**CNPRC**

**Investigator**

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<th>Last Name:</th>
<th>First:</th>
<th>Middle:</th>
<th>email:</th>
<th>Department:</th>
<th>Phone / Fax:</th>
<th>After hrs. #:</th>
</tr>
</thead>
</table>

**Contact**

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<tr>
<th>Last Name:</th>
<th>First:</th>
<th>Middle:</th>
<th>email:</th>
<th>Department:</th>
<th>Phone:</th>
<th>After hrs. #:</th>
</tr>
</thead>
</table>

**Species (common names):** rhesus  
**Number:** 36  
**Source:** CNPRC

**Project Title:** Mechanisms of protection with primate lentiviral vaccines

**Overnight housing location:** CNPRC  
**Day use:** CNPRC (workrooms or animal quarters)

**Animals will be maintained by:** [ X] Vivarium  
[ ] Investigator  
(If investigator maintained, attach husbandry SOPs.)

**Procedures:**

The animals will have intradermal injection of plasmid DNA administered. Blood samples will be collected by venipuncture and mucosal secretions will be collected by saline lavage. In addition, lymph node biopsies will be occasionally obtained.

**Special Husbandry Requirements:**

Describe any special requirements your animals have with respect to food, water, temperature, humidity, light cycles, caging type, bedding, or any other conditions of husbandry.

After immunization animals will need to be placed in indoor housing (non-infectious). After challenge animals will need to be placed in infectious housing.

**Hazardous Materials (only if in the animal room):**

<table>
<thead>
<tr>
<th>Infectious Agents?</th>
<th>Radioisotopes?</th>
<th>Chemical Carcinogens?</th>
<th>Toxic Chemicals?</th>
</tr>
</thead>
<tbody>
<tr>
<td>[ X] Yes</td>
<td>[ X] Yes</td>
<td>[ X] Yes</td>
<td>[ X] Yes</td>
</tr>
</tbody>
</table>

Agent(s): SIV
Summary of Procedures:

a) Briefly describe the overall intent of the study. Include in your description a statement of your hypothesis, the objectives and significance of the study. Your target audience is a faculty member from a discipline unrelated to yours. Do not use jargon.

The primary means of HIV transmission is by sexual contact. We have shown that prior immunization and infection with an attenuated virus can protect animals from vaginal challenge with pathogenic SIVmac239. This project will use the SIV/rhesus macaque model of AIDS to understand the mechanism of protection in this vaccine system by comparing anti-viral immune responses after administration of replication defective specific gene or virus function deletion mutants. We hypothesize that multiple host immune factors as well as vaccine content and will determine the ultimate outcome of SIV challenge.

b) Procedures employed in this project:

Please check the appropriate boxes if any of these procedures will be employed in your project:

- Monoclonal Antibody Production
- Polyclonal Antibody Production
- LD 50 or ID50 studies.
- catheters, blood collection, intubation
- Prolonged restraint. (8 hrs+)
- Monoclonal Antibody Production
- Polyclonal Antibody Production
- LD 50 or ID50 studies.
- catheters, blood collection, intubation
- Prolonged restraint. (8 hrs+)
- Fasting prior to a procedure.
- Food or water restriction
- Non-recovery surgical procedures
- Survival surgical procedures
- Multiple survival surgery
- Special diets; food or water treatment.
- Induced illness, intoxication, or disease
- Death as an endpoint (see i below)
- Trapping, banding or marking wild animals
- Behavioral modification.
- Aversive conditioning.

** If this protocol only describes antibody production, you may use the attached antibody production page in lieu of completing section c below.
c) **Describe the use of animals in your project in detail**, with special reference to any procedures checked above. Include any physical, chemical or biological agents that may be administered. List each study group, and describe all the specific procedures that will be performed on each animal in each study group. Use terminology that will be understood by individuals outside your field of expertise. *(Note: This cell will expand to whatever length you require. You may make this section as long as you wish, but try to be concise. Some projects may require one or two pages.)*

We hypothesize that multiple host immune factors as well as vaccine content and will determine the ultimate outcome of SIV challenge. The studies outlined below will expand on these studies to address this specific hypothesis.

