EFFECT OF ROOTSTOCK ON CITRUS FRUIT QUALITY. A REVIEW

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Abstract

Citrus rootstocks are a relevant tree part that contributes to crops adapting to biotic and abiotic conditions, it becoming a key factor to face the current era of climate change. Although the emphasis of most studies on rootstocks has been placed on the yield and optimization of the citrus fruit grown in different environments, other studies have paid attention to the effect of rootstock on fruit quality. Hence the quality of citrus fruit is becoming increasingly more relevant as consumers demand high internal and external qualities to eat citrus fruit fresh. To better understand how rootstock influences citrus fruit quality, the literature that focuses on physico-chemical parameters, nutritional compounds and physiological disorders has been revised. This review points out the influence that the external aspects surrounding plants has on fruit quality, such as the rootstock/scion interaction, the water uptake capacity of roots, the modification of the photosynthetic rate or availability of nutrients minerals.

Keywords. Nutritional quality, sugars, organic acids, vitamin C, phenolic compound, volatiles, fruit color, rind thickness, peel disorders, juice content

Introduction

Rootstock plays a very important role in commercial citrus production. Selecting rootstock acts as an important tool to adapt crops to adverse environmental conditions, or to biotic (diseases, insect-pests) and abiotic (drought, salinity, water logging, alkalinity, cold) stresses.[1,2] Optimum rootstock selection is crucial as the scion/rootstock interaction influences tree vigor, increases yield, reduces the juvenile period or enhances fruit quality characteristics. These conditions also vary depending on each specific region.[3-5] The effect of rootstock on cultivar performance has been reported to be due mainly to water relations, mineral nutrition and hormonal contribution.[4,6]

The aim of rootstock breeding programs worldwide is to diversify scion-rootstock combinations to help to increase yields and fruit quality, and to extend the harvest season. Therefore, new rootstocks that are tolerant to salinity, iron chlorosis, water stress and flooding conditions, or resist diseases such as citrus tristeza virus (CTV), are being developed.[7-10] In addition, rootstock is key for facing new citriculture challenges, such as optimizing crops in organic farming vs. conventional farming to respect the environment more or to cope with new diseases appearing, such as huanglongbing disease, which has emerged in recent years.[11,12] On the other hand the rootstock becomes a key factor to face the current era of climate change. Global climate changes affect water availability and increase in temperature and are responsible for drought and heat stress which alter the plant morphology, physiology and genetic expression of plants. In this context the evaluation of suitable rootstocks with ability to adapt to changing climate is being recently addressed in different crops including citrus fruit.[13-15]

Citrus rootstocks are selected because they are naturally-occurred species ('Rough' lemon (*Citrus jambhiri* Lush.), 'Cleopatra' mandarin (*Citrus reshni* Hort ex. Tanaka), 'Rangpur' lime (*Citrus limonia* [L.] Osbeck), 'Trifoliate' orange (*Poncirus Trifoliata* [L.] Raf.) or 'Sweet' orange (*Citrus sinensis* [L.] Osbeck)) or hybrids from controlled crosses to view the worldwide use as citrus rootstocks ('Carrizo' citrange (*Citrus sinensis* [L.] Osbeck x *Poncirus trifoliata* [L.] Raf.), 'Troyer' citrange (*Citrus sinensis* [L.] Osbeck X *Poncirus Trifoliata* [L.] Raf.), Macrophylla (*Citrus macrophylla* wester) or 'Swingle' Citrumelo (*Citrus paradise* x *Poncirus trifoliata*). [16] Of these rootstocks, 'Carrizo' citrange is the most widely used in the main citrus-producing countries like Spain, South Africa, Australia, California and Italy. [17] However, 'Trifoliate' orange is preferred in other countries like Japan, China, Argentina and Uruguay, and those with cold environments. [18]

The influence of rootstock on fruit quality has been widely addressed by different authors. Nevertheless, the most studies are focused on a specific cultivar grafted onto different rootstocks, where the rootstocks are selected mainly depending on the relevance they have in the geographical area of study. For this reason, a compilation of the results of different variables (metabolism compounds, physicochemical properties and the incidence of physiological disorders) are needed to understand the effect of rootstock on fruit quality, and this information can be useful for selecting the optimum scion/rootstock combination to meet consumers' product quality demands.

This paper reviews information on the effect of rootstocks on the main fruit quality parameters, and the factors that may cause these effects.

External Color

Fruit coloration is one of the most important attributes to determine citrus fruit quality. In citrus peel, color is due to the accumulation of three main classes of pigments: chlorophylls, carotenoids and anthocyanins. [19] Both content and composition in rind and pulp are specific of different varieties. [19,20]

Many studies have addressed the effect of rootstock on citrus fruit color. Machado et al. [21] found that rootstock impacted color development in 'Ruby Red' grapefruit (*Citrus paradisi* Macf.). Indeed, while fruit grafted onto 'Cleopatra' mandarin developed a yellowish external color, fruit from 'Swingle' citrumelo displayed a pinkish peel color. However, these differences were observed only at the beginning of the season. In the same study, rootstock was seen to not affect 'Star Ruby' grapefruit (*Citrus paradisi* Macf.). color. More recently, Emmanoulidou and Kyriacou [22] reported that the differences observed in the color of the 'Lane Late' (*Citrus sinensis* [L.] Osbeck) and 'Delta' (*Citrus sinensis* [L.] Osbeck) cultivars grafted onto five different rootstocks were associated with maturity state, and not with the effect of rootstock.

