

Applying colour-based feature extraction and transfer learning to develop a high throughput inference system for potato (*Solanum tuberosum* L.) stems with images from unmanned aerial vehicles after canopy consolidation

by Mhango, J.K., Grove, I.G., Hartley, W., Harris, E.W. and Monaghan, J.M.

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1 **Applying Colour-Based Feature Extraction and Transfer Learning to Develop a High**
2 **Throughput Inference System for Potato (*Solanum tuberosum* L.) Stems with Images**
3 **from Unmanned Aerial Vehicles after Canopy Consolidation.**

4 Joseph K. Mhango^a, Ivan G. Grove^b, William Hartley^a, Edwin W. Harris^a and James M. Monaghan^a

5 ^a*Crops and Environment Research Centre, Harper Adams University, TF10 8NB, Edgmond,*
6 *Shropshire, United Kingdom.*

7 ^b*Curious Raven Imagery, Market Drayton, Shropshire, United Kingdom.*

8

9 **Corresponding Author:**

10 **James M Monaghan**

11 **Postal Address:** *Fresh Produce Research Centre, Harper Adams University, TF10 8NB, Edgmond,*
12 *Shropshire, United Kingdom.*

13 **Email Address:** jmonaghan@harper-adams.ac.uk

14

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21 **Abstract**

22 *Potato (*Solanum tuberosum*) stem density variation in the field can be used to inform harvest timing*
23 *to improve tuber size distribution. Current methods for quantifying stem density are manual with*
24 *low throughput. This study examined the use of Unmanned Aerial Vehicle imagery as a high-*
25 *throughput alternative. A colour-based feature extraction technique and a deep convolutional*
26 *neural network (CNN) were compared for their effectiveness in enumerating apical meristems as a*
27 *proxy to subtending stems. Two novel colour indices, named the Cumulative Blue Differences Index*
28 *and Blue Difference Normalized Index, showed significant differences ($P < 0.001$) between*
29 *meristematic leaves and mature leaves in comparison to other indices. The two indices were used to*
30 *generate 500 pseudo-labelled human-corrected images as training data for the CNN. Benchmarked*
31 *against a human labelled test dataset, the CNN performed better with a normalized Root Mean*
32 *Square Error (nRMSE) of 0.09 than the sole use of the image analysis algorithm (nRMSE = 0.3) in*
33 *predicting the number of meristems in a canopy at 52 days after planting. Furthermore, the CNN*
34 *had better precision (Intersection over Union [IOU]: 0.49 and 0.56, respectively) than the image*
35 *analysis algorithm (IOU: 0.33 and 0.13, respectively). Meristem counts in both approaches showed*
36 *a linear relationship with actual subtending stem counts ($P < 0.001$). This study demonstrates the*
37 *validity of using traditional image analysis and CNNs to generate meristem detectors with*

38 *acceptable nRMSE. Transfer learning with CNN is proposed for developing meristem detectors for*
39 *evaluating stem density variation from UAV images in the field.*

40 **Key words:** Vegetation Indices, Deep Learning, Potato, Plant Population, Phenotyping, Machine
41 Vision

42 **1. Introduction**

43 At emergence, Potato (*Solanum tuberosum L.*) seed tubers produce variable sprout numbers
44 depending on the physiological status of the seed, which results in variable stem numbers per potato
45 plant (Knowles & Knowles, 2006). Estimation of spatial variation in plant density is important in
46 potato production, with several studies linking it to tuber size and total yield variations at harvest
47 (Bleasdale, 1965; Gray, 1972; Knowles & Knowles, 2006; Love & Thompson-Johns, 1999; Wurr, 1974).
48 Potato growers normally have a contracted range of optimum tuber size, outside of which the value of
49 the produce declines. Therefore, it is in the interest of growers to determine the factors that cause
50 tuber size variation in the field and tailor management practices to mitigate the effects.

51 At the stem level, a negative correlation between potato stem density and mean tuber size has been
52 widely recognized (Goesser et al., 2012; Mangani et al., 2015), and predictive models have been
53 produced to describe potato tuber size distribution using stem density as a covariate (Bussan et al.,
54 2007). To counteract the negative effect of stem density on average tuber size, several studies propose
55 delayed harvesting to prolonging tuber bulking period as a strategy to increase tuber size (O'Brien &
56 Allen, 1992; Rębarz et al., 2015; Waterer, 2007).

57 With this background, there is interest in techniques for determining stem density within an actively
58 growing crop to enable spatially and temporally variable downstream crop management like vine
59 desiccation, to maximize yield within desired tuber size classes. Manual stem counting in randomly or
60 systematically selected quadrants around the field give approximations of stem densities which can be
61 geospatially interpolated to the whole field, however this is a laborious, sometimes destructive, and a
62 low throughput method. The validity of data interpolation relies on assumption of a random
63 distribution of stem numbers or the unpredictable chance of establishing enough spatial
64 autocorrelation in stem numbers to model the variation with minimal error, which is not always
65 possible.

66 Using the spectral reflectance of potato plants to determine stem numbers from canopies is a potential
67 approach for estimating accurate stem density from RGB or multispectral sensors mounted on
68 Unmanned Aerial Vehicles (UAVs). This approach offers a high-throughput solution to estimate
69 variation in stem population across the entire field without interpolation. Potato stems terminate with
70 leaf primordia forming the growing tip of the stem, which sometimes convert to floral primordia
71 depending on the variety (Firman et al., 1995). The leaf primordia therefore represents a distinct unit
72 which can be used to estimate the total number of stems in a closed canopy. Plant canopies exhibit
73 unique, species-dependent, responses to incident radiation, generally showing high absorption in the
74 ultraviolet and Blue (490–450 nm) spectra, high reflectance in the Green (560–520 nm) spectrum,

75 high absorption in the Red (700–635 nm) spectrum and high reflectance in the near-infrared portion
76 (800-2500 nm) (Gates et al., 1965). Variability in chlorophyll content, water content and cell-to-air
77 space ratio in the leaves directly influences spectral reflectance of plants in the visible (400-700 nm)
78 spectrum (Cochrane, 2000), which can enable the use of computer vision and image analysis
79 techniques to decompose consolidated crop canopies and enumerate features of interest based on
80 their spectral reflectance.

81 Multispectral sensors with the near-Infrared band operating around 750 - 850 nm, enable the use of
82 well-defined vegetation indices like the Normalized Difference Vegetation Index (NDVI) to assess and
83 classify crop canopy components. Sankaran et al. (2015) used NDVI to extract and count emerging
84 potato plant clusters from images taken at 15 metres above ground using a UAV at 32 days after
85 planting with R^2 values of up to 0.82 when regressed to manual plant counts. However, predictive
86 power was lost as the canopy gradually consolidated at 43 days after planting. The most widely used
87 colour index for individual green plant segmentation from canopy remote sensing data is the Excess
88 Green Index (ExG), first proposed by Woebbecke et al. (1995). The index has been used, in
89 combination with other indices, for enumerating plant stands in wheat (Jin et al., 2017), rapeseed
90 (Zhao et al., 2017), and in potatoes (Li et al., 2019), where the index was used to detect potato plant
91 clusters at emergence. These techniques provide sufficient accuracy for counting clusters of stems
92 from the same mother tuber at emergence before canopy closure. However, individual stem
93 enumeration after full crop establishment, which is the level of accuracy required in precision farming
94 for variable desiccation management, has not yet been reported.

