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Effect of Gibberellic Acid (GA₃) on Rootstock on the Evolution of the Mandarin Tree Induction "Nadorcott"

Nabil Rachdi^{1,2*}, Abdelhadi Ait Houssa³, Mohamed Midaoui², Bouchra Et-Takaouy², Kawtar Semaoui⁴, Redouane Bakkali⁵, Mohamed Benbella² and Mohamed Ibriz¹

¹Department of Biology, Faculty of Science, University Ibn Tofail, Morocco. ²Department Science and Technology of Crops, National School of Agriculture of Meknes, Morocco. ³Farming Division, Provedences Vertes Group, Morocco. ⁴Department of Arboriculture, Olive and Viticulture, National School of Agriculture of Meknes, Morocco. ⁵Inorganic Laboratory, Agrilabs Laboratory, Morocco.

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

An application of gibberellic acid (GA₃) was applied to different doses (0; 15; 30 ppm) on Nadorcott trees transplanted on five rootstocks (*Macrophylla citrus*, *Carrizo citrange*, *Troyer citrange*, *C35 citrange* and *Swingle citrumelo*) during the fruits coloration shifting. A monitoring of the evolution of the floral induction by the counting of leaves and the flowers/ fruits for the sensitive stages: flowering, fruiting and end physiological drop, with physico-chemical analyzes of the leaves at the physiological dropping stage, of caliber during the magnification stage. The shoots on the *Swingle Citrumelo* rootstock had more flowers than the Citranges and even more than the *Macrophylla*

*Corresponding author: E-mail: rachdinabil@gmail.com;

citrus. This classification is still valid for fruits during fruiting, results of a standard drop for the different rootstocks with the dominance of leaved inflorescence with unique flower. On the other hand, the physiological drop was different from rootstock to the other, more accentuated in *Swingle citrumelo*, *Troyer* and *Carrizo citrange*, and moderate in *C35 citrange* and *Macrophylla citrus*. While the GA₃ treatment did not significantly affect either the floral induction or the drop in its two phases. It is noted that with the evolution of the flowering there is a significant positive effect of the treatment and of the GA₃ dose on the fruits hooking, with a 23% increase for the dose of 30 ppm GA₃ compared to the control. However, the variation in size is better for fruits on leaved inflorescence with unique flower, as well as on the *Macrophylla citrus* rootstock, followed by Citrange (C35 followed by Carrizo and Troyer), and finally *Swingle Citrumelo* (10% less in comparison with *Macrophylla citrus*).

Keywords: Nadorcott; rootstock; gibberellic acid; fruit coloring; floral induction; type of inflorescence; Caliber.

1. INTRODUCTION

In Morocco, the citrus fruit sector plays an important socio-economic role and it covers in 2016 a surface area of 124.5 kha, of which 109.8 kha is in production. It produces 2.36 MT on average and exports 0.5 MT the equivalent of almost 3 billion DH / year in foreign currency [1]. However, the citrus sector generates nearly 21 million working days per year (60% in orchards and 40% in agribusiness) [2].

The main varieties of Moroccan citrus fruits are Clementine (39%), Maroc Late (29%), Navel (22%), Half season oranges (5%) and other varieties [2]. The Nadorcott variety gained in importance, with an area of 6.6 kha, of which 4.6 kha in production, with a production of 0.18 MT mainly destined for export [1].

This study focuses on the « Nadorcott » highly efficient mandarins, with high quality fruits, without seeds, under certain isolation conditions, easy to peel, and whose maturity is later than that of Clementine, which can be spread out between the end of December until mid-March [3,4].

In order to spread this harvesting and commercialization periods, there are many means that can be used such as: the diversification of rootstocks [5], the spraying of the gibberellins during the period spreading between the end of September and the beginning of July [6-8].

The treatment aiming to delay the harvesting of fruits right before the fruit twist [8-12] happens before or in the beginning of the presumed

period of the installation of the floral induction process [13], occurs in sub-tropical zones during winter as a response to low temperatures [14]. This fact is even more important than the effect of gibberellic acid has on the floral induction, a marked influence depending on the concentration and the application period [13,15], and inhibiting effect over the flowering of the next spring [16-18].

Several studies have shown that floral induction is influenced by environmental conditions and endogenous factors [19-24]. The axillary buds that have undergone floral induction (physiological process) followed by floral differentiation (formation of drafts of the flower organs) will give the spring inflorescences (flowers and floral shoots). The buds that have not undergone floral induction will give vegetative shoots [25].

The picking period is even more reduced with the use of renowned rootstock for their efficiency and the production of big caliber fruits (e.q. Macrophylla citrus. [26-29]. Volkameriana lemon) Βv usina these rootstocks, the quality declines (low proportion of acids and sugars with a rather tasteless savor) [30,31]. However, the hydric stress, under certain conditions, allows a significant amelioration of fruits quality by the increase in dry soluble and titrable acidity [32-40]. Moreover, the coloration and maturity are more advanced [41], the Nadorcott fruits in the coastal Atlantic zone. must be harvested between mid-December and mid-January, which is almost the same time as the clementine [42]. Within these coastal zones and the cited above rootstocks, a delayed picking gives as a result a rapid deterioration of fruits quality, especially after raindrop.

