Standard Operating Procedure

SOP190

Title: Use of Heraeus Megafuge 40 benchtop refrigerated centrifuge

Location: CBE Laboratory Unit H34

1. PURPOSE

The intent of this SOP is to describe the procedures for the safe operation of the Heraeus Megafuge 40 centrifuge.

2. SCOPE

This SOP applies to the Containment Level 2 Laboratory Unit of the CBE and its personnel, specifically users of H34.

SPECIAL NOTES: HEALTH & SAFETY

Hazards associated with the use of centrifuges

- Mechanical failure of rotating parts (often violent).
- Contact with rotating parts.
- Sample leaks causing aerosols, stress corrosion, contamination.
- Sample imbalance causing machine movement/walking.
- Fire/explosion.
- Contact with contaminated components/vapours.

Safe Working Practices (also refer to Safety Note in the Operator Manuals for the Centrifuge you will be using)

Users MUST read the safety notes and instructions, detailed in the Operators Manual for the safe use of these centrifuges.

- 1. Ensure that the centrifuge is placed on a horizontal level surface and there is a clearance of at least 30 cm around the centrifuge when in operation. Make sure that hazardous materials are not stored or placed within this area and there is sufficient ventilation.
- 2. Always observe the start-up checks detailed in this SOP before using the centrifuge
- Do not exceed the specified maximum speed or load for the rotor used. NOTE: Each time a centrifuging run is started, the centrifuge recognizes the rotor code of the installed rotor with the help of a sensor. This means that the nominal speed of the rotor cannot be exceeded.

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- 4. Do not overfill containers. Remember that centrifugal forces drive the fluid up the outside tube wall for tubes used in fixed angle rotors. Use only the correct designated tubes for the rotor. Do not modify them to make them fit unless using attachments authorized by the manufacturer. NEVER USE GLASS CONTAINERS IN THE CENTRIFUGE.
- 5. Only use the rotors (heads) and accessories specified by the manufacturer. If unsure of which rotor or tube to use, ask Responsible Person or call the manufacturer.
- 6. The accessories and contents must be carefully balanced, and the load distributed symmetrically around the head and axis for each carrier before starting the centrifuge.
- 7. Buckets must be correctly located and properly seated in the rotor.
- 8. Before use, the lid must be closed and all externally securing devices firmly placed and fixed.
- 9. The centrifuge must be stopped by its timer or by pressing the Stop button not by switching it off at the mains supply. The lid must not be opened until the rotor has come to rest. The rotation of the rotor should never be stopped by hand.
- 10. Do not centrifuge materials capable of developing flammable or explosive vapours.
- 11. Do not move the centrifuge during operation. Do not lean or rest against the centrifuge during operation.
- 12. Do not centrifuge substances which could damage or affect the mechanical strength of the material of the centrifuge, the rotor, or the buckets. See Operators Manual for details of chemical resistance to plastics.
- 13. Infectious, toxic, pathogenic, or radioactive material MUST be centrifuged in capped/sealed containers inside sealed buckets only.
- 14. Only use rotors which have been approved for the centrifuge (See Operators Manual). If you are unsure of which rotor or tube to use, contact the manufacturer.
- 15. Stay at the centrifuge until it is running smoothly. Shut the machine down immediately if there is any unusual noise or vibration. Report non-resolvable issues to the Responsible Person.
- 16. The bowl, rotor and accessories should be disinfected regularly to counter any contamination from aerosols and droplets. Always clean the equipment using 2 % detergent solution for cleaning. Do not use corroding or aggressive substances.
- 17. Check that the replacement of time-expired components is made according to the manufacturer's Instructions Do not use rotors or buckets which have exceeded their maximum operating life. Rotors and accompanying buckets have a maximum operating life of seven years.