95 This study hypothesized that a colour index to extract clusters of leaf primordia and enumerate them
96 as a proxy to actual stem counts would potentially offer a solution. Following the spectral properties of
97 plants outlined in Gates et al. (1965), an ideal colour index would be one that is sensitive to the
98 differences between Blue and Green reflectance since the meristematic tips have less chlorophyll,
99 thereby exhibiting lower reflectance in the Blue range compared to older leaves. The performance of
100 object detectors based on image colour calculations depends on the acquisition of high quality imagery
101 with optimum light conditions which are not always possible in the field. A deep learning approach
102 therefore potentially presents a more robust approach with respect to variation in image quality.
103 Ground-truth labelled data is essential in deep learning training pipelines and forms the basis of
104 model evaluation in so called supervised learning models. Labelling a large dataset of leaf primordia
105 from closed canopies has a large time cost as it requires expertise in identifying irregular leaf
106 primordia. Semi-supervised learning therefore becomes a potentially important solution. Pseudo-
107 labelling is a widely used technique to train Convolutional Neural Networks from non-labelled data
108 with high accuracy. It involves the generation of an accurate model from a limited labelled dataset
109 then using the model on unlabelled data and selecting all predicted labels that have high confidence as
110 new labels, which helps to expand the labelled dataset.

111 The objective of this study was to use the spectral properties of plants in the visible wavelengths to
112 develop colour indices for enhancing primordial features in canopies and use them to infer variation
113 in actual stem number. The study also tested the use of colour indices for developing an automatic

114 labelling algorithm for generating a training dataset for transfer learning using Faster Regions with
 115 Convolutional Neural Network” (Faster R-CNN) to generate a robust potato meristem detector for
 116 inferring variations in stem number.

117 2. Materials and Methods

118 2.1 Feature Engineering: Development of Colour Indices

119 2.1.1 Data Acquisition

120 Development and evaluation of colour indices was conducted using potato canopy imagery collected
 121 from Harper Adams University, Shropshire, England (52°46'26.8"N, 2°25'48.9"W) on a dark brown
 122 stone-less sandy loam soil. The images were collected from Amora and Maris Piper varieties at 91 and
 123 50 days after planting respectively as shown in Table 1. Four different cameras, with varying sensor
 124 sizes and resolutions were used to generate variable sensor sharpness and colour resolving power.
 125 This enabled evaluation of the ability of the colour indices to distinguish meristem and old leaf pixels
 126 at different sensor sharpness and colour resolving powers so as to select the indices with the most
 127 consistent performance across sensors.

128 *Table 1: Specifications of Unmanned Aerial Vehicle cameras and crop stage used in the study.*

Location	Camera Description (Alias)	Variety	Days after Planting
Colour Index Development			
HAU, Shropshire, England	DJI™ Mavic Air UAV equipped with a 1/2.3-inch CMOS sensor producing 12 MP still images at 88° FOV (Mavic)	Amora	91
HAU, Shropshire, England	DJI™ Inspire equipped with a Zenmuse X3 Camera equipped with 1/2.3-inch CMOS sensor producing 12.4 MP still images at 90° FOV (Inspire)	Amora	91
HAU, Shropshire, England	3DR Solo UAV mounted with a Mapir™ Survey 3N Camera with a Sony Exmor™ R IMX117 sensor, f/3.0 Aperture and 41° FOV (Mapir)	Amora	91
HAU, Shropshire, England	3DR Solo UAV mounted with a GoPro™ Hero 3+ Black Edition camera equipped with a 1/2.3-inch sensor with 12 MP and a fisheye lens with a 94.4° FOV (GoPro)	Maris Piper	50
Model Training Data			
HAU, Shropshire, England	Phantom 4 pro UAV equipped with a Hasselblad L1D-20c aerial camera with a 1 inch CMOS sensor producing 20 MP still images at 70° FOV	Maris Piper	48
HAU, Shropshire, England	Phantom 4 pro UAV equipped with a Hasselblad L1D-20c aerial camera with a 1 inch CMOS sensor producing 20 MP still images at 70° FOV	Pentland Dell	48

HAU, Shropshire, England	Phantom 4 pro UAV equipped with a Hasselblad L1D-20c aerial camera with a 1 inch CMOS sensor producing 20 MP still images at 70° FOV	Amora	82
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Model Testing Data

Shawbury, Shropshire	Mavic Air UAV equipped with a 1/2.3-inch CMOS sensor producing 12 MP still images at 88° FOV at 20 m attitude.	45 different varieties (see Appendix A)	52
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129 MP=Mega pixels. CMOS = complementary metal-oxide-semiconductor. FOV = Field of View, DJI =
130 Dà-Jiāng Innovations™, Shenzhen, China. HAU = Harper Adams University.

131 The crops were grown using standard UK commercial practices for all inputs. For the Amora variety,
132 the aerial images were captured with 4 different visible light (Red, Green and Blue) sensors on UAV
133 systems as outlined in Table 1. The Maris Piper crop was added to increase variation in the dataset as
134 well as capture soil background because the canopy had not fully consolidated to cover the soil.
135 Additionally, the Maris Piper images were collected on a day where the field was partially irrigated,
136 providing the option of sampling pixels from both dry and wet soil. The captured images were
137 manually evaluated to exclude pictures with distortion or blur due to UAV speed and a total of 5
138 images were selected from each camera, resulting in 20 images from which colour indices were
139 evaluated.

140 All image processing was done using Matlab™ R2020a. From each image, pixels from Meristems,
141 Mature Leaves, wet Soil and Dry Soil features were manually selected, and reflectance values for Red,
142 Green and Blue were extracted. For each canopy feature, grayscale values were calculated from all
143 selected pixels and the first 25 pixels from either side of the mean were selected to create 50 pixels per
144 feature per camera. The final dataset for the evaluation of colour indices therefore contained 500
145 labelled data points of features with their Red, Green and Blue values.

146 **2.1.2 Visible Light Colour Index Selection**

147 Several colour indices were evaluated, with inclusion based on a literature search of widely used
148 visible light spectrum indices as shown in Table 2. The ExG is most widely used for segmenting
149 vegetation against soil background, however ~~we are unaware of any~~ evaluation of its potential for
150 differentiating leaf age, health or stress has not been reported. Other indices included the Excess Red
151 (ExR) Index for automatic segmentation of vegetation from soils, the difference between the Excess
152 Green and Excess Red (xGxR), the Colour Index of Vegetation Extraction (CIVE) and the Excess Blue
153 index (ExB), which is analogous to the ExR index. The Normalized Difference Red/Green Redness
154 index (NDGR), normally used for enhancing the contrast between red backgrounds and vegetation
155 was also considered.