On the other side, in adequate rootstock, the maturity of the **Nadorcott** variety is reached in January and the picking could last until mid-February. The fruits remain stiff, and have a satisfying quality [43].

This article is a study on the interactions between the rootstock and the application of gibberellic acid, at the stage of coloration turning over the evolution of floral induction of the **Nadorcott** variety. These interactions have never been studied under the Moroccan conditions.

2. MATERIALS AND METHODS

The studied testing has been carried out during the 2011-2012 campaign in the experimental station of the SEBNAK domain (Maroc-Merchich-Lambert Nord: X=414 559m; Y=464 524m; Z=49 m), in BAHARA OULED AYAD, which is within the Atlantic coastal zone of GHARB in Morocco. On a sandy soil (Table 1) and a good physiochemical quality of irrigation water (Table 2).

The region has only received 227mm/year of raindrop, with a soft weather, not exceeding 41°C during summer, with a limited risk of frost during winters (Fig. 1).

2.1 Monitoring of Floral Induction

The experimental plan is a 3-level split plot, with a complete random block disposition in the first level (Fig. 2), where the rootstock is the main factor, GA₃foliar spraying as secondary factor and the stage is the tertiary factor, using 6 complete random blocks. However, the experimental unit is composed of 4 trees, and only the middle 2 are taken into account, as the two trees in the extremities are discarded due to the boundary effect. Three doses of GA₃ (Berelex 40 SG, Marbar S.A., Casablanca, Maroc, 10% GA₃) have been tested (0, 15, 30 ppm) in the stage of fruit coloration turning (October 26th, 2011), mixed with a non-ionic admixture (Agral 90, Syngenta S.A., Rabat, Morocco, 90% Nonyl phenol ethoxyled) with a rate of 0.5 I /1000 I. Furthermore, five branches were chosen on each orientation, East and West, at the height of a normal man, for the two central trees.

Table 1. Physico-chemical soil properties

Sail	0.20 am	20.60 om
3011	0-30 CM	30-60 CIII
Argil (%)	-	-
Silt (%)	5,00	5,00
Sand (%)	95,00	95,00
pH	7,64	7,87
EC (µS/cm)	240,43	177,13
$CaCO_3$ (%)	0,50	0,50
MO (%)	1,10	0,74
N Dumas (ppm)	562,87	395,53
P (ppm)	34,87	52,60
Ca (meq/100 g)	4,02	4,46
Mg (meq/100 g)	0,69	0,57
K (meq/100 g)	0,17	0,10
Na (meq/100 g)	0,89	0,83

The comments are done in three periods: Flowering (**FLO**): April 8th, 2012, Fruiting (**FRU**): May 12th, 2012. End of physiological drop (**EPD**): 16th July, 2012. We count on each shoot of last summer, the number of leaves, flowers or fruits for every inflorescence.

2.2 Monitoring of the Physiological Drop

The studied factor is the rootstock in a completely random disposition with 6 repetitions. The elementary parcel contains two trees. Five rootstocks have been studied (MC, CC, TC, C35C, SC). The test has been realized between (FRU) and (EPD).

The plantation is five years old, with a density of 2x6 m (833 trees/hectare) and around 3,5m in height. The seeds are transplanted over 5 rootstocks, which are: *Macrophylla citrus* (MC), *Carrizo citrange* [*C. sinensis* (L.) Osb. x *Poncirus trifoliata* (L.) Raf.] (**CC**), *Troyer citrange* (**TC**), *C35 citrange* [*C. sinensis* (L.) Osb. x *Poncirus trifoliata* (L.) Raf.] (**C35C**) and *Swingle* or 4475 *citrumelo* [*C. paradisi* Macf. x *P. trifoliata* (L.) Raf.] (**SC**).

Table 2. Physico-chemical properties of irrigation water

NO3 meq/l	SO₄ ²⁻ meq/l	HCO3 meq/l	Cl meq/l	Ca ²⁺ meq/l	Mg ²⁺ meq/l	Na ⁺ meq/l	K⁺ meq/l
0,95	0,24	2,11	1,55	3,57	0,43	1,7	-
рН	EC μS/cm	B ppm	Fe ppm	Mn ppm	Cu ppm	Zn ppm	
7,85	509	-	-	-	-	-	



