The action of 8-hydroxyquinoline and chlorine in the durability of the postharvest torch ginger variety Red Torch

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Abstract

It had as objective to evaluate the effect of 8-hydroxyquinoline and chlorine, isolated and mixed in and conservation of the stem stick of the emperor. Twenty of the stem torch ginger (Etlingera elatior), variety Red Torch were harvested, transported, and immersed in standardized solutions containing biocides for 48 hours. The biocide used were 100 mg L⁻¹ of chlorine; 250 mg L⁻¹ of 8-(HQC); 500 mg L⁻¹ of 8-(HQC) and 50 mg L⁻¹ of chlorine + 50 mg L⁻¹ of 8-(HQC), the control corresponded to water. The stems stay immersed in water for nine days at 20 ± 4 ºC. Every 24 hours, the water in the container was replaced. During nine days, some measurements were realized of pH of the water in which the stems stay immersed, in the same way, it was measured the consumption/loss volume of the water for each 24 hours in the containers. The stems were weighed and also samples were taken from the base of the stems and pear ligule determination of peroxidase activity. In general, the use of solutions of 8-(HQC), 250 and 500 mg L⁻¹, make easier the flux of water in the vases, measured by the consumption of water; provided lower peroxidase activity at the base, independent of the pH of the solution and maintained lower peroxidase activities in the ligule. Thus, with the results obtained, it demonstrated promise the use of 8-(HQC), in the highest concentrations tested (250 and 500 mg L⁻¹) for quality maintenance of cut stems of Etlingera elatior, variety Red Torch.

Key-words: Conservation, Tropical Plants, Longevity, Enzyme

Introduction

The Etlingera elatior (Jack) R. M. Smith species belongs to the family Zingiberaceae, popularly known as stem stick of the Emperor or torch ginger, and present large and showy inflorescences. In northeastern of Brazil, it is cultivated with inflorescences varied coloring, for exemple, in white pink color called porcelain pink color, in darker pink color Pink Torch and red color Red Torch (Loges et al., 2008) tulipa.

The use of technology increases the production of flowers, especially in post-harvest. The technologies include the use of refrigeration (Aghdam et al., 2016), packaging (Shimizu-Yumoto and Ichimura, 2016), conservative substances such as sugars (Norikoshi et al., 2016), acids (Costa et al., 2016) and growth regulators (Nowak and Rudinick, 1990; Marsala et al., 2014). Still, the use of biocides (Van Doorn and Perik, 1990). These substances are applied by immersion, denominated pulsing or by spraying (Norikoshi et al., 2016).

The citrate of 8-hydroxyquinoline 8-(HQC) and the 8-hidroxiquinolina 8-(HQ) are commonly used as the biocide. The Compounds of microbial action, the genuine 8 hydroxyquinoline or its sulfate ester 8-(HQS) or citrate 8-(HQC) at concentrations of 200 to 600 mg L⁻¹, has been widely used because of its efficiency (Rogers, 1973). They reduce the pH of the maintenance solution by preventing the vascular blockade in cutting-flowers, including cutting-roses (Van Doorn and Perik, 1990). The 8-(HQ) has strong antimicrobial properties that eliminate vascular blockade and increase water absorption by
keeps hydric balance per reducing the transpiration of flower tissues (Rogers, 1973; Jowker, 2005). A solution containing 8-(HQC) and saccharose is routinely used to prolong life vase in cutting-flowers (Łukaszewska and Skutnik, 2003). The maximum life of the vase and the floral diameter of the cultivate cut Vaibhav, Mexican Single, Shringar, Suvasini and Prajwal were obtained when the stems were immersed in sucrose (2%) + 8-(HQC) (200 ppm) + AgNO₃ (50 ppm) in to control (Sudagar et al., 2010).

The stems of the torch ginger, with semi-open inflorescences (Mattos et al., 2017) or with 1/3 or 2/3 of aperture (Carneiro et al., 2014) they had longer durability comparing with the open inflorescences, however, to commercial objective it is considered the consumer preference, the time of the transport and the handling favored by the more closed inflorescences. These stages of open floral influence directly the wastefulness of the fresh substance with high transpiration and/or obstruction of the xylem vase, which cause wilting and lower durability (Nowak and Rudinick, 1990).