156 Following the spectral reflectance properties of meristematic leaves described by Gates et al. (1965), it
157 was hypothesized that the difference between Blue and Red, and Blue and Green reflectance at the
158 pixel level would have the highest potential for maximizing contrast between meristems and older
159 leaves. It was expected that meristematic pixels would reflect more Red than Blue while mature leaves
160 would reverse this order due to darkening of the leaf as chlorophyll accumulated. Due to the darker

161 hue of soil, it was assumed that soil pixels would exhibit negligible differences in reflectance among
162 the three light bands. Two novel indices were therefore derived based on these premises. The Red to
163 Blue difference was plotted against the Green to Blue difference using all the selected pixels from
164 section 2.1.1 as data points. Visual observations showed that the Manhattan Distance or Euclidian
165 Norm of each point as a vector from the Cartesian origin would provide an index that maximizes the
166 difference between meristems and non-meristems. These two distances were therefore simplified into
167 linear expressions, with the Manhattan distance termed as the Cumulative Blue Difference Index
168 (CBDI) and the Euclidian Norm termed as the Blue Difference Norm Index (BDNI).

169

170 *Table 2: Descriptions and equations of popularly cited and custom colour indices based on standard*
 171 *Red, Green and Blue bands.*

Index Name	Formula	Source
Excess Green	$ExG = 2G - R - B$	(Woebbecke et al., 1995)
Excess Red	$ExR = 1.4R - G$	(Meyer & Neto, 2008)
Excess Green minus Excess Red	$ExG - ExR = 3G - 2.4R - B$	(Zhao et al., 2017)
Colour Index for Vegetation Extraction	$CIVE = 0.441R - 0.811G + 0.385B + 18.78745$	(Kataoka et al., 2003)
Excess Blue	$ExB = 1.4B - G$	(Guijarro et al., 2011)
Normalized Difference Blue Index	$NDGR = \frac{R-G}{R+G}$	(Bannari et al., 1995)
Cumulative Blue Difference Index	$CBDI = R + G - 2B$	Generated in this study
Blue Difference Norm Index	$BDNI = \sqrt{(R - B)^2 + (G - B)^2}$	Generated in this study

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173 All colour indices were calculated for all pixel features. The resulting dataset was comprised of pixel
 174 values as observations, the four features as factors and the 8 colour indices as continuous independent
 175 variates. Differences in means from sensors which only contained Meristem and Leaf features were
 176 analysed using the T-Test while data from the GoPro sensor, which contained dry and wet soil features
 177 apart from the canopy vegetation features, was analysed by ANOVA using R version 4.0.2 (R Core
 178 Team, 2020) adopting a Completely Randomized Design and means were compared using Fisher's
 179 unprotected Least Significant Difference. The colour indices with significant highest differences in
 180 index value between meristematic and other features were selected for further analysis.

181 Following selection of colour indices, an algorithm was designed to estimate the number of stems in
 182 an image using Matlab™ R2020a. Briefly, the model consisted of (1) K-means clustering for
 183 segmenting the image into foreground pixels of interest and background pixels and (2) establishing an
 184 objective process for consolidating fragmented pixels of leaflets into single meristem units. These two
 185 components were used to create and test a feature-extraction and object detection process as
 186 illustrated in Fig.1.

187 **2.1.3 Image Segmentation using k-Means Clustering**

188 Both Otsu-based and adaptive threshold methods have been extensively reported in literature for
189 green vegetation segmentation, but they perform poorly in images where the frequency of target and
190 non-target pixels does not result in a bi-modal distribution of intensities (Yang et al., 2012).

191 Meristematic tips constitute a small percentage of the image area in comparison to the rest of the
192 canopy and non-canopy features. Consequently, intensity histograms from indices that maximize the
193 reflectance of meristems against all other features like the CDBI and BDNI are expected to follow an
194 exponential decay, which renders Otsu-based threshold selection unreliable for effective
195 segmentation.

196 Since the images predominantly contained 3 object classes (i.e. Soil, and the non-meristematic canopy
197 component as background and the meristems as the foreground), k-means clustering with three
198 means was applied as the most appropriate method for clustering the three classes and segmenting
199 the foreground without computation of a threshold. Using k-means clustering for segmentation is
200 effective to separate object classes by minimizing the intra-class squared distance (Hartigan & Wong,
201 1979), and is widely applied in foreground segmentation in canopy images collected from UAVs and
202 remote sensing systems (Chen et al., 2019; Cinat et al., 2019; Sun et al., 2019). Accordingly, the pixel
203 with the maximum greyscale intensity in each image was identified, then k-means clustering was
204 performed on each image and the cluster containing the identified pixel was chosen to represent the
205 meristems. The image was then binarized with the selected cluster as foreground and all other clusters
206 as background.

207 **2.1.4 Noise Reduction and Final Bounding Box Generation**

208 Due to the compound nature of potato leaves, the k-means-based segmentation produced
209 unconsolidated meristem binary objects. Therefore, it was necessary to consolidate unconnected
210 meristematic pixels that belonged to the same stem to minimize the chances of double-counting, while
211 ensuring that meristems belonging to adjacent stems were not wrongly attributed. Morphological
212 operations like erosion or dilation have the risk of consolidating some independent but proximal
213 objects in a binary image (Pesaresi & Benediktsson, 2001). To avoid this, a custom noise reduction
214 technique was created by shrinking every binary object in the image to its centroid pixel, followed by a
215 pixel-wise iterative range search to index all other pixels located within a Euclidian distance that
216 corresponded to the average size of a stem in the image. The average size of a stem at the plot level
217 was estimated by calculating the average number of pixels per foreground object in each binary image.
218 As a result, all pixels located in close proximity to each other were indexed together and considered to
219 originate from the same primordia, then joined together. Pixels that were separated by a distance
220 more than the estimated average stem size were not connected and constituted separate instances of a
221 meristem. The number of connected components was then used as an approximation of the number of
222 stems in the image and a minimum bounding box was generated to approximate the location and size
223 of each stem, signalling the end of the algorithm. The flow chart of the algorithm was as illustrated in
224 Fig.1

225 **2.2 Development of a Transfer Learning Model**

226 An aerial survey with a UAV was conducted at Harper Adams University on 09th June 2020 to develop
227 a model training dataset of images images collected at 15 m altitude using a Phantom 4 pro UAV as
228 specified in Table 1. Varieties covered in the survey were Maris Piper (0.5 ha) and Pentland Dell (0.5
229 ha) at 48 days after planting and Amora (9.4 ha) at 82 days after planting. The images were then
230 cropped into 500 images of 500 pixels wide and 1500 pixels long then processed using the developed
231 algorithm to generate bounding boxes around proposed potato meristems. The generated bounding
232 for each of the 500 images were inspected and corrected manually by deleting erroneous detections
233 and adjusting the extent of each valid box to fit the extent to which a human would label the data. Wu
234 et al. (2020) emphasize on the computational constraint of training faster R-CNN object detectors
235 from large UAV images, in their case 5472 X 3648 pixels, which necessitated the cropping of their
236 images to 1000 X 800 pixels for optimized computation. The sensor used in this study produced 5472
237 X 3648 pixels, which were cropped to 1500 X 500 pixels to approximate the size of each plot in a
238 compiled test dataset. All generated bounding boxes were stored as pseudo-labels to create a training
239 dataset for deep learning with a CNN.

