



**STUDY OF MICROBIAL GROWTH AND SPOILAGE FOUND IN THE CITRUS
FRUIT: A REVIEW****DESHPANDE VR¹, UPADHAYAY D², ANDHARE P² AND PRAJAPATI P^{2*}****1:** PG student M.sc (Microbiology), Department of Microbiology, Parul Institute of Applied Science, Parul University, Po-Limda 391760, Ta- Waghodia, Dis- Vadodara**2:** Assistant Professor, Department of Microbiology, Parul Institute of Applied Science, Parul University, Po-Limda 391760, Ta- Waghodia, Dis- Vadodara***Corresponding Author: Dr. Priyanka Prajapati; E Mail: priyanka.mistry82141@paruluniversity.ac.in;****Tel: +919913562946****Received 24th Jan. 2021; Revised 27th Feb. 2021; Accepted 28th March 2021; Available online 1st April 2021****<https://doi.org/10.31032/IJBPAS/2021/10.4.1057>****ABSTRACT**

Citrus fruits, both raw and juiced, are commonly consumed in India, especially during the summer season. Fruit juices and fruits products mainly lemon and orange are introduced like gummies, candies, Jams are commercially advertised for the consumer. These citrus fruits are mostly consumed raw because of their nutritional and vitamin content. When these fruits like orange and Indian gooseberry are consumed raw, they may be infected by bacteria or by fungi. Bacteria can be found from leaves, fruits, small stem and buds of citrus fruits. The most common bacteria found from the citrus fruit surface or juices are *Salmonella*, *E. coli*, *Pseudomonas*, *Lactobacillus* and *Klebsiella spp.* The high amount of fecal form of *E. coli* and *Salmonella* were also isolated from the orange juice preparing machine. There can be presence of mould forming fungi on the outer form of the citrus fruits like *Blue-green mould* this fruit if consumed can form hazardous effect on human health. The spoilage can form watery spot, false odour, powdery formation on surface by the fungi these can also spread other citrus fruits when there is bad handling during packaging. The bacterial growth on the outer part or into the citrus fruit juice may come during the farming through the seeds, during plucking, fruit packaging.

Keywords: *Blue-Green mould, Citrus fruits, Fruit spoilage, E. coli, Salmonella*

INTRODUCTION

History of citrus fruits:

From the ancient time citrus fruit is good source of nutrients, and they also have many medicinal properties [1]. The sweet orange (Malta), also known as sangarta, is a mixture of oranges that is high source of vitamin C. This red blood fruit was not only tasty and juicy but was also most consumed from other Malta varieties [2]. Citrus peels are high in nutrients and contain a variety of phytochemicals; they also used in medicines or dietary supplements. The pH of lemon and lime juices is 2.2–2.4 and 1.8–2.0, respectively [3]. *Kinnow mandarin*, a hybrid of *Citrus nobilis* and *C. deliciosa*, had been one of Punjab's main crops. It also accounts for 45 percent of all fruit production [4]. In Thailand, the lime tree (*Citrus aurantifolia* Swingle) most common commercially grown fruit tree. While growing these limes the problem comes when there is growth of *Xanthomonas citri* spp, this disease is also called citrus canker in the initial weeks there is formation of brown spots and corky appearance on the surface of lime. Citrus canker lesions on fruit and stems are 1 mm deep and look similar to canker lesions on leaves and soil [5]. This disease has caused

the economic loss because of the quality of lemon [6].

Nutritional content of citrus fruits:

Citrus peels are rich in nutrients and phytochemicals including and-sitosterol, glycosides, and volatile oils. Peptic ulcers, eye infections, gum disease, gout, skin treatment, piles, and other conditions benefit greatly from these peels. Citrus limon dried fruit peel has been checked for antimicrobial activity. Alkaloids, saponin, sterols, and terpenoids are phytoconstituents contained in plants that provide an exciting set of circumstances that allow for more rigorous modern therapies against a wide range of microorganisms. This peel was successful against Gram-positive *S. aureus* and Gram-negative *E. coli*, as well as fungi such as *C. albicans* [7].

