Instruction Guide



PlantScreen[™] MorphoAnalyzer

Please read the Guide before operating this product





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The contents of this manual have been verified to correspond to the specifications of the device. However, deviations cannot be ruled out. Therefore, a complete correspondence between the manual and the real device cannot be guaranteed. The information in this manual is regularly checked, and corrections may be made in subsequent versions.

The visualizations shown in this manual are only illustrative.

This manual is an integral part of the purchase and delivery of equipment and its accessories and both Parties must abide by it.

TABLE OF CONTENTS

1	Introduction	5
1.1	Purpose	5
1.2	Layout	5
2	Morpho Analyzer	6
2.1	Data Source Folder	6
2.2	Mask Builder	6
2.2.1	Use of Tray Mask	6
2.2.2	Drawing Tray Mask	6
2.3	Plant Mask Tool	7
2.3.1	Input	7
2.3.2	Color Picker Tool	8
2.3.3	Mask Detection Tools	9
2.3.4	Plant mask settings	13
2.3.5	Masking	15
2.4	Settings	16
2.4.1	Preprocessing Options	16
2.4.2	Plant Mask Settings	16
2.4.3	Tray Mask	16
2.4.4	Computed Parameters	17
2.4.5	Color Segmentation	
2.5	Single Run	
2.6	Batch Run	19
3	Bug Reporting	21
4	List of Figures	21



1 INTRODUCTION

1.1 PURPOSE

The Morpho Analyzer application is designed for users, who need to run morphological and color analysis of plant images stored locally on a computer disc drive. Typical usage is to run an additional analysis or test analysis on plant images exported to a local storage from PlantScreen[™] System database. This manual describes Morpho Analyzer version 1.0.14.3.

1.2 LAYOUT

The application window is based on a horizontal tabbed menu, which displays each tab across the whole window when clicked. The tabbed menu is arranged from left to right as the analysis steps gradually process. The initial windows *Mask Builder, Plant Mask Tool* and *Settings* are used for analysis settings. The final windows *Single Run* and *Batch Run* are used to analyze images based on settings defined in the *Settings* window.



2 MORPHO ANALYZER

2.1 DATA SOURCE FOLDER

The Morpho Analyzer application is designed to process locally stored data, which must be kept in a separate folder. When obtaining plant images with automatic measurement using PlantScreen systems, all data is stored in a database. To create a local folder containing images or mask files obtained with PlantScreen system use the export part of PlantScreen Data Analyzer application. The images are exported only in PNG format, which is currently the only image format supported with MorphoAnalyzer. The file format used for tray mask and plant mask is XML, which is also compatible with Morpho Analyzer.

2.2 MASK BUILDER

2.2.1 USE OF TRAY MASK

The *Mask Builder* window Fig. 1. allows to draw one or more areas subjected to detection of plant specific pixels. To identify the plants, each area is marked with a unique name. Typically, one area is used for detection of a single plant individual. One or more areas designated for the detection of plant masks are referred to as "tray mask".

Before using the tray mask, it's essential to load a template background image onto the viewing panel. (Fig. 1-10). Click the *Load Template* button (Fig. 1-1) and select an image to load from folder. Then, optionally a tray mask can be loaded on the image by clicking the *Load Mask* (Fig. 1-2) button. Once loaded or drawn, the mask can be saved to a local storage with *Save Mask* button (Fig. 1-3).

Load the current mask to *Settings* window with *Use Mask* button (Fig. 1-4) or to the *Plant Mask Tool* with *Use for Plant Mask Tool* button (Fig. 1-5).





2.2.2 DRAWING TRAY MASK

Drawing mode is activated by selecting an area shape button (Fig. 1-6) and marked with a red ribbon on top of the view panel. After choosing a suitable shape type by clicking rectangle , ellipse , pie or polygon button, the shape is drawn by dragging the mouse while holding left-click. To select multiple plants on the image, it is necessary to create multiple nonoverlapping areas, as is shown in Fig. 1.

Polygon shape is drawn differently. Selection of the polygon button activates a mode, where mouse left-clicking adds single points to a newly created shape. When the shape contains at least three points, the drawing can be finished with mouse right-click connecting the last point with the first. If less than three points are present, the right click discards the shape and the drawing mode is canceled.

The drawing mode is automatically deactivated after a shape is drawn. After deactivation, mouse left-click selects an area and when holding the *Shift* key, mouse left-click on another area selects multiple areas. If multiple areas are selected, each of the following operation is applied to all items. To move area(s), place the cursor on the inner part of selected area and hold mouse-left click while dragging the mouse. To resize the area, place the cursor on the border line and hold mouse-left click while moving the mouse. To achieve symmetrical shape of a square or a circle, press *Ctrl* key while resizing. Another tools can be activated with the following buttons (Fig. 1-

7): Discopy selection to clipboard, paste clipboard content on image, delete selection, to clipboard displaying area names, com in the image.

All drawn areas are listed in the *Areas* tab (Fig. 1-9). To select an area, left click the item in the list. Multiple areas can be selected by clicking the items individually. The listed areas are shown in the view panel (Fig. 1-10), either with red border line when selected, or green border line when not selected. The *Selected Item* panel (Fig. 1-8) can be used to edit area name or position by typing new values to textboxes. All changes must be confirmed with *Enter* key.