The term “provirus” is used below and is defined here. Retroviruses (SIV, HIV, SHIV) exists in 2 forms. The first is a discrete virus particle that exists outside a cell. The virus particle consists of an outer lipid membrane surrounding a dense protein capsid. Inside the capsid is the viral RNA, the nucleic acid that encodes all the viral genes. Once the virus particle fuses with a cell the viral RNA is “reverse-transcribed” or converted into viral DNA. This full-length DNA based version of the viral genome is termed the provirus. The provirus serves as the template for transcription of all viral mRNA and genomic RNA using host cell transcriptional machinery. The DNA plasmids that we are injecting as a vaccine in these studies contain the SHIV provirus as the expressed insert with gene or essential genetic elements removed such as to produces viral proteins without producing replication competent virus.

<table>
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<tr>
<th>0</th>
<th>8</th>
<th>16</th>
<th>28 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td>plasmid</td>
<td>plasmid</td>
<td>plasmid</td>
<td>SIVmac239</td>
</tr>
</tbody>
</table>

1) **Intradermal DNA immunization with SHIV 89.6 gp120 deletion mutant provirus will generate protective immunity against SIVmac239 challenge.** The animals will be inoculated intradermally (two sites, both on the back between the shoulder blades) with 100 ug (suspended at 1 mg/ ml in TRIS-EDTA pH 8.0 + 0.15M NaCl) of SHIV 89.6 delta gp120 proviral DNA plasmid. With the removal of the envelope gene non infectious, immunogenic particles as well as other viral proteins will be expressed from transfected cells (cells which pick up the plasmid DNA). DNA will be administered using a 27 gauge needle on a tuberculin syringe with no more than 250 ul injected into one site. Animals will be boosted using the same protocol and additional 2 times with a 1-2 month interval between immunizations (Day 0, 8 weeks, 16 weeks). Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) will be collected from femoral vein at weeks –1, 0, 1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 0,2 and 4 months post immunization to monitor immune responses. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution). The biopsy site will be rotated among the inguinal and axillary nodes. 6 months after the first immunization animals will be challenged with SIVmac239 (am/pm) intravaginally (byatraumatically inserting a 1 ml syringe with $10^5$ TCID$_{50}$ virus in 1 ml saline) and monitored for infection. Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) from femoral vein on the day of challenge, and weeks1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20 post-challenge. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 1,3 and 5 months post challenge. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution) will be obtained no more that once a month to monitor immune responses. For the vaginal challenge, animals may be sedated twice that week, otherwise animals will be sedated no more than once a week. The animals will be euthanized when they develop clinical signs of AIDS or at 12 months after SIV challenge.

2) **Intradermal DNA immunization with SHIV 89.6 integrase deletion mutant provirus will generate protective immunity against SIVmac239 challenge.** The animals will be inoculated intradermally (two sites, both on the back between the shoulder blades) with 100 ug (suspended at 1 mg/ ml in TRIS-EDTA pH 8.0 + 0.15M NaCl) of SHIV 89.6 delta integrase proviral DNA plasmid. With the removal of the integrase gene non infectious,
immunogenic particles with envelope glycoproteins as well as other viral proteins will be expressed from transfected cells (cells which pick up the plasmid DNA). DNA will be administered using a 27 gauge needle on a tuberculin syringe with no more than 250 ul injected into one site. Animals will be boosted using the same protocol and additional 2 times with a 1-2 month interval between immunizations. (Day 0, 8 weeks, 16 weeks). Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) will be collected from femoral vein at weeks –1, 0, 1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 0,2 and 4 months post immunization to monitor immune responses. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution). The biopsy site will be rotated among the inguinal and axillary nodes. 6 months after the first immunization animals will be challenged with SIVmac239 (am/pm) intravaginally (by atraumatically inserting a 1 ml syringe with 10^5 TCID_{50} virus in 1 ml saline) and monitored for infection. Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) from femoral vein on the day of challenge, and weeks1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20 post-challenge. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 1,3 and 5 months post challenge. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution) will be obtained no more that once a month to monitor immune responses. For the vaginal challenge, animals may be sedated twice that week, otherwise animals will be sedated no more than once a week. The animals will be euthanized when they develop clinical signs of AIDS or at 12 months after SIV challenge.

