



Revised: December 1, 2022

## Product Information: **Fluorescent Phospholipids**

| Compound                                      | Storage information                                                | Shelf Life                                                                                 |
|-----------------------------------------------|--------------------------------------------------------------------|--------------------------------------------------------------------------------------------|
| Fluorescent phospholipid                      | Freeze upon receipt<br>< -20 °C<br>Desiccate<br>Protect from light | When stored as indicated,<br>fluorescent phospholipids are<br>stable for at least 3 years. |
| For optical properties see Table 1 on page 2. |                                                                    |                                                                                            |

### Introduction:

Phospholipids play a major role in cell structure and function. Due to their amphiphilic nature and the ability to form lipid bilayers phospholipids are the predominant building block of biological membranes such as plasma- and intracellular membranes etc. These biological barriers control the passage of a large variety of molecules, both between cells and extracellular space and between different compartments within the cells. Membranes are the turntables for crucial processes in neurobiology, muscle contraction, and cell signaling.

To study and investigate cell structures, processes like lipid metabolism, signal transduction, transmembrane diffusion, etc., lipophilic fluorescent probes are very useful tools. Natural phospholipids are generally very similar in structure. However, minor differences e.g. number and length of the fatty acid chains, degree of unsaturation of the fatty acid and nature of hydrophilic head group may result in significant variations of the physical properties and biological activity of such membranes.

### ATTO Fluorescent Phospholipids

**ATTO-TEC** offers a variety of phospholipids based on glycerol carrying one or two fatty acids (lipophilic groups) and a phosphate monoester residue (hydrophilic group). They are labeled at the hydrophilic head group. After incorporation of the fluorescent phospholipid the fluorophore is located at the water/lipid interface of the membrane. We currently provide **1,2-dipalmitoyl-*sn*-glycero-3-phosphoethanolamine** (DPPE), **1,2-dioleoyl-*sn*-glycero-3-phosphoethanolamine** (DOPE), **palmitoyl-*sn*-glycero-phosphoethanolamine** (PPE), **1,2-dimyristoyl-*sn*-glycero-3-phosphoethanolamine** (DMPE) and **1,2-dilauroyl-*sn*-glycero-3-phosphoethanolamine** (DLPE) labeled with **ATTO**-dyes (see Table 2).

### Storage and Handling:

Fluorescent phospholipid derivatives are supplied in solid form and should be stored at -20 °C, desiccated and protected from light. When stored as indicated, **ATTO**-dye labeled phospholipids are stable for at least three years.

For the preparation of stock solutions, we recommend using chloroform/methanol 80:20 as solvent of choice. The stock solution of labeled phospholipids should be stored in the same way as the solid, however the shelf life of such solutions might be significantly reduced.