When multiple areas are selected, only parameters common for all selected areas are displayed in the Selected Item tab.

Area name must be unique within the scope of the mask. If the condition is not met, duplicate names are selected with warning dialog after clicking *Save Mask* or *Use Mask* button.

2.3 PLANT MASK TOOL

The tray mask contains definition of one or more areas, which are used for plant masking. This involves the identification of individual or multiple plants present on a tray. The identification of a plant-masked object within a designated area of the tray mask is achieved either through a pixel-by-pixel evaluation of color formulas or a binary classification of each pixel using random forest model. Additionally, other optional techniques, which re-select and regroup the selected pixels within an area, can be selected. Users have the flexibility to employ either the color formula method or the random forest method in the *Plant Mask Tool* tab.

2.3.1 INPUT

To start using *Plant Mask Tool*, load sample images and then a tray mask to the viewing panel (Fig. 2-4) by clicking the *Load Images* and *Load Tray Mask* (Fig. 2-1) buttons.

<u>Sample images</u>: Within the Plant Mask Tool tab, users can upload any number of images. However, it's important to be mindful that the processing time and computational costs increase with each additional image. By clicking the *Load Images* button, any presently open images are automatically closed and new images can be selected from a designated folder. To display a specific image in the viewing panel, simply click on its corresponding name within the image bar. To navigate among images that are currently not visible, employ the arrows for switching between their names (Fig. 2-3). The presently visualized image is highlighted with bold letters within the image bar for easy identification.

In the **Tab options section** (Fig. 2-2), there is the *Add image* button which allows adding more images to the existing selection. Additionally, the *Close current tab* button is available to swiftly shut the current image display. If you intend to close all loaded images at once, simply press the *Close all* button.

<u>Tray mask</u>: The tray mask can be loaded from the file using *Load Tray mask* button. The tray mask can be also loaded from the *Mask Builder* window - Mask builder tab – *Use for Plant Mask Tool* button (see the previous chapter). The same tray mask is used for all uploaded images.

To improve visibility, re-design the border line of the tray mask. In *Masking* panel (Fig. 2-9), the number represents actual thickness of the line and the colored rectangle on the right represents actual color of the line. To change the color, *Color Selection* window pops-up after right-clicking the colored rectangle.





Fig. 2 Plant Mask Tool Tab Before Color Segmentation

2.3.2 COLOR PICKER TOOL

When the image and the tray mask are loaded and displayed, begin by creating a color dataset comprising both wanted colors, representing plant pixels, and unwanted colors, representing background pixels. Activate color picker tool by flagging either the *Picking mode* or *Drawing mode* checkbox (Fig. 3-1). Then, select several color points (i.e. pixels) or draw one or more shapes by mouse left-clicking or moving mouse with left-click on the area to add colors to corresponding *Wanted Colors* or *Unwanted Colors* tab list (Fig. 2-5, Fig. 2-8).

In either mode of selection, it is necessary to carefully select only pixels corresponding to the plant in *Wanted colors* panel and only pixels corresponding to the background inside the area in *Unwanted colors* panel. When selecting colors, it can be useful to activate the zoom tool (Fig. 2-11) by unflagging the checkbox Fit. Move the slider from left to right across prescribed positions corresponding to 50, 100, 200 or 400 % zoom. Use scroll bars located in the view panel (Fig. 2-4) to navigate around the image.



Fig. 3 Color picker tool

Creating the datasets is a crucial step. A high number of selected colors increases the computation time for both segmentation methods. For Color Formula Computation, using K-means clustering is recommended to reduce the dataset size. However, this approach is not suitable for the Random Forest Method because K-means clustering does not preserve the pixel coordinates, which are essential for its functionality. For the Random Forest Method, it is advisable to use datasets containing up to 500,000 pixels to train a model effectively.

Right-clicking on the table of selected colors allows users to delete the currently selected pixel from the dataset (this can also be done by pressing the delete key), clear the list of colors, or apply K-means clustering to reduce the number of colors. To use K-means clustering, hover the cursor over the *Apply K-means* option and select the desired number of color clusters from the drop-down menu using the left mouse button. (Fig. 3-2).

Before you begin to formulate any segmentation strategy, it's important to consider the balance between your datasets of wanted and unwanted colors. It is advisable to ensure that these datasets are of similar size (balanced) to facilitate optimal segmentation performance. When attempting segmentation, the software evaluates the balance between the datasets of wanted and unwanted colors by calculating the ratio of their sizes. If this ratio falls within the range of 0.7 to 1.3, the datasets are considered balanced, and segmentation proceeds. If the ratio falls outside this range, a warning message is displayed to indicate that the datasets are not balanced and may affect segmentation quality.

2.3.3 MASK DETECTION TOOLS

The *Mask Detection Tools* (Fig. 2-6) include two methods for plant mask creation, accessible from a horizontal tab menu (Fig. 4-1). The first tab page, labeled *RGB Formula Computation*, employs a basic thresholding segmentation method. It is used to generate multiple color formulas, calculate optimum threshold and score precision for any formula of interest.