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3. RESPONSIBILITES

CBE Laboratory Users

- i. Shall ensure that they are familiar with the centrifuge, its controls, requirements, and emergency procedures by reference to this SOP, and Manufacturer's Operating Instructions.
- ii. Shall be an experienced centrifuge user or will have received appropriate training. Before a new user is allowed to operate a centrifuge on their own, competence to operate the centrifuge must have been assessed by the responsible person or another experienced user.
- iii. Shall ensure that the centrifuge is suitable for the work they intend to carry out.
- iv. Shall record alarms, adverse events, non-conformances, or malfunction on the Weekly housekeeping log and notify the Laboratory Manager / Responsible Person.
- v. Shall ensure that the centrifuge and rotors are properly installed and set properly according to the Operators Manual.
- vi. Shall complete the cleaning/maintenance procedures as required and record in the relevant sections on the Equipment Maintenance Record on door to respective laboratory.

Responsible Person (RP)/Laboratory Manager (LM)

- i. Shall ensure laboratory personnel are given suitable information, instruction, training or supervision in the safe use and maintenance of the equipment, providing the following:
 - a. information on the hazards and risks to health
 - instruction in safe procedures; training, where necessary; and effective supervision to ensure, so far as is reasonably practicable, that centrifuges are operated without risks to the health of employees and other persons, i.e. including students and visiting research workers
- ii. Shall coordinate weekly maintenance duties to be performed by laboratory personnel (according to SOP004).
- iii. Shall regularly review maintenance logs and investigate reported alarms, adverse events or nonconformances associated with the centrifuge.
- iv. Shall schedule servicing and preventative maintenance with authorized service representatives. External maintenance and servicing of the equipment can only be performed after it has been suitably disinfected (see SOP003) and a "Decontamination Certificate" has been issued (a proforma is available on the CBE LEARN page, please refer to section 6).

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4. EQUIPMENT AND MATERIALS

- Heraeus Megafuge 40 centrifuge (Cat. No. 11696200)
- TX-1000 swing-out cross rotor (Cat. No. 15133169)
- Tx-1000 buckets (Cat. No. 15163169), biocontainment caps for TX-1000 buckets (Cat. No. 13449289) and 500ml conical centrifuge tube adapters (Cat. No. 15169485)
- Centrifuge tubes: Corning plug seal cap, sterile, polypropylene, non-pyrogenic, RCF:6000xg, max 500mL (Fisher, 10018910)
- 1:50 Chemgene for cleaning
- 2% Detergent solution (Neutracon)
- Anti-corrosion oil
- Rotor grease

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5. PROCEDURE

5.1 Pre-run checks

NOTE: A record for the outcome of routine inspections MUST be maintained. A safety inspection outlined below must be carried out before each use.

NOTE: Lifetime of the Rotor: There is no limitation on the service life of the high-performance rotors. However, it is recommended that after 10 years the rotors and accessories should be inspected by the manufacturers.

NOTE: Autolock system: the centrifuge is equipped with an Autolock system which automatically locks the rotor to the centrifuge spindle. The rotor does not require bolting on to the centrifuge spindle.

CAUTION: Centrifuging can produce aerosols and therefore bio-hazardous material MUST NOT be centrifuged in open containers. Sealed containers provide initial protection but should be reinforced by the use of sealed buckets or rotors, which reduce the hazard if the container collapses.

CAUTION: DO NOT reach into the crack between the lid and the housing. The lid is drawn shut automatically.

5.2 Before each centrifuge run

- i. Inspect all the all safety relevant parts of the centrifuge i.e. rotor and accessories for cleanliness and visible signs of damage, abrasion, discolouration or wear e.g. cracks, nicks, chemical degradation, pitting or corrosion. If necessary, remove any dust, foreign objects, or residue from the chamber. The Autolock and o-ring must be clean and undamaged.
- ii. Inspect buckets and centrifuge tubes for visible signs of damage, malformation, or wear (e.g. cracks, nicks, chemical degradation, pitting or corrosion).

CAUTION: If any sign of visible damage or wear is detected, DO NOT USE the centrifuge. Consult the Responsible Person and record the outcome of the inspection on the Equipment Maintenance of centrifuges Record .

