

Need to separate into different protocols

ScienceBridge Tech Site Standard Operating Procedure

|                                              |                           |                                     |
|----------------------------------------------|---------------------------|-------------------------------------|
| Title: Bacterial Transformation Kit Assembly |                           |                                     |
| Original Issue Date: 8/26/10                 | Revision Date: 10-12-2019 | Pages: 3                            |
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|           |                                                                                                                                      |       |                   |                 |
|-----------|--------------------------------------------------------------------------------------------------------------------------------------|-------|-------------------|-----------------|
| Scope     | For use by biotechnology students at the ScienceBridge tech sites when packaging the ScienceBridge Transformation kits for shipment. |       |                   |                 |
| Objective | This SOP sets the procedural specifications for packaging the Transformation kits.                                                   |       |                   |                 |
| Materials |                                                                                                                                      | 1 kit | Packaged Quantity | Final packaging |
|           | <u>Assembled Protein Purification Kit bag</u>                                                                                        | 1     | 1                 | 1               |
|           | 1. Room temp bag (gallon-sized)                                                                                                      | 1     | 1                 | 1               |
|           | <i>a. Microcentrifuge tubes, 2.0mL, round bottom, clear</i>                                                                          | 30    | 30                |                 |
|           | <i>b. Transfer pipets, (bags of 40)</i>                                                                                              | 40    | 1 bag             |                 |
|           | <i>c. Blue transfer loops (bags of 10)</i>                                                                                           | 10    | 1 bag             |                 |
|           | 2. Wet Bag                                                                                                                           | 1     | 1                 | 1               |
|           | <i>a. Blue 1.5mL microcentrifuge tubes labeled CaCl<sub>2</sub>, containing 1.2mL CaCl<sub>2</sub></i>                               | 10    | 10                |                 |
|           | <i>b. Orange 1.5mL microcentrifuge tubes labeled TE, containing 1mL TE buffer</i>                                                    | 1     | 1                 |                 |
|           | 3. Sterile Bag                                                                                                                       | 1     | 1                 | 1               |
|           | <i>a. Cotton swabs</i>                                                                                                               | 20    | 20                |                 |
|           | <i>b. p200 Pipet tips</i>                                                                                                            | 10    | 10                |                 |
|           | 4. Bacterial stab, inoculated* (Provide one stab <b>per teacher</b> <u>not</u> per kit)                                              | 1*    | 1*                | 1*              |
|           | 5. Transformation Kit Checklist                                                                                                      | 1     | 1                 | 1               |
|           | <u>Media</u>                                                                                                                         |       |                   |                 |
|           | 1. 5cm LB only agar plates                                                                                                           | 10    | ½ Sleeve          | ½ Sleeve        |
|           | 2. 5cm LB only agar plates* (per teacher, not per kit)                                                                               | 10*   | ½ Sleeve*         | ½ Sleeve*       |
|           | 3. 5cm LB/amp agar plates                                                                                                            | 20    | 1 Sleeve          | 1 Sleeve        |
|           | <u>Cold Bags</u>                                                                                                                     | 1     | 1                 | 1*              |