3) Intradermal DNA immunization with SHIV 89.6 LTR deletion mutant provirus will generate protective immunity against SIVmac239 challenge. The animals will be inoculated intradermally (two sites, both on the back between the shoulder blades) with 100 ug (suspended at 1 mg/ ml in TRIS-EDTA pH 8.0 + 0.15M NaCl) of SHIV 89.6 delta LTR proviral DNA plasmid. With the replacement of theLTRs with a CMV promoter and bovine growth hormone poly adenylation signal, non infectious, immunogenic particles with envelope glycoproteins as well as other viral proteins will be expressed from transfected cells (cells which pick up the plasmid DNA). DNA will be administered using a 27 gauge needle on a tuberculin syringe with no more than 250 ul injected into one site. Animals will be boosted using the same protocol and additional 2 times with a 1-2 month interval between immunizations. (Day 0, 8 weeks, 16 weeks). Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) will be collected from femoral vein at weeks –1, 0, 1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 0,2 and 4 months post immunization to monitor immune responses. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution). The biopsy site will be rotated among the inguinal and axillary nodes. 6 months after the first immunization animals will be challenged with SIVmac239 (am/pm) intravaginally (by atraumatically inserting a 1 ml syringe with 10^5 TCID_{50} virus in 1 ml saline) and monitored for infection. Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) from femoral vein on the day of challenge, and weeks1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20 post-challenge. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 1,3 and 5 months post challenge. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution) will be obtained no more that once a month to monitor immune responses. For the vaginal challenge, animals may be sedated twice that week, otherwise animals will be sedated no more than once a week. The animals will be euthanized when they develop clinical signs of AIDS or at 12 months after SIV challenge.

4) Intradermal DNA immunization with empty plasmid will not generate protective immunity against SIVmac239 challenge. The animals will be inoculated intradermally (two sites, both on the back between the shoulder blades) with 100 ug (suspended at 1 mg/ ml in TRIS-EDTA pH 8.0 + 0.15M NaCl) of empty plasmid DNA. DNA will be administered using a 27 gauge needle on a tuberculin syringe with no more than 250 ul injected into one site. Animals will be boosted using the same protocol and additional 2 times with a 1-2 month interval between immunizations. (Day 0, 8 weeks, 16 weeks) Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) will be collected from femoral vein at weeks –1, 0, 1 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20. Lymph node biopsies (peripheral lymph nodes: axillary or
inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 0, 2 and 4 months post immunization to monitor immune responses. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution). The biopsy site will be rotated among the inguinal and axillary nodes. 6 months after the first immunization animals will be challenged with SIVmac239 (am/pm) intravaginally (byatraumatically inserting a 1 ml syringe with $10^5$ TCID$_{50}$ virus in 1 ml saline) and monitored for infection. Blood (10-20 mls, not to exceed 12 ml/kg/month in compliance with Primate Center blood collection guidelines SOP GG-5) from femoral vein on the day of challenge, and weeks 1, 2, 4, 8, 9, 10, 12, 16, 17, 18, and 20 post-challenge. Lymph node biopsies (peripheral lymph nodes: axillary or inguinal, approx 1 gram of tissue per biopsy according to CRPRC Standard Operating Procedure) will be obtained at approximately 1, 3, and 5 months post challenge. This includes anesthetizing the animal using medetomidine hydrochloride as well as topical analgesia via subcutaneous infiltration of Lidocaine (0.1-0.2 ml of 0.25% solution) will be obtained no more than once a month to monitor immune responses. For the vaginal challenge, animals may be sedated twice that week, otherwise animals will be sedated no more than once a week. The animals will be euthanized when they develop clinical signs of AIDS or at 12 months after SIV challenge.

