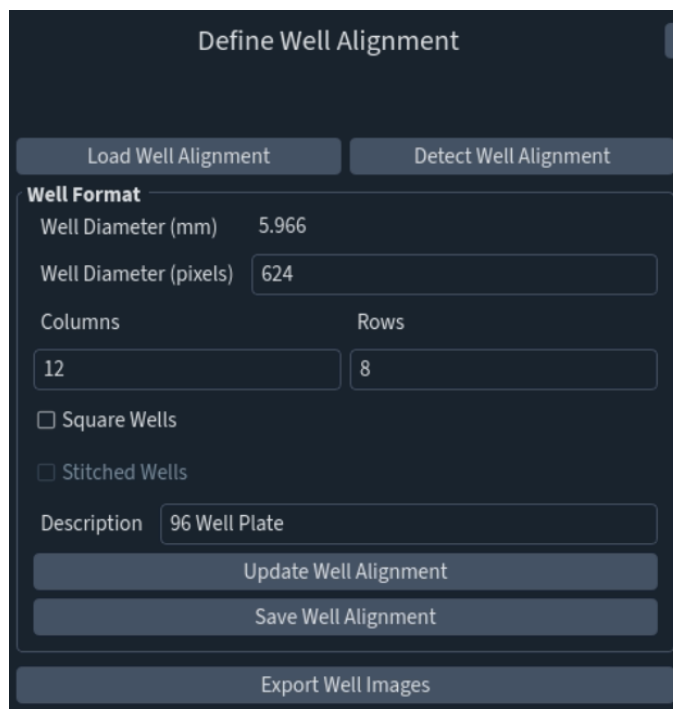


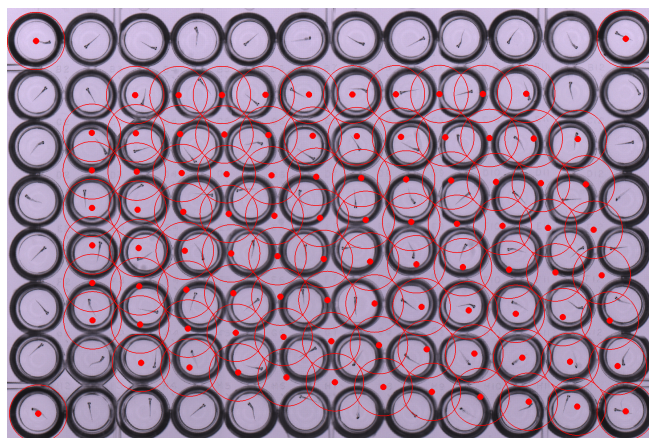
Create a Well Alignment File

This step creates a file that will be used to detect well plate wells for analysis.



The 'Define Well Alignment' dialog box contains the following fields and buttons:

- Buttons:** 'Load Well Alignment', 'Detect Well Alignment', 'Update Well Alignment', 'Save Well Alignment', and 'Export Well Images'.
- Well Format Section:**
 - Well Diameter (mm):** 5.966
 - Well Diameter (pixels):** 624
 - Columns:** 12
 - Rows:** 8
 - ☐ Square Wells
 - ☐ Stitched Wells
 - Description:** 96 Well Plate



Step	Action
1	Open .nc dataset file in MCAM Viewer or open the MCAM Software
2	Go to Tools > Well Alignment to open Well Alignment Panel
3	Under Well Format, enter values into Columns, Rows, and Well Diameter (pixels) fields for well plate depicted in open .nc dataset file that correspond to values in

	Table 1: Well Plate Configuration below Upon entry of the above, Well Diameter (mm) field will auto-update
3a	If well plate contains square wells, check the box beside Square Wells
4	Click Detect Well Alignment. Well Indicators will now appear. To create Well Alignment File for tracking, Well Indicators must be aligned with wells
5	Well Indicators may not be aligned with wells after Step 4. If so:
5a	Double click on center point each of the 4 corner Well Indicators, hold, and drag into alignment with each of the 4 corner wells.
5b	In Well Alignment panel, click Update Well Alignment to align remaining Well Indicators
5c	Make individual well adjustments by clicking and dragging any misaligned Well Indicators to correct locations and clicking Update Well Alignment.
5d	To simultaneously resize all Well Indicators, double click on any Well Indicator boundary, hold, and drag
6	Click Save Well Alignment in the Well Alignment panel to save the Well Alignment File.

Table 1: Well Plate Configuration

The metrics in this table describe common well plate configurations. The MCAM uses them to detect well locations in a .nc dataset file. **Note:** For use with tracking workflows, well diameters must match the specified values. *Pixel widths that do not match these values are not supported, and data sets collected with incorrect values will not be successfully analyzed.*

Table 1 - Creating a Well Alignment File: Well Plate Configuration			
Well Plate Configuration	Columns	Rows	Well Diameter (pixels)
96-well plate	12	8	1024
48-well plate	8	6	1408
24-well plate	6	4	2048

12-well plate (Ramona custom well plate)	3	4	2056
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