of farmers and workers across the country. Over the years, the industry has grown and evolved, adapting to changing market conditions and technological advances to remain competitive in the global market.

I wish that "Indian Citriculture" e-Magazine will be at the forefront of chronicling this transformation, providing in-depth coverage of the latest trends, technologies, and best practices in the industry. With each issue, the magazine should provide valuable insights into the challenges and opportunities facing citrus growers and processors, as well as the scientists and researchers who are working to develop new varieties, improve crop yields, manage pests and diseases and develop innovative food products. I am confident that "Indian Citriculture" e-Magazine will emerge as an indispensable resource for anyone interested in the Indian citrus industry.

I would like to commend the Indian Society of Citriculture, editors, and contributors for their dedication and commitment to excellence in bringing out "Indian Citriculture" e-Magazine.

I look forward to reading future issues with great interest.

(Himanshu Pathak)

Secretary (DARE) and
Director General (ICAR)

डॉ. तिलक राज शर्मा
उप महानिदेशक (बागवानी विज्ञान)

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Message

I am happy that the inaugural issue of "Indian Citriculture", an electronic magazine put out by the Indian Society of Citriculture, is being brought out by the Indian Society of Citriculture. The citrus industry has long been a vital contributor to the horticultural sector in India, providing livelihoods for millions of farmers and workers across the country. Over the years, the industry has grown and evolved. As India continues to develop and modernize its agricultural sector, the citrus industry will undoubtedly play an essential role in driving growth and prosperity.

"Indian Citriculture" e-Magazine is a right step providing the latest information and insights to help stakeholders navigate this rapidly changing landscape. The e-magazine aims to provide comprehensive coverage of the citrus industry in India through articles and reports on the latest developments in citrus cultivation, pest and disease management, processing, marketing, export, extension activities and important events. It also includes success stories of progressive citrus growers and citripreneurs. The content of the magazine is tailored to meet the needs of growers, researchers, and other stakeholders in the industry, making it a valuable resource for anyone interested in citriculture.

I wish that soon this e-Magazine will be a must-read publication for anyone interested in the Indian citrus industry. With its in-depth coverage, expert analysis, and engaging content, it is sure to inform, inspire, and educate readers.

I would like to congratulate the entire team of Indian Society of Citriculture, particularly those engaged in the publication process for this knowledge endeavour.

(T. R. Sharma)

Deputy Director General
(Horticultural Science), ICAR



From the Desk of President, Indian Society of Citriculture

Our country is home to a wide variety of citrus fruits, including mandarins, sweet orange, lemons, limes, grapefruits, pomelo and a plethora of underutilized citrus species which are grown across the length and breadth of the country. Citrus fruits are not only a rich source of nutrition, making them an essential part of Indian cuisine. Citriculture has been an integral part of Indian agriculture for centuries. India is the world's third-largest producer of citrus fruits after China and Brazil, with an estimated production of 14.31 million tons in 2021. Citrus sector also plays a significant role in the Indian economy, providing employment opportunities to millions of people and contributing to the country's GDP. One of the major drivers of the citrus sector's growth is the increasing demand for citrus fruits in both domestic and international markets. Citrus fruits are rich in vitamins, minerals and various bioactives, making them a popular choice for health-conscious consumers. The growing popularity of citrus-based beverages and food products has also contributed to the rise in demand for citrus fruits. In addition to the demand for citrus fruits, the sector has also benefited from technological advancements in farming practices and post-harvest management. These technologies have enabled farmers to increase yields and improve the quality of their produce, resulting in higher profits.

The citrus sector in India has immense potential to contribute significantly to the country's economy by 2047, which marks the 100th anniversary of India's independence. With the right policies and initiatives, the citrus sector has the potential to create substantial employment opportunities, generate substantial revenue, and improve the lives of millions of people across the country, particularly in rural areas. The sector can also create indirect income and employment opportunities, such as transportation, packaging, and marketing. The Government of India is already undertaking a proactive approach for promoting and supporting the horticulture sector including citriculture. This includes investments in research and development to improve improved varieties and farming practices, infrastructure development to facilitate transportation and storage, and policy initiatives.

Given this background, Indian Society of Citriculture is bringing out the e-Magazine "Indian Citriculture" to showcase the diverse aspects of citriculture with specific focus on India, including the latest trends, best practices, and emerging technologies.

The magazine features articles and news from leading experts in the field, providing insights into the challenges faced by the Indian citrus industry and the innovative solutions being implemented to address them. It aims to serve as a common platform for all stakeholders involved in citrus research, outreach, production, processing, marketing and allied activities.

For researchers, it will provide a platform to share their findings and discuss the latest trends and developments in citrus research. This can help to facilitate collaboration between researchers and identify areas for further research. Similarly, for producers, the e-magazine can provide information on best practices, new technologies, and emerging markets. This can help to improve productivity, reduce costs, and increase profitability. The latest processing technologies and techniques to be published in the e-magazine will benefit the processors to improve the quality of processed citrus products and increase their storability. Even for marketers, the publication is expected to provide insights into consumer trends, preferences, and behaviors. This can help to develop effective marketing strategies that meet the needs of consumers and increase demand for citrus products. Furthermore, this e-magazine can serve as a platform for policymakers to share their vision and initiatives for the citrus industry. This can help to promote a conducive policy environment that supports the growth and development of the industry. Therefore, by bringing together diverse perspectives and insights, our e-magazine is expected to play an instrumental role in facilitating communication, collaboration, and innovation within the citrus industry, ultimately leading to a more prosperous and sustainable citrus sector.

I am confident that our e-magazine will serve as a valuable resource for multifaceted stakeholders in the Indian citrus industry. I also believe that it will help to promote greater awareness and appreciation of the importance of citriculture in Indian agriculture and society at large as well as contribute a little towards making our country a self-reliant developed nation.

I, on behalf of Indian Society of Citriculture, would like to thank all the contributors from India and abroad and also the editors who have made the first issue of e-magazine "Indian Citriculture" possible with a zeal to inspire further research, collaboration, and innovation in Citriculture.



Dilip Ghosh
(Dilip Ghosh)
President, ISC

Editorial

Citrus sector has immense importance in India, both economically and culturally. Our country is the third major producer of citrus fruits in the world. Moreover, citrus fruits are known for their nutritional value and health benefits. They are an excellent source of vitamin C, which is essential for maintaining a healthy immune system. They also contain a variety of other vitamins and minerals, all of which contribute to healthy living. In this context, citrus plays a crucial role in ensuring nutritional security for our citizens.

E-magazines are currently playing a significant part in the rapid dissemination of knowledge, the exchange of ideas, and ensuring a level of current awareness regarding the most recent advancements. Given this context, Indian Society of Citriculture, in collaboration with ICAR-Central Citrus Research Institute, took initiative to publish this e-magazine "Indian Citriculture" covering a broad range of subjects, from the most recent developments in citrus production to cutting-edge production and processing technologies.

"Indian Citriculture" features insightful articles written by knowledgeable authors in the field. It offers a wealth of knowledge and in-depth look into the various aspects of citriculture. Additionally, it emphasizes the significance of environmentally responsible business practices for the growth of a strong citrus value chain. The e-magazine is expected to emerge as a valuable resource for citrus enthusiasts who are interested in gaining a deeper understanding about the improved varieties, innovative technologies, processed products, and marketing of citrus fruits.

The editorial board hopes that this e-Magazine will be useful to researchers, academicians, extension functionaries, students, citrus growers and other stakeholders involved in citriculture. We are seeking constructive feedback, innovative suggestions and contributory articles from readers and experts all around the world in order to better the upcoming editions.

Editors

Good Nutrition Management is the First Step to Good Orchard Management

Tripti Vashisth* and Jamie D. Burrow

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Abstract: Good fertilization practice is critical for the optimum production of a crop. The two main objectives of nutrition management in citrus trees are optimum and consistent yield; and building a strong tree that can grow year after year. Therefore, a nutrition program needs to be carefully managed and assessed to ensure both objectives are met.

There are 14 mineral nutrients that are essential for normal plant growth and development in addition to carbon (C), hydrogen (H), and oxygen (O), which plants take up from the surroundings *via* leaves and roots. These nutrients are often viewed simply as plant food necessary for better plant growth and yield. Citrus trees perform best in the optimum range of each nutrient, and nutrient deficiencies or excess of any nutrient can result in poor tree growth and low yield. For example, researchers have demonstrated that the fruit yield and leaf nitrogen concentration have a concave relationship where maximum yield can be achieved between 2.5% to 2.7% nitrogen concentrations in 4- to 6-month-old spring growth of healthy orange trees. Any variation from optimum range on either side of the curve can result in decrease yield. However, mineral nutrition also influences growth and yield by affecting plant resistance or susceptibility to pathogens and pests.

Nutrition and plant-disease interactions

Mineral nutrients are essential for the growth and development of plants and microorganisms and are important factors in plant-disease interactions. How each nutrient affects a plant's response to disease, positively or negatively, is unique to each plant-disease complex. In general, nutrient-pathogen interactions are not well understood due to their complex nature and dependence on a number of external factors. Plant nutrient deficiency or toxicity may affect

disease susceptibility through plant metabolic changes thereby creating a more favorable environment for disease development. When a pathogen infects a plant, it alters the plant's physiology directly or indirectly, particularly with regard to mineral nutrient uptake, assimilation, translocation, and utilization. Pathogens may immobilize nutrients in the soil or in infected plant tissues. They may also interfere with translocation or utilization of nutrients, inducing nutrient deficiencies or toxicities.

Although disease resistance is genetically controlled, it is considerably influenced by environmental factors. Some disease resistance genes in plants are only activated by specific environmental stimuli. Mineral nutrition is an environmental factor that can be easily controlled in agricultural systems, the effects of which can be substantial.

A balanced nutrient supply ensures optimal plant growth and is usually considered optimal for disease resistance as well. As a rule, plants with an optimal nutritional status have the higher resistance (tolerance) to pests and diseases compared to nutrient deficient plants. Susceptibility to disease increases as nutrient concentrations deviate from optimum. The

interaction between plants and disease and pests is complex. However, the roles of mineral nutrients are well established in some areas of host-disease interaction. The goal is to recognize these interactions and see the possibilities and limitations of disease and pest control by mineral nutrition and fertilizer applications.

Nutrition and huanglongbing research

In the last decade, several scientific reports have demonstrated that good fertilization improves the health and productivity of citrus greening or huanglongbing (HLB)-affected trees. However, conflicting reports about the non-conclusive effects of fertilizer on HLB-affected trees has led to confusion. Plant nutrition is a complex topic, number of factors affect such as soil type, form of nutrient, irrigation method, soil pH, and the efficacy of nutrients in plant and soil. There is no limit to the number of combinations (type of fertilizer-location etc.) that can be tested in field for demonstration.

In a recent fertilizer trial, the same fertilizer treatments were applied at two different sites on mature 'Valencia' grafted on Swingle trees of similar age. After five years of the experiment, it was concluded that the use of a constant supply of nutrients along with a 20% higher rate of micronutrients can significantly improve the yield compared to standard fertilization practices. However, the best-performing treatments were not the same at the two sites. The best performer in site 1 was the worst performer at site 2 and *vice versa*. These findings suggest that a fertilizer program cannot be adopted without site-specific adjustments. Multiple factors such as soil profile, tree nutrient level, tree health, HLB status, etc. play a critical role in the success of a nutrition program. Each site is unique, and a fertilizer program should address the specific needs of the

site and grove. Therefore, regular leaf sampling to identify tree nutrient status and requirements should be part of managing a fertilizer program.

Leaf sampling method case study

Collecting the correct leaf sample is the first step in making fertilizer decisions. Random leaf collections around the tree canopy can lead to unreliable outcomes. Thus, using a standard practice for leaf sampling (e.g., leaf samples taken either from fruiting or non-fruiting branches) is important for determining the baseline of tree nutrition requirements. Correct leaf sampling can influence the right diagnosis and treatment. Historically, citrus tree nutrition was studied extensively to determine the best time and method to sample leaves for nutrient analysis. Research indicated sampling in July-August from non-fruiting spring-emerged leaves was when nutrient concentrations were highest, therefore the best time to sample for an accurate evaluation of trees nutrition status. One critical component to the nutrient sampling method is leaf type location- fruiting or non-fruiting branches. In several countries (such as Brazil and South Africa), leaf samples are collected from fruiting branches, whereas in other countries such as the United States leaf samples are collected from non-fruiting branches. The question remains, which is right or is there a right location to select leaf samples? Citrus trees are evergreen, perennial with one of the longest fruit growing periods. Some sweet orange varieties like 'Valencia' have 12–14-month fruit growth period, therefore it always has growing fruit. Citrus tree leaf nutrient accumulation changes seasonally as fruit develops. Huanglongbing management suggests multiple leaf sample analysis to monitor tree nutritional needs in 'real-time' as HLB-affected trees benefit from a constant supply of nutrients. Leaf analysis from fruiting branches is a current

analysis of a tree's needs, whereas non-fruiting branches will be the next season's crop. This further implies the importance of collecting the right leaf to maintain optimum nutrient levels and meet the high nutritional demands of HLB-affected trees.

The recommendation in Florida, USA, is to sample 4-6-month-old spring leaves from non-fruiting branches. However, 'non-fruiting branches' do not always receive much attention. When the fruit is green a very careful assessment of the branch is required. To understand whether it is critical to sample non-fruiting branches for nutrient analysis, we took leaf samples from either fruiting or non-fruiting branches over a year to monitor variations in nutrient concentrations (accounting for seasonal variation).

A case study was done to determine differences and seasonal changes of leaf nutrient concentrations between fruiting and non-fruiting branches of HLB-affected sweet orange trees.

Methods

Ten-year-old 'Hamlin' and 'Valencia' trees with mild HLB symptoms were grown in Central Florida orchards and used during the 2021-2022 production season. Thirty leaves were collected from fruiting (3 to 4 leaves behind the fruit) and non-fruiting branches at four time points (Aug 2021, Dec 2021, Feb

2022, and May 2022). The ground leaf samples were sent to Central Florida Soil Laboratory (Bartow, FL) after washing and drying. Standard nutrient analyses for macronutrients (Nitrogen (N), Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg), and Sulphur (S) and micronutrients (Manganese (Mn), Zinc (Zn), Copper (Cu), Iron (Fe), and Boron (B) were performed. Statistical analyses were performed using a t-test at $\alpha < 0.05$.

Preliminary results

- Significant differences in nutrient concentration were mostly observed in December compared to any other month (Table 1)
- Presence of fruit on a branch influences nutrient concentrations, counter intuitively the differences are observed in the later stages of fruit development than early (Table 1)
- Generally, macronutrients were consistently lower in fruiting branches (Figures 1 and 2)
- Mn and Zn have the same consistent seasonal trends in both fruiting and non-fruiting branch types; lowest nutrient concentrations in May (Figures 3)
- No significant differences between branch type in Mg, Cu, Fe, and B; nutrient concentration levels remained optimum year-round (Table 1)




Fig.1: Fruiting branch
(Photo Credit: T.R. Weeks)



Fig.2: Non-fruiting branch leaves
(Photo Credit: J.D. Burrow, UF/IFAS)

Table 1: Seasonal leaf nutrient concentrations of fruiting (F) and non-fruiting (NF) branches



Nutrient (mg/kg)	February 2022		May 2022		August 2022		December 2022	
	F Mean	NF Mean	F Mean	NF Mean	F Mean	NF Mean	F Mean	NF Mean
N	20600 ^a	24000 ^b	22000 ^b	26200 ^a	22800 ^b	24800 ^a	22400	22400
P	1000 ^b	1300 ^a	1100 ^b	1400 ^a	1300	1500	1200 ^b	1500 ^a
K	11600 ^b	16200 ^a	14200	16800	11000	13000	11400 ^b	15800 ^a
Ca	28800	26400	31200	28800	28400	27800	29000 ^a	19400 ^b
Mg	3600	3500	4800	4700	4500	4500	3600	3600
S	4900	5600	5100	4900	3900	3900	4200 ^a	3100 ^b
Mn	80.8	66.4	31.4	32	100.6	126.4	101.8 ^a	46.2 ^b
Zn	83.4 ^a	55.2 ^b	26.2	24.8	138.4	169.2	116.0 ^a	23.8 ^b
Cu	13.4	11.0	10.4	11.6	11.2	11.2	7.4	5.4
Fe	67.6	65.8	61.6	64.8	63.2	66.4	67.6	60.2
B	61.4	68.4	46.8	50.0	52.0	62.0	49.4	52.0

Indicates optimum nutrient concentration; all other nutrient levels were below optimum except S which was high
 a: significantly higher; b: significantly lower

Graphics: T.R. Weeks, UF/IFAS

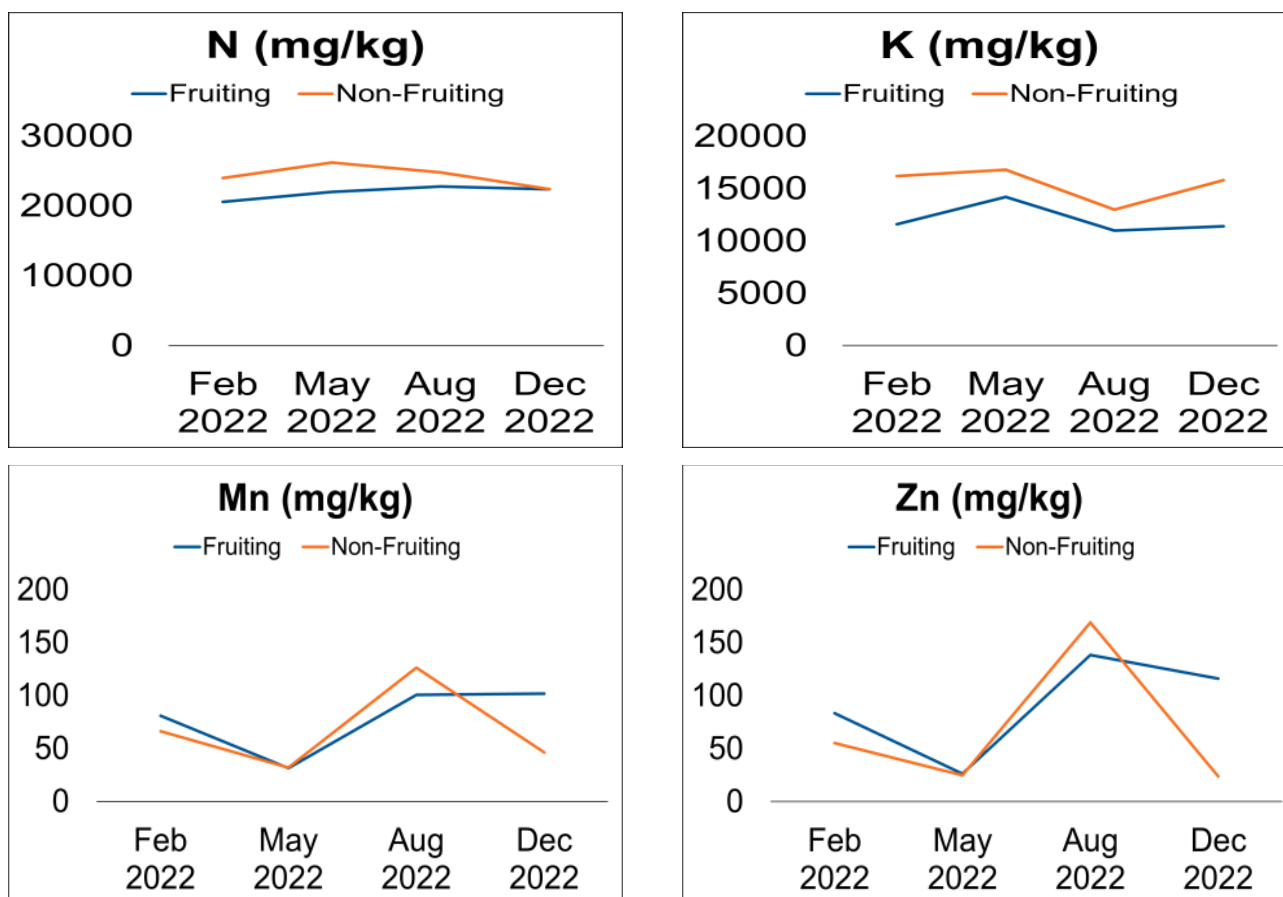


Fig. 3: Seasonal concentration pattern for Nitrogen (A), Potassium (B), Manganese (C), and Zinc (D) in sweet orange leaf

Case study summary

In summary, 'Hamlin' and 'Valencia' showed a similar pattern for nutrient concentrations in fruiting and non-fruiting leaves. Macronutrients (N, P, K) and micronutrients (Mn, Zn) were higher and lower, respectively, in non-fruiting leaves compared with fruiting leaves. In addition, this study also suggests that we need to pay more attention to micronutrients going into fall season when fruit drop increases. It is possible that excessive fruit drop is linked to a decrease in micronutrients in fall. Lastly, this study also highlights the importance of using the correct leaf samples for getting the most accurate leaf nutrient analysis. Consistency is key. Although fruiting branches are lower in the macronutrients year-round, leaf sampling from non-fruiting branches should continue for ease of management decisions (use of one management guideline for both bearing and non-bearing trees). Perhaps increasing the recommended nutrient ranges for citrus should be increased to account for the lower nutrient accumulation in the fruiting leaves of HLB-affected trees.

Summary

Essential nutrients are needed for a citrus tree to produce fruit from bud to harvest. Each nutrient plays a specific role and when a nutrient is deficient or in excess, it affects the growing process and fruit quality. Tree growth can also be affected by disease, but optimal leaf nutrient concentrations can provide a higher resistance (tolerance) to pests and diseases compared to trees that are nutrient deficient. Leaf nutrient analysis is important to determine the nutritional needs of a tree. Collecting the right leaves for analysis is the first step to making proper nutrient management decisions for both healthy and disease-affected citrus trees.

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Protein based Strategies to Combat HLB Disease of Citrus Plants

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Abstract: Huanglongbing (HLB), also known as citrus greening, is a devastating disease affecting citrus plants worldwide. Caused by the bacterium *Candidatus Liberibacter asiaticus* (CLAs) and transmitted by insect vectors, HLB leads to severe economic losses in citrus production. Currently, no effective control strategies exist to manage this disease. In this article, we present protein-based approaches to combat HLB. We focus on targeting essential proteins of CLAs, such as amino acid binding proteins of the ABC transporter family, which are crucial for the survival of the bacterium. Through bioinformatics and in vitro studies, we identified potential inhibitor molecules that showed significant efficacy in reducing CLAs titer in infected plants. Additionally, we explored the role of important defence proteins, including 2S Albumin, Lipid Transfer Protein and Peroxiredoxin, in the plant immune system against HLB. These proteins exhibit antimicrobial properties and have shown promising results in inhibiting disease progression. Furthermore, we discuss the development of stress-tolerant transgenic citrus varieties using plant defense genes, PRpnp and a seed storage protein, to enhance resilience against abiotic and biotic stresses, including HLB. Our findings highlight the potential of protein-based strategies as effective tools for managing HLB disease and improving citrus crop productivity.

Introduction

The world top ten citrus crop producing countries are Brazil, China, United states, Mexico, India, Spain, Iran, Italy, Nigeria, and Turkey. India occupies fifth rank in the production of citrus fruits worldwide. In India, citrus industry is third largest fruit industry after mango and banana. Citrus industries hold an important place in the economy of India as horticultural wealth. Citrus fruits originated in the tropical and sub-tropical regions of South East Asia, particularly India and China. The native place of many citrus species is the North East part of India. Citrus fruits originating in tropical and subtropical southeast Asia (particularly India and China), were among the oldest fruit crops to be domesticated. Citrus fruits are rich in vitamin C and in India various types of citrus fruits like orange (mandarin), sweet orange and lemon are grown for commercial importance. In India, top ten citrus

fruits producing state are Maharashtra, Rajasthan, Madhya Pradesh, Karnataka, Tamil Nadu, Manipur, Jammu and Kashmir, Gujarat, Andhra Pradesh and Arunachal Pradesh.

Citrus greening, often known as Huanglongbing (HLB), is the most devastating disease of citrus family plants and has been identified in East Asia (China, 1940) for more than a century ago. It is caused by endogenous, unculturable, phloem-restricted gram-negative bacterium, *Candidatus Liberibacter asiaticus* (CLAs). *Liberibacter* is transmitted from one tree to another tree by insect vectors citrus psyllids, *Diaphorina citri*. It is classified on the basis of its geographical origin and 16S rDNA sequence into three species, *Candidatus Liberibacter asiaticus* (CLAs), *Candidatus Liberibacter africanus* (CLaf), *Candidatus Liberibacter americanus* (CLam). Among three species, *Candidatus Liberibacter asiaticus* is the most widespread and virulent strain. After the

infection of bacteria, health symptoms in a fruit tree may not appear for two or more years. An early symptom of HLB infected leaf shows blotchy mottling with green islands. As the disease progresses, infected shoots are stunted and the branches gradually die due to disease progress. The fruit from infected trees are small, lopsided having dark aborted seeds tends to drop prematurely. Estimated loss of 30-100% was reported due to decrease in production capacity of HLB- diseased citrus plant.

Till date no effective control strategies have been developed for management of HLB disease. The most probable reason behind this is the late appearance of the general symptoms elicits a delayed defense response in plants and this manipulation of defense response is critical for its survival in plants. The current disease control strategies include minimizing the psyllid population chemically and biologically and prevent trees from becoming infected by removing the infected tree. However, targeting some of the crucial proteins which are essential for survival of the bacterium holds a promising strategy to combat the bacterium. Proteins are the most versatile biological macromolecules in living systems and perform diverse array of functions in essentially all biological processes. They function as transporters, catalysts, provide mechanical support, immune protection, transmit nerve impulses, storage of other molecules such as oxygen and control growth and differentiation. Therefore, one of the potential strategies is to target essential proteins of CLAs, critical for the survival of bacteria. In our lab, we are working on different strategies to combat HLB disease including targeting important proteins of CLAs such as ABC transporter amino acid binding proteins for developing effective potential inhibitor molecules. And we are successful in developing two inhibitor molecules against cystine binding

protein of CLAs. Also we have designed a novel 2S albumin-zinc oxide nanoparticle conjugate where 2S albumin protein was extracted from the seeds of *Cucurbita maxima* has shown excellent results by inhibiting the progression of HLB disease in crop. Our lab also works on defense mechanism of citrus plants using important defense proteins like Lipid transfer protein (LTP) and peroxiredoxins. We have also developed stress tolerant transgenic variety of citrus using PRpnp, a novel PNP family protein from *Putranjiva roxburghii* and a seed storage protein 2S albumin from *Cucurbita maxima*.

Strategies to combat HLB disease

Evaluation of potential inhibitors against essential proteins of CLAs

ABC transporter protein families are present from microorganisms to human beings. They play an important role in transporting the solute across the cell membrane and are targets for inhibitor development. Bacteria have a variety of ABC transporters that are involved in the import of a number of molecules necessary for the bacterial survival. The periplasmic amino acid binding proteins, a component of ABC transport family proteins, are known drug targets in prokaryotes and eukaryotes. CLAs genome analysis showed presence of two putative amino acid binding proteins belonging to ABC transporter family. Of the two, one is specific for cationic amino acid while other is a putative cystine binding protein (CLAsTcyA). The choice of CLAsTcyA, a putative cystine binding receptor, as a target for inhibitor development to control Huanglongbing disease was based on its importance and unique features. The importance of CLAsTcyA, one of the two amino acid binding receptors in CLAs, in survival of the bacterium is quite evident from the fact that it is highly expressed along with its permease in the phloem of the host plant. The

second one with specificity to cationic amino acids is mainly expressed in the host citrus psyllid. The characterization of CLasTcyA, in our lab, revealed unique features in terms of sequence and structure as compared to other related proteins. The crystal structure of CLasTcyA was taken as template for screening of inhibitors. Among many compounds, selected through bioinformatics study, pimozone, clidinium, sulfasalazine and folic acid showed significantly higher affinities and stability in complex with CLasTcyA. Out of the four screened compounds, SPR study showed higher binding affinities of CLasTcyA for Pimozone and Clidinium. Likewise thermal stability studies employing DSC showed higher stability for CLasTcyA in complex with two selected inhibitors. *In planta* studies, carried out to assess the effect of inhibitors on HLB infected Mosambi plants, showed significant reduction in CLas titer in plants treated with inhibitors as compared to control plants. Our results showed that *in vitro* and *in planta* studies were successful in identifying effective inhibitor molecules against CLasTcyA and establishing their efficacy against the pathogen.

