Assessment of leaf cover and crop soil cover in weed harrowing research using digital images

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SHORT TITLE: Assessment of crop soil cover

SUMMARY

Objective assessment of crop soil cover, defined as the percentage of leaf cover that has been buried in soil due to weed harrowing, is crucial to further progress in post-emergence weed harrowing research. Up to now, crop soil cover has been assessed by visual scores, which are biased and context dependent. The aim of this study was to investigate whether digital image analysis is a feasible method to estimate crop soil cover in the early growth stages of cereals. Two main questions were examined: (1) how to capture suitable digital images under field conditions with a standard high-resolution digital camera and (2) how to analyse the images with an automated digital image analysis procedure. The importance of light conditions, camera angle, size of recorded area, growth stage and direction of harrowing were investigated in order to establish a standard for image capture and an automated image analysis procedure based on the excess green colour index was developed. The study shows that the automated digital image analysis procedure provided reliable estimations of leaf cover, defined as the as the proportion of pixels in digital images determined to be green, which were used to estimate crop soil cover. A standard for image capture is suggested and it is recommended to use digital image analysis to estimated crop soil cover in future research. The prospects of using digital image analysis in future weed harrowing research are discussed. **Keywords:** Physical weed control, crop damage, crop tolerance, crop resistance, crop recovery, digital image analysis, the excess green colour index

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Introduction

In a recent paper on guidelines for physical weed control research, Vanhala *et al.* (2004) emphasise the need of unbiased methods to assess the immediate crop damages associated with harrowing.

The importance of crop damage associated with weed harrowing has often been demonstrated (Jensen *et al.*, 2004) and Rasmussen (1991; 1993a) showed that crop soil cover is a valuable input in predictive models that aim to determine the optimal intensity of harrowing.

The immediate crop response to harrowing is most often expressed in terms of crop soil cover, which is the percentage of the above ground crop parts that have been buried in soil (Rasmussen, 1991). This measure is assessed by visual scores, which are context dependent and biased. Even trained people assess crop soil cover rather individually. One assessor may estimate a specified treatment at 20% crop soil cover while another may estimate it at 40% (Rasmussen *et al.*, 1997).

Nevertheless, crop soil cover has been and still is used for the lack of any better (Jensen *et al.*, 2004). The biased nature of visual scores is not vital in experiments where the main objective is to compare different treatments with the same experiment. However, when results from different experiments are of interest, it is indeed very problematic to use visual ratings. For example, Jensen *et al.* (2004) quantified the ability of lupin (*Lupinus albus* L. and *L. luteus* L.) to resist and tolerate crop soil cover from post-emergence weed harrowing without being able to make reliable comparisons to previous studies in pea (*Pisum sativum* L.). They doubted that their assessments of crop soil cover in lupin were comparable to those in earlier studies in pea conducted by Rasmussen (1993b). In consequence, visual assessment of crop soil cover hampers communication and learning within the scientific community.

Crop soil cover is mainly used in Europe (Kurstjens & Kropff, 2001, Jensen *et al.*, 2004, Melander *et al.*, 2005) whereas most research papers from USA and Canada express crop damage as crop density reductions (Mohler & Frisch, 1997; Leblanc & Cloutier, 2000). Jensen *et al.* (2004) discussed advantages and disadvantages of both measures and concluded that crop soil cover is the only practicable real-time method in cereals and grain legumes, because it is impossible to distinguish and count single crop plants immediately after harrowing. Plants are more or less buried in soil, which make them inseparable.

Previously, two objective assessment methods of crop soil cover have been tried out: (1) wooden sticks placed in crop rows to measure the height of the ridges created by the mechanical implements (Melander, 1997; Cirujeda *et al.*, 2003; Melander *et al.*, 2003) and (2) photoelectric sensor techniques where light reflectance from the crop canopy is measured by sensors (Rasmussen, 1996; Rasmussen *et al.*, 1997; Engelke, 2001; Hansen, 2005).

Wooden sticks are not useful for post-emergence weed harrowing because harrowing has no ridging effect but the method has some potential in row-cultivation where soil is thrown into the rows. There has been an increase in work on remote sensing by photoelectric sensors within site-specific weed management (Gerhards & Christensen, 2003; Scotford & Miller, 2005) but results in the context of weed harrowing are either negative or inconclusive when trying to establish a standard method (Rasmussen, 1996; Rasmussen *et al.*, 1997; Engelke, 2001, Hansen, 2005).

Rasmussen (1996) and Rasmussen *et al.* (1997) showed positive correlations between crop soil cover assessed visually and by photoelectric sensors but the relation between assessments was

context dependent. Rasmussen *et al.* (1997) concluded that variability in ground colour rendered sensor assessments inaccurate. Engelke (2001) found that the precision of photoelectric sensors used in the early growth stages of cereals was too low to be useful in automated adjustments of weed harrowing. Hansen (2005) used photoelectric sensors to investigate whether different barley genotypes responded differently to weed harrowing but without indicating the reliability of his assessments.