240 Fuentes et al. (2017) tested the Visual Geometry Group's (VGG-16) CNN (Simonyan & Zisserman,
241 2015) against deeper residual networks in the similar task of deep feature extraction of disease-caused
242 leaf colour changes in tomatoes and found that the VGG-16 performed better than the deeper
243 networks with up to 83% mean average precision. To keep the number of network backbone layers
244 minimal for producing the simplest model with faster training times, A Faster R-CNN model (Ren et
245 al., 2017) with the VGG-16 network backbone and imagenet weights was chosen. Faster R-CNN is a
246 unified framework that learns an object region proposal network from a CNN feature map, classifies
247 each proposed region and localizes the class of the object within the region with the introduction of
248 anchor boxes, from which object bounding boxes are learnt and refined by regression. To convert a
249 VGG-16 CNN into a Faster R-CNN object detector, a region proposal network was trained on the final
250 convolutional feature map and the last max pooling layer was replaced by an ROI-max-pooling layer
251 after which Faster R-CNN's classification and box regression layers were added to achieve object
252 detection and localization. The training was conducted on an Nvidia GeForce GTX 1070 GPU with 8
253 GB Video RAM for 11 hours. The model was trained with a fixed learning rate of 0.0001, a single
254 image mini batch size and 48 anchor boxes. The anchor boxes used in this study were predetermined
255 iteratively by estimating an increasing number of anchor boxes and their sizes with each iteration,
256 then checking their IoU with the ground truth data using the *estimateAnchorBoxes* function in
257 Matlab™ R2020a. The final number and size of anchor boxes was chosen by observing the asymptote
258 of the scatter plot of the determined IoU against the number of anchor boxes. Loss was optimized
259 using the stochastic gradient descent with 0.95 momentum. The model converged after 50000
260 iterations in 100 epochs. The flow chart of the training pipeline is as illustrated in Fig.2.

261 **2.3 Model Testing**

262 **2.3.1 Data Acquisition**

263 The traditional image analysis algorithm and the deep learning model were tested for performance
264 accuracy using a training dataset of images collected from 45 potato varieties (see Appendix A for a list
265 of the varieties) grown at Eaton Upon Tern Runway (Fig.3), Shawbury, Shropshire, England
266 (52°48'19.3"N, 2°30'41.8"W) on a Clayey Loam soil. The potato varieties came from the four
267 determinacy classifications (Group one to four) and some varieties were non-classified by
268 determinacy. Twenty Seven of the 45 varieties were drawn from the top 50 varieties grown in the UK
269 in 2019 in terms of area planted. The varieties acted as a source of variation in stem numbers per unit
270 area and canopy colour intensity. Differential performance between varieties was not considered in
271 order to generalize model accuracy across varieties. The 45 varieties were planted on 2019-04-29,
272 uniformly managed throughout the season and harvested on 2019-09-10. The ground-truth number of
273 above ground stems was manually determined on 2019-09-10 before harvest. The number of visible
274 meristems on top of the canopy was also manually counted. To create the model testing image dataset,
275 two adjacent rows of 5 metres each per variety were imaged on 2019-06-20 at 52 days after planting at
276 20 m altitude using a Mavic Air UAV as specified in Table 1.

277 **2.3.2 Image Processing and Data analysis**

278 The aerial images were cropped manually around each of the 45 varieties plots to create an image for
279 each plot for analysis then bounding box labels were manually defined for all meristems in each
280 cropped image using Matlab™ R2020a's "image labeller" application. For each image in the test set,
281 meristem counts were generated using the image analysis algorithm and the Faster R-CNN model
282 then compared to the manually counted number of meristems. Bounding boxes were generated from
283 the two predictive models. For each image representing a plot, the bounding boxes for the ground
284 truth, Faster R-CNN detections and image analysis detections were converted into binary masks then
285 confusion matrices were computed for the two detection models against the ground truth. The rates of
286 true positives (TP) and false positives (FP) were computed for each of the 45 plots from confusion
287 matrices. From these metrics, classification precision, as a measure of model performance, was
288 computed as follows:

$$289 \quad \textit{Precision} = \frac{\textit{True Positives}}{\textit{True Positives} + \textit{False Positives}}$$

290 Precision was chosen over Recall and F1-score metrics because the image analysis approach was based
291 on the detection of minor colour aberrations at the apex of the plant. This meant that bounding boxes
292 for the image analysis approach were expected to be smaller than the human-verified ground truth
293 where shape features that distinguish a meristem from older leaves were identified and the bounding
294 boxes expanded. This was projected to cause a high rate of false negatives which would penalize Recall
295 and subsequent F1-scores and therefore, the precision metric was used. The most important output of
296 the model for practical decision support is the detection of the presence of a meristem for calculating
297 stem density, while its size and extent are secondary considerations. Furthermore, the Intersection over
298 Union (IoU) was calculated as follows:

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$$Intersection\ Over\ Union = \frac{B_1 \cap B_2}{B_1 \cup B_2}$$

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Where B_1 represents the ground truth and B_2 represents the predicted bounding boxes from the two models.

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The final dataset contained the variety, breeder, number of manually counted above ground stems, number of manually counted meristems, and the number of meristems predicted by the image analysis and Faster R-CNN approaches. Observed vs Predicted plots were plotted for each prediction against the ground truth data to examine the residuals then the Root Mean Square Error (RMSE) was calculated for each model as follows:

307

$$RMSE = \sqrt{\sum_{i=1}^n \frac{(Predicted - Observed)^2}{n}}$$

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The RMSE was divided by the mean of the observed stem or meristem counts to calculate the normalized RMSE (nRMSE).

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3. Results

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3.1 Feature Engineering and Selection of Appropriate Indices

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When the difference between Green and Blue light was plotted against the difference between Red and Blue light, grouped by the pixel source, four distinct clusters were visually discernible in Cartesian space. The data points of the meristems clustered in the first quadrant, the mature leaf data points clustered in the second quadrant while the two soil sources clustered near the origin (Fig.4). Upon visual assessment (Fig.4), the meristem data points were clustered at the largest Euclidian distance from the origin, followed by mature leaves. From this assessment, the CBDI and BDNI were considered to have potential to represent the overall variation linearly and guaranteeing that the meristematic pixels would be at the maxima of this variation's range, therefore enabling a predictable threshold selection. The CBDI and BDNI were calculated and compared with established RGB-based colour indices.

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All the colour indices exhibited significant statistical differences between pixels from meristems and older leaves in mean intensity values ($P < 0.05$), a trend sustained across all sensor types except for the ExR in the Mapir camera (Table 2). This suggests that the features of interest occur in exclusive quantiles of the range of each index, and therefore a k-means clustering approach would adequately segment the image into meristems and background, eliminating the need for determination of a subjective threshold. To maximize the chances of accurate segmentation, it was necessary to select colour indices that maximize the value of the meristematic pixels while maintaining a large separation with all other features. To achieve this, the separation between the maximum and minimum quantile of mature leaves and meristems was evaluated and the percentage overlap was calculated, with the aim of selecting the colour index with the largest difference between the values of meristem and non-meristem features.