Studies of plant defense mechanism using important defense proteins of Citrus

We are very aware of the defense mechanisms and immune system of human body to fight against the bacterial pathogens, viruses, fungal infections. The immune system is so well developed that it can keep the memory of previous infections experienced long ago and develop resistance against the pathogen attack in the form of IgG antibodies. But, have you ever thought that plants can also have some defense system and their own strategies to tackle the devastating plant pathogens? Plant immune system is divided into two categories as innate immunity and systemic acquired resistance. During the course of co-evolution of plants and

pathogens, plants have evolved in such a way that as soon as they encounter any invading pathogen or physical damage they start activating signalling pathways. Messenger molecules activate other genes for the expression of specialized protein molecules called defence proteins and secondary metabolites that directly or indirectly inhibit the pathogen to reduce crop damage. It is very interesting to understand exactly how the defense protein protects the plant from attacking pathogens and insect pests. We are investigating the effect and mechanism of action of 2S albumin protein from Pumpkin and Lipid Transfer Protein from Citrus using various advanced techniques involving structure-function relationship. 2S albumin is a storage protein found in seeds which protects the seed from insect pests during its dormant stage. 2S albumin is a multifunctional protein having various unique characteristics like very high temperature stability which makes it suitable to keep its structure intact even at harsh environmental conditions. Its small size and diverse functionality like antimicrobial, insecticidal property, inhibition of various proteases from pathogens makes it a promising agent to use as a broad spectrum therapeutic for treating plant diseases. We have identified its novel DNA degrading potential which gives us more insights into its mechanism of action as an antimicrobial agent. Our research team has designed a novel 2S albumin-zinc oxide nanoparticle conjugate to enhance the stability of 2S albumin and the product has shown excellent results by inhibiting the progression of HLB disease in crop. The results suggest that 2S albumin - nanoparticle conjugate has a great potential to come up as a therapeutic agent for the better management of crop associated diseases at large scale. Lipid Transfer Protein from Citrus crop is a versatile protein having very high temperature and pH stability

belonging to PR14 family of plant defense proteins. It has been observed that LTP protein is upregulated upon infection of *Candidatus Liberibacter asiaticus* along with some other plant defense protein. It is evident that LTP protein plays some important role in activating the defense response in plants but what kind of pathways it follows during these complex biochemical processes and protect the plant from attacking pathogens need to be investigated. Hence we found it very fascinating to study this enigmatic protein and explore its biotechnological potential for the management of plant diseases and insect vectors. First step to study any protein's function is to understand its structure and properties. Hence, we expressed Citrus LTP protein in bacterial system using recombinant DNA technology and purified the protein to study its biochemical and structural characteristic. Surprisingly, LTP protein showed very wide range of antimicrobial potential against bacterial and fungal phytopathogens along with some human pathogens even at very low concentrations. LTP protein is found to be very effective against the destructive pest cotton ball worm which affects several economically important crops like cotton, tomato, chickpea, sunflower, corn etc. in central and southern parts of India. Incorporation of LTP protein in the diet of cotton ball worm drastically inhibited the larval stage of its life cycle which is the most damage causing stage. Our study has proved that LTP protein from plants is a very potent candidate for the development of transgenic crops to protect the crop from phytopathogens and insects.

The antioxidant defense system is also an important domain for the study and exploration of its role in plant survival and development. Peroxiredoxin is the most significant antioxidant defense protein that participates actively to neutralize the effect of oxidative

stress. The characterization of peroxiredoxin was explored in detail and report the potent inhibitor molecules against that. The conoidin and celastrol, inhibitor molecules were used against peroxiredoxins from CLA to control the HLB. The specific activity of CLaBCP is mostly inhibited by conoidin and celastrol as compared to CsPrx. After inhibitor binding, the changes in secondary structure and fluorescence quenching were observed in CLaBCP and CsPrx. Similarly, the inhibitor-bound CLaBCP is thermally more stabilized as compared to CsPrx. The binding of conoidin and celastrol by using surface plasmon resonance (SPR) showed a strong affinity with CLaBCP as compared to CsPrx. The bioinformatics study was carried out to screen potent inhibitor molecules against peroxiredoxins from CLA. The 3-D model of CLaBCP and Prx was used to screen the inhibitors from the drug library. Strong binding affinity molecules were docked at the active site of CLaBCP and Prx. Molecular dynamics analysis such as RMSD, Rg, SASA, hydrogen bonds, and PCA results indicated that inhibitor complexes had lesser fluctuations and were more stable and compact complexes. MMPBSA results confirmed that the identified compounds could bind at the active site of CLaBCP and Prx to form a lower energy inhibitors complex. Hence, the characterization of peroxiredoxins, inhibitor binding studies, and screening of lead molecules may play important role in the development of inhibitors against CLA for the management of HLB disease.

Development of stress tolerant transgenic variety of citrus

The world population is projected at 7.9 million as of 2022 and is estimated to exceed 10 million by the end of 2050. However, fruit production has consistently been insufficient to meet the required demand for increased population. The lack of proper fruits in the human diet leads to a

lack of several micro- and macronutrients thus, causing malnutrition. In children and elderly people, fruits provide essential immunity to fight against several diseases. Citrus is one of the major fruit crops, grown in the tropical and subtropical regions of more than 140 countries and includes many species of economic importance, such as limes, oranges, lemons, grapefruit, and tangerines. Among the three classes of limes, is the globally predominant natural hybrid and represents 70% of total global annual citrus production. The prime producers of citrus include India, China, the USA, Brazil and Mexico. Despite being an important horticultural crop, the cultivation of citrus, including and is remarkably limited by several abiotic and biotic stresses including drought, salinity, Huanglongbing (HLB), alkalinity, extreme temperatures, oxidative stress and several bacterial, viral and fungal diseases. Therefore, developing stress tolerant variety is imperative. Among all the available methods, the most efficient and reliable approach for the development of stress resistant citrus cultivars is based on *Agrobacterium*-mediated transformation. In our study, we selected two plant defense genes i.e., PRpnp, a novel PNP family protein from *Putranjiva roxburghii* and a seed storage protein from *Cucurbita maxima* as overexpression targets to develop different stress resistant transgenic citrus. The transgenic Mexican lime overexpressing PRpnp had enhanced tolerance against salt, drought, alkaline pH and oxidative stress and two pests, *Papilio demoleus* and *Scirtothrips citri*. Further, to know the underlying reason behind overall improved resilience in transgenic Mexican lime, we performed comparative global transcriptome study. Under normal growth conditions, the differentially expressed genes (DEGs) involved in abscisic acid and jasmonic acid biosynthesis and signaling pathways, plant defense, growth

and development were significantly up regulated by PRpnp overexpression. We also anticipated the role of purine salvage enzyme Rpnp, in cytokinin interconversion reactions. Finally, we confirmed a higher accumulation of endogenous ABA, JA and CK in transgenic plants through LC-MS/MS analysis. Thus, PRpnp overexpression positively regulates multiple stress tolerance in Mexican lime by enhancing endogenous ABA and JA, which interact synergistically and it also inhibits trypsin proteases in the insect gut. Additionally, upon transient expression of PRpnp in tobacco cells, it showed nuclear-cytoplasmic localization which revealed its possible role in maintaining the intracellular purine reservoir. Overall, overexpression of PRpnp enhanced overall plant vigour in transgenic *C. aurontifolia*. In another study, we overexpressed a seed storage protein from *C. maxima* in *C. aurontifolia* and *C. sinensis* in phloem followed by testing their efficacy against HLB tolerance. The antimicrobial and defensive properties of the seed storage protein reduced the bacterial titer in HLB infected parts and prevented the infection development in the transgenic plants.

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Grapefruit – A Potential Citrus Crop of India

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Abstract: Grapefruit, a traditional citrus fruit, is primarily cultivated in the United States and Europe. Originating in the West Indies, grapefruit juice has antioxidant activity, vitamin C, and anti-tumor properties. In India, it is cultivated on small farms. After successful introduction and evaluation by ICAR-CCRI, the State Variety Release Committee of Maharashtra endorsed the Flame grapefruit cultivar in 2021. Grapefruits reach maturity between 210 and 260 days and must be harvested in stages. With a 5 x 5 m spacing, the yield potential is 32-36 tonnes per hectare, with 330-350 crops per tree. Proper nutrient management, crop regulation, mulching, intercropping and integrated plant protection measures are essential for successful commercial production, optimal growth, higher yield and better quality.

Introduction

Citrus is one of the important fruit crops in India. Although traditional citrus fruits, *i.e.* mandarin, acid lime and sweet oranges are popular, other citrus fruits such as grapefruit and pummelo are gaining popularity due to their nutritional properties. Grapefruit (*Citrus paradisi*) is believed to have originated in the new world (West Indies) and commercially it is grown mainly in USA and European countries. Grapefruit juice is ranked among the highest in antioxidant activity. It is very good for health and the most common fruit juice in Europe and America. From the nutritional security point of view this will be one of the future fruits in India. Grapefruit is also an excellent source of vitamin C, a vitamin that helps to support the immune system and may help to reduce cold, symptoms or severity of cold. The rich pink and red flesh colours of grapefruit are due to lycopene, a carotenoid phytonutrient (Lycopene is only found in pink and red grapefruit). Lycopene appears to have anti-tumor activity.

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Among the common dietary carotenoids, lycopene has the highest capacity to help fight free radicals, which can damage cells. It is reported to reduce risk of prostate cancer, as per research published in peer reviewed research articles.

In India, grapefruit is commercially grown in Punjab but on very limited scale with area of few hundred acres only. It is considered as exotic fruit and not known in other parts of the country. Now many farmers and farmers' groups are interested in planting for good variety of grapefruit due to its health benefits. Therefore, there is a need to have high yielding variety with desirable fruit quality *viz.*, attractive flesh colour and high juice content.



Fig. 1: Grapefruit cv. Flame

Keeping this need in view, grapefruit material of variety "Flame" was introduced from California, USA at ICAR-CCRI during 1990's. The budded plants were produced and evaluated in post-quarantine facility of the institute for freedom from insect-pest and diseases. When the plants were found free from any exotic or new insect-pest and disease, the shoot-tip grafting technique was utilized to get further clean material. Following the research trials, grapefruit cv. Flame was recommended for commercial cultivation by State Variety Release Committee in 2021.

The following are the salient features of the grapefruit cv. Flame.

Yield at 5 m x 5 m spacing	: 30-35 t/ha
Plant canopy	: Medium and suitable for 5 x 5 m spacing
Juice content	: 50-52%
TSS	: 9-11 ^o Brix
Titrateable acidity	: 0.7-1.2%
Fruit maturity (<i>Ambia</i> crop)	: October first fortnight
Fruit weight	: 260-320 g
Fruit colour	: Greenish yellow with characteristic red blush on surface
Flesh colour	: Deep red
Fruit bearing habit	: Inside the canopy



Fig. 2: Bearing Plant of Grapefruit cv. Flame



Fig. 3: Fruits and juice of Grapefruit cv. Flame

Plant propagation

Grapefruit (*Citrus paradisi*), is a natural hybrid citrus species and polyembryonic in nature. Therefore, it is propagated by budding on recommended rootstocks viz. rough lemon, Rangpur lime and Alemow. For healthy grapefruit trees, it is essential to procure planting material from certified citrus nurseries.

Soil and climatic requirements

Grapefruit can be grown on varied soil types and under different climatic conditions that prevail in different parts of India. A well-drained, medium deep (1 to 1.5 meters) soil with a pH of 6.5 to 7.5 is ideal for better growth and yield. The temperature range between 13°C to 37 °C is optimum for active growth of vegetative and reproductive parts. The areas which are warm with moderate humidity and free from frost, strong winds and excessive rainfall are ideally suited for its cultivation. Grapefruit is sensitive to frost conditions.

Plantation

Proper layout of Grapefruit orchard is necessary. Most commonly practiced system for Grapefruit is square system. In square planting method, the orchard space is divided into squares and the trees are planted at the four corners of the square, in straight rows running at right angles. Now a days planting on a raised bed is also recommended. Since Grapefruit plants grow luxuriantly in all directions it is advisable to provide a spacing of 6 x 6 m depending upon the soil type. For light soils even a distance of 5 x 5m is adequate. Recently, under high density planting 5 x 2.5 m was also found suitable on raised bed plantations.

The new plantations may be done by digging pits. Pits should be filled before the rainy season for proper settling of the loose soil. In light soils 60 x 60 x 60 cm planting pits are sufficient while in heavy soils 90 x 90 x 90 cm pits should be prepared. At hill slopes, the pits should be of 1 x 1 x 1 m size along the contour. The pits should be filled with good quality soil mixture, containing farm yard manure, neem cake, vermi-compost, *Trichoderma* rich manures and bio-fertilizers. As a common initial dose, *Trichoderma* enriched FYM (30-40 kg), Single Super Phosphate or bone meal (500 g) and about 10 g Carbofuran 3% CG granules (to take care of soil borne insects) are mixed with soil and applied to one pit. The upper level of pit is kept 15 cm above the field level. After filling, the pits should be irrigated to settle down the soil.

Orchard management

Flowering and crop regulation: Grapefruit trees produce cyclic growth flushes twice the year. In order to have concentrated commercial bloom, crop regulation is necessary. In the case of Grapefruit, mostly *Ambia bahar* (February-March bloom) is taken. Seldom *Mrig bahar* (June-July bloom) is taken as the natural stress

during summer induces flowering at the onset of monsoon rains in central and southern parts of India. During flowering, adequate water management with double ring method or drip irrigation is advocated. Foliar application of 2,4-D or GA₃ (15 ppm) along with urea (1 %) reduces fruit drop and increases fruit size.

Mulching: In Grapefruit plants (7-8 years old) at 5 x 5 m spacing, black polyethylene mulch has been more effective. The difference between moisture content under different mulches has not been very significant at initial stages, however, after six months, soil moisture was found to be higher under black polyethylene than paddy straw and grasses. The fruit yield of Grapefruit has been highest in black polyethylene sheet followed by white polyethylene sheet, grasses, soybean straw and paddy straw, respectively.

Intercropping: Leguminous crops like soybean, black gram, gram, green gram can be taken in Grapefruit plantation. Generally, 2-3 feet space should be left from tree trunk on both sides and intercrop can be taken in straight rows so that trees are not damaged. Intercrops should not be taken after five years. Care should be taken that water do not stagnate near tree trunk when intercrops are irrigated.

Nutrient management: With application of 800 g N, 200 g P and 300 g K per plant yield of 50-55 kg, 70-75 kg and 78-80 kg fruit per plant, can be obtained in first, second and third year, respectively of commercial production *i.e.* sixth, seventh and eighth year of tree age. Plants need sufficient organic manure to produce this yield.

Out of different micronutrients, nutrients like Fe, Mn, Zn and B are more responsive on Grapefruit. Foliar application of 0.5% FeSO₄ or 0.5% MnSO₄ or 0.5% ZnSO₄ give good result. In order to prepare 0.5% of FeSO₄ / MnSO₄ / ZnSO₄ solution, dissolve 500g FeSO₄ / MnSO₄ / ZnSO₄ in 100 liters of water and spray on the

plant till drench. Generally, it is better to give micronutrient application as foliar sprays when new leaves are completely developed / grown but still young. Ring should be made at outer periphery of plant canopy and fertilizers should be mixed in soil followed by watering. FYM (full dose) and half of NPK dose should be applied just before monsoon. The remaining half of NPK should be applied in October-November followed by irrigation.

Irrigation: For the sustainable Grapefruit fruit production and quality, it is necessary to supply adequate irrigation in the dry season, and proper drainage during the wet season. It is important to provide the right amount of water and fertilizers at different growth stages not only to enhance the growth of trees, but also to improve yield and fruit quality. For efficient water management of Grapefruit trees, consideration should be given to (i) water requirement of the crop (ii) scheduling of irrigation (iii) irrigation method, and (iv) other practices such as *in situ* water harvesting and moisture conservation coupled with use of mulches, etc.

Insect-pests management: The major citrus insect pests include citrus psylla, leaf miner, thrips, fruit piercing moth and mites. The standard control measures are well placed to control major pests normally.

Prophylactic measures for disease management: Pre-monsoon and post-monsoon application of Bordeaux Paste (1 kg CuSO₄: 1 kg Lime: 10 lit. of water) on tree trunks. The neutrality of the paste was checked before application. Foliar spraying of fosetyl Al (0.25%) (twice during August and October at 40 days interval). Removal of dry twigs (if any) immediately after harvest, followed by a fungicidal spray (Copper oxychloride @ 0.3% or Thiophanate methyl @ 0.1 %).

Fruit harvesting and marketing: Bearing habit of grapefruit is in bunches of 5 to 12 fruits.

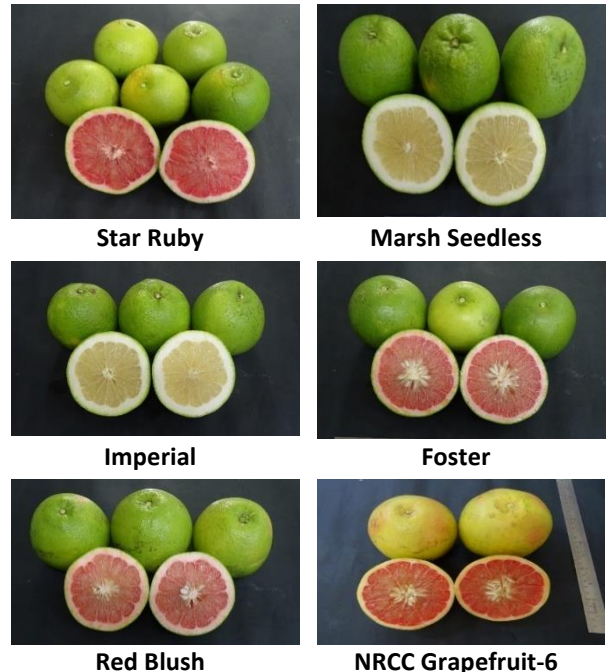


Fig. 4: Different varieties of grapefruit

Grapefruit fruits are harvested when they turn from dark green surface to light green yellowish smooth surface with characteristic pink blotch. Fruits mature within 210-260 days from fruit set and need to be harvested in instalments as all the fruit do not mature at a time. Fruits are manually harvested with a hook attached to a bamboo pole or by shaking branches. Yield potential is about 32 - 36 ton/ ha if planted at 5 x 5 m spacing bearing about 330-350 fruits/ tree.

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New Acid Lime Variety for the Plains of North-western Regions of Jammu

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Abstract: Acid lime (*Citrus aurantifolia* Swingle) is an important commercial fruit crop, cultivated in subtropical areas as well as mid hill areas of Jammu region. However, production and productivity is very low due to various reasons including lack of high yielding varieties, infestations by various diseases and pests. In this context, determination of genetic variation at molecular level is fundamental to citrus breeders for the development of elite cultivars with desirable traits. In the present study, JMU- acid Lime (Sel-70) is an acid lime variety developed by SKUAST-Jammu. It is precious and prolific bunch bearing variety of acid lime having vigorous and spreading growth habit with low density of spines. As per the citrus descriptor the data was collected on morpho-physiological and molecular characterization of JMU- acid Lime (Sel-70).

Acid lime (*Citrus aurantifolia* Swingle) is the third most important citrus species in India after mandarin and sweet orange. In Jammu, lime (acid lime and sweet lime) accounts for an area of 4.97 thousand ha with the production of 12.74 thousand MT (2018-19, Department of Horticulture, Jammu). Acid lime (*Citrus aurantifolia* Swingle) is one of the largest growing and ever-demanding commercially growing fruit cultivated in Jammu province. JMU- acid Lime (Sel-70) is a medium sized, hardy, semi vigorous, upright growth, thorny fruit tree. Fruits are round to oval. Fruits mature in an irregular pattern throughout the year are greenish yellow in colour and have a very acidic juice. Seeds are small, smooth and cotyledons whitish. JMU- Acid lime exhibits cyclical growth flushes throughout the year. The three blooming seasons for acid lime in Jammu conditions are Mrig bahar (July - August bloom), Hasta bahar (September–October bloom) fruits fetch high price in the market, and Ambe bahar (February to March bloom).

Acid lime cultivation is becoming very popular in Jammu sub-tropics because of its high productivity and good fruit quality. The studies conducted on biodiversity of acid lime in Jammu subtropics showed promising results among the lime genotypes of seedling origin. During the investigation, the survey was conducted in major lime growing districts viz., Jammu, Samba, Kathua, Udhampur and Reasi of Jammu province, UT of J&K. In these regions, the maximum areas of existing old citrus plantation are predominantly from seedling origin as plants are naturally propagated through selected seed from meritorious indigenous. A wide range of variability with respect to fruit attributes were noticed in various locations of Jammu province. On the basis of preliminary field observations and interactions with owners, the trees which were bearing extremely small sized, poor quality fruits and low yielding were not included. Finally, plants of seventy (70) seedling origin lime genotypes with divergent characters were selected at fruit maturity stage

on the basis of size and fruit quality. Coding was allotted to each selection on the basis of their location and geo tagging was done on selected plants.

The research work was carried out at SKUAST-J, Chatha, Jammu. Thus, among selected genotypes after screening of wild genotypes of acid limes in Jammu region, genotype code JMU-Acid Lime (Sel-70) was given to the selected acid lime genotypes of seedling origin since, there were no standardized names for the selected seedling origin acid lime genotypes therefore codes were allotted to each selection as JMU-acid lime (Selection-1, 2, 3). This brand name was given by SKUAST-Jammu for all its released varieties with the prefix “JMU”. Regular visits were made during the period of flowering, fruit setting, fruit maturity and ripening stage.



Fig. 1: Full grown tree of promising lime genotype JMU-Acid Lime (Sel-70)

As per the citrus descriptor given by IPGRI 1999 the data was collected where maximum average fruit weight (56.26 g), fruit diameter (49.57 mm), fruit length (55.70 mm), total yield (160.28 Kg/tree), TSS (8.89 °brix), acidity (8.35 per cent), ascorbic acid content (36.81 mg/100ml juice) and juice per cent (57.13 per cent) were recorded highest in seedling lime selection coded with JMU- acid Lime (Sel-70). After analysing the data of the collected lime genotypes on the basis of citrus descriptor, the bud wood of best seedling lime genotype

selection JMU- acid Lime (Sel-70) was collected and budded on the rough lemon rootstock at SKUAST-Jammu for its further multiplication.

Molecular characterization of acid lime genotypes

Molecular characterization of seventy indigenous acid lime genotypes of Jammu region was carried out to determine genetic diversity among acid lime genotypes by using 25 SSR primers out of 25 SSR markers, genotype JMU- acid Lime (Sel-70) amplified and showed high levels of polymorphism/allelic diversity in 21 primers.

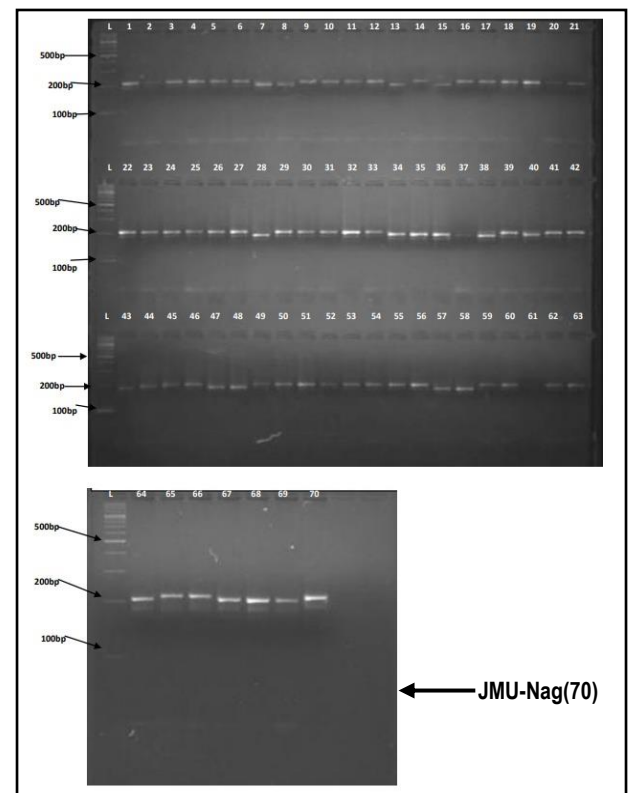


Fig. 2: Amplification profile of 70 acid lime genotypes using BQ624796 primer (L stands for ladder 100bp)

Propagation and rootstock

The propagation of quality plants need extreme attention as citrus is vulnerable to viruses and mutation. The quality of nursery plants has a

major influence in the productivity of citrus orchard. Most of the citrus fruits are generally propagated through T-budding. Lime seeds are poly-embryonic and generally multiplied sexually (seeds). To maintain duplicate clones in limes asexual propagation is practised. Choose disease-free mother plants while planting through layering. The most suitable rootstock for lime is Jattikhathi.

Rootstock raising

The seeds used for rootstocks should be taken from healthy fruits harvested from vigorous trees. Fruits that are very near to the ground should not be taken for extraction of seeds as there is a high chance of Phytophthora infection. The seeds of Jattikhathi are extracted in the month of August-September. The fresh seeds of rootstocks are sown in beds at 2 cm x 10 cm distance and 1.0 cm deep which germinate 3 weeks after sowing. For obtaining higher productivity, selection of nucellar seedlings by eliminating weak seedlings is an ideal step. When the seedlings reach to a height of about 15 cm, the seedling should be transplanted in nursery where layering operations are to be performed by using moss grass or roots of water hyacinth as a rooting media. The following points need to be considered, while selecting and preparing scion wood for grafting.

1. The scion sticks and rootstock should be of the same thickness.
2. The scion sticks should be taken from terminal non-flowered shoot, that is 3 to 4 months old.
3. The scion stick should be defoliated leaving a portion of petiole 7-10 days before their detachment from the mother plant.
4. Under north Indian conditions, ground layering can be done from February to March.

Land Preparation

Land should be well-ploughed to full tilth and properly leveled. Acid lime plants are enormously susceptible to water stagnation during rainy season, A proper drainage system is a must-have.

Planting

The best time to plant limes (*Citrus aurantifolia* Swingle) is between June and August, with a planting distance of 5-6 metres. Plant acid lime plants in pits measuring 50 cm x 50 cm x 50 cm or 75 cm x 75 cm x 75 cm and irrigate afterward. Before planting, apply FYM at a rate of 5-10 kg per pit. During the winter, young plants should be protected from the cold.



Fig. 3: Flowering and Fruiting in JMU-Acid Lime (Sel-70)

Irrigation

Always opt for light irrigation, as a higher frequency is more favourable to plant growth, especially in the early stages. Water must not contain salts in excess of 1000 parts per million (PPM), as this is harmful to citrus plants. To protect plants from root and collar decay, avoid flooding them with water. The frequency and amount of watering should be determined based on the soil texture and rainfall conditions. During the summer, bearing plants should be watered every 10-15 days, while a gap of 15-20 days is preferable during the winter. Irrigate the young plant (3-4 years) at least once a week.

Micro irrigation, also known as deep irrigation, is a method of conserving water while also providing plants with the water and nutrients they require.