The interaction between rootstock and scion can modify the effect of rootstock on the external fruit color. The dwarfing hybrid rootstock Forner-Alcaide 418 ((*Citrus sinensis* [L.] Osbeck x *Poncirus trifoliata* [L.] Raf.) x *Citrus deliciosa* Ten. & Pasq.)) has been reported to induce the lower color index in 'Navel' orange (*Citrus sinensis* [L.] Osbeck var. *brasiliensis Tanaka*) scion than the rootstocks 'Carrizo' citrange, and other dwarfing hybrid rootstock (Forner-Alcaide 517 (*Citrus nobilis* Lour. x *Poncirus Trifoliata*)). [23] Nevertheless, Forner-Alcaide 418 induced the highest external color index when evaluating three different lemon cultivars ('Fino 49' *(Citrus limon* [L.] Osbeck), 'Fino de Elche' *(Citrus limon* [L.] Osbeck), and 'Verna 50' (*Citrus limon* [L.] Burm) (grafted onto three rootstocks (Forner-Alcaide 5 (*Citrus reshni* Hort. Ex Tan. X

Poncirus trifoliata [L.] Raf.), Forner-Alcaide 418 and Forner-Alcaide 2324 (*Citrus sinensis* [L.] Osbeck x *Poncirus trifoliata* [L.] Raf.) x *Citrus reshni* Hort. ex Tan.)). [24]

The effect of rootstock in the citrus coloration has been related with the content of different components or with external conditions (Table 1).

Table 1. Effect of rootstock on external color of citrus fruits.

Effect of rootstock	**Reference**
Full Coloration	
Pigments content	*[10]; [25]*
Color Breakdown	
Abiotic stress	*[26]; [27]*
Increase Sucrose content	
Reduced Nitrogen content	*[28]; [29]*
	[30]

This difference in citrus peel color could be due to the influence of rootstock on the accumulation of pigments, which has also been addressed in different studies. With ‘Owari’ mandarin (*Citrus unshui* Marc.) grafted onto two rootstocks, fruit budded onto ‘Cleopatra’ mandarin rootstock had higher levels of carotenoids β-cryptoxanthin and violaxanthin (in flavedo and albedo tissue) than those grafted onto ‘Troyer’ citrange. [25] Although most studies have reported the effect of rootstock on external color, some authors have also evaluated this effect on internal color, above all in blood orange cultivars. Continella et al. [10] analyzed the effect of eight rootstocks on ‘Tarocco Scirè’ sweet blood orange (*Citrus sinensis* [L.] Osbeck). They reported that the fruit budded onto the hybrid rootstock ‘Bitters’ (*Citrus sunki* x *Poncirus Trifoliata*) had the highest external color, while fruit grafted onto ‘Swingle’ citrumelo had the lowest. Moreover, the anthocyanin content measured in juice followed the same pattern. Conversely, Incesu et al. [31] observed that the influence of six rootstocks on ‘Moro blood’ (*Citrus sinensis* [L.] Osbeck) oranges was different between peel and pulp color because red color rind was higher in the fruits grafted onto ‘Carrizo’ and ‘Troyer’ citranges, while the fruit budded onto rootstocks Yuzu (*Citrus junos*) and ‘Cleopatra’ mandarin had the highest color in pulp.

Abiotic conditions can also influence the effect of rootstock on citrus color. While comparing the performance of ‘Clemenules’ (*Citrus clementina* Hort. ex Tan.) mandarin grafted onto ‘Carrizo’ citrange and ‘Cleopatra’ mandarin, Navarro et al. [26] found no differences in external fruit color. Nevertheless, the same study carried out under deficit irrigation conditions revealed that external coloration delay was more evident in the scion budded onto ‘Cleopatra’ mandarin fruits than in those on ‘Carrizo’ citrange. [27]

Color breakdown from green to orange is the result of chloroplast to chromoplast conversion, which involves chlorophyll degradation and carotenoid accumulation. [32,33] This change is known to be influenced by environmental and stress conditions, nutrient availability and hormonal action. Reduced nitrogen content and a rise in sucrose levels have been reported to promote color changes in the flavedo of citrus fruits. [34,35]

Rising sugar levels have been positively related to color break in citrus peel. [35,36] These observations are consistent with the known effects of soluble sugars, especially hexoses, on the down-regulation of the genes encoding chlorophyll and photosynthetic enzymes. [37,38] As indicated below in the “Sugars and Organic Acids” section, rootstock has a major effect on levels of sugars in citrus fruit. Legua et al. [28] reported that ‘Lane Late’ oranges budded onto ‘Cleopatra’ mandarin, which exhibited higher color index than the fruit budded onto Macrophylla and ‘Volkamer’ lemon (*Citrus Volkameriana* Ten. & Pasq.), also had higher sucrose contents in juice. Nevertheless in a study about ‘Navelina’ orange (*Citrus sinensis* [L.] Osbeck) grafted onto 14 rootstocks, fruit budded onto ‘Cleopatra’ mandarin exhibited the lowest external color but, in this case, fruit also gave the lowest TSS:TA ratio. [29] Despite the few studies available, it has been proved that rootstock can modify nitrogen uptake and translocation to the main sink (fruit) and, therefore, the amount in citrus peel. Hass [30] found different amounts of nitrogen in dry matter in ‘Valencia’ (*Citrus sinensis* [L.] Osbeck) orange peel as regards which rootstocks were grafted. The fruit from the scion grafted onto rootstocks ‘Sour’ orange (*Citrus aurantium* L.) and ‘Rough’ lemon had the highest total nitrogen content, while those from ‘Trifoliate’ orange had the smallest amount. However, no studies were found in which variations in nitrogen due to rootstock have been related directly to fruit color change.

Peel Thickness

Peel is the first fruit barrier against abiotic and biotic factors during pret and postharvest periods. Peel thickness is one of the most important factors involved in the peel disorders manifested during shipping and storage. [39] So peel thickness is a characteristic that is usually taken into account when studying the effect of rootstock on citrus fruit quality.

Table 2 shows the most relevant results of the effect of rootstock on rind thickness. Although the effect of the rootstocks on the peel thickness depends on the scion-rootstock interaction, it can be highlighted than in the most of the studies Macrophylla, ‘Volkamer’ lemon and ‘Rough' lemon induced the thickest peel, while rootstock ‘Cleopatra’ mandarin led to the thinnest peel.

Table 2. Effect of rootstock on rind thickness of citrus fruits.