RGB formula computation

To quickly compute a set of color formula definitions, click on *Suggest Formula Faster*. For more accurate computation with higher precision score, use *Suggest Formula Better* button (Fig. 4-2). Each formula name in the list of suggested color formula definitions contains bracketed addition behind the formula, which marks whether the image was pre-processed for better performance with reflection reduction (*RR*). If only standard color formula computation was used (i.e. without RR), then the item is marked with (*std*) addition (Fig. 4-3).



Fig. 4 RGB Formula Computation tab page in Mask Detection Tools

After selecting a color formula by left-mouse click, add the formula to the formula box (Fig. 4-4) by right-clicking anywhere on the table to show a drop-down menu with the *Add Selected Formula* option. If the formula uses reflection reduction, the checkbox *Use reflection reduction* is flagged automatically. The *Show* option in the drop-down menu can be used to adjust maximum number of formulas shown in the list. To remove a single formula item, click the *minus* (-) button. It is also possible to type a new formula definition directly to the upper text-box and add it to the formula box with *plus* (+) button (Fig. 4-4)

The optimum threshold for each color formula is computed by *Auto threshold* tool. After adding the formula to the formula-box, click the button *Compute* to gain the threshold line for standard and for reduced reflection conditions. The *Show Graph* button (Fig. 4-5) opens graphs of individual computed color values divided by the threshold line (Fig. 5).





Fig. 5 Graph display performance of the selected threshold line

To load the selected color formula with threshold to the *Plant Mask settings* tab (Fig. 2-7), click the *Use selected formula button* located on the bottom of the *Mask Detection Tools* (Fig. 4-6) panel.

Machine learning computation

The second page tab in the *Mask Detection Tools* section, *Machine Learning Computation* (Fig. 6), adopts a more advanced approach by using a machine learning model to analyze more complex images. This tab features pixel binary classification through the use of a random forest model.



Fig. 6 Machine Learning Computation tab page in Mask Detection Tools

This method employs four preprocessing approaches, three of which can be adjusted by user. Upon clicking the *Preprocessing Options* button (Fig. 6-1), a new window appears (*Fig. 7*), where the size of these three features can be selected. The purpose of each feature is detailed in Tab. 1. All three incorporate matrices with sizes that can be modified in the *Preprocessing Options* window (*Fig. 7*-1, *Fig. 7*-2, *Fig. 7*-3). After choosing the preferred sizes, confirm your selections by clicking the *Ok* button (*Fig. 7*-4).

These approaches, when integrated as features in the random forest model, not only enhance its performance but also significantly improve the quality of segmentation by enabling more accurate differentiation between image regions. This leads to more precise and valuable segmentation outcomes.

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1	Gauss Fil Size: 7	lter: ≑	
2	Sobel Op Size: 7	erator	:
3	Entropy: Size: 7	÷	
4		ОК	

Fig. 7 Preprocessing Options for machine learning plant mask computation

Tab.	1 Brief	description	of Preproces	sing Option	s representing	g Random	Forest model	features
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Feature name	Description
Gauss Filter	The Gauss Filter, also known as the Gaussian blur, is a smoothing technique used in image processing to reduce noise. This results in a softening effect, making it useful for blurring images, enhancing image quality. Valid range: 4 – 25. Set to 0 and this feature won't be used.
Sobel Operator	Sobel Operator employs edge detection, a gradient-based method used in image processing to identify the edges of objects within an image by detecting changes in brightness. Valid range: 3 – 15. Set to 0 and this feature won't be used.
Entropy	In the context of image texture analysis, entropy measures the randomness or complexity within an image's texture, quantifying the amount of information or variability in the intensity patterns. This metric is used to differentiate between areas of an image with varying levels of detail or texture, with higher entropy values indicating more complex, information-rich textures. Valid range: $1 - 25$. Set to 0 and this feature won't be used.
Histogram Equalization	Histogram equalization is a technique to adjust the contrast of images, enhancing features by evenly distributing the intensity levels. This preprocessing step can improve the model's ability to recognize patterns and distinctions within the data. This feature is created from the image blurred with Gauss filter, therefore depends on its values range.

Examples of different size values of Preprocessing Options: Gauss Filter

Size = 25 (max. value)



Sobel Operator













After selecting the *Preprocessing Options*, the model's training is initiated by clicking the *Train Model* button (Fig. 6-2). The computation time for this operation depends on the number of loaded images, tray mask areas, and the size of the color dataset. The training uses 80% of the dataset, while the remaining 20% is reserved for model evaluation. The results of this evaluation are displayed in a table (Fig. 6-3), which includes the model name, its accuracy (the proportion of true results among the total number of cases examined), AUC (Area Under the ROC Curve, measuring the model's ability to distinguish between classes), F1 score (a harmonic mean of precision and recall, indicating the balance between the model's accuracy and its completeness), and time it took to train the model in milliseconds.

The name of the trained model appears in the drop-down list below, from which you can select the *Chosen Model* to be used for segmentation. This list also includes models added when you click the *Load Model* button (Fig. 6-4). Trained models can be saved as a ZIP file to any directory by clicking the *Save Model* button (Fig. 6-5).