Fig. 1. Weather data of the experimental station in SEBNAK domain

	Bloc 05					B	oc 0	3		Bloc 01				
R_2T_2	R₅T₂	R_4T_2	R_1T_2	R_3T_2	R_4T_3	$R_4T_3 \ R_5T_3 \ R_3T_3 \ R_1T_3 \ R_2T_3$					R_4T_2	R₃T₂	R_2T_2	R_1T_2
R_2T_1	R_5T_1	R_4T_1	R_1T_1	R_3T_1	R_4T_1	R₅T1	R_3T_1	R_1T_1	R_2T_1	R₅T₃	R_4T_3	R₃T₃	R_2T_3	R_1T_3
R_2T_3	R₅T₃	R_4T_3	R_1T_3	R_3T_3	R_4T_2	R_4T_2 R_2T_2 R_3T_2 R_1T_2 R_2T_2					R_4T_1	R_3T_1	R_4T_1	R_1T_1
Bloc 06					Bloc 04									
	Bl	oc 0(6			B	oc 04	4			В	loc 0	2	
R ₂ T ₃	BI R₅T₃	<mark>oc 0</mark> R₄T₃	5 R₁T₃	R ₃ T ₃	R ₄ T ₁	B R ₅ T ₁	oc 04 R ₃ T ₁	4 R ₁ T ₁	R_2T_1	R ₅ T ₁	B R ₄ T ₁	oc 0	2 R ₂ T ₁	R_1T_1
R_2T_3 R_2T_2	Bl R ₅ T ₃ R ₅ T ₂	OC 00 R ₄ T ₃ R ₄ T ₂	5 R ₁ T ₃ R ₁ T ₂	R_3T_3 R_3T_2	R ₄ T ₁ R ₄ T ₂	B R ₅ T ₁ R ₅ T ₂	OC 04 R ₃ T ₁ R ₃ T ₂	4 R ₁ T ₁ R ₁ T ₂	R_2T_1 R_2T_2	R₅T₁ R₅T₃	B R ₄ T ₁ R ₄ T ₃	OC 0 R ₃ T ₁ R ₃ T ₃	2 R ₂ T ₁ R ₂ T ₃	R_1T_1 R_1T_3

Fig. 2. Experimental Plan

R₁: Macrophylla citrus; R₂: Carrizo citrange; R₃: Troyer citrange; R₄: C35 citrange; R₅: Swingle citrumelo; T₁: 0 ppm d'GA₃; T₂: 15 ppm d'GA₃; T₃: 30 ppm d'GA₃;

The samples have been taken on a daily basis, by counting the dropped fruits, which are recovered using a net installed under the two threes, with heightened boundaries in order to avoid any interferences with neighboring trees.

2.3 Nutritional State in the Physiological Drop Stage

The test has only used four blocks (1, 2, 3 and 4) as shown in the plan in Fig. 2, this is a Randomized Complete Block Design. The samples are taken in the physiological drop stage (May 21st, 2012), at the average height of a man around the tree, with a representative sample of 100 leaves, on mature 3 months old shoots (spring shoots).

Once taken, the samples are directed towards the laboratory, where they are cleaned using distilled water (Milli-Q Water), drained, weighted, dried using a stove (Labtech) at 80°C for 8 hours, crushed using a hammer mill, sift using a 2 mm sieve and then re-weighted. A 1-g porcelain crucible is weighted and its concentration of N analyzed following the Dumas process: this method consists of dry combustion up to 950°C, using oxygen as an accelerator. The products of this combustion (H_2O , CO_2 , SO_2 , NO_X , N_2) are filtered. NO_x is reduced into N_2 using copper, and is swept by gaseous helium through a sensor with thermal conductivity where the concentration is measured (DNA 701 VELP). Another one is introduced into an oven (Labtech) at a temperature of 450°C. After a calcination during 2 hours, we remove the crucible and we let it cool down at ambient temperature, then we add a few drops of Milli-Q water to fix the ashes, then we add 2ml of concentrated nitric acid. After filtration, we take the solution into a 100ml flask and fill it up with Milli-Q water. We transverse the obtained solution into a plastic bottle. The obtained solution is to be used for an analysis of chloride by spectrophotometry (Jenway), of phosphorus by colorimetry (Jenway), and of metals by spectrometry of inductive plasma atomic emissions (Agilent Technology 700 ICP OES Series). A computer program allows us to compare every new result to the referential values stored in the database. The measures are taken in an inorganic laboratory, while determining the chemical compositions of the foliar samples (N, P, K, Ca, Mg, Cl, Na, Zn, Mn, Cu, Fe and B).

2.4 Monitoring of the Caliber

By the end of the physiological drop (July 16th, 2012), we have identified the type of 270 inflorescences with flowers/fruits [44,14,45,46], of three trees for five rootstocks (MC, CC, TC, C35C, SC), following the counting methodology.

2.5 Inflorescence Types

Under the experiment's conditions, we note the dominance of inflorescences with a unique fruit (Table 3).

The framing method consists of counting the number of each inflorescence type with the help of a square frame with the length of a side being 33,3 cm. The results show also that only LSFI

and LsSFI carry the production of the current year (Table 4).