In a recent review on canopy spectral remote sensing, Thorp and Tian (2004) concluded that the presence of variable soil backgrounds still complicates the spectral response and hinders the analysis of vegetative cover. Unfortunately, the review was mostly concerned with the canopy reflectance in the near infrared (NIR) and red wavebands. The study by Marchant *et al.* (2001) who utilized three wavebands, red, near-infra-red (NIR) and green, was not included in the review. They obtained satisfactory segmentation of vegetation from background with a combination of these three wavebands plus introduction of a novel classification method (alpha-method). Unfortunately, this method requires a dedicated sensor and it has not been tested in crops that have been disturbed by mechanical weed control.

In order to develop a standard for objective and reproducible assessment of crop soil cover, we chose digital image analysis instead of photoelectric sensors because digital cameras are widespread and because image processing is used widely in research on leaf cover assessment (Thorp and Tian, 2004).

Our objectives were (1) to investigate whether digital image analysis provides reliable estimations of crop soil cover in the early crop growth stages of cereals and (2) to suggest a standard for the image capture procedure.

Materials and methods

Terminology and experimental approach

In this study, leaf cover is defined as the proportion of pixels in digital images determined to be green, and crop soil cover, defined as the percentage of leaf cover that has been buried in soil due to weed harrowing, is calculated as the leaf cover differences between control plots and harrowed plots divided by the leaf cover in control plots within each block replication. Leaf cover and crop soil cover are both expressed in percentage by multiplying by 100.

This study focus on factors that could be assumed to influence the estimation of leaf cover and thereby crop soil cover from digital images such as camera tilt angle, light conditions, size of recorded areas, direction of harrowing and growth stage of the crop. It is based on an innovative approach, which started with two main questions, (1) how to acquire useful digital images with a standard high-resolution digital camera (the image capture challenge) and (2) how to develop an appropriate algorithm and automated analysis procedure within a standard software package (the image analysis challenge) to calculate the proportion of green pixels in digital images. There was no attempt to discriminate crop and weeds because it was considered unimportant in the perspective of early post-emergence weed harrowing. In most cases weeds are assumed to make up only a few percent of the total leaf cover.

The challenges associated with the image capture and the digital image analysis were mutually connected. The image-processing procedure was changed several times during the study to cope with the different characteristics of the images acquired and the work with the image analysis also influenced the image-capture scheme.

The innovative working process with the image analysis challenge is described and illustrated in the materials and methods section, whereas the outcome of the work with the image capture challenge is described the result section.

Field experiments

Digital images originated from two field experiments (experiment 1 and 2) with weed harrowing in organic winter wheat (cv Complet) mixed with approximately 10% winter rye (cv Caroass). The mixture was arranged in order to guarantee that the harvested crop, could be distinguished from other non-organic winter wheat.

Both experiments were conducted on a sandy loam at Bakkegården, which is an experimental farm owned by The Royal Veterinary and Agricultural University, Denmark. The farm is organic, which means that pesticides were not used.

Experiment 1 was originally planned to investigate the importance of timing of weed harrowing in order to achieve efficient weed control and positive crop yield response, and the results have been reported elsewhere (Rasmussen & Nørremark, 2006).

Weed harrowing was carried out at three growth stages (BBCH), 12, 22 and 23 and in a combination of all growth stages (12+22+23), hereafter called the combined growth stage. Harrowing was on 9 December 2003, 14 April 2004 and 30 April 2004. At each growth stage, the crop was harrowed in the same direction 1, 2, or 3 times on the same day to create a progressive series of intensities. The planned targets of the graded levels of harrowing in each of the three specific growth stages were 0 (control), 30, 60 and 90% crop soil cover in order to cover the whole range of intensities from normal to very aggressive. The practical adjustment of the aggressiveness of harrowing was adjusted on the basis of visual assessments of the whole plots. Driving speed and tine angle was adjusted so one pass gave approximately 30% crop soil cover. After the settings of driving speed and tine angles had been chosen, all plots were harrowed with the same adjustment.

Harrowing was done with a 3 m wide weed harrow manufactured by Einböck (Einböck GmbH & CoKG, A-4751 Dorf an der Pram, Austria). At growth stage 12, the angle of tines was adjusted to the highest negative value possible (Vanhala *et al.*, 2004) giving a very gentle treatment. Driving speed was 3 km h⁻¹. At growth stage 22 and 23, the angle of tines was adjusted to the highest positive value possible (Vanhala *et al.*, 2004) giving the most aggressive treatment. Driving speed was 8 km h⁻¹. Higher driving speed did not increase the intensity in terms of crop soil cover.

The planned targets were practicable in autumn 2003 but not in spring 2004 where the soil was too compacted to achieve high degrees of crop soil cover. Based on visual assessments, about 20% crop soil cover was achievable after 3 successive passes at growth stage 22 (14 April 2004) and at growth stage 23 (30 April 2004) only about 20% crop soil cover was achievable.

