

# JotDyes for Membrane Labelling

## Introduction

Jotbody's JotDyes are water-soluble, transmembrane conjugated oligoelectrolytes (COEs). These lipophilic dyes fully embed themselves across the lipid bilayer of their target membranes, minimising "leakage" of dye from their targets. JotDyes are characterised by a high quantum yield (>25%). The significant increase in fluorescence is only observed when the dyes are bound to their target membranes, and this stark difference in emission profiles between bound and unbound dyes underlie the "light on" mechanism. Furthermore, JotDyes are highly water soluble and do not form micelles or nanoparticles when reconstituted in aqueous buffer, minimising the occurrence of false positives in flow cytometry analyses.

## Storage Conditions

JotDyes can be shipped at ambient temperature but should be stored at 2 to 8°C in sterile conditions and protected from light. In its dried form, these products are stable for at least 1 year when stored as directed. Solubilised dyes can be stored for up to 1 month at 2 to 8°C.

## Dye Reconstitution

Table 1. Preparation of JotDyes stock solution.

Product	Number of Tests	Solvent	Volume of Solvent to Add	Stock Concentration
JotDye-430	25 Tests (Trial Size)	Aqueous Buffer	25 µL	25 µM
JotDye-490				
JotDye-600	100 Tests		100 µL	
JotDye-800				

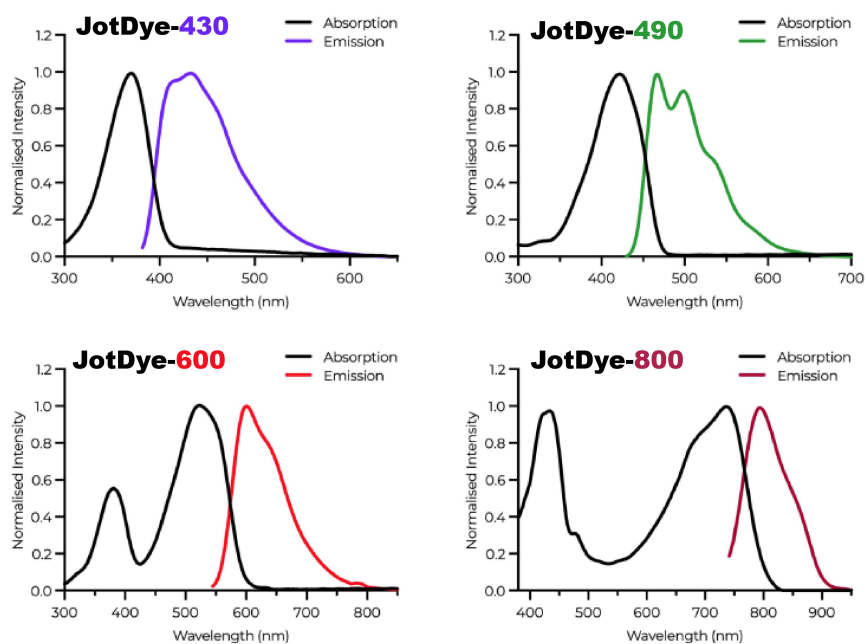
1. Allow all buffers and reagents to come to room temperature before use.
2. To get a 25 µM stock solution, reconstitute the dried dye in an aqueous buffer of your choice (e.g. sterile, filtered PBS). Refer to Table 1 above for the volume of solvent to add into the dye vial based on the product size (number of tests).
3. Vortex the dye for 1 minute to fully dissolve the solid, then do a quick spin to collect the dye at the bottom of the tube.
4. Incubate the dye for 15 minutes at RTP<sup>#</sup> to allow the dye to solubilise.
5. Repeat Step 3.
6. Dyes perform best when freshly reconstituted. Solubilised dyes can be stored for up to 1 month at 4°C. However, dyes should be allowed to equilibrate to RTP<sup>#</sup> before each use.

*<sup>#</sup>If a sonicator is available, sonicate the dye stock solution for 5 minutes at 40°C in pulse setting to ensure complete dissolution before each use.*

## Photophysical Properties

**Table 2. Photophysical properties of JotDye-430, JotDye-490, JotDye-600 and JotDye-800.**

Dye Name	JotDye-430	JotDye-490	JotDye-490	JotDye-800
Appearance	Colourless	Yellow	Purple	Green
$\lambda_{abs, with SUVs}$ (nm)	369	422	525	735
$\lambda_{em, with SUVs}$ (nm)	403 - 460	458 - 508	586 - 635	775 - 818
Suitable Laser(s)	UV (355 nm)	Violet (405 nm)	Blue or Yellow (488 or 561 nm)	Red (638 nm)
Quantum Yield (%)	94	60	27	13.8
Fluorescence Lifetime (ns)	1.1	0.9	1.9	1.6
IC <sub>50</sub> (μM)	>256	>256	170	>272



**Figure 1. JotDye-430, JotDye-490, JotDye-600 and JotDye-800 absorption and emission spectra.**

Please feel free to email [jotbody@jotbody.com](mailto:jotbody@jotbody.com) for assistance with troubleshooting of the staining protocol.