To carry out the segmentation and generate the plant mask, click the *Compute Plant Mask With Chosen Model* button (Fig. 6-6). This action applies the model selected from the drop-down list, referred to as the *Chosen Model*, to all pixels within the tray mask areas, thereby creating the Plant Mask. The model name is then transferred to the Plant Mask settings section, where additional postprocessing and filtering are applied to the created Plant Mask.

2.3.4 PLANT MASK SETTINGS

Image analysis can be optionally enhanced with other techniques displayed in *Plant Mask Settings panel*. The settings can be saved or loaded with *Load* or *Save* buttons located on the bottom of the *Plant Mask Settings* tab (Fig.8-2).

Depending on the segmentation method chosen, there are two configurations for Plant Mask settings. For the basic method, known as *RGB Formula Computation*, the settings are visually depicted in Fig. 8, while for the more advanced *Machine Learning Computation* method, the settings are shown in Fig. 9. A brief description of all the parameters available for each method can be found in Table 2.



Tab. 2 Summary of Image Analysis parameters

	RGB Formula Computation
Parameter name	Description
Formula	Color formula definition. Use whole numbers (1, 2, 3, etc.), operators (+, -, *, /, min, max, ln, log, sqrt, ^) and pixel color intensity variables (R, G, B).
Threshold	Thresholding the computed color formula value is used to distinguish between plant containing pixels (above threshold) and the background (below threshold).
Median Filter Size	Median 2D filter is used to reduce noise by filtering with pixel window $n \ge n$. When plants are clearly visible, higher n values can be used to increase efficiency.
Min Size	Set value of minimal size of a single plant mask selected area [px].
Min Hole Size	Minimal size of holes in the mask objects in pixels, typically tens of pixels. The holes smaller than this value are closed and taken into the object pixels. [px]
Crop Object on Borders	Flag the checkbox to crop protrusions or unrelated objects (e.g. leaves of other plants) appearing on the borders of the tray mask.
CropObjectsOnBordersMin PreserveSize [px]	Minimum size of objects that will not be removed even if they are on the edge of the area specified by the tray mask (objects smaller than this value will be cropped on the borders if CropObjectsOnBorders option is set to true)
Use Reflection Reduction	Option normalizing RGB values in each pixel (ratio between channels is then considered for thresholding rather than absolute pixel values). Useful for harsh imaging conditions – e.g. light reflection from petri dishes.
Skip Bad Exposed Points	Flag the checkbox to crop poorly exposed pixels from the plant mask. Poorly exposed pixels have at least one color at minimum or maximum value.
	Machine Learning Computation
Parameter name	Description
Chosen Model	Name of a model selected for segmentation; trained or loaded.
Set Decision Threshold	The random forest model assigns each pixel a certain probability of belonging to one of two categories: plant or background. The default threshold for classification is set at 0.5, but this can be adjusted.
Dilation	Increases the size of the plant areas by applying a disk of a specific, adjustable size; potentially connecting nearby plants and filling in small gaps within the plant shapes.
Erosion	Reduces the size of the plant areas in a plant mask by using a disk of a specific, adjustable size; helping to remove small, isolated patches mistakenly identified as plants and sharpening the boundaries between plants and the background.
Opening	Conducted by first applying erosion and then dilation with a disk of a specific, adjustable size; separates closely situated plants in a plant mask by removing small connections between them and eliminating tiny specks or noise, clarifying the individual plant shapes.
Closing	Performed by first applying dilation followed by erosion using a disk of a specific, adjustable size; smooths the edges of plant shapes in a plant mask, closes small holes, and connects close parts, making the plant areas appear more cohesive.



Fig. 8 Plant mask settings for RGB Formula Segmentation

Fig. 9 Plant mask settings for Machine Learning Computation

2.3.5 MASKING

Clicking the *Recompute Plant Mask* button (Fig. 2-8, Fig. 8-3) in case of *RGB Formula Computation* generates Plant Mask. The color formula and optimum threshold is computed for each pixel of the image and other optional image analysis techniques listed in *Plant Mask Settings* are applied. On the other hand, in case of *Machine Learning Computation* the *Recompute Plant Mask* button (Fig. 9-3) serves to filter the existing Plant Mask according to the parameters set in *Plant Mask Settings*.

Then, the view panel (Fig. 11-2) is refreshed with the resulting masked image. The image can be viewed with plant mask border line placed on top of the image and/or in combination with tray mask border line and with or without background. The view options are defined by flagging checkboxes in the Masking tab (Fig. 10). The border lines can be restyled by thickness and color options. The thickness is determined by the number in the text-box and the color can be set by clicking the colored rectangle and selecting a color from Color pop-up window.



Fig. 10 Masking



If the analysis yields unsatisfactory results, consider adding new colors to the existing list in color picker tool (Fig. 11-1) and computing a new set of formulas, or train a new model. Alternatively, you can select a different configuration of filtering parameters in *Plant Mask Settings* (Fig. 8).





Fig. 11 Plant Mask Tool Tab After Color Segmentation

In the end, the settings, which yield most accurate masking result are sent to the *Settings* window by clicking the *Use Plant Mask Settings* (Fig. 11-3) button. It is recommended to also save the file separately to a local storage for later use or reference (Fig. 8-3, Fig. 9-3, button Save).