Thereby, for every experimental unit, we follow the caliber, with the help of a numbered sliding foot, of 10 fruits per direction East and West, for the two inflorescence types: LSFI and LsSFI. The caliber recordings have started after the end of the physiological drop (August 8th, 2012) and have been measured regularly every week until the end of autumn (December 29th, 2012). The experimental plan is a slit plot (Fig. 2) with 4 completely random blocks (1,2,3 and 4).

The relative growth rate (**RGR**) has been calculated using the following equation [47]:

$$RGR = \frac{D2 - D1}{t2 - t1}$$

RGR is the relative growth rate in mm/day, **D** is the measured value of the growth parameter (equatorial diameter of the fruit) at instant t (in mm) and **t** is the time between measures in days.

The statistical analysis has been carried using the help of the IBM SPSS Statistics program (21^{st} version; IBM Corporation), we use the Duncan test for the means comparisons at P=5%.



Fig. 3. Inflorescence types

VI: Vegetative Inflorescence; LSFI: Leafy Single Flowered/Fruited Inflorescence; LMFI: Leafy Multi-Flowered/Fruited Inflorescence; LsSFI: Leafless Single Flowered/ Fruited Inflorescence; LsMFI: Leafless Multi-Flowered/Fruited Inflorescence

Inflorescence type	Rootstock							
	тс	C35C	SC	CC	MC			
LSFI	78% A ^z a ^v	75% Aa	75% Aa	77% Aa	77% Aa			
LMFI	0% Ca	0% Ca	0% Ca	0% Ca	0% Ca			
LsSFI	22% Ba	25% Ba	25% Ba	23% Ba	23% Ba			
LsMFI	0% Ca	0% Ca	0% Ca	0% Ca	0% Ca			
Zfor a given restated	the perceptages f	allowed by the er	ma conital lattar	ara not cignifican	thy different			

Table 3. Effect of rootstock on inflorescence type

²for a given rootstock, the percentages followed by the same capital letter are not significantly different (SNK, 5%)

^Yfor a given type of inflorescence, the percentages followed by the same tiny letter are not significantly different (SNK, 5%)

Dose GA ₃	Inflorescence type			Rootstock	S	
		ТС	C35C	SC	CC	MC
0 ppm	LSFI	95%	90%	96%	100%	95%
	LsSFI	5%	10%	4%	0%	5%
15 ppm	LSFI	100%	100%	97%	100%	100%
	LsSFI	0%	0%	3%	0%	0%
30 ppm	LSFI	100%	89%	88%	88%	100%
	LsSFI	0%	11%	13%	13%	0%

Table 4. Effect of the rootstock and AG₃ on the inflorescence type

3. RESULTS

3.1 Monitoring of Floral Induction

The Floral Induction variable (**FI**) is calculated by the formula:

 $FI(\%) = \frac{Number \ of \ inflorescences \ with \ flowers}{Total \ number \ of \ inflorescences} x100$

The results are expressed as a percentage in order to compensate for the size differences between branches chosen for the counting.

The variances analysis shows that there is no effect of the interaction of the rootstock and the GA_3 treatment over the floral induction. However, it shows a very significant effect of both rootstocks and GA_3 .

Fig. 4 shows that the *Macrophylla citrus* rootstock has a low floral induction, followed by the Citranges group (C35C, TC, CC) and at the end the *Swingle citrumelo* rootstock.

However, the GA₃ treatment has had a slight positive effect on the floral induction, insignificant, of the GA₃dose. This being said, GA₃ treatment has an inhibitory effect over the flowering during the floral induction. This allows us to suggest that the floral induction process either has not yet started, or there are other factors that reversed or cancelled the inhibitory effect of GA₃.

In order to better understand each rootstock behavior and how the GA_3 treatment acts on the flowering process, we have tried to deepen our study, by examining the effect of different rootstocks alongside with GA_3 treatment on every type of flowering. The results are shown in Table 5. For floral inflorescences, we notice the dominance of mixt inflorescences, notably those with a unique flower, this type being the most influenced by the rootstock and GA_3 treatment. Our study is also interested in the flowering evolution, and more specifically the hooking rate around the end of the physiological drop, which is conditional for the production. To do so, it is better to study both the flower drop (Drop I) and the fruits drop (Drop II): physiological drop or June drop.

The variances analysis shows that there is neither a rootstock nor a GA_3 treatment effect on Drop 1, no matter the inflorescence type, while the statistical results show that there is a highly significant effect of rootstock on Drop II.

Fig. 5 and Table 6 show that the shoots on the Swingle Citrumelo rootstock have had more flowers than the Citranges group and much more than the Macrophylla citrus. This ranking is also valid for fruits during the fruit set, result of a standard drop for the different rootstocks, with a domination from the leaved inflorescence with a unique flower. On the other side, the physiological drop is different from one rootstock to another, more pronounced on the Swingle citrumelo, the Troyer citrange and the Carrizo citrange, while being moderate on the C35 citrange and Macrophylla citrus. It has also led to an important hooking on the Swingle citrumelo and the C35 citrange, followed by Macrophylla citrus and at the end the Carrizo and the Trover citrange rootstocks.