Cultivar (Country)	Number of Rootstocks	Effect of rootstock		Reference
		Thickest	Thinnest	
Orange				
'Navelina' (Spain)	14	Forner-Alcaide 418	'Carrizo' citrange; 'Cleopatra' mandarin; Forner-Alcaide 13	[29]
'Olinda Valencia' 'Washington Navel' (Saudi Arabia)	9	Macrophylla 'Rough' lemon	'Cleopatra' mandarin; Citrus amblycarpa; 'Sour' Orange	[40]
'Lane Late' (Spain)	4	Macrophylla; 'Volkamer' lemon	Gou Tou Chen; 'Cleopatra' mandarin	[28]
'Queen' (Iran)	6	No differences		[41]
Mandarin				
'Nova' (Cyprus)	11	'Palest. Sweet' lime; 'Volkamer' lemon	'Troyer' citrange	[42]
'Nova' 'Robinson' (Turkey)	3	No differences		[43]
'Duong' (Vietnam)	3	'Matt' Orange	'Tau' lemon; 'Carrizo' citrange	[44]
Grapefruit				
'Piemonte' (Brazil)	14	'Volkamer' lemon	'Cleopatra' mandarin Sunki; Riverside; TSCK x CTSW-028	[45]
'Marsh' (Saudi Arabia)	7	Shaub; 'Rough' lemon	'Volkamer' lemon	[46]
'Rio Red' (Turkey)	7	1st Season 'Volkamer' lemon 2nd Season No differences	Calamondin; Smooth Flat Seville	[47]
'Henderson' (Turkey)	5	No differences		[48]
Lemon				
'Allen Eureka' (Saudi Arabia)	7	Macrophylla; 'Volkamer' lemon; 'Rough' lemon	'Cleopatra' mandarin; 'Swingle' citrumelo; Citrus amblycarpa; 'Sour' Orange	[39]
'Fino 49','Verna 50' 'Fino de Elche' (Spain)	3	Forner-Alcaide 2324	Forner-Alcaide 418	[24]
Lime				
'Mexican' (Saudi Arabia)	7	Shaub; 'Rough' lemon	'Cleopatra' mandarin	[46]
'Persa' (Mexico)	8	'Volkamer' lemon; Dulce de Palestina	Flying dragon	[49]

It is also noteworthy that biotic and abiotic factors can affect rind thickness (table 3).

Table 3. Influence of external factors on the effect of rootstock on rind thickness of citrus fruits.

Factor influencing thethe rootstock's effect	**Reference**
Season	[46,47]
Mineral uptake	[42, 50]
Endogenous hormone metabolism	[51]

Despite some studies offering repetitive results for different seasons, in other studies the differences found among rootstocks for one season were not observed for the following season, such as grapefruit 'Marsh' and 'Rio Red' (*Citrus paradisi* Macf). [46,47]

Citrus peel thickness is known to be highly dependent on mineral nutrition. Although it is known that rootstock influences nutrient uptake, [52,53] very few studies report the relation between the influence of rootstock on peel thickness and its nutritional composition. In 'Nova' mandarin (*Citrus clementina* Hort. ex Tan. X (*Citrus reticulata* Blanco x *Citrus paradisi* Macf)), this correlation was observed in the fruit from the scion budded onto rootstock 'Troyer' citrange which the thinnest peel and the tree leaves had the largest amounts of phosphorous and the least amounts of potassium. [42] Nevertheless, when Dubey and Sharma [50] compared four rootstocks for 'Kagzi Kalan' lemon, they found that the thickest peel was exhibited by the fruit from the trees grafted onto Karna Khatta (*Citrus Karna* Raf.), with the highest phosphorous content in leaves instead of the lowest as expected.

Recently, the influence of rootstock on the endogenous hormone metabolism that regulates peel thickness has been explored. [51] 'Kiyomi' tangor fruits had higher rind thickness values in the scion grafted onto *Citrus Junos* rootstock than in 'Trifoliate' orange, which coincided with the highest levels of IAA, GA3 and ZT hormones in rootstock *Citrus Junos*. These hormones correlated directly with increased peel thickness. Regarding hormone ABA, despite the amount in the cultivar budded onto

Citrus Junos was higher than in those budded onto ‘Trifoliate’ orange, no correlation with increased peel thickness was found.

Juice Content

Juice content is an important quality parameter for citrus fruit since a requirement for its commercialization. It has been reported that rootstock influences the juice content, which is associated to its effect on water uptake, peel thickness and granulation incidence (Table 4).

Table 4. Effect of rootstock on juice content on different cultivars associated to its effect on tree water status, peel thickness and granulation incidence.

	Cultivar	Rootstock		Reference
		Higher Juice Content	*Lower Juice Content*	
Tree Water Status				
	‘Valencia Tuxpan’ and ‘Pinneapple’ sweet orange		‘Rough’ lemon	[54]
	‘Valencia’ orange	‘Carrizo’ citrange	‘Rough’ lemon	[55]
Peel Thickness		*Thinnest peel*	*Thickest peel*	
	‘Clemenules’ mandarin	‘Cleopatra’ mandarin	‘Carrizo’ citrange	[56]
	‘Lane late’ and ‘Delta’ orange	‘Volkamer’ lemon		[22]
	‘Fino 49’ lemon	Forner-Alcaide 2324	Forner Alcaide 418	[24]
Granulation Incidence		*Lower disorder*	*Higher disorder*	
	‘Nagpur’ mandarin	Karna Khatta; ‘Rangpur’ lime and Gandharaj	Kumquat and ‘Rough’ lemon	[57]

The lower juice content reported for ‘Rough’ lemon in different varieties was associated with a lower juice osmotic potential. [54,55] Nevertheless, they pointed out that the effects of rootstock on fruit juice depended on annual climate variations and edaphic conditions

In the most of the studies, the thicker rind has been related with less juice content. [22,24,56] . Although it has been reported that thicker rind could predispose fruit to better postharvest performance, it also tended to negatively impact its juice content

We ought to take into account that juice content can lower due to disorders like granulation. Sau et al. [57] worked with 'Nagpur' mandarin (*Citrus reticulata* Blanco) and observed a negative correlation, more juice content and less granulation. The fruit budded onto Kumquat (*Fortunella* sps.) and 'Rough' lemon rootstocks obtained the lowest percentage of granulation and the highest juice content. In the same way, the fruit grafted onto Karna Khatta, 'Rangpur' lime and Gandharaj (*Citrus lemon* Burn.) rootsotcks exhibited the highest granulation incidence with the lowest juice content.