d) Study Groups and Numbers: Define, in the form of a table, the numbers of animals to be used in each experimental group described above. The table may be presented on a separate page as an attachment to this protocol if you prefer. The Normal format should be three columns: Study Group, Procedure, Number of animals. The number of rows should follow from the number of study groups; you may add as many rows as you require. The chart must fully account for the number of animals you intend to use under this protocol. Assign each group to an invasiveness category according to the chart below.

<table>
<thead>
<tr>
<th>Group</th>
<th>Procedures / Drugs</th>
<th>Number of Animals</th>
<th>Category</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>ID SHIV 89.6 delta gp120 / SIV Challenge</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>2</td>
<td>ID SHIV 89.6 delta integrase / SIV Challenge</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>ID SHIV 89.6 delta LTRs / SIV Challenge</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>4</td>
<td>ID empty plasmid / SIV Challenge</td>
<td>18</td>
<td>3</td>
</tr>
</tbody>
</table>

Categories of invasiveness

<table>
<thead>
<tr>
<th>Category</th>
<th>Description</th>
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</table>
| 1        | Little or no discomfort or stress  
*Examples*: domestic flocks or herds being maintained in simulated or actual commercial production management systems; the short-term and skilful restraint of animals for purposes of observation or physical examination; blood sampling; injection of material in amounts that will not cause adverse reactions by the following routes: intravenous, subcutaneous, intramuscular, intraperitoneal, or oral. |
| 2        | Minor stress or pain of short duration  
*Examples*: cannulation or catheterization of blood vessels or body cavities under anesthesia; minor surgical procedures under anesthesia, such as biopsies or laparoscopy; short periods of restraint beyond that required for simple observation or examination, but consistent with minimal distress |
| 3        | Moderate to severe distress  
*Examples*: major surgical procedures conducted under general anesthesia, with subsequent recovery; prolonged (several hours or more) periods of physical restraint; induction of behavioral stresses such as maternal deprivation |
| 4        | Severe pain near, at or above the pain tolerance threshold  
*Examples*: exposure to noxious stimuli or agents whose effects are unknown; exposure to drugs, chemicals, or infectious agents at levels that markedly impair physiological systems and which cause death, severe pain, or extreme distress; Surgical experiments which have a high degree of invasiveness. |

Further descriptions of these categories are included in the instructions following this document.
e) **Rationale for species and numbers:** How did you determine that 1) the species choice was appropriate and 2) the number of animals in each study groups was the minimum number necessary to achieve sound scientific results?

1) The SIV rhesus macaque model of HIV heterosexual transmission has become the recognized standard for studies on pathogenesis and prevention of HIV vaginal transmission. The reason for this is that SIV is closely related to HIV biologically and genetically. Dr. [Name] created this model and has published more than 50 peer-reviewed articles involving studies using this model.

2) We have decided on six monkeys per group, which will allow us to determine statistically significant differences between groups (using a student T test) using viral load post-challenge. Each group from groups 1-3 will require 6 monkeys from group 4 as controls. Therefore, we require 18 monkeys total for group 4.

f) **Surgery:** If the project involves survival surgery, where will the surgery be conducted?

<table>
<thead>
<tr>
<th>Building</th>
<th>Room</th>
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<tbody>
<tr>
<td></td>
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</table>

Who will be the surgeon?

<p>| |</p>
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</table>

h) **Neuromuscular blocking agents** can conceal inadequate anesthesia and therefore require special justification. If you are using a neuromuscular blocking agent, please complete the following:

Why do you need to use a neuromuscular blocking agent?