Growth stage was assigned to main plots, with the four intensities of harrowing applied as a subplot treatment. Growth stage was on main plots because this saved time when plots were harrowed. Each sub-plot was 14 m long and 3 m wide.

All digital images were taken in 2004, which means that there were no recordings from the earliest growth stages in autumn 2003. To investigate the utility and possible limitations of the digital image analysis procedure in very early growth stages, a second experiment (experiment 2) was carried out in autumn 2004 to question whether it is possible to discriminate treatments when leaf cover approaches 1-3% of the ground surface.

In experiment 2, three progressive series of harrowing with 4 graded levels of harrowing was carried out in growth stage (BBCH) 11 when the first developed leaf was about 4-5 cm long. One series was harrowed along the crop rows, one across the crop rows and one in both directions, which means that harrowing was done in plots that were drilled in two perpendicular directions (double seed rate). The double seed rate was used because it was doubted whether it would be possible to discriminate treatments at the normal seed rate due to very low levels of leaf cover. The experiment was designed as three randomised block experiments within each direction; along, across and both. The angle of the tine on the Einböck harrow was adjusted to give the gentlest treatment possible, and driving speed was adjusted to give the graded levels of treatment (0 km h⁻¹, 2 km h⁻¹, 3.5 km h⁻¹ and 5 km h⁻¹). The driving speed was adjusted instead of the number of passes as in experiment 1 because an increasing number of passes created too aggressive treatments.

Image capture

In all photo sessions, four images were taken in each plot, and a total of 2112 images were recorded and analysed. Of these, 512 images were used to interpret the weed control experiment (Experiment 1) as presented in Rasmussen & Nørremark (2006).

The images, 2288 pixels horizontally by 1712 pixels vertically with 24-bit depth, were taken using a red, green, blue (RGB) digital camera, Olympus C750UZ (Olympus Optical Co., Ltd.). An 11 mm focal length lens was used with a fixed F-stop of 3.2. The digital image analysis procedure makes no special demands on the camera in terms of filters, white balancing, shutter speed or aperture value. The only requirement is that the images are focused and correctly exposed. To avoid random variation in the camera angle, a tripod was used to fix the position of the camera.

To investigate the importance of light source, a series of images were captured in bright and diffuse sunlight on 14 April and 30 April in experiment 1 (Table 1). To create diffuse sunlight, the sun was screened with a bright cloth, which made shadows from the crop plants imperceptible. Images captured on 14 April were taken with camera tilt angle 30^{0} and on 30 April with camera angle 45^{0} according to Fig. 1.



Fig. 1 Illustration of camera angles relative to the ground plane and the direction of harrowing

To investigate the importance of the directed angle of the camera relative to the ground plane, three different angles were compared, 0^0 , 30^0 and 45^0 (Table 1). By increasing angles, the camera was turned in the same direction as the plots were harrowed for the majority of recordings (Fig. 1). However, also one camera angle against driving direction was tried out on 30 April at growth stage 23 in bright and diffuse sunlight (Table 1).

 Table 1 Image capture schemes from experiment 1.

	Harrowing dates				
Angle of camera	Growth stage 12	Growth stage 22 14 Apr. 2004		Growth stage 12, 22, and 23 9 Dec. + 14 Apr. + 30 Apr.	
according to Fig. 1	9 Dec. 2003				
	Image capture dates and conditions				
Along driving direction	Overcast	Sunlight	Direct	Sunlight	Direct
		screened	sunlight	screened	sunlight
0°	7 Apr. 04 [‡]	14 Apr.		30 Apr.	
		04^{\ddagger}		04^{\ddagger}	
30°	7 Apr. 04 [‡]	14 Apr.	14 Apr.		
		$04^{\dagger},^{\ddagger}$	04^\dagger		
45°	7 Apr. 04 [‡]	14 Apr.		30 Apr.	30 Apr.
		04^{\ddagger}		$04^{\dagger}, {}^{\ddagger}, {}^{\$},$	$04^{\dagger},^{\$}$
Against driving direction					
45°				30 Apr.	30 Apr.
				$04^{\$}$	$04^{\$}$

[†]Data is presented in Fig. 5

[‡]Data is presented in Fig. 6

[§]Data is presented in Fig. 7

As the camera height over ground was kept constant at 110 cm, images represented increasing areas (in the range of 0.32 m^2 to 1.00 m^2) and decreasing resolutions (mm²/pixel) by increasing camera angles. To investigate the importance of area and to break the correlation between area and camera angle, two series of images covering 0.23 m^2 and 0.32 m^2 , respectively, were acquired with camera angle 0^0 on 7 April in plots that were harrowed on 9 December 2003 in experiment 1. Camera height was reduced in order to reduce area.

To investigate whether the digital image analysis could discriminate treatments carried out at very early growth stages, a series of images were captured on 2 November 2004 in experiment 2 in a crop with only one 4-5 cm leaf (BBCH 11). The camera angle was 0° and images were captured in bright sunlight. All plots were photographed before and after treatment on the same day to investigate whether pre-treatment images would improve the accuracy of the estimated crop response curves.