Sugars and Organic Acids

Many studies have addressed the effect of rootstock on sugars and acids, which are important factors that strongly influence the characteristic flavor of citrus fruit.

Although the effect on the content of sugars and acids depends on the rootstock/scion interaction, some rootstocks have been observed to exert the same effect on different cultivars. This happened with 'Sour' orange, which has been reported to increment total soluble solids (TSS) and titratable acidity (TA) levels in different citrus species grafted onto this rootstock by delaying commercial maturity compared to other rootstocks. McCollum et al. [58] studied the effect of different rootstocks on 'Marsh' grapefruit, and found the highest TSS content and acidity in the fruit grafted onto 'Sour' orange. In their study, the fruit grafted onto 'Carrizo' citrange had the lowest acidity, but the lowest TSS content was found in the fruit grafted onto 'Smooth Flat Seville' orange (*Citrus aurantium* putative hybrid). In 'Washington Navel' oranges (*Citrus sinensis* [L.]

Osbeck), Hifny et al. [59] obtained fruit with higher TSS and TA levels when they were grafted onto rootstock 'Sour' orange than onto rootstock 'Volkamer' lemon. More recently, this effect has been reported in a study about six rootstocks on 'Lane Late' and 'Delta' oranges. [22] In both cultivars, the fruit grafted onto rootstock Volkamer' lemon had the lowest TSS and TA contents, while the fruit grafted onto 'Sour' orange and 'Carrizo' citrange had the highest levels. This was also reflected in organic acids and individual sugars. Nevertheless, Bassal et al. [60] found different results when evaluating four rootstocks for 'Marisol' clementine (*Citrus reticulata* Blanco) as the fruits grafted onto 'Carrizo' citrange had higher TSS and TA than those from 'Sour' orange.

In some studies, although an effect of rootstock has been observed only on TSS, no differences in acidity content were detected. Hussain et al. [5] worked with 'Clementine' mandarin from nine rootstocks and found that the highest TSS values went to the fruit grafted onto 'Trifoliate' orange and the lowest values were for the fruit grafted onto 'Carrizo' citrange. However, these authors found no significant differences in acidity content. In 'Arrayana' mandarin (*Citrus reticulata* Blanco), grafted onto six different rootstocks, once again no effect was found in fruit acidity, but the influence of rootstock was strong on TSS content. [61] Likewise, in 'Ray Ruby' grapefruit (*Citrus paradisi* Macf.) grafted onto seven rootstocks, McCollum and Bowman [62] ran different assays and found that the scion grafted onto rootstock US-897 (*Citrus reticulata* Blanco x *Poncirus trifoliata* [L.] Raf.) had highest TSS content, but found no major differences in fruit acidity.

The influence of rootstock on sugars and acid content has been related to the inherent rootstock differences that affect plant water relations. These differences include root distribution, water uptake ability, hydraulic conductivity and leaf or stem water potentials. [63-65] Reflecting the plant water relation, in a study of 'Valencia' sweet oranges, it was reported that the fruit grafted onto ‘Carrizo’ citrange had more TSS than the fruit from ‘Rough’ lemon, which was explained by ‘Rough’ lemon experiencing less water stress than ‘Carrizo’ citrange due to the larger root distribution and greater hydraulic conductivity of ‘Rough’ lemon. [55]

The effect that the rootstock exert on the content of sugars and acids in fruit has also been linked with the differences that it induces in the leaf photosynthetic rates of scion and photosynthetic product distribution. [66] The better the scion-rootstock’s photosynthetic capacity, the more carbohydrate compounds transported from leaves to fruits. [67] Moreover, accumulation of sugars in fruit has been related to vascular resistance to sucrose transport at the rootstock’s budding union. So the reduced translocation of photoassimilates from leaves to roots limits root development, and also contributes to these compounds being more available in the scion, which results in increased carbon transport toward fruit. [23,68]

Phenolic Compounds

In recent years, more attention has been paid to the phenolic compounds of citrus fruits because many epidemiological studies have indicated that eating polyphenol-rich foods is associated with a reduced risk of cardiovascular diseases and certain cancer forms. It has been suggested that these compounds play an important role in the antioxidant capacity of citrus fruits. [69] Moreover, the presence of phenolics contributes to the sensory quality of fruit and juice through their effect on color, bitterness, astringency and flavor. [70] Among phenolic compounds, special attention has been paid to flavonoid compounds given their potential beneficial effects for human health.

In most species, abiotic stress induces the expression of flavonoid biosynthetic genes and their subsequent accumulation because flavonoids are involved in the regulation of environmental stress responses. [71,72]

To explain the effect of rootstock on phenolic content, different theories have been proposed and are based on tree vigor, water stress, uptake and the transport of water and minerals and mobile signals. [2,73-75] Tavarini et al. [73] reported that rootstocks affect internal fruit quality through the interaction of water with nutrient availability in soil, which may consecutively affect the synthesis of phytochemicals. Thus reduced water supply may also lead to high levels of phytochemicals, such as phenolic compounds.

In table 5, it has been listed the effect of rootstock on phenolic compounds, which is affected by tree water status and grafting union.

Table 5. Effect of rootstock on juice content on different cultivars associated to its effect on tree water status and grafting union.