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What physiologic parameters are monitored during the procedure to assess adequacy of anesthesia?

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Under what circumstances will incremental doses of anesthetics-analgesics be administered?

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i) **Adverse effects:**

Describe any potential adverse effects of the experiment on the animals (such as pain, discomfort; reduced growth, fever, anemia, neurological deficits; behavioral abnormalities or other clinical symptoms of acute or chronic distress or nutritional deficiency)
Any injection, venipuncture, or biopsy has the potential to cause minor pain or discomfort, but the animals are immobilized for the procedure and should not experience pain.

Lymph node biopsies in both SIV infected and uninfected animal have potential risk of secondary infection. Animals will be observed for several days post biopsy and treated at the discretion of the CRPRC veterinary staff.

SIV infection of rhesus macaques can result in a fatal immunodeficiency and wasting syndrome. The animals will be euthanized when they experience 3 of the following: weight loss >15% in 2 weeks or >30% in 3 months; persistent hypothermia <96°F even with heat supplementation; leukopenia (total WBC <3,000); lymphopenia (lymphocytes <800); anemia (hemoglobin <10); dehydration >10%; nonresponsive to therapy for opportunistic infections; persistent anorexia (> 3 days); animal significantly obtunded. These criteria are based on CRPRC guidelines.

How will the signs listed above be ameliorated or alleviated? If signs are not to be alleviated or ameliorated by means of post-operative analgesics or other means, explain why this is necessary.

All possible efforts will be made to minimize animal pain and discomfort.

Analgesics have no effect on the proposed studies and they will be administered at the discretion of the CRPRC veterinary staff.

Note: If any unanticipated adverse effects not described above do occur during the course of the study, a complete description of those effects and the steps taken to mitigate them must be submitted to the committee as an amendment to this protocol.

Is death an endpoint in your experimental procedure?  [ ] Yes  [X] No

(Note: “Death as an endpoint” refers to acute toxicity testing, assessment of virulence of pathogens, neutralization tests for toxins, and other studies in which animals are not euthanized, but die as a direct result of the experimental manipulation). If death is an endpoint, explain why it is not possible to euthanize the animals at an earlier point in the study. If you can euthanize the animals at an earlier point, describe the clinical signs which will dictate that an animal will be euthanized.

j) Literature search for alternatives and unnecessary duplication:

Federal law specifically requires this section. You are required to conduct a literature search to determine that either 1) there are no alternative methodologies by which to conduct this class/lab, or 2) there are alternative methodologies, but these are not appropriate for your particular class/lab. “Alternative methodologies” refers to reduction, replacement, and refinement (the three R’s) of animal use, not just animal replacement. You must also show that this use of animals is not unnecessarily duplicative of other studies.

UC Davis provides on-line access to a number of databases that can be used to search for alternatives. Visit http://trc.ucdavis.edu/jawelsh/Databases_Med_Vet_Researchers.htm (email: jawelsh@ucdavis.edu)
or http://www.vetmed.ucdavis.edu/Animal_Alternatives/main.htm (email: mwwood@ucdavis.edu)

What was the date on which you conducted this search?  6/9/03

List the databases searched or other sources consulted (there should be more than one). Include the years covered by the search.

<table>
<thead>
<tr>
<th>Database Name</th>
<th>Years Covered</th>
<th>Keywords / Search Strategy</th>
</tr>
</thead>
<tbody>
<tr>
<td>PubMed</td>
<td>1990–present</td>
<td>Primate, SIV, SHIV, vaccine, vaginal transmission</td>
</tr>
<tr>
<td>Reference Update</td>
<td>1999–present</td>
<td>Primate, SIV, SHIV, vaccine, vaginal transmission</td>
</tr>
<tr>
<td>Current Contents</td>
<td>1990–present</td>
<td>Primate, SIV, SHIV, vaccine, vaginal transmission</td>
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</tbody>
</table>
What were your findings with respect to alternative methodologies?