The image analysis procedure

The objective of the image analysis was to obtain a binary image where green plant leaves were segmented from soil surface, shadows, stones, dead plant residues and other debris. In a standard colour camera the spectrum received has a dimension of three, corresponding to a response in the red (R) (560-700nm), green (G) (480-600nm), and blue (B) (380-480nm) bands giving the so-called RGB tristimulus. Usually 24 bits of information for each pixel is stored in the image from standard digital cameras. This is apportioned with 8 bits each for red, green and blue, giving a range of 256 possible intensities for each hue. An image can be defined as a two-dimensional function f(x, y), where *x* and *y* are spatial (plane) coordinates, and the amplitude of *f* at any pair of coordinates (*x*, *y*) is called the intensity of the image at that point.

The segmentation was based on the three-component (RGB) data vector that describes each point in the image. The first stage of the segmentation, transforms the original RGB image into greyscale (monochrome) image by applying the excess green index introduced by Woebbecke *et al.* (1995) and Meyer *et al.* (1998):

$$Q_{x,y} = 2 \times G_{x,y} - R_{x,y} - B_{x,y}$$
(Equation 1)

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where $R_{x,y}$, $G_{x,y}$, $B_{x,y}$ and $Q_{x,y}$ are the non-normalized red, green, and blue intensities (0-255) and excessive green index respectively for each pixel coordinate (*x*, *y*) in the image. Q was rescaled into the range of 0-255 by adding number of pixels for greyscale values below this range to 0 and above this range to 255.

In the resulting greyscale image, green plants appear bright in contrast to a dark, almost uniform background where the soil surface, including shadows, stones, straw, and other debris have disappeared.

The next step was to determine the greyscale threshold (Fig. 2), which sets the contrast breakpoint between pixels containing vegetation and pixels containing non-vegetation. The threshold is depending on illumination conditions. That is, the brightness magnitude varies if the greyscale image of vegetation and non-vegetation is light, dark, low-contrast or high-contrast (Fig. 3). Therefore, the threshold should be set automatically in order to adjust for differences in illumination.

The automatic threshold determination consists first of a least square polynomial curve fit to a histogram of the greyscale image (Equation 2 and Fig. 2). The histogram has greyscale levels in the range $[0, 2^8-1]$ and is a discrete function $h(r_k) = n_k$, where r_k is the *k*th greyscale value and n_k is the number of pixels in the image having greyscale value r_k .

$$h(r_k) = \sum_{i=0}^n a_i r_k^i$$
 (Equation 2)

where k = 5,..., m, and m = 95 (i.e. a section of the greyscale value range), i = 0,..., n and n = 6 and a_i is the coefficients of the polynomial. Then, the local minima and maxima for $h(r_k)$ were found by determining both real and complex roots of the derivative of the 6th degree polynomial ($h'(r_k) = 0$).

The complex roots came in conjugate pairs whereof the real number was considered as a root. The smallest roots $(r_{k,l})$, determined the threshold on the greyscale range. If the number of pixels $(h(r_k))$, estimated for the smallest root $(r_{k,l})$, was larger than a limited number of pixels (L), depending on image size, the procedure looked for the next root $(r_{k,2})$. The *L* parameter was introduced in order to compensate for 'false' threshold values experienced at low crop densities and at illuminations conditions that provide 'hard shadows' (Fig. 4, left). Hard shadows are soil regions in the shade of leaves where the colour is biased towards the colour of the vegetation as the light as first transmitted through the plant leaves (Andersen, 2002). The L-parameter was determined empirically based on a visual study of the segmentation results of 60 images analysed by running the program without the L parameter. This showed that if the number of pixels estimated for the smallest root was above 15000, the root should be omitted in order to eliminate noise from hard shadows. The L-parameter value corresponds to 0.383% of the total number of pixels (2288 x 1712 pixels) and should be adjusted to other image sizes. The *L*-parameter had no impact on the segmentation value when pictures were without hard shadows.

The range of k [5, 95] was within the brightness of interest and based on studying 60 greyscale images. The images covered different illuminations at growth stage (BBCH) 11, 12, 22, and 23. The polynomial degree i in equation 2 were determined by using histograms of the same 60 greyscale images. For all 60 histograms, iteration of the polynomial degree i until obtaining the optimum R² values while providing a viable threshold at the same time (visual assessment) was done. The result was as stated above that n = 6. However, for some histograms, the least square 6th degree polynomial curve fit was badly conditioned. A solution would be to increase the degree of the polynomial equation, but it was experienced that higher order polynomials can be highly oscillatory leading to unwanted false local minima and maxima. Instead, it is more relevant to obtain the effect of averaging out questionable data points, rather than distorting the curve to fit data exactly. Nevertheless, the methodology was always able to find a root that could set a viable greyscale threshold.

The obtained greyscale threshold was then used to transform the rescaled excess green image into a binary image. The transformation assigned 0 to all of the pixels below the threshold value and 1 to all of the pixels above the threshold (i.e. vegetation). Finally, a 3 by 3 median filter was applied to reduce the binary image noise due to segmentation errors. Thus, it was assumed that a group of four or less connected pixels was noise. From the filtered image, the proportion of white pixels corresponding to green pixels on the original colour image was counted and termed leaf cover.

