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# Stress detection by laser-induced fluorescence in *Zantedeschia aethiopica* planted in subsurface-flow treatment wetlands

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## ABSTRACT

In developing countries, the use of constructed wetlands for domestic wastewater treatment has been limited despite their advantages over conventional treatment systems. In order to make the use of constructed wetlands more attractive in these countries, where the economic resources are very scarce, it is necessary to find a way that the resources invested in the construction and maintenance of the system can be recovered in a relatively short time. This could be accomplished through the combination of wastewater treatment and the production of commercially valuable ornamental plants.

There are recent studies on the feasibility to produce flowers in treatment wetlands, but there are no studies conducted to evaluate the health of the plants to assess if the plants are suffering stress from the flooding conditions of the wetlands. In this work, laser-induced fluorescence (LIF) and physical measurements were used to evaluate and compare the health of an ornamental plant – *Zantedeschia aethiopica* – under two patterns of flow in subsurface-flow constructed wetlands. The plants were studied when 7 months old and 9 months old. The fluorescence spectra and the calculated ratio F690 nm/F740 nm indicated that the plants in a horizontal flow wetland (HFW) were healthier than those in a vertical flow wetland (VFW). The physical measurements led to the same conclusion. The results suggest that the plants in the VFW were stressed because of the less water available for them under the cyclic flooding and draining characteristic in this type of wetland. It is also possible that the use of non-stratified media influenced by reducing the water–root contact time.

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## 1. Introduction

Constructed wetlands have been widely used during the past decades for the treatment of domestic wastewater because of their good efficacy at improving water quality and low operational costs. These natural treatment systems are simple to

operate and very efficient in removing nutrients and toxic organic pollutants from wastewater (Belmont et al., 2006). The emergent vegetation most commonly used in these systems includes cattail (*Typha* spp.), reed (*Phragmites* spp.) and bulrush (*Scirpus* spp.) (USEPA, 2000; Sim, 2003; Stottmeister et al., 2003; Belmont et al., 2004) basically due to the good adaptation

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that they have shown under different climatic conditions. The vigorous growth of these species in the constructed wetlands implies that they have to be harvested to assure the good performance of the system. This represents an investment of economic resources during the operation of the treatment wetland without recovering costs since these plants are generally valueless once harvested (Kivaisi, 2001). This aspect of the constructed wetlands is a great obstacle for its implementation in developing countries where the financial resources are very scarce. In this regard, recent studies have evaluated the possibility to substitute these hydrophytes with plants of commercial value with the aim that the resources invested in the construction and maintenance of the system can be recovered (Belmont and Metcalfe, 2003). Belmont et al. (2004) observed good results using ornamental plants in subsurface-flow constructed wetlands. Vaillant et al. (2004) studied the resistance of plants of pharmaceutical interest in a hydroponic system that was fed with domestic wastewater, observing good results regarding the effluent quality but very poor results on the plant growth. Zurita et al. (2006) studied five different species of ornamental plants in subsurface-flow constructed wetlands and observed good quality of the effluent as well as good development of the plants. However, these studies have omitted a quantitative evaluation of the physiological state of the plants, and therefore there is no certainty that the ornamental plants are not suffering from stress. Only Vaillant et al. (2004) have evaluated the health of medicinal plants in wetlands by means of chlorophyll fluorescence measurements. Chlorophyll fluorescence under continuous stimulation of light is, to a first approximation, inversely proportional to photosynthesis or carbon assimilation (Cervantes-Martínez et al., 2002). This means, that fluorescence intensity emitted by plants depends fundamentally on the chlorophyll content and on the photosynthetic efficiency (Subhash and Mohanan, 1994). Several studies have shown that an increase in chlorophyll fluorescence indicates a reduction in the photosynthetic activity as a result of plant stress caused by nutrient deficiency, water scarcity, and presence of disease-causing microorganisms,

such as bacteria, virus and fungi (Cervantes-Martínez et al., 2002; Gopal et al., 2002; Muñoz-Muñoz et al., 2007). The fluorescence spectrum of chlorophyll is characterized by two peaks in the red (685–690 nm) and near-infrared region (735–740 nm). The ratio of fluorescence at these wavelengths in the red portion of the spectrum (F690/F740) has already been established as a non-invasive indicator of stress in plants (Lichtenthaler and Rinderle, 1988). There are no references on the use of this powerful tool for the evaluation of the physiological state of ornamental plants used in treatment wetlands. The objectives of this study were (a) to evaluate the stress in *Zantedeschia aethiopica* under different flow patterns in subsurface-flow constructed wetlands using laser-induced fluorescence (LIF), and (b) to determine the best flow regime for the development of this ornamental plant that produces flowers with a high market value in Mexico (Belmont and Metcalfe, 2003).

## 2. Materials and methods

### 2.1. Description of the treatment wetland

This study was carried out in Ocotlán, Jalisco, Mexico where the climate is classified as warm and wet with rainfall in summer ( $AC_w$ ). The pilot-scale treatment system consisted of an 1100-L feeding tank (FT) used to store prescreened wastewater from the inlet of the wastewater treatment plant of Ocotlán. This wastewater was fed into an HFW measuring  $3.6\text{ m} \times 0.9\text{ m} \times 0.3\text{ m}$  ( $L \times W \times H$ ) and on a VFW measuring  $1.8\text{ m} \times 1.8\text{ m} \times 0.7\text{ m}$  ( $L \times W \times H$ ) (Fig. 1). Both wetlands had a planted surface of  $3.24\text{ m}^2$ . The main difference between these two wetlands was the pattern of flooding with domestic wastewater. In the HFW, the domestic wastewater was fed continuously at a rate of  $128\text{ L/d}$ , which corresponds to a hydraulic retention time of 4 days. Therefore, the roots of the plants were permanently under flooded conditions with a low level of dissolved oxygen (Langergraber and Haberl, 2001; Tousignant et al., 1999). In the VFW, the domestic wastewater

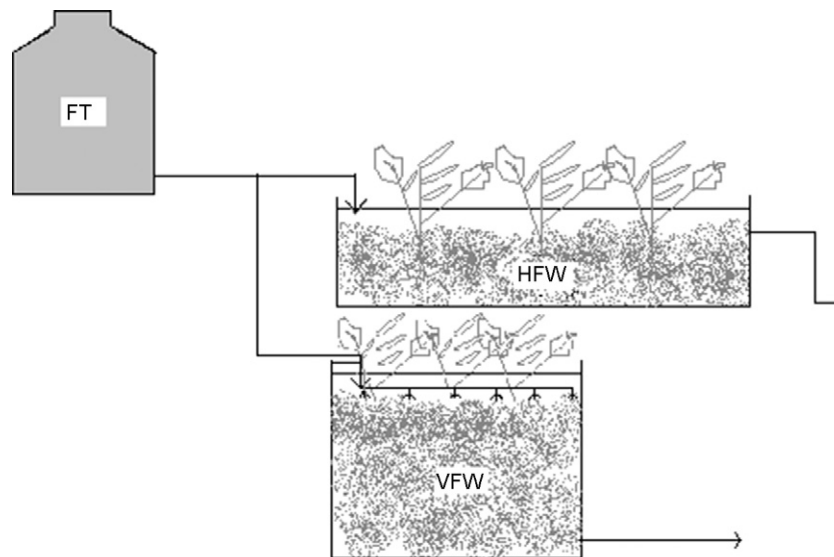


Fig. 1 – Wetland system for the treatment of domestic wastewater. Feeding tank (FT), horizontal flow wetland (HFW), vertical flow wetland (VFW).

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