How To Use CalMorph

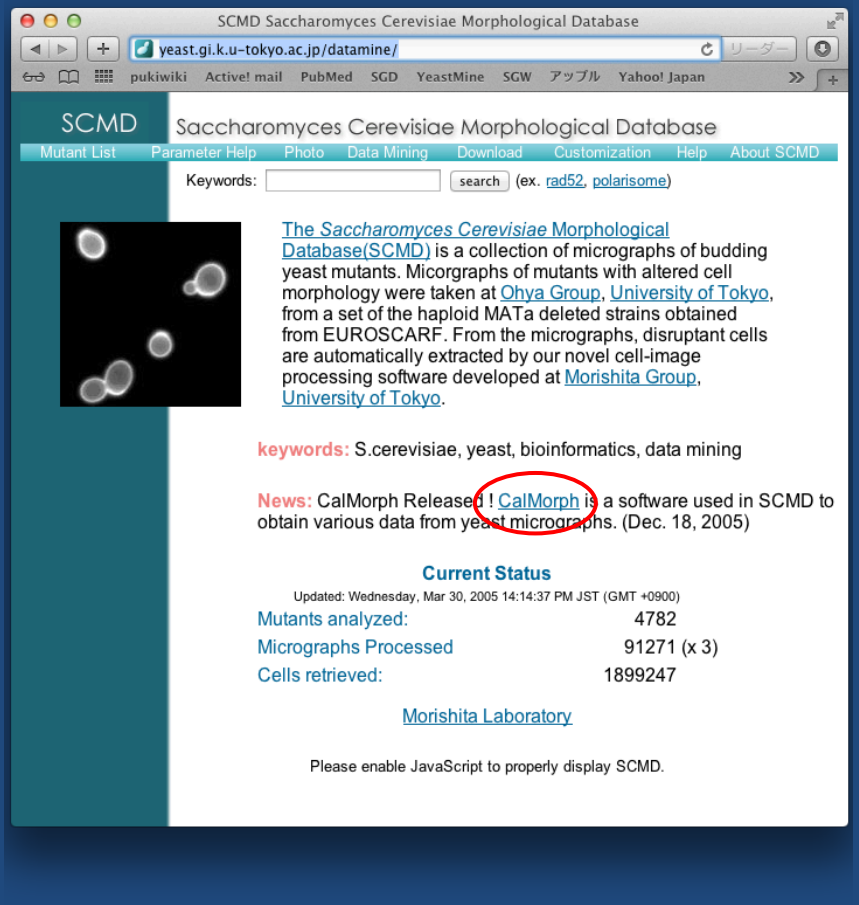
Yoshikazu OHYA

Univ. Tokyo

Topics

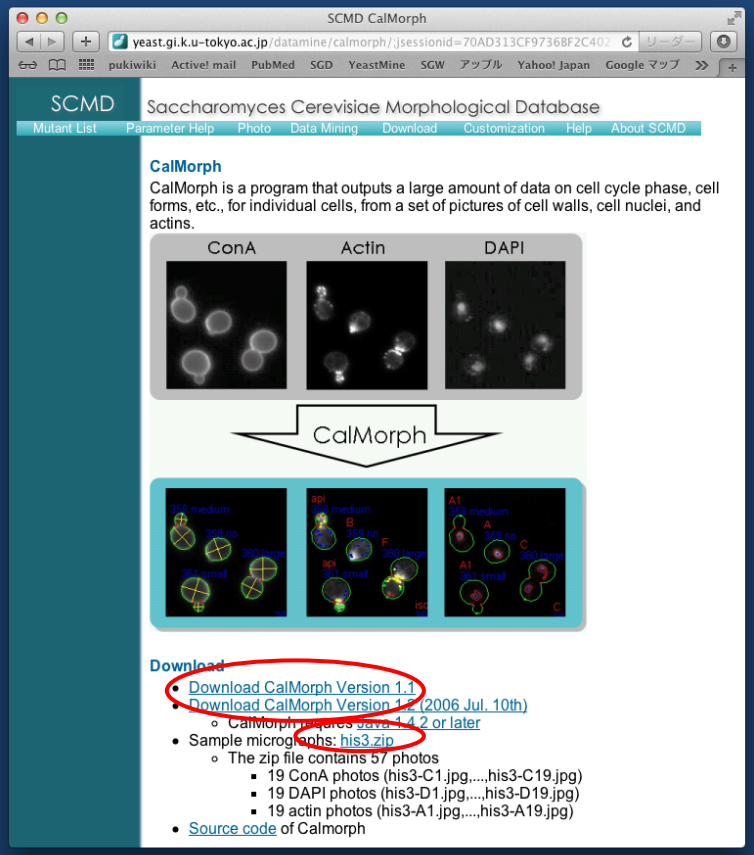
- Download
- Start CalMorph
- Set Input Files
- Set Output Folder
- Set Analysis Option
- Start Image Analysis
- Check Output files
- Requirements
- Helps

Access to SCMD
http://scmd.gi.k.u-tokyo.ac.jp/datamine/



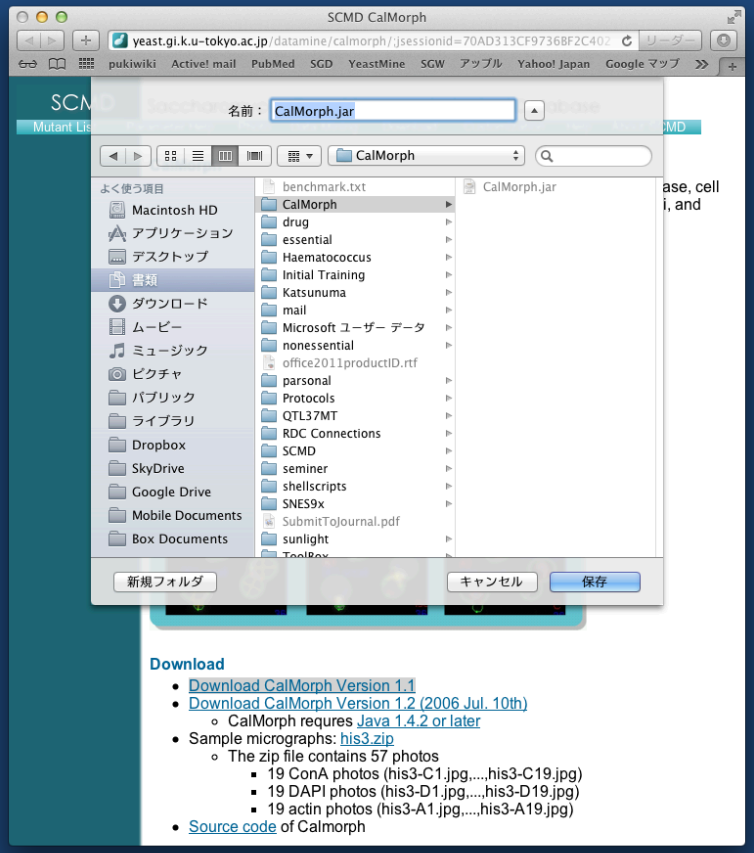
- You can see a home page of SCMD
  - *Saccharomyces Cerevisiae* Morphological Database
- Click CalMorph at the middle of the page

Download CalMorph from SCMD



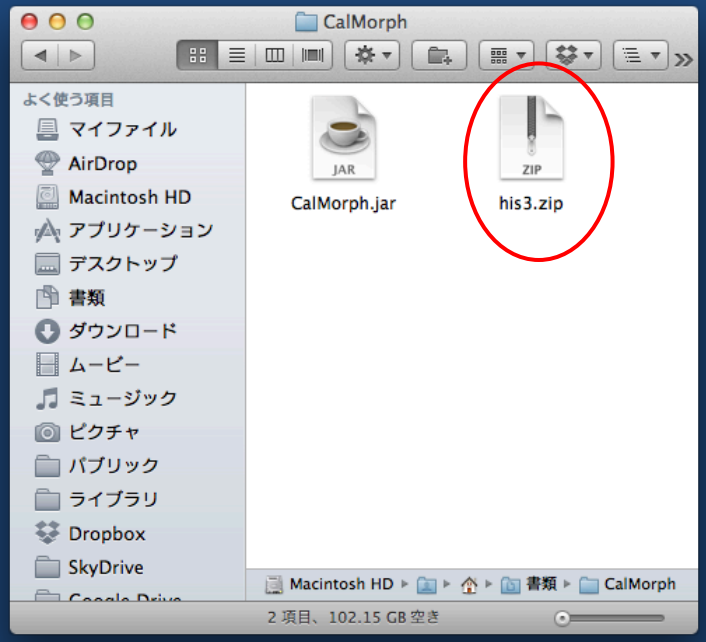
- Click “Download CalMorph Version 1.1”
- Click “his3.zip”
  - Test images

