Optical Flow to Measure Minute Increments in Plant Growth

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Abstract

Computer motion analysis was tested as an ultra-sensitive imaging technique to detect minute displacement of corn seedling growth. The motion analysis software displayed motion in vector form, i.e., as both magnitude and direction of seedling elongation. The system was able to detect increments of growth in a non-intrusive, non-contact fashion as minuscule as 5 microns.

Keywords: Optical Flow, Plant Growth

1 Introduction

Measurement of minute increments in plant growth, using displacement transducers, have been used for a number of years [5, 7, 9]. Using transducers, resolution of measurement of growth can be as sensitive as $10^{-6}$ meters. However, the transducer technique for growth measurement requires mechanical coupling of the transducer to the plant and the effect of this mechanical engagement has not been documented. A non-contact optical technique of growth measurement had also been demonstrated using interferometry [2, 4]. However, this method of measuring plant growth requires an elaborate setup with mirrors, lasers and other instruments.

In the present study we used a variant of an optical method, i.e. processing of image sequences to determine optical flow, an approximation to the local 2D image motion, to measure seedling elongation. The instrumentation necessary for this technique is comprised of a black and white camera, frame grabber and computer with appropriate software. In our particular setup, a color camera (a Sony 3 CCD camera, model DXC-327 with a zoom lens, model VCL-712BX, having a focal length range of 7.5-90 mm) was used and the $480 \times 512$ pixel color images were converted to $150 \times 150$ grayvalue images by using a YIQ color to grayvalue transformation$^3$ and by partitioning out the same subareas of each of the images that contained the corn seedling before optical flow analysis. Corn seeds were sown singly in modified peat moss growing medium in black polyethylene tubes 10 cm long and of 1 cm inner diameter. A tube with a corn seedling at the primary leaf stage was

$^3$Each pixel of a grayvalue image was computed as 0.299, 0.587 and 0.114 respectively of the corresponding red, green and blue pixels of a color image.
inserted into a glass bottle containing water and coils from water baths set at various temperatures. The seedling shoot was exposed to the ambient room air temperature of 24°C while the roots in the tube were exposed to the water bath temperature. The seedlings were grown initially at a temperature of 24°C for about 1 hour. Following this period the water temperature was reduced to 12°C, held at that temperature for a period and then raised back to room temperature. Constant scene illumination was maintained during the growth sequence, with fluorescent lighting having an intensity of 0.5 micro einsteins per meter squared per second\textsuperscript{1}. The windows in the experimental room were also blocked so that the room was devoid of natural light. This setup results in the scene being illuminated by ambient light, thus ensuring constant spatio-temporal illumination for all visible parts of the scene. Surface reflectivity was minimized by using a deep nap black rayon velvet cloth behind the seedling as the image background and placing the camera with a macro lens in close proximity to the front of the seedling. An image was acquired every 2 minutes during this activity. This sampling rate was determined from trial and error.

2 The Computation of Optical Flow

We computed optical flow from spatiotemporal derivatives of filtered versions of the images in a sequence taken of a growing seedling. Such differential techniques are based on the assumption of local image translation

$$\ I(x, t) = I(x - v\ t, 0) \ ,$$

where $I(x, t)$ is the image intensity at $x$ in image $t$ and $v = (u, v)^T$ is an approximation of the 2D image motion, also known as the optical flow or image velocity. The validity of this equation depends not only on the local translation and constant illumination assumptions but also on the assumption that the scene is rigid, i.e. the motion of the plant due to growth is much greater than the motion due to shape changes. From a 1st order Taylor expansion of (2.1) or more generally from an assumption that intensity is conserved, $dI(x, t)/dt = 0$, the gradient constraint equation is easily derived:

$$\nabla I(x, t) \cdot v + I_t(x, t) = 0 \ ,$$

where $I_t(x, t)$ denotes the partial time derivative of $I(x, t)$, $\nabla I(x, t) = (I_x(x, t), I_y(x, t))^T$ is the spatial intensity gradient and $\nabla I \cdot v$ denotes the usual dot product. From this equation, we can see that constant scene illumination is necessary, otherwise non-zero intensity derivatives due to illumination changes may lead to the detection of “false” image motion where none exists. Equation (2.2) yields the normal component of image velocity relative to spatial contours of constant intensity. Figure 1a illustrates what is commonly called the aperture problem \textsuperscript{3}. If a contour, for example a straight

