

## **New Procedure for Transformation**

If your school is close to one of our partners, you might be receiving competent cells in dry ice even if you are running the pARA-R sequence. The transformation efficiency of these cells is extremely high, far better than scraping colonies off an LB plate. However, it's important that these cells remain frozen until transformation day. Although you will receive the cells on dry ice, you'll need to place the entire styrofoam chest, including its ice pack, into your freezer. The styrofoam chest will act to buffer changes in freezer temperatures.

We have found that schools will get excellent transformations if we deliver frozen, competent cells to you the day before you do transformation. Even though your students are working with pARA-R, we suggest that you follow the directions for **Lab 5**. The only difference will be the plasmid used for transformation. You will be using pARA-R not the ligated plasmid.

We've included LB broth with your refrigerated items. You will *not* be using CaCl<sub>2</sub> or picking your cells from a plate streaked with LMG cells.

On the day of transformation, take out the number of tubes your class will need for transformation- each group will need 100 µL of cells; each tube contains approximately 500 µL of cells. Place these tubes in wet ice to defrost. Cells will defrost in 10-15 minutes. Be certain to resuspend the cells before aliquoting. This is accomplished by gently pumping in and out with the P-200 pipette. After aliquoting the cells, place the aliquots in wet ice. For efficient transformation, it is important the cells remain cold until the heat shock step.

Please refer to the Teacher Guide for additional information. Don't hesitate to call or e-mail us with any questions.