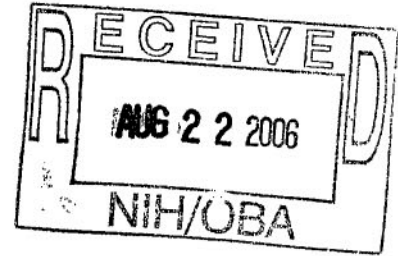




THE UNIVERSITY
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Date: 18 August 2006

To: Office of Biotechnology Activities
National Institutes of Health
6705 Rockledge Drive, Suite 750, MSC 7985
Bethesda, MD 20892-7985

From: *Jan Klein*
Jan Klein, Biological Safety Officer
Safety Department

RE: Report of Laboratory Incident

The purpose of this memo is to report a research-related incident that occurred in a research laboratory at the University of Wisconsin – Madison, per the NIH Guidelines Section IV-B. The incident involved recombinant *Brucella melitensis* 16M, modified to contain a plasmid (pBR-mcs4 (Amp^r with lux operon under *Brucella* 16M promoters). This pathogen is infectious at a low dose via inhalation. There is uncertainty about whether there was a breach of containment. We believe the likelihood that a breach occurred to be low, but it cannot be disregarded and we have taken precautionary steps.

On Monday August 7, a supervisor reported a possible breach in containment from a BSL-3 laboratory. The following provides a brief summary of the events and our assessment of the potential risks.

- Tubes (24 14-ml tubes containing 3 ml each) with culture medium were inoculated with *Brucella melitensis* 16M on Friday (8/4/06) and placed in a shaker unit over the weekend to propagate the microbe. When examined on Monday, it was discovered that caps had come off some of the tubes.
- Loss of culture medium from the tubes was not evident and none of the tubes was broken. There was a significant air gap between liquid culture medium and the top of the tube, reducing the likelihood that droplets could escape.
- The tubes had been placed within a plastic holder box that has a tight-fitting lid taped in place. The lid of the shaker unit was closed but does not have a seal.
- Two researchers may have been exposed to the microbe when they entered the lab on Monday. They were wearing appropriate personal protective equipment, including a respirator (N95, fit-tested), which should have provided adequate protection if any microbe was actually released within the lab. They were evaluated by a physician from the University Hospital Infectious Disease Unit. A serum sample was taken, and they were given antibiotics as a precaution. A follow up examination is scheduled for 1 month post-incident. These individuals participate in a medical surveillance program where serum is tested for exposure to *Brucella* every 6 months.
- It is possible but unlikely that some of the agent escaped from the lab into the environment surrounding the building where the incident occurred. The air from the lab exhausts directly outside from the roof of the building via a high velocity fan which rapidly dilutes the exhaust air. The risk to the public was reduced by low probability of

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escape of the microbe from the plastic box, the rapid dilution of the lab exhaust air, and the susceptibility of the pathogen to desiccation.

- The lab was under negative pressure, preventing exposure of other occupants within the building. The equipment and surfaces within the lab were decontaminated with a disinfectant that is known to inactivate this pathogen.
- Notifications were given to the CDC Select Agent Program and to the county public health officer.

The primary factor that contributed to this incident was a modification of the routine procedures. Tubes with snap cap lids were substituted for those with screw caps that normally are used. These tubes do not fit within the centrifuge safety cups that routinely are used in the shaker unit; the plastic box was substituted. The tubes do not touch the bottom of the box and instead rest on the rim of the cap. The shaking action was sufficient to knock the cap off. This modified procedure had previously been used without incident, but tubes with snap caps recently were acquired from a new supplier and they did not hold the seal as well as those previously used.

Follow up will focus on 1) ensuring that modifications to existing procedures are reviewed by a supervisor prior to implementation and possibly tested with an initial dry run; 2) training is provided to increase awareness of activities that potentially could generate aerosols and the importance of secondary containment when a BSC is not used for such activities.

Please feel free to contact me for additional information. I may be reached by phone (608-263-9026) or email (jklein@fpm.wisc.edu).

cc: Susan Paskewitz, IBC Chair
James Tracy, Responsible Official and
Assoc. Dean, SVM