\textsuperscript{1}An einstein is $6.022 \times 10^{23}$ photons of light.
line, moving up and to the right with velocity \( \mathbf{v} \), is viewed locally through an aperture, only the component of velocity normal to the line's orientation, i.e. the velocity component to the right, \( \mathbf{v}_n \), can be recovered. The tangential component of velocity, \( \mathbf{v}_t \), is the component of velocity in the up direction and cannot be recovered from a single local measurement. The normal speed \( s \) and the normal direction \( \mathbf{n} \) are given by

\[
\begin{align*}
    s(x, t) &= \frac{-I_t(x, t)}{\|\nabla I(x, t)\|} \quad \text{and} \quad
    \mathbf{n}(x, t) &= \frac{\nabla I(x, t)}{\|\nabla I(x, t)\|}.
\end{align*}
\]

(2.3)

Hence, each measurement of \( I_x, I_y \) and \( I_t \) at an image point allows the recovery of a normal image velocity, \( \mathbf{v}_n = s\mathbf{n} \), at that point. Full velocity is the sum of the normal and tangential velocities, i.e. \( \mathbf{v} = \mathbf{v}_n + \mathbf{v}_t \).

There are two unknown components of full velocity \( \mathbf{v} \) in (2.2), constrained by only one linear equation. This equation describes a line in velocity space as shown in Figure 1b. Any velocity on this line satisfies (2.2). The velocity on the line with the smallest magnitude is the normal velocity \( \mathbf{v}_n \). Another velocity on the line (at unknown location) is the full velocity \( \mathbf{v} \). Hence, further constraints in addition to (2.2) are necessary to solve for both components of \( \mathbf{v} \).

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Figure 1: (a) The aperture problem: only the component of velocity normal to the line's orientation, \( \mathbf{v}_n \), can be recovered, the tangential component of velocity, \( \mathbf{v}_t \), cannot be recovered. (b) The Motion Constraint Equation (2.2) yields a line in \( \mathbf{v} = (u, v) \) space, the velocity with the smallest magnitude on that line is \( \mathbf{v}_n \). Another velocity on the line is the correct full velocity \( \mathbf{v} \).

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Our computation of optical flow is based on Lucas and Kanade's technique [6] with modifications proposed by [10]. A study of various performance issues of this and other techniques is presented in [1] and those results led us to believe that optical flow might be suitable for measuring plant growth.
We implemented a weighted least-squares fit of local first-order constraints (2.2) to a constant model for \( \mathbf{v} \) in each small spatial neighbourhood \( \Omega \) by minimizing

\[
\sum_{\mathbf{x} \in \Omega} W^2(\mathbf{x}) \left[ \nabla I(\mathbf{x}, t) \cdot \mathbf{v} + I_t(\mathbf{x}, t) \right]^2,
\]

where \( W(x) \) denotes a window function that gives more influence to constraints at the centre of the neighbourhood than those at the periphery. The solution to (2.4) is given by

\[
A^T W^2 A \mathbf{v} = A^T W^2 \mathbf{b},
\]

where, for \( n \) points \( \mathbf{x}_i \in \Omega \) at a single time \( t \),

\[
A = [\nabla I(x_1), ..., \nabla I(x_n)]^T, \\
W = \text{diag}[W(x_1), ..., W(x_n)], \\
\mathbf{b} = -(I_t(x_1), ..., I_t(x_n))^T.
\]