By applying the procedure as outlined above, each image resulted in one value, leaf cover, which was the proportion of pixels that were determined to be green in the original image. The leaf cover was expressed in percentage and could easily be related to crop soil cover as the percentage of loss of leaf cover as a result of harrowing. The image processing approach did not discriminate crop and weeds. An experienced drawback of the segmentation process is that plant leaves or leaf parts will not be recognised as vegetation when illumination and plant conditions provides 100% surface reflection of incident light.

The analysis procedure was programmed in MATLAB and MATLAB Image Processing Toolbox (MathWorks, Inc, MA, USA). For 20 images the clock time at the beginning and ending of processing was stored so that the elapsed time to analyse each image could be calculated. MATLAB ran on a standard laptop PC with a Pentium M processor operating at 1.6 GHz under a Windows XP

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operating system. The mean elapsed time of the segmentation was 1.77 s with a standard deviation of 0.03s.

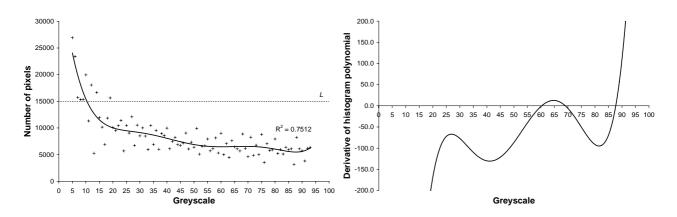


Fig. 2 Example of polynomial fit to histogram (left) and the derivative of the histogram polynomial (right). The found threshold values on the greyscale were 26.4 (the real part of a complex root), 60.3, 69.0, and 87.7. The data is from the segmentation presented in Fig. 3, left column. For the illustrated segmentation the threshold value 26.4 was used.



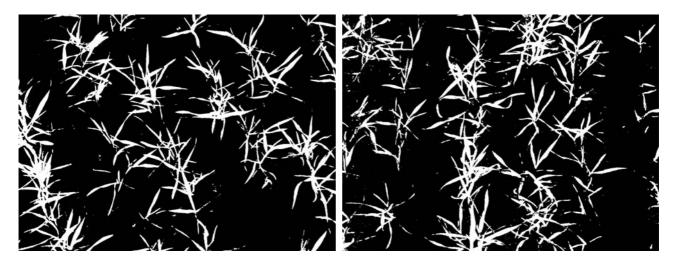


Fig. 3 Examples of images taken under shielded (left) and un-shielded conditions (right) on a sunny day and the corresponding binary images from the digital image analyses. Leaf cover was 15.9% for both images calculated on basis of the automated image processing segmentation. Threshold values of 25.4 (left) and 29.9 (right) was determined by the segmentation process. Images are from control plots that have not been harrowed.



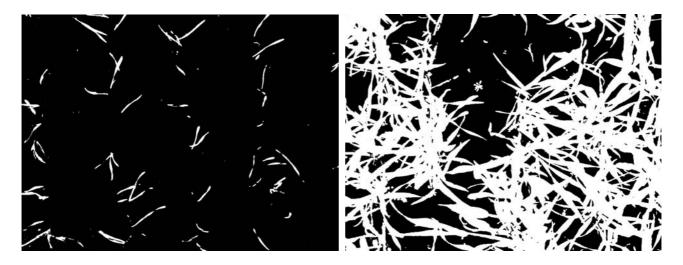


Fig. 4 Examples of images with low and high leaf cover in harrowed plots and the corresponding binary images. The left column image was taken under illumination conditions that resulted in 'hard shadows' and the right column image in "soft shadows". The low leaf cover was determined 2.57% and the high leaf cover was determined to 53.4%. Images are from plots that have been harrowed.

Statistics

To investigate whether digital image analysis provides reliable assessments of the immediate crop response, percentage leaf cover was used in the statistical analysis and not crop soil cover. Leaf cover is a more appropriate response parameter than crop soil cover, because it is an absolute measure in opposite to crop soil cover, which is expressed relative to untreated plots.

Regression analysis was used to describe how light conditions, camera angle, growth stage, size of photographed area and angle of camera and direction of harrowing influenced the digital assessment of leaf cover. Number of passes (intensity of harrowing) was the independent regression variable (covariate) and leaf cover was the response variable in different mixed models with light condition, camera angle, direction of harrowing, growth stage, size of photographed area and block as qualitative variables. Regression models were tested against analysis of variance models to test the

lack-of-fit. In order to omit non-significant factor or factor combination effects on parameters, successive approximative F-tests were made to reduce the complexity in models. Statistical analyses of leaf cover were performed with PROC MIXED in SAS (SAS version 8, SAS Institute, Cary, USA). In all cases, it was decided to analyse the logarithm of the leaf cover after inspecting the residuals.