Training and Pruning

Citrus trees may be pruned at any time, but if we want to grow a tree with a solid stem it is better to avoid those periods when the trees are in active growth. Support the main stem with bamboo stick that helps them stand erect during high winds or downpour. The ideal time for pruning of bearing trees is after the fruit harvest during late winters or early spring. Make sure that branches are evenly distributed (as per as practicable) to both sides and remove cross twigs or water suckers in early stage. Branches that are diseased, drooping, or wounded should be pruned on a regular basis, but for bearing plants, little or no trimming is suggested.

Intercropping and weed control

In bearing orchards intercropping should be avoided. But in young and non-bearing orchards, intercropping with shallow rooted, short duration leguminous crops upto four years is desirable. Independent irrigation and fertilizers should be added to fulfil the requirements of intercrops. In order to control different kinds of weeds light cultivation of the field should be done.

Maturity indices of lime

Unlike some other fruits, citrus fruit do not ripen further once they have been removed from the tree, so it is important that they are picked at the right stage of maturity. Maturity is measured depending on different characteristics such as color, juice content, level of soluble solid (sugars) and solids to acid ratio. Citrus fruits are typically harvested by hand.

Harvesting and yield

In general, maturity of acid lime depends largely on certain factors including nutrition, farming techniques, climatic condition, moisture availability etc. Among all citrus groups of fruits, acid lime takes a shorter maturity period and matures in about 5-6 months from the time of flowering.

In north India, the main harvesting season is in the month of August-September. Greenish yellow fruits picked by hook and total 6-8 pickings per season. The mature fruits of 'JMU-acid Lime (Sel-70)' fruit are available throughout the year, however, a tree gives production of 160 kg/tree.

Plant protection

Acid lime is affected by numerous diseases and pests, resulting in variable losses, starting from slight effects on leaves, roots, stems or fruits to reduction in growth and yield or complete decline.

Citrus tristeza virus (CTV), exocortis, and ring spot virus, for example, have a significant impact on citrus production around the world.

1. Citrus Tristeza Virus (CTV): CTV is a serious citrus disease. Millions of citrus trees were declined as a result. Veinlet clearing in young leaves, necrosis of cells at the bud union, honeycombing of the main stem, and inverse pits on the bark are all symptoms of this disease.

Control

- Disease-free bud wood and only tolerant rootstocks, such as Jatti Khatti, should be utilised to control the disease.
- Use insecticidal sprays to control the insect vector (aphid) that spreads the disease.

2. Exocortis (*Citrus exocortis viroid*): It is not common on Kinnow and sweet orange budded on Jatti Khatti. In some species like Lime, it

causes yellow blotches and cracks on shoots with stunting of trees.

Control

- a) Use of bud wood that is disease free.
- b) Only rootstocks with high resistancesuch asJatti Khatti should be used.
- c) Through the cutting tools, it spreads. To prevent infection from spreading to healthy trees, sterilise them after each interaction with diseased plants.

3. Ring Spot (*Citrus ring spot virus*): The disease's initial signs are yellow rings on mature leaves. The number of rings per leaf varies from one to numerous, and their diameter varies. By covering the entire leaf, these rings can join to form enormous patches. The heavily diseased plant shows signs of die-back and decline, as well as limited fruit yield. The virus is passed from infected bud wood to infected bud wood.

Control

Only use virus free plants for raising the citrus orchard.



Fig. 4: Harvested fruits of JMU-Acid Lime (Sel-70)

Conclusion

Acid lime is highly demanded fruit crop of Jammu Province however, Jammu is not self-sufficient in Acid lime production and large volume has to be imported from India to fulfill the market demand. Although, geo-climatic condition of Jammu is highly suitable for acid lime cultivation, its production per hectare is comparatively very low in

comparison to other states in the country. Therefore, on the basis of genetic assessment and molecular characterisation, the seedling origin genotype JMU- acid Lime (Sel-70) performed best for most of the horticultural traits and was found to have good potential for commercial cultivation under subtropical climate of Jammu region therefore, on the basis of the studies conducted, the JMU- acid Lime (Sel-70) is ready to get released for commercial cultivation in Jammu region.

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The Enigmatic World of Citrus Flowering: A Mystery Worth Discovering

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Abstract: Flowering is an essential phase in the development of citrus trees, as it initiates the production of delectable fruit. Flowering is influenced by tree age, climate, nutrient availability, and hormonal balance. The timing and intensity of flowering are affected by environmental factors such as light, temperature, and humidity. Gibberellins, cytokinins, and auxins play a crucial role in the hormonal equilibrium that regulates flowering. Blooming is controlled by an intricate network of genes and signalling pathways, including *SOC1*, *FLC*, *WUSCHEL*, *FT*, and gibberellin production and signalling pathways. The *FLC* gene suppresses blooming, while the *FT* gene stimulates flowering. The gibberellin biosynthetic enzymes *GA20ox* and *GA3ox* are upregulated during flower induction and development, while gibberellin receptor *GID1* and downstream transcription factors *DELTA* are downregulated. Biotechnology techniques like genetic engineering and genome editing have gained popularity in recent years, allowing for overexpression of *FLC* and *FT* genes to stimulate early and prolific flowering. Further study in this field could lead to significant improvements in citrus horticulture and production.

Citrus flowering is a critical process in the development of citrus trees that leads to the production of high-quality fruits. It is a complex physiological process that involves the regulation of gene expression, hormone balance, and environmental factors. Understanding the mechanisms that regulate citrus flowering is critical for the development of new citrus cultivars with improved flowering and fruit production traits, as well as for the optimization of citrus production practices to maximize yield and quality.

Factors influencing citrus flowering

The timing and intensity of citrus flowering are influenced by various endogenous and exogenous factors, including tree age, environmental conditions, nutrient status, and hormonal balance. One of the most critical factors influencing citrus flowering is tree age. Citrus trees typically start flowering after they reach a certain age, which varies depending on

the cultivar and growing conditions. For example, sweet orange trees may start flowering after 3-5 years of growth, while some mandarin cultivars may take up to 8 years to flower.

Environmental conditions also play a critical role in citrus flowering. The availability of light, temperature, and water can affect the timing and intensity of citrus flowering. For example, low temperatures and short photoperiods can delay or inhibit flowering, while high temperatures and long photoperiods can promote early and abundant flowering. Water availability can also affect citrus flowering, as water stress can delay or reduce flowering and fruiting.

Hormonal balance is another critical factor influencing citrus flowering. Several hormones, including gibberellins, cytokinins, and auxins, play a role in the regulation of citrus flowering. The balance between these hormones is tightly regulated, and any disruption in their balance can affect flower induction and development.

For example, high levels of gibberellins can promote flowering, while high levels of cytokinins can inhibit it.



Fig. 1: Citrus flower in full bloom

Molecular mechanisms regulating citrus flowering

Understanding the molecular mechanisms that regulate citrus flowering is critical for the development of new citrus cultivars with improved flowering and fruit production traits, as well as for the optimization of citrus production practices to maximize yield and quality. Advances in molecular biology techniques, such as transcriptomics, proteomics, and metabolomics, have enabled researchers to study the expression patterns and interactions of various genes and signaling pathways involved in citrus flowering in more detail.

The molecular mechanisms that regulate citrus flowering are complex and involve the interactions between various genes and signaling pathways. Several key genes and molecular pathways have been identified as regulators of citrus flowering, including the *SOC1* (*SUPPRESSOR OF OVEREXPRESSION OF CONSTANS 1*) gene, *FLOWERING LOCUS C* (*FLC*), *WUSCHEL* gene, the *FLOWERING LOCUS T* (*FT*) gene, and the gibberellin biosynthesis and signaling pathways.

The *FT* gene is another critical regulator of citrus flowering. The *FT* gene encodes a protein that promotes flowering by activating genes that regulate flower development and maturation. The expression of *FT* is regulated by various environmental and endogenous factors, such as light, temperature, hormones, and nutrient status. Studies have shown that overexpression of the *FT* gene can promote early and abundant flowering in citrus trees, while its suppression can delay or inhibit flowering.

The *FLC* gene is involved in the vernalization induction of flowering in citrus. The *FLC* gene encodes a transcription factor that represses the expression of genes involved in floral development, thereby delaying the onset of flowering. The regulation of *FLC* expression involves complex interactions between various environmental and endogenous factors, including temperature, light, and hormonal signals. The study of the *FLC* gene has provided important insights into the molecular mechanisms that control flowering time in citrus.

The gibberellin biosynthesis and signaling pathways are also critical for citrus flowering. Gibberellins are hormones that promote vegetative and reproductive growth in plants, and their balance is tightly regulated by feedback mechanisms and interactions with other hormones. Several genes involved in the gibberellin biosynthesis and signaling pathways have been identified as regulators of citrus flowering. For example, the genes encoding the gibberellin biosynthetic enzymes GA20ox and GA3ox are upregulated during flower induction and development, while genes encoding the gibberellin receptor *GID1* and the downstream transcription factors *DELLA* are downregulated.

In recent years, there has been increasing interest in the use of biotechnology tools, such as genetic engineering and genome editing, for the improvement of citrus flowering and fruit production. For example, the overexpression of key genes involved in citrus flowering, such as the *FLC* and *FT* genes, has been shown to promote early and abundant flowering in citrus trees. Similarly, the use of genome editing tools, such as CRISPR-Cas9, has the potential to generate new citrus cultivars with improved flowering and fruit production traits.

Conclusion

Citrus flowering is a critical process in the development of citrus trees that leads to the production of high-quality fruits. The timing and intensity of citrus flowering are influenced by various endogenous and exogenous factors, and the regulation of citrus flowering involves the interactions between various genes and signaling pathways. Understanding the molecular mechanisms that regulate citrus flowering is critical for the development of new citrus cultivars with improved flowering and fruit production traits, as well as for the optimization of citrus production practices to maximize yield and quality. Further research in this area, particularly in the use of biotechnology tools, is likely to lead to significant advances in citrus horticulture and production.

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Physiological Disorders in Citrus: Causes and Strategies to Overcome the Problems

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Abstract: Physiological disorder is any disturbance in the normal development of a plant or fruit, that is not associated with any pathological cause. The occurrence of physiological disorders is a common problem in fruit crops, which are associated with imbalances in nutrients and exposure to abiotic stresses. Such disorders can cause harm by interfering with the tree's physiology, leading to stunted growth, decreased yield, and poor fruit quality and storability. Despite being the second-largest fruit-producing nation globally, India is only ranked seventh in terms of productivity. The prevalence of physiological disorders is a significant factor contributing to this lower productivity. These disorders affect all stages of fruit cultivation, starting from the nursery to the storage and transit of harvested produce. In citrus, physiological disorders affect the quality and yield at pre-harvest as well as post-harvest stages. Diagnosis and management of these disorders can improve fruit quality as well as crop productivity. Classification of citrus fruit disorders can be based on their probable origin time, which comprises of pre-harvest nutrition-related disorders (such as exanthema and boron deficiency), weather-related disorders (like zebra skin and sunburn), maturity-related disorders (including sloughing and granulation), harvest-induced disorders (such as styler end breakdown), and post-harvest origin disorders (like chilling injury and aging). Having a thorough comprehension of these disorders and their management can aid in enhancing the production of quality citrus fruits.

Physiological disorders are common in fruit crops, which are caused by non-pathological reasons such as extreme in nutrients, abiotic stresses (light, temperature, moisture stress, air pollution etc.), resulting the physical damage through the alteration in tree physiology, and subsequently affecting the tree growth, yield and quality of fruit. India is the second largest fruit-producing country in the world (107.24 million MT) just next to china (253.89 million MT), however, in terms of productivity, it ranks on 7th position (15.12 tonnes/ha) among the top ten fruit producing countries throughout the world. This low fruit productivity is caused by a variety of factors. Of these, high occurrence of various physiological disorders in fruit crops is one of the most important reasons for the low productivity of fruit crops in India. These physiological disorders severely affect fruit

cultivation at all stages, right from the plants in the nursery to the fruits in storage or transit. Therefore, causes, proper diagnosis and management of these disorders can help to a great extent in improving the productivity of fruit crops.

Non-pathological disorders of citrus fruits can be grouped according to their most probable time of origin, such as nutrition-related disorder at the pre-harvest stage: exanthema (ammoniation), boron deficiency (arsenic toxicity), creasing, 'Pineapple' pitting etc. Weather-related disorder at pre-harvest stage includes zebra skin of tangerines, water spotting, sunburn, freeze injury, endoxerosis (wither tip) of lemons, wind scar, concentric rind stipple of grapefruit etc. Maturity related disorders at pre-harvest stage are sloughing of red grapefruit, rind staining of Navel oranges,

puffing of tangerines, rumple of lemons, granulation of oranges etc. Harvest induced disorders are oleocellosis, styler-end breakdown of limes, blossom-end clearing of grapefruit, stem-end rind breakdown of oranges etc. Postharvest origin disorders are chilling injury (peel pitting, oil gland, darkening, watery breakdown, scalding, brown staining, albedo browning), aging, freezing, red epidermal lesions etc. The major disorders seen frequently in citrus orchards have been discussed hereunder.

Fruit splitting

It is a frequent problem of lemon, orange and mandarin orchards, usually developed at the styler end and reaching or even extending beyond the equatorial zone. It is due to seasonal water deficit followed by heavy rainfall at cell enlargement and fruit development stage. Splitting is also developed because of disruption between peel and pulp growth.



Fig. 1: Fruit splitting in lemon



Fig. 2: Fruit splitting in grapefruit

Management

- Regular irrigation at 7-10 days interval during summer after fruit setting.
- Spray calcium nitrate (2%) and borax (0.8%) at the beginning of the cell enlargement stage significantly reduces the fruit splitting.
- Application of a mixture of GA₃ (20 mg l⁻¹) and 2,4-D (20 mg l⁻¹) after June drop significantly reduces the problem of fruit splitting.
- Use mulch and compost to maintain uniform moisture levels into the soil.
- Use slow release fertilizers, which help to facilitate the uniform uptake of nutrients.

Granulation

The juice sac of the affected fruit becomes tough, enlarged, grayish in colour, turn insipid and tasteless. TSS, acid and juice content decrease gradually with the increase of pectin, lignin and other polysaccharides. High temperature and relative humidity during spring, frequent irrigation, age and vigour of the plant, high nitrogen supply, use of vigorous rootstocks and delayed harvesting are highly associated factors with the occurrence of granulation disorder.

Management

- Less frequent irrigation, limited use of N-fertilizer and spraying of 2,4-D (16 ppm) is highly effective to control the malady.
- Spray of nutrients mixture containing Zn, Cu and K (0.25% each) at monthly interval during August to October
- Single spray of lime at 18-20 kg/450 litre water during the summer season *i.e.* March-May is highly effective, helps to reduce the occurrence of granulation.

- d) Avoid use of vigorous rootstocks like sour orange and rough lemon, and grow resistant varieties (Kinnow, Minneola, Orlando Tangelo and Duncan grapefruit)
- e) Use resistant rootstocks (Troyer citrange and Cleopatra mandarin).

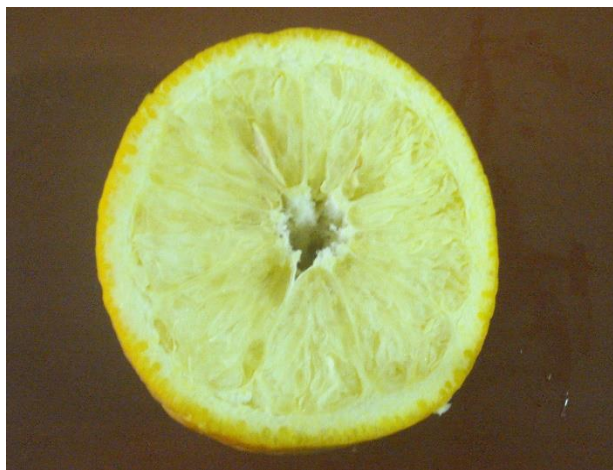


Fig. 3: Citrus fruit affected by granulation

Citrus exanthema

It is caused by a deficiency of copper (Cu). It causes development of dark green leaves of typical 'S' shaped twigs, commonly called ammoniation, as usually occur due to excessive nitrogen fertilization. This produces yellowish blotches at the leaf node due to phloem choking, preventing the sugar translocation from leaf to stem. The fruits exhibit brown gum-soaked eruptions with irregular blotches around central pith. Often there are brown or reddish brown gum excretions on the rind, sometimes with small to large splits in the rind. Dieback symptoms are observed on twigs.

Management

Spray of copper sulphate (0.5%) fortnightly interval or spray of Bordeaux mixture solution as basal + 2 foliar sprays of copper sulphate at 500 g/100 liter of water twice at 30 days interval before flowering can be beneficial for controlling this disorder.

Creasing

Creasing (albedo breakdown) is a physiological disorder that affects the albedo of citrus fruits. The physiological basis of creasing development is unknown, but several hypotheses have been suggested. Creasing is normally detectable at maturity or post-colour break stage, and inclines to rise with the approach of fruit maturity. Visual symptoms of creasing development are characterized by separations of cells at the middle lamella of the albedo tissue, resulting in fractures in the albedo and collapse of the flavedo, showing creases on the fruit surface. Creasing is associated with increased polygalacturonase activity resulting in a higher content of water-soluble pectin in the creased fruit. Imbalances in hormone levels (especially GA_3) are also associated with creased fruit. It is usually more prevalent on thin-skinned and fully mature fruit. Creased fruits generally exhibit poor keeping quality, because affected areas often split during handling, thus providing injuries that allow entry of decay pathogens.

Management

Foliar application of an aqueous solution of urea phosphate (0.5%) can be beneficial in reduction of creasing in citrus fruit.

Zebra skin

This disorder usually develops only on mandarins (easy peeling/zipper mandarin), and not very often on any of the other groups. 'Satsuma' mandarins (*Citrus unshiu*) are especially susceptible to the disorder. Zebra skin markings can appear a few days after packing, as well as during transport and marketing. Due to the rind being damaged during the packing process, the fruit has a higher water loss rate leading to a more wilted and soft rind. Fruit from older trees as well as weakly coloured fruit

(yellow fruit from the inside of the canopy) have more susceptibility, in addition to smaller fruit and those with a thin rind. The disorder can lead to increased decay as well as off-flavours in the fruit due to physical damage.

Management

The delayed harvesting until adverse weather conditions have passed (wind, high temperature, and low RH) as well as fruit retention on tree for adequate colour development. After harvest, it is essential to decrease the field heat of the fruits in order to decrease moisture loss from the rind. Thereafter, the fruits should be placed in degreening rooms for the least required period. Packing lines should be designed to prevent damage the mandarin rind by reducing line speed as well as the use of brushes in the pack line in order to avoid over-brushing

Oleocellosis

All types of citrus may develop oleocellosis, but lemons and Navel oranges are particularly highly sensitive. It is a physiological rind disorder of citrus fruit that is caused by the action of phytotoxic rind oils on the rind tissue. These oils are released from glands located in the rind following mechanical damage to the fruit. Oleocellosis can result from various types of damage, including insect attack, hail damage or wind rub, and can also be developed in undamaged fruit that comes into contact with damaged fruit. The oleocellosis damaged rind has a sunken and discoloured appearance, and immature fruit fails to develop colour normally, leaving a green/ yellow area. Upon physical injury, peel oil discharged from oil glands in the rind is phytotoxic, and causes necrosis and collapse of surrounding healthy epidermal cells. These cells remain green following degreening and later become darkened. The oil glands are most easily ruptured, if the fruit has a high turgor pressure, which occurs early in the

morning, when fruit water potential is highest or shortly after irrigation or rainfall.

Water spotting

Water spot occurs in mature citrus fruit during prolonged wet winter. Severity surge, when mature fruits remain on the tree longer than required. Symptoms are observed usually on the lower half of fruits, characterized by soft, water-soaked spots with no marked colour change. Prolonged fine weather after incidence renders these spots dry and papery, whereas prolonged wet condition leads to the development of common moulds, eventually leading to the rotting of fruits.

Management

It can be controlled by timely harvesting of fruits and keeping them in dry conditions.

Mesophyll collapse

Mesophyll collapse disorder of citrus of unknown cause. Leaves of oranges, lemons and grapefruit are worse affected. The disorder commences as translucent, irregular areas of different sizes scattered throughout the leaf blade. One or more light green spots may appear, later turning brown as cells within the tissue die. Only the soft tissue inside the leaf between the veins (mesophyll) collapses at first, giving these areas a translucent appearance. Palisade and epidermal tissues are unaffected initially, but may die later. Symptoms appear readily following periods of low soil moisture and hot dry winds. For this reason, water deficiency is often thought to cause mesophyll collapse, but mite feeding, damaged root systems and excessive transpiration also are associated with mesophyll collapse. Nutritional imbalances also are implicated in the mesophyll collapse of citrus. This disorder can be managed using budlings on RLC-4 or rough lemon rootstocks.

Sun scald

Sun scald affects all tree species, but young trees with smooth bark are usually most vulnerable. Sunscald is caused in young and soft vulnerable parts of the plants by weather and temperature changes, and occurs most often on the tree's southwest side, where it is exposed directly to the sunshine. The winter sun heats the bark, causing the cells to become active. When the sun sets, cool temperatures arrive and kill these cells, causing cracks or splits in the bark. A citrus tree with sunscald may experience fruit rotting, stunted growth, and any number of opportunistic diseases, while it is not fatal to a healthy grapefruit tree. The frequent sunscald can cause the tree to decline and eventually die.

Management

This problem can be prevented by wrapping the tree with a trunk guard during winter months. Preventing the young plant from high-intensity of heat is the only controlling method.

Freeze injury

Citrus freeze injury is initiated due to the formation of ice crystals in intercellular spaces. Freezing injury in transit is more likely to occur in the fruit next to the side walls and along the floor of the car or van, or near the air delivery, than in fruit in the body of the load. Freezing damage in oranges is best identified by cutting off both ends of the fruit, then cutting through the rind of the central part remaining, and pulling the segments apart. If the fruit has been frozen, the membrane between the segments looks soaked and usually contains a number of white specks, which are hesperidin crystals (naringin in grapefruit) resulting from the freezing. The hesperidin/naringin crystals occur in the pulp as well as on the segment walls, and are, therefore, apparent in cross section. If the freezing has been acute, the peel may exhibit

impacts ranging from a typical brown stain to gray discoloured areas of variable size. The affected rind tissues may or may not be sunken. When severely frozen, they usually become soft and mushy, and are underlain by mushy pulp.



Fig. 4: Freezing injury of citrus

Endoxerosis (wither tip)

Citrus endoxerosis another water-related physiological disorder which mainly affects lemons. It occurs mainly due to internal water stress, and external factors, which affect the water balance of the plant. Endoxerosis also called an internal decline, dry core, yellow tip, which often accompanies or follows it. Internal tissues back of the stylar end break down, dry and become pinkish or brownish in colour. Gum commonly forms in the core and either in or next to the rind. Green fruits lose lustre frequently but do not always, develop a yellow colour in circular areas surrounding the stylar end. The cut fruit shows the gummy pinkish to the brownish mass of partially dried and collapsed tissue. Gumming may even extend into the twig bearing the affected fruit. When the fruit turns colour, the malady is more difficult to detect without cutting. The cause is believed to be related to water and the physiological conditions within the tree and fruit, and temperature conditions in the air and soil influencing transpiration and water stress.

Management

It may be controlled by maintaining proper water requirements as needed by the plant.

Wind scar

Injury from wind occurs often to young fruit during the first 3 weeks after petal fall when leaves, twigs, or thorns rub against the rind. Scarring is less frequent after fruit is 12 weeks of age. Areas of the fruit surface affected by wind scar expand as the fruit enlarges during growth.

Management

It can be prevented by raising windbreaks, which breaks wind speed. Planting of trees should be done in the orientation of the wind. Keep soil moist, if the wind is predicted earlier.

Sloughing

Sloughing is a serious disorder of red grapefruit, which primarily occurs in Florida region, but may occur in other areas too. It was discovered by Grierson and Nehwall in 1955. It was named as 'sloughing' because it is confined to albedo and flevado, the necrotic tissue remains moist, and it is easily separated from the underlying healthy plant. Symptoms become visible in one week of harvested fruits. It is partially associated with high rainfall interval.

Management

Storing the fruits at low temperatures is effective to manage the sloughing.

Albedo browning

Albedo browning is the discolouration of the white, spongy inner tissues of the rind. The disorder may show externally as a minor, pebbly, brown-to-gray darkening of the rind, where the discoloured albedo shows through the surface layer. It tends to develop on fruit in

storage at low temperatures (0 °C) and with poor ventilation, especially fruit that was harvested at an immature or dark-green stage. This disorder affects only lemons.

Rumple

Rumple is common in lemons. The characteristic symptoms of rumple are the wrinkling of the fruit and chlorotic spots on the rind, in which, the oil glands stand out prominently. The spots later become brown, firm, and filled with gum, which penetrates deeply into the albedo. Directly underneath the spot, the albedo becomes snow white. Many of the fruit have gum in the albedo, most frequently under the calyx, sometimes in the columella, and less frequently in areas of the peel that have no external symptoms. The cause and control measures are unknown.

Blossom end clearing (BEC)

BEC of grapefruit is a postharvest physiological disorder that typically seems as a wet translucent area at the blossom end of the fruit. The disorder is also known as waterlog, water bottom, wet bottom, or wet wick, and continues to lead to citrus significant commercial losses as fruit appearance. This peel injury occurs primarily in thin-skinned, fully mature, seedless grapefruit in late season. The disorder develops when rupture of juice vesicles arises during handling at harvest and packing, which causes outflow of juice into the central fruit cavity. The juice causes a soaked area to develop, primarily at the fruit stylar-end, which is susceptible to fungal decay.

The formation of a wet area on the fruit's surface is the result of internal bruising and juice leakage out of the vesicles into the rind due to physical mishandling. Visual symptoms can develop 24 hours after damage to the fruit due to ruptured juice vesicles at the blossom end,

and the movement of released juice into the rind, especially the white albedo.

Management

In order to control this disorder, careful consideration should be given to all steps in the postharvest handling chain of grapefruit, from the picking action to transport and processing in the packinghouse. During packaging, any action taken that could result in dropping the fruit such as re-binning or a too high packing-line speed should be avoided in order to reduce the physical stress on the fruit. Furthermore, proper handling of fruits during packaging, and removal of field heat prior via drenching before packaging, and shock proof packaging help in reduction of BEC incidence.

Chilling injury

Chilling injury is the collapse of distinct areas of the grapefruit rind that develops after at least one month of storage at temperatures below 10–15.4 °C. Other symptoms may be displayed as a discoloured scald or water-soaked area of the rind. Grapefruit are most susceptible during the early and late months of the harvesting season. Low-temperature storage is the most widely applied technology to extend the postharvest life of citrus fruit. Moreover, the presence of certain pests such as fruit flies requires mandatory treatment at very low temperatures (below 2 °C for most citrus species except lemon) for exportation to certain markets. Because the *Citrus* spp. is of subtropical origin, it is a cold-sensitive crop, and the fruit of many of its species and varieties may develop damage and a disorder that is referred to as chilling injury (CI), when exposed to temperatures below 5–10 °C. However, fruit sensitivity to this disorder ranges widely among the different *Citrus* species and cultivars, exhibiting that genetic background is the first endogenous factor that determines susceptibility to cold stress.

Moreover, CI is greatly influenced by pre-harvest climatic factors, growing conditions, and postharvest management, which together modulate the timing and intensity of CI symptoms develop on the fruit. It is extensively accepted that symptoms of CI are a consequence of oxidative stress in the tissues occurring when reactive oxygen species (ROS) such as hydrogen peroxides, superoxides and hydroxyl radicals are in excess of the scavenging capacity.

