

<b>TITLE:</b>	<b>Monoclonal Antibody Production</b>
<b>Policy Number:</b>	2014-014
<b>Responsible Department:</b>	Institutional Animal Care and Use Committee
<b>Policy Contact:</b>	<a href="mailto:IACUCOffice@westernu.edu">IACUCOffice@westernu.edu</a>
<b>Approval Date:</b>	7/9/14
<b>Reviewed:</b>	6/7/17; 2/12/2020
<b>Revised:</b>	6/7/17 (Updated 3Rs link); 2/12/2020 (Added ascites "procedure must be outlined in an approved IACUC protocol or SOP")

**Purpose of Policy:** To ensure that the ascites method for the production of monoclonal antibodies (mAbs) in laboratory animals is used only if *in vitro* methods for their production cannot accomplish the same goals and thus avoid unnecessary pain or distress to laboratory animals.

**Policy Information:** Federal regulations require investigators to consider alternatives to procedures that may cause more than momentary or slight pain or distress to animals. The National Research Council's [Guide for the Care and Use of Laboratory Animals](#) states that certain procedures require special consideration due to the potential for unrelieved pain or distress and that the Institutional Animal Care and Use Committee (IACUC) must "weigh the objectives of the study against potential animal welfare concerns." [Section F8 of the Office of Laboratory Animal Welfare's \(OLAW\) FAQs](#) concurs with the recommendations of the National Research Council [Monoclonal Antibody Production](#) in that "during the accumulation of ascites there is likely to be pain and distress". The Report concluded that there is a scientific necessity for this method but that tissue-culture methods for the production of mAbs should be used unless there is clear evidence that they cannot be used.

As stated in the *Guide* cited above, the [3Rs](#) "have become an internationally accepted approach...to apply when deciding to use animals in research" and thus every effort should be made to replace animal models with non-animal models when possible.

The IACUC shall require rigorous scientific justification for the use of the ascites method for the production of mAbs in laboratory animals. Investigators must first document that *in vitro* methods for the production of mAbs have been unsuccessful and that:

- the proposed use of laboratory animals for this purpose is scientifically justified and
- methods that avoid or minimize discomfort, distress, and pain (including *in vitro* methods) to the animals have been considered.

Convenience of the ascites method or cost of purchasing mAbs from an outside source will not be accepted as justifications for the use of the ascites method unless the cost of purchasing the mAbs is so great that the IACUC considers the mAbs unavailable.

Problems associated with the use of the ascites method in mice include, but are not limited to:

- respiratory distress and circulatory shock,
- difficulty walking, drinking and eating,
- development of solid tumors.

Therefore, if the ascites method is approved by the IACUC, the procedure must be outlined in an approved IACUC protocol or SOP and the following conditions will apply:

- Animals must be monitored daily, including weekends and holidays, for the degree of abdominal distension and signs of illness.
- Ascites fluid must be tapped **before**
  - ascites fluid volumes exceed 20% of the animal's baseline body weight prior to hybridoma cell inoculation.
  - the animals are expected to experience unalleviated pain or discomfort from accumulation of fluid in the peritoneal cavity. Then the animals may be listed under Pain Category D (pain or distress, drug relieved). However, if significant inflammation or otherwise unrelieved pain is expected, Pain Category E (pain or distress, no relief provided) may be appropriate.
  - Abdominal distension is so great as to interfere with normal activity. Any animal with a grossly distended abdomen in which the skin is drawn tight must be tapped and/or euthanized.
- Animals must be observed **continuously** by trained personnel for at least 30 minutes following abdominal paracentesis for signs of hypovolemic shock and distress. If an animal appears hunched or lethargic, an equal volume of warm saline should be administered subcutaneously.
- **At a maximum, animals may be tapped twice and allowed to recover. The third tap, if one occurs, must be conducted after the animal has been euthanized.**
- Mice must be euthanized promptly if they show severe signs of pain or distress or illness such as huddling, ruffled coat, hunched posture, anorexia, dehydration, pallor, weight loss, inactivity, difficulty in ambulation, tachypnea or dyspnea.
- Scientific justification must be given for the use of priming agents other than incomplete Freund's adjuvant (IFA) or pristane.