Save “CalMorph.jar” and “his3.zip”

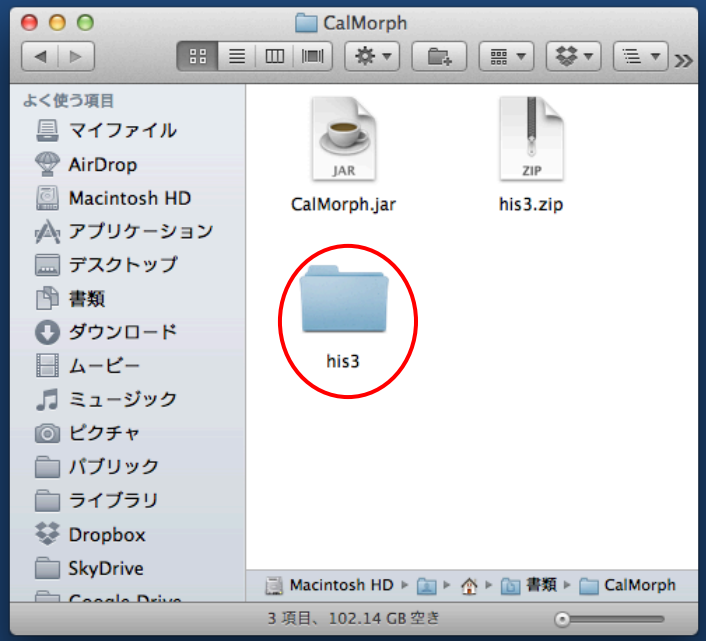


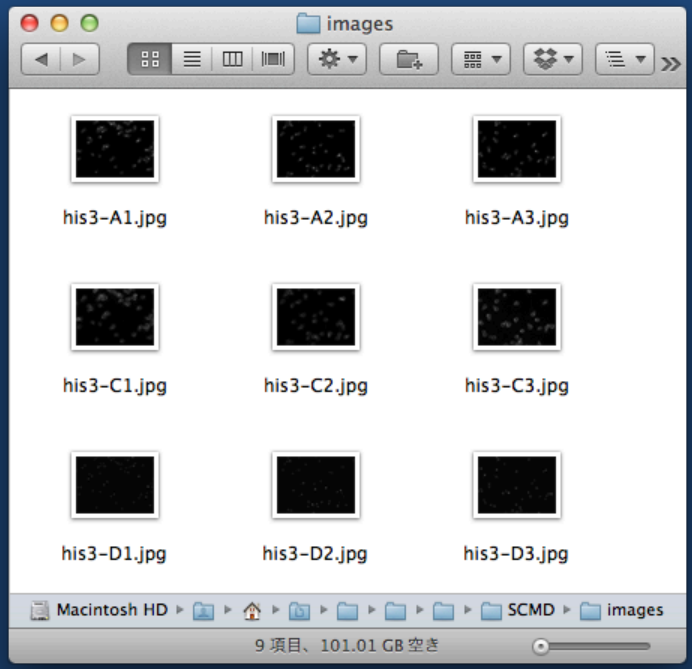
- In this case, the files are saved at ~/Documents/CalMorph

Unzip “his3.zip”



