

## Oxygen sensing

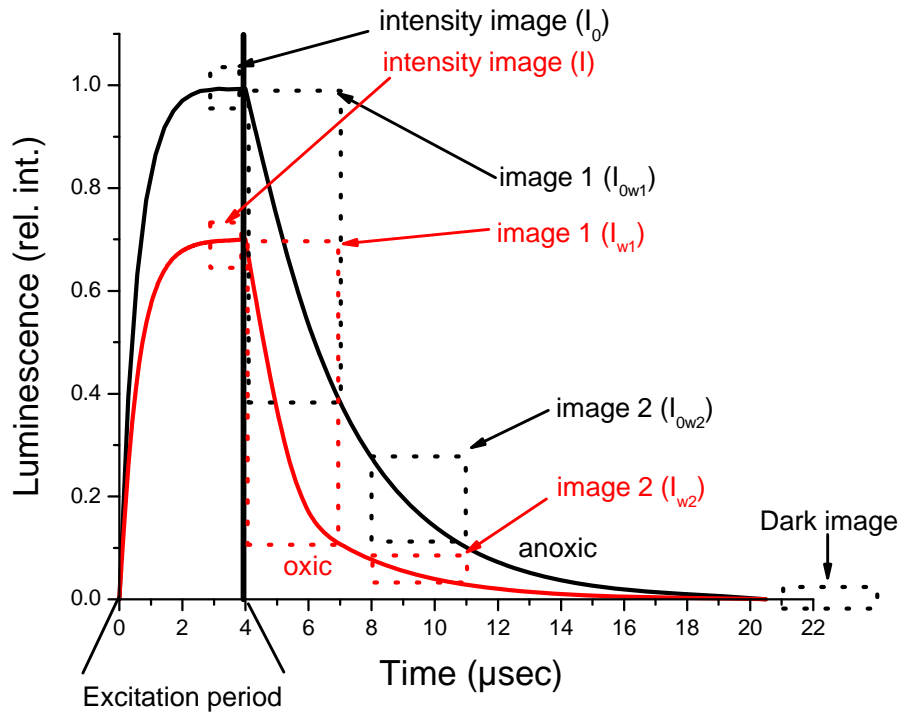


Figure X-1. Scheme to explain the different images taken for the construction of the oxygen images. The black line indicates the luminescence signal under anoxic conditions during one light pulse, while the red line indicates the luminescence signal under oxic conditions. The boxes indicate when the different images are taken. The table below denotes which cameras, images and equations are used for the calculation of the oxygen images.

Table X-1. Overview of the types of images and calculations made to come to the different oxygen images. Dark images are subtracted from all fluorescence images to correct for background grey values.

Camera	Method	Image	Calculation	Illumination	Integration time (s)
PCO	Lifetime	$I_{w1}, I_{w2}$	$\Delta t / \ln(I_{w1}/I_{w2})$	Modulated	0.5
PCO	Ratiometric	$I_{ow1}, I_{w1}$	$I_{ow1}/I_{w1}$	Modulated	0.5
PCO	Ratiometric	$I_0, I$	$I_0/I$	Continuous	0.1
$\mu$ Eye	Ratiometric	$I_0, I$	$I_0/I$	continuous	0.2

## Rhodamine assay

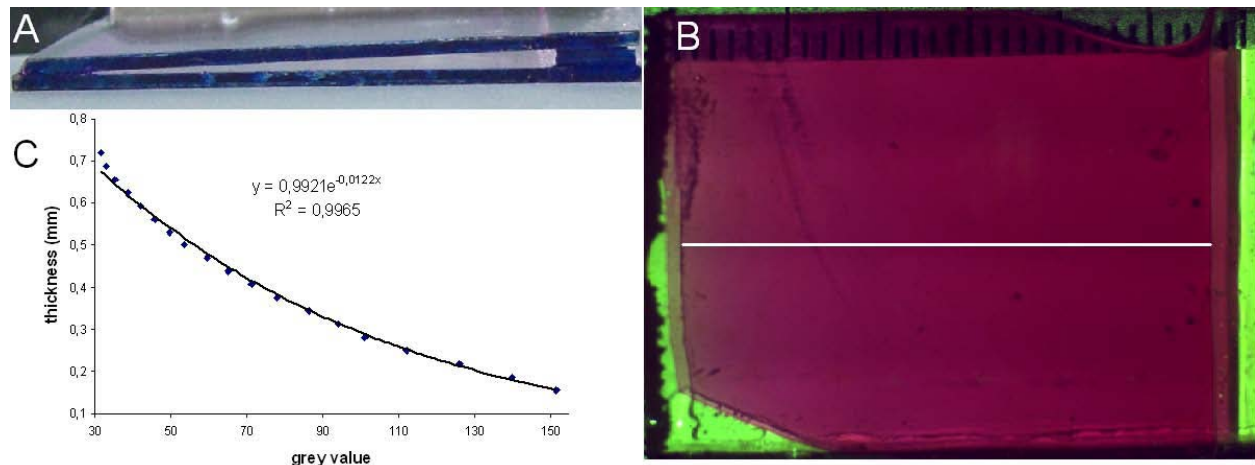


Figure X-2. Calibration cuvette for the water thickness calculations. (A) Top view of the cuvette: two transparent blades are glued in order to create an angle. A gradient of thickness is obtained. (B) Calibration image. The red dye is introduced in the calibration tool, resulting in a gradient in absorption. (C) The calibration curve links the thickness of the B-rhodamine solution to the grey values of the green channel of the picture in panel B. The curve corresponds to the line drawn on image (B).

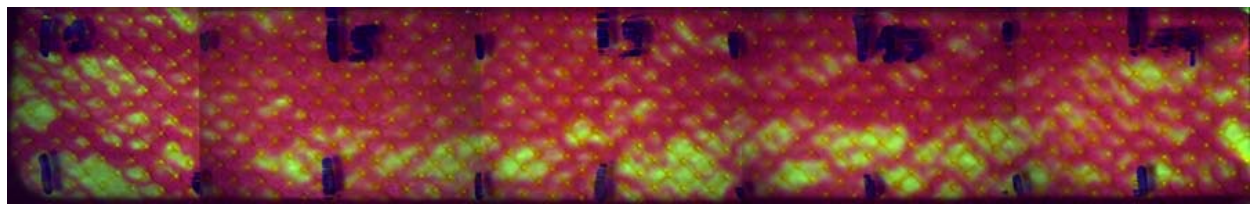


Figure X-3. An example of an image showing the biomass distribution. 5 images are stitched together to cover the whole MFS to give a good idea on the biomass development in the incubator. The water is colored by the rhodamine WT solution, allowing to discriminate the areas where the water flows (red) and the areas covered by a biofilm (yellow).