The solution to (2.5) is \( \mathbf{v} = [A^T W^2 A]^{-1} A^T W^2 \mathbf{b} \), which is solved in closed form when \( A^T W^2 A \) is nonsingular, since it is a \( 2 \times 2 \) matrix:

\[
A^T W^2 A = \begin{bmatrix}
\sum W^2(x) I_x^2(x) & \sum W^2(x) I_x(x) I_y(x) \\
\sum W^2(x) I_y(x) I_x(x) & \sum W^2(x) I_y^2(x)
\end{bmatrix},
\]

where all sums are taken over points in the neighbourhood \( \Omega \). Spatial neighbourhoods \( \Omega \) were \( 5 \times 5 \) pixels, and the window function \( W^2(x) \) was separable and isotropic; its effective 1-d weights are \((0.0625, 0.25, 0.375, 0.25, 0.0625)\) as in [10]. The solution to this system of equations is a least squares fit of a single full velocity to a local \( 5 \times 5 \) neighbourhood of normal velocity measurements, i.e., a constant local velocity model is assumed. With respect to Figure 1b, this can be viewed as a least squares computation of the intersection point of all the motion constraint lines arising from the normal velocities in the neighbourhood. In this way, normal velocity is a precursor to a full velocity calculation.

Equations (2.4) and (2.5) may also be viewed as weighted least-squares estimates of \( \mathbf{v} \) from estimates of normal velocities \( \mathbf{v}_n = \mathbf{n}, \) i.e., (2.4) is equivalent to minimizing

\[
\sum_{\mathbf{x} \in \Omega} W^2(\mathbf{x}) \left[ \mathbf{v} \cdot \mathbf{n}(\mathbf{x}) - s(\mathbf{x}) \right]^2
\]

where the coefficients \( w^2(\mathbf{x}) \) reflect our confidence in the normal velocity estimates; here, \( w(\mathbf{x}) = ||\nabla I(\mathbf{x}, t)||.\)

The Sony camera used in our experimentation has non-square pixels with a height of \( 9.8 \times 10^{-6} \) meters and a width of \( 8.4 \times 10^{-6} \) meters. To take this into account we scaled all the computed horizontal components of full velocity by \( \frac{84}{98} \). This scaling has only a minor effect on the final computed full velocities as most unscaled full velocities were almost vertical.
3 Image Prefiltering

Our implementation first smoothed each image in the sequence. This filtering helped to attenuate temporal aliasing and quantization effects in the input. We used balanced smoothing which involves using a 3D separable Gaussian filter with a standard deviation ($\sigma$) of 1.5 pixels-frame. Good digital approximations to Gaussians can be found using $n$ coefficients, where $n$ is the smallest odd integer greater than or equal to $6\sigma + 1$. For $\sigma = 1.5$, to produce one smoothed image at frame $i$ we used images $i - 5$ to $i + 5$.

4 Image Differentiation

For balanced smoothed images, we computed $I_x$, $I_y$ and $I_t$ using 4-point central differences with mask coefficients of $\frac{1}{12}(-1, 8, 0, -8, 1)$. Hence the temporal support for a single flow computation was 15 frames (we needed 5 smoothed adjacent images for differentiation and 11 images to produce one smoothed image), i.e. we needed frames $i - 7$ to $i + 7$ to compute flow for frame $i$.\(^5\)

5 Flow Thresholding

Following Simoncelli et al. [10], unreliable velocity estimates were identified using the eigenvalues of $A^T W^2 A$, $\lambda_1 \geq \lambda_2$, which depended on the range of magnitudes and orientations of spatial gradients in local image neighbourhoods.\(^6\) The greater the range of magnitudes and orientation the larger $\lambda_2$ is and the more confident we can be in the computed velocity. If $\lambda_2$ is zero the matrix is singular and no velocity can be recovered. In the instance where there is little variation in the local normal velocities, $\lambda_2$ will be quite small, meaning full velocity cannot be reliably recovered. We used $\lambda_2$ as a confidence measure on each of the computed velocities. That is, we use the value of $\lambda_2$ to ascertain the reliability of computed full velocities. In our implementation, if $\lambda_2$ (and hence $\lambda_1$) was greater than a threshold $\tau$, then $v$ was computed from (2.5), otherwise no full velocity was recoverable at that location. We have found in previous work [1] that $\tau$ is a good threshold; large $\tau$ values produce accurate but sparse flow fields while small $\tau$ values produce inaccurate but dense flow fields. For the results in this paper we used $\tau = 1.0$. This number is based on empirical observations [1] made on a large number of synthetic and real image sequences, including the image data analyzed for this paper. The choice of $\tau$ is independent of physical meaning it might have.