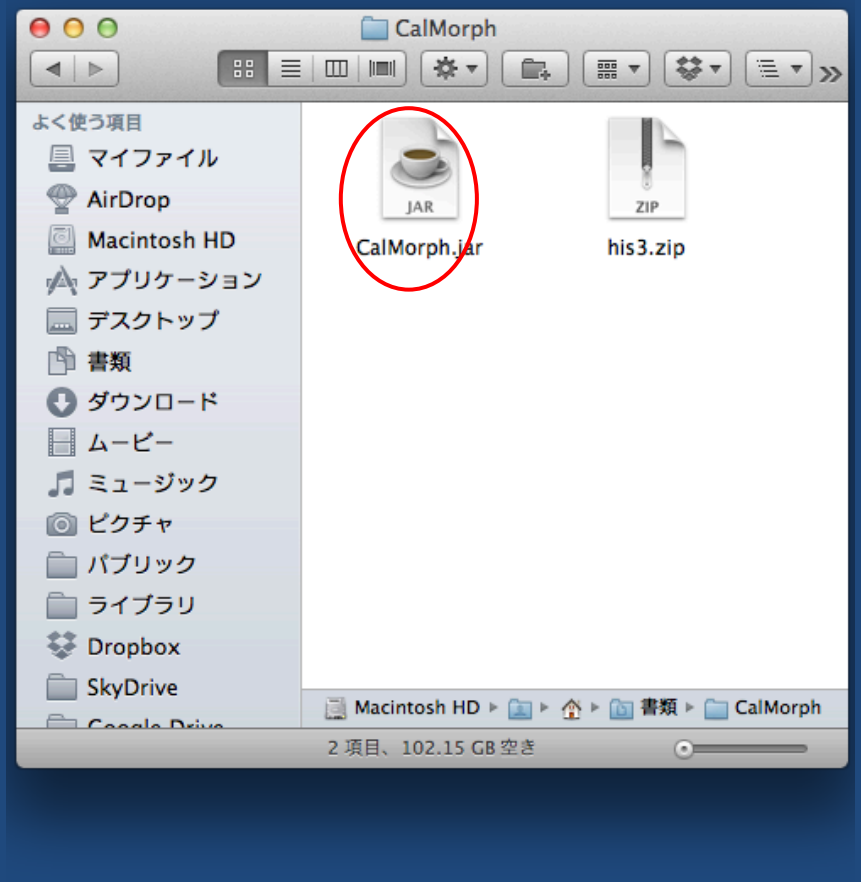
Check file names





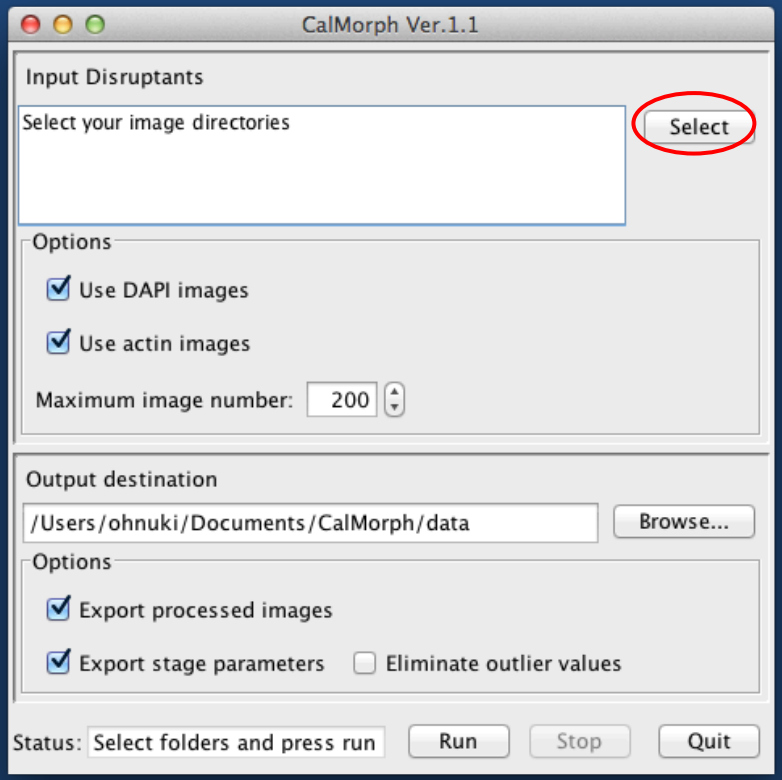
[folder name]-[A|C|D][number].jpg

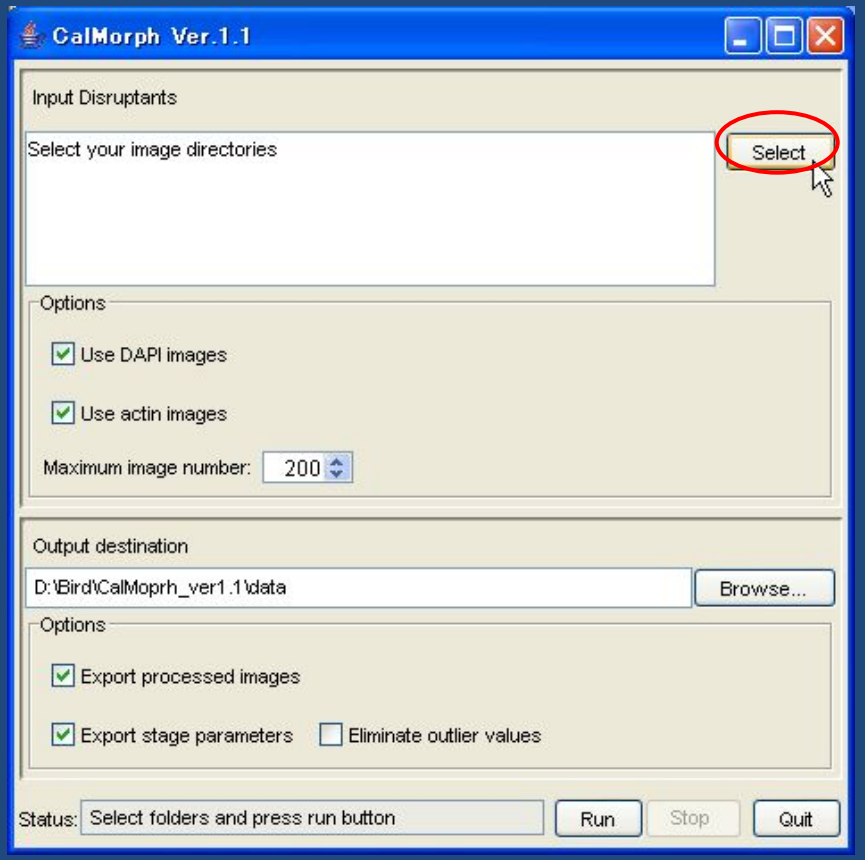
Start CalMorph



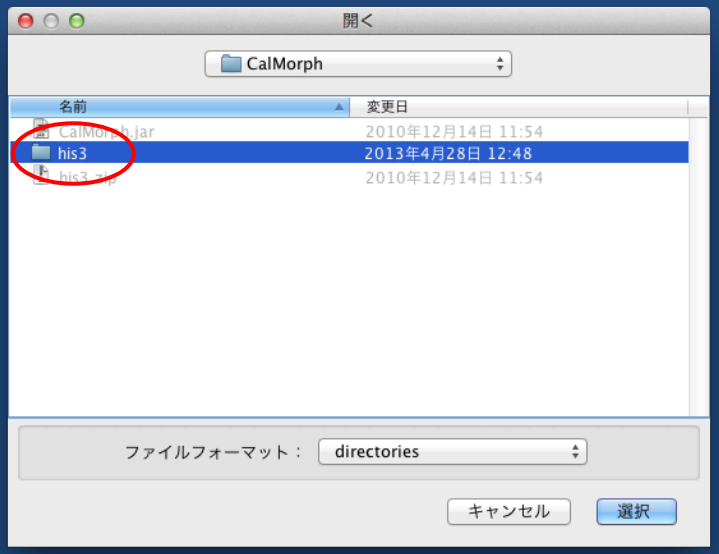
- Double click “CalMorph.jar”

Set Input Files



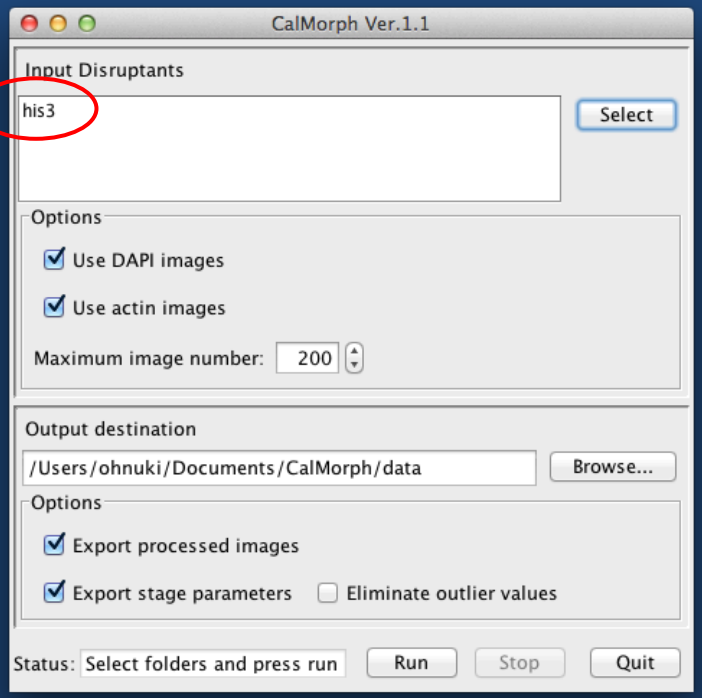


Select “his3” folder



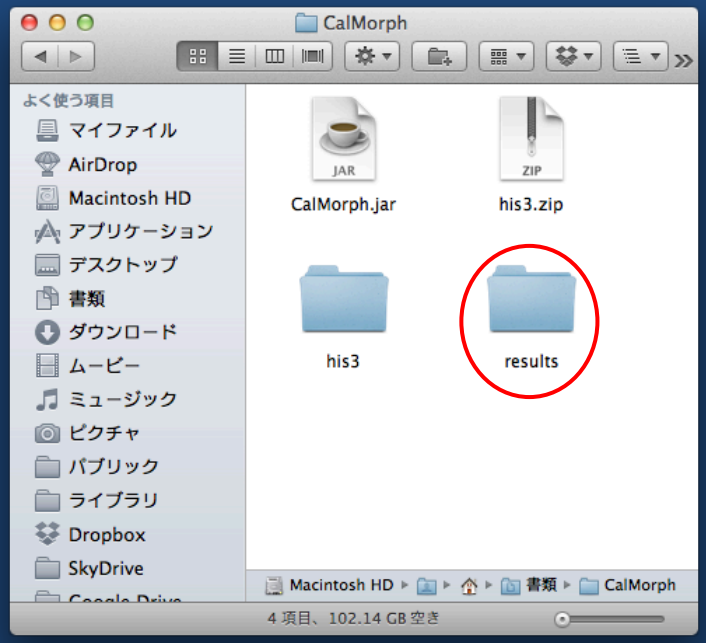
- If you want to analyze two or more sets of images, you can select multiple folders in this step by saving the folders of images at a same folder in advance

Check the selected folder name



- If you selected multiple folders, all selected folders appear here

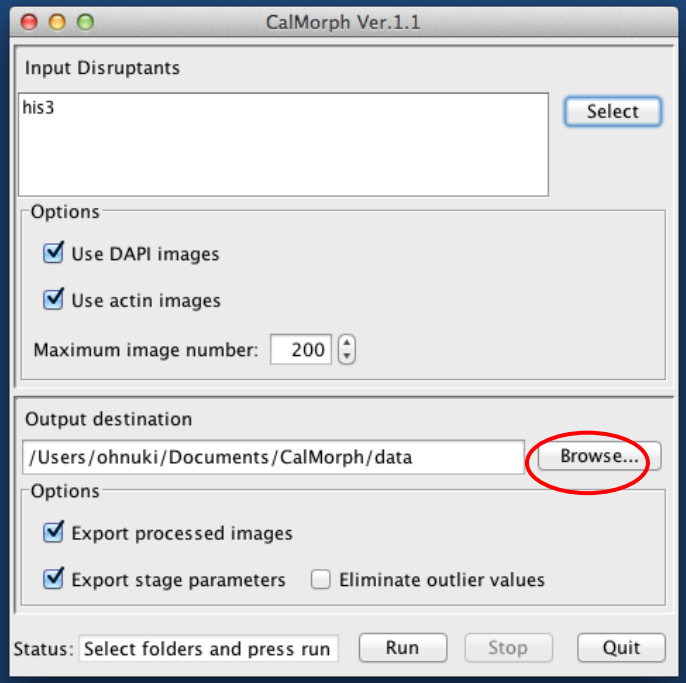
Make a folder “results” for output



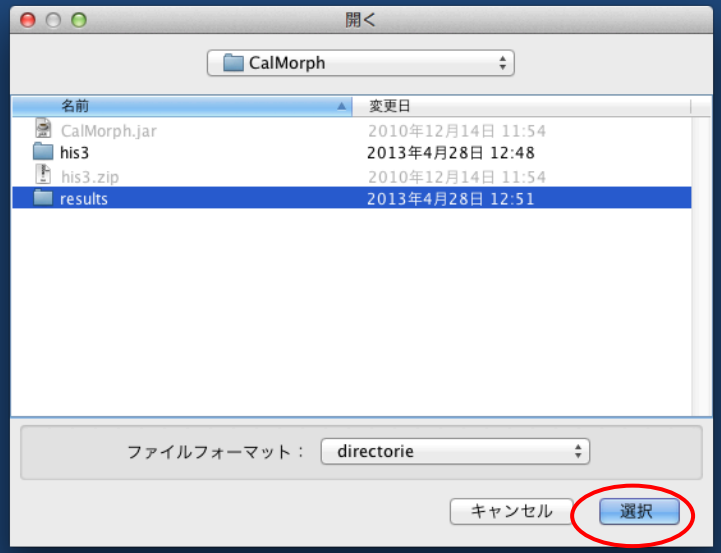
- Output files from CalMorph will stored in this folder

