Automatic detection of tulip breaking virus (TBV) in tulip fields using machine vision

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Abstract

Tulip breaking virus (TBV) causes severe economic losses for the Netherlands. Infected plants must be removed from the field as soon as possible to prevent further spread by aphids. Until now screening is done by visual inspection in the field. As the availability of human experts is limited there is an urgent need for a rapid, automated and objective method of screening. Based on laboratory experiments in 2008, we developed a vision method for use in the open field. From 2009 – 2011 field trials were carried out and the techniques were tested and improved. The final score of our system in the last experiment (2011) approached the scores obtained by the human crop experts.

Key words: Plant virus, Image processing, Autonomous robot.

1. Introduction

Tulip and other bulbous ornamental crops are often infected by viruses. One of the most important viruses in tulips is the potyvirus TBV (tulip breaking virus). Symptoms of the virus can manifest themselves in different ways, including striping of the leaves and abnormal flowers (Dekker et al. 1993). The presence of viruses causes a reduction in the quantity and quality of the product, which leads to sales and export restrictions. In the Netherlands the tulip area covers about 11000 ha, which is about 50% of the total area of flower bulb cultivation worldwide. In 2011, about 1.5% of the tulips were infected with TBV. In the Netherlands the annual cost associated with tulip breaking virus in flower bulb culture is estimated at 9 M€ (van der Vlugt 2006). A current method to keep the spread of viruses under control is by application of crop protection agents to control the aphids, which spread the virus. In addition to this, crop experts remove plants with characteristic symptoms.

A major problem with visual assessment whether or not plants are infected is the difficulty of observing the subtle and complex symptoms, which require expert interpretation. The visibility of the symptoms is also largely influenced by the tulip cultivar and weather conditions, and can often only be seen during a limited period of the growing season. This requires a large number of trained personnel, which are difficult to find for such a short period of time.

In a previous study, four proximal optical sensing techniques for the detection of TBV in tulip plants were evaluated. These techniques were compared with visual assessment by crop experts and ELISA (enzyme immunoassay) analysis of the same plants (Polder et al. 2010). In this study it was concluded that the most discriminating TBV symptoms are the red or purple spot patterns on the leaves. These patterns can be quantified using a hyperspectral - or RGB color camera. Because of the promising results (Polder et al. 2010) field trials were carried out in the period 2009 – 2011.

2. Material and methods

2.1. Field trial

To test the automated vision system, 500 bulbs of cultivar Yokohama (10%TBV) were planted after standard disinfection at $4/m^2$ on the 25th of November 2010 (plot 1). Also 100 healthy bulbs and 300 100% infected bulbs of the same cultivar were planted (plot 2 and 3). After the plants became visible in early spring, standard crop protection treatments to curb weeds, and to prevent infection by aphids were applied. On the 5th and 6th of April the tulips were evaluated for the presence of TBV symptoms, and the diseased tulips were labeled by tagging a QR code to these plants.

2.2. ELISA tests

After measuring the plants with the automated vision system, the labeled plants, together with 50 non-labeled plants (presumed to be healthy) were tested with a biochemical immunoassay (ELISA) for the presence or absence of TBV. The assay uses TBV-specific antisera in a validated protocol (Derks et al. 1982). These ELISA measurements give a reliable indication of infection of the bulb and plant with the virus and were used as reference to compare the human visual assessment and the machine vision results.

2.3. Imaging platform

A universal mobile platform was designed for imaging the rows of tulips in the field. In previous field experiments several types of cameras and illumination sources were tested.



FIGURE 1: The imaging platform in the field. The inset shows the interior, with the two color cameras.

The best performing system was obtained with two 5-megapixel RGB color cameras (Prosilica GC2450) under a zenith angle of 45 degrees, at 90 degrees difference in azimuth angle, viewing two sides of the plants, and illumination by fluorescent daylight (6000k) lamps. A light resistant canvas cover was used to exclude ambient light (Fig. 1).

