Particle Counting and Analysis.

Problem: Count and determine the size distribution of a collection of echinoderm embryos. (Open embryos image via Select File → Open Samples → Embryos)

- Draw line over the scale bar and select Analyze → Set Scale
  In Set Scale window enter 100 into the 'Known Distance' box and change the 'Unit of Measurement' box to um, check 'Global'

- Confirm that the measurement scale is correct.

- Convert the image to grayscale:
  Image → Type → 8-bit

- Threshold the image using the automated routine:
  Process → Binary → Make Binary

- Surround the scale bar with the rectangular selection tool and clear the contents (Edit → Clear)

- Analyze Particles:
  Analyze → Analyze Particles
  Enter 20 as the minimum particle size, toggle 'Show Outlines', check 'Display Results', 'Summarize' and 'Record Stats' and click 'OK'
  Twenty five embryos are counted, numbered and outlined. The data window lists the area (in um²) for each embryo. These data could be copied to a spreadsheet.

| Threshold: 0-0 |
| Count: 25     |
| Total Area: 3177.6 μm² |
| Average Size: 127.1 μm² |
| Area Fraction: 3.72%  |

A summary of the particle count is also shown in another data window.

As mentioned in the previous example, this technique should be manually validated before collecting experimental data.