During senescence of the tissues produce enzymes like the peroxidases (POD) , which act in the darkening of the tissues. The use of some substances such as gibberellic acid (GA₃) in Heliconia (Guimaraes et al., 2014) and 8-hydroxyquinoline in gladioli (Casares et al., 2017) can intervene in decrease of the enzyme activity and reduce the phenolic compounds, that operate in the darkening occasioned by these enzymes.

To minimize the loss of the stem stick of emperor in the postharvest it was realized stowage with the bactericide 8-(HQC) and of course per 48 hours, with the objective to verify their effects in the durability of their actions in the base structures of the stem and ligule.

**Material and methods**

*Obtaining and transporting the main material*

Twenty stem of the torch ginger, variety Red Torch (*Etlingera elatior* (Jack) R. M. Smith) were harvested with closed bracts, the Mumbecas Farm is located in Paulista, Pernambuco, Brazil, with geographical coordinates 7º56'20” S and 34º55'06,57” W. The stems were hydrated, immediately after harvesting, and transported to the Laboratory of the Vegetable Production Nucleus program of the Postgraduate in Plant Production of the Federal Rural University of Pernambuco, Academic Unit of Serra Talhada, in Serra Talhada, Pernambuco State, Brazil (7º56'20” S; 38º17'31” W and altitude of 499 m), Brazilian Semiarid Region.

**Handling and disposal of treatments**

The stems were standardized to 40 cm in length, duly identified and distributed, randomly. The stems bases were immersed for 48 hours, “pulsing” with different concentrations biocides 8-(HQC) and Chlorine: 100 mg L⁻¹ Chlorine; 250 mg L⁻¹ of 8-(HQC); 500 mg L⁻¹ 8-(HQC) and 50 mg L⁻¹ Chlorine + 50 mg L⁻¹ of 8-(HQC), the control corresponded to water immersion (Fig. 1). After 48 hours, the stems were replaced by water treatment to 20 ± 4 °C, for nine days. Every 24 hours the water of each container was replaced to prevent the proliferation of microorganism.

![Fig. 1. Overview of the experiment with floral stems torch ginger (*Etlingera elatior* (Jack) R. M. Smith), variety Red Torch in PET bottle.](image)

**Temperature and humidity during storage were controlled by a thermo-hygrometer (Model 7663.02.0.00, Incoterm Inc., Porto Alegre, Brazil) and were continuously recorded.**

**Realized Analyzes**

It was measured the volume of the solution consumption, the pH of the fresh material weighs every two days, while the determination of peroxidase activity in the base of the stem and ligule was performed every three days for nine days. The peroxidase activity were labeled, frozen in liquid nitrogen, and stored in a freezer (-80 °C).

**Solution Consumption**
Initially it was placed 1000 mL of water was placed in the respective containers. Consumption of the solution volume was measured by measuring for each two days the difference between the initial volume and the final volume, using a beaker in which water exchange occurred every 24 hours.

**pH of the solution**

The pH measured of the solution was performed, every three days, using a digital bench potentiometer model of bench model PHS-3E EVEN. It was calibrated in standard solution (pH 6.7 and 4.5) before and after immersion of the base of the stems.

**Loss of fresh mass**

The loss of fresh mass was obtained by the perceptual relation between the initial stem mass and the final mass, on each day of evaluation, express percentage (Awad et al., 2017), according to the following equation: 

\[
PMF = \frac{(MFI - MFF) \times 100}{MFF},
\]

wherein: PMF = loss of fresh mass (%), MFI = initial fresh mass (g) and MFF = final fresh mass (g), in an interval of two days.

**Extraction and assay of peroxidase (POD)**

It was concluded the extraction was according to the methodology described and adapted by Simões et al. (2015). With the support of liquid nitrogen, 50 mg of fresh tissue of the stem base and the ligule, it was homogenized separately, in 6 mL of 0.2 M sodium phosphate buffer (pH 6.0) previously maintained at 4 °C. The extract was centrifuged at 9,000 rpm for 21 minutes at 4 °C. The POD assay was determined by the addition of 60 μL of phosphate stopgap 0.2 M (pH 6.0), 100 μL of guaiacol (0.5%) and 150 μL of the supernatant for enzymatic extract to ligule and base of stem, respectively, to the reaction form, containing 1.0 mL of phosphate stopgap 0.2 M (pH 6.0), 100 μL of guaiacol (0.5%) and 150 μL of hydrogen peroxide (0.08%). The reading process was completed by the spectrophotometer (Biochrom; scale model S8) at 470 nm, at a temperature of 30 °C, every 30 seconds for two minutes. The peroxidase activity was expressed in UE gMF⁻¹ min⁻¹.