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|-------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------|-----------|-----------|
|                   | <ol style="list-style-type: none"> <li>Green 0.6mL microcentrifuge tube labeled <b>PM1</b>, containing 15uL plasmid</li> <li>Purple 0.6mL microcentrifuge tube labeled <b>PM2</b>, containing 15uL plasmid</li> </ol>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 | 1         | 1         |           |
| Equipment         | <ol style="list-style-type: none"> <li>Sharpie</li> <li>Clear tape</li> </ol>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         | 1<br>2 in | 1<br>2 in | 1<br>2 in |
| Supplemental Aids | <ul style="list-style-type: none"> <li>list each video separately- (hyperlink the address to their title)</li> <li>list any worksheets or other documents needed to complete the job</li> </ul>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                       |           |           |           |
| Safety            | Take care to ensure that liquid-filled tubes do not open during handling.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                             |           |           |           |
| Quantity          | <p>*items are 1 per teacher (NOT per kit)</p> <p>Prepare 1 room temperature kit for each order</p> <p>Prepare 1 cold bag for each teacher (all orders into 1 bag)</p> <p>Prepare media for each order (if ordered multiple kits- combine 2 half sleeves to make 1 full sleeve as appropriate)</p>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |           |           |           |
| Protocol          | <p><b>Part I: Kit Bag Preassembly (Prior to Pick Up Date)</b></p> <ol style="list-style-type: none"> <li>Fill out and affix 1 teacher bag delivery sticker as well as other labels to the top right corner of the bag for each kit ordered.</li> <li>Tape one printed Bacterial Transformation Kit Checklist on the same side of the bag as the labels.               <ol style="list-style-type: none"> <li>If a teacher orders multiple kits, write on the checklists of bags 2-5 the bag number in which the bacterial stab will be located, since each teacher is only supposed to have one bacterial stab.</li> </ol> </li> </ol> <p><u>For the next three steps, it would be better to put in one item bag type into all the kits being assembled instead putting in all three bags in one kit at a time. Also, when you sign off on the checklist, you are only making sure the items are present. Do not spend time counting the items because that is what Quality Assurance does.</u></p> <ol style="list-style-type: none"> <li>Add one room temp bag to each kit, sign off on checklist.</li> <li>Add one sterile bag to each kit, sign off on checklist.</li> <li>Add one wet bag to each kit, sign off on checklist.</li> <li><b>QC: Use the checklist to make sure that all room temperature contents, wet bag contents, and sterile bag contents are present and tubes are completely sealed</b></li> <li>Immediately send bags to Quality Assurance.</li> </ol> <p><b>Part II: Kit Preassembly of Temperature Sensitive Supplies</b></p> <ol style="list-style-type: none"> <li>Fill out 1 teacher bag delivery sticker and affix to a sandwich bag.</li> <li>Before starting the packaging process make sure you keep the pm1 and pm2 tubes in boxes of ice to make sure they don't defrost. The pm1 and pm2 tubes should be placed in ice before and after they are put into the sandwich bag.</li> <li>Package 1 green <b>PM1</b> tube and 1 purple <b>PM2</b> tube per kit ordered into one of the labeled sandwich bags. Make sure to work quickly to keep the tubes from defrosting.</li> <li><b>QC: Make sure that the PM1 and PM2 tubes are sealed and do not defrost. Recount the number of purple and green</b></li> </ol> |           |           |           |

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|               | <p><b>tubes and make sure that for each color it is the same as the # ordered on the delivery sticker.</b></p> <p><b>Part III: Kit Bag Assembly the Week of Pick Up Date</b></p> <p><b>12.</b>Open up the bacterial stab spreadsheet to see which bacterial stabs can be used. Here are a list of criteria for how to choose the bacterial stabs:</p> <ol style="list-style-type: none"> <li>Bacterial stabs must be QC'd at most 2 months ago.</li> <li>Out of these stabs, they must have the "Active" status.</li> <li>Out of those stabs, choose the oldest stab (these will have the smallest number), still making sure it was QC'd at most 2 months ago.</li> </ol> <p><b>13.</b>Provide one bacterial stab <b>per teacher</b> (<del>not</del> per kit). Place in the bag 1 of #. You need to get these bags back from Quality Assurance as they will not be in your possession right now.</p> <p><b>14.</b>Send an email to senior managers to tell them which stabs have been used, including numbers. Senior managers will then change the bacterial stab spreadsheet.</p> <p>(Part IV is packaged by TechSite 2/ Senior Managers. This will include plates. The LB sterile plates and the LB streaked plates will be packaged together.)</p> <p><b>Part IV: Kit Assembly on Pick Up Date (Box 1/X)</b></p> <p><b>15.</b>Add twenty 5cm LB only agar plates and twenty 5cm LB/amp agar plates into each bag for each kit ordered.</p> <p><b>16.</b>The pm1 and pm2 tubes will be given to the teachers separately, most likely in an ice box. Make sure they are still cold.</p> <p><b>17.</b>Place completed kits in alphabetical order and grouped by teacher (and bag order #) in delivery pickup area.</p> <p><b>QC: Once all kits are finished make sure that all the bags for each order are present and have been fully checked off on their check off sheet.</b></p> |
| Documentation | <p><b>All work and any variance from the protocol must be documented</b><br/>         (who do you need to inform &amp; what information, include what document/tracking log to fill out)</p>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |
| Storage       | <p><b>All materials should be stored in the (kit type) cabinet.</b></p> <p><b>Always start with the oldest dates on supplies and complete partially finished or open bags first.</b></p> <p><b>Product Storage</b></p> <ul style="list-style-type: none"> <li>(Where do completed parts/products go,</li> <li>where do unfinished parts/products go and how to label)</li> </ul> <p><b>Supplies Storage Locations</b></p> <ul style="list-style-type: none"> <li>(Where are the supplies stored?)</li> </ul>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |

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|--------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------|
| Quality Control                | Use the Transformation Kit Checklist as a guide when packing the boxes. Include the checklist in each box, and send packed boxes to QC immediately. |
| When                           | (How often does this job need to be repeated and/or when should it be done)                                                                         |
| Tech Site Kit:<br><i>Group</i> | Bacterial Transformation : <i>Order Fulfilment</i>                                                                                                  |