

Source Plates:

The Foundation to a Successful Transformation!

Over the years, we have improved our protocols to provide consistent results. We have found that a well-streaked source plate is one of the most important parts in guaranteeing a successful transformation!

What is a Source Plate?

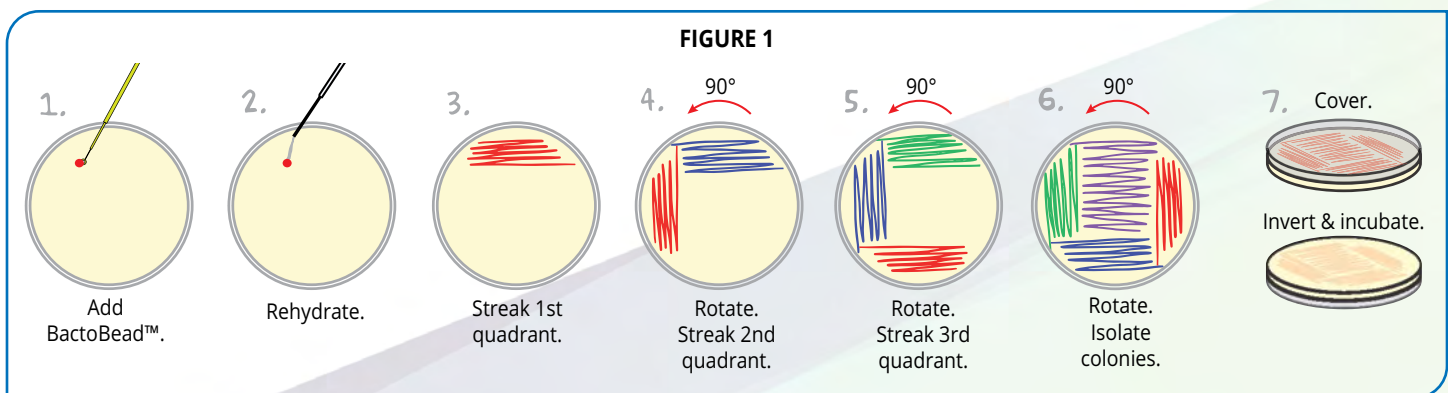
A source plate is a standard LB Agar plate that is used to grow the initial colonies of bacteria that will be used for the transformation process. Source plates allow scientists to observe the growth of the bacteria to make sure it is growing well and there is no contamination. Because of the way source plates are streaked, students are able to select well-isolated colonies to get the proper amount of bacteria for their transformation!

We have found that when the source plates are older than 24 hours, and when clumps of bacteria are used rather than individual colonies, transformation is far less likely to succeed. Always pick the well-isolated colonies on your plate that are 1-1.5 mm in diameter.

Source Plates Should Be:

- Prepared the day before the lab and incubated overnight at 37°C.
- Used as soon as possible after the incubation period (18-22 hours) has finished.
- Streaked properly to ensure the growth of well-isolated colonies (see Figure 1, below).

Streaking Source Plates:



1. Add a fresh BactoBead™ to an LB Agar plate.
2. Rehydrate the BactoBead™ with 10 μ L of recovery broth.
3. Start a dense patch of bacteria in the first quadrant by spreading the BactoBead™ back in forth.
4. Rotate the plate 90° and drag the loop through the bacteria in the first quadrant. Streak back and forth making sure not to dip back into the previous quadrant.
5. Repeat this on the 3rd quadrant.
6. Drag the loop across the rest of the empty space on the agar to isolate colonies.
7. Cover and invert the plates, then place them in a 37°C incubator overnight.



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Tips for making source plates:

- You can practice this technique by using a marker on the base of the plate and mimicking the steps in Figure 1.
- Your students can practice this too! Pass out pieces of paper or white boards and have your students master streaking.
- Ensure the agar is fresh and not dried out. Avoid puncturing the plate with the inoculation loop when streaking the bacteria.
- Incubate the plate inverted at 37°C overnight.
- Make sure that the BactoBead™ has been stored correctly: Keep the vial capped tightly and in the refrigerator, as exposure to air and moisture will ruin the BactoBead™.
- Make sure the BactoBead™ is rehydrated properly with 10 µL recovery broth.

Watch our instructional video here:



All our instructional videos can be found at [youtube.com/edvotekinc](https://www.youtube.com/edvotekinc)

After the incubation:

After the overnight incubation has finished it is important to check on the status of your source plates. The source plate should have isolated colonies that are about 1-1.5 mm in diameter, all the same color, and the agar should not be dried out or hard.

Picking colonies:

When it is time to perform the transformation, students will want to pick up between 5-10 well-isolated colonies for the experiment. To do this, provide the students with sterile inoculation loops or toothpicks and gently swipe the colonies to get them adhered to the loop or toothpick. Then these colonies will be resuspended in ice cold calcium chloride to make them competent! Make sure your students are gentle, and do not pierce the agar with the loop or toothpick.