**Design experimental and statistical analysis**

The experimental design was randomized, 5x4 factorial scheme, it has been: five treatments: water; Chlorine 100 mg L⁻¹; 250 mg L⁻¹ 8-(HQC); 500 mg L⁻¹ 8-(HQC) and 50 mg L⁻¹ Chlorine + 50 mg L⁻¹ 8-(HQC) and four days trial, with four replications. Each experimental unit was composed of a floral stem. Analysis of variance was performed to detect differences between treatments and were compared by Tukey test at 5% probability.

**Results and discussion**

The agents used in the present work, Chlorine and 8-(HQC), have a biocidal action, preventing the proliferation of microorganisms (Van Doorn and Perik, 1990; Macnish et al., 2008), which can cause vascular block (Rani and Singh, 2014).

It was verified that the consumption of solutions, containing biocidal or not (control) was reduced in the process of the conservation of stems (Table 1). Observing that the stems that were in the solution of 8-(HQC), they were which showed at the end of the conservation period the consumption the biggest values of consumption, average 26.3 mL (Table 1). While the stems were submitted to the mixture containing chlorine and 8-(HQC) present a medium consumption of 11.2 mL (Table 1). This demonstrate that the biocidal solutions can have contributed, in different way, reflecting in different degrees in the blocking of water in the conduction per xylem (Rani and Singh, 2014). Furthermore, the mixture of biocide solution made possible the lower consumption during storage (Table 1). On the other hand, the stems subjected to treatments containing only 8-(HQC) showed higher consumption of solutions and, consequently, a better durability postharvest stems.

### Table 1. Mean values of solution consumption (mL) by the stems of torch ginger (*Etlingera elatior* (Jack) R. M. Smith) variety Red Torch.

| Treatments                          | Storage Time (days) | CV%  |
|-------------------------------------|---------------------|------|
|                                     | 2       | 4       | 6       | 8       |       |
| T1 - Control                        | 32.50 aA  | 26.25 aA | 18.75 aAB| 10.00 aB|       |
| T2 - 100 mg L⁻¹ Cl                  | 38.75 aA  | 21.25 aB| 20.00 aB | 15.00 aB|       |
| T3 - 250 mg L⁻¹ 8-(HQC)             | 35.00 aA  | 26.25 aAB| 21.25 aAB| 8.75 aB |       |
| T4 - 500 mg L⁻¹ 8-(HQC)             | 35.00 aA  | 22.50 aAB| 22.50 aAB| 8.75 aB |       |
| T5 - 50 mg L⁻¹ Cl + 50 mg L⁻¹ 8-(HQC)| 28.75 aA  | 20.00 aA | 17.50 aA| 17.50 aA|       |
| Value of F                          | 0.3828 ns  | 0.3238 ns | 0.3051 ns | 0.6206 ns |       |
| CV%                                 | 23.09    | 24.44    | 24.79    | 30.04    |       |

Means followed by the same vertical letters (lowercase) for storage time and horizontal (uppercase) for treatments do not differ from each other by the Tukey test at 5% probability. *: significant at 5% probability. **: significant at 1% probability. ns: not significant.
The 8-Hidroxiquloline, beyond the biocide action, also appears to influence in the levels of polyamines in bird-of-paradise (Vieira et al., 2017). In the present work, the solutions with 250 and 500 mg L\(^{-1}\) of 8-(HQC) were effective in the strengthening of the stem torch ginger. In roses, one of the effects of using 8-(HQC) is to reduce the pH, preventing vascular blockage (Van Doorn and Perik, 1990). In the present work, the solutions containing only the mixture of Chlorine and Chlorine and 8-(HQC) were the more reduced pH, even though significantly until the second day (Fig. 2).

![Fig. 2. pH of solutions containing Chlorine and 8-(HQC), either individually or mixed during storage of torch ginger (Etlingera elatior (Jack) R. M. Smith), variety Red Torch.](image1)

When it was quantified the loss of the accumulated fresh mass of the stems, it was verified that those kept chloro (isolated) and 8-(HQC) at 500 mg L\(^{-1}\), demonstrated higher loss of weight values of fresh mass at the end of the experiment (Fig. 3). However, these minimum values were about 4 and 3% for the stems held in chloro isolated and 8-(HQC) 500 mg L\(^{-1}\), respectively. For the other treatments was observed mass gain at the end of the conservation period, demonstrated by the negative values of the accumulated percentage of the loss fresh mass (Fig. 3).

