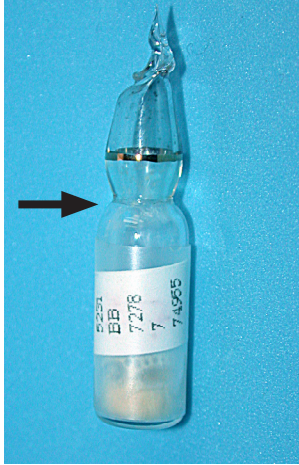


# Recovery of Lyophilized Cultures

Your shipment includes one or more glass, pre-scored and flame-sealed ampules or aluminum and rubber-capped serum bottles holding freeze-dried fungal isolates that must be rehydrated with sterile water before inoculating a suitable culture medium such as that indicated on the ARSEF packing slip. Rehydrate each isolate by the following steps:

## FROM PRE-SCORED AMPULES:



1. Disinfect the ampule by wiping with 70% ethanol.
2. Wrap the scored area (arrow at the narrow neck below the gold-colored band) with the ethanol-dampened tissue (Kimwipe, etc.) to protect your fingers. The tissue should not be so wet that alcohol enters the ampule.
3. Bend and break the ampule at the narrow, pre-scored area (arrow). The alcohol-dampened tissue provides good cushioning and protection against cuts for this step.
4. Aseptically add 0.2–0.5 ml of sterile water.
5. Using a sterile pipette, gently aspirate the contents several times to mix the suspension thoroughly.
6. Let the suspension to rest for 15-30 minutes.
7. Inoculate the suspension onto an appropriate medium, and incubate.

## FROM SERUM BOTTLES:



1. Some bottles have aluminum rings through which the rubber cap is pre-exposed. If an aluminum cap covers the rubber seal (as shown here), either remove the cap or bend back the pre-cut disk to expose the diaphragm of the rubber seal.
2. Disinfect the bottle's upper part and exposed rubber seal by wiping with 70% ethanol.
3. Add 0.5 ml of sterile water either with a sterile syringe to inject water through the seal's diaphragm (leave the syringe in place during rehydration) **OR** remove the rubber seal before adding sterile water by pipette.
4. Gently agitate or aspirate the contents several times to mix the suspension thoroughly.
5. Let the suspension to rest for 15-30 minutes.
6. Use the syringe or a sterile pipette to inoculate the suspension onto an appropriate medium, and incubate.

The labels on our lyophilized cultures have changed over time; we try to change these older labels before shipment for the sake of uniformity and easier interpretation by the receiving laboratory. The labels on units you receive should have the ARSEF accession number of the isolate in the top line and the fungus key (the isolate's abbreviated identification) in the second line. All numbers on any lower lines are locator information generated by the collection's database management software and are not important for you as the end user *unless* you need to communicate with us directly about any apparent problem the fungus you have recovered from the vial.

Lyophilized units can be stored indefinitely at 4 °C in a refrigerator before opening them, and *should* be refrigerated if they are not likely to be opened within a few days after their receipt. There is no need to revive such material immediately upon receipt. These units were maintained in a refrigerator before being sent from ARSEF, and should not suffer being exposed to warmer temperatures during shipment. Most freeze-dried cultures should show some initial signs of growth within 24-48 hours after rehydration and plating.