Set Output Folder

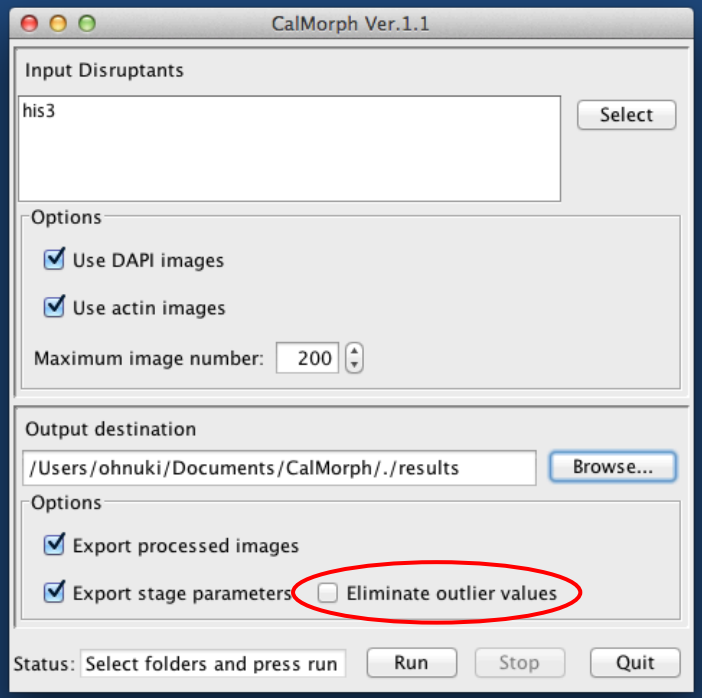
Click “Browse” button



Select output folder



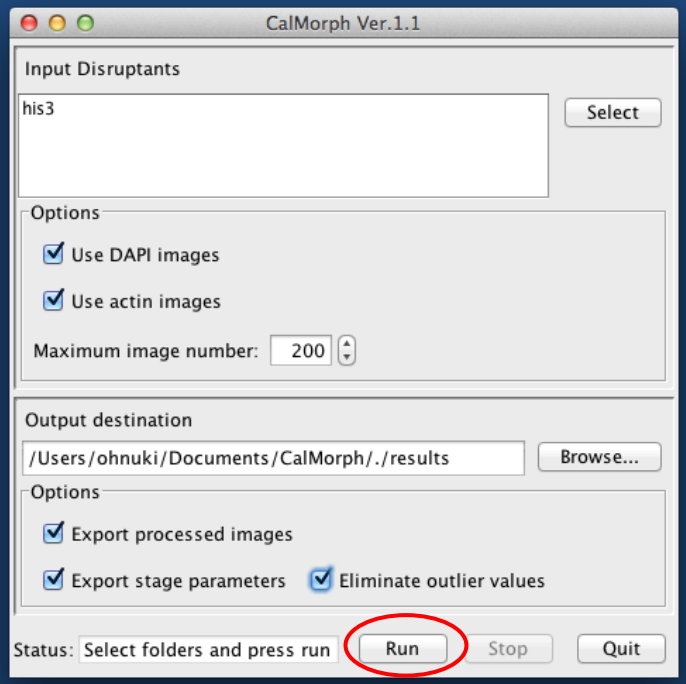
Set Analysis Option



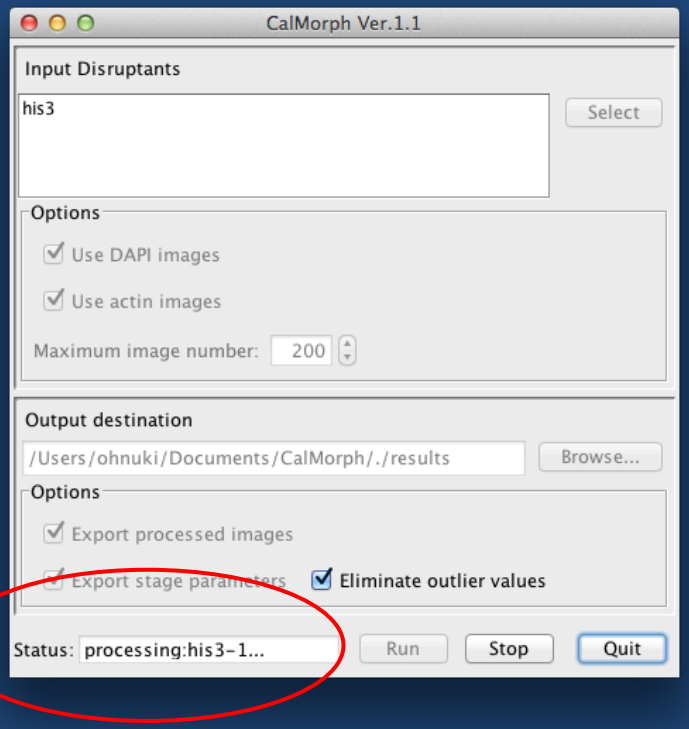
- Select “Eliminate outlier values”
- CalMorph eliminate 1% outliers before calculating mean values in each parameter