2.4 SETTINGS

The *Settings* window displays final analysis settings, which is then used in *Single Run* and *Batch Run* windows. The application stores the settings automatically when closed and restores them after restart.

2.4.1 PREPROCESSING OPTIONS

If the advanced *Machine Learning Computation* method is selected in the *Plant Mask Tools* tab, the *Preprocessing Options* settings (Fig. 12-1) become visible. Here, the settings are passed through and can be changed.

2.4.2 PLANT MASK SETTINGS

Plant Mask Settings tab (Fig. 12-2) contains correction settings for barrel lens distortion and plant mask detection. To correct for barrel (fish-eye) distortion, specify a correction degree with a number from -1 to 1. The checkbox x- fixed or y- fixed omits correction in x or y axis when flagged, which is useful for correcting images acquired in line-scan mode where distortion occurs only in one direction. When check-boxes for both axis are flagged, none correction is done. Centering of image correction can be done after flagging the *Change fish -eye center* checkbox and filling new values of x- or y- axis.

2.4.3 TRAY MASK

The button *Load Tray Mask* in *Tray Mask Panel* (Fig. 12-3) is used to select and load a tray mask for use in analysis. In gradually processed analysis steps starting in Mask Builder window, the tray mask should be already added to the *Settings* window, which is indicated by green colored *Load Tray Mask* button. If the button remains gray, the tray mask must be loaded by clicking the *Load Tray Mask* button and selecting the mask file in *Open* window. The tray mask can be also added with the *Use Mask* button (Fig. 1-4) in the *Mask Builder* window. Then, preview the mask on a selected image after clicking the *Show Tray Mask* button (Fig. 12-3).

R	J Morpho Analysis - 1.0.14.2 Mask Builder Plant Mask Tool Settin	2 ³⁵ Single Run Baich Run	4	5			
	Preprocessing Options	Plant Mask Settings	Computed Parameters	Color Segmentation			
	Gauss Filter Size: 7	Barrel (Fish-eye corr.): -0.005 👟	Side View Parameters	Color Definitions:			
	Sobel Operator Size: 7	X Fixed Y Fixed	Top View Parameters	R= 66 ,G= 60 ,B= 38	Add		
	Entropy Size: 7	Change fish-eye center	Area	R= 101 (G= 36 (B= 35	Delete		
		X: 0.0 🗘 Y: 0.0 🗘	Perimeter		Delete All		
		Median Filter Size: 6 🚖	Roundness				
		Chosen Model: Loaded:2002202	Roundness2				
1		Set Decision Threshold: 0.50 🛬	Isotropy				
		Min. Object Size: 700 🚖	Compactness				
		Min. Hole Size: 400					
		Crop Small Objects on Borders					
		Dilation Frosion	Width				
			Height]		
				Load Definitions)		
		Load Save		Store Definitions			
	Tray Mask	Colors Generating					
	Load Tray Mask	Auto-Detect Cluster Count		Finished		ſ	<u> </u>
3	Show Tray Mask						ю
		Generate Colors					

Fig. 12 Settings Tab

2.4.4 COMPUTED PARAMETERS

Morphology parameters are selected in the Computed Parameters panel (Fig. 12-4). Clicking the button Side View or Top View Parameters automatically flags a set of relevant parameters. All morphology parameters are listed and briefly described in *Tab. 3*.

Parameter	Description
AREA_PX	Total area covered with plant. Unit: pixel
PERIMETER_PX	Length of plant perimeter. Unit: pixel
ROUNDNESS	Ratio between area and perimeter of plant surface.
Formula:	4 * PI * Area / Perimeter ²
ROUNDNESS2	Ratio between area and perimeter of plant convex hull.
Formula:	4 * PI * Convex_Hull_Area / Convex_Hull_Perimeter ²
ISOTROPY	Isotropy of the object, computed as ratio of area and perimeter of the polygon created by object
	vertices.
Formula:	4 * PI * Polygon_Area / Polygon_Perimeter ²
COMPACTNESS	Ratio between area and surface of convex hull enveloping particular plant. More "compact" plants
	usually have shorter petioles and massive leaf blades.
Formula:	Area / Convex_Hull_Area
ECCENTRICITY	Parameter describing the degree of difference between convex hull area and circle, which has center
	in plant centroid.
Formula:	{Area (circle only) + Area (convex hull only)} / Area (intersection)
RMS	Rotational Mass Symmetry, based on the fitting of the ellipsis with identical second central moment
	as the analyzed object.
Formula:	2 * {sqrt ((1/2 * Major_Axis_Length) ² - (1/2 * Minor_Axis_Length) ²)} / Major_Axis_Length
SOL	Slenderness of Leaves.
Formula:	Skeleton Perimeter ² / Area
WIDTH_PX	Width of bounding box enveloping plant. Unit: pixel

Tab. 3 Morphology Parameter Description



HEIGHT_PX

Heigth of bounding box enveloping plant. Unit: pixel

2.4.5 COLOR SEGMENTATION

After masking the image, color segmentation can be used to classify the pixel-by-pixel color representation of the masked object. In color segmentation, the number of colors is reduced by k-means clustering technique, which classifies colors based on similarity. Each class shares properties similar to a color defined as representative.

