## Improved Use of Foliar Urea on Clementine Mandarin to Manipulate Cropping in a Sustainable Production System

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#### Abstract

Cadoux', Clementine mandarin (Citrus reticulata Blanco) is an alternatebearing cultivar with problems of low fruit set and small fruit size. Properly timed foliar-applied urea has been shown to increase flower intensity, fruit set, fruit size and yield of citrus. In this research, we investigated effect of some spray factors on efficacy of foliar urea on fruit set, fruit size, and yield of 'Cadoux' clementine mandarin. In one trial, urea (containing 46%N) was applied as a foliar spray to fully mature trees during the periods of flower initiation-differentiation, fruit set and 'June drop'. Urea was used at the rate of 1 kg/100 l and trees were sprayed to the point of run-off. The experiments were run for two consecutive years. Urea increased leaf area, leaf specific dry weight (mg/cm<sup>2</sup>), leaf N levels and total yield. The yield increase was due to an increase in both fruit number per tree and fruit size. As a consequence, the total number of export grade fruit (diameter > 51 mm) was increased by  $\approx 50\%$  regardless of timing of urea application. More fruit were of export grade by the first harvest date indicating an indirect effect of urea on earliness. Incorporation of Mn and Zn in the spray mix with an application at flower initiation-differentiation increased yield to a level similar to that obtained with acidification to pH 6.0. Multiple urea applications during flower initiation-to-bloom did not lead any additional benefit when compared to a single application. In another 2-year trial we investigated the effect of the pH of the spray solution and found that lowering the pH from 7.6 to 6.0 increased leaf N and cumulative yield.

#### **INTRODUCTION**

Urea is an efficient and low cost fertilizer that can be used in citrus foliar nutrition for its numerous benefits. If used at the proper time and concentration, it can replace, at least in part, soil N applications and thus reduces the risk of leaching of the commonly used nitrates into the ground-waters (Council for Agricultural Science and Technology, 1985). In particular, in winter, when the root system is less active, N nutrition through leaves can supply the nitrogen quantities necessary for cell/tissue renewal and activity and, particularly in species such as citrus, Lovatt and her co-workers have reported a correlation between levels of some N-compounds such as the polyamines and NH<sub>3</sub>-NH<sub>4</sub><sup>+</sup> in the leaves, and flower intensity (Ali and Lovatt, 1994; Lovatt et al., 1992a). In citrus, flower initiation mostly occurs in winter months when nutrient absorption by the roots is reduced or even lacking (Lovatt et al., 1992a). Urea uptake by leaves is reported to be under the influence of factors such as the chemical formulation of the urea used, leaf age, pH of the spray mix and environmental conditions (Cook and Bonyton, 1952; Bondada et al., 2001; Orbovic et al., 2001).

Many clementine mandarin cultivars such as 'Cadoux' and 'Nour' have an alternate bearing pattern in that in one year (the "on year"), it flowers profusely and sets a heavy crop with small-sized fruit often of an unmarketable value and the next year (the

"off year"), it produces very few flowers that set less than optimum number of fruits often too large in size with low overall quality. Therefore, enhancing flowering when an "off year" or a low flower intensity is predicted may regulate year-to-year yields and improve fruit quality. In earlier reports, we have documented that properly timed winter urea sprays (at 0.8 to 1.6%) enhanced flowering and that the response was greater on trees coming out of an "on year" (El-Otmani et al., 1998a, 1998b). We later showed that the December-January treatment gave the greatest increase in yield in kg and in fruit counts with an increase in fruit size (El-Otmani et al., 2000).

This paper addresses the following questions: i) is urea beneficial at all stages of flower/fruit development?, ii) does the pH of the spray mix have any effect on leaf N uptake and on tree response? iii) to reduce the number of necessary sprays/year and increase spray efficacy, can the spray mix include other compounds such as micronutrients (Mn, Zn, for example) or pH modifiers without any negative impact such as phytotoxicity, reduced effect of urea, ...)?

#### MATERIALS AND METHODS

The experimental orchards were located in the Souss Valley of Southern Morocco characterized by a semi-arid climate with rainfall of  $\approx 220$  mm/year and minimum and maximum temperature of  $\approx 13$  and 25°C, respectively. Soils are of the clay silty type with pH  $\approx 8.4$  and the water has a pH  $\approx 7.4$ . Irrigation was by microsprinklers and the fertilizers were routinely supplied to the soil with the irrigation water.

Commercially available urea (containing 46% N) was used in these experiments and quantities of N supplied by the treatments used are in addition to the routine N applications made by the grower (i.e. 180-222 Kg N/ha applied from February to August)..