Start Image Analysis

Click “Run” button

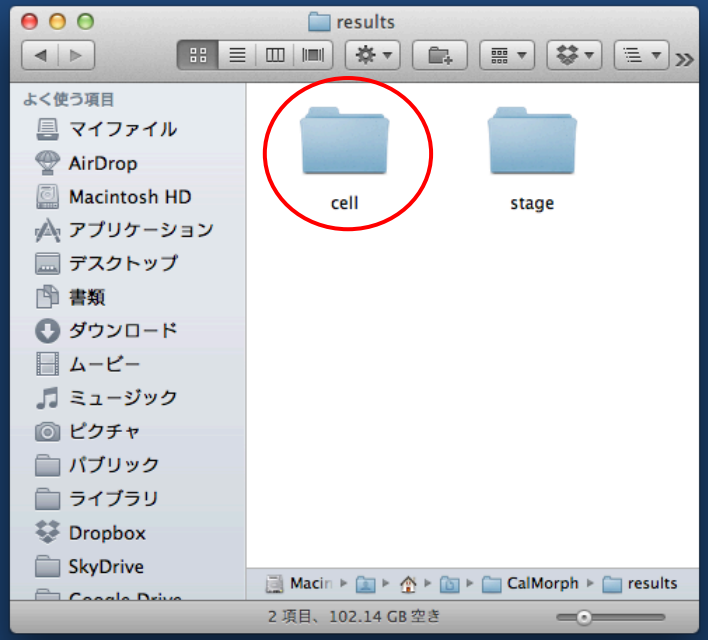


Check status

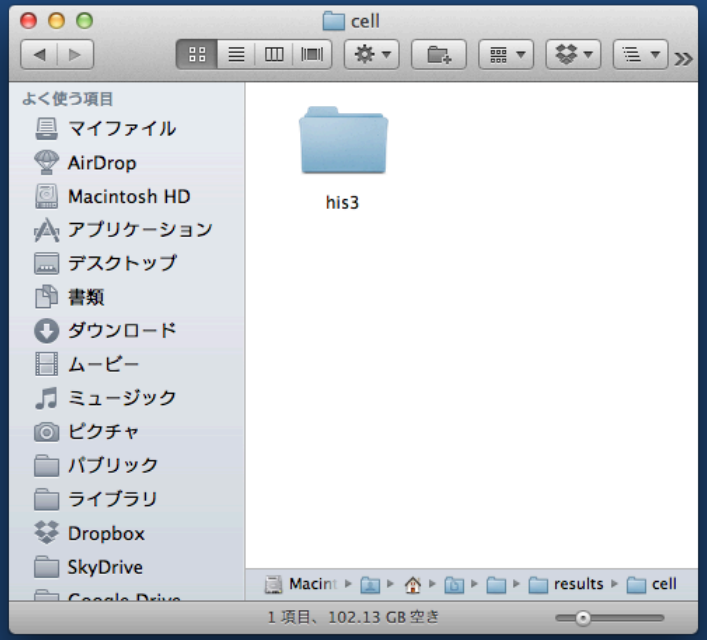


Check Output Files

“results” folder

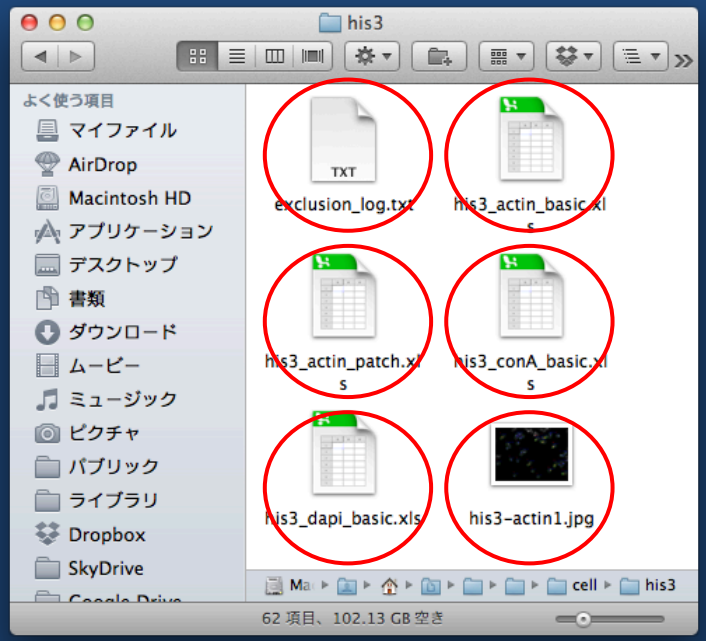


“cell” folder



Folders of same names as inputs will be made in a “cell” folder

“cell” folder



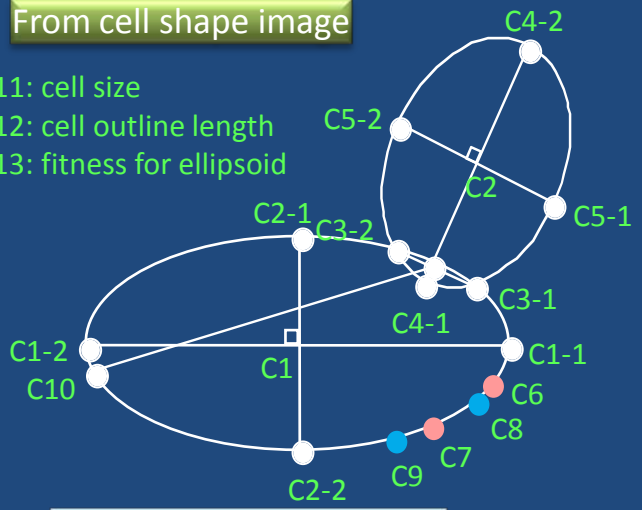
- JPG files of analyzed image
- Error-log file
  - exclusion_log.txt
- Excel files for basic parameter values
  - his3_actin_basic.xls
  - his3_actin_patch.xls
  - his3_conA_basic.xls
  - his3_dapi_basic.xls

Basic Parameters

Feature points that are extracted directly from the images
to calculate biologically significant parameters

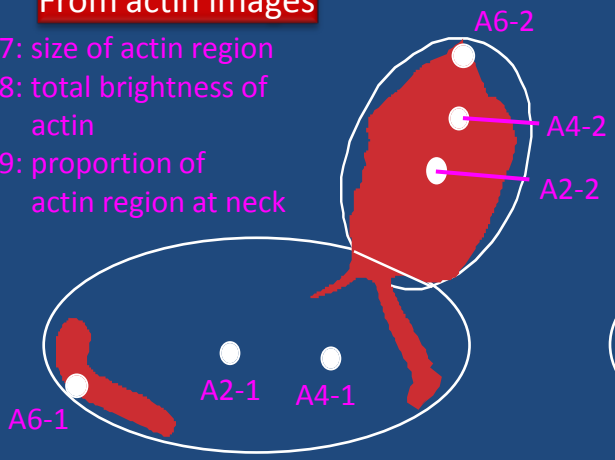
From cell shape image

C11: cell size
C12: cell outline length
C13: fitness for ellipsoid



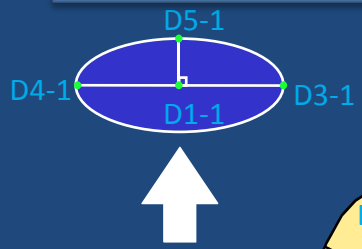
From actin images

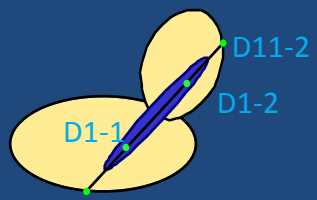
A7: size of actin region
A8: total brightness of actin
A9: proportion of actin region at neck

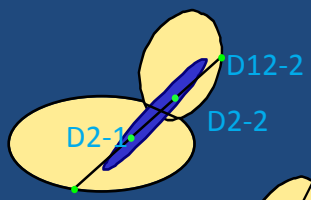


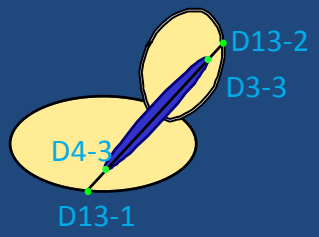
A1: Number, location, size, brightness in each actin patch

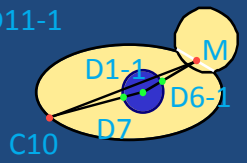
From nuclear DNA images

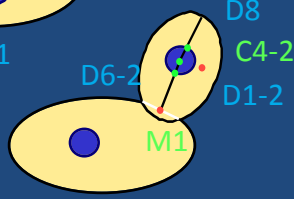




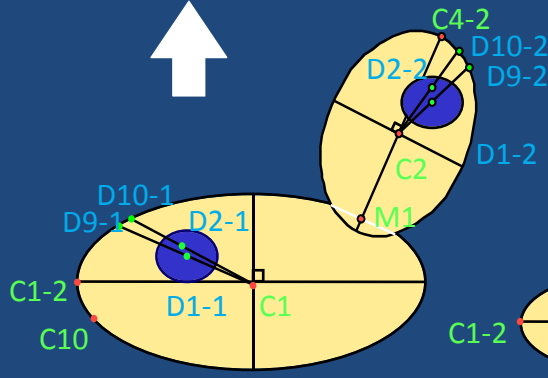


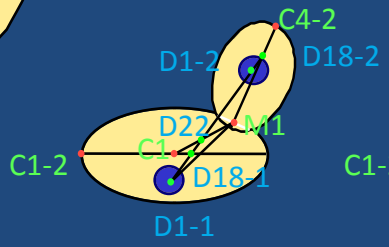


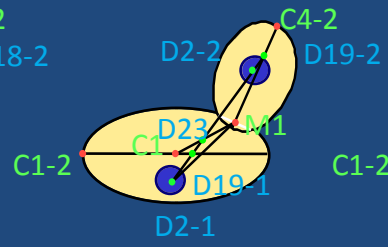


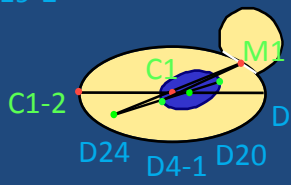


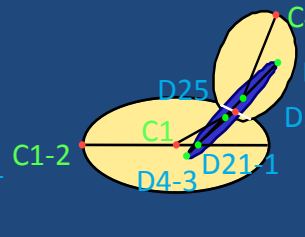
D14: nuclear size
D15: sum of nuclear brightness
D16: maximal brightness of nuclei