Light conditions were analysed starting with a mixed linear model with harrowing, light conditions and time (and all interactions) as fixed effects, and plot, block and the interaction between time and block (whole plot) as random effects. Due to the fact that the variation within plots decreased with angle, the within plot standard deviation was allowed to depend on angle. Estimated regression lines based on the reduced model are presented in Fig. 5.

Camera angle and size of photographed area were analysed starting with a mixed linear model with harrowing, angle, and time (and all interactions) as fixed effects, and plot, block and the interaction between time and block (whole plot) as random effects. As variation within plots increased with angle, the within plot standard deviation was allowed to depend on angle. Estimated regression lines based on the reduced model are presented in Fig. 6. The importance of size of the photographed area was analysed by using the same mixed linear model, only replacing angle with size (within plot variation decreased with size).

Direction of camera was analysed starting with a mixed linear model with harrowing, light, and camera direction as fixed effects (with all interactions). Block, plot, and the interaction between plot and direction (sub-plot) were included as random effects. There was a larger within plot variation against the driving direction compared to along the driving direction, and the within plot standard

deviation was allowed to depend on driving direction. Estimated regression lines based on the reduced model are presented in Fig. 7.

The importance of harrowing direction in experiment 2 was analysed using the same model, only replacing camera direction with harrowing direction. Estimated regression lines based on the reduced model are presented in Fig. 8.

Results

Light conditions

Analysis of variance showed that there was no significant three-way interaction between growth stage, harrowing, and light conditions (P = 0.08) and subsequently no interaction between harrowing and light conditions (P = 0.61) when leaf cover was the response parameter. The remaining two interactions were significant, between harrowing and time (P = 0.04) and between time and light (P = 0.005). Regression analysis showed that number of harrowings could be included in the statistical analysis as a covariate and that leaf cover decreased exponentially by increasing number of passes with the harrow. No significant light effect was found on 14 April (P = 0.61) but on 30 April the general level of percentage leaf cover was assessed as 10% (95%-CI: 7% - 13%) higher in diffuse sunlight compared to direct light (Fig. 5). The assessed impact of harrowing was unaffected by light conditions in terms of percentage reduction of leaf cover but growth stage influenced the impact of harrowing. On 14 April each pass reduced leaf cover by 11% (95%-CI: 6% - 16%) and on 30 April by 19% (95%-CI: 15% - 22%) independently of light source.

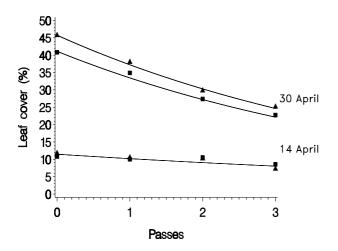


Fig 5 Impact of light source on the assessment of leaf cover on 14 April and 30 April. ■= direct and
▲ = diffuse sunlight. Observed treatment means and estimated regression lines. Further details in
Table 1.

Camera tilt angle

Analysis of variance showed that there was a clear interaction between camera tilt angle and growth stage (P < 0.0001), number of harrowings and growth stage (P = 0.022), but no three way interaction (P = 0.90) or interaction between harrowing and camera angle (P = 0.22) when leaf cover was the response parameter. Regression analysis showed that number of harrowings could be included in the analysis as a covariate showing that leaf cover decreased exponentially by increasing number of passes with the harrow.

Camera angles 0^0 and 30^0 gave inseparable assessments on 7 April and 14 April (P = 0.60) but the within plot standard deviation was clearly decreased by increasing camera angle (P < 0.0001).

The assessed impact of harrowing was unaffected by camera angle in terms of percentage reduction of leaf cover, whereas growth stage influenced the impact (Fig. 6). On 7 April, each pass reduced leaf cover by 8% (95%-CI: 3% - 12%), on 14 April by 14% (95%-CI: 10% - 19%), and on 30 April by 20% (95%-CI: 16% - 24%) independently of camera angle.

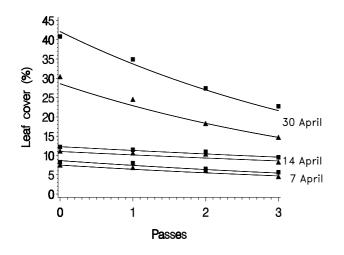


Fig. 6 Influence of camera tilt angle on the assessment of leaf cover on 7 April, 14 April, and 30 April. \blacktriangle = average of 0⁰ and 30⁰ (except on 30 April where data on 30⁰ did not exist) and \blacksquare = 45⁰. Further details in Table 1. Observed treatment means and estimated regression lines.

When the camera was angled away from 0^0 , it was important whether the direction was along or against the driving direction (+/- 45^0). There was no three-way interaction between camera direction, harrowing, and light (P = 0.65). Subsequently there was no interaction between harrowing and light (P = 0.67) or between camera direction and light (P = 0.54). As in the previous analyses, harrowing could be included as a covariate. There was no effect of direction on the intercept (P = 0.73) but light conditions influenced the intercept (P < 0.0001) (Fig. 7). An average leaf cover was assessed as being 9% (95%-CI: 5% - 13%) higher in plots with shaded light

compared to bright sunlight. The assessed impact of harrowing was unaffected by light conditions. The impact, however, was influenced by camera direction. When camera direction was along the driving direction ($+45^{\circ}$), each pass was estimated to reduce leaf cover by 19% (95%-CI: 15% - 23%) and when the direction was against the driving direction (-45°) by 25% (95%-CI: 21% - 28%).