	Cultivar	Reference
Tree Water Status		
Flooding soils	True-to-type rootstocks	[76]
Drought stress	'Valencia Delta' sweet orange	[77]
Drought stress and warm conditions	True-to-type rootstocks	[78]
Grafting Union		
	'Delta' and 'Lane late' oranges	[22]

In response to soil flooding, tolerant citrus rootstocks have been reported to have more flavonoids in leaves than sensitive ones, [76] which apparently constitutes an adaptive response.[79] Souza et al. [77] evaluated the interaction of 'Valencia Delta' sweet orange (*Citrus sinensis* [L.] Osbeck) with two rootstocks that displayed different tolerances to drought stress. These authors found that the fruit grafted onto the tolerant rootstock to long drought stress had flavonoid biosynthesis pathway genes that were overexpressed. The effect of being able to tolerate combined heat and drought conditions has also been studied in true-to-type ‘Cleopatra’ mandarin and ‘Carrizo’ citrange’ rootstocks, where ‘Carrizo’ citrange was selected as being tolerant and ‘Cleopatra’ mandarin was the sensitive rootstock. Leaves from the scion grafted onto rootstock ‘Cleopatra’ mandarin had greater activation of flavonoid biosynthesis, which could mitigate the greater oxidative damage observed in this genotype. [78]

There have also been reports that phenolic metabolism can be modified by rootstock due to either wounding or stress inflicted by grafting and graft union formation. [80] Emmanouilidou and Kyriacou [22] evaluating the performance of oranges ‘Delta’ and ‘Lane Late’ grafted onto six rootstocks, found that phenolic content was higher in fruit grafted onto the least compatible Gou Tou Cheng (*Citrus aurantium* [L.]) and lower in the highly productive Volkamer’ lemon.

The effect of rootstock on the main individual citrus flavonoids has also been recorded. [25,81] In general, it has been reported that the dominant flavanone glycosides in Sweet oranges are hesperidin and narirutin, whereas in ‘Sour’ orange the two predominant flavanone glycosides are neohesperidin and naringin as reflected in different studies. [82] In this sense, Hemmati et al. [83] studied four orange scions grafted onto four rootstocks, and found that the highest hesperidin content was in the peel of the four scions grafted onto rootstock Shelmahalleh (*Citrus sinensis* var. shelmahalleh), followed by ‘Swingle’ citrumelo. Nevertheless, the same study indicated that the highest naringin content was detected in the peel and pulp of all the cultivars grafted onto ‘Sour’ orange. Another study addressed the effect of six rootstocks on ‘Kinnow’ mandarin (*Citrus nobilis* Lour. x *Citrus deliciosa* Ten. & Pasq.). [84] ‘Sour’ orange induced the largest amount in flavanones (hesperidin, naringin, narirutin, naringenin and neoeriocitrin) and dihydroxy-B-flavanols (rutin and quercetin). Moreover, in the same study, ‘Rough’ lemon-2 had a suppressing effect on large amounts of phenolic compounds on the scions grafted onto it, which led to a marked reduction in these metabolites compared to other rootstocks.

Among the individual flavonoids, naringenin chalcone is considered necessary for the synthesis of flavonoids. Babazadeh-Darjazi [85] reported how fruit grafted onto Flying dragon (*Poncirus trifoliata* [L.] Raf. var. *monstruosa*) rootstock had higher levels of naringenin and total flavonoids than five other studied rootstocks. These authors assumed a specialized function for this molecule that could be better performed by Flying dragon. In 'Daisy' mandarins (*Citrus reticalata* Blanco), Feng et al. [86] observed how the highest naringenin content was found in fruit grafted onto 'Rough' lemon when referring to other rootstocks like 'Carrizo' citrange and 'Trifoliate' orange. Hesperitin and eriodyctol were also present in large amounts in fruit grafted onto 'Rough' lemon, as explained by these compounds deriving from naringenin. However, another study conducted in two grapefruit cultivars revealed that while 'Marsh Seedless' grapefruit (*Citrus paradisi* Macf.) grafted onto 'Sour' orange had the highest total phenol content, the highest naringenin content was found in the fruit grafted onto 'Rough' lemon fruit. [87]

Vitamin C

Vitamin C is considered one of the most important nutrients in citrus fruit, and is a water-soluble antioxidative component and an excellent reducing agent. [88] Vitamin C content in citrus sp. depends on species and cultivar, [89,90] but also on the other factors like maturity stage, climate and other different agronomic factors. [91,92] It has been stated that rootstock affects vitamin C production in citrus fruit, as reported by Magwaza et al. [93], who collected relevant data about the preharvest factor affecting vitamin C.

Some of the most relevant studies to have addressed the effect of rootstock on vitamin C content in citrus are shown in Table 6.

Table 6. Effect of rootstock in vitamin C in citrus fruit juice.

Cultivar (Country)	Number of rootstocks	Effect of rootstock		Reference
		Highest Vit. C	Lowest Vit. C	
Orange				
'Washington Navel' (Egypt)	2	'Sour' orange	'Volkamer' lemon	*[59]*
'Washington Navel' (Egypt)	4	'Volkamer' lemon	'Sour' orange 'Troyer' citrange 'Rangpur' lime	*[94]*
'New Hall', 'Navelina', 'Lane Late', 'Spring', 'Fisher' and 'Fukumoto' (Egypt)	2	'Volkamer' lemon	'Sour' orange	*[95]*
'Lane Late' (Spain)	6	'Cleopatra' mandarin	Forner-Alcaide 41	*[96]*
'Delta' and 'Lane Late' (Cyprus)	6		'Volkamer' lemon	*[22]*
Mandarin				
'Marisol' (Egypt)	4	'Cleopatra' mandarin 'Carrizo' citrange 'Swingle' citrumelo	'Sour' orange	*[60]*
'Clemenules' (Spain)	14	'Volkamer' lemon		*[8]*
'Balady' and 'Fremont' (Egypt)	2	'Sour' orange	'Volkamer' lemon	*[97]*
'Nagpur' (India)	7	Kumquat		*[57]*
'Kinnow' (India)	6	'Rough' lemon	Shekwasha Pectinifera	*[84]*
Grapefruit				
'Marsh' 'Ruby Red' (Iran)	8	C.Amblycarpa 'Volkamer' lemon 'Cleopatra' mandarin 'Sour' orange		*[98]*
'Marsh Seedless' 'Redblush' (India)	8	Attani-2		*[4]*
Lemon				
'Kagzi Kalan' (India)	8	RLC-4 'Troyer' citrange	Karna Khatta	*[50]*