There are no alternative methodologies for assessing genital lentiviral transmission.

Has this study been previously conducted?  [X] Yes  [ ] No

If the study has been conducted previously, explain why it is scientifically necessary to replicate the experiment.

This study has been ongoing in the laboratory, the groups listed in this protocol have not yet been tested.

k) Disposition of animals:  At what point in the study, if any, will the animals be euthanized?

Animals will be either euthanized after systemic infection is documented, or possibly recycled into a therapeutic SIV study.

l) Methods of euthanasia:  Even if your study does not involve killing the animals, you should show a method that you would use in the event of unanticipated injury or illness.  If anesthetic overdose is the method, show the agent, dose, and route.

<table>
<thead>
<tr>
<th>Species</th>
<th>Method</th>
<th>Drug</th>
<th>Dose (mg/kg)</th>
<th>route</th>
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<tbody>
<tr>
<td>rhesus</td>
<td>IV</td>
<td>pentobarbital</td>
<td>60 mg/kg</td>
<td>IV</td>
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m) Surplus animals:  What will you do with any animals not euthanized at the conclusion of the project?

Animals will be either euthanized after systemic infection is documented, or possibly recycled into a therapeutic SIV study.
**Project Roster:** Please provide the names of all the individuals who will work with animals on this project. This page will not be made available to the public. Give either the University Employee ID # or a valid UC Davis email address so that we can document training and occupational health compliance for regulatory agencies. Include all investigators, student employees, post-doctoral researchers, staff research associates, post-graduate researchers and laboratory assistants who will actually work with the animals. You don't need to include the staff of the vivarium in which your animals will be housed.

The principal investigator is responsible for keeping this roster current. If any staff is added or subtracted from this project, you must amend the protocol by sending the campus veterinarian a memo describing any changes.

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<tr>
<th>Last Name</th>
<th>First Name</th>
<th>Middle Name</th>
<th>UC ID Number or SSN</th>
<th>Email Address</th>
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**Occupational Health Program:**

Supervisors must enroll their employees in the campus Occupational Health Program if the workers are at increased risk of illness or injury (such as allergy, physical injury, or infectious disease) because of their work. Enroll workers by having them complete an "Animal Contact History Form", available from Employee Health Services (phone 752-2330). For further information, visit our web site at [http://ehs.ucdavis.edu/animal/health/](http://ehs.ucdavis.edu/animal/health/) or read the UC Davis Policy & Procedure Manual 290-25.

**Training:**

Supervisors are responsible for insuring that their employees are adequate trained, both in the specifics of their job and in the requirements of the Federal Animal Welfare Act. EH&S offers free, basic wet labs in laboratory animal handling and techniques, and lecture format classes in the requirements of the Animal Welfare Act. To schedule a class for your unit, contact EH&S at 2-2364. Information is available on the world wide web at [http://ehs.ucdavis.edu/](http://ehs.ucdavis.edu/).
Assurances for the Humane Care and Use of Vertebrate Animals:

Principal Investigator's Statement:

I have read and agree to abide by the UC Davis Policy and Procedure Manual section 290-30 (Animal Use and Care). This project will be conducted in accordance with the ILAR Guide for the Care and Use of Laboratory Animals, and the UC Davis Animal Welfare Assurance on file with the US Public Health Service. (These documents are available from the Campus Veterinarian and at http://ehs.ucdavis.edu/). I will abide by all Federal, state and local laws and regulations dealing with the use of animals in research.

I will advise the Animal Use and Care Administrative Advisory Committee in writing of any significant changes in the procedures or personnel involved in this project.

<table>
<thead>
<tr>
<th>Principal Investigator</th>
<th>Rank / Title</th>
<th>Date</th>
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**Conditions necessary for Committee Approval:**

Final Disposition of this protocol:

- _________ Approved
- _________ Not Approved
- _________ Withdrawn by Investigator

Date of Action: _____ / _____ / _____

I verify that the Institutional Animal Care and Use Committee of the University of California, Davis, acted on this protocol as shown above.