In general, we notice that the drop of *Macrophylla citrus* is the least one comparing with other rootstocks, especially with *Swingle citrumelo* reaching a difference of around 89%.

Although there is no significant effect between GA_3 treatment and fruit drop in these two phases. It is observed that with the flowering evolution there is a positive and significant effect of treatment and of the GA_3 dose on the fruits hooking, with a 23% increase for the 30 ppm GA_3 dose compared to the control.

Under the experiment condition, and the results in Fig. 6, we notice that a great number of flowers coming from multiple flowers inflorescences have the tendency to completely drop by the end of the physiological drop, which confirms the results of Table 3. The flowers coming from unique flower inflorescences have the tendency to hook more.

Fig. 6 show the drop rhythm during the two phases, for the whole types set of inflorescences, brought to light by the existence of two rootstock groups, the first consists of *Macrophylla citrus* and *C35 citrange*, which have a slower pace of drop, while the second group consists of *Swingle citrumelo*, *Carrizo* and *Troyer citrange*.

3.2 Monitoring of the Physiological Drop

The monitoring of the drop only concerns the second drop (Drop II), recognized by the physiological drop. Statistical analysis unveils a

highly significant effect of the rootstock on the number of fruits per tree by the end of the physiological drop. Also, a very highly significant effect of rootstock on the percentage of the physiological drop of fruits (**PFD**).

The **PFD** variable is calculated as follows:

$$PFD(\%) = \frac{Number of dropped fruits (DROP II)}{Number of fruits (FRU)} x100$$

Fig. 7 and Table 7 show that the number of fruits dropper is approximately the same for all five rootstocks. In terms of percentage, we find the highest rate in the group containing TC, CC and SC, while the lowest percentages are found in MC and C35C.

The *Macrophylla citrus* could hook the maximum of fruits, followed by the *C35 citrange* and then the other rootstocks.





Fig. 5. Effect of rootstocks and GA₃ treatments on fruit drop

Paramete	ers			Rootstock	(GA ₃ Treat	nent	
		SC	тс	CC	C35C	MC	Sig	0 ppm	15 ppm	30 ppm	Sig
S	Vegetative inflorescence	45% a	53% b	55% b	56% b	61% c	***	57% a	53% b	51% b	**
ъ	Floral inflorescences	55% a	47% b	45% b	44% b	39% c	*	43% a	47% b	49% b	***
cer	Mixt inflorescences	43% a	37% b	35% bc	35% bc	32% c	***	32% a	38% b	39% b	***
es	Single flowered	41% a	35% b	34% b	34% b	31% b	**	31% a	36% b	37% b	***
lo	Multi-flowered	2%	2%	1%	2%	1%	NS	1%	1%	2%	NS
of infl	Generative inflorescences	12% a	10% ab	10% ab	8% bc	7% c	*	10%	9%	9%	NS
	Single flowered	9%	8%	8%	7%	6%	NS	8%	8%	8%	NS
%	Multi-flowered	2%	2%	1%	1%	1%	NS	2%	1%	1%	NS
ee دو	Floral inflorescences	63 a	54 b	48 cd	49 bc	42 d	**	48 a	51 ab	55 b	*
ene	Mixt inflorescences	48 a	41 b	37 cd	39 bc	34 d	***	35 a	40 b	44 c	***
nn	Single flowered	41 a	35 b	34 b	34 b	31 b	***	31 a	36 b	37 b	**
s n ore	Multi-flowered	8	6	2	6	3	NS	4	4	6	NS
ower's 0 inflo	Generative inflorescences	15	13	11	10	9	NS	13	11	11	NS
	Single flowered	9	8	8	7	6	NS	8	8	8	NS
E 6	Multi-flowered	5	5	3	3	2	NS	4	3	3	NS

Table 5. Rootstock and GA₃ treatment effect over the inflorescence type and flowering

The percentages or the numbers followed by the same tiny letter are not significantly different. Sig: Significance (Duncan: NS, *, **, *** Not significant or significant at P = 0.05, 0.01, 0.001, respectively)

Table 6. Effect of rootstock and GA₃ treatment on flowering evolution

Stages		Rootstock							GA ₃ Treatment (ppm)		
	MC	C35C	CC	тс	SC	Sig	0	15	30	Sig	
FLO	42a	49b	47b	52b	62c	**	47a	51ab	55b	**	
FRU	34a	39b	38b	41b	50c	***	37a	40a	44b	***	
EFD	22b	23bc	19a	19a	25c	**	20a	22b	24c	***	
Dropl	9a	10a	10a	12a	12a	NS	10a	11a	10a	NS	
Dropll	11a	16b	18bc	22cd	25d	**	17 a	18a	20a	NS	

The numbers followed by the same tiny letter are not significantly different. Sig: Significance (Duncan: NS, *, **, *** Not significant or significant at P = 0.05, 0.01, 0.001, respectively)



Fig. 6. Evolution of the number of flower by 100 inflorescences for every GA₃ treatment and each rootstock between flowering, fruit set and physiological drop

3.3 Nutritional State in the Physiological Drop Stage

The results of physio-chemical analysis of **Nadorcott**'s leaves on each rootstock are presented in Table 8. The results show a satisfying level of most of mineral elements, except for very low foliar levels of B and Cu, and low levels of Ca, Mg and P in the *Macrophylla Citrus*; P in *Troyer* and *Carrizo citrange*, N in *Macrophylla Citrus* and *Carrizo citrange*. however, we notice very high levels of S, Zn, Ma and Na.

