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Research Paper

# A new colour vision system to quantify automatically foliar discolouration caused by insect pests feeding on leaf cells

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Leaf colour and leaf area are key parameters for many plant studies, including plant protection. The quantification of foliar bleached area is often used to evaluate the strength of plant attack by pests or diseases, to compare the efficiency of different treatments, the resistance of some populations, and to set up some control methods. The quantification of such area by human vision often lacks accuracy and reliability. Image analysis systems could bring a more accurate and objective measure, and could be automated to treat a great number of samples. Such an automated tool has been developed in order to measure quantitatively foliar bleaching due to the sycamore lace bug, *Corythucha ciliata* (SAY) on Plane tree. This tool was built up with a colour image capture bench and a fast automated segmentation process based on an original chlorophyll histogram unsupervised classification. Dedicated software was developed and integrated in an operational image processing platform capable of routine use by non-specialists in image analysis. The accuracy of the tool was determined by comparison to human expert segmentation. A very low error rate was observed in the absence of artefacts, but artefacts such as powdery mildew symptoms were not well distinguished and lead to weaker performance. Comparing its reliability and robustness to classical visual estimation and classification method, the tool performance was similar to the most experienced rater. The advantage of such a system is the possibility to treat automatically a large number of pictures and produce accurate, reliable, repeatable and non-subjective quantitative measurements.



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## 1. Introduction

Leaf area and leaf colour are key parameters in plant science, because they reflect some strong physiological processes and plant health in response to biotic or abiotic stresses (Murakami, Turner, van den Berg, & Schaberg, 2005; O'Neal, Landis, & Isaacs, 2002). In the field of plant protection, pest damage or disease symptoms could be characterised on the plant leaves by foliar area removed, change in leaf colour, or a combination as the proportion of bleached area (Bakr, 2005; Wijekoon, Goodwin, & Hsiang, 2008). The precise quantification of these parameters is an essential element of many fundamental and experimental studies of plant-pathogen and plant–pest interactions. Measuring pest damage enables us to study the biology and dispersion of pests and to assess experimentally treatment efficiency or resistance level of a plant variety (Strickland, 1961). At an operational level, damage quantification is involved in detection and monitoring of pest populations, and therefore in the planning and the launching of pesticides or biological treatments thanks to the determination of economic action thresholds (Pedigo, Hutchins, & Higley, 1986).

Visual analysis methodologies, including visual damage estimation and classification by scorers are often developed to measure foliar damage (Strickland, 1961). These methods are easy to implement and rapid to apply, but show some disadvantages like subjectivity and lack of accuracy (Moya, Barales, & Apablaza, 2005). Considering leaf discolouration damage, human vision cannot differentiate weak variations of colour, and can confuse damage with other possible alterations. Visual area estimation is quite difficult when damage occurs in multiple split areas, which often leads to overestimation (Kerguelen & Hoddle, 1999; Sherwood, Berg, Hoover, & Zeiders, 1983). Finally the number of classes and the thresholds between the classes are difficult to determine and strongly affect the results (Morris, 1960). Digital image analysis could provide an objective and accurate measurement of leaf colour and many methods, from supervised to automated procedures have been developed to avoid visual methodology bias (reviewed in Bock, Poole, Parker, & Gottwald, 2010; and Barbedo, 2013). However, these methods often require specific hardware and software, lots of plant manipulations and image adjustments, which make them of limited practicality and time-consuming. The conception of an automated tool for image analysis can enable a very large number of samples to be processed rapidly with an accurate measure of areas presenting slight differences of colour (Kerguelen & Hoddle, 1999; Schaffer, Peña, Coils, & Hunsberger, 1997). Several imaging technologies have been used for the identification of foliar damage due to fungi or animals. Images can be obtained by means of a scanner (Wijekoon et al., 2008), a video camera (Wang, Yamamoto, & Ibaraki, 2008) or even a digital camera (Philipp & Rath, 2002). In each case, the nature and position of light sources determines the ability of the system to highlight the areas of interest (Philipp & Rath, 2002; Valous, Mendoza, Sun, & Allen, 2009).

Cell content feeding pests, such as some spider mites, thrips and hemipterans, destroy and empty plant cell content which leads to the death of the cell and its bleaching. This symptom is associated with the loss of the cells' photosynthetic activity, which could alter plant resistance to physiological stresses, diseases or pests but also its productivity and aesthetic aspects at high pest density. Moreover, these pests can transmit phytopathogenic organisms such as viruses or phytoplasms. Visual estimation of bleached leaf proportion and classification, by comparing samples to reference images with defined amounts of damage, is often used to estimate pest density (Bakr, 2005; Kerguelen & Hoddle, 1999). However, visual estimation of such area, corresponding to summing up multiple weakly aggregated bleached pixels and determining their proportion of the total leaf area, is very uncertain by visual analysis (Sherwood et al., 1983), leading to inaccurate and subjective measurement (Nita, Ellis, & Madden, 2003). The development of an objective and accurate computerised tool should be of interest for testing and setting up new solutions in plant protection against these cell content feeding pests.

The sycamore lace bug *Corythucha ciliata* (Say) (Hemiptera: Tingidae) is a cell content feeding insect, almost specific to plane tree *Platanus* sp (Hamamelidales: Platanaceae). This pest leads to aesthetic damage (leaf bleaching) and public nuisance (honeydew release) on urban plane trees, without harming their survival (Chauvel, 1988). Several beneficial organisms have been tested *in vitro* and their efficiency in controlling sycamore lace bug populations and to reduce the nuisance should be checked *in situ* (Verfaille et al., 2011; in press). These experiments require an accurate measurement of leaf bleaching and the analysis of many samples coming from different experimental sites.

In order to characterise automatically the leaf bleaching due to the sycamore lace bug and to evaluate the efficacy of the organisms used, we have developed an innovative tool producing quantitative measures by colour image analysis and here we present the materials and software. The accuracy of the tool was determined by comparison to segmentation by human experts. Its reliability was compared to that of a classical method, consisting of visual estimation and classification by human raters. The robustness of the tool and the classical method toward intra- and inter-rater variability and toward the presence of artefacts is compared. Finally, we discuss the advantages and limits of such a tool compared to more classical approaches.

## 2. Materials and methods

### 2.1. *Biological material*

The plane tree leaf is palmatilobed, having five lobes with five main veins placed in a fan. The leaf is covered by a layer of trichomes, mainly on the lower surface.

Mobile stages of the sycamore lace bug stand on the lower surface of the leaf and suck intracellular liquids, leading to discolouration of the upper surface of the leaf. The exact mechanism of nutrition and punctured tissues is poorly understood (Chauvel, 1988; Fraval, 2006). Discolourations first appear at the intersection between main and secondary veins, then spread along these veins to finally affect the whole leaf. Subsequently, the leaf can have a chlorotic aspect and then become brown (Halbert & Meeker, 1998).