Management

Postharvest heat treatments can be used to encourage fruit tolerance to cold temperatures, and to inhibit the development of CI symptoms during cold storage and cold quarantine. Hot water dipping at 41°C for 20 minutes can control chilling injury in Navel and Valencia late orange



Fig. 5: Chilling injury of grapefruit

Stem end rind breakdown (SERB)

This is post-harvest disorder roughly takes one-week time to develop. So, cause of the disease is the drying period, usually between harvesting and wax polishing, and over all due to water-loss. Oranges and Temples are highly sensitive to SERB. This disorder is caused by dehydration of the rind around the stem-end of the fruit that leads to darkening and collapse of the surface cells. Cells close to the button usually remain normal. SERB occurs more widely on small,

thin-skinned fruit when excessive moisture is lost, normally between harvesting and waxing during packing. SERB occurs in oranges after harvest, particularly if they are held for 2 to 3 days at low relative humidity before washing and waxing. A zone of flavedo tissue starting 3 to 5 mm from the button, and extending 10 to 20 mm toward the equator is susceptible. The area becomes sunken and eventually brown if SERB develops.

Management

Field treatments with anti-transpirant 1 to 2 months before harvest to reduce water stress can reduce the incidence of SERB in the packed fruit. It can also be controlled by narrowing the period between harvesting and waxing, keeping fruit in shade after harvesting, and maintaining low temperatures during storage.

Petaca

Petaca spot of lemons is a postharvest physiological disorder associated with the collapse of individual oil glands, usually between harvest and cold storage. The incidence of peteca has been recorded as early as 1924 in the USA in lemon fruit, shipped from Italy, from which it appears that this name was adopted from petecchia, as it was known in Italy. Initial symptoms can develop 3–5 days after harvest, and consist of the darkening of an individual oil gland as it collapses, leaking its content into the albedo tissue. After a few days, the rind tissue surrounding the collapsed oil gland sinks and adjacent oil glands can also collapse. This collapse of the oil glands occurs in green and yellow fruit, but the sunken lesion is clearly seen in yellow fruit. Regarding the mechanism of this highly problematic and costly disorder, no clear cause has been identified; however, several pre- and postharvest conditions have been recognized to influence the incidence of petaca. The main factor to take into account is that, in

contrast to oleocellosis, peteca develops without any mechanical damage to the rind tissue, and occurs due to physiological breakdown (senescence) of the oil gland. Cold and wet conditions (especially dramatic changes in day-night temperature) close to harvest have been linked with a higher incidence of peteca. Petaca has been previously associated with calcium (Ca) imbalances in the rind, but no direct evidence of Ca content or Ca-oxalate crystals being a causal factor of cell damage has subsequently been reported.

Management

Postharvest ambient conditions and handling can aggravate petaca in lemon fruit, and the exposure of fruit to 5% CO₂ increased the incidence of the disorder, whereas ethylene gas (3 ppm) reduced it. Currently, the most sustainable commercial action that can control the incidence of petaca includes delayed harvesting, allowing the fruit rind to mature.

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Management of Citrus Fruit Drop: A Complex Problem in Central India

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Abstract: Citrus fruit drop is a severe disorder that lowers fruit quality and yield. Fruit drop in citrus occurs at various stages viz., post setting drop, pea to marble size and pre-harvest drop causing huge loss to the farmers. A single component does not just cause fruit drop; it is also influenced by several physiological factors, such as high humidity, extreme heat, and water stress, as well as a number of pathological and entomological causes, such as the spread of several groups of pathogens and insect pests. Hence based on the causal factors, the fruit drop can be classified broadly as i) Physiological drop ii) Entomological drop and iii) Pathological drop. The physiological fruit drop is mostly associated with hormonal imbalance and poor nutritional management. In order to limit the problem of fruit drop in citrus, a number of different approaches should be taken in an integrated manner with attention to proper nutrition, irrigation, pest and disease management, pruning, weather conditions, and potential hormonal treatments.

Citrus is the third most important fruit crop in India. Nagpur mandarin and sweet orange are the two most popular citrus cultivars of Central India. Though widely cultivated, these crops face a major problem called fruit drop. For the past few years fruit drop has become a major challenge for citrus growers of central India. But flowering, fruit set, subsequent fruit drop and ultimate fruit retention are affected by several environmental and physiological factors. Most commercially important citrus cultivars bloom prolifically producing as many as 1,00,000-2,00,000 flowers on a mature tree. However fewer than 1-2% of these flowers produce harvestable fruit. The rest of the flowers and fruits are dropped by the plants due to several reasons. Fruit drop in citrus occurs more or less in four distinct waves:

- a. The first wave (post-setting drop) occurs soon after fruit set in January-February months due to natural over production and is not of much concern to the grower,
- b. A second wave occurs from March to April (summer drop). It is particularly severe in

the hot dry weather conditions with injudicious watering practices,

- c. Third wave occurs during July-September during rainy season owing either to intermittent dry spells or incessant rains conducive to pathogenic infections leading to heavy economic losses,
- d. The fourth wave, called the pre-harvest fruit drop is the drop of mature fruits before the harvest attributable to both abiotic and biotic factors. This drop is again of economic loss to growers as in this stage almost completely matured fruits are shed.

However, based on the causal factors involved, fruit drop can be classified broadly as: physiological, pathological and entomological.

Physiological fruit drop

Physiological fruit drop refers to the abscission of fruitlets as they approach 0.5-2.0 cm in diameter. Physiological drop is a disorder most probably related to competition among fruitlets for carbohydrates, water, hormones and other

metabolites. Fruit drop is regulated by the balance of two endogenous plant hormones, auxin and ethylene. When the ratio of ethylene to auxin is higher, it induces the enzymes which dissolve cell wall components in the abscission zone between the fruit and stem (peduncle) at the button, which separates the fruit from the tree. Ethylene is produced in response to stress factors such as water stress, physical injuries, frost damage, and decay of the fruit. When the fruit is injured, ethylene gas production is triggered, which may cause fruit to drop. The problem, however, is greatly aggravated by stress factors especially high temperatures or water deficit.



Fig. 1: Physiological fruit drop

Consequently, physiological drop (Fig. 1) is usually most severe where leaf temperatures may reach 35-40°C and where water stress is a problem in several parts of central India.

Management

- Application of 50g FeSO₄ and 50g ZnSO₄/plant mixed with 5 kg FYM or vermicompost along with recommended

dose of fertilizers in the basin of these trees in the month of July.

- Foliar spray of GA₃ 1.5 g + KNO₃ 1.5 kg in 100 litres of water may be done in first week of July.
- Foliar spray of 2,4-D or NAA 1.5 g + monopotassium phosphate (00:52:34) 1.5 kg may be done in third week of July.
- If there is long dry spell of more than a week with maximum temperature of >35°C, foliar application of Kaolin 40 g/lit. water may be done in the months of April-May.

Pathological fruit drop

Pathological fruit drop generally starts during/after rainy season (July-August) and continues till harvest. Sometimes a bacterial infection (fruitlet blight) also causes the drop of small fruitlets that set immediately after *Ambia* bloom. In continuously wet weather conditions for about 24 hrs. or more, *Phytophthora* sp. causes a typical brown rot of fruits (Fig. 2) triggering severe fruit drop, especially in Mosambi sweet orange. Several stem end rot (SER) fungal pathogens viz. *Lasiodiplodia theobromae*, (Fig. 3) *Colletotrichum gloeosporioides* (Fig. 4) and in some cases *Alternaria* sp. cause the drop of mature fruits prior to harvest (pre-harvest drop).



Fig. 2: Fruit Drop due to brown rot infection



Fig. 3: *Lasiodiplodia* stem end rot



Fig. 4: *Colletotrichum* stem end rot

In recent times association of citrus greening bacterium has also been detected with pre-harvest fruit drop.

Management

- Foliar spray of Copper oxychloride 50 WP @ 2.5g / litre water for controlling fruitlet blight infection of *Ambia* crops during March-April.
- Foliar spraying of fosetyl AI (0.25%) (twice during August and September at 40 days interval) should be undertaken for brown rot of fruits. Prophylactic spraying of 1% Bordeaux mixture or 0.3% copper oxychloride helps in preventing the infection.
- The pathological fruit drop (due to *Collectotrichum* and *Diplodia*) affected

trees should be sprayed (twice) with carbendazim or thiophanate methyl at the concentration of 0.1% formulation basis (i.e. 1 g/l). The second spray should be repeated after 10 days of first spraying. Spraying of (azoxystrobin + difenoconazole) combination fungicide @ 0.5 - 1 ml/lit. can also be taken up as an alternative.

In case of greening-infected trees (showing typical lower end greening fruit symptoms or confirmed positive for infection), integrated application of Tetracycline hydrochloride 600 ppm (6g / 10 litres water) + ZnSO₄ + FeSO₄ (200 g each). Tetracycline hydrochloride should be applied as foliar spray twice at 45 days interval. ZnSO₄ & FeSO₄ should be applied in tree basins.

Removal of dry and dead twigs/ shoots and destruction by burning after harvest, followed by two fungicidal sprays (Copper oxychloride @ 3 g/liter water or carbendazim / thiophanate methyl @ 1 g/ liter water). The fallen fruits must be removed from the orchard as they act as carrier for several diseases.

Entomological fruit drop

Entomological fruit drop in Nagpur mandarin and sweet orange in central India occurs mainly due to two insect pests namely; fruit flies (*Bactrocera* sp.) and fruit sucking moth (*Eudocima* sp.). The incidence of fruit sucking moth (Fig. 5) is seen in August – November on colour breaking Nagpur mandarin and sweet orange fruits of *Ambia* season. Moths do not breed on citrus plants but spend their early life on wild shrubs and only adult moths get attracted towards maturing fruits. They suck juice from fruits at night and rotting follows due to secondary infections through skin puncture (Fig. 6) leading to fruit drop. Such fruit drop is heavier during September-October in central India. Fruit flies affect both *Ambia* and

Mrig season fruits during colour breaking stage but peak activity period of fruit flies is from August to November.



Fig. 5: Adult fruit sucking moth (FSM)



Fig. 6: FSM infested fruits

The adult fly (Fig. 7) punctures the ripening fruit and lays eggs inside the rind. On hatching, the maggots bore the ripening fruit and feed on soft pulp and makes the fruit unfit for consumption. The infested fruits show depressions with dark greenish punctures, get deformed and due to bacterial and fungal activity, fruits rot and fall down.

Management

- For management of fruit flies, install methyl eugenol-based traps @ 15-20 traps per hectare for mass trapping of fruit flies. The

lures should be changed at every 15-20 days interval for effective control.

- Collection of fallen fruits and their destruction at regular intervals would prevent the development of puparia and thus reduce the fruit fly population in next subsequent generations. Fallen fruits also attract fruit sucking moths.
- Systematic destruction of larval host plants (Gulvel, Vasanvel etc) of fruit sucking moths during rainy season in the vicinity and surrounding the orchards in a mass campaign mode reduces the pest population.
- Generation of smoke in the late evening hours in orchards repels the fruit sucking moth adults.
- Poison baiting with 10 ml malathion + 100 ml mandarin Juice + 100 g Jaggary + 900 ml water in wide mouth plastic container helps to attract and kill the adults of fruit sucking moths.
- Foliar application of Neem oil 1% or Petroleum spray oil (Horticulture Mineral Oil) @ 2% at 10-15 days interval during colour break stage to till the harvest effectively control the fruit sucking moth.



Fig. 7: Adult fruit fly

Conclusion

Citrus naturally adjusts the amount of fruit carried on the tree, so some fruit drop is normal. Excessive drop of small and mature fruits may have several causes: lack of water or fertilizer, heavy pruning, sudden changes in temperature (especially sudden hot temperatures at fruit set or shortly after), or insect/ disease infestation. It is not always possible to avoid excessive fruit drop, but it can be kept to a minimum by giving the trees the best possible care. It is especially important to irrigate carefully, avoid excessive pruning and keep disease and insect pests under control. Hence orchard management should be focused on reducing any type of stress on the tree while managing irrigation and nutrition. Finally, it is expected any successful treatment that improves tree health will lead to more leaf retention and better root growth as well as reduced fruit drop.

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An Approach for Conservation of Citrus Germplasm through Cryopreservation for Long Term Storage

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Abstract: Researchers, breeders, and industry participants have access to a variety of genetic resources and these are typically maintained in field, Field Gene Bank (FGB) or screen house rendering them vulnerable to biotic and environmental dangers. Certain cultivars can only be grown vegetatively since they lack seeds. In FGB, where they are still susceptible to pests, diseases, and other natural dangers including drought, weather damage, human mistake, and vandalism, Citrus genetic resources are currently stored as complete plants (Withers and Engels 1990). Cryopreservation is now the only option for the long-term preservation of problematic crops. Seeds (Mumford and Grout 1979), ovules (Bajaj 1984), embryonic axes (Radhamani and Chandel 1992), somatic embryos (Marin and Duran-Vila 1988), embryogenic calluses, and cell suspensions have all been used for the cryopreservation of Citrus germplasm (Sakai et al. 1990, 1991; Niino and Sakai 1992; Engelmann et al. 1994). An urgent need to implement complementary conservation strategies to protect these species and guarantee their availability for use in the future has resulted from the alarming rate of forest cover loss and lack of cultivation of some Citrus species. The creation of techniques for their multiplication, regeneration, and *in-situ* and *ex-situ* propagation must be given top priority. Since Citrus germplasm conservation is important, a global network for Citrus germplasm was established in 1997 and in 2023, a Cryo preservation unit has been established at AAU- Citrus & Plantation Crops Research Station, Tinsukia for long term conservation of Citrus germplasm of North East Region.

Introduction

One of the world's most important fruit crops, citrus is produced in large quantities throughout various subtropical areas (FAO, 2013). Southeast Asia, which includes southern China, north-eastern India, Burma, Malaysia, and surrounding regions, is where Citrus originated. There are further closely similar species in Australia and Papua New Guinea. Citrus is primarily an understory tree found in subtropical forests, a biome whose area has already been significantly decreased by logging and agricultural development and is now further endangered by climate change. Citrus has a narrow genetic base since the major commercial cultivars, which make up a sizable portion of Citrus production in many nations, are derived from a small number of genotypes. For instance,

sweet oranges are the most significant commercial variety, and virtually all sweet orange cultivars have evolved through the selection of mutations from a single complicated hybrid progenitor. Many other commercial groups such as clementine and grapefruit are now known to be F1 hybrids of sweet orange (Moore, 2001; Wu *et al.*, 2014).

Many tribes from various communities are employing various Citrus germplasm for treating human ailments in the north-eastern region of India, which is home to a wealth of distinct Citrus species. Many species, landraces, and possible hybrids of Citrus can be grown carelessly in the area thanks to its edaphic and climatic conditions as well as its physiographic state. Yet, as the population grows and farmers' attitudes towards some recently introduced

remunerative cash crops, the genetic resources of these important resources are being eroded, and the majority of them are on the edge of extinction. Therefore, it is imperative that the scientific community utilise all of the unique and imperilled Citrus resources of the north-eastern region while also conserving them.

The fine structure of cells is preserved through the process of cryopreservation, which keeps biological samples frozen at cryogenic temperatures for any significant amount of time. It began in 1948, following the unintentional discovery by C. Polge *et al.* that fowl spermatozoa could be successfully frozen to 70°C using glycerol. It is a technique for keeping cells alive at extremely low temperatures, such liquid nitrogen. In 2023, a North-East Cryo Gene Bank unit have been established at the AAU-Citrus & Plantation Crops Research Station, Tinsukia.

Significance of conservation

Seeds of several Citrus species exhibit refractory or intermediate storage behaviour, making dry storage at low temperatures impossible. The created seeds are heterozygous as well, making it unable to preserve certain gene combinations through seed storage. Certain cultivars can only be grown vegetatively since they lack seeds. In field gene banks, where they are still susceptible to pests, diseases, and other natural dangers including drought, weather damage, human mistake, and vandalism, Citrus genetic resources are currently conserved as complete plants (Withers and Engels 1990). Cryopreservation is now the only option for the long-term preservation of problematic crops. Seeds (Mumford and Grout 1979), ovules (Bajaj, 1984), embryonic axes (Radhamani and Chandel, 1992), somatic embryos (Marin and Duran-Vila 1988), embryogenic calluses, and cell suspensions

have all been used for the cryopreservation of Citrus germplasm (Sakai et al., 1990, 1991; Niino and Sakai, 1992; Engelmann et al., 1994).

Conservation

Citrus genetic resources are conserved utilising a variety of methods at the genotype, gene pool, species, and eco-system levels. The vast genetic diversity of Citrus fruits includes a variety of genetic stocks, wild and semi-wild species' germplasm, and farmer's selection, among other things. It is therefore stressed that the optimum choice for accomplishing safe conservation of endangered Citrus species under severe threat of extinction would be a complementary conservation strategy comprising the deployment of more than one applicable approach. Indian culture places a strong emphasis on the preservation and sustainable use of biological resources based on regional knowledge systems and customs.

1. In-Situ Conservation: An integrated strategy to ensure the conservation of plant germplasm must include both in-situ conservation and on-farm conservation of crop genetic diversity. These places are now endangered as a result of several development initiatives and the changing climate, and it is necessary to preserve and collect the valuable plant diversity that already exists for secure in-situ preservation.

2. On-farm conservation: The preservation of plant diversity on farms through farmer participation has been ardently promoted. Such a strategy is very advantageous to small and marginal farmers living in inhospitable and fragile terrain that contain a significant genetic variety of different Citrus fruits.

3. Ex-situ conservation/Field gene banks: Wild Citrus fruit ex situ conservation is crucial to preserve the genetic diversity already present and to enable access to germplasm for genetic

improvement to create desirable cultivars or types. Field gene banks play a significant role in the preservation of clonally propagated species. There are already numerous field gene banks operating all over the world for various horticultural plant species. Citrus field gene banks are being kept up to date in India by the State Agricultural Universities (SAUs), the State Horticultural Research Stations, and the ICAR Horticultural Institutes.

4. Long term Cryobanking (Cryo Gene Bank):

Cryopreservation of seeds, embryos, and embryonic axes is the sole method that can be used to store resistant seeds for an extended period of time. Under the direction of the National Bureau of Plant Genetic Resources, New Delhi, the Indian Institute for Horticultural Research is preserving priceless germplasm using cutting-edge methods incorporating pollen, seed, and asexual propagules of numerous fruit plants.



Fig. 1: Preservation in liquid nitrogen



Fig. 2: Cryo Gene Bank at AAU-CPCRS, Tinsukia

Conclusion

The current state of Citrus genetic variety is concerning because massive habitat degradation is occurring in order to meet human needs in numerous ways. To maintain a parallel repository of *ex-situ* germplasm in remote regions, specialised drives must be created. If diversity goes extinct, the species will still be available for use in the future. Moreover, every effort should be taken to maintain the integrity of the on-site germplasm that already exists. *Citrus indica* is one of the rare and wild species whose accessions failed to survive in the field gene bank. In the event that any of the two procedures fails, this will safeguard one alternative to the other. Such an effort should focus more on locations for collecting where seemingly primitive types might still exist in pristine condition. The forest and the isolated, inaccessible environment have a lot of promise for producing primitive and wild Citrus varieties, and they require our immediate attention. It is possible to conduct a thorough assessment and compile a rare collection of Citrus accessions in the special region where a select few can withstand challenging environmental circumstances (biotic and abiotic).

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A Glance at Citriculture in Punjab

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Abstract: Citrus journey in Punjab started off with cultivation of the sweet oranges, vernacularly pronounced as *malta* in the state. The entry of high performing ‘Kinnow’ [a hybrid evolved from the cross of King and Willow leaf mandarins] changed the cultivation scenario in Punjab. Now most of the citrus area is under Kinnow mandarin with small acreage under sweet oranges, limes and lemons. Punjab has a subtropical climate and the winter provides the necessary resting period that induces flowering during February-March. Growing citrus effectively requires a soil pH between 5.5 and 7.5; however, with the help of Jatti khatti rootstock and canal water for irrigation, citrus can be cultivated in soils with a pH as high as 8.5. A complete package for cultivation of citrus fruits has been developed by the Punjab Agricultural University. Young plants need regular pruning for a better tree-framework while light annual pruning is essential for regular fruiting. Insect and mite pests *viz.*, psylla, leaf miner, thrips, mites and fruit flies and diseases *viz.*, foot rot/gummosis, fruit drop, canker and greening cause substantial damage to tree health and productivity. Integrated management options in the form of pesticides, growth regulators or biologicals are available to tackle them. Use of the elite plants coupled with adoption of modern technologies like drip irrigation can ward off the onset of various diseases. Punjab state has several public sector nurseries to meet the plants’ demand. Technological and chemical interventions are needed to address problems such as waterlogging, soil salinity, and temperature variations especially during blooming. Punjab farmers sell their produce to middlemen who market it through roadside stands and larger supermarkets across the country. The export and long-distance promotion of citrus fruits are also facilitated by organizations like APEDA and PAIC.

Citriculture refers to the cultivation of different citrus fruits namely mandarins, sweet oranges, grapefruits, lime and lemons. The journey of citrus cultivation in Punjab started off with promotion of sweet orange plantation (popularly known as Malta) under a ‘Garden Colonies Scheme’ meant for rehabilitation of migrants (after partition) in Indian Punjab. In this scheme, to promote citrus cultivation, the growers were allotted land, supplementary canal water and Malta saplings at a subsidized rate. The scheme proved to be a crop diversification incentive for the state. But the citrus cultivation got a real boost with the entry of Kinnow mandarin, a hybrid of King and Willow leaf mandarins introduced from the USA in 1959. Now, almost all the citrus fruits are grown in state in varying proportions. Due to the well adaptation of citrus to the Punjab agro-climatic conditions, it

constitutes the numero uno fruit industry of the state. Fazilka, Shri Muktsar Sahib, Hoshiarpur and Bathinda are the leading citrus growing districts of the state. Fazilka alone holds 70% of the total area under citrus cultivation.

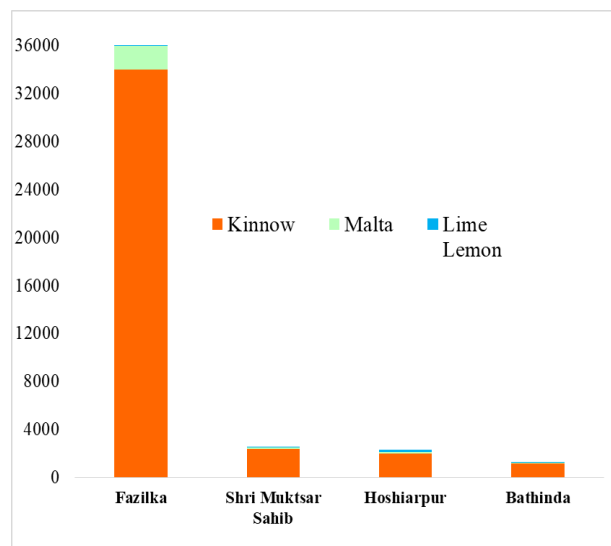


Fig. 1: Major citrus growing districts in Punjab

Punjab has a subtropical climate. This subtropical climate induces one main crop of citrus in Punjab and the flowering of which occurs in February-March. Due to poor quality sub-surface water, canal water is the main source of irrigation.

Varietal landscape of citrus in Punjab

The citriculture in Punjab started with cultivation of sweet oranges, which were soon replaced by the Kinnow mandarin. The Kinnow

mandarin, due to its high yield potential has revolutionized the fruit industry of Punjab. The area under Kinnow mandarin in 1970's was 500 ha, which has expanded to 44,752 ha in 2020-21. The development of low seeded Kinnow mandarin has further increased the scope for future area expansion under mandarins. A small area is also occupied by sweet oranges (2,788 ha) and limes & lemons (2,655 ha). Of the total citrus production (1.22 million MT), Kinnow accounts for 1.18 million MT (96.3% of total).

Table 1: Varietal landscape of citrus in Punjab

Particulars	Cultivated citrus groups			
	Mandarins	Sweet oranges	Lime and Lemons	Grapefruit
Features and use	Easy to peel and used for table purpose	Hard to peel and used for juice purpose	Highly acidic and used for juice and shikanjavi	Highly rich in antioxidants and has medicinal value
Important varieties	Kinnow, PAU Kinnow-1, W-Murcott and Daisy	Early Gold, Mosambi, Jaffa, Blood Red and Valencia Late	Kagzi Nimboo, PAU Baramasi Lemon-1, Sweet lime Local (<i>Mitha</i>)	Marsh Seedless, Red Blush and Star Ruby
Rootstock	Jatti khatti and Carrizo citrange	Jatti khatti	Jatti khatti	Jatti khatti
Scale of cultivation	Commercial	Commercial	Commercial	Home scale

Soil suitability: The ideal pH range for citrus cultivation is 5.5-7.5, but with the use of Jatti khatti (Rough lemon) rootstock, and canal water as source of irrigation, citrus can be successfully grown even in soils having pH up to 8.5. The other permissible soil limits for successful citrus cultivation include-electrical conductivity of 0.5 mmhos/cm, calcium carbonate concentration of up to 5 per cent and lime concentration up to 10 per cent.

Propagation and planting: After evaluation of different rootstocks, Jatti khatti, a strain of rough lemon (*Citrus jambhiri* Lush.) has been found to be the most suited rootstock for propagation (budding) of different citrus varieties in Punjab. In sub-mountainous regions of Punjab (Hoshiarpur district) soil pH remains

in optimum range, hence, the plants propagated on Carrizo citrange are preferred. There are few public sector nurseries which supply quality planting material to the growers (Table 2).

Table 2: Public sector citrus nurseries in Punjab

1. Department of Fruit Science, Punjab Agricultural University (PAU), Ludhiana
2. PAU-JC Bakhshi Regional Research Station, Abohar, Fazilka
3. PAU-Fruit Research station, Jhallowal
4. PAU-Regional Research Station, Bathinda
5. Centre of Excellence for Fruits, Khanaura, Hoshiarpur

The citrus plants are planted at a spacing of 6 m X 6 m. However, for higher returns per unit area,

a dense spacing of 6 m (between rows) X 3 m (between plant to plant) is also recommended for Kinnow mandarin for areas where tree life span is short due to high water table or high salinity. After 15 years, when the trees start mixing with each other, uprooting of alternate trees brings the planting to the normal (6 m X 6 m) spacing. The plants can be planted during Feb-March and August-September.



Fig. 2: A View of citrus nursery at JC Bakhshi Regional Research Station, Abohar



Fig. 3: High density planting (6 m X 3 m) of Kinnow mandarin



Fig. 4: Kinnow mandarin plantation at normal spacing (6 m X 6 m)

Regular training and removal of angular shoots (water shoots) and rootstock sprouts (suckers) is essential for maintenance of plants during initial years of plantation. During fruiting phase, a light annual pruning is required for regular fruiting.

During the initial 3-4 years, when the citrus trees are not in fruiting, the soil health can be improved by growing leguminous crops like guar, moong, cowpea, daincha, pea, gram etc. Tall and exhaustive crops like cotton, jowar, bajra or water guzzling crops like berseem should not be used as intercrops. The irrigation of the citrus trees should be given depending on the tree age, soil type and weather conditions. The installation of drip irrigation with drippers of capacity @2-10 litres/hr ensures supply of water as per tree requirement and reduces excessive water loss and also checks the spread of *Phytophthora* disease.

Nutrition: A Kinnow plant aged 8 years or above requires to be supplied with 1920 g urea and 2750 g super phosphate. In central and sub-montaneous districts of Punjab, an additional 1465 g muriate of potash is also given to the bearing trees. Half of the nitrogen dose along with full phosphorous and potash dose are applied in February-March (before flowering) and the remaining half nitrogen in April (after fruit set). Application of 100 Kg/plant of farmyard manure (FYM) improve soil physical conditions and improve the availability of synthetic fertilizers. In addition, three sprays with KNO₃ (1.0%) during end of May, June and July improves fruit size and yield in Kinnow mandarin. Citrus plants grown on soils having high pH exhibit micronutrients (Zn and Mn) deficiencies, which can be corrected by sprays of 4.7 g zinc sulphate + 3.3 g manganese sulphate per litre of water in end April and mid-August.