The Variance analysis results have not shown any significant effect neither of the interaction of rootstock-GA₃ treatment nor of the GA₃ treatment on the mineral composition of leaves. Moreover, they unveil a significant effect of rootstocks on the P and Zn, a highly significant effect on Ca and P and a very highly significant effect on Mg and S.

Table 8 shows that the *Macrophylla citrus* has low foliar levels of Ca and Mg when compared to

the other rootstocks. *Carrizo citrange* displays the lowest levels when it comes to Zn and P, followed by *Troyer citrange* and *C35citrange*, *Swingle citrumelo* and then *Macrophylla citrus*.

3.4 Caliber Monitoring

The variance analysis results show a significant effect of the interaction between the rootstock and the inflorescence type in the initial date (**DI**: August 4 and 5, 2012). Otherwise, at the final date (**DF**: December 29 and 30, 2012), we find a very highly significant effect as for the rootstock as for the inflorescence type.

The caliber measurements at the initial date represent a result of the tree's equilibrium after the physiological drop, while the measurements at the final date represent the fruit bulking consequence, thus pushing us towards studying the caliber evolution.

The variance analysis of the caliber evolution (**CE**) between the initial and final dates, show a significant effect of the interaction between the rootstock and inflorescence type.



Fig. 7. Effect of rootstocks on the number of fruits at physiological drop

Table 7. Effect of the rootstock on the evolution of the number of fruits at physiological drop

	Rootstock							
	MC	C35C	CC	SC	TC	Sig		
FRU	684a	606a	554a	563a	615a	NS		
EFD	467a	400ab	322bc	299c	291c	**		
DROPII	218a	207a	233a	265a	324a	NS		
PFD	32%a	33%a	41%b	47%bc	51%c	***		

The numbers followed by the same tiny letter are not significantly different. Significance (Duncen: NS, *, **, het significant et significant et B = 0.05, 0.01, 0.001, respo

Sig: Significance (Duncan: NS, *, **, *** Not significant or significant at P = 0.05, 0.01, 0.001, respectively)

The Table 9 shows that there is a highly significant effect of the inflorescence type over the fruit caliber, between the beginning of the bulking until maturity, while there is no significant effect on the caliber evolution. We note again that the leaved inflorescences with unique flowers give bigger fruits than non-leaved inflorescences with a difference of 6,1% at the final stage.

However, the rootstock has a highly significant effect on both the fruit caliber at initial and final dates and the fruit evolution. At the initial date, we notice that the Troyer citrange gives the smallest caliber, followed by Carrizo citrange, C35 citrange, Swingle citrumelo, thus leaving the best caliber to the Macrophylla citrus rootstock. At the final date, we notice that the Trover citrange is the one that always has the smallest caliber, this time followed by Swingle citrumelo then the Carrizo citrange and C35 citrange, to leave the best caliber to Macrophylla citrus. This ranking readjustment is mainly due to the fact that the caliber evolution rhythm differs from one rootstock to another. To know that the best caliber evolution rate come back to Macrophylla citrus followed by Carrizo citrange (C35 citrange followed by Carrizo and Troyer citrange), and at

the end the *Swingle citrumelo* (10% less comparing to *Macrophylla citrus*).

The final caliber profile is a discounted result, the latest being commercially standardized by various groups of calibers for the **Nadorcott**, with a specific weight, as shown in the Table 10.

The Fig. 8 shows that the *Macrophylla citrus* rootstock the most important caliber, followed by the *C35 citrange*.

According to Fig. 9 and Table 10, we can deduct that the fruit average weight for each rootstock will be different, which will directly affect the yield.

From the results shown in Table 9 and the statistical analysis results, we have shown a highly significant correlation between the temperature and the caliber evolution rate for every rootstock (r>0.8). The caliber evolution rate oscillates between 0.24 and 0.36 mm/day with temperatures varying from 20° to 29°C, and afterwards, the rate slows down when the temperatures are below 20°C, while stabilizing at temperatures below 12.5°C.