Changing leaf angles by increasing number of passes caused the interaction between the camera direction and harrowing. After 3 passes the crop plants were clearly angled in the driving direction.

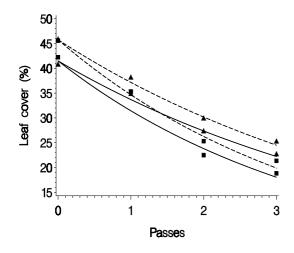


Fig. 7 Influence of camera direction relative to driving direction on the assessment of leaf cover on 30 April. \blacktriangle = camera direction along driving direction and \blacksquare = camera direction against driving direction. Broken lines indicate shaded sunlight and full lines bright sunlight. Observed treatment means and estimated regression lines. Further details in Table 1.

Size of recorded area

There was a clearly larger variation in the determined leaf cover when small areas (0.23 m²) were photographed compared to large areas (0.32 m²) (P = 0.0005). There was no systematic effect of size on leaf cover (all P values associated with size and interactions including size were larger than

0.24). The within plot variance in large area images was only 53% of the within plot variance in small area images.

Direction of harrowing in early growth stages

Covariance analysis including pre-treatment assessment of leaf cover as a covariate did not improve the statistical analysis of leaf cove after treatments due to non-significant effects of the pretreatment assessment. In consequence, pre-treatment assessment was excluded from further analysis.

There was no interaction between direction of harrowing and forward speed (P = 0.76) and the analysis showed that each km h⁻¹ increase in driving speed reduced leaf cover by 13% (95%-CI: 9% - 16%) independently of direction of harrowing (Fig. 8). There was no indication that the crop response was better assessed in plots that were drilled both ways and thereby had a higher leaf cover.

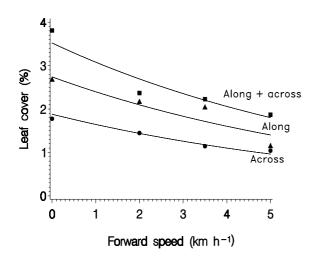


Fig. 8 Influence of harrowing direction in experiment 2 relative to forward driving speed on the assessment of leaf cover on 2 Nov. Observed treatment means and estimated regression lines.

Discussion

The main challenge in our study, in terms of the digital image analysis procedure, was to achieve segmentation robustness in outdoor field images under varying lighting conditions and to automate the digital image analysis. The image analysis procedure used was based on the discrimination of plant and background by thresholding the excess green colour index (Mayer *et al.*, 1998). Our contribution to the generation of a standard procedure was that we automated the determination of the grey level threshold, which sets the breakpoint between vegetation and non-vegetation. A fixed threshold as used in other studies (Meyer *et al.*, 1998; Tang *et al.*, 1999) did not apply in our study with different growth stages and light conditions.

We evaluated that the automated procedure provided reliable assessments. However, we did not apply a "true" reference, which could quantify the accuracy of the automated image analysis procedure. Some authors (Ngouajio *et al.*, 1998) compared operator-assisted classification of pixels (crop, weeds and soil) with automated digital image analysis, but this method was too labour intensive in our study. We controlled our image analysis procedure by careful comparison of the original colour images and the segmented binary images by randomly checking a number of images representing different lighting conditions and growth stages in the range of 1% to 40% leaf cover. Compared with the visual assessments of crop soil cover, which may by be influenced by a factor 2 of the individual assessor (Rasmussen *et al.*, 1997), our digital image analyse procedures represents a huge improvement in precision.

We did not compare our digital image algorithm with modified forms of the excess green colour index (Ribeiro *et al*, 2005) or our ordinary digital camera with more sophisticated 3CCD cameras (Onyango & Marchant, 2001). We were satisfied with the excess green colour index because our algorithm was fully automatic i.e. no manual settings of parameters were needed and it worked for image capture with auto white balancing. This was not the case for other methods used for vegetation segmentation in digital images (Onyango & Marchant, 2001; Grundy *et al.*, 2005; Ribeiro *et al.*, 2005).

Our image analysis procedure is fairly simple compared to image analysis systems that discriminate crop and weeds (Lemieux *et al.*, 2003; Onyango & Marchant, 2003, Grundy *et al.*, 2005) and our next step will be an attempt to convert the procedure into a simple software package that can be used by non-specialists into image analysis or without sophisticated software.