NOTE: Slight scratches and cracks can cause severe inner damage to the rotor materials, which are difficult to detect with the eye. If in doubt, DO NOT USE the rotor.

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> iii. Make certain the Autolock and o-ring are clean and undamaged. Inspect the rotor and check that it sits on the autolock correctly and locked in place by pulling at its handle.



- Make sure the bowl is dry and that the drive spindle is clean. In the event of condensation water ίV. formation, dry the centrifugal chamber by wiping out with an absorbent cloth
- Fill sample containers only within the range recommended by the manufacturer. Never fill ٧. sample containers when they are mounted in the centrifuge unless contained in a head specifically designed for this method of filling.
- Distribute the load symmetrically around the rotation assembly so that it is evenly balanced and vi. complies with the manufacturer's instructions in this respect. Always consider all components when balancing.
- vii. Select a centrifuging speed up to the maximum recommended by the manufacturer according to the head and accessories to be used or to the recommended reduced speed if the sample has a high density.
- Follow the manufacturer's instructions for starting and (where the control is not automatic) for viii. increasing the speed of rotation.

5.3 Operation

5.3.1 Inserting the rotor

NOTE: It is not permitted to use any other rotor other than those designated by the manufacturer.

- i. Open the centrifuge by pressing the lid open key.
- Place the rotor over the centrifuge spindle and let it slide slowly down the centrifuge spindle. ii. The rotor clicks automatically into place.

CAUTION: do not push the rotor down using force. If the rotor is very light, then it may be necessary to press it onto the centrifuge spindle with a bit of pressure.

iii. Check if the rotor is properly installed and locked by pulling on the handle. If the rotor can be pulled up, then it must be re-clamped to the centrifuge spindle.

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CAUTION: if the rotor cannot be properly locked in place after several attempts, then the Autolock is defective; DO NOT use the rotor, check for damage, and place a DO NOT USE sign on the equipment.

iv. If available close the rotor with the rotor lid. Close the centrifuge lid by lightly pressing it down. The lid should audibly click into place.

5.3.2 Removing the rotor

- i. Open the centrifuge lid.
- ii. Grab the rotor handle with both hands and press against the green Autolock key. At the same time pull the rotor directly upwards with both hands and remove it from the centrifuge spindle. Make sure not to tilt the rotor while doing this.



NOTE: When using Aerosol tight rotor lids the rotor can only be removed with the lid closed. This is to protect the user and the samples.

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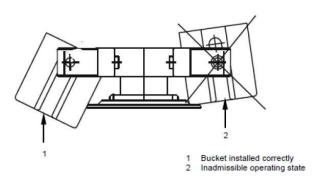
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5.3.3 Placing the tubes in the swing-out rotor (TX-1000)

- i. Distribute the load symmetrically in the inserts within the buckets so that the rotor is evenly balanced. It is essential that opposing buckets carry an identical load, and in particular are filled with a liquid of density not grossly dissimilar to that being centrifuged. Always consider all components (e.g. inserts and trunnion rings) when balancing. There should be no more than 100g difference between buckets next to each other
- ii. Avoid contaminating the outside of a sealed bucket. Never fill sample containers when they are mounted in the centrifuge.
- iii. Place the buckets correctly on the trunnions, at a slight angle, pointing into the middle. ALWAYS insert all four buckets to ensure correct running of the centrifuge
- iv. Close the lid of the centrifuge by lightly pressing it down. Ensure that this is accompanied by a clicking sound. The lid will be locked once the centrifuge is put into operation.



Note: Maximum RCF permitted for the TX-1000 is 4122 x g.