Fig. 8. Fruit caliber progress (a) and fruit caliber evolution rate (b) of Nadorcott on each rootstock, alongside with weather data

Param	neters			Roots	tock		
		MC	C35C	CC	тс	SC	Sig
Са	%	2,99 a	3,49 b	3,59 b	3,53 b	3,74 b	**
Mg	%	0,25 a	0,34 b	0,37 b	0,35 b	0,35 b	***
K	%	1,3 bc	1,28 bc	1,14 a	1,19 ab	1,36 c	**
Р	%	0,16 a	0,18 b	0,18 b	0,17 ab	0,18 b	*
S	%	0,34 a	0,37 ab	0,39 b	0,38 b	0,43 c	***
Ν	%	2,27	2,31	2,28	2,38	2,36	NS
Zn	ppm	91 c	86 bc	67 a	71 ab	78 abc	*
Mn	ppm	101	96	75	84	86	NS
В	ppm	31	30	34	30	33	NS
Fe	ppm	138	182	174	156	147	NS
Cu	ppm	1,17	1,25	1,28	1,4	1,63	NS
CI	ppm	399	348	471	416	429	NS
Na	ppm	675	696	803	689	739	NS

Table 8. Effect of rootstocks on physio-chemical composition of leaves at physiological drop

The numbers followed by the same tiny letter are not significantly different. Sig: Significance (Duncan: NS, *, **, *** Not significant or significant at P = 0.05, 0.01, 0.001, respectively)

Table 9. Rootstock and inflorescence type	effect on caliber evolution
---	-----------------------------

Date		Rootstock						orescence t	зуре
	MC	C35C	CC	тс	SC	Sig	LSFI	LsSFI	Sig
ID	34,2c	31,6b	31,5b	29,7a	31,8b	***	33,6b	29,9a	***
FD	69,2d	65,8c	64,0b	62,0a	63,2b	***	66,8b	62,9a	***
CE	35,0d	34,3c	32,5b	32,4b	31,4a	***	33,2	33,0	NS

The numbers followed by the same tiny letter are not significantly different. Sig: Significance (Duncan: NS, *, **, *** Not significant or significant at P = 0.05, 0.01, 0.001, respectively)

Table 10.	Measurements	and number	of fruits f	or everv	commercial	caliber in	10ka	packag	ina

Caliber	Range	Number of fruits	Caliber	Range	Number of fruits
HC	< 44	-	C 1 X	68 - 71	75
C 6	44 - 46	225	C 1 XX	72 - 73	68
C 5	47 - 50	196	C 1 XX	74 - 77	60
C 4	51 - 54	168	C 1 XXX	78 - 81	53
C 3	55 - 58	140	C 1 XXX	82 - 84	42
C 2	59 - 64	108	C 1 XXX	≥ 85	36
C 1 X	65 - 67	90			



Fig. 9. Caliber profile for fruits from each rootstock

4. DISCUSSION

The GA₃ treatment on the mandarin **Nadorcott** transplanted on five different rootstocks was realized at the beginning of floral induction process period (Databases non published). Thereby, an inhibiting effect over the flowering of the following spring should be expected [17,18]. The results in Fig. 4 show that the gibberellic acid increases the floral induction during the flowering in the following season. The gibberellins (GAs) are responsible for the activation of the cell division process and the stretching of the shoots [48], and consequently, the growing of the shoots [49], or the control of both the growing and stretching of the shoots [50,51]. However, as was found on the field, the treatment was coincident with temperatures favorable to the development of the autumn shoot, which actually favored the growth thereafter. The trees that were treated with Gibberellic Acid had more developed shoots and could accumulate more carbon hydrates during winter [52,53], which have positively influenced the floral induction [54], and followed by the flowering of the next campaign [25] especially before the drop of temperatures [21] at an early date [19]. However, the GA₃ treatment serves to increase the number of leaved inflorescences [55,56] especially with unique flowers instead of minimizing the number of non-leaved inflorescences [16]. This is maybe further explained by the non-coincidence of the GA₃ application with the two peaks of sensitivity to the response of the citrus plant to the gibberellins. [57,16,19]: The first is timed with the beginning of the translocation of an unknown flowering signal from the leaves to the buds [58] and the second coincides with the beginning of the morphological flowers differentiation [59,16].

The rootstock influences the vegetative growth, the charge, the size and the fruits quality [60-64]. In our experiment, the rootstock has also significantly influenced the floral induction, which is explained probably by the fruits charge in the going season, or the lack of carbohydrates or the release of safety compounds, essentially the gibberellins [19,65]. Nonetheless, *Macrophylla citrus* has produced many more vegetative shoots followed by the Citranges group, then the *Swingle citrumelo*, in total harmony with the ongoing season charge.

In our experiment's conditions, the rootstock did not only influence the flowering evolution by the floral induction control, but also the fruit drop, Rachdi et al.; JEAI, 18(6): 1-18, 2017; Article no.JEAI.36958

namely in the physiological drop phase (Drop II). This drop has been considered as an autoregulation mechanism that adjusts the number of fruits to the capacity of the tree's procurement metabolite [66].