Biological Parameters

From Cell Shape Image

C101 whole cell size
C102 whole cell outline length
C103 long axis length in mother
C104 short axis length in mother
C105 neck position
C106 bud direction
C107 long axis length in bud
C108 short axis length in bud
C109 neck width
C110 distance between bud tip and mother long axis extension
C111 distance between bud tip and mother short axis extension
C112 distance between middle point of neck and mother center
C113 distance between bud tip and mother long axis
C114 bud axis ratio
C115 mother axis ratio
C116 axis ratio ratio
C117 outline ratio
C118 size ratio
C119 no bud ratio
C120 small bud ratio
C121 medium bud ratio
C122 large bud ratio
C123 small bud ratio to bud
C124 medium bud ratio to bud
C125 large bud ratio to bud
C126 brightness difference
C127 thickness difference
C128 distance between middle point of neck and C10

From Actin Image

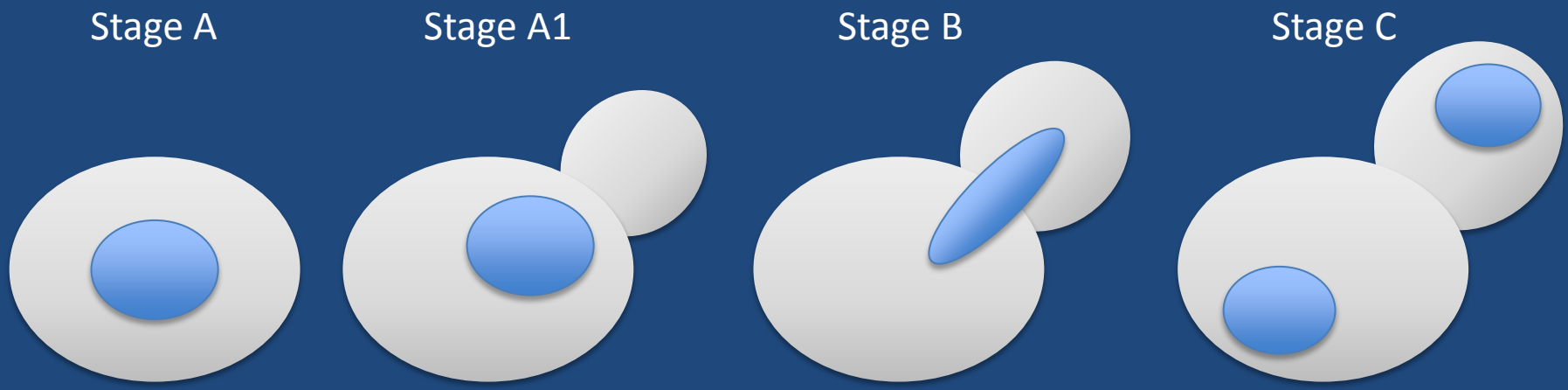
A101 actin region ratio
A102 bud actin region ratio
A103 relative gravity center of weighted actin region in mother
A104 relative gravity center of weighted actin region in bud
A105 ratio of the no bud cells with delocalized actin
A106 ratio of no bud cells with localized actin
A107 ratio of budded cells with bud-tip localized actin
A108 ratio of budded cells with actin dispersed in bud
A109 ratio of budded cells with delocalized actin
A110 ratio of budded cells with neck localized actin
A111 ratio of cells with delocalized actin
A112 ratio of cells with localized actin
A113 ratio of cells with no actin
A114 ratio of delocalized actin to budded cells
A115 ratio of localized actin to budded cells
A116 ratio of bud-tip localized actin to budded cells
A117 ratio of dispersed actin to budded cells
A118 ratio of delocalized actin to budded cells
A119 ratio of neck localized actin to budded cells
A120 total length of actin patch link
A121 distance between the most distant patches
A122 the number of bright actin patches
A123 ratio of actin patches to actin region

From Nuclear DNA Image

D101 D1-1C1-1 or D1-1C1-2
D102 D1-1C1-2
D103 D1-1M1
D104 D1-2M1
D105 D1-3-1M1
D106 D1-3-2M1
D107 D1-1D1-2
D108 D1-1C1
D109 D1-3-1C1 or D1-3-2C1
D110 D1-2C2
D111 D1-3-1C2 or D1-3-2C2
D112 D1-2C4-2
D113 D1-3-1C4-2 or D1-3-2C4-2
D114 D1-1C10
D115 D1-3-1C10 or D1-3-2C10
D116 D2-1C1-1 or D2-1C1-2
D117 D2-1C1-2
D118 D2-1M1
D119 D2-2M1
D120 D2-3-1M1
D121 D2-3-2M1
D122 D2-1D2-2
D123 D2-1C1
D124 D2-3-1C1 or D2-3-2C1
D125 D2-2C2
D126 D2-3-1C2or D2-3-2C2
D127 D2-2C4-2
D128 D2-3-1C4-2 or D2-3-2C4-2
D129 D2-1C10
D130 D2-3-1C10 or D2-3-2C10
D131 D6-1M1
D132 D6-2M1
D133 D7C10
D134 D8C4-2
D135 D1-1C1 / C1D9-1
D136 D2-1C1 / C1D10-1
D137 D1-2C2 / C2D9-2
D138 D2-2C2 / C2D10-2
D139 D6-2M1 / D6-1M1
D140 D6-1M1 / D7C10
D141 D6-2M1 / D8C4-2
D142 ∠D1-1C1C1-2
D143 ∠D2-1C1C1-2
D144 ∠D1-2C2C4-2
D145 ∠D2-2C2C4-2
D146 ∠D1-1D18-1C1-2
D147 ∠D2-1D19-1C1-2
D148 ∠D4-1D20-1C1-2
D149 ∠D4-3D21-1C1-2
D150 ∠D1-1D22C1
D151 ∠D2-1D23C1
D152 ∠D4-1D24C1
D153 ∠D4-3D25C1
D154 ∠D1-2D18-2C4-2
D155 ∠D2-2D19-2C4-2
D156 ∠D3-3D21-2C4-2
D157 ∠D1-1M1C1
D158 ∠D2-1M1C1
D159 ∠D4-1M1C1
D160 ∠D4-3M1C1
D161 D1-1D3-1
D162 D1-2D3-2
D163 D1-3-1D3-3 or D1-3-2D3-3
D164 D3-1D4-1
D165 D3-2D4-2
D166 D3-3D4-3
D167 D1-1D5-1
D168 D1-2D5-2
D169 D1-3-1D5-3 or D1-3-2D5-3
D170 D3-1D4-1 / D1-1D5-1
D171 D3-2D4-2 / D1-2D5-2
D172 D3-3D4-3 / D1-3D5-3
D173 D1-1D1-2 / D11-1D11-2
D174 D2-1D2-2 / D12-1D12-2
D175 D3-3D4-3 / D13-1D13-2
D176 D1-1D2-1
D177 D1-2D2-2
D178 D1-3D2-3
D179 D15-1 / D14-1
D180 D15-2 / D14-2
D181 D15-3 / D14-3
D182 D16-1 / (D15-1 / D14-1)
D183 D16-2 / (D15-2 / D14-2)
D184 D16-1 / (D15-1 / D14-1)
D185 D14-2 / D14-1
D186 D15-2 / D15-1
D187 A_ratio
D188 A1_ratio
D189 B_ratio
D190 C_ratio
D191 D_ratio
D192 E_ratio
D193 F_ratio
D194 A_ratio_to_no_bud
D195 A1_ratio_to_bud
D196 B_ratio_to_bud
D197 C_ratio_to_bud