Fruit drop and its management: The fruiting citrus trees face high summer fruit drop in May-June (drop of small fruits) and again in September-October (drop of about to mature fruits). Proper pruning and burning of dead and diseased wood during winters reduces the intensity of fruit drop in next year crop. Spraying the trees with a combination of fungicides and growth regulators like 1250 ml of Ziram 27 SC or 500 ml of Propiconazole 25 EC or 500g of Carbendazim 50 WP in combination with 5 g 2,4-D (sodium salt of horticultural grade) in 500 litres of water during mid of April, August and September controls both kinds of fruit drop. Two additional sprays of any of the above fungicides during July end and September end is beneficial for checking pathogenic fruit drop. In case cotton or any other broad-leaved crops is growing in the vicinity of the orchard, GA3 (Gibberellic acid) at 5 g / 500 litres of water is sprayed instead of 2,4-D.

Harvesting: Kinnow and other citrus fruits ripe on tree itself. They must be harvested at fully ripe stage to get the best fruit quality (Table 3). The best period for harvesting Kinnow fruit is from mid-January to mid-February when it

attains TSS/acidity ratio of 12:1 to 14:1. Harvesting is done by clippers by retaining green button along with the shortest possible stalk and pick the fruits in bags. The long stalk with fruits may cause mechanical injuries to the adjacent fruits during handling and transport. Harvesting of Daisy, and sweet orange, and grapefruit varieties is done according to the month of their ripening. The delay in the harvesting of the sweet oranges and Daisy leads to drying of juicy vesicles, hence has a short harvesting window.



Fig. 5: PAU Kinnow -1 (less seeded Kinnow)

Table 3. Time of maturity of different citrus varieties

Citrus group	Variety	Time of maturity
Mandarin	Daisy	1 st fortnight of November
	PAU-Kinnow 1, W-Murcott	January
	Kinnow	Mid-January to mid February
Sweet orange	Early Gold	End of October to Mid November
	Mosambi	November
	Jaffa	December
	Blood Red	December-January
	Valencia Late	February-March
Lime and Lemons	PAU Baramasi Lemon-1	July
	Kagzi Nimboo	Summer months
	Sweet lime Local	September
Grapefruit	Star Ruby, Red Blush	November
	Marsh Seedless	December

Marketing: Punjab growers generally sell over their ready to harvest crop to the contractors for further marketing. The contractors sell the fruit through roadside fruit stalls and in local and distant markets within country. The progressive growers/ contractors who sell their produce after grading and packing have created their own market in different parts of the country. Many agencies like APEDA and Punjab Agro Industrial Corporation (PAIC) also facilitate distant marketing and even export of Kinnow and other citrus fruits.

Insect pests and diseases: Large number of insect-pests like citrus psylla, citrus leaf miner, lemon butterfly, thrips, mites and fruit flies and diseases like foot rot/ gummosis, canker, fruit drop, sooty mould, greening and citrus ring spot viruses cause substantial damage to tree health and productivity. Among these, the incidence of viruses and virus like diseases can be managed by use of healthy, disease-free planting material. The citrus psylla is managed with sprays of 0.4 ml imidacloprid or 0.33 g thiamethoxam or 12.5 ml of MaK Horticultural mineral oil (HMO)/ litre of water at the start of new growth (during March and September). The foot rot/ gummosis caused by *Phytophthora* is the major disease which induces tree death. Two applications of Ridomil Gold/ Curzate M8 as paint (2 g/100 ml of linseed oil) to the infected trunk portion and as soil drench (25 g/10 litres of water/ tree) at

the base of the tree in February-March and again in July-August helps in managing the foot rot and gummosis. Soil drenching is followed by sprays of Alliette @ 2.5 ml/ litre during April and September months.

There are also a few upcoming challenges like water logging, soil salinity and sudden increase of temperature during flowering and fruit set. These need to be addressed with technological and chemical interventions.

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Soil Sampling for Isolation of Entomopathogenic Nematodes from Citrus Orchards and their Identification

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Abstract: Entomopathogenic nematodes are microscopic organisms that are capable of infecting and killing insect pests, making them a valuable tool in the control of pests in citrus orchards. The isolation and identification of these nematodes is crucial for the development of biological control programs in citrus orchards. EPNs are effective biological control agents against various insect pests and are commonly found in the soil of citrus orchards. The isolation and identification of EPNs from soil samples can aid in the selection of the most effective biocontrol agents and the development of integrated pest management strategies. The identification of EPNs from soil samples involves morphological, molecular, and biochemical techniques. Morphological techniques involve the observation of characteristic features of EPNs, such as body shape and size, head and tail morphology, and the presence or absence of specific structures. Molecular techniques, such as polymerase chain reaction (PCR) and sequencing, rely on the analysis of specific DNA regions to distinguish between different EPN species and strains. Biochemical techniques, such as enzyme assays, can also aid in the identification of EPNs. A combination of different techniques is recommended for accurate identification. Future integrated pest management programmes in citrus orchards that aim to control of insect pests will need to find and identify EPN isolates that are adapted to the local environmental and climatic conditions.

Introduction

Entomopathogenic nematodes (EPNs) are parasites of several insects that are obligatory soil dwellers. They can be found anywhere, in both cultivated and uncultivated soils. EPNs are widely dispersed and have been observed on all continents of the world with the exception of Antarctica (Campos-Herrera *et al.*, 2012; Bhat *et al.*, 2020). EPNs belonging to the genera *Steinernema* and *Heterorhabditis* have a symbiotic relationship with enterobacteria *Xenorhabdus* and *Photorhabdus* respectively (Stuart *et al.*, 2006). These bacteria are capable of killing host within 24-48 hours after they have been released into the insect's gut. Through this life cycle, EPNs have thus far being recognized as the best candidates for biological control of problematic insects (Adams, 2009, Aryal *et al.*, 2022). EPNs are considered insect pathogens rather than insect

parasites because symbiotic bacteria that exist within the gut of these nematodes are released when the nematode enters an insect host and are ultimately responsible for the death of the insect.

Since their discovery in 1927, EPN have been considered valuable alternatives to chemical pesticides as they can parasitize a wide range of insects that are agricultural pests (Gaugler, 2002, Gaugler & Kaya, 1990, Adams *et al.*, 2006).

Most nematodes are amphimictic such as the *Steinernema* genera while *Heterorhabditis* nematodes are hermaphrodites or parthenogenetic (requiring females only) (Griffin *et al.*, 1994). Most of these identified EPNs live for several weeks to months in the infective stage so they can persist for a long time in the soil. Their life cycle also contribute to why they are cheap and easy to culture, sub-

culture and maintain hence they are considered the best biological control organisms which can be used to replace or substitute for harmful synthetic chemical pesticides (O’Leary *et al.*, 2001).

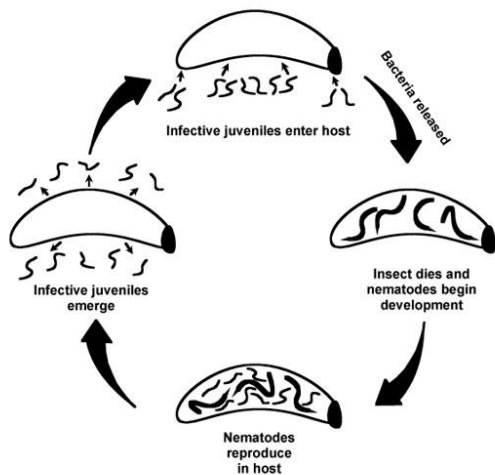


Fig. 1: EPNs life cycle (Source: Adams et al, 2009)

EPN’s Life cycle

Infective juveniles (IJs) enter the insect and release enterobacteria into the hemocoel of the host which then kill the insect. New generation of EPNs feed on the insect cadaver and proliferate together with the symbiotic bacteria. IJs are then released into the soil and they migrate in search for new live host (Fig. 1) (Adams, 2009). The objective of this article is to discuss the methods and techniques most commonly used for soil sampling and isolation of EPN from soil and their identification.

Sampling

Depending on the purpose of the study, two sampling strategies can be considered: Stratified sampling, b) Random sampling.

The stratified sampling is usually used as part of an intensive study in a specific or demarked area over a given period of time. In general, a transect is demarked and soil samples are taken

at stipulated intervals in the transect over a period of time (Stuart & Gaugler, 1994).

The random sampling is generally employed when focusing on an extensive area. This sampling strategy has been used for studying the diversity of EPN from a large geographic area or region (Stock & Gress 2006). A number of factors can be considered depending on the focus of the study including a diverse range of elevations, soil textures and habitats (e.g., cultivated fields, forests, pastures, *etc.*).

Baiting

The insect-baiting technique has been described, which was originally described by Bedding and Akhurst, 1975 and represents a simple and selective alternative for recovering EPN from soil samples. This is a selective procedure that is based on the premise that the nematodes (IJ stage) will be attracted to an insect host and parasitize it.

Modified White trap technique is also explained which is used for retrieving nematode progeny from infected insect hosts (Kaya & Stock, 1997). This is an easy method that offers the advantage of retrieving a ‘clean’ nematode progeny free of debris from the degrading insect cadavers.

These methods and techniques are key steps for the successful establishment of EPN cultures in the laboratory and also form the basis for other bioassays that consider these nematodes as model organisms for research in other biological disciplines.

How to Detect Entomopathogenic Nematodes

The use of a living organism to survey for a biological agent is termed “bioassay.” Waxworms are caterpillars of the greater wax moth (*Galleria mellonella*) and are often used to detect the presence of soil pathogens at a given

site. Waxworms in their natural state are parasites of honey bee hives, feeding on the wax and normally never coming in contact with the soil. Thus, wax worms have no natural immunity to soil pathogens and will most likely become infected when they come in contact with EPN. Because symptoms of EPN infection include a color change, waxworm caterpillars are ideal organisms for bioassays.

EPNs occur in soils worldwide and it is very easy to survey any area for their presence. Rather than examining soil samples for IJ's, which are very tiny, the soil is baited with a living organism that will attract IJ's from the soil and then exhibit symptoms indicating infection.

A. Soil collection

1. Soil should be collected from the site of interest.
2. Collect soil when it is damp – not saturated and not completely dry.
3. Using a trowel, collect soil sample from the top 3 to 5 inches of soil; take several trowels full of soil from the surface to fill an approximately 500 g sample.
4. Each 500g soil sample should be placed into individual zipper-type bags. Close the bag and knead gently to break up the soil.
5. The bag should be labeled with the date and location sampled as well as a description of the sample area.
6. EPN tend to be patchy in their distribution, so soil samples should be collected from areas that are subject to different applications (e.g., different types of plants, different tillage types, different soil types, etc.). Keep in mind that sampling designs may vary, depending on the type and size of habitat you are sampling (e.g., a diverse orchard will be different than a crop field).

Keep samples in a cooler (8 - 15 °C) during transport to the laboratory.

7. Take a portion of the soil sample for analysis to obtain information on soil composition, texture, moisture, electrical conductivity or other desired soil parameters.

B. Nematode Isolation from Soil Samples: Insect-baiting Technique

1. Remove any debris (*i.e.*, rocks, pieces of wood or bark, leaves, *etc.*) collected with your samples to avoid contamination with saprobic microorganisms.
2. Add water to moisten the soil and facilitate the movement of nematodes.
3. Place approximately 200 to 250 cc of moist soil in a clean plastic container with a lid.
4. Add insect baits. Consider 5 to 10 insects to the soil surface of each sample.
5. Cover the container with a lid and turn containers upside down.
6. Maintain containers in the dark and at room temperature (usually 22-25 °C).
7. Check containers every 2-3 days and remove dead insects. Add additional healthy insects to each container for further baiting of the soil sample.
8. Remove dead insects from the containers. Cadavers with a brown or ochre coloration are usually parasitized by Steinernematids, whereas brick red to dark purple cadavers are parasitized by Heterorhabditids.
9. Rinse cadavers in sterile water.
10. Place cadavers in a modified White trap for recovery of nematode progeny.

C. Culturing and Bioassay for Native EPNs

1. Place infected cadavers on a moist piece of filter paper in a covered dish.
2. Place the dish inside a plastic container and seal it most of the way to hold in moisture,

- but not so much that air cannot get into the bag.
3. Place the container with dish in a dark place at room temperature and check it daily.
 4. When IJ's begin to emerge, place the petri dish into a larger "catch" dish that has a little water in it.
 5. Nematodes will crawl into the water, where they may be poured off into a flask containing tap water that has been allowed to sit for at least 48 hours. The flask should be stored in the refrigerator with a loosened cap so nematodes have access to oxygen. The flask should not be filled completely, nematodes should not be too concentrated in the flask (i.e., the water should not appear to be thick and cloudy). Use multiple flasks to accommodate these storage requirements. Be sure to label flasks with the same data on the covered dish from which the nematodes emerged.
 6. Nematodes in solution may then be used to inoculate healthy waxworms by pouring a few milliliters of the collected nematode solution over 5 to 10 waxworms which should be placed on a clean damp piece of paper towel in a clean pre-labeled petri dish. Repeat steps 1 to 5.
 7. Stored nematodes should be used within three weeks for optimal efficacy.

Identification of Entomopathogenic Nematodes

To identify species of *Steinernema* and *Heterorhabditis*, the following should be considered:

- a) IJ morphometrics usually are insufficient for species identification, and male and female characteristics must be used.
- b) IJ produced on artificial media (laboratory reared or commercial products) are shorter

(rarely longer) than those produced in vivo, and usually do not meet the criteria of the original description. Males and females collected 4 or 5 days after the host dies, and IJ collected for one week after they first appear from cadavers, usually meet original species descriptions.

Morphology and morphometry and molecular identification of EPNs

The identification of EPNs is crucial for understanding their biology, ecology, and effective utilization as biocontrol agents. Several techniques have been developed for the identification of EPNs from citrus orchards, including morphological, molecular, and biochemical methods.

Morphological techniques involve the observation of different life stages of IJs, adults of 1st and 2nd generations' characteristic features, such as the shape and size of the body, head, tail, spicules, and the presence or absence of specific structures such as the excretory pore. These techniques are useful for initial identification, but their accuracy is limited, and they may require taxonomic expertise. Morphological features of EPNs include their cylindrical body shape, a distinct cuticle, and a pointed head with two sensory organs called amphids. EPNs also have a ventral gland and a digestive tract with a posterior anus. Morphometry can be used to measure the body length, width, and shape of EPNs, as well as the size and location of their amphids and other anatomical structures and most of the characters such as distance from anterior end to excretory pore, distance from anterior end to nerve ring, tail length, mucro length, a, b, c, SL, GL SW, GS, and D%. Biochemical techniques, such as enzyme assays and isozyme analysis, can also be used to identify EPNs. These techniques rely on the differences in the biochemical properties of different species and strains of EPNs.

However, they are less commonly used than morphological and molecular methods.

Molecular identification involves using genetic techniques to identify EPNs at the species level. This typically involves extracting DNA from the nematodes and using polymerase chain reaction (PCR) to amplify specific genetic markers, such as the internal transcribed spacer (ITS) region, *COI* gene etc. The amplified DNA can then be sequenced and compared to known sequences in DNA databases to identify the species.

Molecular identification has become an important tool in EPN research and application, as it allows for more accurate identification of species and strains, as well as the detection of new and previously unknown EPNs. Combined with morphological and morphometric analysis, molecular identification can provide a comprehensive understanding of the diversity and biology of EPNs, which can aid in their use as biological control agents for insect pests.

Family Steinernematidae

Steiner (1923) described the genus and species *Aplectana kraussei*, but the genus name was preoccupied so Travassos (1927) renamed the genus as *Steinernema*. Two year later Steiner described another genus and species *Neoaplectana glaseri*, close to *Steinernema kraussei*. In 1934, Filipjev placed *Steinernema* and *Neoaplectana* in the new subfamily Steinernematinae, he stated that *Neoaplectana* was probably congeneric with *Steinernema*. Chitwood and Chitwood 1937 raised the subfamily Steinernematinae to family Steinernematidae. Wouts *et al.* 1982, studied the two nematodes and concluded that *Neoaplectana* was a junior synonym of *Steinernema*, leaving the family with a single genus. In 1994, Nguyen and Smart described the

new genus *Neosteinerinema* and added this nematode to the family.

Presently, the family has two genera: *Steinernema* and *Neosteinerinema*.

Diagnosis

(After Nguyen and Smart, 1994): Cephalooboea, Rhabditida. Obligate insect parasites. Infective juveniles carry symbiotic bacteria in the bacterial chamber of the intestine. Both males and females are necessary for reproduction.

Females: Large, size variable. Cuticle smooth or annulated. Lateral fields absent. Excretory pore distinct. Head rounded or truncate, rarely offset. Six lips present, partly or completely fused, each lip with one labial papilla, sometimes additional papilla-like structures present near labial papillae. Four cephalic papillae. Amphids present, small. Stoma collapsed; cheilorhabdions pronounced, forming a ring resembling two large sclerotized dots in lateral view. Other parts of stoma forming an asymmetrical funnel with thick anterior end. Esophagus rhabditoid with metacarpus slightly swollen, narrow isthmus surrounded by nerve ring, and large basal bulb with reduced valve. Esophago-intestinal valve usually pronounced. Reproductive system didelphic, amphidelphic, reflexed. Vulva at mid-body, sometimes on a protuberance, with or without epiptygma. Females oviparous or ovoviviparous with juveniles developing up to the infective stage (IJ) before emerging from the body of the female. Tail longer or shorter than anal body width, with or without prominent phasmids.

Males: Smaller than female. Anterior end usually with six labial papillae, four large cephalic papillae and usually with perioral disc. Esophagus similar to that of the female. Testis single, reflexed; spicules paired;

gubernaculum long, sometimes as long as spicule; bursa absent. Tail tip rounded, digitate or mucronate. One single and 10 to 14 pairs of genital papillae present with 7 to 10 pairs precloacal.

Infective juveniles (=third-stage infective juvenile): Stoma collapsed. Body slender, with or without a sheath (cuticle of second-stage juvenile). Cuticle annulated. Lateral fields present with 4-9 incisures and 3-8 smooth ridges. Esophagus and intestine appearing reduced. Excretory pore distinct. Tail conoid or filiform. Phasmids, located about mid-tail, prominent, inconspicuous, or not observed.

Type genus: *Steinernema* Travassos, 1927

Other genus: *Neosteinerema* Nguyen and Smart, 1994

***Steinernema*, *Neosteinerema* species, names and authorities**

Genus *Steinernema* Travassos, 1927

Type species:

Steinernema kraussei (Steiner, 1923) Travassos, 1927

syn.

Aplectana kraussei Steiner, 1923

Steineria kraussei (Steiner, 1923) Travassos, 1927

Oxysomatium kraussei (Steiner, 1923) Skrjabin, Shikhobalova and Mozgovoï, 1951

Other species: *S. arenarium* (Artyukhovsky, 1967) Wout, Mracek, Gerdin and Bedding, 1982

syn.

Neoaplectana arenaria Artyukhovsky, 1967

Neoaplectana anomali Kozodoi, 1984

Steinernema anomalae (Kozodoi, 1984) Curran, 1989

S. abbasi Elawad, Ahmad & Reid, 1997

S. aciari Qiu, Yan, Zhou, Nguyen and Pang, 2005

S. akhursti Qiu, Hu, Zhou, Pang and Nguyen, 2005

S. apuliae Triggiani, Mracek and Reid, 2004

S. affine (Bovien, 1937) Wouts, Mracek, Gerdin and Bedding, 1982

syn. *Neoaplectana affinis* Bovien, 1937

S. backanense Phan, Spiridonov, Subbotin and Mones, 2006

S. bicornutum Tallosi, Peters & Ehlers, 1995

S. beddingi Qiu, Hu, Zhou, Pang & Nguyen, 2005

S. carpocapsae (Weiser, 1955) Wouts, Mracek, Gerdin and Bedding, 1982

syn.

Neoaplectana carpocapsae Weiser, 1955

Neoaplectana feltiae sensu Stanuszek, 1974, nec Filipjev, 1934

Neoaplectana feltiae pieridarum Stanuszek, 1974

Steinernema feltiae pieridarum (Stanuszek, 1974) Wouts, Mracek, Gerdin and Bedding, 1982

Neoaplectana carpocapsae pieridarum Stanuszek, 1974

Neoaplectana dutkyi Turco, Thames and Hopkins, 1971

Steinernema dutkyi (Turco, Thames and Hopkins, 1971) Wouts, Mracek, Gerdin and Bedding, 1982

S. caudatum Xu, Wang and Li, 1991

S. ceratophorum Jian, Reid and Hunt, 1997

S. cubanum Mracek, Hernandez and Boemare, 1994

S. cumgarensis Phan, Spiridonov, Subbotin and Moens, 2006

S. diaprepesi Nguyen and Duncan 2002

S. eapokense Phan, Spiridonov, Subbotin and Moens, 2006

S. feltiae (Filipjev, 1934) Wouts, Mracek, Gerdin and Bedding, 1982

syn.

Neoaplectana feltiae Filipjev, 1934

Neoplectana bibionis Bovien, 1937

Steinernema bibionis (Bovien, 1937) Wouts, Mracek, Gerdin and Bedding, 1982

Neoaplectana leucaniae Hoy, 1954

Steinernema leucaniae (Hoy, 1954) Wouts, Mrack, Gerdin and Bedding, 1982

S. glaseri (Steiner, 1929) Wouts, Mracek, Gerdin and Bedding, 1982

syn. *Neoaplectana glaseri* Steiner, 1929

- S. guangdongense* Qiu, Fang, Zhou, Pang and Nguyen, 2004
- S. intermedium* (Poinar, 1985) Mamiya, 1988
syn. *Neoaplectana intermedia* Poinar, 1985
- S. jollieti* Spiridonov Krasomil-Osterfeld, Moens, 2004
- S. karii* Waturu, Hunt and Reid, 1997
- S. khoisanae* Nguyen, Malan and Gozel, 2006
- S. kushidai* Mamiya, 1988
- S. leizhouense* Nguyen, Qiu, Zhou and Pang, 2006
- S. longicaudum* Shen and Wang, 1992
- S. loci* Phan, Nguyen and Moens, 2001
- S. monticolum* Stock, Choo and Kaya, 1997
- S. neocurtillae* Nguyen and Smart, 1992
- S. oregonense* Liu and Berry, 1996
- S. pakistanense* Shahina, Anis, Reid, Rowe and Maqbool. 2001
- S. puertoricense* Romin and Figueroa, 1994
- S. rarum* (de Doucet, 1986) Mamiya, 1988
syn. *Neoaplectana rara* de Doucet, 1986
- S. riobrave* Cabanillas, Poinar and Raulston, 1994
- S. ritteri* de Doucet and Doucet, 1990
- S. sasonense* Phan, Spiridonov, Subbotin and Moens, 2006
- S. scapterisci* Nguyen and Smart, 1990
syn. *Neoaplectana carpocapsae* 'Uruguay strain' of Nguyen and Smart, 1988
- S. sichuanense* Mracek, Nguyen, Taillier, Bomoemare and Chen, 2006
- S. silvaticum* Sturhan, Spiridonov and Mracek, 2005
- S. tami* Luc, Nguyen, Reid and Spiridonov, 2000
- S. thanhi* Phan, Nguyen and Moens, 2001
- S. thermophilum* Ganguly and Singh, 2000
- S. websteri* Cutler and Stock, 2003
- S. weiseri* Mracek, Sturhan and Reid, 2003

S. yirgalemense Nguyen, Tesfamariam, Gozel, Gaudler and Adams, 2004

Genus *Neosteinerinema* Nguyen & Smart, 1994

Type and only species:

Neosteinerinema longicurvicauda Nguyen and Smart, 1994

Family Heterorhabditidae

This family was erected by Poinar in 1976 when he described the genus and species *Heterorhabditis bacteriophora*. The family contains only one genus *Heterorhabditis*.

Diagnosis

Rhabditoidea, Rhabditida. Obligate insect parasites. Infective juveniles carrying symbiotic bacteria. Both hermaphroditic and amphimictic females present.

Hermaphroditic females: After entry into an insect host, infective juveniles developing into hermaphroditic females. Head truncate to slightly rounded, six conical lips well developed, separate, each with a terminal papilla; one or two small raised structures sometimes visible at the base of each lip; amphidial opening small. Stoma wide but shallow; cheilorhabdions present, forming a ring, in lateral view resembling two refractile elongate structures. Other parts of the stoma fused to form a collapsed posterior portion. Posterior part of stoma covered by esophagus. Esophagus without metacarpus; isthmus slender; basal bulb swollen; valve in basal bulb reduced. Nerve ring at middle of isthmus. Excretory pore usually posterior to end of esophagus. Vulva slight anterior to mid-body (V%=43-48), slit-like, surrounded by elliptical rings; ovotestis amphidelphic, reflexed. Oviparous, later becoming ovoviviparous. Tail pointed, longer than anal body width, postanal swelling usually present.

Amphimictic females: Similar to, but usually smaller than, hermaphroditic female; labial papillae prominent. Reproductive system amphidelphic. Vulva not functional for egg deposition, but functional for mating.

Males: Testis one, reflexed. Spicules paired, separate, slightly curved ventrally. Spicule head short, offset from lamina by a constriction. Gubernaculum usually about half as long as spicule length. Bursa peloderan with nine pairs of genital papillae.

Infective juveniles: Third-stage infective juvenile usually with sheath (cuticle of second-stage juvenile). Sheath with anterior tessellate pattern and longitudinal ridges; IJ cuticle striated with one smooth band marginated by two ridges in lateral fields. Head with prominent dorsal tooth. Mouth and anus closed. Stoma appearing as a closed chamber with parallel walls. Esophagus and intestine reduced. Excretory pore posterior to nerve ring. Symbiotic bacterial cells found in intestine. Tail pointed.

Type and only genus: *Heterorhabditis* Poinar, 1976

Heterorhabditis species, names and authorities

Genus *Heterorhabditis* Poinar, 1976

Type species:

Heterorhabditis bacteriophora Poinar, 1976

syn.

Chromonema heliothidis Khan, Brooks and Hirschmann, 1976

Heterorhabditis heliothidis (Khan, Brooks and Hirschmann, 1976) Poinar, Thomas and Hess, 1977

H. argentinensis Stock, 1993

Other species:

H. amazonensis Andalo, Nguyen and Moino, 2006

H. baujardi Phan, Subbotin, Nguyen and Moens, 2003

H. downesi Stock, Griffin and Burnell, 2002

H. floridensis Nguyen, Gozel, Koppenhofer and Adams, 2006

H. indica Poinar, Karunaka and David, 1992

syn. *H. hawaiiensis* Gardner, Stock and Kaya, 1994

H. marelatus Liu and Berry, 1996

syn. *H. hepialius* Stock, Strong and Gardner, 1996

H. megidis Poinar, Jackson and Klein 1987

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H. taysearae Shamseldean, El-Sooud, Abd-Elgawad and Saleh, 1996

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Snippets of Successful Citrus Farming from Small Holders of Maharashtra

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Abstract: The Indian farming community is comprised of a large chunk of small and marginal farmers. Citrus is a commercial crop and generally large farmers of Maharashtra are the big players in this domain. But a handful of small farmers are also striving their way towards success through their progressive outlook and perseverance and adopting scientific farming practices. The snippets of such successful small holders provided in the article send a message of encouragement to all citrus farmers throughout the country. Sagar Dnyaneshwar Chikte of Amravati district and Sunanda Santosh Salodkar of Nagpur district of Maharashtra are the shining examples of this edition.

Introduction

Farmers are the cornerstone of agricultural prosperity of India. The citrus growers of India have etched their success stories in not only bumper production but also in terms of export of their fruits. But often the small farmers are the ones who are left behind in this race in spite of the fact that 120 million small holder farmers exist in India who have less than 2 hectares of land as operational holding.

