

WT Microorganism Risk Assessment

FullSerial No	
Version	1
Title	Cholera toxin induced IgA production in-vivo
Containment Level	Containment Level 2
PI responsible	Cantrell, Prof Doreen
Division	Cell Signalling and Immunology
Building	Wellcome Trust Biocentre
Lab No	DAC
Name of assessor	Sarah Thomson
Approval date	
Review date	

1. Brief description of project

The aim of these experiments are to track antigen-specific IgA responses in WT and GA mice in response to oral immunization with cholera toxin.

2. Hazards to human health associated with the microorganisms

Cholera toxin (CTX) is protein complex secreted by the bacterium *Vibrio cholerae* and is responsible for the massive, watery diarrhea characteristic of cholera infection.

In a laboratory setting, the main hazard is that the operator's hands are contaminated with infected material and that the toxins are transferred to other surfaces and food. The use of gloves, disinfectant and due care and attention is sufficient to reduce this hazard to a minimum.

3. Assign a provisional containment level

Containment Level 1 Containment Level 2 Containment Level 3

4. Hazards to the environment associated with the microorganisms

No –The risk of infection to humans is minimal as and is further reduced by: wearing appropriate PPE; housing animals in sealed, individually ventilated cages; adhering to strict disinfection and cleaning regimes; use of a Class II microbiological safety cabinet. Cage waste is removed within a Class II cage-change cabinet, double bagged then autoclaved before disposal. Used cages are treated with Sanosil Super 25 (efficacy data available on manufacturer's web site) prior to regular

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5a. Brief description of nature of work (include maximum culture volumes)

Cholera toxin is delivered from List Biological Laboratories (United States) as a lyophilized powder in vials of 1 mg. The toxin is reconstituted in water (less than 1ml volumes) in a fume hood, using double nitrile gloves and a spill tray. Any spills will be neutralized with bleach. After reconstitution, the cholera toxin will be stored in a locked box in the fridge. Only authorized people will have access to the keys and an inventory of the amount of cholera toxin will be kept. Transport and handling of cholera toxin will follow the safe operating procedures as detailed in SOP document 157.

Mice housed in sealed ventilated cages will be infected by oral gavage of a small volume (200µl) of 2.5ug active cholera toxin and blood and tissue assessed 10 days later. Oral administration will be done by highly skilled and experienced personal license holders. Great care will be taken to avoid accidental systemic administration of the toxin and mice will be culled using Schedule 1 procedures if showing severe clinical signs,

5b. Is a microbiological safety cabinet or isolator required to protect the worker from aerosol transmission?

5c. Waste disposal

Critical waste processing criteria:

- (a) Do ANY of the microorganisms covered by this risk assessment have the potential to cause harm to human health or the environment? Yes
 No
 Don't know
- (b) Do ANY of the microorganisms covered by this risk assessment have the capacity to survive, establish and/or multiply in the environment? Yes
 No
 Don't know
- (c) Do ANY of the microorganisms covered by this risk assessment have capacity to transfer genetic material to other microorganisms? Yes
 No
 Don't know

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5d. Are sharps required? Yes or no. If yes, justify use.

No

5e. If the work involves experimental infection of animals is it known if the animal will shed the microorganisms?

If yes, give details and measures to prevent exposure.

YES – can be shed to the bedding of animals and can spread to animals within the same cage by fecal matter. The risk of infection to humans is minimal as and is further reduced by: wearing appropriate PPE; housing animals in sealed, individually ventilated cages; adhering to strict disinfection and cleaning regimes; use of a Class II microbiological safety cabinet. Cage waste is removed within a Class II cage-change cabinet, double bagged then autoclaved before disposal. Used cages are treated with Sanosil Super 25 (efficacy data available on manufacturer's web site) prior to regular cage washing procedure. Carcasses are double bagged and autoclaved prior to disposal.

5f. If the work involves experimental infection of plants what is known about the likely route of transmission of the microorganisms?

No

5g. Where will the microorganisms be stored?

Cholera toxin will be stored in a labelled locked box in the fridge in DAC lab. Only authorised people will have access to the keys and an inventory of the amount of cholera toxin will be kept.

5h. How will the microorganisms be transported within/between buildings to minimise risk of spillage/escape?

Samples will be doubly contained during transport & clearly labelled with a contact name & number, the nature of the sample & the biohazard symbol. Inner container/tube will be robust & leak-proof. Outer container will be robust, leakproof & contain enough absorbent material to absorb the total volume of sample should the inner container leak.

5i. Will staff/students receive any vaccination or health surveillance? If yes, give details.

No

5j. Emergency plan, if required.

Small scale CL2 activity therefore not required

5k. Monitoring

As per the standard procedures on the SLS H&S web site

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6. Final classification

Containment Level 1 Containment Level 2 Containment Level 3

7. Additional information