## **Monoclonal Antibody (Mab) Production Guidelines**

## I. Overview:

Antibodies are important tools used by many investigators in their research. Producing monoclonal antibodies (mAb) requires immunizing an animal, usually a mouse with an adjuvant and the protein of interest; obtaining immune cells from its spleen; and fusing the cells with a cancer cell (such as cells from a myeloma). A tumor of the fused cells is called a hybridoma, and these cells secrete mAb. The development of the immortal hybridoma requires the use of animals; no commonly accepted nonanimal alternatives are available. Production of mAbs needs to be scientifically justified in the animal care and use protocol. In addition, the investigator needs to justify the number of animals used in the production of monoclonal antibodies, and must describe procedures taken to minimize pain and distress used in the production of antibodies.

## II. Guidelines:

- No more than 0.1 ml Pristane, or 0.25 ml Freund's Incomplete Adjuvant, may be used for peritoneal cavity priming.
- Animals must be observed at least three times per week the first week and daily thereafter, including weekends and holidays, to monitor the degree of abdominal distension, to relieve it as needed, and to look for other signs of illness.
- Fluid should be removed before abdominal distension is great enough to cause discomfort, or labored breathing, or to interfere with normal activity.
- Animals must be anesthetized prior to peritoneal tap.
- Ascites fluid may be harvested only once with recovery of the animal. Subsequent harvesting shall be performed as a terminal procedure.
- Ascites fluid should not exceed 20% of the baseline body weight.
- Animals that become moribund, cachexic, or that are otherwise unable to obtain food and water must be euthanized immediately.
- III. References:
  - 1. Monoclonal Antibody Production. Committee on Methods of Production Monoclonal Antibodies, Institute for Laboratory Animal Research, NRC. 1999