

## **<sup>32</sup>Si Sample Processing - HOT Cruises**

### **At Sea: (NEW)**

1. Rinse a 250ml round PC bottle three times with sample water then fill to brim. Place bottle in bottle carrier and keep covered with dark plastic bag to protect from light.
2. Take samples to rad van. Rinse pipette tips 1x with 1N HCl and 3x with Nanopure water to clean it. Separate the P (planchette) bottles from the C (counter) bottles. Find the PK (planchette kinetics) bottle and inject 1000 $\mu$ l of 5mM sodium metasilicate solution into the bottle, cap and invert several times. Reset the pipetter for 300 $\mu$ l. Using the 10-100 $\mu$ l pipetter, clean a pipette tip and draw/expel <sup>32</sup>Si stock several times to wet tip with isotope (isotope sticks to plastic) then inject 100 $\mu$ l of 0.1 $\mu$ Ci/ml <sup>32</sup>Si stock into all the P bottles (**300 $\mu$ l into the PK bottle only**), cap, invert several times to mix and record the time of injection. Using the 100-1000 $\mu$ l pipetter, inject 300 $\mu$ l of 0.1 $\mu$ Ci/ml <sup>32</sup>Si stock into all the C bottles, cap, invert several times to mix and record the time of injection.
3. Place injected samples back in bottle carrier and cover with dark plastic. Take carrier out to deck so bottles can be hung on PP array line. For deep bottles (150m and 175m), place in dive bag and tie bag to PP array line with zip ties – be sure to zip tie the dive bag shut as well.
4. Incubate for 24hours (gas array) or dawn-dusk (production array).
5. Prior to filtration, rinse all filtration towers in the Nalgene bath (cubic container) with dilute HCl (~10% or less) in order to keep background <sup>32</sup>Si activity low. Filtration towers can be left inverted in the acid bath for a few hours. After acid rinse, wash towers with Nanopure to remove HCl, by dipping into a series of plastic beakers (filled with Nanopure). After rinsed, replace the towers in the manifold. If any significant amount of time will pass between rinsing the towers and filtering the samples (e.g. >1 hour), cover all rinsed towers with a plastic Ziploc bag in order to keep towers clean.
6. Collect bottles from PP array – keep in bottle carrier covered with dark plastic.
7. Filter each sample through a **0.6 $\mu$ m DTTP 25mm filter (Millipore Brand)**. If DTTP filters are exhausted use 0.6 $\mu$ m PCTE 25mm filters (Whatman brand); however, the 250ml towers will require a backing filter because the Whatman PC filters are too thin to make a seal (Millipore filters require NO backing filter). Use one of the 25mm paper filters as a gasket, punch at least 3 holes in the centre leaving a full rim of paper for the tower to seal to the base. Rinse the sample bottle with 3 small FSW rinses using a squirt bottle.
8. Prior to sample running dry, rinse inside of the incubation bottle with an FSW, vigorously shake and pour FSW rinse into the corresponding filtration tower (i.e. this is to rinse any diatoms sticking to the interior of the bottle. Use enough volume to allow bottle interior to be coated by FSW upon shaking, a 2-3 second squeeze of the FSW-squirt bottle should be sufficient. Do this rinse, shake, filter 2 or 3 times. FSW has been filtered through a 0.2 $\mu$ m filter.
9. When the sample goes dry, rinse the tower with 3 small FSW rinses using a squirt bottle.
10. **FOR LSC FILTERS:** Place filter in a pre-labeled 20ml plastic scintillation vial, particle side up, flat on the bottom. Vial label (write only on the cap, not on the vial) should be clear and easily readable, label as follows: Cruise #, Cast ID, Bottle ID (e.g. 199 S2C2 1C). Place cap in vial flat and place the open vial on top of the cap. Let the filters air dry for 24 hours before recapping.
11. **FOR BETA COUNTER FILTERS:** Place filter on a pre-labeled planchette (remove the locking ring from the planchette), particle side up, flat and centered on the disc. The underside of the planchette and the lid of the small petrie dish should both be labelled with Cruise #, Cast ID, Bottle ID (e.g. 199 S2C9 7P). Let the filters air dry for 24 hours. Centre the planchette in the small petrie dish and lay a circle of mylar over it. Place the locking ring over your finger and place that finger on the centre of the planchette to hold the mylar in place. Using the other hand, push the locking ring down over the mylar, lifting your finger off the mylar as you push the ring down so that the mylar will stretch across the planchette forming a smooth cover over the filter. Place the lid on the petrie dish and stack the petrie dishes into a “burrito” which you can then wrap with foil to keep together.
12. Empty the filtration waste reservoir into the rad waste container provided by UH rad people. While the UH waste container is open, rinse each incubation bottle three times with Nanopure, disposing of

rinse water directly in rad waste container. Use the squirt bottle to rinse the towers down with Nano after filters have been removed.

13. After the cruise, the rad bottles will need a rinse with 10% HCl (to prevent anything from growing in them while they sit around until the next cruise), followed by 3 Nano rinses. The bottles can be left in the partitioned grey box to dry if there is clean bench coat on the bottom of the box.

14. **SAMPLE SHIPPING:** The radiation level of the samples does not require external rad/hazardous labelling (less than 2.5 $\mu$ Ci) so they can be put in the FedEx box and sent to UCSB directly.