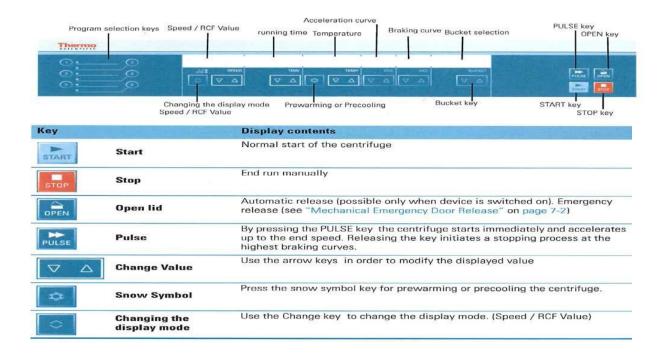
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5.4 Display/program options



5.4.1 Setting the run speed/RCF

- i. Press the Arrow keys below the SPEED display in order to open the speed/ RCF value menu.
- ii. Press the Change display mode key to toggle between the two modes and select the correct parameter. Note: speed parameter is measured in rpm; RCF parameter is measured in g.
- iii. Press the arrow keys repeatedly until the desired value shows.
- iv. Acceleration/deceleration can be adjusted by selecting a pre-set curve (refer to operator manual) in the same manner using the arrow keys below the ACC or DEC displays, there are 9 curves to select from.

5.4.2 Setting the run time

- i. Press the arrow keys below the TIME display in order to open the runtime selection menu. The display shows the message "set time".
- ii. Enter the run time in H.mm. by pressing the arrow key repeatedly until the desired value shows.

NOTE: During continuous operation, the centrifuge will continue running until you stop it manually. The display will state HOLD, Non-Stop Mode.

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5.4.3 Bucket selection

NOTE: Bucket selection is only possible for swing-out rotors. The bucket code corresponds to the last four digits of the bucket catalogue number.

- i. Press the BUCKET key or arrow keys below the BUCKET display. The display shows the message "Set bucket code".
- ii. Press the BUCKET key or arrow keys repeatedly until the bucket being used is displayed.

5.4.4 Setting the run temperature (-10° to +40°C)

NOTE: The centrifuge run temperature should be set at least 30 minutes before use to allow the centrifuge to equilibrate to the set temperature.

- i. Press the TEMPERATURE key or arrow keys below the TEMPERATURE display. The display shows the message "Set temperature".
- ii. Press the arrow keys repeatedly until the temperature required is displayed.

NOTE: To <u>save programmes</u>, enter the programme parameters then press any of the programme store keys for 4 seconds.

5.4.5 Pulse function

For short term centrifugation the centrifuge has a PULSE-function. By pressing the PULSE key, spinning will start and continue until the key is let go. The centrifuge accelerates and brakes at maximum power regardless of which rotor was installed, any rpm / RCF is overridden.

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5.5 Starting and stopping the centrifuge

Once the rotor has been properly installed, the main switch turned on and the lid closed the centrifuge can be started.

- i. Close the lid of the centrifuge, lock must audibly latch
- ii. Press the start key in order to start a centrifugation run.
- iii. The centrifuge accelerates to the pre-set speed with the time and temperature displays active.

NOTE: If the RCF setting is higher than the maximum permissible for the rotor then the display will show the message "max 4122 x g" once the centrifuge has been started. Within 15 seconds you can apply this value by pressing the start key again, and the centrifuge programme will continue. Otherwise the centrifuge will stop, and you will have to enter a valid number. You cannot open the lid as long as the centrifuge is running.

CAUTION: If the load is imbalanced, this will be indicated at speed higher than ~200 x g by the message "imbalance load". The run will terminate. Check the loading and once balanced restart the centrifuge (refer to operator manual for information on proper loading).

- iv. Once the pre-selected time has come to an end the centrifuge slows down, comes to a stop and the message "End" will appear on the display.
- v. The lid can then be opened by pressing the OPEN key. Run can be stopped at any time by pressing the STOP key.
- vi. If you selected continuous operation you will have to stop the centrifuge manually by pressing the STOP key. The centrifuge will decelerate at the designated rate, the "End" message will be displayed and then the lid can be opened.

NOTE: Refer to the operator manual for procedures for emergency (manual) lid release, for more information on troubleshooting, and instructions on entering the system menu (language, auto lid open and cycle count option selection).

