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| Standard Operating Procedure for:**Use of anaerobic chamber** | **PPE required:** |
| The anaerobic chamber is a heavy duty, flexible PVC chamber with airlock access designed to minimise oxygen ingress. However, each time the airlock is opened after cycling approximately 200-400 ppm oxygen is introduced into the chamber**. For this reason please try to minimise opening.** Remove any rings or watches that might puncture the chamber gloves. If working with sharp objects (only blunt scissors or syringes allowed) please wear a pair of cotton gloves to avoid punctures. |
| **Operation of chamber**1. Open nitrogen and N2/H2 mix cylinders until pressure is up to mark on regulator (50 psi) NO HIGHER. **(MUST HAVE BASIC GAS CYLINDER TRAINING)**
2. **Wearing nitrile gloves** open front door and place items inside airlock.
3. Containers being transferred into chamber should be open when placed in the airlock to reduce oxygen transfer into chamber. Consult a technician if this is not possible. If you are transferring liquids they must have been degassed. Containers of powder should be covered with parafilm with small vent holes.
4. Securely close door, turn on airlock and press the cycle ‘start’ button. **Run cycle twice**. Wait until the second cycle is complete and chamber reads ‘anaerobic’.
5. **Wearing nitrile gloves** place hands into glove port neoprene gloves, open internal door and transfer items into chamber. Securely close door.
6. To remove items: make sure airlock is anaerobic, open internal door, place items into airlock, securely close internal door, open external door and remove items, then securely close external door. **NEVER HAVE MORE THAN ONE DOOR OPEN AT THE SAME TIME.**
7. If you have finished: then turn off gas cylinders and empty regulators, turn off the air lock, put airlock under vacuum (15-20 inch. Hg).

**Carrying out work inside the chamber**1. Check monitor to confirm oxygen level and hydrogen level are appropriate for your work.
2. **Check gloves for pin holes.** Make sure you keep the chamber and the gloves clean to avoid cross-contamination. Wear an extra pair of gloves on top of the neoprene gloves if necessary.
3. Remove all waste and unnecessary equipment at the end of your work session.

**Storing samples in chamber**1. The lid of every bottles/containers stored inside the chamber should be properly closed, especially for liquids and acids. To avoid corrosion do not store large amounts of concentrated acids in the chamber.
2. Proper labelling on everything (name of user, content, hazards, date). Non-labelled stuff will be removed and/or disposed without asking.
 | **Hazard symbols:** Compressed gashttp://www.hse.gov.uk/chemical-classification/images/pictogram-gallery/irritant.gif |
| **Significant hazards:**Compressed gas cylinders  |
| Hazard phrases:**H280, H332** |
| **Can it be done out of hours?**Anaerobic chamber can be used out of hours only if user’s assessment of specific experimental risk allows it. |
| **This SOP is not relevant in the following circumstances:**1. SOP does not cover specific experimental risk these must be covered by user’s assessments
2. Any other situation where the procedure may result in harm to yourself or others.
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| Standard Operating Procedure for:**Maintenance of the anaerobic chamber** | **PPE required:** |
| There is a min. 0.5-2 ppm/min diffusion of oxygen through chamber surfaces. To keep the O2 level below detection limit (20 ppm) reusable palladium-coated alumina pellets-containing stainless steel mesh frames are mounted in front of recirculating fans. The palladium acts as a catalytic surface for the reduction of oxygen using the H2 from the chamber gas, and the alumina support absorbs the water formed by the reaction. Additionally, a bowl of silica crystals is placed into the chamber to keep humidity low (< 20 %). |
| **Maintaining atmosphere inside chamber**To be carried out when hydrogen level falls below 2.0%, usually weekly depending on use.1. Open nitrogen and N2/H2 mix cylinders until pressure is up to mark (50 psi) NO HIGHER. **(MUST HAVE BASIC GAS CYLINDER TRAINING)**
2. Make sure airlock is anaerobic, open internal door. **NEVER HAVE MORE THAN ONE DOOR OPEN AT THE SAME TIME.**
3. Depress vacuum switch at back and push on sides of chamber until chamber becomes deflated.
4. Depress “Mix” switch at back to add nitrogen/hydrogen mix to chamber.
5. Repeat this process until hydrogen content is around 4.0%.
6. When finished close the internal door, depress the vacuum switch at back until airlock is under vacuum (15-20 in. Hg). Then turn off gas cylinders.
7. Record in log book.

**Drying catalyst and silica gel**To be carried out weekly or as closely as possible:1. Remove four (of the six) catalyst plates and the two bowls of silica gel. **Ensure you follow SOP for using the anaerobic chamber**. **NEVER HAVE MORE THAN ONE DOOR OPEN.**
2. Place catalyst plates and bowls of silica gel in an oven at 200°C overnight. Silica gel is dark orange/reddish when recharged and dry, and turns pale orange as it takes up water vapour. **WEAR HEAT PROOF GLOVES.**
3. Once they are cool enough to handle, remove catalyst plates and bowl of silica gel from oven and return to chamber. **WEAR HEAT PROOF GLOVES.**
4. Record in log book.

**Monthly clean**1. Wipe the rubber doorframes and the glass clean using isopropanol and re-cover the rubber with a thin layer of silicon vacuum grease once a month,
2. Record in log book.
 | **Hazard symbols:**Compressed gashttp://www.hse.gov.uk/chemical-classification/images/pictogram-gallery/irritant.gif |
| **Significant hazards:**Gas cylindersHigh temperatures |
| Hazard phrases (H):**H280, H316, H332** |
| **Can it be done out of hours?****Maintenance should not be performed out of hours** |
| **This SOP is not relevant in the following circumstances:**1. Any other situation where the procedure may result in harm to yourself or others.
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