**Table 1:** Properties of available **ATTO**-label:

| <b>Label</b> | $\lambda_{\text{abs}}$ ,<br>nm | $\epsilon_{\text{max}}$ ,<br>M <sup>-1</sup> cm <sup>-1</sup> | MW, g/mol<br>DPPE | MW, g/mol<br>DOPE | MW, g/mol<br>PPE | MW, g/mol<br>DMPE | MW, g/mol<br>DLPE |
|--------------|--------------------------------|---------------------------------------------------------------|-------------------|-------------------|------------------|-------------------|-------------------|
| ATTO 390     | 387                            | 24000                                                         | 1017              | 1069              |                  | 961               |                   |
| ATTO 425     | 433                            | 45000                                                         | 1075              | 1127              |                  |                   |                   |
| ATTO 430LS   | 441                            | 32000                                                         | 1285              | 1337              |                  |                   |                   |
| ATTO 465     | 467                            | 75000                                                         |                   | 1135              |                  |                   |                   |
| ATTO 490LS   | 498                            | 40000                                                         | 1348              | 1444              |                  |                   |                   |
| ATTO 495     | 503                            | 80000                                                         | 1025              |                   |                  |                   |                   |
| ATTO 488     | 508                            | 90000                                                         | 1264              | 1316              | 1025             | 1207              | 1150              |
| ATTO 520     | 513                            | 110000                                                        | 1040              | 1092              | 802              | 984               |                   |
| ATTO 532     | 543                            | 115000                                                        | 1320              | 1372              | 1081             | 1264              |                   |
| ATTO Rho6G   | 535                            | 115000                                                        |                   |                   |                  | 1232              |                   |
| ATTO 540Q    | 546                            | 120000                                                        |                   | 1285              |                  |                   |                   |
| ATTO 542     | 548                            | 120000                                                        | 1588              |                   |                  |                   |                   |
| ATTO 550     | 565                            | 120000                                                        | 1382              | 1420              | 1130             | 1312              |                   |
| ATTO 565     | 555                            | 120000                                                        | 1299              | 1209              | 596              | 1128              |                   |
| ATTO Thio12  | 579                            | 120000                                                        | 1276              |                   |                  |                   |                   |
| ATTO Rho101  | 584                            | 120000                                                        | 1364              |                   |                  |                   |                   |
| ATTO 590     | 595                            | 120000                                                        | 1379              | 1417              | 1127             | 1309              |                   |
| ATTO 594     | 606                            | 120000                                                        | 1480              | 1532              | 1241             | 1424              | 1368              |
| ATTO 620     | 621                            | 120000                                                        | 1186              |                   |                  |                   |                   |
| ATTO 633     | 630                            | 130000                                                        | 1326              | 1378              | 1088             | 1270              |                   |
| ATTO 643     | 648                            | 150000                                                        |                   | 1584              |                  |                   | 1420              |
| ATTO 647     | 653                            | 120000                                                        | 1267              | 1319              |                  | 1211              |                   |
| ATTO 647N    | 646                            | 150000                                                        | 1420              | 1485              | 1194             | 1364              | 1308              |
| ATTO 655     | 657                            | 125000                                                        | 1316              | 1368              | 964              | 1146              | 1203              |
| ATTO 665     | 665                            | 160000                                                        |                   | 1349              |                  |                   |                   |
| ATTO 680     | 678                            | 125000                                                        | 1314              | 1366              |                  |                   |                   |
| ATTO 700     | 694                            | 120000                                                        | 1353              | 1292              |                  |                   |                   |
| ATTO 740     | 745                            | 120000                                                        | 1142              | 1194              |                  |                   |                   |
| ATTO MB2     | 658                            | 100000                                                        | 1130              |                   |                  |                   |                   |

$\lambda_{\text{abs}}$ : longest-wavelength absorption maximum in chloroform:methanol 80:20 (V/V);  $\epsilon_{\text{max}}$ : molar extinction coefficient at the longest-wavelength absorption maximum.

## Application

Membrane incorporation of fluorescent lipid analogs can be performed as described in literature<sup>1</sup>. Generally, a complex of the fluorescent labeled phospholipid and Bovine Serum Albumin (BSA) is prepared, dried, preferably redissolved in ethanol and simply injected to cell containing aqueous medium. The densities of the labeled species in a plasma membrane varies with the concentration of the BSA-lipid-complex and conditions (incubation time and temperature).

For recent applications using **ATTO**-dye labeled phospholipids we refer to Literature<sup>2-6</sup>.

## References

1. Eggeling C, et al., *Direct observation of the nanoscale dynamics of membrane lipids in a living cell*, Nature **457** (2009) 1159–1163.
2. Honigsmann, A.; Walter C. et al., *Characterization of Horizontal Lipid Bilayers as a Model System to Study Lipid Phase Separation*, Biophysical Journal **98** (2010), 2886-2894
3. Vicidomini, G.; Ta, H. et al., *STED-FLCS: An Advanced Tool to Reveal Spatiotemporal Heterogeneity of Molecular Membrane Dynamics*, Nano letters **15** (2015), 5912-5918.
4. Cardoso Dos Santos, M.; Vézy, C.; Jaffiol, R., *Nanoscale characterization of vesicle adhesion by normalized total internal reflection fluorescence microscopy*, Biochimica et Biophysica Acta – Biomembranes **1858** (2016), 1244-1253.
5. Schmid, E. M.; Bakalar, M. H. et al., *Size-dependent protein segregation at membrane interfaces*, Nature Physics **12** (2016), 704-711.
6. Johnson, A.; Bao, P. et al., *Simple, Direct Routes to Polymer Brush Traps and Nanostructures for Studies of Diffusional Transport in Supported Lipid Bilayers*, Langmuir **33** (2017), 3672-3679.