333 Of all the colour indices, the BDNI (Fig.5-a) and CBDI (Fig.5-b) had the most consistent maximization
334 of the index values for meristematic pixels, with distinct visual separation between the targeted and
335 background features in boxplots of the indices. Boxplots of ExG, (Fig.5-c) and CIVE (Fig.5-d) showed
336 an overlap of index values between the mature leaves and meristems. The minimum quantiles of
337 meristematic pixels were consistently higher than the maximum quantiles of the mature leaves in only
338 the CBDI and BDNI (Table 3). The opposite trend was consistently observed in the ExB, where the
339 meristematic pixels had negative values due to a Blue colour deficit while the soils and mature leaves
340 had excess Blue, resulting in a positive index, except in the Mavic and Mapir images. Unlike the EXG-
341 ExR (Fig.5-e), NDGR (Fig.5-f) ExR (Fig.5-g), indices, the ExB index (Fig.5-h) achieved a linear
342 positioning of features that would enable clustering. However, there were overlaps in the index values
343 of meristem and mature leaf features from images obtained with all cameras except the GoPro, with
344 the Inspire having a 28% overlap (Table 3).

345 As illustrated in table 3, the performance of the indices was consistent across camera type used to
346 collect the data. Index values of the features of interest overlapped in 6 out of 8 indices in images
347 taken with the Mapir Camera. The Mavic, Inspire and GoPro cameras had overlaps in five, four and
348 two indices respectively. The ExG index consistently showed overlapping in all the cameras while the
349 CBDI and BDNI indices only overlapped in the Mapir camera images. Based on this analysis, the
350 CBDI and BDNI were chosen for use in the k-means clustering. In all further images, the two indices
351 were calculated at each pixel and the resulting images normalized to 8-bit range then the two indices
352 were combined into a single channel by averaging. The resultant grayscale image was then used for k-
353 means clustering and subsequent stem count generation and bounding box approximation for both
354 the image analysis and Faster R-CNN approaches.

355 *Table 3: Mean values of colour indices calculated from the pixels of Leaves, Meristems, Wet soil and Dry soil using four different cameras*

356

Camera ^a	Feature	BDNI	CBDI	CIVE	ExB	ExG	xGxR	ExR	NDGR ^c
GoPro	Leaf	40.3 (2.7)	12.3 (11.7)	-15.6 (2.3)	19.8 (9.8)	87.8 (5.6)	97.5 (11.2)	-9.7 (7.6)	-20 (3.0)
GoPro	Meristem	82.1 (17.8)	115.6 (25.2)	-6.7 (5.6)	-32.1(13.0)	70.2 (14.6)	27.2 (15.8)	42.9 (8.9)	-3.0 (0.2)
GoPro	Wet Soil	4.9 (1.9)	-10.0 (3.2)	23.6 (1.0)	56.9 (5.9)	-8.3 (2.1)	41.1 (9.2)	33.6 (7.7)	1.0 (0.2)
GoPro	Dry Soil	7.4 (2.4)	5.2 (4.3)	21.8 (1.6)	39.1 (5.8)	-1.6 (3.8)	-64.3 (9.0)	62.7 (5.9)	1.0 (0.3)
GoPro	Gap ^b	0.3***	1.4***	-3.0***	0.1***	-3.0***	0.1***	2.5***	27***
Inspire	Leaf	33.5 (4.9)	-16.5 (4.9)	-3.1 (7.1)	19.3 (4.8)	54.0 (17.5)	75.8 (26.6)	-21.5 (10.3)	-3.0(1.0)
Inspire	Meristem	87.8 (14.7)	115.4 (22.6)	-28.7 (5.2)	-28.2 (16.4)	125.0 (13.0)	103.9 (19.8)	21.2 (11.1)	-1.0 (0.4)
Inspire	Gap ^b	1.0***	1.5***	-1.0***	-2.8***	-0.4***	-7.0***	0.1***	0.1***
Mapir	Leaf	81.2 (17.9)	114.3 (25.6)	-6.6 (5.6)	-31.7 (13.2)	69.9 (14.7)	27.5 (15.6)	42.4 (8.4) ^{NS}	-3.0 (0.02)
Mapir	Meristem	150.1 (16.1)	211.20 (22.5)	-31.9 (8.3)	-91.4 (15.9)	135.6 (21.04)	93.2 (27.7)	42.41 (9.6) ^{NS}	-0.6 (0.02)
Mapir	Gap ^b	-1.0***	-1.0***	1.9***	2.4***	-1.6***	-2.1***	-0.1	-1.0***
Mavic	Leaf	50 (5.5)	43.20 (10.9)	-23.6 (3.9)	-14.9 (14.6)	107.2 (9.5)	131.8 (17.9)	-24.6 (12.0)	-3.0 (0.7)
Mavic	Meristem	127.4 (16.6)	170.40 (24.3)	-47.5 (7.7)	-77.5 (18.8)	171.80 (17.9)	170.5 (28.3)	1.4 (13.2)	-2.0 (0.3)
Mavic	Gap ^b	1.5***	2.1***	-0.1***	-0.8***	-0.2***	-4.5***	-5.1***	28***

357 a = Camera alias, unless otherwise stated, there was significant difference in mean index values between meristem and non-meristem features in each camera
358 (P<0.05). Standard deviations from each mean are expressed in parentheses. b = the interval between the minimum value of meristems and the maximum value
359 of a mature leaf, expressed as a proportion of the range x 10⁻¹ (x 10⁻² for NDGR), negative values indicate that the ranges of the two features overlap. c = NDGR
360 x 10⁻². NS=No significant difference between meristem and non-meristem features. CBDI = Cumulative Blue Difference Index, ExG = Excess Green Index,
361 NDGR = Normalized Difference Green Redness, ExR = Excess Red, ExB = Excess Blue, xGxR = Excess Blue to Excess Red difference, CIVE = Colour Index of
362 Vegetation Extraction, BDNI = Blue Difference Normalized Index.

363 **3.2 Model Testing**

364 **3.2.1 Mean Stem Counts**

365 *Table 4: Performance of image analysis and Convolutional Neural Network approaches*
 366 *in the enumeration of meristems in all the varieties. The varieties are grouped into*
 367 *determinancy types for presentation purposes and “Actual Stem Number” information is*
 368 *included to illustrate the difference between meristem and stem counts.*

Variety Group	Actual Meristem Number	Actual Stem Number	Image Analysis Meristem Prediction	CNN Meristem Prediction
1	77.5 (2.1)	40.5 (3.5)	82.0 (2.8)	70.0 (2.8)
2	82.6 (12.2)	50.3 (13.2)	102.0 (32.1)	75.0 (8.5)
3	78.4 (7.9)	47.4 (6.7)	87.1 (22.6)	73.0 (8.0)
4	67.3 (3.1)	48.7 (5.1)	74.0 (16.1)	67.7 (1.5)
UG ^a	81.1 (9.1)	49.3 (11.5)	92.2 (28.2)	75.8 (10.0)
Grand Mean	79.8	48.7	90.6	74.5
RMSE^b			24.1	7.3
nRMSE^b	-	-	0.3	0.1
RMSE^c			46.9	26.8
nRMSE^c	0.7	-	0.9	0.6

369 RMSE = Root Mean Square Error. nRMSE = Normalized Root Mean Square Error. a =
 370 Unknown variety group. b = RMSE or nRMSE with meristem ground truth as the
 371 observed variable. c = RMSE or nRMSE with manual stem counts as the observed
 372 variable

373 Actual main stem counts from the field validation showed that the average number of
 374 above-ground stems per determinancy group had low variation ranging from 47 to 50
 375 stems while there was more variation in the actual number of meristems counted, ranging
 376 from 67.3 in group 4 varieties to 82.6 in group 2 varieties (Table 4). Faster R-CNN had a
 377 better predictive accuracy for the total number of meristems (nRMSE=0.09) than the
 378 image analysis method (nRMSE=0.3). Both Faster R-CNN and image analysis algorithms
 379 had low accuracy in predicting the actual number of stems in the plot (nRMSE was 0.6
 380 and 0.9 respectively) and the same observation was made when manually labelled
 381 meristem were compared to the manual stem counts (nRMSE = 0.7) as shown in table 4.