Snippet 1: Sagar Dnyaneshwar Chikte

Shri Sagar Dnyaneshwar Chikte is a citrus grower of Dawargaon taluka of Amravati district of Maharashtra. His main source of income is Nagpur mandarin cultivation in little over 2 hectares (about 6.5 acres).



Fig. 1: Shri Sagar Dnyaneshwar Chikte

Shri Sagar Dnyaneshwar Chikte is a dedicated citrus grower with a curiosity for learning advanced scientific package of practices of mandarin cultivation and replicating them in his orchard. He is also the opinion leader of various farmers' groups where he shares his knowledge of farming techniques with fellow farmers and guides them.

About his orchard

Shri Sagar Dnyaneshwar Chikte has an orchard of 2 hectares 73 R (6.5 Acre) with the trees planted at a spacing of 16 x 16 feet. He has 700 Nagpur mandarin plants that are 15 years old (purchased from private nursery), as well as 220 plants that are 2 years old (purchased from ICAR-CCRI, Nagpur). He cultivates only the *ambia bahar* of Nagpur mandarin. He has drip irrigation system in orchard.

Package of practices adopted

Shri Sagar Dnyaneshwar Chikte tries to follow the scientific package of practices as recommended by ICAR-CCRI, Nagpur and Dr. P.D.K.V Akola University. He does timely pruning of the trees for deadwood removal. As per CCRI recommendations he does spraying of fungicides and applies fertilizers to the trees. He

is aware of the fact that citrus orchards require good drainage facility. So, he ensures proper drainage in his orchard. He managed a good drainage system for rainwater in the monsoons which helped him to control root rot and *Phytophthora* infestation from his orchard. He also uses *Trichoderma* in the months of June-July as fungal disease management measure.

Every year he applies jivamrut (10 kg FYM +2 kg jaggery+ 2 kg besan+ 10 liter gomutra in 200 lit water) to his mandarin trees. He also applies waste decomposer to the soil (200 lit water + 2 kg jaggery+ 1 to 5 liter waste decomposer culture). He applies fertilizer judiciously after doing soil testing from CCRI (2012-13) and last year from PDKV Akola.

He also uses scientific methods to control the fruit drop problem in his orchard during the monsoons. For this purpose he uses Gibberellic Acid (GA) 3 gm, 2,4-D 3 gm, Bavistin, potassium nitrate, 0:52:34 or urea 3 kg and during heavy rains uses Aliate 500 gm in 200 lit water. According to him, Carbendazim and Thiophanate methyl also works well against fruit drop as a fungicide. He also provides adequate nutrients to the plants during the monsoons when there is high chance of fruit dropping.



Fig. 2: Shri Sagar Chikte following scientific package of practices in his citrus orchard



Fig. 3: Shri Sagar Chikte following scientific package of practices in his citrus orchard

Yield and income realized

Shri Sagar Dnyaneshwar Chikte gets an average yield of 35 to 40 tons of ambia bahar fruits from 700 Nagpur mandarin plants annually. Experimentation, planning and practicing scientific solutions for diseases and pests has resulted in quality fruits which fetch good market prices. He sells his produce at Rs. 25,000/ tons (buyers from Maharashtra and other states). So, he obtains an average annual income of around Rs. 8 to 10 lakhs per year from citrus farming. Before contact with ICAR-CCRI, there was a major fruit drop problem in his orchard which is now in control. The yield has also improved from nearly 25 tons to 35-40 tons per year.



Fig.4: Shri Sagar Chikte at his bearing Nagpur mandarin orchard

Progressive Outlook

Sagar maintained a steady communication with scientists of ICAR-Central Citrus Research Institute, Nagpur from year 2012-13 to till date and followed their advice and scientific methods to efficiently manage his orchard. Sagar wishes to involve his children in farming too. He often guides other citrus farmers through farmers field school organized by state agriculture department and ICAR-CCRI. He also shares his knowledge through social media platforms and even phone calls.

Awards and recognitions received

He was awarded by "Progressive farmer award" from ICAR-CCRI Nagpur on 28 July 2020 on the occasion of Foundation Day of CCRI. He delivered an invited lecture at training programme organized by VANAMATI named as "Trainers Training Programme on orange" on 8th July, 2022. He is an active member of Orange Growers Association of India. He delivered lecture in Farmers Field School organized by Agriculture department at Pipari, Ta- Karanja (Gh), Dist -Wardha on 1st September, 2021. He delivered lecture in Farmers Field School organized by Agriculture department for Mrigbahar Citrus Production at Hetikundi, Ta- Karanja (Gh), Dist -Wardha on 12th March, 2021. He delivered lecture in Farmers Field School organized by Agriculture department for Ambia bahar Citrus Production at Karajgaon, Ta- Karanja (Gh), Dist -Amravati on 29th September, 2021. He delivered lecture in Farmers Field School organized by Agriculture department for Ambia bahar Citrus Production (Flowering to Harvesting) at Karajgaon, Ta- Karanja (Gh), Dist -Amravati on 30th November, 2021. Dr. PDKV Akola University certified him for adoption of improved technologies of citrus farming and for active participation in transfer of technologies to other citrus growing farmers.

Snnipet 2: Sunanda Santosh Salodkar

Smt. Sunanda Santosh Salodkar (Jadhav) is a 44 years old woman citrus farmer from Sonegaon, taluka (P.O: Kalmeshwar) of Nagpur district, Maharashtra. She has 26 years of experience in citrus farming.



Fig. 5: Smt. Sunanda Santosh Salodkar

About her orchard

The plants were bought from a private nursery in Mohapa, Ta-Kalmeshwar, Nagpur district. 150 Nagpur Mandarin plants were planted in 1.5 acre area, 125 Sweet Orange plants in 1 acre area, and 25 Acid Lime plants in 0.25 acre area. She takes ambia bahar fruits for Nagpur mandarin and sweet orange.

Package of practices adopted

Smt. Sunanda is involved in farming operations like ploughing, harrowing and for other crops does sowing too. She has struggled 14 years in the farming sector to survive as a sole woman farmer. Initially she faced domination on various aspects but her dream to achieve success and become financially independent, served as her major drive. Her message to other women farmers is practicing farming with own ideas, doing small demonstration first and if the demonstration is successful then planning for large scale production.



Fig. 6: Smt. Sunanda Santosh Salodkar following scientific package of practices in her citrus orchard

Yield and income realized

The production of Nagpur Mandarin in her orchard is 50 -60 kg fruits / plants, Sweet orange 40-50kg / plant and Acid lime 20-25 kg/plant. On an average total production of Nagpur Mandarin from her orchard are 6-7 tons/year, Sweet orange 5-6 tons/year and Acid lime 2 tons/year. She sells Nagpur Mandarin at around Rs. 50,000/ton generating Rs. 3 to 3.5 lakhs per year, Sweet orange at Rs. 60,000/ton

yielding Rs. 2 to 2.5 lakhs per year and acid limes at Rs.100-125/kg generating Rs. 60,000 to Rs. 70,000 per year. So, on average she earns Rs. 5 to 6 lakhs from citrus farming annually.



Fig. 7: Smt. Sunanda Santosh Salodkar at her bearing sweet orange orchard

Progressive outlook

Sunanda is firm in taking her own decisions related to citrus farming but only after carefully analyzing the results in her small demonstration plot. Her future planning is taking two flowerings of citrus in a year and also utilize her land for cultivating other crops throughout the year. She also wishes to involve her children in farming.

Awards and recognitions received

Sunanda has received many awards and recognitions for her outstanding work as a woman farmer. Amongst the list the notable awards are Vasantarao Naik (Encouraging Award) (2009-2010), Jijamata Krushibhushan award (2010), Panjabrao Krushiratna Award (2020-21). The Marathi movie "Kapus

Kondyachi Goshta" (released in 2016) which had won 13 International Awards and even an Oscar nomination, was based on her life.

Conclusion

These farmers have proved that where there is a will there is a way. They have achieved recognition for their passion towards farming and are further planning to expand their activities. Such success stories are always an inspiration for small farmers to adopt scientific methods of farming and reap the benefits.

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Managing Gummosis and Root Rot Diseases in Citrus through Integrated Approach

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Abstract: Citrus is one of the major fruit crops grown in more than 50 countries including USA, China, India and Mediterranean region being the leading producers and exporters in the Northern hemisphere while Brazil is a major producer in the Southern hemisphere. In India, citrus is the third largest fruit crop after mango and banana. The major citrus producing states in India are Maharashtra, Andhra Pradesh, Karnataka and Punjab. Among them, Maharashtra is considered as topmost citrus growing state in terms of area and second in production. Commercial citrus cultivars grown in different regions require different soil and climate conditions to thrive. It demonstrates wide adaptability of citrus to wide range of soil and climatic conditions. However, average productivity of citrus fruits is alarmingly low as compared to the developed countries. Various constraints are responsible for low productivity and widespread problem of citrus decline in India. The disease gummosis and root rot has a global distribution, affecting all citrus varieties in tropical and subtropical citrus production regions, especially in warm, humid climates. The most difficult phase is to identify feeder root rot which marks the beginning of the disease. These early symptoms are not noticed by citrus growers till there is occurrence of lesion formation and gummosis on the trunk region. For timely and effective management, it is imperative to identify the disease at early stages and adopt integrated approaches.

In India, citrus is the third largest fruit crop after mango and banana cultivated over an area of 1064 thousand ha with production of 1,40,71 thousand MT. The major citrus producing states in India are Maharashtra, Andhra Pradesh, Karnataka and Punjab. Among them, Maharashtra is considered as top most citrus growing state in terms of area and second in production. The important commercial citrus fruits in India are mandarins (*Citrus reticulata*), sweet orange (*C. sinensis*) and acid lime (*C. aurantifolia*). Average productivity of citrus fruits is alarmingly low as compared to the developed countries. There are number of biotic and abiotic factors that are responsible for low productivity and widespread problem of citrus decline in India. Among the biotic factors, gummosis and root rot are the most serious and economically important diseases in major citrus-growing areas of India. It is increasingly becoming the major limiting factor in the

successful cultivation of citrus in the states where citrus cultivation is a predominant horticultural activity. Every year 5-10% plants die due to root rot/gummosis. It causes hidden damage by decaying the feeder roots of nursery as well as grown-up plants. Gummosis and root rot diseases are caused by *Phytophthora nicotianae* which attack all species and varieties of citrus being grown in India. These diseases reduce the productive life span of trees, and considerably reduce fruit yield and quality. It is very essential that citrus growers pay much attention to the timely detection of these diseases and adopt integrated approaches to effectively manage them in a sustainable way for profitable citriculture. The information regarding symptoms, suitable conditions for spread of these diseases along with their integrated management approach are given hereunder for the benefit of the orchardists.

Where and when do these diseases occur?

Heavy losses occur in citrus orchards where flood irrigation is used if trees are planted on any of the susceptible rootstocks (e.g. Rough lemon). Within orchards, flood irrigation water greatly spreads the fungus. Various cultural practices like low level of budding, deep planting, use of infected planting material, heavy and ill drained soil, injuries to the trunk and roots during orchard operations, piling of manure near the tree trunk, thick weed growth under the tree canopy and use of the same site for nursery raising make the trees more susceptible to the disease. Gummosis and root rot diseases commonly occur seriously during July-October following periods of high rainfall with warm temperatures. *Phytophthora* fungus infects roots and trunks under high soil moisture conditions. High susceptibility occurs when roots are stressed or damaged. Soils with drainage restricted by clay layers or those with low water table provide favourable conditions for infection of fibrous roots.

Symptoms

Gummosis: An important symptom is sap or gum oozing from cracks in the bark near the base of the tree. Because of this symptom, the disease is popularly known as “gummosis”. Later on the rotting of the bark occurs on the trunk or crown near the ground level. The colour of the bark and wood below the bark becomes dark brown. The bark develops vertical cracks. Removal of the soil around the base of the trunk of an affected tree will show bark that appears water-soaked, slimy, and reddish-brown to black. Foot rot lesions may develop as high as 45-60 cm from the ground level on the trunk and may extend below the soil line. The lesions may spread around the circumference of the trunk, slowly girdling the tree, leading to the death of the tree. Such girdled trees show typical pale green leaves with heavy defoliation, produce

large number of flowers and die before fruit maturity. Young trees of small trunk circumference are rapidly girdled and killed.

Root rot: It causes a slow decline of citrus trees, which becomes clear particularly under water stress. In dry season, the dead bark becomes firm, breaks away from healthy bark, curls and splits. Usually the disease is confined to feeder roots and remains unnoticed by the growers. The fungus causes a decay of feeder roots leaving only the white thread-like stele. Appearance of dull chlorotic leaves is the first symptom of such affected trees where mid-rib and main lateral veins become yellow. Such vein chlorosis is often confused with nitrogen deficiency. The diseased trees have comparatively fewer fibrous roots than healthy ones. In severe cases, where the production of new feeder roots does not keep pace with the rate of root rot, the affected trees grow poorly with sparse foliage with somewhat naked branches, stunted growth and die-back of twigs. Sometimes, these trees bear profusely and collapse when fruit are still on the trees by showing acute wilting symptoms.



Fig. 1: Oozing of gummy drops from bark



Fig. 2: Oozing of sap from bark

Management

Management aspects should focus on preventing conditions that encourage infection and disease development. Prevention through cultural practices and good sanitation is the most

important step in reducing the incidence and spread of gummosis and root rot diseases. In trees affected by these diseases, fungicidal treatments are needed to minimize damage. However, fungicide applications are not expected to be highly effective if cultural practices such as judicious need-based irrigation and improved drainage system are not followed. It is important to use an integrated approach for effective and cost-effective management of gummosis and root rot diseases. The useful integrated management practices are given below:

1. Preventive measures:

The disease is soil-borne and once it enters in nurseries and orchards, it becomes very difficult to eradicate the disease. So “prevention is better than cure”.

- Obtain the planting material from reputed nurseries. Such nurseries use containerized nursery system, sterilized potting mixture, and a separate set of nursery implements for growing plants in net/screen houses.
- Use *Phytophthora* tolerant rootstock such as Carrizo. But pH of orchard soil should be less than 8.0.
- Keep the bud union 9 inches above the ground level at the time of planting so that irrigation water does not touch the scion.
- Plant trees in well drained fields.
- Maintain good tree health. Provide only need-based irrigation. Do not give frequent heavy irrigations, as they favour rapid disease development and spread.
- Prevent flood irrigation water from reaching the trunk, as it promotes infection.
- Keep the tree basin slightly elevated to avoid water stagnation around the trunk.

- Provide a drainage ditch at the end of fields to allow excess soil water to leave the field during heavy rainfall periods.
- If possible, use drip irrigation system as it helps in proper water management to check the spread of *Phytophthora* fungus.
- Adopt proper sanitation practices in orchards. Remove weeds and grass, especially under the trees. Keep the area under the tree clean, dry and free of weeds.
- Don't pile soil around the trunk.
- Avoid using tractor-discs for removing weeds from under the trees and near the tree canopy to prevent any injury to the trunk bark and roots. Injuries provide entry points for fungal infection.
- Remove severely damaged (with more than 50% girdling) and unproductive trees from the orchard because they spread the fungus. Destroy the dead wood, to prevent it from becoming a source of infection for healthy trees.
- Intercropping should not be done in bearing orchards. In young and non-bearing orchards, intercropping up to four years with leguminous crops such as *Guara*, *Moong*, *Mash*, Cowpea, Gram and Pea may be done. Do not grow incompatible crops like *Berseem*, Potato and creeper type vegetables in citrus orchards. Because these intercrops need frequent irrigations in winter which is conducive for spread of the fungus.

2. Chemical control:

- Scrap the infected bark portion along with some healthy green part and disinfect the wounds with disinfectant solution. Collect and destroy the scrapped diseased bark to avoid further spread of the fungus in the soil. Don't keep it in the orchard. Cover the

wounds with Bordeaux paste which when dries up, apply Bordeaux paint followed by spray of Bordeaux mixture (2:2:250).

OR

Treat trunk lesions with Curzate M 8. Apply Curzate M 8 as paint (2g/100 ml of linseed oil) to the surgical portion with the help of brush, twice in a year during February-March and July-August.

- Clean the root zone of the tree by hoeing. Dissolve 25 g of Curzate M 8 in 10 litres of water for treating one tree and drench the root area with the solution followed by light irrigation in February-March and July-August.

OR

Give application of Bordeaux paste on tree trunk as pre-monsoon (in May) and post-monsoon (in October) with foliar spray (two sprays- pre-monsoon and post-monsoon) of potassium phosphonate @ 3g/litre water.

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Studies on Combined Influence of Pruning and Exogenous Application of Growth Regulators on Fruit Set, Yield and Quality of Sweet Orange (*Citrus sinensis* Swingle.) cv. Sathgudi

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Abstract: Among the different interaction treatments tested, to initiate flowering, improve behavior and quality improvement in sweet orange (*Citrus sinensis* Swingle) cv. Sathgudi, T₂ (P₁C₂ – Pruning 10 cm + NAA @ 100 ppm) recorded maximum fruit set percentage, more number of fruits per shoot, more number of fruits per tree, minimum fruit drop percentage, highest fruit yield per tree, highest juice content, maximum total soluble solids, low titrable acidity, more ascorbic acid content, maximum total sugars content. Highest fruit volume and fruit diameter were measured under T₁ (Pruning 10 cm + NAA @ 50 ppm). Maximum fruit weight and highest reducing sugars content were recorded in T₉ (Pruning 15 cm + GA₃ @ 50 ppm).

Introduction

The sweet orange (*Citrus sinensis* Swingle.) occupies the first position among all the commercial citrus species grown in the world. It prefers dry, sub-tropical climate for good growth, yield and for producing quality fruits. Sweet orange plants are generally planted at a spacing of 5 x 5 meters, and economical orchard life varies from 15 to 25 years depending upon the rootstock used, management practices followed and the prevailing agro-climatic conditions in a particular area. It is observed that on attaining the age of seven to eight years, the canopy of the sweet orange plant becomes dense and overcrowded, besides, excessive growth of the leaders and laterals may result in shade which may result in reduced pollination and decreases in yield.

Pruning is done to restrict excessive vegetative growth and to maintain a balance between leaf/fruit ratio, fruit size, fruit colour and other quality attributes.

**Part of the M.Sc. (Hort.)-Fruit Science thesis submitted to Dr. Y.S.R. Horticultural University by the first author.*

Flowering in sweet orange is recurrent under tropical and sub-tropical conditions unless synchronized into well defined period of extreme stress. Since the demand for the fruit remains very high during summer it is very essential to regulate flowering that gives fruiting in the months of April and May which fetches higher returns to the grower compared to the income receive during other seasons. There is difficulty in fruit set because of incomplete pollination, hence plant growth regulators may be effectively used to increase fruit set.

Hasta-bahar (September-October) management through the use of plant growth regulators and chemicals play an important role to get maximum fruit yields during summer (Lakshmi *et al.* 2014). Hence there is a need to test the plant growth through the use of plant growth regulators and chemicals for their role inducing flowering for the *Hasta bahar* crop.

Material and Methods

The experiment was carried out to find out the combined influence of pruning and growth

regulators at sweet orange orchard, College of Horticulture, Anantharajupeta, YSR Kadapa Dist., which was located in southern agro-climatic zone of Andhra Pradesh at an elevation of 184 m (606 feet) above mean sea level between 13° 99' North latitude and 79° 33' East longitudes. The experiment was laid out in Factorial Randomized Block Design with single control with three replications and one tree per replication. Two factors were taken, the first factor is pruning and the second one is growth regulators.

Pruning was done in two levels i.e. 10 cm pruning from the terminal portion of the shoot and 15 cm pruning from the terminal portion of the shoot done in each tree under each replication. The growth regulators were applied @ two concentrations each growth regulator i.e. NAA @ 50 ppm and 100 ppm, GA₃ @ 50 ppm and 100 ppm, KNO₃ @ 2% and 3%. Combination of pruning and growth regulators having 12 treatments and one control were taken. They are as follows.

- T₁ - Pruning 10 cm + NAA @ 50 ppm
- T₂ - Pruning 10 cm + NAA @ 100 ppm
- T₃ - Pruning 10 cm + GA₃ @ 50 ppm
- T₄ - Pruning 10 cm + GA₃ @ 100 ppm
- T₅ - Pruning 10 cm + 2% KNO₃
- T₆ - Pruning 10 cm + 3% KNO₃
- T₇ - Pruning 15 cm + NAA @ 50 ppm
- T₈ - Pruning 15 cm + NAA @ 100 ppm
- T₉ - Pruning 15 cm + GA₃ @ 50 ppm
- T₁₀ - Pruning 15 cm + GA₃ @ 100 ppm
- T₁₁ - Pruning 15 cm + 2% KNO₃
- T₁₂ - Pruning 15 cm + 3% KNO₃
- T₁₃ - Control

Pruning of the trees was done in the orchard by following two levels. They were Pruning (Heading back) was done in the second week of

September. Spraying of plant growth regulators is done twice at fortnightly intervals during October.

Results and Discussion

1. Fruiting parameters

1.1 Percentage of fruit set (%): The effect of pruning, growth regulators and their combinations significantly increased percentage of fruit set (Table 1). Effect of pruning has shown the highest percentage of fruit set (79.72%) under pruning treatment P₁ (Pruning 10 cm), which was showed significant superiority over pruning treatment (74.38%) in P₂ (Pruning 15 cm). The maximum percentage of fruit set (82.95%) was observed under treatment C₂ (NAA @ 100 ppm) which was significantly superior as compared to control and all other treatments whereas, minimum percentage of fruit set (69.95%) was observed under C₄ (GA₃ @ 100 ppm).

Combined effect of pruning and growth regulators revealed that the maximum percentage of fruit set (85.65%) was noted significantly superior under treatment combination T₂ (Pruning 10 cm + NAA @ 100 ppm), followed by T₃ (82.98%), T₁₁ (80.56%) and T₈ (80.24%) while, minimum percentage of fruit set (64.51%) was recorded under T₁₀ (Pruning 15 cm + GA₃ @ 100 ppm). The increase in the fruit set percentage might be due to the increased availability of nutrients from leaves by NAA while it may also be due to varietal genetic capability to set high or low percentage of fruits.

In the findings of present research all treatments showed a significant increase in fruit set of sweet orange cultivars as compared to control treatment. GA₃ also increased the fruit set, might be due to the translocation of carbohydrates and increased pollen viability and fertilization. These were lined with the findings of Mishra *et al.* (2012), Garcia-Martinez and Garcia-papi, (1979) and Ullah *et al.* (2014) in sweet orange cv. Mosambi and Kaur (2017) in litchi cv. Dehradun.

Table 1: Percentage of fruit set, number of fruits per shoot and number of fruits per tree as influenced by pruning and exogenous application of plant growth regulators in sweet orange cv. Sathgudi.

Pruning	Fruit set (%)	Number of fruits per shoot	Number of fruits per tree
P₁ - Pruning 10 cm	79.72	10.86	142.33
P₂ - Pruning 15 cm	74.38	9.38	120.55
SE m(±)	0.04	0.03	0.12
CD @ 0.05	0.12	0.08	0.36
Growth Regulators			
C₁ - NAA @ 50 ppm	77.46	8.50	122.16
C₂ - NAA @ 100 ppm	82.95	13.05	162.33
C₃ - GA ₃ @ 50 ppm	80.80	11.40	137.66
C₄ - GA ₃ @ 100 ppm	69.95	7.33	104.33
C₅ - 2% KNO ₃	80.10	11.55	131.16
C₆ - 3% KNO ₃	71.07	8.90	130.99
SE m(±)	0.02	0.02	0.07
CD @ 0.05	0.07	0.05	0.21
Interaction			
T₁ (P₁C₁) - Pruning 10 cm + NAA @ 50 ppm	77.41	8.10	109.66
T₂ (P₁C₂) - Pruning 10 cm + NAA @ 100 ppm	85.65	12.00	192.00
T₃ (P₁C₃) - Pruning 10 cm + GA ₃ @ 50 ppm	82.98	10.50	155.65
T₄ (P₁C₄) - Pruning 10 cm + GA ₃ @ 100 ppm	75.38	8.66	116.00
T₅ (P₁C₅) - Pruning 10 cm + 2% KNO ₃	79.64	9.29	121.32
T₆ (P₁C₆) - Pruning 10 cm + 3% KNO ₃	77.27	9.70	159.33
T₇ (P₂C₁) - Pruning 15 cm + NAA @ 50 ppm	77.50	9.00	134.66
T₈ (P₂C₂) - Pruning 15 cm + NAA @ 100 ppm	80.24	11.09	132.66
T₉ (P₂C₃) - Pruning 15 cm + GA ₃ @ 50 ppm	78.62	9.29	119.66
T₁₀ (P₂C₄) - Pruning 15 cm + GA ₃ @ 100 ppm	64.51	8.00	92.66
T₁₁ (P₂C₅) - Pruning 15 cm + 2% KNO ₃	80.56	9.80	141.00
T₁₂ (P₂C₆) - Pruning 15 cm + 3% KNO ₃	64.86	8.20	105.66
T₁₃ (P₀C₀) - Control	73.81	9.00	102.00
SE m(±)	0.06	0.04	0.17
CD @ 0.05	0.17	0.11	4.65

*NS = Non-significant

1.2 Number of fruits per shoot: The effect of pruning, growth regulators and their combinations significantly increased number of fruits per shoot (Table 1). The maximum number of fruits per shoot at (10.86) was recorded under pruning treatment P₁ (Pruning 10 cm), which was showed significant superiority over pruning treatment (9.38) in P₂

(Pruning 15 cm).The maximum number of fruits per shoot (13.05) was observed under treatment C₂ (NAA @ 100 ppm) which was significantly superior as compared to control and all other treatments whereas, minimum number of fruits per shoot (7.33) was observed under C₄ (GA₃ @ 100 ppm).

The combined effect of pruning and growth regulators, the maximum number of fruits per shoot (12.00) was recorded under T₂ (Pruning 10 cm + NAA @ 100 ppm) significantly higher than others, followed by T₈ (11.09), T₃ (10.50) while, minimum number of fruits per shoot (8.00) was recorded under T₁₀ (Pruning 15 cm + GA₃ @ 100 ppm) was followed by T₁ (8.10), T₁₂ (8.20). Pruning and plant growth regulators (PGR's) have been commonly used in modifying various physiological processes with advantage in plant growth, flowering, fruit yield and other attributes in phalsa crop. Pruning and PGR's give the significant results like increasing the yield and quality of phalsa. Application of growth substances *viz.*, auxins and gibberellins has been effective in increasing fruit set and yield in several fruit crops including phalsa. Similar findings were concluded by Lakra *et al.* (2018) in phalsa, Randhawa *et al.* (1959) and Saleem *et al.*, (2008) in sweet orange cv. Blood red.

1.3 Number of fruits per tree: The effect of pruning, growth regulators and their combinations significantly increased number of fruits per tree (Table 1). Pruning alone, the maximum number of fruits per tree (142.33) was recorded under pruning treatment P₁ (Pruning 10 cm), which was showed significant superiority over pruning treatment (120.55) in P₂ (Pruning 15 cm). The maximum number of fruits per tree (162.33) was observed under treatment C₂ (NAA @ 100 ppm) which was significantly superior as compared to control and all other treatments whereas, minimum number of fruits per tree (104.33) was observed under C₄ (GA₃ @ 100 ppm).

Regarding combined effect of pruning and growth regulators, the maximum number of fruits per tree (192.00) was noted under treatment combination T₂ (Pruning 10 cm + NAA @ 100 ppm) was found to be significantly

higher than other treatments, followed by T₆ (159.33), T₃ (155.65), T₁₁ (141.00) while, minimum number of fruits per tree (92.66) was recorded under T₁₀ (Pruning 15 cm + GA₃ @ 100 ppm) followed by T₁₃ (102.00).