501 parameters

Classification of Biological Parameters

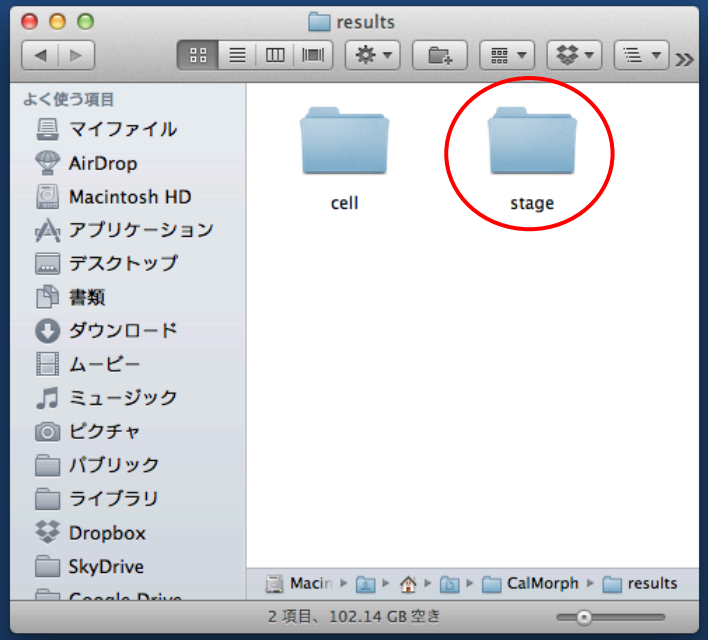


Category of Parameters	Cell Wall	Actin	Nucleus	Total
Stage A	16	17	40	73
Stage A1B	55	32	68	153
Stage C	55	32	148	235
Population	7	15	18	40
Total	133	96	272	**501**

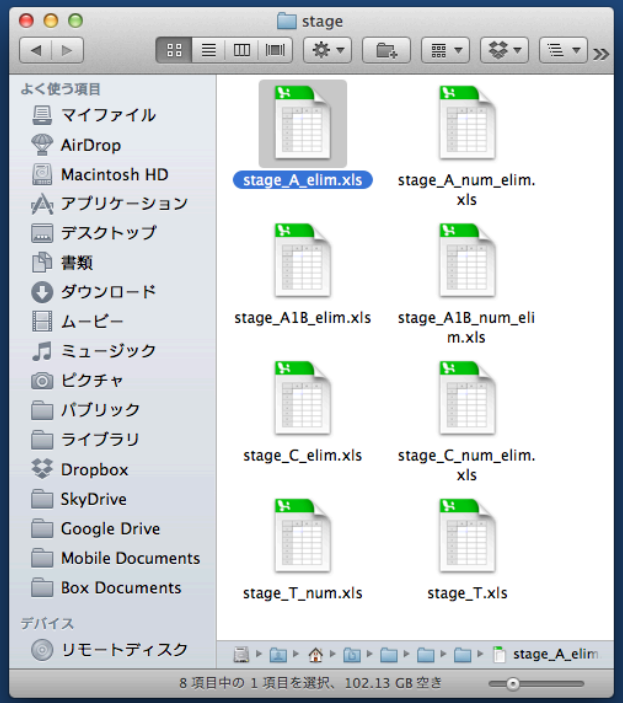
CalMorph outputs 501 parameter values in an Excel format

Check Output Files in “stage” folder

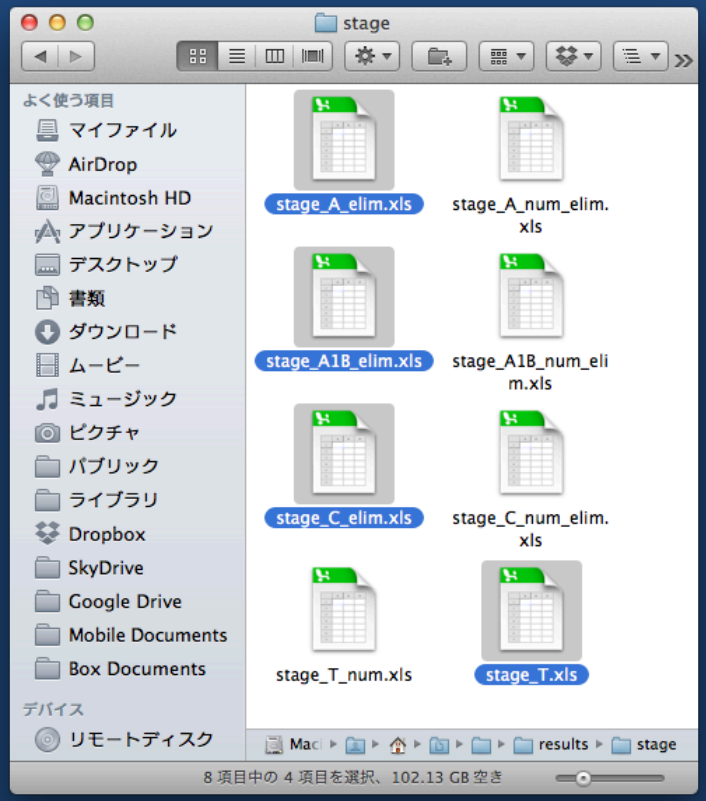
“results” folder



“stage” folder

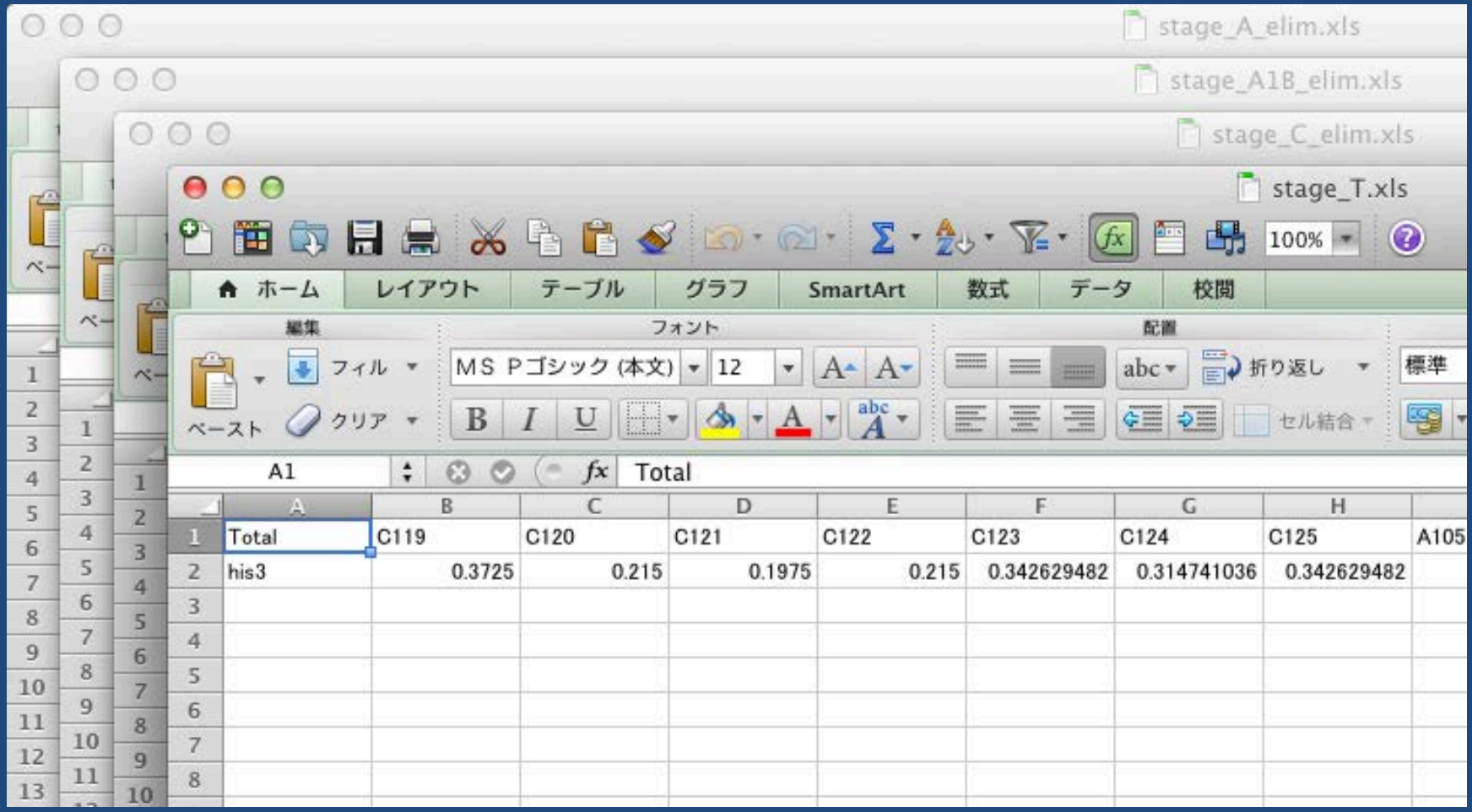


Files of Biological Parameters



- stage_A_elim.xls
- stage_A_num.xls
- stage_A1B_elim.xls
- stage_A1B_num.xls
- stage_C_elim.xls
- stage_C_num.xls
- stage_T_num.xls
- stage_T.xls