![Fig. 3. Loss of accumulated fresh mass (%) on torch ginger (Etlingera elatior (Jack) R. M. Smith), variety Red Torch, chlorine containing biocidal solutions end 8-(HQC), insulated and mixed.](image2)

The (loss of fresh mass presented in this study may be divided to the process of transpiration, increased respiration) (Mansouri, 2012) and, as well, the reduction of water conductivity during senescence of the floral stems (Dias-Tagliacozzo et al., 2005). The process of transpiration consist in an of the principal causes of deterioration, resulting, principally, in a change in the appearance, due to wilting (Ribeiro et al., 2010). This was most evident in the handing with 100 mg L\(^{-1}\) Cl.

They were determined peroxidase (POD) activities on the basis of the ligule stems (Fig. 4 and 5). It was observed that the POD activity increased, regardless of the treatments imposed on the stems (Fig. 4 and 5). Furthermore, it was observed that in absolute value the activity of POD was bigger in the ligule in relation to the base of the stems, presumably, by the biggest metabolic activity of the organ.

![Fig. 4. Peroxidase activity (10\(^4\) x UE gMF\(^{-1}\) min\(^{-1}\)) in the base of the torch ginger (Etlingera elatior (Jack) R. M. Smith), variety Red Torch submitted to different strengthening solutions.](image3)

![Fig. 5. Peroxidase activity (10\(^4\) x UE gMF\(^{-1}\) min\(^{-1}\)) in the ligula of torch ginger (Etlingera elatior (Jack) R. M. Smith) variety Red Torch submitted to different strengthening solutions.](image4)

It was also observed that the stems held in 100 mg L\(^{-1}\) of Chlorine and 500 mg L\(^{-1}\) 8-(HQC) demonstrated smaller increases in the POD activity in the base and the ligule, comparing the beginning and end of the storage (Fig. 4 and 5). On another
hand, the largest increments in the POD activity in the stem base, until 6 days, were observed in the immersed stems in the water and mixture of chlorine and 8-(HQC) (Fig. 4 and 5). This may suggest a bigger vascular blockade, as observed in the lower water consumption (Table 1). The most consistent activity manipulated by the handing 250 mg L⁻¹ 8-(HQC) and 500 mg L⁻¹ 8-(HQC) and 500 mg L⁻¹ 8-(HQC) may have been favorable, because the POD is a combat enzyme of free radical (Scandalios, 1993), permitting better maintenance of the membranes and the cellular metabolism in the long , and consequently benefiting the quality of the flowers. In this study, this variation of the activity according to the function of the treatments should be further assessed, considering that the POD enzyme is directly linked to the lignifications of the tissues, which polymerizes lignin from the hydroxyl oxidation of phenolic groups (Pascholati and Leite, 1995; Castro et al., 2005), which makes important to defense and sustain the plant.

The reduction of pH in the range 0.5 to 0.7 units, until the first two days in the stems held in the mixture of chlorine and 8-(HQC) (Fig. 2) does not result in falling in the POD activity, such as reported by Vieira et al. (2016). In the present work it was reverse, the mixture of biocides, Chlorine and 8-(HQC), in the tested concentrations stimulate the POD activity until 6 days, similar to the control. This suggests that the stems maintained in the solutions containing 250 and 500 mg L⁻¹ of 8-(HQC) maintained lower lignification and less obstruction of xylem (Van Doorn and Vaslier, 2002), as evidenced by the lower POD activities (Fig. 4 and 5) and increased water consumption (Table 1).

In general aspect, the use of solutions based on the 8-(HQC), 250 and 500 mg L⁻¹ to conservation postharvest of Etlingera elatior stem, variety Red Torch, facilitated the water conduction in the vases, the measure by the water consumption (Table 1) and lower activity of POD in the base (Fig. 4) independently of the solution pH. Thus, as they maintained lower POD activities in the ligule (Fig. 4).

**Conclusion**

Thus, the results obtained demonstrated, shown promising to using of 8-HQC, in the highest concentrations tested (250 and 500 mg L⁻¹) in the quality of Etlingera elatior cutting stems maintenance, variety Red Torch. More studies are necessary to understand the physiological mechanisms.

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