2.4. Image classification

The first step in Image classification is to separate the plant from the soil background. This is accomplished by thresholding the excessive-green image ((2G-R-B)>0), resulting in a mask image per plant. The main characteristic of TBV infected plans is a red/purple spot pattern on the leaves. This pattern is quantified using the following features:

- 1. Fraction of red pixels $\frac{\sum((R-G)>15)}{area}$
- 2. Mean normalized red value $\frac{\sum R}{\sum (R+G+B)}$
- 3. Mean normalized green value $\frac{\sum G}{\sum (R+G+B)}$
- 4. Ratio of contour pixels of spots $\frac{\sum(spot \ contour)}{area}$

Where *R*, *G* and *B* are the red green and blue pixel values within the mask of each plant. The *area* is the total size of the plant, which was calculated as the pixel-count (Σ) of the mask. The spot contour pixels are the red pixels touching green pixels ((*R*-*G*)<15), which is an indication of the patchiness of the spot pattern.

Linear discriminant classification was used to compare both the image analysis results and the visual assessment with the ELISA score (ground truth). Classification was done for each side view. When at least one side was classified as diseased, the whole plant was considered to be diseased. The rationale behind this is that it is sufficient if the symptoms are visible from at least one viewing angle,

3. Results

From the first plot of 500 plants, 24 diseased plants were found and all imaged. Additionally 34 plants of the remaining healthy plants were randomly selected for image analysis. The crop experts also scored these plants. The confusion matrixes for the crop expert's scores versus ELISA are tabulated in table 1.

Elisa\Crop Expert	Healthy	TBV	Total
Healthy	30	4	34
TBV	0	24	24
Total	30	28	58

TABLE 1: Confusion matrix for crop experts versus ELISA

The crop expert missed no diseased plants, but 12% of the healthy plants were wrongly scored as diseased. The crop expert scored the additional plots with 100% healthy and 100% diseased plants correctly.

For the machine vision, when using excessive green to separate the plant from the background 71% of the diseased plants were found, but 24% of the healthy plants scored as diseased. The confusion matrix is tabulated in table 2.

Elisa\Machine Vision	Healthy	TBV	Total
Healthy	26	8	34
ТВV	7	17	24
Total	33	25	58

TABLE 2: Confusion matrix for machine vision (excessive green) versus ELISA

This relatively high error is mainly caused by a wrong mask image due to the color of the red spots on the leaves. The color of these spots is difficult to distinguish from the color of the sandy soil. This is not a problem for plants with only green pixels on the outer contour of the mask, but often red spots are connected to the background resulting in a wrong mask. Figure 2a shows an example.



а

b

FIGURE 2: (a) Wrong mask due to red spots on plant touching background, (b) Manual fixed mask.

We simulated the effect of correct segmentation by manual adaption of the masks (Figure 2b). When using a manually outlined mask for the plants 83% of the diseased plants were found, while 9% of the healthy plants scored as diseased. Table 3 shows the confusion matrix.

TABLE 3: Confusion	matrix for mac	hine vision (mai	nual mask) vers	sus ELISA
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Elisa\Machine Vision	Healthy	TBV	Total
Healthy	31	3	34
TBV	4	20	24
Total	34	24	58

The score for infected plants is slightly better (one more plant correctly classified) than the score of the crop experts but the score for the healthy plants is worse, where 4 plants are wrongly classified as infected.

As a validation experiment, 144 plants from the two additional plots were imaged (53 healthy and 88 infected plants respectively).

The classifier obtained from the first plot is applied on these images. The mask is obtained from the excessive green image, without manual adaptations. Table 4 shows the confusion matrix.

Elisa\Machine Vision	Healthy	TBV	Total
Healthy	49	4	53
TBV	7	81	88
Total	56	85	141

TABLE 4: Confusion matrix for machine vision versus ELISA (plot 2 and 3)

4. Discussion

These results of automated virus detection in tulips growing in the open field are promising, and further research will be done to develop an autonomous robot for the detection and removal of diseased tulip plants in the open field. The application of this robot system will reduce the amount of insecticides and the considerable pressure on labor for selecting diseased plants by the crop expert.

The manual segmentation has to be replaced by an automatic segmentation. Earlier experiments showed that a good segmentation result could be obtained by using a near infrared (NIR) camera, since the soil strongly absorbs NIR and appears dark, whereas plants have high reflection, resulting in a clear segmentation. Instead of using manual outlined masks, in the next experiment we plan to use a multispectral (RGB + NIR) camera and use the NIR-channel to discriminate between plant and background.

Reference list

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