Check Parameter Values



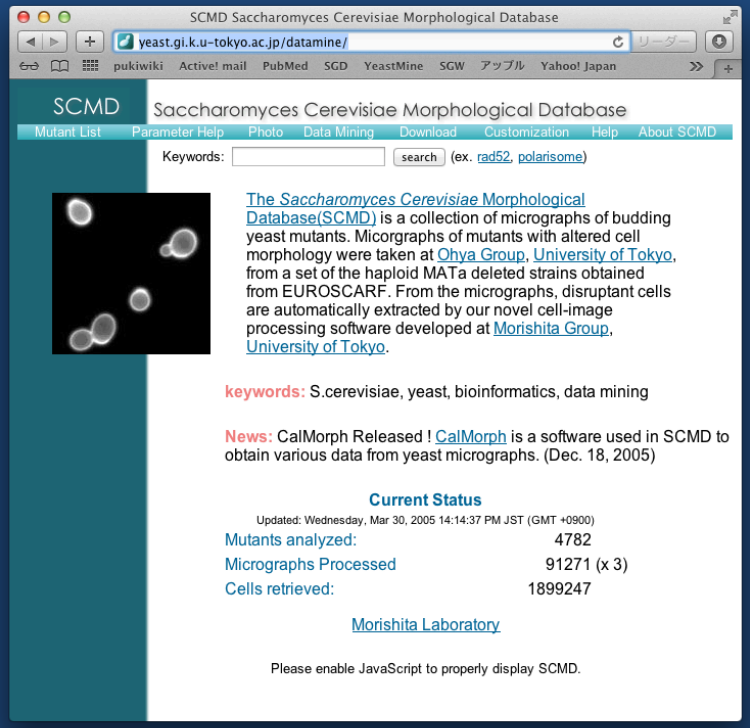
If you input multiple samples by selecting multiple folders, results of each sample will be stored at each row in the files

Requirements of CalMorph

- Java runtime version: 1.4.2 or later
- Image size: 520 x 696 pixels
- Format of image files: 8 bit gray scale JPG
  - Images of cell shape (cell wall stained by FITC-ConA)
  - Three images for each view window
- Format of file names:
  - [folder name]-[A|C|D][number].jpg
- Pixel size: 0.129 x 0.129 μm^2/pixel
  - x100 object lens
  - 2 x 2 binning
  - CCD of 6.45 μm/pixel (ex. Photometrix CoolSNAP HQ)

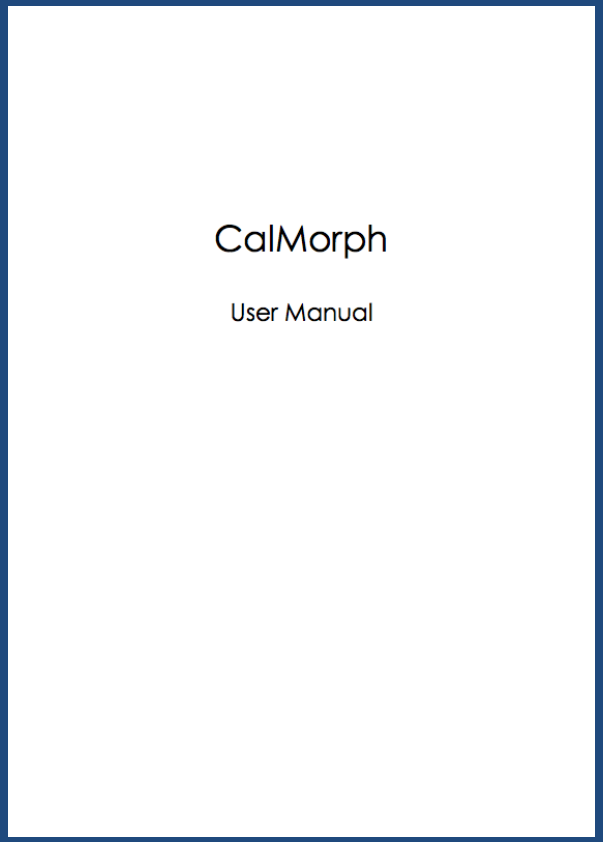
Helps for CalMorph

Saccharomyces cerevisiae morphological database



http://scmd.gi.k.u-tokyo.ac.jp/datamine/

CalMorph User Manual



Available at download page of SCMD

Helps for Parameters

High-dimensional and large-scale phenotyping of yeast mutants

Ohya et al., 2005, PNAS

Figure 5 for Basic Parameter

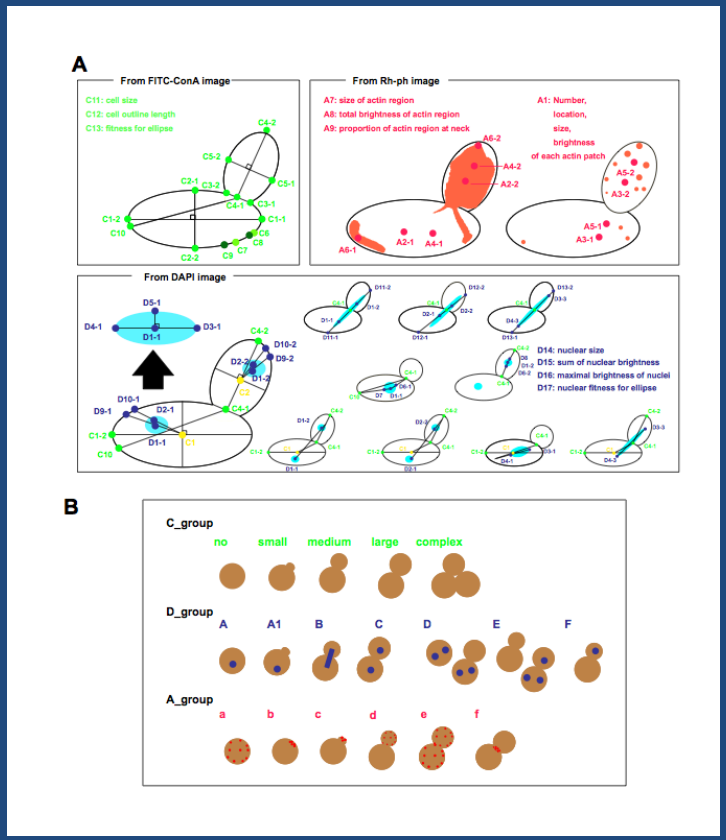
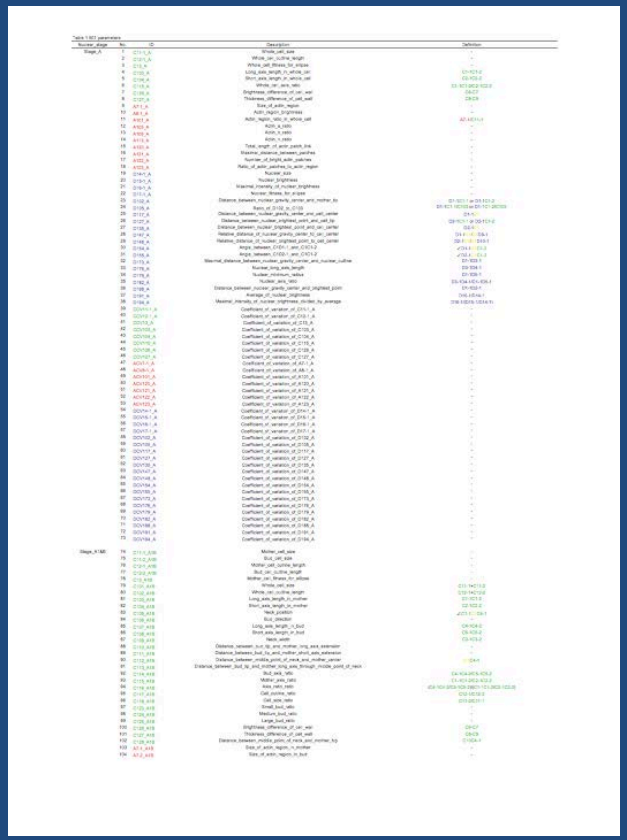


Table 1 for Biological Parameter



Thank you for your attention!

http://scmd.gi.k.u-tokyo.ac.jp/datamine/