In weed science, leaf cover used in competition models is assessed as the vertical projection of plant canopy on the ground (Ngouajio *et al.*, 1998; Lemieux *et al.*, 2003), which is closely related to leaf area in early growth stages. We used different camera tilt angles because we focused on plant populations with different degrees of disturbance. It could be hypothesised that vertical projection would result in overestimation of leaf cover in harrowed plots because harrowing affects the deflection of the crop plants and may even flatten them in the driving direction. In contrast, it could be hypothesised that angled projections in the driving direction, would result in overestimation of leaf cover in undisturbed plots. Interactions between camera angle and number of passes with the harrow were expected if camera angle is important for the assessment of leaf cover. Our study, however, showed no significant interactions between camera angle and the effects of weed harrowing in terms of percentage reduction of leaf cover (Fig. 6), which indicates that vertical

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projection is suitable to assess leaf cover in crops that have been disturbed by harrowing. Camera angles against the driving direction, however, clearly influenced the assessed impact of harrowing (Fig. 7) and should be avoided.

Based on the findings, vertical projections should generally be preferred and definitely be used if driving direction is variable within a field or experiment. An angled camera may result in overestimation of the general level of leaf cover but the estimated impact of weed harrowing in terms of percentage reduction of leaf cover (crop soil cover) was not affected (Fig 6).

The captured area in the image recording procedure is inversely related to image resolution. The resolutions used in this study were in the range of 0.06-0.25 mm²/pixel, which is a higher resolution than 1 mm²/pixel used by Ngouajio *et al.* (1998). Ngouajio *et al.* (1998) reported how increasing recording area (0.33 to 1.63 m²) influenced leaf cover assessments at the expense of resolution (0.6 to 3.2 mm²/pixel). They found that average leaf cover estimates from increasing areas were less variable even if the resolution was lower. Their image analysis procedure, however, was not automated and the resolution limit is in our automated programme is unknown. It is assumed that the resolution used in this study could be further reduced.

Objective estimations of leaf and crop soil cover add new perspectives to research because postemergence weed harrowing is a trade-off between crop damage and weed removal effectiveness. This explains why mechanical weed control may cause yield decrease compared with untreated plots at low weed pressure and yield increase at high weed pressure as found by Rasmussen (2004) and Rasmussen & Nørremark (2006). Objective assessment of crop soil cover makes it practicable to distinguish two important aspects of crop tolerance to weed harrowing, resistance and recovery. Resistance reflects the ability of the crop to resist leaf cover reduction and recovery reflects the ability of the crop to regenerate from crop soil cover in weed-free environments. In this perspective, crop tolerance is the combined capacity of the crop to resist and recover from crop soil cover associated with harrowing. The importance of gaining knowledge about crop tolerance was illustrated in the weed control experiment, which constituted the basis of this methodology study. Crop resistance and crop recovery were highly affected by timing of weed harrowing and of major importance in order to optimize weed harrowing in terms of crop yield response (Rasmussen & Nørremark, 2006)

The lack of objective assessment of crop soil cover has resulted in oversimplified guidance and decision support. In Denmark, it is recommended that crop soil cover should not exceed 10-20% in spring cereals (Berthelsen, 2003). This guidance implies (1) that the specific range of crop soil cover is generally reasonable and (2) that farmers are capable of forming fairly accurate estimates of crop soil cover. Both preconditions are questionable. Weed species and densities, selectivity conditions and crop tolerance all influence the range of acceptable crop soil cover. Experiments have shown that increasing intensity of harrowing may result in yield gains in the entire range of 0-80% crop soil cover under given conditions, whereas other conditions make it impossible to achieve yield gains even at very low levels of crop soil cover (Rasmussen, 1991; 1993a).

Only few decision support models have considered post-emergence weed harrowing (Kristensen & Rasmussen, 2002) and no systems have been developed to help farmers to adjust the intensity and timing of harrowing. Kurstjens & Kropff (2001) developed a conceptual model of the harrowing process, which helps to get an understanding of the involved mechanisms but their model is too

complicated to serve as the framework in a practical decision support system. A practical way to integrate knowledge about crop tolerance in guidance and decision support systems still has to be evolved.

Conclusion

This study shows that leaf cover and crop soil cover can be estimated from images captured by an ordinary digital camera by using our automated digital image analysis procedure, which is based on the excess green colour index. Our results show that images should be captures in stable lightning conditions. The estimated values of crop soil cover were unaffected by lightning conditions under the condition that they were constant. Shifting lightning conditions while images are captured add experimental error to the estimated values and should be avoided. A camera angle of 0^0 (vertical projection) is preferable because an angled camera influences the general level of assessment. As for lightning conditions, the estimated values of crop soil cover were unaffected by camera angle under the condition that the camera is angled along the driving direction of the harrows. Image resolution of 0.25 mm²/pixel and four 0.32 m² images per experimental plot in experiments with four block replications provided high precision assessments. Larger recording areas at the expense of image resolution would most likely improve assessment precision but this was not tested. In conclusion, our study shows that digital image analysis provides a feasible method to assess crop soil cover in weed harrowing research, and we recommend that the digital image analysis procedure and the image capture standard proposed in this paper is used to quantify crop soil cover in future research into post-emergence weed harrowing.

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