The first wave of abscission (Drop I) is tightly linked to the hormonal equilibrium, particularly the gibberellins [67,68]. As a result, two flows of GA_3 during the flowering have most likely destroyed any significant effect of rootstock or GA_3 treatment on the drop I.

Other than the environmental conditions, such as the high temperature [69], rain, insufficient light that also affect the physiological drop, most of researchers [70-78] indicate that the abscission during physiological drop is particularly dependent on metabolic factors (carbohydrates). The carbohydrates competition between the fruits, or between fruits and shoots (inexistent in our case), probably causes a fruit drop when the fruits cannot reach a certain carbohydrates concentration threshold [72], explains the drop of most of the fruits located on non-leaved inflorescences, followed by multiple flowers inflorescences. The rootstock affects the tree vigor [63], thus affecting the carbohydrates development, which means that it directly contributes to the physiological drop control. Even though the Macrophylla citrus rootstock, known by its vigor [27], have had a low drop in fruits percentage, followed by C35 citrange and then the other rootstocks, explained by the number of leaves and the inflorescences foliar area [79], especially for unique flower leaved inflorescences, the statistical results show a significant effect of the rootstock on the number of leaves in the fruit set stage, with a number superiority for the Macrophylla citrus. Otherwise, by the mineral absorption rhythm.

The citrus rootstock affects the yield by influencing the nutritional status of the tree [80] by the water absorption capacity [81] and the minerals [60], by taking in consideration the other parameters such as the edapho-climatic conditions and the cultural practices which simulate the rootstock behavior [80].

The rootstock had a significant effect on the foliar levels of Ca, Mg, K, P, S, Zn, matching with Georgiou's results [82]. However, the rootstock effect on Mg is already verified, the "Lemons" (*Macrophylla citrus*), on the contrary to the Citranges have generally problems with the Mg absorption [82], thus explaining the low levels.

	Rootstock							
	MC	C35C	CC	SC	тс	Sig		
AWF(g)	129	111	103	99	94	-		
NSS	2126 a	2120 a	1531 b	1040 c	1386 b	***		
YLD	50 a	37 b	28 c	25 c	23 c	***		

Table 11. Estimated yield for each rootstock in tons/hectare

The numbers followed by the same tiny letter are not significantly different

Sig: Significance (Duncan: NS, *, **, *** Not significant or significant at P = 0.05, 0.01, 0.001, respectively)

More than Mg, *Macrophylla citrus* also have low foliar levels when it comes to Ca and P, while as Sanz [70] says P decreases during the flowering and then increases progressively in the following months, this being associated to a high demand by the hooked fruits, producing an accentuated deficit. Nevertheless, K is at low levels among *Carrizo* and *Troyer citrange*.

A level varying from normal to low of N, as reported by Georgiou [80], is maybe a result of the conducting of a fertilization/irrigation process or of the dilution effect [83]. We also note low foliar levels of B and Cu, essentially due to the low levels of this elements in the soil and the water. Highly to very highly levels of sodium are caused by a bad management of potassium fertilizers [RACHDI, unpublished data]. Moreover, *Macrophylla citrus* shows a relatively high level in Mn, as it has been described by Wutscher [60].

Finally, the rootstock effect evaluation on the trees nutritional states, cannot be independently studied of its biotic and non-biotic surroundings, such as the stock levels of nutritive elements, weather condition or cultural practices....

The fruit caliber evolution is mainly influenced by the rootstock and the type of inflorescence from a side, and by the temperature on the other side. However, the rootstock *Macrophylla citrus* allows the most important evolution to the fruit, with a more advanced caliber profile, followed by *C35 citrange*.

From the gathered data, we could predict the yield (**YLD**), the number of spring shoots (**NSS**) for each rootstock. The yield in tons/hectare is computed from the number of fruits per tree and the average weight of the fruit (**AWF**), Also the spring shout from the number of drop in fruits per tree and per shoot.

The drop study should always englobe the number of fruits per shoot as the number of

shoots, and if these conditions are not met, we will have false results.

The components of the yield are as follows: the number of fruits per shoot, the number of shoots and the average weight of a fruit. Therefore, the tree always finds rise of compensation between the different components, taking into account the biotic conditions, for each physiological stage.

5. CONCLUSION

To conclude, the application of gibberellic acid during the coloration turn of the fruit, especially with the existence of autumn shoots does not represent any risk on the floral induction and the flowering evolution of the previous year, nor on the nutritional state of the trees, while we could reach very satisfying results by increasing the dose.

We intend to favor the Citrange C35 rootstock as an appropriate choice for the Atlantic coastal zone of northern Morocco

The diversification of rootstocks is always researched in order to face the complex set of risks we can encounter. In the experiment condition, we count to favor the *C35 citrange* rootstock as an appropriate choice for the coastal zones in north of Morocco, known by a sandy soil, giving us a good adaptation potential, yield and fruit caliber, while focusing on the fruit quality and the production lateness.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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