The trials were carried out in commercial groves of 'Cadoux' clementine trees (*Citrus reticulata* Blanco) grafted on sour orange rootstock (*Citrus aurantium* L.) and planted in 1969 at a spacing of 6 m x 6m. The trees were apparently healthy and showed no nutritional deficiency or disease symptoms.

# Experiment 1: Effect of urea application at various flower/fruit development stages with acidification or incorporation of MnZn in the spray mix used at flower initiation-differentiation

Urea was applied at 1% (w/v) and at pH = 7.6 (which corresponds to the normal pH of the spray mix using well water of the region) to whole trees at various stages of flower/fruit development and for 2 consecutive years (1997, an "on year" and 1998, an "off year") (Table 1). An additional 2 treatments applied at flower initiation-differentiation were tested in 1998: 1) urea at 1% (w/v) acidified to pH=6.0 using nitric acid, and 2) urea at 1% (w/v) plus Mn and Zn (at the recommended rate of 300 g of a granular product (Mangozinc®) containing 16% Mn and 11% Zn, in 100 l).The treatments were applied to the point of run-off with  $\approx$  10 l of the spray mix/tree. Because leaf N is greatest at 48 hours after treatment (Lea-Cox and Syvertsen, 1995; El-Otmani et al. 2002) total leaf N content was analysed on leaf samples taken 48 hours following urea application. Six to 7 months old leaves were sampled in October from the current year's spring growth flush. Composite samples of 20 leaves each were obtained from each 2 adjacent trees using the four tree quadrants. Four replications were used for each treatment. Total N was obtained using the Kjeldahl method (Chapman and Pratt, 1978).

Harvest was done according to fruit size and color and fruit was sorted according to size using a precalibrated packing line. Total yield (in kg and in fruit counts), and potentially exportable yield (i.e. fruit of diameter > 51 mm) were determined. A completely randomized experimental design was used with 16 single-tree replicates in both years (1997 and 1998).

Data were subjected to an analysis of variance and mean separation was carried out using the Newman-Keuls test at the 5% level.

# Experiment 2: Effect of pH of the urea solution and of single vs multiple urea applications on leaf N and fruit yield parameters

In one test, a multiple application (using 3 sprays) of a 1% urea solution at pH = 7.6 was compared to a single application of the same solution at the same pH. In another test, effect of the pH of the 1% urea spray mix was assessed using applications at pH = 7.6, 6.5, 6.0 and 5.5. The pH was lowered using phosphoric acid. The experiment was repeated for 2 consecutive years (1989 and 1999). Applications were made during the winter period to early spring (Table 2) and treatments were applied to the point of run-off with each tree receiving  $\approx 101$  of the spray mix. In 1998, the trees were in the "off year", whereas in 1999 they were in the "on year".

For both seasons, total leaf N content following urea application was monitored during 30 days after each application. Sampling of the leaves begun immediately before each urea application and continued at 2, 7, 15 and 30 days after. Composite samples of 20 leaves were obtained from each 2 adjacent trees. Leaves were sampled from the four quadrants of the tree using the summer growth flush of the current growth cycle located on the outer canopy. Four replications were used for each treatment. Total N was obtained using the Kjeldahl method (Chapman and Pratt, 1978).

At harvest, fruit was sorted and yield was analyzed as for Experiment 1. A completely randomized experimental design was used with 13 single-tree replicates in 1998 and six two-tree replicates in 1999.

Statistical analyses were run on the data as for Experiment 1 and, in addition, a linear regression analysis was run for cumulative yield and leaf N content against pH of the urea solution.

#### **RESULTS AND DISCUSSION**

#### Effect of Repeated Applications of Urea on leaf N Content and Yield

Urea significantly increased leaf N levels for 1998 (Fig. 1) and 1999 (El-Otmani et al., 2002). The greatest leaf N content was recorded 2 days after each application and declined thereafter, confirming information reported by Lea-Cox and Syvertsen (1995).

In 1998 (i.e. the "off year"), at the start of the experiments on 24 January, leaf N content was  $\approx 1.70 - 1.79\%$  and, 30 days after the 3rd urea application, leaf N was  $\approx$ 1.83% in the untreated trees and 1.98% in the urea-treated trees; these differences are not significant, however. In 1999 (i.e. "on year"), leaf N was 1.83% at the start of the experiment (23 Jan. 1999), but thirty days after the 3rd application, it was 1.81% for untreated trees and 2.07% for the urea-treated trees and this difference was significant at the 5% level. It is not clear whether the effect was on flowering or was due to N deficiency overcome by treatment but we had reported that urea applied as a foliar spray during the period of October-January increased flower intensity and/or fruit set (El-Otmani et al., 1998b). Taken together, these results indicate that tree coming out of an "on year" (i.e. 1997) and going into an "off year" (1998) had their N reserves depleted and that those trees going into an "on year" (1999) had greater reserves but were also depleted. In addition, during the "off year" (i.e. 1998), it can be speculated that absorbed N was probably rapidly mobilized to stronger sinks such as new growth, new roots, etc. where needs are much greater. Besides that, El-Hila (1996) reported that increased leaf size and leaf specific dry weight (see below) indicating an enhancement of dry matter synthesis in the leaf as a result of urea application.