A positive effect of 'Volkamer' lemon on Vitamin C has been reported in some Navel oranges cultivars (*Citrus sinensis* [L] Osbeck) ('New Hall', 'Navelina', 'Lane Late', 'Spring' Navel, 'Fisher' and 'Fukumoto', 'Washington' Navel), mandarins ('Clemenules') and grapefruits ('Marsh' and 'Ruby Red'). [8,94,95,98] Nevertheless in other studies on 'Washington Navel' oranges and 'Balady' (*Citrus reticulata* Blanco) and ‘Fremont’ mandarins, which compared the effect of ‘Volkamer’ lemon and ‘Sour’ orange on vitamin C, the fruit grafted onto ‘Sour’ orange had higher vitamin C levels than those from ‘Volkamer’ lemon. [59,97] Similarly, with oranges ‘Delta’ and ‘Lane Late’, the fruit grafted onto ‘Volkamer’ lemon exhibited the lowest vitamin C content than the fruit from five other rootstocks. [22]

‘Cleopatra’ mandarin is another rootstock that some studies have reported as being able to induce incremental vitamin C content. The ‘Lane Late’ oranges grafted onto ‘Cleopatra’ mandarin exhibited the highest vitamin C levels of the fruit from six other evaluated rootstocks. [96] Similar results were found by Bassal [60], who evaluated vitamin C content in ‘Marisol’ clementines grafted onto four different rootstocks, and found that fruit grafted onto ‘Cleopatra’ mandarin had the largest amount, but observed no significant differences with ‘Carrizo’ citrange and ‘Swingle’ citrumelo. Ramin and Alirezanezhand [98] also reported that ‘Cleopatra’ mandarin is one of the rootstocks that led to fruit having the highest vitamin C content of eight evaluated rootstocks.

On the other hand, it has been reported that the content of vitamin C are related to the amount of some macro and micronutrients. So, while the presence of high phosphorous and nitrogen contents reduces the amount of vitamin C, [91,99] high potassium and zinc concentration increase the amount of vitamin C. [99-101] Although it is known that rootstock interferes with the nutritional status [102,103], no studies have been found in which the relation between the influence of rootstock on vitamin C content and the mineral uptake ability of rootstocks has been specifically addressed.

Fruit Disorders

Rootstock has been reported to influence the development of citrus disorders (Table 7).

Table 7. Effect of rootstock on peel disorders of citrus fruit

Cultivar (Country)	Number of Rootstocks	Rootstock's Effect		Reference
Albedo Breakdown		**High disorder**	**Low disorder**	
'Bellamy' Navel orange (Australia)	5	Normal Conditions 'Carrizo' citrange* 'Troyer' citrange* 'Trifoliate' orange	'Cleopatra' mandarin** 'Sweet' orange	*[104]*
		*: <50% Reduction after Deficit Irrigation **: 50% Reduction after Deficit Irrigation		
'Navelate' Sweet orange (Spain)	3	'Carrizo' citrange	'Sour' orange	*[108]*
'Chislett' Navel orange	4	'Carrizo citrange' C-35	'Cleopatra' mandarin Forner-Alcaide 5	*[109]*
'Nova' mandarin	2	'Carrizo citrange'	Forner-Alcaide 5	
'ClemenRubi' mandarin (Spain)	2	'Carrizo citrange'	'Trifoliate' orange	
Oleocellosis				
'Hamlin' Sweet orange (China)	3	'Rangpur' lime	Lichi 16-6 Trifoliate Gou Tou Cheng	*[110,111]*
Peel Pitting				
'Nadorcott' mandarin (South Africa)	2	'Rough' lemon	'Carrizo' citrange	*[112]*
'Valencia' orange (Australia)	3	'Symons Sweet' orange	'Rangpur' lime 'Emperor' mandarin	*[113]*
'Nova' and 'Fremont' mandarin (Turkey)	3	'Carrizo' citrange	'Sour' orange	*[114,115]*
Stem-End Rind Breakdown				
'Marsh' grapefruit (Florida)	6	'Sour' orange		*[58]*
'Valencia' orange	5	US-952	Gou Tou Cheng	*[116]*
'Ray Ruby' and 'Oroblanco' grapefruit (Florida)	4 9	No differences		
Chilling Tolerance				
'Clementine' mandarin (France)	2	Tetraploid 'Carrizo' citrange	Diploid 'Carrizo' citrange	*[117]*
Decay				
'Washington' orange (Egypt)	2	'Sour' orange	'Volkamer' lemon	*[59]*
'Marsh' and 'Ruby Red' grapefruit (Iran)	8	'King' mandarin	'Volkamer' lemon	*[118]*
Granulation (Pulp)				
'Valencia' orange (Greece)	2	*Citrus aurantium*	'Trifoliate' orange	*[119]*
'Kinnow' mandarin (India)	3	Sohsarkar	'Troyer' citrange	*[120]*
'Kinnow' mandarin (Pakistan)	9	'Volkamer' lemon	'Rough' lemon citrange (x3)	*[121]*
'Hamlin' orange (Oman)	6	'Cleopatra' mandarin 'Volkamer' lemon	'Acid' lime 'Sour' orange	*[122]*

Albedo breakdown (also referred to as 'creasing') is one of the most prominent pre-harvest rind disorders to limit the suitability of mandarins and oranges for fresh fruit markets. It is characterized by the outer colored portion of flavedo folding inwardly into channels to form the underlying albedo tissue as adjacent albedo cells separate. [104] This disorder has been reported to be influenced by scion, rootstock, rind thickness and Ca levels. [104-107]

The effect of rootstock on albedo breakdown has been related to water uptake capacity. Treeby et al. [104] demonstrated the major influence of rootstock genotype on the response to irrigation management. A study carried out during two seasons established that the response to albedo breakdown incidence in 'Bellamy' Navel oranges (*Citrus sinensis* [L.] Osbeck) highly depended on rootstock. 'Troyer' and 'Carrizo' citrange and 'Trifoliate' orange induced higher percentages of affected fruit than 'Cleopatra' mandarin and 'Sweet' orange. Under deficit irrigation conditions, while fruit budded onto Sweet orange halved the incidence of this disorder, those grafted onto 'Troyer' and 'Carrizo' citrange exhibited a major reduction, and the fruit grafted onto 'Cleopatra' mandarin and 'Trifoliate' orange displayed no effect.