The *Color Definitions* list in the *Color Segmentation* tab (Fig. 12-5) is used to define representative colors. The *Add* and *Delete* buttons allows to add or remove one selected color definition to/from the list. You can edit the RGB values for any representative color in the list by selecting and double-clicking the item. From the *Color* pop-up window select a definition from the *Basic colors* part or define a custom color in the *Custom colors* part.

The *Color generating* tab (Fig. 12-6) is a tool for automatic generation of color classes of plant(s) masked according to the settings definition in the *Setting* window (Fig. 12-1 and Fig. 12-2).

To set the number of clusters for the K-means algorithm, you can either specify the number manually or select the *Auto-Detect Cluster Count* checkbox. The software will then employ the Silhouette method to assess clustering effectiveness with 2 to 20 clusters, choosing the number that yields the highest score.

Then click *Generate colors* button. In the *Open file* pop-up window select a folder of source images used for masking and automatic color definition. The progress bars in *Color generating* tab (Fig. 13-1) show how the color generator progresses. When done *Finished* appears on the upper bar (Fig. 13-2).

Colors Generating Auto-Detect Cluster Count Generate Colors	Finished	- 2
Generate Colors		
Colors Generating	9-35-PS_Tray_042-RGB2-RahEyeCorrected	- 1

Fig. 13 Color generating in progress (1) and finished (2)

Color definitions can be exported as a TXT file with the *Store Definitions* button or loaded with the *Load Definitions* button in the *Color Segmentation panel* (Fig. 12-5). The exported file containing the color hue definitions, can subsequently be imported into the Local Analysis section of the PlantData Analyzer software. This allows for the use of the generated color channels to run color segmentation on data stored in the database.

The result of the color segmentation analysis is formulated as a number of pixels in each color class for each plant mask selected object.

2.5 SINGLE RUN

After settings up analysis in the *Settings* tab, click *Show Tray Mask* button or select *Single Run* window in the horizontal tabbed menu. The *Single Run* window (Fig. 14) is used to analyze and preview a single sample image to verify analysis settings.

1	Morpho Analysis - Mask Builder Plant M	- 1.0.14.2 lask Tool Settings	Single Run Batch	Run											
	Load Image	Area Name	AREA_PX	AREA_MM	PERIMETER_PX	PERIMETER_MM	COMPACTNESS	WIDTH_PX	WIDTH_MM	HEIGHT_PX	HEIGHT_MM	[66:60:38]	[101		
		vea 1	60551	6594.0039	1476.704760933	487.31257110789	0.780517672536	320	105.6	329	108.57	7868	5268		
2	Run Analysis	vea 2	306823	33413.0247	3943.756851698	1301.439761060	0.709952472846	731	241.23	834	275.22	262541	4428		
		lvea 3	173379	18880.9731	3019.2673862422	996.3582374599	0.675733399849	763	251.79	605	199.65	166360	7019	·	4
	Run ML Analysis	vea 4	274328	29874.3192	3997.715221138	1319.246022975	0.755849451700	606	199.98	800	264	137784	1365		
5	Show Image — O Grgmal O Masked O Color Segm.					×			ŕ					[6
7	Save Plant Mask Save Image														

Fig. 14 Single Run Tab

In the tab *Single Run* window load a sample image with the *Load Image* button (Fig. 14-1). Click the button *Run Analysis* (Fig. 14-2) to start analysis using the basic *RGB Formula Computation*. If you want to run the advanced *Machine Learning Computation*, click the *Run ML Analysis* button (Fig. 14-3). After completion of analysis, a table with calculated results appear in the table view panel (Fig. 14-4).



The values shown in the result table can be selected by clicking the table panel and selecting required cells. Then, the cells can be copied to clipboard and inserted to a text or spreadsheet editor.

The resulting masked image is displayed in the view panel (Fig. 14-6). In *Show Image* panel (Fig. 14-5) pick one of three viewing modes: *original, masked* and *color segmented* to visualize the masking and color segmentation result. To save the plant mask or the image click *Save Plant Mask* or *Save Image* button (Fig. 14-7).

2.6 BATCH RUN

The Batch Run tab (Fig. 15) is used for analysis on a set of images stored in a local storage folder.



Fig. 15 Batch Run Tab



After verifying correct analysis settings in the *Single Run* tab, load a whole set of input images to the *Batch Run* tab. Click the *Select Images Folder* button (Fig. 15-1) and select the image folder in the *Browse for Folder* pop-up window. In the *Storing Path* text-box (Fig. 15-2) define a path for storing analysis result. Click *Select Path* button (Fig. 15-2) to create and select the folder in the *Browse for Folder* pop-up window.

Then, click *Run Analysis* (Fig. 15-3) and the basic *RGB Formula Computation* image analysis starts. If you want to run the advanced *Machine Learning Computation*, click the *Run ML Analysis* button (Fig. 15-4).

During analysis, the result is continuously updating on the table view panel (Fig. 15-5) and on the image view panel (Fig. 15-6), the last processed image is shown. Analysis results are being exported to the output folder defined in the *Storing Path* (Fig. 15-2). The folder contains a result table, the used plant mask and three sets of images: fish-eye corrected, masked and color segmented.