Total yield and potentially exportable yield were significantly increased as a result of single urea application and the degree of increase was  $\approx$  76% for total yield (in kg/tree) and 91% for export yield in 1998 which is an "off year" and  $\approx$  23% and 13%, respectively, in 1999 which is an "on year" (Table 3), indicating that this treatment if properly timed can significantly reduce alternate-bearing, which is a major challenge for clementine citrus growing and particularly for producing a seedless but economically valuable crop. In addition, increased yield was mainly the result of increased fruit number since fruit size was not changed (Table 3), completing results of our earlier work when we have shown that urea applied prebloom increased flower number and fruit set and that the effect was greater in the "off year" (El-Otmani et al., 1998a; 1998b).

Furthermore, total yield (both in fruit number and weight) as well as potentially exportable yield were not statistically different between trees receiving a single urea application and those receiving three applications at one-month interval during the period of prebloom-to-full bloom (Table 3). This indicates that at this physiological stage, one application (at 1%) would be sufficient to achieve an increase in yield. Moreover, it can also be assumed that at this specific stage there is a level of saturation in N (probably corresponding to that of the leaves 48 hrs after the first application) beyond which the tree does not yield any additional benefits in terms of fruit counts and total weight. Excess N might be allocated to other sinks such as vegetative growth and root growth whose activity also starts at this time for citrus. In addition, because total yield is always smaller in the trees that are in the "off year" regardless of the number of urea applications it is assumed that some other factor or factors are limiting since the production potential of these trees is greater than 100 kg of fruit as shown in Table 3. This limiting factor may be of nutritional (carbohydrates) or hormonal nature (Lovatt et al., 1992b).

Urea application also increased yield at the first harvest early both in weight and in proportion relative to total yield indicated an indirect enhancement of earliness due to this treatment (Table 3).

#### Effect of pH of the Urea Solution on leaf N and Cumulative Yield

For the 2 years of trial, urea increased leaf N content (measured 48 hours following treatment application) at all pH values tested with the greatest increase recorded in the pH range of 5.5-6.0 (Table 4). Furthermore, the equation of the linear regression between the leaf N means (Y in %) and the pH of the urea spray mix (X) was in the form of Y=2.97 - 0.12X (with r = 0.91 with 2 degrees of freedom) for 1998 and Y=3.19–0.07X (r = 0.89) for 1999 indicating that 81% of the variation observed in leaf N was due to the pH of the spray mix.

For the trees coming out of an "on year" and treated with urea (i.e. treatments applied in 1998), leaf N content was lower and the increase in leaf N was smaller than that recorded for the same trees coming out of an "off year" (i.e. the treatments applied in 1999) indicating depletion of tree N reserves in the "on year" (i.e. 1997) and their reconstitution during the "off year" (i.e. 1998). In addition, the results also indicate that N absorption was greater in the leaves that already have high N levels. A similar conclusion was reported on apples by Cook and Bonyton (1952) who showed a direct relationship between high urea absorption and high N levels in the leaves. In citrus, Bondada et al. (2001) also reported that N deficient leaves had lower N absorption than N sufficient leaves. As a result, yield was lower in 1998 and significantly greater in 1999 (El-Otmani et al., 2002). In addition, the linear relationship between treatment means of cumulative yield (Y in kg/tree for 1998 + 1999) and pH of the urea spray solution (X) was in the form Y = 285.2 - 10.8X with a correlation coefficient r = -0.726 with 2 degree of freedom indicating that 53% of the variation observed in yield was due to the pH of the spray mix. It can be assumed that the effect of treatment on yield was likely due to enhanced flowering and/or fruit set due to increased leaf N as a result of lowering the pH of the urea spray solution.

# Effect of a Single Urea Spray (with or without Acidification or MnZn) Applied at Various Stages of Flower/Fruit Development

For all of the treatments used in this study, leaf N (measured in leaf samples taken in October 1998 from the previous spring flush) was in the range of 2.38-2.55% which is considered optimum for the region, whereas that of untreated trees was  $\approx 2.07\%$ .