Highest fruit production in open-canopy plants might be due to greater penetration of sunlight and better air circulation, creating a micro-climate conducive to synthesis of carbohydrates and phyto-hormones. The maximum number of fruits per plant with NAA might be attributed to less dropping of flowers and fruits, as the application of growth regulators made up the deficiency of endogenous auxin, which prevented formation of abscission layer possibly through the inhibition of enzymatic activity at higher temperature. The combined application of NAA along with zinc, iron and manganese significantly increased number of fruits and yield. Zinc being the nutrient responsible for auxin synthesis, might have influenced the higher fruit retention and increased the number of fruits per tree. These results are in line with Ghosh and Bera (2014) in sweet orange, Neware *et al.* (2017) in sweet orange cv. Mosambi, Sweety *et al.* (2018) in sweet orange cv. Jaffa and Nawaz *et al.* (2008) in Kinnow mandarin, Hussain *et al.* (2011).

1.4 Fruit drop (%): Data revealed that the effect of pruning and exogenous application of growth regulators on fruit drop was not found to be significant (Table 2). The decreased fruit drop percentage (28.79%) was recorded under pruning treatment P₁ (Pruning 10 cm). The increased fruit drop percentage (29.57%) was recorded under pruning treatment P₂ (Pruning 15 cm). The lowest fruit drop percentage (26.18%) was recorded under growth regulators treatment C₂ (NAA @ 100 ppm). The highest fruit drop percentage (30.62%) was recorded under growth regulators treatment C₄ (GA₃ @ 100 ppm).

Regarding combined effect of pruning and growth regulators on fruit drop percentage, the minimum fruit drop percentage (23.10%) was recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm). The maximum fruit drop percentage (39.31%) was recorded under treatment T₁₀ (Pruning 15 cm + GA₃ @ 100 ppm).

The NAA treatment significantly decreased fruit drop by the suppressing the formation of abscission layer. The beneficial role of sweet orange for reducing fruit drop may be explained from the fact that it maintains the ongoing physiological and biological process of inhibition of abscission. Similar findings were observed by Somwanshi *et al.* (2017) in sweet orange cv. Nucellar and Tomaszewska and Tomaszewska, (1970), Frolov, (1967).

The fruit drop synchronizes with the period of low auxin production in the fruit and suggested for application of auxin which would be helpful in increasing auxin level and thereby resulted in reduce fruit drop. Final fruit retention at harvest is influenced by fruit abscission occurring during various stages of development. Retention of more fruiting shoots in terms of length and numbers along with higher fruit set might have favoured the more number of fruits in control. The decrease in fruit drop with the application of growth regulator might be attributed to the fact that making up the deficiency of endogenous auxin prevented the formation of abscission layer possibly through the inhibition of enzymatic activity caused by pectinase and polygalactouronase enzyme. These findings are in consistent with the previous reports of Masalkara *et al.* (2009), Sharma and Singh (2018) in pomegranate cv. Kandhari Kabuli, Sweety *et al.* (2018) in sweet orange cv. Jaffa and Luckwill (1957).

1.5 Fruit yield per tree (Kg): The fruit yield per tree was recorded and presented in Table 2.

Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on fruit yield per tree. The increased fruit yield per tree (20.04 kg) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased fruit yield per tree (18.67 kg) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum fruit yield per tree (22.37 kg) was recorded under growth regulators treatment C₂ (NAA @ 100 ppm). The minimum fruit yield per tree (14.14 kg) was recorded under growth regulators treatment C₄ (GA₃ @ 100 ppm).

Interaction effect of pruning and growth regulators on fruit yield per tree has shown the significant increase in fruit yield per tree (25.03 kg) was observed under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm), followed by T₃ (22.43 kg), T₁ (20.61 kg), T₁₁ (20.32 kg). The minimum fruit yield per tree (12.08 kg) was observed under treatment T₁₀ (Pruning 15 cm + GA₃ @ 100 ppm) followed by T₁₃ (12.47 kg).

The maximum number of fruits per plant with NAA might be attributed to less dropping of flowers and fruits, as the application of growth regulators made up the deficiency of endogenous auxin, which prevented formation of abscission layer possibly through the inhibition of enzymatic activity at higher temperature. The results of present investigation are in conformation with the findings of Greenberg *et al.*, 2006, Sweety *et al.* (2018) in sweet orange cv. Jaffa and Hifny *et al.* (2017) in sweet orange cv. Washington Navel Orange. NAA application after fruit set reduced pre-harvest fruit drop and increased in fruit yield, fruit weight of Kinnow mandarin trees compared to control. The results are in contrast with the findings of Munoz- Fambuena *et al.* (2012) who found that GA₃ applied at the floral bud induction period significantly increased fruit yield per tree of Washington navel orange.

GA₃ is being used to improve the fruit set of parthenocarpic cultivars that tend to flower profusely.

The hormones may play a sequential and synergistic role in the retention and growth of fruitlets. Higher yield in aged plants may be

attributed to the pruning practices, best fruit retention and to production of weighable fruits followed on the tree. Results were in line with the findings of Ghosh and Bera, (2014), Joubert *et al.* (2000) in sweet orange and grapefruit and Nawaz *et al.* (2008) in Kinnow mandarin.

Table 2: Fruit drop percentage, fruit yield per tree and fruit length as influenced by pruning and exogenous application of plant growth regulators in sweet orange cv. Sathgudi.

Pruning	Fruit drop (%)	Fruit yield per tree (Kg)	Fruit length (cm)
P ₁ - Pruning 10 cm	28.79	20.04	6.68
P ₂ - Pruning 15 cm	29.57	18.67	6.54
SE m(±)	0.41	0.12	0.10
CD @ 0.05	NS	0.36	NS
Growth Regulators			
C ₁ - NAA @ 50 ppm	26.73	19.16	6.77
C ₂ - NAA @ 100 ppm	26.18	22.37	6.60
C ₃ - GA ₃ @ 50 ppm	28.99	20.81	6.72
C ₄ - GA ₃ @ 100 ppm	30.62	14.14	6.71
C ₅ - 2% KNO ₃	29.30	19.98	6.35
C ₆ - 3% KNO ₃	29.56	16.68	6.50
SE m(±)	0.24	0.07	0.06
CD @ 0.05	NS	0.21	NS
Interaction			
T ₁ (P ₁ C ₁) - Pruning 10 cm + NAA @ 50 ppm	24.28	20.61	6.77
T ₂ (P ₁ C ₂) - Pruning 10 cm + NAA @ 100 ppm	23.10	25.03	6.70
T ₃ (P ₁ C ₃) - Pruning 10 cm + GA ₃ @ 50 ppm	29.73	22.43	6.65
T ₄ (P ₁ C ₄) - Pruning 10 cm + GA ₃ @ 100 ppm	28.49	15.80	6.99
T ₅ (P ₁ C ₅) - Pruning 10 cm + 2% KNO ₃	29.75	19.64	6.39
T ₆ (P ₁ C ₆) - Pruning 10 cm + 3% KNO ₃	28.36	18.73	6.56
T ₇ (P ₂ C ₁) - Pruning 15 cm + NAA @ 50 ppm	27.55	19.71	6.76
T ₈ (P ₂ C ₂) - Pruning 15 cm + NAA @ 100 ppm	29.26	19.70	6.49
T ₉ (P ₂ C ₃) - Pruning 15 cm + GA ₃ @ 50 ppm	28.24	19.19	6.79
T ₁₀ (P ₂ C ₄) - Pruning 15 cm + GA ₃ @ 100 ppm	39.31	12.08	6.43
T ₁₁ (P ₂ C ₅) - Pruning 15 cm + 2% KNO ₃	28.85	20.32	6.31
T ₁₂ (P ₂ C ₆) - Pruning 15 cm + 3% KNO ₃	32.88	14.62	6.44
T ₁₃ (P ₀ C ₀) - Control	30.63	12.47	6.33
SE m(±)	0.58	0.18	0.14
CD @ 0.05	NS	0.51	NS

*NS = Non-significant

1.6 Fruit length (cm): The length of the fruit was recorded and presented in Table 2. Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on fruit length. The increased fruit length (6.68 cm) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased fruit length (6.54 cm) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum fruit length (6.77 cm) was recorded under growth regulators treatment C₁ (NAA @ 50 ppm). The minimum fruit length (6.35 cm) was recorded under growth regulators treatment C₅ (2% KNO₃).

Both the treatments combined effect of pruning and growth regulators on fruit length, the maximum fruit length (6.99 cm) was noticed under treatment T₄ (Pruning 10 cm + GA₃ @ 100 ppm). The minimum fruit length (6.31 cm) was observed under treatment T₁₁ (Pruning 15 cm + 2% KNO₃).

Increase in fruit length might be due to ability of gibberellic acid to increase cell enlargement, thus enhancing fruit growth. The reason for increase fruit size in term of length and girth due to GA₃ application might be due to increased level of carbohydrate in shoots which stimulated cell division and cell elongation resulting in larger fruit size as reported by Babu and Lavaniya (1985) in Pant lemon-1. In rainy season crop, reduction in size and weight of the fruits in unpruned plants was associated with the heavier crop loads which caused the drain on the food reserves of the plants and increasing competition among the growing fruit population for the food supply. Similar findings also reported by Pilania *et al.* (2010) in guava. It is evident from the findings of Ranganna *et al.* (2017) in acid lime cv. Balaji, Sah *et al.* (2018) in guava cv. Pant Prabhat and Jagtap *et al.* (2013) in Kagzi lime.

1.7 Fruit diameter (cm): The fruit diameter was recorded and presented in Table 3. Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on fruit diameter. The increased fruit diameter (6.76 cm) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased fruit diameter (6.70 cm) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum fruit diameter (6.90 cm) was recorded under growth regulators treatment C₁ (NAA @ 50 ppm). The minimum fruit diameter (6.52 cm) was recorded under growth regulators treatment C₅ (2% KNO₃).

Pruning and growth regulators combined together on fruit diameter, the highest fruit diameter (7.10 cm) was recorded under treatment T₁ (Pruning 10 cm + NAA @ 50 ppm). The lowest fruit diameter (6.30 cm) was recorded under treatment T₁₃ (Control).

The present results may be attributed to stimulative influence of this bio regulator on cell extension and/or cell division. The increase in fruit size may be attributed to the increase in cell division and cell elongation. Maximum fruit length and diameter resulted from cell elongation and cell division. Similar results were reported by Ranganna *et al.* (2017) in acid lime cv. Balaji, Hifny *et al.* (2017) in sweet orange cv. Washington Navel Orange and Dabbarma and Hazarika (2016) in acid lime. Increased fruit diameter associated with GA₃ application is probably due to involvement of GA₃ in mobilizing there serve foods to the growing apices as reported by Krishnamoorthy (1993).

1.8 Fruit weight (g): The weight of the fruit was recorded and presented in Table 3. Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on fruit weight.

The maximum fruit weight (148.08 g) was recorded under pruning treatment P₁ (Pruning 10 cm). The minimum fruit weight (146.13 g) was recorded under pruning treatment P₂ (Pruning 15 cm). The highest fruit weight (151.55 g) was recorded under growth regulators treatment C₃ (GA₃ @ 50 ppm). The lowest fruit weight (142.72 g) was recorded under growth regulators treatment C₆ (3% KNO₃).

As regards the combined effect of pruning and growth regulators on weight of the fruit, the maximum fruit weight (152.57 g) was recorded under treatment T₉ (Pruning 15 cm + GA₃ @ 50 ppm). The minimum fruit weight (130.97 g) was recorded under treatment T₁₃ (Control).

The increase in fruit weight may be due to the rapid increase in the size of cells or it is also due the fact that foliar application of GA₃ increased the fruit weight eventually by maintaining lighter level of auxins in various parts of the fruits which helped in increasing the fruit growth. These results were in lined with the findings of Haq *et al.* (2013). The results are having contrast with the findings of Kaur (2017) in litchi cv. Dehradun and Sharma *et al.* (1997) in Kinnow mandarin.

1.9 Fruit volume (cc): The fruit volume was recorded and presented in Table 3. Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on fruit volume. The increased fruit volume (160.51 cc) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased fruit volume (158.75 cc) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum fruit volume (167.29 cc) was recorded under growth regulators treatment C₃ (GA₃ @ 50 ppm). The minimum fruit volume (154.14 cc) was recorded under growth regulators treatment C₄ (GA₃ @ 100 ppm).

With regard to the combined effect of pruning and growth regulators on fruit volume, the increased fruit volume (167.87 cc) was recorded under treatment T₁ (Pruning 10 cm + NAA @ 50 ppm). The decreased fruit volume (143.83 cc) was recorded under treatment T₁₃ (Control).

The present findings are also in agreement with Dabbarma and Hazarika (2016) in acid lime. The reason for increase in fruit volume with the application of growth regulators could be due to increase in fruit size and accumulation of more pulp. The increase in fruit volume seems again due to their role in cell enlargement and division, increase in intercellular spaces in the mesocarpic cells and higher translocation of photosynthates and mineral nutrients from vegetative parts towards the developing fruits that are extremely active metabolic sink (Krishnamoorthy, 1993). Similar results were obtained from the findings of Singh and Singh (2015) in Narendra aonla-6, Parauha and Pandey (2019) in mango cv. Amrapali, Ranganna *et al.* (2017) in acid lime cv. Balaji.

2. Quality parameters

2.1 Juice (%): The percentage of juice was calculated and tabulated in Table 4. Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on juice percentage. The increased juice percentage (40.95%) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased juice percentage (40.85%) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum juice percentage (42.37%) was recorded under growth regulators treatment C₂ (NAA @ 100 ppm). The minimum juice percentage (39.31%) was recorded under growth regulators treatment C₅ (2% KNO₃).

Results on combined effect of pruning and growth regulators on juice percentage, the

maximum juice percentage (42.57%) were recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm). The minimum juice percentage (38.60%) was recorded under treatment T₁₃ (Control). Increase in juice percentage could be due to the fact that humic acid and fulvic acid fraction of the soil organic matter contributed by the organic sources

(vermicompost) would have probably formed water soluble micronutrient, thereby increasing their availability and uptake resulting in better quality. The present findings are also in agreement with Ghosh and Bera (2014) in sweet orange, Joubert *et al.* (2000) in sweet orange and Ghosh *et al.* (2017) in lemon cv. Assam lemon.

Table 3: Fruit diameter, fruit weight and fruit volume as influenced by pruning and exogenous application of plant growth regulators in sweet orange cv. Sathgudi.

Pruning	Fruit diameter (cm)	Fruit weight (g)	Fruit volume (cc)
P ₁ - Pruning 10 cm	6.76	148.08	160.51
P ₂ - Pruning 15 cm	6.70	146.13	158.75
SE m(±)	0.07	1.38	2.16
CD @ 0.05	NS	NS	NS
Growth Regulators			
C ₁ - NAA @ 50 ppm	6.90	150.95	164.56
C ₂ - NAA @ 100 ppm	6.79	145.73	159.41
C ₃ - GA ₃ @ 50 ppm	6.71	151.55	167.29
C ₄ - GA ₃ @ 100 ppm	6.53	147.63	154.14
C ₅ - 2% KNO ₃	6.52	144.05	155.18
C ₆ - 3% KNO ₃	6.82	142.72	158.21
SE m(±)	0.04	0.80	1.25
CD @ 0.05	NS	NS	NS
Interaction			
T ₁ (P ₁ C ₁) - Pruning 10 cm + NAA @ 50 ppm	7.10	150.67	167.87
T ₂ (P ₁ C ₂) - Pruning 10 cm + NAA @ 100 ppm	6.80	146.30	159.63
T ₃ (P ₁ C ₃) - Pruning 10 cm + GA ₃ @ 50 ppm	6.80	150.53	167.11
T ₄ (P ₁ C ₄) - Pruning 10 cm + GA ₃ @ 100 ppm	6.51	149.73	153.46
T ₅ (P ₁ C ₅) - Pruning 10 cm + 2% KNO ₃	6.65	147.23	158.09
T ₆ (P ₁ C ₆) - Pruning 10 cm + 3% KNO ₃	6.71	144.03	156.90
T ₇ (P ₂ C ₁) - Pruning 15 cm + NAA @ 50 ppm	6.94	151.23	159.24
T ₈ (P ₂ C ₂) - Pruning 15 cm + NAA @ 100 ppm	6.78	145.17	159.19
T ₉ (P ₂ C ₃) - Pruning 15 cm + GA ₃ @ 50 ppm	6.62	152.57	167.47
T ₁₀ (P ₂ C ₄) - Pruning 15 cm + GA ₃ @ 100 ppm	6.56	145.53	154.81
T ₁₁ (P ₂ C ₅) - Pruning 15 cm + 2% KNO ₃	6.41	140.87	152.26
T ₁₂ (P ₂ C ₆) - Pruning 15 cm + 3% KNO ₃	6.92	141.40	159.53
T ₁₃ (P ₀ C ₀) - Control	6.30	130.97	143.83
SE m(±)	0.10	1.95	3.05
CD @ 0.05	NS	NS	NS

* NS = Non-significant

Table 4: Juice percentage, total soluble solids and titrable acidity as influenced by pruning and exogenous application of plant growth regulators in sweet orange cv. Sathgudi.

Pruning	Juice (%)	TSS (^o Brix)	Titrable acidity (%)
P₁ - Pruning 10 cm	40.95	11.55	0.49
P₂ - Pruning 15 cm	40.85	11.45	0.51
SE m(±)	0.38	0.20	0.01
CD @ 0.05	NS	NS	NS
Growth Regulators			
C₁ - NAA @ 50 ppm	40.43	11.66	0.49
C₂ - NAA @ 100 ppm	42.37	11.68	0.50
C₃ - GA ₃ @ 50 ppm	40.89	11.46	0.51
C₄ - GA ₃ @ 100 ppm	41.80	11.80	0.48
C₅ - 2% KNO ₃	39.31	11.01	0.54
C₆ - 3% KNO ₃	41.20	11.38	0.51
SE m(±)	0.22	0.12	0.01
CD @ 0.05	NS	NS	NS
Interaction			
T₁ (P₁C₁) - Pruning 10 cm + NAA @ 50 ppm	41.43	11.50	0.49
T₂ (P₁C₂) - Pruning 10 cm + NAA @ 100 ppm	42.57	14.70	0.48
T₃ (P₁C₃) - Pruning 10 cm + GA ₃ @ 50 ppm	40.44	11.28	0.56
T₄ (P₁C₄) - Pruning 10 cm + GA ₃ @ 100 ppm	41.28	13.96	0.48
T₅ (P₁C₅) - Pruning 10 cm + 2% KNO ₃	39.47	11.06	0.49
T₆ (P₁C₆) - Pruning 10 cm + 3% KNO ₃	40.53	11.63	0.57
T₇ (P₂C₁) - Pruning 15 cm + NAA @ 50 ppm	39.42	11.83	0.50
T₈ (P₂C₂) - Pruning 15 cm + NAA @ 100 ppm	42.17	11.60	0.53
T₉ (P₂C₃) - Pruning 15 cm + GA ₃ @ 50 ppm	41.34	11.63	0.46
T₁₀ (P₂C₄) - Pruning 15 cm + GA ₃ @ 100 ppm	41.72	11.53	0.56
T₁₁ (P₂C₅) - Pruning 15 cm + 2% KNO ₃	39.16	10.96	0.52
T₁₂ (P₂C₆) - Pruning 15 cm + 3% KNO ₃	41.86	11.13	0.51
T₁₃ (P₀C₀) - Control	38.60	10.53	0.58
SE m(±)	0.53	0.28	0.02
CD @ 0.05	NS	NS	NS

* NS = Non-significant

2.2 TSS (^o Brix): The total soluble solids in the fruit juice was measured and tabulated in Table 4. Data revealed that the effect of pruning and exogenous application of growth regulators was insignificant effect on total soluble solids. The increased total soluble solids content (11.55 ^oBrix) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased total soluble solids content (11.45 ^oBrix) was recorded under pruning treatment P₂ (Pruning 15 cm). The

maximum total soluble solids content (11.80 ^oBrix) was recorded under growth regulators treatment C₄ (GA₃ @ 100 ppm). The minimum total soluble solids content (11.01 ^oBrix) was recorded under growth regulators treatment C₅ (2% KNO₃).

The cumulative effect of both the factors like pruning and growth regulators has revealed, the increased total soluble solids content (14.70 ^oBrix) was recorded under treatment T₂

(Pruning 10 cm + NAA @ 100 ppm). The decreased total soluble solids content (10.53 °Brix) was recorded under treatment T₁₃ (Control). The increased TSS due to the spray of NAA might be the increased translocation of sugars from source to the sink. The increase in the sugar: acid ratio and reduces the fruit pressure, which is an index of fruit hardness or softness. The softening of fruit may be explained through its action on cell wall hydrolysis. Increase in membrane permeability in plant cells which might facilitate accelerated breakdown of organic acids stored in cell vacuoles with consequent increase in TSS content. During ripening carbohydrates convert into simple sugar, which may ultimately increase the TSS content of fruits. Similar results were reported by Ghosh and Bera (2014) in sweet orange, Joubert *et al.* (2000), Sweety *et al.* (2018) in sweet orange cv. Jaffa and Ahmad *et al.* (2008).

2.3 Titrable acidity (%): The titrable acidity in the fruit juice was calculated and tabulated in Table 4. Results revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on titrable acidity. The decreased titrable acidity (0.49%) was recorded under pruning treatment P₁ (Pruning 10 cm). The increased titrable acidity (0.51%) was recorded under pruning treatment P₂ (Pruning 15 cm). The minimum titrable acidity (0.48%) was recorded under growth regulators treatment C₄ (GA₃ @ 100 ppm). The maximum titrable acidity (0.54%) was recorded under growth regulators treatment C₅ (2% KNO₃).

The combined effect of pruning and growth regulators on titrable acidity revealed that, the minimum titrable acidity (0.48%) was recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm). The maximum titrable acidity (0.58%) was recorded under treatment T₁₃

(Control). The decreased titrable acidity content due to the spray of NAA might be because of the increased translocation of sugars from source to the sink. The findings are also supported by. The increase in the sugar: acid ratio and reduces the fruit pressure, which is an index of fruit hardness or softness. The softening of fruit may be explained through its action on cell wall hydrolysis. The reduction in titratable acidity with chemicals may be due to its action on the fast conversion of organic acids and starch into reducing and non-reducing sugars and their derivatives through higher respiration and carbon assimilation activity during rapid ripening process. Similar findings were reported by Sweety *et al.* (2018) in sweet orange cv. Jaffa, Ghosh *et al.* (2017) in lemon cv. Assam lemon, Ahmad *et al.*, (2008) and Yadav *et al.* (2001).

2.4 Ascorbic acid content (mg/ 100 ml of juice): Results on the ascorbic acid content in the fruit juice tabulated in Table 5. Data revealed that the effect of pruning and exogenous application of growth regulators was showed significant effect on ascorbic acid content.

More ascorbic acid content (57.10 mg/ 100 ml of juice) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased ascorbic acid content (56.08 mg/ 100 ml of juice) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum ascorbic acid content (59.00 mg/ 100 ml of juice) was recorded under growth regulators treatment C₅ (2% KNO₃). The minimum ascorbic acid content (53.84 mg/ 100 ml of juice) was recorded under growth regulators treatment C₁ (NAA @ 50 ppm).

Combined effect of pruning and growth regulators results reveal that there was significant increase in ascorbic acid content (59.90 mg/ 100 ml of juice) recorded under treatment T₂ (Pruning 10 cm + NAA @ 100

ppm) compared to all other treatments, which was followed by T₁₁ (57.29 mg/ 100 ml of juice), T₃ (56.82 mg/ 100 ml of juice), T₉ (56.76 mg/ 100 ml of juice). The minimum ascorbic acid

content (51.37 mg/ 100 ml of juice) was recorded under treatment T₁₃ (Control), on par with T₁ (52.56 mg/ 100 ml of juice).

Table 5: Ascorbic acid content, reducing sugars and total sugars content as influenced by pruning and exogenous application of plant growth regulators in sweet orange cv. Sathgudi.

Pruning	Ascorbic acid content (mg/100 ml of juice)	Reducing sugars (%)	Total sugars (%)
P₁ - Pruning 10 cm	57.10	3.04	6.90
P₂ - Pruning 15 cm	56.08	3.12	6.28
SE m(±)	0.61	0.07	0.20
CD @ 0.05	1.77	NS	0.59
Growth Regulators			
C₁ - NAA @ 50 ppm	53.84	3.01	6.66
C₂ - NAA @ 100 ppm	58.27	2.89	7.05
C₃ - GA ₃ @ 50 ppm	57.79	3.19	6.65
C₄ - GA ₃ @ 100 ppm	54.50	3.10	7.29
C₅ - 2% KNO ₃	59.00	3.18	6.98
C₆ - 3% KNO ₃	56.15	3.10	6.10
SE m(±)	0.35	0.04	0.12
CD @ 0.05	1.02	NS	0.34
Interaction			
T₁ (P₁C₁) - Pruning 10 cm + NAA @ 50 ppm	52.56	2.98	6.71
T₂ (P₁C₂) - Pruning 10 cm + NAA @ 100 ppm	59.90	3.07	8.29
T₃ (P₁C₃) - Pruning 10 cm + GA ₃ @ 50 ppm	56.82	3.01	6.71
T₄ (P₁C₄) - Pruning 10 cm + GA ₃ @ 100 ppm	54.52	3.08	7.19
T₅ (P₁C₅) - Pruning 10 cm + 2% KNO ₃	56.71	3.02	7.11
T₆ (P₁C₆) - Pruning 10 cm + 3% KNO ₃	56.12	3.09	6.20
T₇ (P₂C₁) - Pruning 15 cm + NAA @ 50 ppm	55.13	3.05	6.61
T₈ (P₂C₂) - Pruning 15 cm + NAA @ 100 ppm	56.64	2.71	6.61
T₉ (P₂C₃) - Pruning 15 cm + GA ₃ @ 50 ppm	56.76	3.36	6.60
T₁₀ (P₂C₄) - Pruning 15 cm + GA ₃ @ 100 ppm	54.47	3.13	7.39
T₁₁ (P₂C₅) - Pruning 15 cm + 2% KNO ₃	57.29	3.35	6.85
T₁₂ (P₂C₆) - Pruning 15 cm + 3% KNO ₃	54.17	3.11	5.99
T₁₃ (P₀C₀) - Control	51.37	2.91	5.54
SE m(±)	0.86	0.10	0.29
CD @ 0.05	2.51	NS	0.84

*NS = Non-significant

High ascorbic acid content in fruits might be due to proper supply of nutrients and induction of growth hormones, which stimulate cell division, cell elongation, increase in number and weight of fruits, better root development, and better translocation of water and deposition of nutrients. This might be attributed to improved fertilizer use efficiency with the application of

organic source of nutrients. Increase in ascorbic acid content might be due to more availability of nutrients both in leaf and soil under this particular treatment. The results have accordance with the findings of Ghosh and Bera (2014) in sweet orange, Ghosh *et al.* (2017) in lemon cv. Assam lemon and Joubert *et al.* (2000).

2.5 Reducing sugars (%): The reducing sugars in the fruit juice was calculated and tabulated in Table 5. Data revealed that the effect of pruning and exogenous application of growth regulators was not showed significant effect on reducing sugars. The increased reducing sugars content (3.12%) was recorded under pruning treatment P₂ (Pruning 15 cm). The decreased reducing sugars content (3.04%) was recorded under pruning treatment P₁ (Pruning 10 cm). The maximum reducing sugars content (3.19%) was recorded under growth regulators treatment C₃ (GA₃ @ 50 ppm). The minimum reducing sugars content (2.89%) was recorded under growth regulators treatment C₂ (NAA @ 100 ppm).