382 Least squares linear models of the predicted meristem counts against manual meristem
 383 counts showed an R² value of 0.57 (Fig.6) and 0.73 (Fig.7) for the image analysis method
 384 and Faster R-CNN learning method respectively. Additionally, there was a significant
 385 (P<0.001) relationship between manual counts of main stems and Faster R-CNN meristem
 386 detections (Fig.8) as well as counts from the image analysis approach (Fig.9).

387 3.2.2 Localization Accuracy

388 The image analysis method had a low mean IoU (0.3) and Precision (0.1) compared to the
 389 Faster R-CNN method (IoU = 0.5, Precision = 0.6) against the ground truth bounding
 390 boxes (Table 5). The Image Analysis algorithm had an average bounding box size that was
 391 closer to the average size of the ground truth boxes than observed in the faster R-CNN
 392 model (Table 5). The Inter-quartile Range (IQR) showed that there was more spread in the
 393 bounding box predictions of the image analysis method than the Faster R-CNN method,
 394 which predicted more equally sized bounding boxes (IQR=159.6).

395 *Table 5: Means and standard deviations (in parentheses) of detection and localization*
 396 *performance metrics of the image analysis and Convolutional Neural Network against*
 397 *manually labelled meristem data*

Variety Group	Ground Truth	Image Analysis			Faster R-CNN		
	BB ^a Size	IoU ^b	Pr ^c	BB ^a Size	IoU ^b	Pr	BB ^a Size
1	2991.1 (657.5)	0.4 (0.3)	0.2 (0.2)	1739.8 (584.2)	0.5 (0.5)	0.6 (0.2)	3161.1 (45.2)
2	2382.7 (745.9)	0.31 (0.5)	0.1 (0.7)	1253.8 (222.2)	0.4 (0.5)	0.4 (0.1)	3168.9 (130.6)
3	2985.4 (366.9)	0.32 (0.7)	0.1 (0.6)	2468.9 (304.2)	0.5 (0.7)	0.6 (0.2)	3159.0 (111.4)
4	2851.7 ()	0.3 (0.5)	0.1 (0.7)	938.8 (343.7)	0.4 (0.3)	0.6 (0.1)	3015.9 (172.2)
UG ^d	2996.3 (553.5)	0.34 (0.9)	0.4 (0.9)	2132.6 (204.9)	0.5 (0.5)	0.6 (0.1)	3125.9 (110.6)
Mean	2930.2	0.3	0.1	2009.8	0.5	0.6	3129.2
IQR^e	814.4			609.9			159.6

398 a = Bounding Box. b = Intersection over Union, standard deviation values are x 10⁻¹. c =
 399 Precision, standard deviation values are x 10⁻¹. d = Unknown Variety Group. ^eInterquartile
 400 Range

401 4. Discussion

402 **4.1 Feature Engineering and Development of Colour Indices**

403 Both the CBDI and BDNI indices achieved better classification of the meristematic leaves
404 than the other indices compared. The CBDI and BDNI indices were derived in such a way
405 as to take advantage of the theory that plant leaves exhibit variable reflectance of the Blue
406 wavelength based on the age of the leaves, and the maximization of index values in
407 meristematic features in line with the projected spectral signature of Gates et al. (1965).
408 The Excess Blue index equally agrees with the findings of Gates et al. (1965) as it shows
409 sensitivity to the diminished level of Blue light reflectance in meristematic structures,
410 leading to lower index values than older leaves and soil.

411 In agreement with findings from Woebbecke et al. (1995), The Excess Green index
412 adequately separated soils from canopy features. However, the index showed insensitivity
413 to the amount of reflected green light between the meristematic structures and leaves,
414 though the matured leaves had a higher mean reflectance than the meristems. The range of
415 the Excess Green index and all other indices (Fig.5) in meristematic leaves overlaps with
416 the range of the matured leaves, reflecting different levels of chlorophyll in meristematic
417 leaves as affected by the age of the leaf. This is expected as noted by Gates et al. (1965) that
418 a sharp drop in Red reflectance accompanies the continued increase in green reflectance
419 with leaf age as proto-chlorophyll is converted to chlorophyll.

420 Though potatoes generally contain a larger concentration of the lighter shaded chlorophyll-
421 a than chlorophyll-b (Anžlovar et al., 1996), a noticeable difference in Blue reflectance can
422 be expected in mature leaves compared to the meristems which still have proto-chlorophyll.
423 This is confirmed by Gates et al. (1965) who illustrates a slightly higher reflectance in the
424 Blue range from mature leaves than younger leaves in reflectance curves. The CBDI and
425 BDNI achieve better classification of meristematic leaves because they take this Blue light
426 reflectance into account in relation to green reflectance. The difference between these two
427 wavelengths is responsible for the high Manhattan distance and Euclidian norm from the
428 origin in the meristems (Fig.4). The results also show that the difference between Blue and
429 green reflectance is minimal in soils, showing more reflectance in the green range than the
430 Blue range in dry soils. This is in agreement with soil reflectance curves reported by Huete
431 (2004) which show a linear increase in reflectance from Blue to Near Infrared.
432 Baumgardner et al. (1986) reported similar curves which consistently show more Red than
433 Blue reflectance in soil. The findings for dry soils in this study concur with Baumgardner et
434 al. (1986), however, wet soils were found to reflect more Blue light than Red. Huete (2004)
435 and Baumgardner et al. (1986) discussed a decrease in reflected energy which makes soils
436 appear darker, consistent with the high reflectance of Blue wavelength observed. These
437 findings make the CBDI and BDNI ideal as they minimize the index values of soils and
438 mature leaves in comparison to meristems. Comparison of the boxplots of the two indices
439 additionally shows that the BDNI can be used as a general colour index as it additionally

440 separates vegetation from soils, while the overlap between mature leaves and soils in the
441 CDBI would make it unsuitable as a general colour index.

442 When targeting sparse features that do not show a peak in the feature space's histogram,
443 Otsu-method binarization of an image is known to produce non-satisfactory segmentation.
444 K-means segmentation adopted in this study provides an alternative that formulates
445 clusters of features based on the variation in the feature space (Yang et al., 2012) rather
446 than a subjective segmentation threshold. Where the feature space is defined by the
447 Manhattan distances using the CDBI or Euclidian distances in the BDNI, automatic
448 selection of a cluster of interest as a basis for binarizing the image is made possible since
449 the meristems cluster is bound to occur at the upper quantiles of the histogram.