- *results.csv*: This file contains comma separated values of parameters defined in analysis settings. The file format is compatible with most text spreadsheeed editors.
- PlantMask_originalfilename.xsel: Plant mask definition file.



FEC_originalfilename.png

Original image corrected for fish-eye distortion.



WithMask_originalfilename.png:

Fish-eye corrected image with cropped plant masked objects.

Coloring_originalfilename.png

Image with masked plants colored based on the result of color segmentation.



In addition to shape definition, tray masks used in PlantScreen systems also contain information about pixel size. When pixel size value defined in the tray mask is not equal to zero, several morphological parameters with pixel units are also calculated with milimeter units. These parameters, such as Area (mm2), Perimeter(mm), Width (mm) and Height (mm), are automatically included in the results table (Tab. 4).

Tab. 4 Results Table

Image Name	Area Name	AREA_PX	AREA_MM	PERIMETER_PX	PERIMETE	HEIGHT_MM	[110;111;90]	[90;98;58]	[72;84;58]
18-12-PS_Tray_102-RGB2	A1	12175	335.5669	1029.401154	170.8991	27.89101728	22	6230	412
18-12-PS_Tray_102-RGB2	A2	10787	297.3108953	1081.058008	179.475	33.03757404	57	6253	342
18-12-PS_Tray_102-RGB2	A3	13360	368.2278262	1201.283405	199.4346	37.85209488	69	7976	390
18-12-PS_Tray_102-RGB2	A4	9321	256.9050575	991.5432893	164.614	28.05703524	7	4292	247
18-12-PS_Tray_102-RGB2	A5	14333	395.0456163	1280.479364	212.5826	36.85598712	109	6486	426
18-12-PS_Tray_102-RGB2	B1	6130	168.9548335	875.1341264	145.288	23.2425144	0	2191	371
18-12-PS_Tray_102-RGB2	B2	5781	159.3357083	845.6782823	140.3978	22.4124246	27	3265	298
18-12-PS_Tray_102-RGB2	B3	9997	275.5369445	1126.454978	187.0118	26.06481972	161	6253	254
18-12-PS_Tray_102-RGB2	B4	9908	273.0839298	1072.472222	178.0497	25.23472992	139	5179	263
18-12-PS_Tray_102-RGB2	B5	12221	336.8347503	1222.98189	203.037	33.03757404	32	5951	346
18-12-PS_Tray_102-RGB2	C1	8961	246.9827508	829.7787873	137.7582	23.74056828	0	4687	388
18-12-PS_Tray_102-RGB2	C2	9742	268.508644	929.1341264	154.253	33.03757404	8	5661	382
18-12-PS_Tray_102-RGB2	C3	15504	427.320675	1418.805266	235.5472	37.02200508	634	8373	432
18-12-PS_Tray_102-RGB2	C4	6086	167.7421071	857.8204179	142.4136	25.06871196	3	3378	227
18-12-PS_Tray_102-RGB2	C5	13158	362.6603097	1146.070201	190.2682	29.21916096	57	6664	600
18-12-PS_Tray_102-RGB2	D1	8155	224.7678086	929.5605333	154.3237	25.40074788	0	3643	331
18-12-PS_Tray_102-RGB2	D2	10231	281.9864439	964.8742418	160.1865	28.88712504	59	5344	317
18-12-PS_Tray_102-RGB2	D3	11602	319.7738952	1295.709812	215.1111	31.37739444	99	7114	484
18-12-PS_Tray_102-RGB2	D4	12397	341.6856558	1233.082395	204.7138	28.88712504	54	7404	360
18-12-PS_Tray_102-RGB2	D5	9044	249.2703938	1143.827561	189.8959	28.05703524	0	3477	411
18-15-PS_Tray_102-RGB2	A1	12550	345.9026362	1035.744299	171.9522	27.89101728	11	6811	410
18-15-PS_Tray_102-RGB2	A2	11158	307.5363836	1100.957503	182.7787	34.53173568	68	6801	389

3 BUG REPORTING

Even though we are extensively testing each released version of the software, some bugs still may pass our testing process. Please consider reporting bugs to make this software better. If you encounter a bug or application crash please send a mail with "Morpho Analyzer bug" text in the subject to <u>support@psi.cz</u>. In the message body, describe the sequence of the steps leading to the error.