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5.6 Cleaning and maintenance

- i. Pre-run checks include daily inspection of the rotor chamber, rotor, and accessories before use.
- ii. Weekly safety checks as part of weekly housekeeping tasks.
- iii. Recommended cleaning and maintenance intervals further to (i) are when polluted, monthly for the cabinet and every six months for the ventilation holes.
- iv. Refer to operator manual for cleaning procedure. In brief, ensure the centrifuge is turned off, unplugged and all parts removed. Use warm water with neutral solvent (neutracon) and rinse with distilled water. Dry all parts with a soft cloth. Treat the entire surface of aluminum parts with anti- corrosion oil (including cavities). Treat the bold of the swing out rotor with bold grease.
- v. For cleaning before and after use, use 1:50 Chemgene solution to wipe down all accessible surfaces, taking care not to remove the grease from the rotor bold and bucket hinges.
- vi. Centrifuges must be inspected and serviced every 2 years by Centriservices.
- vii. Any maintenance should be recorded in the equipment maintenance record sheets on door to respective laboratories.

CAUTION: DO NOT allow liquids to get onto the drive shaft or the bearings of the centrifuge, the shaft could freeze up or the grease in the bearings could be broken down.

5.7 Spill Response and sterilisation and Disinfection of centrifuge parts

NOTE: The centrifuges and the accessories consist of different materials and possible incompatibility must be considered. Before using detergents or decontamination agents, which have not been recommended by the manufacturer, the user should contact them to make sure that such procedure would not damage the centrifuge.

Where a spill, leak or breakage is detected within the centrifuge, the centrifuge MUST be disinfected immediately. The procedure will depend upon the risk group of the agent. If there is reason to believe that a breakage may have occurred whilst the centrifuge was running, adopt the following procedure:

- i) If centrifuge contamination is identified after the lid of the centrifuge is opened, carefully close the lid and turn off the centrifuge.
- ii) If centrifuge contamination is identified whilst the centrifuge is running, turn off the centrifuge.
- iii) In both cases DO NOT open lid as the bowl may contain an aerosol if the centrifuge tube or its seals have failed. ADOPT THE FOLLOWING PROCEDURE:
- iv) Wait at least 30 minutes before opening the lid to allow aerosol to settle.

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- v) Place notice on the lid to alert others and notify Responsible Person.
- vi) Put on clean disposable gloves and full-face protection. Respiratory equipment may be required seek advice from Responsible Person.
- vii) After 30 minutes, transfer the centrifuge to BSC only if the external surface is dry. If the external surface appears to be wet, wipe the external surface with virkon solution followed by 70 % IMS solution.CAUTION: DO NOT EXPOSE METAL PARTS TO VIRKON FOR MORE THAN 10 MINUTES.
 - (viii) Carefully open the lid inside the BSC.
 - (ix) Carefully retrieve any broken tubes with forceps and place in sharps container.
 - (x) Remove the rotor, bucket, accessories, and the lid. If practicable autoclave or alternatively soak in Virkon solution for 10 minutes. Thoroughly wipe down with 70% IMS.

 Place the rotor back into the centrifuge after ensuring the rotor and the components are dry.
 - (xi) Remove PPE and contaminated clothing. Place all contaminated PPE in an autoclave bag/container for decontamination (reusable items) or disposal as biohazardous waste.
 - (xii) Wash hands and other potentially contaminated areas with soap and water.
 - (xiii) Inform lab staff when clean-up is complete.
 - (xiv) Inspect the centrifuge for defective and damaged components. Retain all components and debris for inspection. Do not use centrifuge or rotor again until authorised by the Responsible Person.

5.8 Centrifuge Malfunction

(i) If any part of the equipment fails or malfunctions, including faults or defects, indicated by vibration, noise or by failure to operate, the user should contact the Laboratory Manager/Responsible Person. With permission of the Laboratory Manager or Responsible Person the user should consult the Operator Instruction Manuals to access fault finding, error displays and troubleshooting procedures.

