

SCHOOL OF MEDICAL SCIENCES

General Risk Assessment – Within labs or offices

Department: Physiology and Pharmacology	Room number: LG04	Date assessed: 6 March 2004
Procedure title: Preparing diluted aliquot of CD4 Dynabeads.	Assessed by: Trevor Lewis	Supervisors signature:

Description of Procedure:

The CD4 Dynabeads (Dyna) are supplied in a concentrated stock solution in a sealed glass vial, which contains 1.4×10^8 beads / ml in phosphate buffered saline, pH7.4, containing 0.1% bovine serum albumin and 0.2% sodium azide as a preservative. CD4 Dynabeads are superparamagnetic polystyrene beads coated with a primary monoclonal antibody specific for the CD4 membrane antigen. A 50 μ l aliquot is placed in a microcentrifuge tube with 500 μ l of filter sterilised HEPES buffered saline and centrifuged to pellet the CD4 beads. The supernatant is removed and placed in a waste tube. The beads are rinsed by resuspending in 500 μ l of filter sterilised HEPES buffered saline. The tube is again centrifuged to pellet the beads and the supernatant removed to a waste tube. The beads are then finally resuspended in 500 μ l of filter sterilised HEPES buffered saline.

Apparatus/Equipment used:

Displacement pipetters; microcentrifuge tubes; bench-top microcentrifuge; syringe tip filter units (0.22 μ m mesh).

Chemical Hazards – Properties of chemicals to be used

Hazardous substance	Hazards (see label and MSDS) Indicate if relevant								MSDS viewed
	Flammable	Corrosive	Toxic	Harmful	Irritant	Oxidizing	Radioactive	Safe	
Reactants or Products									
0.2% Sodium azide (NaN_3)			Yes		Yes				Yes

First Aid Notes:

Sodium azide is toxic if swallowed – this is an unlikely route in the laboratory and the volume used is very small.

Acute human intoxication is characterized by profound hypotension (unresponsive to presser drugs) and may include; tachycardia (rapid heart rate), rapid breathing, hypothermia, acidosis (i.e. acidic body fluids), convulsions, and severe headache.

Sodium azide is an irritant if exposed to the skin or to eyes. Affected areas of skin should be washed with soap and water. Eyes should be irrigated with water for at least 15 minutes, keeping the eye lids open. Seek medical attention.

Review of Hazard/Risk

Step in process List major steps or tasks in process	Hazard	Risk (Harm)	EXISTING CONTROLS List all current controls that are already in place or that will be used to undertake the task e.g. -personal protective equipment - identify facility type, location - existing safety measures	Risk rating with existing controls?			ADDITIONAL CONTROLS REQUIRED Additional controls may be required to reduce the risk rating e.g. - greater containment - specific induction / training - additional personal protective equipment	Risk rating with additional controls?		
				Consequences	Likelihood	Rating		Consequences	Likelihood	Rating
1. Opening glass vial containing the stock suspension of CD4 beads and dispensing aliquots of suspension	Aerosols of CD4 beads suspension	Potential for the inhalation of the beads and solution containing sodium azide	<ul style="list-style-type: none"> Care is taken to expel the contents of pipetters below the surface of any solution to reduce the chances of producing aerosols. Appropriate personal protective equipment is to be worn at all times. Training of all lab personnel in good laboratory practice. Yearly inspection and testing of power cord and equipment (inspection tags attached). Appropriate personal protective equipment is to be worn at all times. Appropriate personal protective equipment is to be worn at all times. 	3	E	M	<ul style="list-style-type: none"> All operations with the CD4 beads are carried out in a class II biological safety cabinet. 	2	E	L
	Spills or splashes of suspension	Potential irritation of skin, or damage / irritation to the eyes		2	D	L				
4. Centrifuging the aliquot to pellet the CD4 beads	Electrical	Electric shock		2	E	L				
	Spills or splashes of suspension	Potential irritation of skin, or damage / irritation to the eyes		2	D	L				
5. Removing the supernatant and resuspending the beads	Spills or splashes of suspension	Potential irritation of skin, or damage / irritation to the eyes		2	D	L				

<u>Containment Facility used</u>	<u>Sterilization/Decontamination requirements</u>	<u>Disinfectant</u>	<u>Location of Spill Kit</u>	<u>Emergency information</u>
Location: LG04 PC level: PC1	Disinfect contaminated surfaces with bleach solution for at least 10 minutes	Type: Bleach solution Suitability:	Room: LG04 near the storage cabinet for corrosives	Contacts: Prof Peter Barry Dr Trevor Lewis Dr Andrew Moorhouse