4 LIST OF FIGURES

Fig. 2 Plant Mask Tool Tab Before Color Segmentation8Fig. 3 Color picker tool8Fig. 4 RGB Formula Computation tab page in Mask Detection Tools9Fig. 5 Graph display performance of the selected threshold line10Fig. 6 Machine Learning Computation tab page in Mask Detection Tools10Fig. 7 Preprocessing Options for machine learning plant mask computation11Fig. 8 Plant mask settings for RGB Formula SegmentationFig. 9 Plant mask settings for Machine Learning ComputationFig. 10 Masking15Fig. 11 Plant Mask Tool Tab After Color Segmentation16Fig. 12 Settings Tab17Fig. 13 Color generating in progress (1) and finished (2)18Fig. 14 Single Run Tab19Fig. 15 Batch Run Tab19	Fig. 1 Mask Builder Tab	6
Fig. 3 Color picker tool 8 Fig. 4 RGB Formula Computation tab page in Mask Detection Tools 9 Fig. 5 Graph display performance of the selected threshold line 10 Fig. 6 Machine Learning Computation tab page in Mask Detection Tools 10 Fig. 7 Preprocessing Options for machine learning plant mask computation 10 Fig. 8 Plant mask settings for RGB Formula Segmentation Fig. 9 Plant mask settings for Machine Learning Computation Fig. 10 Masking 15 Fig. 11 Plant Mask Tool Tab After Color Segmentation 16 Fig. 12 Settings Tab 17 Fig. 13 Color generating in progress (1) and finished (2) 18 Fig. 14 Single Run Tab 19 Fig. 15 Batch Run Tab 19	Fig. 2 Plant Mask Tool Tab Before Color Segmentation	8
Fig. 4 RGB Formula Computation tab page in Mask Detection Tools9Fig. 5 Graph display performance of the selected threshold line10Fig. 6 Machine Learning Computation tab page in Mask Detection Tools10Fig. 7 Preprocessing Options for machine learning plant mask computation11Fig. 8 Plant mask settings for RGB Formula SegmentationFig. 9 Plant mask settings for Machine Learning ComputationFig. 10 Masking15Fig. 11 Plant Mask Tool Tab After Color Segmentation16Fig. 12 Settings Tab17Fig. 13 Color generating in progress (1) and finished (2)18Fig. 14 Single Run Tab19Fig. 15 Batch Run Tab19	Fig. 3 Color picker tool	8
Fig. 5 Graph display performance of the selected threshold line10Fig. 6 Machine Learning Computation tab page in Mask Detection Tools10Fig. 7 Preprocessing Options for machine learning plant mask computation11Fig. 8 Plant mask settings for RGB Formula SegmentationFig. 9 Plant mask settings for Machine Learning ComputationFig. 10 Masking15Fig. 11 Plant Mask Tool Tab After Color Segmentation16Fig. 12 Settings Tab17Fig. 13 Color generating in progress (1) and finished (2)18Fig. 14 Single Run Tab19Fig. 15 Batch Run Tab19	Fig. 4 RGB Formula Computation tab page in Mask Detection Tools	9
Fig. 6 Machine Learning Computation tab page in Mask Detection Tools10Fig. 7 Preprocessing Options for machine learning plant mask computation11Fig. 8 Plant mask settings for RGB Formula SegmentationFig. 9 Plant mask settings for Machine Learning ComputationFig. 10 Masking15Fig. 11 Plant Mask Tool Tab After Color Segmentation16Fig. 12 Settings Tab17Fig. 13 Color generating in progress (1) and finished (2)18Fig. 14 Single Run Tab19Fig. 15 Batch Run Tab19	Fig. 5 Graph display performance of the selected threshold line	10
Fig. 7 Preprocessing Options for machine learning plant mask computation 11 Fig. 8 Plant mask settings for RGB Formula Segmentation Fig. 9 Plant mask settings for Machine Learning Computation 15 Fig. 10 Masking 15 Fig. 11 Plant Mask Tool Tab After Color Segmentation 16 Fig. 12 Settings Tab 17 Fig. 13 Color generating in progress (1) and finished (2) 18 Fig. 14 Single Run Tab 19 Fig. 15 Batch Run Tab 19	Fig. 6 Machine Learning Computation tab page in Mask Detection Tools	10
Fig. 8 Plant mask settings for RGB Formula Segmentation Fig. 9 Plant mask settings for Machine Learning Computation 15 Fig. 10 Masking 15 Fig. 11 Plant Mask Tool Tab After Color Segmentation 16 Fig. 12 Settings Tab 17 Fig. 13 Color generating in progress (1) and finished (2) 18 Fig. 14 Single Run Tab 19 Fig. 15 Batch Run Tab 19	Fig. 7 Preprocessing Options for machine learning plant mask computation	11
Fig. 10 Masking15Fig. 11 Plant Mask Tool Tab After Color Segmentation16Fig. 12 Settings Tab17Fig. 13 Color generating in progress (1) and finished (2)18Fig. 14 Single Run Tab19Fig. 15 Batch Run Tab19	Fig. 8 Plant mask settings for RGB Formula Segmentation Fig. 9 Plant mask settings for Machine Learning Computation	15
Fig. 11 Plant Mask Tool Tab After Color Segmentation 16 Fig. 12 Settings Tab 17 Fig. 13 Color generating in progress (1) and finished (2) 18 Fig. 14 Single Run Tab 19 Fig. 15 Batch Run Tab 19	Fig. 10 Masking	15
Fig. 12 Settings Tab. 17 Fig. 13 Color generating in progress (1) and finished (2) 18 Fig. 14 Single Run Tab. 19 Fig. 15 Batch Run Tab 19	Fig. 11 Plant Mask Tool Tab After Color Segmentation	16
Fig. 13 Color generating in progress (1) and finished (2) 18 Fig. 14 Single Run Tab 19 Fig. 15 Batch Run Tab 19	Fig. 12 Settings Tab	17
Fig. 14 Single Run Tab	Fig. 13 Color generating in progress (1) and finished (2)	
Fig. 15 Batch Run Tab	Fig. 14 Single Run Tab	19
	Fig. 15 Batch Run Tab	19