Leaves, roots, and fruit become immune to infection when they grow unless they are injured. Within the first six weeks of growth, almost all infections begin on the leaves and stems. Lemon fruits are an important predictor during an infection. Since the fruits are still young, the infection on the fruit is most likely to occur after the petal falls [8]. In terms of food poisoning species, frozen citrus fruits are obviously not a public health danger. Frozen citrus fruits, such as oranges, are healthy to eat

because they avoid the growth of bacteria as well as mold-forming fungi. Despite good sanitary practises, coliform bacteria were discovered on the orange surface. These coliform bacteria are an indication that no human hepatitis is present [9]. Postharvest fungal diseases on fruit have gotten more attention from researchers in a variety of fields, especially horticulture, plant health, and food science. Biocontrol agents, natural compounds, UV, hot water treatment, and salts are among the most popular nonchemical treatments for postharvest fungal diseases on fruit. Researchers have used transgenesis or genome editing to achieve their goals [10].

Citrus fruit Spoilage:

Spoilage can be found at any point in the food chain as a result of physical damage, the action of indigenous enzymes in animal or plant tissue, or microbial infection. Some microbes need oxygen to expand, while others are killed by it, and still others are facultative. During storage and packing, handling at temperatures below ambient temperature can delay or prevent the growth of certain microbes. The noxious odour generated by microbes in spoiled fruits is used to repel large animals and maintain a food supply for them. Pathogens can infect a wide range of plant tissues, with the leaf being one of the most

common. Some of the microbes like *E. coli* and *Xanthomonas* as found on the surface of lemon or orange during packaging process [11].

Microbial growth can be seen in seed or crop growth in the field, as well as post-harvesting and harvesting handling, distribution, and storage. On all the surface of fruits *Salmonella* survival was statistically distinct, with compared to orange and lime. fruit juices do have high content of water and moisture which are mostly favourable for the growth of many microbes. The spoilage of fruits is mainly caused by the bacteria. The other most common bacteria are *Lactobacillus* these are acid tolerance as they can survive in the citrus fruits these bacteria are rod shaped and can survive in 5.5 to 5.8 pH [12].

Fruits become poisonous and less palable when they are spoiled by fungi, which can also affect the taste of fruits or fruits products. Fruit–pathogen interactions have recently received a lot of attention, and we've learned a lot about fungal pathogenicity and fruit resistance as a result. Citrus fruits are eaten raw as well as processed into candies, jams, and juices. Flavonoids, steroids, and alkaloids present in citrus fruits have antimicrobial properties. *Pseudomonas spp*, were the most common bacteria present in the citrus fruits out of all the samples [13].

The spoilage of fruits may cause change in smell, colour and appearance. Fruit spoilage is mainly caused by bacteria. [14]. Fruits have a high moisture content, which is conducive to the growth of many microorganisms. The key cause of fruit spoilage is an extrinsic factor: improper handling and pre-harvesting. *Aspergillus spp.* is primarily responsible for the rotting or spoilage of oranges [15]. The samples of citrus fruits contain a large number of microorganisms. The identification of pathogenic and indicator bacteria such as *Klebsiella spp.*, *Listeria spp.*, *Staphylococcus spp.*, and *Vibrio spp.* is near 107 cfu/g from the sample, which is a major source of concern for buyers [16].

Fungal infection in citrus fruits:

The detection of potential mycotoxins in fruits infected with postharvest fungal pathogens is important for quality control and food protection. Toxigenic fungi are isolated from rotten fruits. *Collectotrichum Glocosporoides*, a fungal infection caused by *Phytophthora spp.*, may infect during the pre-harvesting period. Certain moulds can produce mycotoxins and it is well known for the danger it poses to human and animal health around the world [17]. Fungi such as blue and green mould cause a soft, watery spot on the fruit surface are seen like from Indian gooseberry. These bacteria

and fungi are harmful to human health and can cause pneumonia, urinary tract infections, and food poisoning. These moulds can infiltrate tissue and have an impact on plant development, fruit production, and photosynthesis formation. These fungal can grow on the surface of fruit, branches and leaves [18]. Sooty mould produces a velvety layer of black fungal growth on the surface of leaves, twigs, and flowers, primarily in Indian gooseberry. Some of the dangerous *penicillium* fungi, such as *P. Citrinum*, which grow on amla, and *P.expansum*, produce mycelia that are harmful to human health, causing necrotizing esophagitis disease and asthma. Fruit juices like orange and sweet lime have pH ranges of 4.34 and 5.08, respectively. Mould growth was also isolated from juices, including *Aspergillus Niger*, *Aspergillus flavus*, and *Penicillium digitatum*. Lactic acid bacteria prefer a pH range of 2.9-3.5, while enteric bacteria prefer a pH range of 3.6-4.5 [19]. If a sweet orange gets spoiled, changes in hue, or tastes bad, it may be harmful to human health. Since spoiled fruit contains fungal such as *Aspergillus fumigatus*, *Aspergillus niger*, and *Aspergillus flavus*, it should be handled carefully and kept refrigerated.