Physical Hazards

- | | | | | |
|--|---|--|--|---------------------------------------|
| <input type="checkbox"/> Explosion | <input type="checkbox"/> Hot liquids | <input checked="" type="checkbox"/> Spillage/Splash | <input type="checkbox"/> Chemical fumes | <input type="checkbox"/> Fire |
| <input type="checkbox"/> Heavy objects | <input type="checkbox"/> Cold liquids | <input type="checkbox"/> Sharps | <input type="checkbox"/> Hot equipment | <input type="checkbox"/> Other people |
| <input type="checkbox"/> Radiation | <input type="checkbox"/> Cold environment | <input type="checkbox"/> Ignition source | <input checked="" type="checkbox"/> Biological | <input type="checkbox"/> Other |
| <input type="checkbox"/> Pressure/vacuum | <input type="checkbox"/> Noise | <input checked="" type="checkbox"/> Electrical current | <input type="checkbox"/> UV/X-Ray/laser | |

Comments:

Any splashes or spills are expected to be of small volumes, as the stock suspension is a total of 2 ml and the dispensed aliquots are resuspended in a volume of 500 µl. Such small volumes can be easily contained should a spill occur. Protection from any aerosols produced is provided by the class II biological safety cabinet. The bench top microcentrifuge should be checked to see that it has a valid electrical inspection tag. It is not known what effect inhalation of an aerosol of CD4 beads would have. Any spills on exposed skin may produce an irritation.

Control Measures Needed

- | | | | |
|--|---|---|---|
| <input checked="" type="checkbox"/> Protective clothing | <input checked="" type="checkbox"/> Fume cupboard | <input type="checkbox"/> Hearing protection | <input checked="" type="checkbox"/> PC1 lab |
| <input checked="" type="checkbox"/> Appropriate eye protection | <input type="checkbox"/> Appropriate footwear | <input type="checkbox"/> Handling aids | <input type="checkbox"/> PC2 lab |
| <input checked="" type="checkbox"/> Gloves | <input type="checkbox"/> Appropriate face mask | <input checked="" type="checkbox"/> training received | <input type="checkbox"/> Hot room |
| <input type="checkbox"/> Nitrile gloves | <input type="checkbox"/> Remove ignition source | <input type="checkbox"/> Assistance needed | <input type="checkbox"/> S.O.P viewed |

Comments:

Dispensing aliquots of CD4 beads, centrifuging the aliquots and resuspension of aliquots should all be carried out in a class II biological safety cabinet, to protect the user from aerosols and to protect the product from contamination. Personnel should be trained in the use of the biological safety cabinet. The rubber stopper should be carefully replaced in the stock suspension of CD4 beads and sealed with 'parafilm' laboratory wrap. Standard personal protective equipment (lab coat or gown, eye protection, latex gloves) should be worn at all times. Hair should be tied back. Contact lenses should not be worn.

Other Control Measures

Location of nearest Spill Kit: LG04 near corrosives cabinet	Location of nearest Eye Wash: First aid kit, LG04
Location of nearest First Aid Kit: LG04 near corrosives cabinet	Nearest Safety Shower: Not available

Clean up and disposal of contaminated waste

<input type="checkbox"/> Non halogenated hydrocarbons	<input checked="" type="checkbox"/> Gloves	<input checked="" type="checkbox"/> Other (please specify)
<input type="checkbox"/> Halogenated hydrocarbons	<input checked="" type="checkbox"/> Biological	<input type="checkbox"/> Radioactive
<input checked="" type="checkbox"/> Aqueous (not heavy metal)	<input type="checkbox"/> Sharps	
<input type="checkbox"/> Aqueous heavy metals	<input type="checkbox"/> Glass bin	

Clean up procedure:
The first rinse of the CD4 beads should be disposed of as cytotoxic waste, for it contains low levels of sodium azide (NaN₃). The external surface of the CD4 suspension vial should be kept clean and clear of spills or splashes by wiping down with alcohol and disposing the wipe as cytotoxic waste. Pipette tips and gloves can generally be disposed of as biological waste. In the case of any spills or splashes of the CD4 suspension onto gloves, they should be removed as soon as practicable and disposed of as cytotoxic waste. Microcentrifuge tubes containing used or old aliquots of CD4 beads can be disposed of as biological waste. Any spills are expected to be of small volumes (approx. 1ml or less) and can be wiped up and surfaces decontaminated with bleach solution.

Overall Risk Assessment:

<input checked="" type="checkbox"/> Low risk (simple assessment). File and keep this assessment.
<input type="checkbox"/> Risks are significant but can be effectively controlled. File and keep this assessment.
<input type="checkbox"/> Risks are significant and not easily controlled. Carry out a more detailed risk assessment and report. File and keep Consider other control measures and provide training.

Signed:	Date: 6 Mar 04	Date for review: 6 Mar 05
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