With regard to the combined effect of pruning and growth regulators on reducing sugars content, the maximum reducing sugars content (3.36%) was recorded under treatment T₉ (Pruning 15 cm + GA₃ @ 50 ppm). The minimum reducing sugars content (2.71%) was recorded under treatment T₈ (Pruning 15 cm + NAA @ 100 ppm).

The increase in sugar content in treated plants may be due to the accelerated activities of hydrolytic enzymes, which is associated with high metabolic changes in fruits, leading to the conversion of complex polysaccharides and organic acids into simple sugars through higher respiration and carbon assimilation activity (Yadav *et al.* 2001). Increase in reducing sugars might be due to the effect of gibberellic acid (Tymowska-Lalanne and Kreis, 1998) on the activity of invertase enzyme, which break down sucrose into fructose and glucose and hence results in increased reducing sugars. Increased fruit quality could be due to the fact that the different sources of organic and inorganic nutrients enhanced the nutrient availability by enhancing the capability of plants for better uptake of nutrients from rhizosphere resulting in

the conversion of acid to sugar and their derivatives by the reversal glycolytic pathway. Increase in reducing sugars might be due to the effect of gibberellic acid (Tymowska-Lalanne and Kreis, 1998) on the activity of invertase enzyme, which break down sucrose into fructose and glucose and hence results in increased reducing sugars. Similar findings were reported by Ghosh *et al.* (2017) in lemon cv. Assam lemon, Singh *et al.* (2018) in Kinnow mandarin and Saleem *et al.* (2008) in sweet orange.

2.6 Total sugars (%): The total sugars content in the fruit juice was calculated and tabulated in Table 5. Data revealed that the effect of pruning and exogenous application of growth regulators was showed significant effect on total sugars content. The increased total sugars content (6.90%) was recorded under pruning treatment P₁ (Pruning 10 cm). The decreased total sugars content (6.28%) was recorded under pruning treatment P₂ (Pruning 15 cm). The maximum total sugars content (7.29%) was recorded under growth regulators treatment C₄ (GA₃ @ 100 ppm). The minimum total sugars content (6.10%) was recorded under growth regulators treatment C₆ (3% KNO₃).

Results on combined effect of pruning and growth regulators total sugars content, there was significant increase in total sugars content (8.29%) recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm), followed by T₁₀ (7.39%), T₄ (7.19%), T₅ (7.11%). The minimum total sugars content (5.54%) was recorded under treatment T₁₃ (Control), on par with T₁₂ (5.99%), T₇ and T₈ (6.61%).

The increase in sugar content in treated plants may be due to the accelerated activities of hydrolytic enzymes, which is associated with high metabolic changes in fruits, leading to the conversion of complex polysaccharides and organic acids into simple sugars through higher

respiration and carbon assimilation activity (Yadav *et al.* 2001). Higher total sugar may be due to increased leaf to fruit ratio which attribute towards more synthesis of carbohydrates other metabolites and their translocation to the fruit tissues. The present findings are also in agreement with Joubert *et al.* (2000), Ghosh and Bera (2014) in sweet orange, Gurjar *et al.* (2018) in guava cv. G-27, Ghosh *et al.* (2017) in lemon cv. Assam lemon and Bhagawati *et al.* (2015).

Conclusion

From the investigation it is clear that pruning and exogenous application of growth regulators are responsible for manipulating the vegetative growth and flowering behavior in sweet orange. With regard to the maximum percentage of fruit set (85.65%) was noted significantly superior under treatment combination T₂ (Pruning 10 cm + NAA @ 100 ppm) compared to other treatments, followed by T₃ (82.98%), T₁₁ (80.56%).

The combined effect of pruning and growth regulators, the maximum number of fruits per shoot (12.00) was recorded with T₂ (Pruning 10 cm + NAA @ 100 ppm) significantly higher than others, followed by T₈ (11.09), T₃ (10.50). Maximum number of fruits per tree (192.00) was noted under treatment combination T₂ (Pruning 10 cm + NAA @ 100 ppm) was found to be significantly higher than other treatments, followed by T₆ (159.33), T₃ (155.65). The minimum fruit drop percentage (23.10%) and fruit yield per tree (25.03 kg) was recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm). followed by T₃ (22.43 kg), T₁ (20.61 kg).

Fruit quality as indicated by many observations like fruit size, weight, juice percentage, was influenced by the application of pruning and growth regulators. The maximum fruit length

(6.99 cm) was noticed under treatment T₄ (Pruning 10 cm + GA₃ @ 100 ppm). The highest fruit diameter (7.10 cm) and increased fruit volume (167.87 cc) were recorded under treatment T₁ (Pruning 10 cm + NAA @ 50 ppm). Maximum fruit weight (152.57 g) was recorded under treatment T₉ (Pruning 15 cm + GA₃ @ 50 ppm).

The maximum juice percentage (42.57%) was recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm). With regards to the biochemical characters like TSS, titrable acidity, ascorbic acid and reducing and total sugars content, the present study showed little influence to increase quality. The increased total soluble solids content (14.70 °Brix), minimum titrable acidity (0.48%) and increase in ascorbic acid content (59.90 mg/ 100 ml of juice) were recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm). The combined effect of pruning and growth regulators on reducing sugars content, the maximum reducing sugars content (3.36%) was recorded under treatment T₉ (Pruning 15 cm + GA₃ @ 50 ppm). There was significant increase in total sugars content (8.29%) recorded under treatment T₂ (Pruning 10 cm + NAA @ 100 ppm), followed by T₁₀ (7.39%), T₄ (7.19%).

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Some Good Practices for Genetic Diversity Enhancement and Conservation as well as Assuring Sustained Farmer's Income

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Abstract: India's farmers are implementing best practices to preserve genetic diversity and ensure consistent income. These practices include planting multiple fruit trees, cultivating multiple varieties, planting seedlings alongside grafted plants, domestic gardening, and religious ceremonies. These practices provide year-round income, increase fruit supply, protect against pests and diseases, and promote horticulture. Multi-variety cultivation ensures consistent income, increased fruit availability, and pest and disease control. This genetic diversity enhancement can be applied to cultivar improvement programs. Kitchen gardening provides fresh food and conservation of genetic material, while maintaining rare fruit varieties for religious celebrations and health benefits. Encouraging these practices in rural communities can preserve and improve the genetic diversity of fruit commodities, boosting agricultural communities' health and income.

In India, many good practices are being followed by the farmers, which are not only providing the sustainable conservation of the genetic diversity, but also helping in getting assured and regular income over a period. These practices are being followed in almost all the states of our country, but their purpose may be somewhat different depending on the area. These practices are being passed from one generation to next generation. Because of the perennial nature of the fruit crops and their cross-pollinated behaviour, such practices are helping in maintenance, conservation as well as creation of genetic variability that can be selected and released in the form of new cultivars and thus helping in the evolution of the fruit crops.

Methodology followed for data collection and validating the good practices

The information on the good practices being followed by the farmers was collected from both primary and secondary sources. Data were collected by semi-structured surveys and several

unstructured interviews and interactions with key farmers. The survey included interviews in the form of a questionnaire and group discussions with the farmers, while non-survey data collection included the information collected through field visits made for clonal selection in citrus, mango and litchi orchards. Both qualitative and quantitative data were collected. Data of fruit and other crop diversity maintained by the farmers were collected.



Fig. 1: Group meetings for getting feedback/information from the farmers

The impact of these good practices was recorded in the form of fruits and vegetables consumption by the farming families and increase in their

family income from the sale of surplus fruits and vegetables produced by them. For the good practices, which we have come across during the surveys carried out for the clonal selection in citrus, litchi and mango and during discussion with the stakeholders were reported and some of the important practices are as under:

Multiple fruit tree planting in the field

Many farmers used to plant many fruits in their fields (*e.g.*, citrus, mango, litchi, banana, sapota, bael, aonla, *etc.*). These fields act as diversity gardens and help in maintenance of intergeneric as well as interspecific diversity. The practice gives all the year-round income to the farmers by sale of different types of fruits in different seasons.



Fig. 2: Multiple fruit tree planting

Multi variety planting of a fruit crop

The farmers plant many varieties of fruit (*e.g.*, citrus, mango, litchi) in the orchard. This ensures regular income, increased fruit

availability duration and control of many insect-pests and diseases. This also ensures the conservation of the intraspecific diversity. As most of the fruit crops are out-crossing, this will also lead to evolution of new types as well as some natural hybrids, that can be selected and released in the form of new cultivars. This practice is particularly important in the case of citrus, litchi and mango, where the farmers plant multiple varieties of these crops. This will help in extending the harvesting period as well as avoiding the market glut, leading to longer fruit availability and better price for the farmer's produce. The genetic diversity enhancement results naturally, which can be utilized for the varietal improvement programmes.

By following this practice, the farmers get income regularly even if the fruit trees do not bear regularly. This practice is being followed by many fruit tree growers. Even if some natural calamity or some pest break happens in one fruit crop, the farmer can get some income from other fruits. Moreover, the practice also provides some sort of protection from the damages caused by insect-pest and diseases as well as the natural calamities. The practice can also prove helpful in promoting horti-tourism in the near future.



Fig. 3: Multi-variety fruit crops planting

Planting of seedlings along with the grafted plants

This practice is generally followed in mango, where on the border, a line or two of the seedling plants are planted in addition to the grafted varieties. This provides opportunity for the evolution, development of new varieties and adds to the income of the growers. From the seedling plants, new selections can be made, because of the out-crossing nature of majority of the fruit crops. The seedlings can also provide the pollination services and act as wind breaks. Owing to the variable season of ripening of the fruits of seedlings, they will expand the harvesting period and provide income to the farmers for a longer period. For the sale of fruits of these seedlings, the practice of packing of diversity boxes might be followed, where a box of fruit carries fruits from multiple seedlings.

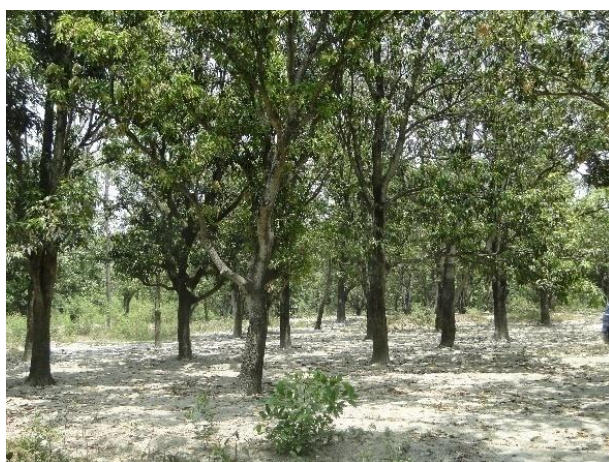


Fig. 4: Growing of varieties and seedlings of mango

The fruits from such orchards can also be sold in the form of roadside fruit sale. The seedlings are generally free from the virus and virus-like pathogens carried out by the mother trees; the chances of their longer survival are always there.



Fig. 5: Roadside sale of fruits from mixed planting

Kitchen gardening

In many house-holds, the kitchen gardening is a common practice where in addition to the floricultural and vegetable crops, some fruit trees are also grown, generally in the backyard of the house. This practice helps in the resource saving, provide fresh eatables in the form of vegetables and fruits, maintains different types of fruits and vegetables, ultimately leading to germplasm conservation and maintenance. The kitchen garden is a status symbol in the society and is mainly maintained by the house-wives and thus helps in giving opportunities to them. Many farmers used to plant unique type of fruits in these gardens leading to broadening of the genetic base. Generally, the seeds of the best fruits are planted in these gardens, which lead to the evolution of new plant types that ultimately

leads to the development of new fruit plant varieties. Sometimes, the farmer tries to have unique types and if they are successful, the new fruit crops can be grown on large scale.

Different types of fruits such as papaya, mango (dwarf type, sucking type, pickling type and common commercial varieties) citrus (mandarin, sweet orange, pummelo, lime and lemon), banana, guava, pomegranate, aonla, seasonal vegetables (leafy vegetables, cucurbits, beans, cabbage, chilli, coriander, tomato, onion, garlic, *etc.*) and some flowering plants (China rose, marigold, *etc.*) are planted. These provide fresh fruits, vegetables and flowers throughout the year. The seeds used for flowering plants and vegetables are from the farmers own seed savings and thus entail high genetic variability. Some of the farmers are interested in maintaining unique types of fruits and vegetables out of hobby, thus increasing the inter- and intraspecific biodiversity. The households plant many seedling types of different fruits, thus giving a chance for the selection of improved types from these fruit crops. During the clonal selection survey for the selection of improved types in mango, litchi and citrus, we have identified many different types from the farmer's fields. The origin of different types of mango and pummelo might be due to their cross-pollinating nature and monoembryonic seeds, respectively.



Fig. 6: Kitchen gardening in a village

Religious celebrations

In many religious ceremonies, many fruits are offered to the gods that leads to maintenance and conservation of different fruit species. Many house-holds plant fruit trees along with seasonal vegetables and ornamental plants in the premises of the house for multi-purpose functions like consumption, aesthetics, beautification, hobby and for some religious celebrations like the *Chhath Puja*, particularly in the states of Bihar, Uttar Pradesh, and Jharkhand. The fruits are used as special offering (*puja*) performed by the women of the village a week after Diwali celebrations in October or November as part of local Hindu tradition in which they offer 7, 9 or 11 different fruits to the God of the Sun. Several fruit species like pummelo, coconut and late mango seedlings are maintained specifically to supply fruits for this religious occasion. The majority of the fruits offered in this pooja have one or the other medicinal property and thus help in improving the health of the people.



Fig. 7: Chhath Puja celebrations

Maintenance of sacred groves

At many places and particularly in the rural areas, people plant the fruiting trees in the proximity of village and do religious ceremonies inside the grove and no one among the villagers cut these trees, ultimately leading to *ex situ* germplasm conservation. Such groves are common near the places of *Puja*, like temples. In these groves, many unique types of fruits can be found. This provides a sort of facilities for the testing of the new fruit types so that their large-scale plantations can be made for commercial purposes. The cutting of fruit trees as well as commercial sale of the fruits is generally prohibited from these groves, meaning the villagers can enjoy the fruits without paying any money, ultimately leading to their good health.

Such practices should be advocated in the rural communities so that the genetic diversity of the fruit crops is conserved and enhanced. The enhanced farm produce will improve the health of the farming communities and help in increasing their income, which will in turn will improve their social status and the increased incomes can be utilized by the rural communities for the better education of their children.

CONTRIBUTOR



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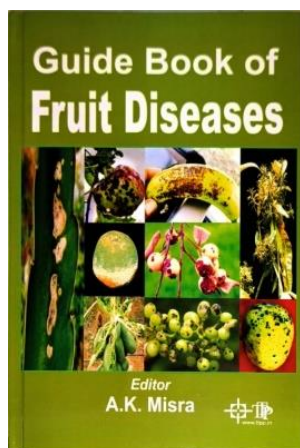


Review of “Guide Book of Fruit Diseases”

H. B. Singh

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*Reviewer’s Email: hbs1@rediffmail.com



The Guide Book of Fruit Diseases (ISBN: 9789391734398), edited by Dr. A.K. Misra, former Project Coordinator, AICRP on Subtropical fruits and Former Head, Division of Crop Protection, ICAR-Central Institute for Subtropical Horticulture, Lucknow is

published by Today & Tomorrow’s Printers and Publishers, New Delhi, in May 2023.

This is a recent publication in the field of fruit diseases. The book is fully printed in colour, depicting typical symptoms, causal pathogens and disease cycles. The book comprises 485 pages and contains 15 chapters covering more than 142 diseases with 521 coloured photographs of 14 fruit crops. The fruits covered are apple, banana, citrus, grapes, guava, mango, papaya, litchi, jackfruit, dragon fruit, ber, bel, aonla and sapota. The geographical distribution, crop losses, symptoms, causal organisms, favourable conditions, disease cycle and efficient management with new advances of each crop are presented in a precise manner. The chapters are contributed by prominent scientists and crop specialists namely Drs. Dilip Ghosh, A.K. Misra, A.K. Das, R. Selvarajan, S. Sriram, S.K. Singh, Dr. Indu Sawant, Dr. Sanjay Sawant, Dr. Vinod Kumar, Dr. K.P. Singh and Sangeetha Ganesan, In this volume the diseases of citrus is systematically dealt in greater detail by Dr. Dilip Ghosh, Director ICAR-CCRI, Nagpur along with Dr. A.K. Das, Principal Scientist, Plant Pathology, ICAR-CCRI, Nagpur.

The special quality of the book is that with each disease, separate references are cited, which is easier for the readers to gather more information on the disease. The book also provides nicely drawn disease cycles of most of the diseases, which will help the readers to understand about the diseases and their critical stages. Photographs of most of the pathogens are also provided with each disease, which is very useful for scientists and students of plant pathology working on these crops.

The book is dedicated to the late Prof. A.P. Misra, a world-renowned great Plant Pathologist of his time. The book is excellently printed with quality, uniformity and with no error. Publisher has taken special care to maintain the quality of publication with excellent setting and clarity of photographs. It is a very useful publication, which was required for a very long time and it is hoped that this book would serve as ready source of information for plant pathologists, horticulturists, nursery managers, teachers, young researchers, students, exporters, importers, extension specialists and those who are associated with temperate, tropical and subtropical fruit crops.



The book was formally released at ICAR-CISH, Lucknow on 4th July, 2023 by Dr. A.N. Mukhopadhyay, Former Vice-Chancellor AAU, Jorhat and a leading Plant Pathologist of India.

Momentous Events



Visit of Hon'ble Union Minister of Agriculture Shri Narendra Singh Tomar ji and Maharashtra State Agriculture Minister Shri Abdul Sattar ji to ICAR-CCRI Stall at VAMNICOM Exhibition held on 1st November, 2022 in Pune, Maharashtra



July to December, 2022

Shri Nitin Gadkari Ji, Hon'ble Union Minister for Road Transport and Highway and Dr. Dilip Ghosh, Director, ICAR-CCRI during Inauguration of Outreach Programme on "Export Potential for Agri Crops, Fruits & Vegetables" on 10th September, 2022



Dr. Himanshu Pathak, Secretary (DARE) and Director General (ICAR) Interacting with ICAR Scientists



Signing of MoU for Technology Licensing between ICAR-CCRI, Nagpur and M/s Shivar Nursery on 5th December, 2022



Celebration of 38th Foundation Day of ICAR-CCRI on 28th July, 2022



Signing of MoU for Academic Collaboration between ICAR-CCRI, Nagpur and SKUAT-Jammu on 19th October, 2022



Tree Plantation Drive at Panchwati Vridhashram (Old Age Home) and Shradhanand Anathalay (Orphanage) on 12th August, 2022



Celebration of Independence Day at ICAR-CCRI



ICAR-CCRI Exhibition Stall in AGROVISION-2022

Address by Dr. Dilip Ghosh, Director, ICAR-CCRI in AGROVISION-2022



Signing of MoU for Collaborative Research between ICAR-CCRI, Nagpur and IIT – Roorkee in 2022

January to June, 2023



Dr. Dilip Ghosh, Director, ICAR-CCRI Presenting and Achievements and Future Plan of the Institute before Hon'ble Union Minister of Agriculture Shri Narendra Singh Tomar ji on 26th May, 2023



Dr. Dilip Ghosh, Director, ICAR-CCRI interacting with Shri Nitin Gadkari Ji, Hon'ble Union Minister for Road Transport and Highways on 25th January, 2023



Foreign Delegates from Asian Development Bank Visiting Citrus Nursery at ICAR-CCRI



Participation of ICAR-CCRI at 108th Indian Science Congress from 6th to 8th January, 2023



Dr. Dilip Ghosh, Director, ICAR-CCRI Interacting with Foreign Delegation and NHB Officials on Clean Plant Programme on 25th January, 2023 at ICAR-CCRI



Signing of MoU for Technology Licensing between ICAR-CCRI, Nagpur and Jain Irrigation Systems Ltd. on 19th April, 2023



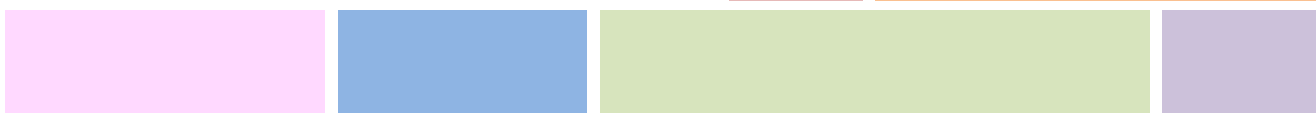
Signing of MoU for Collaborative Outreach between ICAR-CCRI, Nagpur and ICAR-ATARI, Kolkata on 7th May, 2023



Participation of ICAR-CCRI in the Workshop on 'Implementable Technologies for Chotanagpur Plateau Region of Eastern India', Organized by ICAR-ATARI, Kolkata from 22nd to 23rd May, 2023



Signing of MoU for Academic Collaboration between ICAR-CCRI, Nagpur and UBKV, West Bengal on 11th May, 2023



Glimpses of Outreach Activities by ICAR-CCRI in Different Parts of the Country

Achievements of ISC Members



Visit of Dr. Dilip Ghosh, Director, ICAR-CCRI to USA under Clean Plant Programme from 11th to 17th June, 2023



Dr. Dilip Ghosh, Director, ICAR-CCRI Receiving Dr. R. S. Paroda Award Conferred by CHAI



Dr. N. Vijayakumari, Principal Scientist, ICAR-CCRI with Other Delegates in International Citrus Congress – 2022 held in Turkey



Dr. A. A. Murkute, Principal Scientist, ICAR-CCRI assumed the Charge of Director, Mahatma Gandhi Institute of Rural Industrialization, Ministry of MSME, Wardha, Maharashtra.



Dr. Dinesh Kumar, Principal Scientist, ICAR-CCRI assumed the Charge of Head, Division of Food Science and Post Harvest Technology, ICAR-Indian Agricultural Research Institute, New Delhi



Dr. G. T. Behere, Principal Scientist, ICAR-CCRI assumed the Charge of Head, Division of Crop Protection, ICAR-Central Institute of Cotton Research, Nagpur, Maharashtra.



US Patent Granted on “Compositions and Methods for systemic delivery of cargos in vascular plants” (Patent No. 11229608), **Inventors:** Swadeshmukul Santra, Ali Ozcan, Dilip K. Ghosh, Sunil Kokane, Pranav Kumar and Ashwani Kumar Sharma

Indian Patent Granted on “DNA based diagnostics for identification of citrus rootstock cultivars” (Patent No. 407695), **Inventors:** Aswath C., Lakshmana Reddy D.C., A. K. Das, N. Vijayakumari, I. P. Singh and V. J. Shivankar

Indian Patent Granted on “Sparkling clear ready-to-serve beverage” (Patent No. 370936), **Inventors:** Dinesh Kumar, Milind Shivratn Ladaniya, Thiruvoth Uchambally and Sunil Kumar

Indian Society of Citriculture

Indian Society of Citriculture (ISC) was established on 24th August 1983 in New Delhi (Registration No. S/13760 of 1983). Later, the Headquarter of the Society was shifted from IARI, New Delhi to ICAR-Central Citrus Research Institute, Nagpur, Maharashtra (Formerly known as National Research Centre for Citrus) in November 1997 to boost its activities and pursue the objectives more effectively, being placed in hub of citrus industry. Since its inception, the ISC brings together researchers, academics, citrus growers, industry experts, and policymakers from across India. The ISC works closely with the government and other stakeholders to address the challenges faced by the Indian citrus industry. Overall, ISC plays a crucial role in advancing the citrus industry in India and ensuring its long-term sustainability and profitability. Its efforts are helping to promote citrus cultivation as a significant contributor to India's horticultural sector and to the country's economy as a whole.

Objectives of the Society

1. To promote and advance the research in horticulture with special reference to citrus and development work being done in different countries.
2. To promote the development of a sound Citrus Industry in India.
3. To carry out researches on Citriculture in accordance with accepted modern principles.
4. To publish original research works on Citriculture carried out anywhere.
5. To establish and maintain liaison with well-known libraries relating to relevant and allied subjects.
6. To establish regional offices at convenient centers in India to promote the activities of the society.
7. To organize seminars, exhibitions, lectures, training programmes etc. to promote the development of a sound Citrus Industry in India

Membership: ISC membership is open to persons actively engaged in citrus research, teaching and extension including the organizations/institutions dealing with citrus and allied subjects related to citrus industry.

Classification of Members

Life membership: All the persons who pay the specified fee for the class of life membership. The life membership fee may also be paid through installments (maximum two installments within the calendar year.)

Patron: An individual who contributes the specified or more amount to the society once.

Other membership: Non-profitable educational institutions / Research organizations, NGOs who become members by paying specified amount once.

Corporate membership: Any company/firm who subscribes to the society only once.

Membership Fee

Membership Category	Membership Fee
Patron	Rs. 25000.00
Life Member (India)	Rs. 5000.00
Life Member (Indian Students limited to 5 years)	Rs. 2500.00

Membership Category	Membership Fee
Life Member (SAARC Countries)	Rs. 10000.00
Life Member (Other than SAARC Countries)	USD 200.00
Corporate Member	Rs. 50000.00
Others	Rs. 30000.00

Bank Account details	QR code for UPI Payment	Mode of Payment
Account Name: Indian Society of Citriculture Account Number: 10199461448 Bank Name: State Bank of India Branch: Ravinagar, Nagpur IFSC code: SBIN0007504 PAN: AACAI5124Q		The membership fee (non-refundable) can be paid through internet banking/UPI/Demand draft (drawn in favour of "Indian Society of Citriculture" payable at Nagpur).

INDIAN SOCIETY OF CITRICULTURE

ICAR - CENTRAL CITRUS RESEARCH INSTITUTE

AMRAVATI ROAD, NAGPUR - 440 033, MAHARASHTRA, INDIA



MEMBERSHIP APPLICATION FORM

1. Name : _____
2. Correspondence Address : _____
(Complete address including pin code)
3. Permanent Address : _____
(Complete address including pin code)
4. E-mail : _____
5. Mobile No. : _____
6. Highest Degree : _____
7. Field of Specialization : _____
8. Nationality : _____
9. Date of Birth : _____
10. Membership Category : *Please put tick mark (✓) on appropriate check box*

Patron	<input type="checkbox"/>	Life Member (India)	<input type="checkbox"/>	Life Member (SAARC Countries)	<input type="checkbox"/>
Life Member (Other than SAARC Countries)	<input type="checkbox"/>	Life Member (Indian Students limited to 5 years)	<input type="checkbox"/>	Corporate Member	<input type="checkbox"/>
Others (Please specify your occupation _____)					<input type="checkbox"/>
11. Payment Details of Membership Fee : Transaction Ref./ID: _____ Date: _____
Demand Draft No.: _____ Date: _____

Date:
Place:

Signature

Please send the Membership Form to "The Secretary, Indian Society of Citriculture, ICAR-Central Citrus Research Institute, Amravati Road, Nagpur – 440 033, Maharashtra, India" by speed post/registered post/courier service OR **Email the Scanned Copy of the Signed Application Form to** info@iscindia.org.in and iscCRI@gmail.com

Indian Society of Citriculture

Hqs: ICAR-CCRI, Amravati Road, Nagpur - 440033, Maharashtra



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Nagpur, Maharashtra

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Asian Citrus Congress - 2023

Advancing Citriculture for Agro-economic Prosperity

Organized by

28-30 October, 2023
Nagpur, Maharashtra, India



Indian Society of Citriculture
Nagpur, Maharashtra, India



In Association with



ICAR - Central Citrus Research Institute
Nagpur, Maharashtra, India



**Asia-Pacific Association of
Agricultural Research Institutions**
Bangkok, Thailand



**Korean Society for Citrus and
Subtropical Climate Fruits**
Jeju City, South Korea



Ceremonial Launching of ACC-2023
Website by Shri Nitin Gadkari Ji,
Hon'ble Union Minister for Road
Transport and Highways, GoI

For more information, please visit:
<https://accindia2023.iscindia.org.in>