CAUTION: DO NOT ATTEMPT TO REPAIR THE CENTRIFUGE YOURSELF

- (ii) All problems and corrective actions should be recorded in the Equipment Maintenance record.
- (iii) If the equipment fails to work or malfunctions and cannot be rectified according to troubleshooting procedures detailed in the Operator and Users Manuals the Laboratory Manager must be informed and the centrifuge must be tagged and locked-out or "Do Not Use" notice posted on the equipment. Contact

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the manufacturer for advice and coordinate with the Lab Manager for external maintenance and servicing.

NOTE: Centrifuges in need of repair should be tagged and locked-out (or "Not in Use" label applied) while awaiting service.

(iv) External maintenance and servicing of the equipment can only be performed after it has been suitably disinfected (refer to SOP003 for further details) and a 'Decontamination Certificate' has been issued (a proforma is available on the CBE website). NOTE: A 'Declaration of decontamination'; available in the Operators Manual may also be required. Permit to works should be used for external contractors.

5.7 Centrifuge/Rotor Decommissioning Checklist

- (i) If the centrifuge or rotor needs to be decommissioned, for example due to relocation or change of use, it is necessary to ensure no hazardous materials are left behind and that the unit has been decontaminated and made safe for future use or removal. A checklist that can be used to record that the centrifuge or rotor has been suitably decommissioned is provided in Section 6.
- (ii) On completion, the form should be forwarded to DSO or another responsible person to request its disposal. If multiple items need to be disposed of together from the same room, it may be possible to use one form — as long as <u>each item</u> is listed and each has a "Safe for Disposal" sticker / note on it to confirm that appropriate cleaning/disinfecting has been carried out. A completed decommissioning checklist precludes the need for maintenance staff and contractors to be issued with a Laboratory Permit to Work.

6. **DOCUMENTATION**

The following records are outputs of this SOP and can be found on the CBE internal website:

- 6.2 QS- FORM-006 Centrifuge decommissioning checklist
- 6.3 QS-FORM-009 Generic equipment decontamination certificate
- Weekly Housekeeping sheet
- Lab Equipment Maintenance Record.

These records shall be filed in the Equipment File and stored in the CBE Office or otherwise archived for future review or retrieval.

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		Number
08/06/2020 J.Bowdrey	Major review Removed the centrifuge maintenance sheet, and added use of the Weekly Housekeeping log and Equipment Maintenance log.	002
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Annex 1 : Safe Use of Centrifuges (Extracts from BS4402)

Moving parts in centrifuges can reach very high kinetic energies. Breakage of a rotor or accessories inside the centrifuge releases large amounts of energy, and debris can be expelled at high speeds from the centrifuge; bodily movement of centrifuges for a distance of several metres can also occur in this situation.

Modern centrifuges must be designed and manufactured to British Standard BS 4402: 1982, Safety Requirements for Laboratory Centrifuges. This Standard is intended to ensure that the casing of the centrifuge will contain the debris of components which break inside it and to ensure that centrifuges are fitted with necessary interlocks and other safety devices.

It is important that all users of centrifuges are properly trained in the safe use and maintenance of the equipment.

Appendix B of BS 4402 gives advice on the use of centrifuges and part of it reads:

'RECOMMENDATIONS ON THE USE OF CENTRIFUGES'

B.IRecommendations applying to all centrifuges

- B.I.I. **Introduction**: When running, a centrifuge has considerable kinetic energy and even a small 'bench centrifuge' not complying with the requirement of this British Standard can expel debris, or even move bodily, for a distance of several metres in the event of mechanical failure. While considerable effort goes into the design and construction of centrifuges to try to minimise the hazards, sensible use is also important for the safety of operators and others in the laboratory.
- B.1.2 **Installation**: It is essential that all centrifuges be fixed to the working surface, following the specifications and recommendations of the manufacturer for suitable fixings and fixing details. Care should be taken to ensure that the working surface and its substrate are strong enough to withstand the maximum forces of rotation and translation expected ac he fixings in the event of a disruption of the rotation assembly (see instruction manual) A centrifuge may move abruptly in the event of such a disruption, in spite of the fixings, but the distance will be limited if the centrifuge is correctly fixed.
- B.1.3 **Training of Operators**: Training in the operation of the centrifuge should be carried out by an experienced user or (by arrangement) by the manufacturer's representative, and should be given to all operators of the centrifuge before they are permitted to use the centrifuge.
- B.1.4 **Precautions to be taken when using centrifuges**: Note: attention is also drawn to recommendations given in BS5345 and to the "User Guide for the Safe Operation of Centrifuges" published by the Institution of Chemical Engineers
 - B.1.4.1. Inspect each sample container and head accessory and all seals (if any) before filling. Reject any which are damaged or not authorised by the manufacturer, since such components could cause unsatisfactory sealing, breakage or jamming in the centrifuge head.