Huanglongbing HLB (citrus greening):

Citrus greening is the most serious threat to the citrus industry in the United States, and a prime example of how invasive species can destabilise an industry. Given the fact that HLB (huanglongbing), also known as Citrus greening, has been known in China for over a century, the disease's bacterial caused by *Xanthomonas citri* was only discovered in the 1970s. This citrus greening can cause serious disease to many fruits like orange, mandarins and grapefruits. There are no citrus varieties that are resistant to HLB, and the disease has yet to be eradicated from any region [20].

Citrus cultivation is continuously threatened by pathogenic bacteria, resulting in financial loss. Transgenic and genome-edited plants have attracted a lot of attention as a way to improve plant resistance to pathogenic fungi and bacteria. Citrus canker (CCK), citrus variegated chlorosis (CVC), and Huanglongbing (HLB) are three common diseases that reduce citrus fruit production. Citrus fruits should not be produced in any infected areas because of these diseases. The *Xanthomonas citri* has left a yellow-brown mark on the lemon [21].

Xylella fastidiosa, a bacterial citrus pathogen that causes citrus variegated chlorosis (CVC), was the first plant

pathogen to have its entire genome sequenced by Brazilian scientists from over 200 national laboratories.

Candidatus Liberibacter asiaticus (CLas) is spread by the phloem-feeding insect *Diaphorina citri*, and there is no established genetic resistance in the Citrus genera. Incubation cycles in citrus seedlings and nursery plants, according to Canale *et al.*, range from 2.5 to 5 months [22]. As a result, farmers have experienced setbacks, and the whole industry has shrunk. Growers in Florida have tried a variety of strategies to combat the disease and its vector, including increased foliar nutritional applications, extensive insecticide uses to suppress the ACP, antibiotics, a cooperative area-wide pest management programme, and thermotherapy of diseased trees, with little, if any, success. The Florida citrus industry's strategic plan to combat HLB included a voluntary area-wide management programme for psyllid control. In this study, *B. subtilis* LE24, *B. Amyloliquefaciens* LE109, and *B. Tequilensis* PO80, endophytic bacteria isolated from healthy citrus plants, were found to inhibit the growth of *X. citri* subsp. *citri*. One of the benefits of endophytic bacteria is their ability to replicate inside the host plant tissues. Bacillus strains are thus important in the

biological management of citrus canker disease on leaves [23].

The method found for the identification number of bacteria in the citrus fruits are the counting bacteria in Cfu unit of the bacteria or fungi and if heat is provided to the fruits still the presence of fungal or bacteria are seen or not. A dual culture technique was used to test the ability of endophytic bacteria to inhibit *X. citri subsp. Citri*. Homogenisation was used to remove DNA from the selected bacteria. The extraction of crude bioactive compound was done for the endophytic bacteria.

Computational methods play a key role in the automated detection and early diagnosis of diseases in citrus fruits, which helps to improve growth and efficiency. To design a diagnostic system to classify diseases in citrus plants, technique as image processing such as pre-processing, image segmentation, features extraction, and classification are used. The diseased regions in leaves can be segmented using the k-means clustering method, and colour and texture features can be extracted. To choose the most appropriate features, the ANOVA F-test is used [24].

CONCLUSION

Citrus fruit juices are often consumed during the winter season and are prepared in companies and sold as well as prepared

by small-scale vendors. Bacteria infect these fruits through the soil, during packaging, and during plucking. The most of the spoilage in citrus fruit is caused by the bacteria. The bacteria found may be gram + and also gram -. Citrus Fruit that are infected on the surface, such as *Blue-green mould*, may also infect other fruits during packaging. Indicator bacteria, such as *E. coli* and *Staphylococcus*, signify the existence of pathogenic bacteria.

Faecal form of *E. coli* was also discovered on the orange fruit juice preparation unit. The most hazardous citrus disease which is citrus greening which have no cure can also be studied further to understand more. From which type of citrus fruits do bacteria have found and are they hazardous for human health can be studied further. The type of illness or disease caused by citrus fruits can also cause death because of the fungal infection caused on the citrus fruit these fungi can also form mycelium which when entered into the human body and can penetrate their mycelium into organs.

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