450 **4.2 Model Testing**

451 Observed vs predicted plots of the number of meristems in the image analysis and Faster
452 R-CNN methods had R^2 values of 0.57 and 0.73 respectively (Fig.6 and 7). Faster R-CNN
453 has an advantage over image analysis with a low nRMSE of 0.09 compared to 0.3 nRMSE
454 observed in the image analysis. With no previous studies on potato stem detection, these
455 results can be benchmarked against models that detect variation in leaf colour and shape
456 due to viral leaf yellow mottling and crinkling akin to the underdeveloped leaves of
457 meristematic tips. In this respect, the Faster R-CNN performs comparably to findings by
458 Duarte-Carvajalino et al. (2018) where convolutional neural networks achieved a
459 maximum of 0.82 R^2 value for the detection of incidences of Late Blight (*Phytophthora*
460 *infestans*) on potato leaves when compared to manually labelled ground truth data.
461 Comparably, Sugiura et al. (2016) similarly developed an image analysis protocol for
462 estimating the severity of late blight with R^2 of 0.77. The results presented here show that
463 the Faster R-CNN approach is as efficient as other studies that aim to detect objects of
464 interest in potato canopies that are humans identify based on colour and leaf shape. The
465 difference between predicted counts and observed counts in the image analysis approach
466 show the need to account for more variation within the image by improving the image
467 segmentation and the algorithm's inclusion criteria of an independent stem. Improvements
468 in the image segmentation can be achieved by further feature engineering to generate more
469 robust colour indices. Furthermore, although K-means clustering and subsequent cluster
470 segmentation overcomes the problems of Otsu-based segmentation in non-bimodal data,
471 the hard-coding of cluster number introduces the possibility of misclassification of
472 ambiguous pixels, a double-edged sword that caused both false positives and false negatives
473 (Kanungo et al., 2002). More in-depth studies into possible adaptive threshold selection
474 techniques at the image level are needed to generate robust clustering and threshold
475 selection rules to improve accuracy. Differences between predictions and observations in
476 the Faster R-CNN model can partially be attributed to the limited variation in the training
477 dataset, generated from two potato varieties, against the testing dataset which contained
478 45 varieties with variable canopy characteristics.

479 The performance of region-based CNNs is influenced by the adequate determination of the
480 number of anchor boxes and their sizes at the training phase (Zhao et al., 2019). The
481 irregularity of potato meristems means there needs to be a representative compendium of
482 anchor boxes to cover the high variation in ground truth bounding box sizes. In this study,
483 the ground truth bounding boxes had a high IQR of 814.38 pixels compared to the predicted
484 bounding boxes of the CNN (159.61) and image analysis (609.86) on the test dataset (table
485 5). The CNN model produced regular (equally-sized) but larger bounding boxes than the
486 ground truth while the image analysis approach produced smaller bounding boxes than the
487 ground truth but were more variably sized, more naturally representing the variation in
488 sizes of meristematic tips. In subsequent studies with the CNN approach, a more exhaustive
489 method of anchor box size estimation is warranted, but equally so is the development of
490 the model from lower resolution imagery at higher UAV altitude to reduce the ground truth
491 IQR of the test dataset and potentially improve the model accuracy, though this comes at a
492 cost of more errors in labelling low resolution imagery. These observations signal potential
493 improvements to the data collection and hyper-parameter settings which may improve
494 model accuracy in future studies. The small bounding boxes in the image analysis approach
495 were reflective of the results of k-means clustering on the novel colour indices which were
496 highly optimized to maximize values of meristematic pixels against mature leaves.
497 However, the high R^2 values observed in both models show that there is a significant
498 correlation between the predicted and actual meristem counts, as well as actual main stem
499 counts (Fig.8 and 9), which shows that both models can be used in mapping this variation
500 at field scale, a key desire for farmers who seek to vary vine desiccation dates based on stem
501 density to manage potato tuber sizes and their distribution at harvest.

502 The faster R-CNN model achieved higher precision (0.56) and mean IoU (0.49) across the
503 variety groups compared to the image analysis method (0.13 and 0.33 precision and IoU
504 respectively), showing better efficiency at learning the features that a human labeller would
505 identify with meristems, as well as the effect of the human-verification and adjustment of
506 training labels in section 2.2 on the final model. In the absence of potato meristem
507 segmentation studies, precision scores were benchmarked against the Potato Virus Y
508 (Polder et al., 2019), whose primary symptom is chlorotic foliage akin to the signal being
509 detected by the image analysis approach to label stems. Polder et al. (2019) found precision
510 scores between 0.23 and 0.54 when a fully convolutional network was used to achieve
511 semantic segmentation of Potato Virus Y. This is comparable with the performance of the
512 faster R-CNN approach but outperforms the image analysis method. While the image
513 analysis approach also adequately identifies the presence or absence of a meristem, the size
514 and centroid of its resultant bounding boxes is less consistent since the system is purely
515 based identifying the extent of the colour aberration at the very tip of the youngest leaves
516 and not learning any other advanced features as in the Faster R-CNN. As a result, the image
517 analysis approach produces highly variable meristem sizes within an image as shown by
518 the high IQR. However, its inclusion in the Faster R-CNN pipeline is justified as it speeds

519 up the labelling of a large dataset, allowing a human-labeller to only correct the computer
520 generated labels. Potato meristems are not difficult to annotate for domain non-experts.
521 The image analysis method allows the generation of initial annotations to guide labellers
522 and train non-expert labellers to identify canopy features of interest from which they can
523 simply adjust bounding box extents and hence speed up the annotation process.

524 For the purposes of deriving a management or phenotyping tool for evaluating variations
525 in stem sizes across different stem densities, the establishment of a significant linear
526 relationship between predicted stem counts and actual counts is important despite the
527 presence of residuals because the linear relationship can be used to model spatial
528 variation in stem density at field scale. While Sankaran et al. (2015) reported a predictive
529 model with R^2 values of 0.83 for modelling plant density variation at emergence using the
530 NDVI, they observed that predictive accuracy was lost as the canopy consolidated and
531 they were not able to successfully run the prediction after 43 days from emergence.
532 Furthermore, the effective unit of plant density in the potatoes is the stem, which can only
533 be evaluated when all potential stems have developed, after plant canopy consolidation
534 (Wurr & Morris, 1979). The overall 0.73 R^2 value in this study's CNN method gives a level
535 of accuracy that is comparable to Sankaran et al. (2015) while offering the desired ability
536 to enumerate the preferred unit of plant density, which can be incorporated in vine
537 desiccation decision support systems for manipulating tuber size distribution at harvest
538 and in high throughput phenotyping. With 40 tubers planted per plot, the actual stem
539 counts found in this study mean that the average number of above ground stems per plant
540 (1.21) falls within the ranges (1-4.4) reported in literature (Wurr & Morris, 1979). Most
541 plants had one or two primary stems due to physiologically young seed tubers, stored
542 below induced dormancy-breaking temperature. While the meristems represent the
543 termination of both primary and secondary stems as well as sympodial branches, it can be
544 noted that the average number of meristems per plant (2.01) also falls within the range of
545 the number of main stems per potato plant reported in literature (Wurr & Morris, 1979)
546 and further suggests that most plants in this study produced one or two primary stem and
547 one secondary stem. The potato main stem always terminates with a meristem in all
548 varieties and a sympodial branch continues growth in indeterminate varieties
549 (Almekinders & Struik, 1996). The average number of secondary branches per stem
550 reported in literature is minimal ranging from 0.5 to 0.9 branches per main stem (Vos &
551 Biemond, 1992; Wurr & Morris, 1979). Therefore, while the number of meristems does
552 not directly correspond to the number of main stems, its density variation across the field
553 is a predictable proxy for stem density variation, which is the main desired unit of potato
554 plant density whose determination at field-scale had so far been elusive (Wurr & Morris,
555 1979).