Further thresholding of the computed flows was necessary to remove surviving outliers. We first remove all velocities with a negative vertical component (there were only a few of these). Second, we impose a similar velocity constraint on the computed flow on the basis that all velocities computed should

\(^5\)We also considered \underline{unbalanced} smoothing and forward/forward differences to compute flow on both sides of a temperature discontinuity but we found no appreciable difference in those results and the ones presented here.

\(^6\)The units of the eigenvalues are $(\text{units of } \frac{1}{\text{units of length}})^2$. 
be the same if the plant is growing uniformly for some short time interval. We do this using a number
of thresholds. Viewing a velocity \( \mathbf{v} = (v_1, v_2)^T \) as a 3 component vector, \( \mathbf{v} = \frac{1}{\sqrt{u^2 + v^2 + 1}} (u, v, 1)^T \) we can measure the angle between any two velocities, \( \mathbf{v}_1 \) and \( \mathbf{v}_2 \), as
\[
\psi = \arccos(\mathbf{v}_1 \cdot \mathbf{v}_2).
\] (5.8)

This metric was used as an error measure in [1]. In this paper, all velocities not within 3.5° of the
average velocity in local 11 \times 11 neighbourhoods (using (5.8) to measure the angles) were thresholded.
Finally, we computed the average magnitude and angle plus standard deviations of the remaining
velocities and removed all those velocities not within 1.0 standard deviation of these averages. The
final result of all this thresholding was a flow field with nearly uniform velocities.

![Corn Seedling](image1.png)  ![Normal Flow](image2.png)  ![Full Flow](image3.png)

Figure 2: An image of the corn seedling sequence and its normal and full optical flow fields. The corn
seedling's root temperature was 24°C, the normal and full flow fields were sampled by 2 and scaled by
50.0. The flows were computed using balanced smoothing and differentiation. The full flow (c) was
computed from neighbourhoods of local normal flow (b) and then thresholded as described in the text.

## 6 Results

Our program computed the rate of growth in pixels per frame. We converted this to \(10^{-4}\) meters per
frame by imaging a metric ruler in the same 3D plane as the plant was in and computing the number
of pixels per centimeter, we found that vertical 1 pixel corresponds to \(7.853 \times 10^{-5}\) meters. Since the
images were acquired at 2 minute intervals this corresponded to speeds in the order of \(10^{-6}\) meters
per second.

Figure 2a shows an image of the corn seedling, Figure 2b its normal flow field and Figure 2c the
full flow field computed from the normal flow field after thresholding as described above. The average
rate of growth for this image (the average full velocity magnitude) was $1.982 \times 10^{-7} \pm 8.393 \times 10^{-9}$ meters/second with a density of 93 velocities. Note that full velocities can only be computed where there is sufficient variation in the normal velocities (as indicated by the magnitude of $\lambda_2$), i.e., at the tip of the seedling in this case as shown in Figure 2b.

![Graph](Figure 3: The growth rate plus standard deviation versus time (the solid disks) and the temperature versus the time for the corn seedling image sequence (the open circles). Flow was computed using Gaussian smoothing ($\sigma = 1.5$) and 4-point central differentiation.)

Figure 3 shows the average magnitudes with standard deviation bars of the optical flow for each time (image) in the corn seedling sequence (as solid disks) and the seedling’s root temperature versus time (as open circles) for a sequence of 100 images.

7 Conclusions

The data in Figure 3 indicate that there was a good correlation between increased/decreased growth rate and higher/lower temperature. The average magnitude of the flow plus its standard deviation served as an alternative metric for plant growth. We note that we may be able to measure even smaller growth increments than $10^{-6}$ meters, the limitation in this paper is due to the corn seedling’s rapid
growth. Slower growth may be measured by reducing the temporal sampling rate.

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