Agustí et al. [108] evaluated the influence of three rootstocks on the rind breakdown of 'Navelate' sweet oranges (*Citrus sinensis* [L] Osbeck). Fruit grafted onto 'Carrizo' citrange had the highest rind breakdown level, followed by fruit grafted onto 'Cleopatra' mandarin and 'Sour' orange. This fact was correlated with fruit-tree water relations due to the average xylem vessel diameter of the peduncles fruit grafted onto 'Carrizo' citrange being 6% and 17% larger than those on 'Cleopatra' mandarin and 'Sour' orange, respectively. Similarly, in 'Chislett' Navel orange and 'Nova' and 'Clemenruby' mandarins, the higher percentages of damage due to rootstocks coincided with a larger vessels diameter. [109]

Oleocellosis (or oil spotting) is a physiological disorder that occurs after the peel oil gland ruptures, which causes visible pitting due to the released oil that is phytotoxic to pericarp cells. [123,124] Zheng et al. [110,111] studied the tolerance of on-tree oleocellosis in 'Hamlin' sweet oranges (*Citrus sinensis* [L.] Osbeck) grafted onto three different rootstocks. They observed that the fruits grafted onto 'Rangpur' lime exhibited the lowest oleocellosis incidence, which was related to the ability to adjust the water potential and antioxidant enzyme activities in trees. The peel of the fruit grafted onto 'Rangpur' lime maintained a higher water potential and relative water content.

Peel pitting is a common postharvest citrus disorder that can be manifested at chilling or non-chilling temperatures. This disorder affects subepidermal cells on fruit surfaces. [125,126] This postharvest disorder has been reported to be affected by rootstock. In 'Nadorcott' mandarin (*Citrus reticulata* Blanco) greater non-chilling peel pitting incidence was found in the fruit budded onto 'Rough' lemon rootstock compared to those budded onto 'Carrizo' citrange. This fact was related to the lesser ability of the fruit rind from the scion grafted onto 'Rough' lemon to prevent water loss. [112]

As for peel pitting appearing as a response to chilling temperatures, El-Zeftawi et al. [113] stated that 'Newton late Valencia' oranges (*Citrus sinensis* [L.] Osbeck), stored at 5-15°C for 18 days, had a higher peel disorder incidence when grafted onto rootstock 'Symons Sweet' orange (*Citrus sinensis* [L.] Osbeck) than those grafted onto rootstocks 'Rangpur' lime or 'Emperor' mandarin (*Citrus reticulata* Blanco). Özdemir et al. [115] studied 'Nova' mandarin and corroborated the influence of rootstock on peel pitting during storage at 4°C and 6°C. The fruit budded onto 'Troyer' and 'Carrizo' citrange exhibited higher percentages of fruit with this disorder than the fruit budded onto 'Sour' orange. 'Fremont' mandarin grafted onto 'Carrizo' citrange also displayed a higher incidence of physiological disorders than when the fruit were grafted onto 'Sour' orange after cold storage. [114] With grapefruit cultivars, higher incidence of chilling injury was observed in 'Marsh' grapefruit stored at 5°C when fruit were grafted onto 'Sour' orange than when grafted onto five other rootstocks. [58] Recently, Oustric et al. [117] studied the effect of tetraploid and diploid rootstocks on the chilling tolerance of common clementine, and reported better chilling resistance for the leaves of clementines grafted onto tetraploid rootstocks compared to diploid ones, which was related to the antioxidant system. Hence it would be interesting to study how this effect would be reflected on fruit quality.

Rootstock has also been reported to affect the development of stem-end rind breakdown (SERB). [116,127] This disorder has been related to water peel status, and the water stress that leads to SERB in citrus fruit may regulate phospholipase gene expression and involves ABA signaling. [128] Ritenour et al. [116] examined the rootstock effect on SERB incidence, and observed how 'Valencia' Navel oranges consistently showed considerable SERB damage on the fruit grafted onto rootstock US-952 ((*Citrus paradisi* x *Citrus reticulata*) x *Poncirus trifoliata*), but less damage on the fruit grafted onto rootstock GouTou Cheng. However, these authors did not report any rootstock effect on grapefruits 'Ray Ruby' and 'Oroblanco' (*Citrus grandis* [L] Osbeck x *Citrus paradisi* Macf.).

The influence of rootstock on citrus fruit decay has also been reported. Alirezanezhad and Ramin [118] evaluated eight different rootstocks in grapefruits 'Marsh' and 'Ruby Red' and observed the highest decay percentage in fruit grafted onto 'King' mandarin (*Citrus nobilis* Lour.) and the lowest incidence in those grafted onto 'Volkamer' lemon. The decay incidence in 'Washington' Navel oranges was also lower in the fruit grafted onto 'Volkamer' lemon than on those grafted onto 'Sour' orange. [59] In other studies, no influence of rootstock has been observed on the decay incidence. [115,129]

Regarding internal quality, granulation is one of the most important preharvest disorders to seriously affect citrus commercialization. It is a disorder of juice sacs of citrus fruit when they become hard, dry and enlarged, take a grayish color and have only a little free juice. [127] In most cases, a positive relation between the granulation incidence and rootstock vigor has been reported, and this effect has been related to the vigor that rootstock confers. [130] Kotsias et al. [119] reported that 'Valencia' oranges grafted onto 'Trifoliate' orange, a dwarfing rootstock, showed less granulation than the fruit grafted onto *Citrus Aurantium*, a vigorous rootstock. In 'Kinnow' mandarin, the granulation incidence was very high in the fruit from the trees grafted onto vigorous rootstock Sohsarkar (*Citrus Karna* Raf.), but was very low in the fruit from the trees grafted onto 'Troyer' citrange, a dwarfing rootstock. [120] Those authors also found that pectinesterase and diastase enzyme activities, which are closely related to granulation, were less marked in granulated fruit than in unaffected fruit, and were significantly influenced by rootstocks.