**Table 2: ATTO-Labeled Phospholipids Order Information:**

| Dye        | Order Code                   |                              |                          |                          |                          |
|------------|------------------------------|------------------------------|--------------------------|--------------------------|--------------------------|
|            | DPPE 1 mg<br>5 mg            | DOPE 1 mg<br>5 mg            | PPE 1 mg<br>5 mg         | DMPE 1 mg<br>5 mg        | DLPE 1 mg<br>5 mg        |
| ATTO 390   | AD 390-151<br>AD 390-155     | AD 390-161<br>AD 390-165     |                          | AD 390-191<br>AD 390-195 |                          |
| ATTO 425   | AD 425-151<br>AD 425-155     | AD 425-161<br>AD 425-165     |                          |                          |                          |
| ATTO 430LS | AD 430LS-151<br>AD 430LS-155 | AD 430LS-161<br>AD 430LS-165 |                          |                          |                          |
| ATTO 465   |                              | AD 465-161<br>AD 465-165     |                          |                          |                          |
| ATTO 495   | AD 495-151<br>AD 495-155     |                              |                          |                          |                          |
| ATTO 488   | AD 488-151<br>AD 488-155     | AD 488-161<br>AD 488-165     | AD 488-181<br>AD 488-185 | AD 488-191<br>AD 488-195 | AD 488-241<br>AD 488-245 |
| ATTO 490LS | AD 490LS-151<br>AD 490LS-155 | AD 490LS-161<br>AD 490LS-165 |                          |                          |                          |
| ATTO 520   | AD 520-151<br>AD 520-155     | AD 520-161<br>AD 520-165     | AD 520-181<br>AD 520-185 | AD 520-191<br>AD 520-195 |                          |
| ATTO 532   | AD 532-151<br>AD 532-155     | AD 532-161<br>AD 532-165     | AD 532-181<br>AD 532-185 | AD 532-191<br>AD 532-195 |                          |

| Order Code  |                                |                   |                              |                   |                              |  |                            |  |                            |  |
|-------------|--------------------------------|-------------------|------------------------------|-------------------|------------------------------|--|----------------------------|--|----------------------------|--|
| Dye         | DPPE 1 mg<br>5 mg              | DOPE 1 mg<br>5 mg | PPE 1 mg<br>5 mg             | DMPE 1 mg<br>5 mg | DLPE 1 mg<br>5 mg            |  |                            |  |                            |  |
| ATTO Rho6G  |                                |                   |                              |                   | AD Rho6G-191<br>AD Rho6G-195 |  |                            |  |                            |  |
| ATTO 540Q   |                                |                   | AD 540Q -161<br>AD 540O -165 |                   |                              |  |                            |  |                            |  |
| ATTO 542    | AD 542-151<br>AD 542-155       |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO 550    | AD 550-151<br>AD 550-155       |                   | AD 550-161<br>AD 550-165     |                   | AD 550-181<br>AD 550-185     |  | AD 550-191<br>AD 550-195   |  |                            |  |
| ATTO 565    | AD 565-151<br>AD 565-155       |                   | AD 565-161<br>AD 565-165     |                   | AD 565-181<br>AD 565-185     |  | AD 565-191<br>AD 565-195   |  |                            |  |
| ATTO Thio12 | AD Thio21-151<br>AD Thio12-155 |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO Rho101 | AD Rho101-151<br>AD Rho101-155 |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO 590    | AD 590-151<br>AD 590-155       |                   | AD 590-161<br>AD 590-165     |                   | AD 590-181<br>AD 590-185     |  | AD 590-191<br>AD 590-195   |  |                            |  |
| ATTO 594    | AD 594-151<br>AD 594-155       |                   | AD 594-161<br>AD 594-165     |                   | AD 594-181<br>AD 594-185     |  | AD 594-191<br>AD 594-195   |  | AD 594-241<br>AD 594-245   |  |
| ATTO 620    | AD 620-151<br>AD 620-155       |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO 633    | AD 633-151<br>AD 633-155       |                   | AD 633-161<br>AD 633-165     |                   | AD 633-181<br>AD 633-185     |  | AD 633-191<br>AD 633-195   |  |                            |  |
| ATTO 643    |                                |                   | AD 643-161<br>AD 643-165     |                   |                              |  |                            |  | AD 643-241<br>AD 643-245   |  |
| ATTO 647    | AD 647-151<br>AD 647-155       |                   | AD 647-161<br>AD 647-165     |                   |                              |  | AD 647-191<br>AD 647-195   |  |                            |  |
| ATTO 647N   | AD 647N-151<br>AD 647N-155     |                   | AD 647N-161<br>AD 647N-165   |                   | AD 647N-181<br>AD 647N-185   |  | AD 647N-191<br>AD 647N-195 |  | AD 647N-241<br>AD 647N-245 |  |
| ATTO 655    | AD 655-151<br>AD 655-155       |                   | AD 655-161<br>AD 655-165     |                   | AD 655-181<br>AD 655-185     |  | AD 655-191<br>AD 655-195   |  | AD 655-241<br>AD 655-245   |  |
| ATTO 665    |                                |                   | AD 665-161<br>AD 665-165     |                   |                              |  |                            |  |                            |  |
| ATTO 680    | AD 680-151<br>AD 680-155       |                   | AD 680-161<br>AD 680-165     |                   |                              |  |                            |  |                            |  |
| ATTO 700    | AD 700-151<br>AD 700-155       |                   | AD 700-161<br>AD 700-165     |                   |                              |  |                            |  |                            |  |
| ATTO 740    | AD 740-151<br>AD 740-155       |                   | AD 740-161<br>AD 740-165     |                   |                              |  | AD 740-191<br>AD 740-195   |  |                            |  |
| ATTO MB2    | AD MB2-151<br>AD MB2-155       |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO 565    | AD 565-151<br>AD 565-155       |                   | AD 565-161<br>AD 565-165     |                   | AD 565-181<br>AD 565-185     |  | AD 565-191<br>AD 565-195   |  |                            |  |
| ATTO Thio12 | AD Thio21-151<br>AD Thio12-155 |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO Rho101 | AD Rho101-151<br>AD Rho101-155 |                   |                              |                   |                              |  |                            |  |                            |  |
| ATTO 590    | AD 590-151<br>AD 590-155       |                   | AD 590-161<br>AD 590-165     |                   | AD 590-181<br>AD 590-185     |  | AD 590-191<br>AD 590-195   |  |                            |  |