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- B. 1.4.2 Fill sample containers and head accessories only within the range recommended by the manufacturer and avoid contamination outside the seal of a sealed head or sealed bucket. Never fill sample containers when they are mounted in the centrifuge unless contained in a head specifically designed for this method of filling.
- B. 1.4.3Distribute the load symmetrically around the rotation assembly so that it is evenly balanced and complies with the manufacturer's instructions in this respect. If more than one sample container is placed in a bucket, use an appropriate insert to ensure their correct positioning. It is essential that balancing buckets carry an identical load, and in particular are filled with a liquid of density not grossly dissimilar to that being centrifuged. Always consider all components (e.g. inserts and trunnion rings) when balancing.
- B. 1.4.4 Select a centrifuging speed up to the maximum recommended by the manufacturer according to the head and accessories to be used or to the recommended reduced speed if the sample has a high density (i.e. above 1200 kg/m3).
- B. 1.4.5 Follow the manufacturer's instructions for starting and (where the control is not automatic) for increasing the speed of rotation.
- B. 1.4.6 Allow the rotation assembly to come to rest before attempting to open the centrifuge lid. NEVER attempt to slow down a head by hand.

B. 1.5 Maintenance

- B. 1.5.1 any liquid spilt in the centrifuge should be removed immediately.
- B. 1.5.2 At the end of each day's use of the centrifuge, clean and dry the inside of the centrifuge chamber, all parts of the rotation assembly and any head accessories used. In order to reduce corrosion, inserts and adaptors should be removed after use to allow adequate cleaning and drying. Ensure that the manufacturer's recommendations on cleaning materials are followed.
- B. 1.5.3 periodically inspects the clean and dry rotation assembly for any signs of corrosion or mechanical defect. If such signs are seen, do not use it until advice has been obtained from the manufacturer or his representative
- B. 1.5.4 it is advisable to have the centrifuge heads, buckets and accessories checked by the manufacturer's representative, at the recommended intervals.
- B. 1.6 Procedures after a disruption of the rotation assembly.
 - B. 1.6.1 Whenever a mechanical failure (other than the simple breakage of sample containers) of any part of the rotation assembly is suspected to have occurred, the procedure detailed in B. 1.6.2 to B.I.6.5 is the simplest procedure that should be followed. (For hazardous materials, a more extensive procedure applies.)
 - B. 1.6.2 Immediately isolate the centrifuge from the electrical supply. Do not attempt to open the centrifuge chamber until at least 30 minutes after all motion has ceased.

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Location: CBE Laboratory Unit H34

- B. 1.6.3 Avoid moving defective and damaged components, unless it is necessary for decontamination of hazardous materials, and in any event retain all components and debris for inspection by the manufacturer's representative.
- B. 1.6.4 Note all the details of the incident and any measures subsequently taken and inform the relevant authority of the laboratory.
- B. 1.6.5 Do not use the centrifuge or the rotation assembly again until the necessary repairs and replacements have been completed and the manufacturer's representative has given an assurance that its safety is not impaired.' The Appendix of BS 4402 goes on to deal with extra precautions required when centrifuging hazardous materials (see below).

Rotors

It is essential that rotors are properly handled, used and maintained.