Al-Hosni et al. [121] also stated that the number of granulated fruits in 'Hamlin' oranges was affected by rootstock selection. The most affected fruit was that grafted onto 'Cleopatra' mandarin followed by 'Volkamer' lemon, while fruit grafted onto 'Acid' lime (*Citrus aurantifolia* (Cristm.) Swingle) and 'Sour' orange had lower fruit damage incidence. 'Kinnow' mandarin gave a higher granulation incidence when grafted onto 'Volkamer' lemon than onto rootstocks 'Rough' lemon and three citrange ones. [122] Nevertheless, it is noteworthy that besides the influence of rootstock, the incidence of this disorder depended on other factors like cultivar, agroclimatic conditions and genetic differences. [120]

Volatile Compounds

Essential oil (EO) is the volatile liquid fraction that is usually distilled by steam stripping. EO contains mixtures of terpenic hydrocarbons and oxygenated compounds, such as aldehyde, alcohol, ketone and ester derivatives, which are responsible for fruit aroma. The principal component in all citrus fruit EOs is the monoterpene limonene. [131]

Scarce information exists about the effect of rootstock on the volatile compounds on citrus fruit. Regarding 'Page' mandarin [(*Citrus reticulata* var. 'Dancy' × *Citrus paradisi* var. 'Duncan') × *Citrus clementina* Hort. ex Tan.] grafted onto eight rootstocks, Babazadeh-Darjazi et al. [132] stated that juice and peel oil from the fruit budded onto 'Swingle' citrumelo and Yuzu rootstocks had the highest aldehydes content, which is one of the most important fractions of citrus volatile compounds.

Benjamin et al. [133] carried out an in-depth study into the influence of five rootstocks on the volatile composition of pulp fruit of mandarins 'Or' and 'Odem' (*Citrus reticulata* Blanco), 'Valencia' oranges and 'Redson' grapefruits (*Citrus maxima* (Burm.) Merr, cv. 'Chandler') × *Citrus paradisi* Macf., cv. 'Hudson'). The results revealed that despite the wide variability in volatile compounds due to the grafting combination, the fruit budded onto 'Volkamer' lemon had the lowest levels of volatile compounds. Regarding mandarins, 'Or' budded onto 'Sour' orange had the highest levels of aldehydes nonanal and decanal, and 'Odem' budded onto rootstock US-812 (*Citrus reticulata* x *Poncirus trifoliata* [L.] Raf., hybrid) presented the most aroma volatiles. In the last cultivar, these differences were due to the levels of linalool (alcohol), perillaldehyde and dodecanal (aldehydes), β-pinene, limonene and γ-terpinene (monoterpenes). The 'Redson' grapefruit grafted onto 'Sour' orange had larger amounts of 19 volatile compounds than fruit grafted onto 'Volkamer' lemon and Macrophylla. In 'Valencia' oranges, not all the volatile compounds were affected by rootstock in the same way. While carvone compound levels were higher in the fruit budded onto 'Sour' orange fruit than in those grafted onto 'Volkamer' lemon and x639 (*Citrus reticulata* x *Poncirus trifoliata* [L.] Raf., hybrid), the levels of α-pinene, sabinene, β-pinene, limonene (terpenes) and copaene (sesquiterpene) were higher in the fruit grafted onto x639 than in those onto 'Sour' orange and 'Volkamer' lemon.

Saini et al. [84] studied four different rootstocks and found that most volatile compounds were differently affected depending on rootstock. The juice of the 'Kinnow' mandarin grafted onto Pectinifera (*Citrus depressa* Hyata 'Pectinifera') had the highest total volatile contents because this rootstock induced the highest limonene levels. Nevertheless, 'Kinnow' mandarin grafted onto Shekwasha (*Citrus depressa* Hyata 'Shekwasha') had the highest levels of β-pinene, dodecyl aldehyde, octanal, α-terpineol, terpinen-4-ol, perialdehyde, nonanal, isoleucine, linalool and hexanal, while fruit budded onto 'Sour' orange had the highest trans-β-ionone concentrations. Even the 'Cleopatra' mandarin rootstock, which induced the lowest levels of total volatile compounds, led to the highest ethyl acetate levels.

Apart from the influence of the amount of volatile compounds on fruit, rootstock has also been described as having an effect on the presence or absence of certain volatile compounds. This was the case of 'Persian' lime (*Citrus latifolia* Tan.) grafted onto five rootstocks, in which β-myrcene was found only in the fruit grafted onto 'Sour' orange and Flying dragon. Likewise, β-thujene and dodecane were detected only in fruit budded onto 'Volkamer' lemon and C-35, respectively. Linalool was present in the fruit grafted onto all the rootstocks, except in the 'Sour' orange fruit. [134]

Conclusion

Rootstocks significantly impact the internal and external quality parameters of citrus fruit. Nevertheless, the influence of a specific rootstock on citrus quality is highly dependent on the cultivar, and also on climate conditions and cultural practices. They all need to be taken into account when making decisions about the rootstock to be used in each specific case.

The influence of the rootstock on fruit quality has been linked with the water uptake capacity of roots. Nevertheless, there are few studies that address in depth the absorption of nutrients from the soil by rootstocks. The knowledge about the nutrients translocation thought the rootstock-cultivar graft union and its effect on fruit quality is crucial to select the optimal rootstock in soils with a certain mineral composition.

On the other hand, many studies have addressed the influence of rootstock on endogenous production of primary and secondary metabolites such as sugars, acids, volatiles and vitamins, which in turn are involve on the fruit quality. Nevertheless, few attempts have made to elucidate the mechanism underlying rootstock-induced phenotypic changes. The molecular mechanism related to fruit quality affected by rootstock is unclear and it becomes essential due to increasing consumer demand for high quality fruit.

Metabolomics and transcriptomics tools are needed to explore the candidate genes involved in the metabolic processes affected by rootstock. Knowledge of changes in gene expression caused by the rootstock-scion interaction can would provide useful information to develop or select genotypes for future rootstock breeding programs.

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