556 The number of main stems formed by a potato is largely variable and contingent upon the
557 physiological age, plant population density and other agronomic and management factors

558 (Knowles and Knowles, 2006). The number of secondary stems formed is also dependant
559 on factors that affect apical dominance like inherent determinacy characteristics and frost
560 events (Chang et al., 2014). Additionally, differences in growth rates between stems means
561 some meristems are occluded from view at the top of the canopy by other leaves, hence
562 cannot be captured by UAV. These factors all contribute to the residuals between the
563 number of actual main stems and the number of meristems detected at the top of the
564 canopy. The results of this study suggest that the number of meristems visible at the top of
565 the canopy can be predicted using a CNN with low residuals (nRMSE = 0.09). Predicting
566 the actual number of stems from the meristems proved to be less accurate due to the
567 influence of secondary stems that also terminated in a meristem. However, this study
568 established that the predicted number of meristems at the top of the canopy explains a large
569 portion of the variation in the actual number of stems, providing a statistical route for
570 generating 2D density maps of the variation in stem density from UAV, using meristem
571 density as a proxy. Future studies must focus on generating methods for distinguishing a
572 meristem originating from a main stem from those originating from branches and
573 secondary stems. Unlike the physiologically young seed used in this study, temperature-
574 primed physiologically old seed is mostly used in commercial production, with low apical
575 dominance, forming multiple primary main stems at emergence and only branching late in
576 the season after flowering (Knowles and Knowles, 2006). To partially solve the problem of
577 secondary stems, it is therefore suggested that the meristem detection models should be
578 used before significant branching occurs. Future studies must also focus on determining
579 the optimum timing of imagery for minimizing the probability of detecting secondary
580 meristems.

581 **5. Conclusion**

582 This study represents the first attempt to enumerate potato stem number after canopy
583 consolidation using UAV based sensors. The prospect of accurately mapping variation in
584 stem density across a field enables the possibility of using precision agriculture techniques
585 to manipulate potato tuber size distribution through variable harvesting dates and other
586 in-season management practices. This study provides evidence that deep learning and
587 image analysis approaches can be used to accurately enumerate potato meristems and
588 estimate stem density variation in 45 UK potato varieties. Based on the spectral properties
589 of plants, the colour indices developed in this study should also have potential applicability
590 in mapping physiological maturity and leaf discolouration due to biotic or abiotic stress.
591 More studies to test the wider applicability of these indices are therefore recommended.
592 The study has also demonstrated the validity of automated labelling for generating a large
593 dataset of pseudo-labelled ground truth data which can be more rapidly quality-checked
594 and adjusted by a human labeller then used to train deep learning models that learn the
595 features of interest and achieve high IoU with manually labelled test data.

596 **6. Declarations**

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600 Number 11140054].

601 **6.2 Conflicts of interest/Competing interests**

602 The authors declare that they have no conflict of interest.

603 **6.3 Availability of data and material**

604 The data for this study shall be made available upon request.

605 **6.4 Code availability**

606 The custom code for this study was written in Matlab™ R2020a. The code shall be made
607 available upon request.

608 **CRedit Authorship Contribution Statement**

609 Conceptualization, Methodology, Software, Investigation, Writing – Original Draft,
610 Formal Analysis, Visualization, **Joseph K. Mhango**; Investigation, Funding Acquisition,
611 Writing – Review & Editing, Supervision, **Ivan G. Grove**; Conceptualization, Writing –
612 Review & Editing, Supervision, **William Hartley**; Writing – Review & Editing,
613 Supervision **Edwin Harris**; Writing – Review & Editing, Supervision, Funding
614 Acquisition, Project Administration, **James M. Monaghan**.

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762 **Figure Titles**

- 763 **Fig.1** Flow chart of the image analysis algorithm for generating meristem objects and
764 inferring stem number
- 765 **Fig.2** Flow chart of the Faster R-CNN algorithm for training a potato meristem object
766 detector
- 767 **Fig.3** Aerial image of the testing site for the image analysis and convolutional neural
768 network algorithms
- 769 **Fig.4** The difference between Green and Blue colour plotted against the difference

770 between Red and Blue colour in pixels selected from four prevalent features in a
771 potato canopy

772 **Fig.5** Index values of Meristems, Leaves, Dry soil and Wet soil using eight colour indices,
773 from images taken before canopy consolidation of partially irrigated Sandy Loam
774 soil. a - Blue Difference Normalized Index, b - Cumulative Blue Difference Index, c -
775 Excess Green Index, d - Colour Index of Vegetation Extraction, e - Excess Green
776 minus Excess Red Index, f - Normalized Difference Green Redness Index, g - Excess
777 Red, h - Excess Blue Index

778 **Fig.6** Observed vs Predicted of the number of meristems in potato canopies when
779 predictions were made using the traditional image analysis approach

780 **Fig.7** Observed vs Predicted number of meristems when predictions were made using a
781 Convolutional Neural Network-based object detector

782 **Fig.8** Observed number of stems vs Predicted number of meristems when predictions
783 were made using a Convolutional Neural Network

784 **Fig.9** Observed number of stems vs Predicted number of meristems when predictions
785 were made using the traditional image analysis approach

786 **Appendices**

787 **Appendix A** A list of the varieties used to test the object detection models

Purpose	Breeder	Variety
Chipping	Agrico	Agria
Crisping	HZPC	Alcander
Chipping	HZPC	Althea
Chipping	HZPC	Alverstone Russet
Crisping	Agrico	Arsenal
Chipping	HZPC	Asterix
Unknown	Unknown	Babylon
Crisping	PepsiCo	Brooke
Chipping	HZPC	Challenger
Crisping	Agrico	Corsica
Prepack	Agrico	Desiree

Prepack	Agrico	Estima
Crisping	HZPC	Heraclea
Chipping	HZPC	Innovator
Chipping	HZPC	Ivory Russet
Prepack	Greenvale	Jelly
Prepack	Unknown	King Edward
Crisping	Meijer	Lady Clair
Prepack	Branston	Lanorma
Prepack	Branston	Laura
Salad	Agrico	Maris Peer
Chipping	Agrico	Maris Piper
Crisping	Agrico	Markies
Prepack	Meijer	Melody
Prepack	HZPC	Mozart
Prepack	IPM	Nectar
Prepack	HZPC	Panther
Chipping	SCRI	Pentland Dell
Chipping	Agrico	Performer
Chipping	Norika	Pirol
Chipping	Higgind Group	Ramos
Chipping	IPM	Rooster
Chipping	McCains	Royal
Chipping	Unknown	Russett Burbank
Chipping	HZPC	Sagitta
Crisping	Stet	SHC1010
Crisping	PepsiCo	Shelford
Unknown	Unknown	Sorentina

Prepack	HZPC	Sunita
Crisping	HZPC	Taurus
Unknown	Unknown	Thalassa
Crisping	Unknown	Titan
Crisping	HZPC	Triple 7
Unknown	Unknown	VDW 07-197
Crisping	Stet	VR808

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