An ultracentrifuge rotor, for example, experiences 600,000 times the force of gravity. Each gram of it will effectively weigh over 600kg. Under these conditions a small flaw in part of it y lead to failure and the virtual explosion of the rotor within the centrifuge.

The centrifugal forces cause rotors to stretch. At low and normal speeds, the change of size is elastic and the rotor returns to its original size when stationary. At high or over speeds the elastic limit of the material may be exceeded and plastic deformation occurs. If this process recurs micro cracks may be formed and grow at high stress points within the rotor and subsequently cause catastrophic failure.

Corrosion of the rotor by moisture or chemicals greatly reduces the level of stress at which micro cracks form and grow. This increase the probability of early failure (by corrosion fatigue or stress-corrosion cracking). For these reasons it is essential to conform to the manufacturer's conditions on maximum rotor speeds and maximum running times, and to follow closely their cleaning and maintenance procedures

Useful advice on the care of rotors is given in the cited references. The publication Centrifuges by L.W. Price, University of Cambridge, is particularly use the following list of some causes of rotor failure.

- 1. Misplaced trunnions on MSE Minors
- 2. Use of wrong accessories e.g. i h horizontal rotors. In a laboratory where several makes of centrifuge are in use, it is possible to mix up accessories which are not interchangeable. Keep them separate.
- 3. Failure to reduce speed abo e specific gravity of 1.2.
- 4. Powdered glass in rotor.
- 5. Failure to use speed restriction curves to avoid recrystalisation with CsC1 gradients.
- 6. Miss hooked buckets on swinging bucket rotors.

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- 7. Over speeding because de rated over speed disc not fitted.
- 8. Running badly corroded and weakened rotors or buckets, stress corrosion failure.
- 9. Loose lid on angle rotor.
 - 10. Incorrect installation of rotor on drive shaft.'

Bench Centrifuges

Although the kinetic energy attained by the components of bench centrifuges is lower than that involved in larger models their improper use can lead to dangerous failures.

Breakages of glass tubes or improper balancing can cause considerable vibration and 'wandering' of an unfixed centrifuge over the bench top. This may, in extreme cases, allow the mains cable to become wrapped around the centrifuge body with consequent strain and damage to the cable. The centrifuge may move completely off the bench-top to the floor. This may cause the chamber lid to open and the spinning rotor or buckets to be ejected at high speed.

A visual check should always be made to see that all tubes are filled to the same equal level and are not overfilled, that glass tubes are not cracked, and that a cushion is installed in each bucket.

Centrifuging Hazardous Materials

Hazardous materials must be centrifuged in the proper type sealed container. The basic principle of safe containment is an air-tight, leak proof seal.

Most equipment is unsafe for the centrifuging of flammable materials. A sealed chamber is required, and the necessary design features are laid down in BS4402 which allows only approved centrifuges to be marked with 'sc' (sealed chamber).

Advice on sealed containers for centrifuging hazardous chemicals is given in BS 4402 and in the book by Price. These also mention biologically hazardous materials, and these are dealt with in greater detail in Laboratory-Acquired Infections, CH Collins, Butterworths, 2ndEdition 1988.

If pathogenic materials are used it may be necessary to sterilise parts of a centrifuge and its accessories. Price gives the following relevant advice.

Before sterilising **always** remove and dissemble tube cap assemblies. Tubes and bottles for autoclaving should be separated from cap parts to avoid damage and deformation. Cellulose tubes cannot be autoclaved, and they may explode if autoclaving is attempted. Polyallomer tubes distort.

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Polycarbonate containers with join seams should not be autoclaved and this method of sterilisation is possible only for seamless polycarbonate containers. Even then severe distortion will probably render the container useless. Cold sterilisation is recommended.

Cold methods of sterilisation that can be used on container materials include ultraviolet radiation, zephiran chloride, ethylene oxide gas and 70% ethanol (except cellulose)

Remember that it is important to follow the manufacturer's instructions in all aspects of centrifuge use. Only trained persons should use centrifuges. If in doubt about the operation of a centrifuge - ask somebody else.