| Order Code |                            |                            |                            |                            |                            |
|------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|
| Dye        | DPPE 1 mg<br>5 mg          | DOPE 1 mg<br>5 mg          | PPE 1 mg<br>5 mg           | DMPE 1 mg<br>5 mg          | DLPE 1 mg<br>5 mg          |
| ATTO 594   | AD 594-151<br>AD 594-155   | AD 594-161<br>AD 594-165   | AD 594-181<br>AD 594-185   | AD 594-191<br>AD 594-195   | AD 594-241<br>AD 594-245   |
| ATTO 620   | AD 620-151<br>AD 620-155   |                            |                            |                            |                            |
| ATTO 633   | AD 633-151<br>AD 633-155   | AD 633-161<br>AD 633-165   | AD 633-181<br>AD 633-185   | AD 633-191<br>AD 633-195   |                            |
| ATTO 643   |                            | AD 643-161<br>AD 643-165   |                            |                            | AD 643-241<br>AD 643-245   |
| ATTO 647   | AD 647-151<br>AD 647-155   | AD 647-161<br>AD 647-165   |                            | AD 647-191<br>AD 647-195   |                            |
| ATTO 647N  | AD 647N-151<br>AD 647N-155 | AD 647N-161<br>AD 647N-165 | AD 647N-181<br>AD 647N-185 | AD 647N-191<br>AD 647N-195 | AD 647N-241<br>AD 647N-245 |
| ATTO 655   | AD 655-151<br>AD 655-155   | AD 655-161<br>AD 655-165   | AD 655-181<br>AD 655-185   | AD 655-191<br>AD 655-195   | AD 655-241<br>AD 655-245   |
| ATTO 665   |                            | AD 665-161<br>AD 665-165   |                            |                            |                            |
| ATTO 680   | AD 680-151<br>AD 680-155   | AD 680-161<br>AD 680-165   |                            |                            |                            |
| ATTO 700   | AD 700-151<br>AD 700-155   | AD 700-161<br>AD 700-165   |                            |                            |                            |
| ATTO 740   | AD 740-151<br>AD 740-155   | AD 740-161<br>AD 740-165   |                            | AD 740-191<br>AD 740-195   |                            |
| ATTO MB2   | AD MB2-151<br>AD MB2-155   |                            |                            |                            |                            |

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