Urea applied at fruit set or during the "June drop" period increased yield (cumulative as well as per year) with the same magnitude as it did when applied at flower initiation-differentiation (Table 5). This increase was due to an augmentation in both fruit counts and weight. The increase due to fruit counts was somewhat greater for all of the treatments applied during flower initiation-differentiation compared to these applied at "June drop" indicating a greater effect of urea on flower intensity and confirming reported results by El-Otmani et al. (1998a, 1998b). The opposite trend was observed for fruit size (Table 5) indicating a greater effect on fruit size when urea was used during early stages of fruit enlargement (i.e. "June drop" stage). In addition, export yield was almost doubled as a result of the double effect of the treatment on fruit counts and on fruit size (Table 5). In our laboratory, El-Hila (1996) showed that urea applied at these stages of flower/fruit development increased leaf size, leaf dry weight and leaf specific dry weight(mg/cm<sup>2</sup>). It, therefore, can be assumed that increased leaf size and increased leaf specific dry weight were a feed-back response of the tree to increased number of generative organs (flowers, fruits) as these are strong sinks and it can also be assumed that they exert a stimulatory effect on synthesis of dry matter in the leaf which is then translocated to the developing sinks (flowers and fruits in particular). In addition, leaf specific dry weight had the biggest value (= $9.3 \text{ mg/cm}^2$ ) when the treatments were applied in April (i.e. fruit set), the smallest value (= $6.3 \text{ mg/cm}^2$ ) when the treatments were applied in December (i.e. flower initiation) and an intermediate value when the treatments were used in February or March (i.e. flower differentiation-to-bloom).

Furthermore, acidifying the solution from pH = 7.6 to pH = 6.0 had an additional increase in yield when the application was made prebloom (Table 5) .This, additional increase was due to an increase in fruit number but fruit size was not affected. This, together with the above study on pH effect, indicates the beneficial effect of acidifying the urea spray treatment. It is noteworthy that Orbovic et al. (2001) reported that changing urea solution pH from 8.0 to 4.0 could have a negative effect on the amount of urea penetrating the cuticle through the loss of urea from breakdown possibly due to hydrolysis. However, these authors did not measure the direct effect of solution pH on cuticular penetration or on leaf N but they examined the stability of urea in the solutions of different pH using Na-citrate buffer solution (50 mM) adjusted with NaOH. However, the chemistry of the solution once in the leaf may play a major role in the response obtained. Our results also show that adjustment of the pH of the spray mix from pH 7.6 to pH 6.0 using either phosphoric acid (Table 4) or nitric acid (data not shown) increased leaf N by  $\approx 0.15$ -0.18 units indicating a similar effect of both acids on leaf N absorption. Moreover, adding MnZn into the spray mix applied prebloom also enhanced urea effect on fruit counts with no additional benefit on fruit size (Table 5). Export yield was, therefore, not different from that of urea alone. A synergistic effect between urea and Mn and Zn was reported by El-Hila (1996) and by Rajput and Kumar (1990) who indicated greater yield benefits from urea sprays when they were combined with Zn or Mn.

As conclusions, in foliar nitrogen nutrition, and for optimum results, timing of urea application and conditions of its use are critical. Our results indicate that a single application during flower initiation-differentiation gave similar results as multiple application. Single application during fruit set and "June drop" were also efficient in increasing yield. Such applications should therefore be included in fertilizer programs to meet, at least partially, the nitrogen needs of trees during periods of high demand (flower initiation-differentiation, fruit set, fruit growth, ...). This practice will minimize N loss and the potential for increased soil salinity and for increased soil and water pollution by other N sources such as nitrates. It also appears to be one efficient way to reduce alternate-bearing of 'Cadoux' clementine mandarin. Acidification of the spray mix to pH = 6.0 and incorporation of micronutrients such as MnZn in the spray mix improve efficacy of such treatments in improving tree leaf N status and dry matter content and increasing yield.

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### **Tables**

Table 1. Stage and time of application of a 1% (w/v) urea solution (pH=7.6) to 'Cadoux' clementine mandarin trees.

Stage of flower/fruit development	Treatment date
Flower initiation-differentiation	3 February in 1997 and 21 January in 1998
Fruit set	21 March in 1997 and 18 March in 1998
End of "June drop"	16 May in 1997 and 4 May in1998

Table 2. Phenological stage and timing of single and multiple application of a 1% (w/v) urea solution to 'Cadoux' clementine mandarin.

Treatment	Date of treatment		Phenological stage
	1998	1999	
Untreated control	-	-	-
Repeated applications	24 Jan. +	23 Jan +	Flower initiation-differentiation +
(pH=7.6)	20 Feb.+	2 Mar. +	Match head green-to-white buds +
	24 Mar.	2 Apr.	Full bloom
Single applications at			
pH=7.6, 6.5, 6.0, 5.5	24 Jan.	23 Jan.	Flower initiation-differentiation

Table 3. Effect of a single *vs* a multiple application of a 1% (w/v) urea foliar spray (pH=7.6) during the period of flower differentiation-to-full bloom on yield parameters of 'Cadoux' clementine mandarin during 2 consecutive years (1998 and 1999).

Year	Treatment	Total yield		Yield at first		Fruit size	Export
				harv	harvest		yield
		kg/tree	Fruit	kg/tree	% of		(kg/tree)
			counts/		total		
			tree				
	Untreated	45.6b <sup>z</sup>	721b	35.2	77	63.2	29.1b
	Single						
	application	80.2a	1251a	68.7	86	64.1	55.7a
	Multiple						
1998	applications	80.4a	1285a	64.5	80	62.6	53.4a
	Significance						
	level <sup>y</sup>	***	***	-	-	NS	***
	Untreated	85.5b	1376b	62.3	73	62.2	51.0b
	Single						
	application	109.1a	1775a	87.3	80	61.5	67.0a
1999	Multiple						
	applications	104.9a	1763a	79.3	76	59.5	57.6a
	Significance						
	level	***	***	-	-	NS	**
	Untreated	131.1	2097	97.5	74	-	80.1
Cumulative	Single						
1998+1999	application	189.3	3026	156.0	82	-	122.7
	Multiple						
	applications	185.3	3048	143.8	78	-	111.0

<sup>2</sup> Within a given year and for a given columns, means followed by the same letter are not significantly different at 5% level (Newman-Keuls test)

<sup>y</sup> Differences are not significant (NS) or are significant at 1% (\*\*) or 0.1% (\*\*\*) level

Table 4. Effect of pH of a 1% (w/v) urea solution, applied during flower initiationdifferentiation, on cumulative yield (i.e. 1998+1999) and on total leaf N content 48 hours following a foliar prebloom application to whole 'Cadoux' clementine mandarin trees in 2 consecutive years.

Treatments	Leaf N (%	Cumulative vield	
	Year 1998	Year 1999	(kg/tree)
Untreated trees	$1.77c^{z}$	1.83b	131.1
Urea-treated trees:			
pH = 7.6	2.08b	2.65a	189.3
pH = 6.5	2.11b	2.69a	227.9
pH = 6.0	2.26ab	2.80a	212.4
pH = 5.5	2.33a	2.78a	224.5
Significance level	0.1%	0.1%	_

<sup>z</sup> Within columns, means followed by the same letter are not significantly different at 5% level (Newman-Keuls test)

Table 5. Effect of a single urea spray (at 1% w/v) at various stages of flower/fruit development on yield parameters of 'Cadoux' clementine mandarin during 2 consecutive years (1997 and 1998).

Treatment	Year 1998				Cumulative (1997+1998)	
	Total	Fruit	Fruit	Exportable	Total yield	Export
	yield	counts/	size	yield	(kg/tree)	yield
	(kg/tree)	tree	(g/fruit)	(kg/tree)		(kg/tree)
Untreated	$40.9c^{z}$	735c	55.0b	26.5c	84.1b	53.6b
Urea at flower initiation-						
differentiation (pH=7.6)	63.1b	1051ab	61.6a	43.3b	154.3a	106.7a
Urea at fruit set (pH=7.6)	60.8b	979b	62.4a	40.3b	146.8a	100.0a
Urea at "June drop"						
(pH=7.6)	57.9b	909b	64.8a	40.0b	150.8a	106.8a
Urea at flower initiation-						
differentiation (pH=6.0)	79.5a	1300a	62.3a	47.0a	X	-
Urea at flower initiation-						
differentiation + MnZn	72.5a	1172ab	62.7a	47.3a	-	-
Significance level <sup>y</sup>	*	*	***	**	***	***

<sup>z</sup> Within columns, means followed by the same letter are not significantly different at 5% level (Newman-Keuls test)

<sup>y</sup> Differences are not significant (NS) or are significant at 5% (\*), 1% (\*\*) or 0.1% (\*\*\*) level

<sup>x</sup> not determined for 1997

## Figures



Fig 1. Changes in leaf nitrogen following repeated foliar applications of 1% urea solution to mature clementine trees. Time of treatment application was 24 Jan., 23 Feb. and 25 Mar. 1998. Significant differences (p<5%) at all determination dates except at day 0, 30, 60 (i.e. immediately prior to application) and 90 (i.e. end of experiment).