<table>
<thead>
<tr>
<th>Campus Veterinarian</th>
<th>Date</th>
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</thead>
</table>
**ANIMAL ROOM SAFETY INFORMATION**

Complete this form if you will be using biohazards, radioisotopes, carcinogens, or toxic chemicals in the animal room.

<table>
<thead>
<tr>
<th>Identity of Hazard:</th>
<th>SIV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Investigator Last Name:</td>
<td></td>
</tr>
<tr>
<td>First Name:</td>
<td></td>
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<tr>
<td>Department:</td>
<td></td>
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<tr>
<td>Phone:</td>
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<tr>
<td>Email:</td>
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<td>Fax:</td>
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</tbody>
</table>

**Provide a short description of the agent:**

SIV is a primate lentivirus that is genetically similar to HIV and causes fatal immunodeficiency (AIDS) in infected rhesus macaques. SIV can infect humans, but it is unknown whether SIV causes human disease.

**This agent / material is hazardous for:**

- [ ] Humans only
- [ ] Animals only
- [X] Humans and Animals

**The agent can be spread by:**

- [X] Blood
- [X] Saliva/nasal droplets
- [X] Other: All mucosal secretions can be contaminated.
- [ ] Feces/urine
- [ ] Does not leave animal

**Describe any human health risk associated with this agent:**

SIV can infect humans; thus, it is possible that SIV could cause fatal, AIDS like disease in humans. Infectious virus and SIV antibodies have been detected in SIV-infected humans but there have been no reports of disease in SIV infected people.

**The precautions checked below apply to this experiment:**

- [ ] The researcher or his/her technicians are responsible for the feeding and care of these animals.
- [ ] The following items must be assumed to be contaminated with hazardous material and must be handled only by the researcher or his/her technicians.
  - [ ] Cage
  - [ ] Stall
  - [ ] Water Bottle
  - [ ] Animal Carcasses
  - [ ] Bedding
  - [ ] Other:
    - [X] Cages must be autoclaved before cleaning.
    - [X] Label cages and remove label after decontamination.
    - [X] Animal carcasses must be labeled and disposed of as follows:
      - [ ] Incineration
      - [X] Biohazardous Waste Container
      - [ ] Bag and Autoclave
      - [ ] EH&S will pick-up (2-1493).
    - [ ] All contaminated waste (soiled bedding or other animal waste) must be properly labeled and disposed of as follows:
      - [ ] Incineration
      - [ ] Biohazardous Waste Container
      - [ ] Bag and Autoclave
      - [ ] EH&S will pick-up (2-1493).

**Personal Protective Equipment Required:**

- [X] The following personal protective equipment must be worn/used in the room:
  - [X] Lab Coat/Coveralls
  - [X] Shoe Covers/Booties
  - [X] Disposable Gloves
  - [X] NIOSH Certified Dust Mask
  - [X] Eye Protection/Face Shield
  - [ ] Fitted Respirator
    - Type: Plastic disposable gown/coveralls
  - [X] Other: Describe:

- [ ] Personal protective equipment must be removed before leaving the room.
- [ ] Personal protective equipment must be discarded or decontaminated at the end of the project
- [ ] Hands, arms, and face must be thoroughly washed upon leaving the room
- [ ] Full shower, including washing of hair, must be taken upon leaving the room.
- [ ] Decontaminate Room (Inform ARS area supervisor when cage and/or room can be returned to general use).

**Provide any other information needed to safely work in this room:**

Biosafety level 2+ (BSL 2+) precautions must be used at all times.
Attached is a revised version of protocol 10677 which hopefully addresses all your questions. We did consult with the SRAs responsible for obtaining lymph node biopsies and they indicated the prefer the use of Medetomidine and Atipamezole instead of Ketamine or Telazol.

At 09:29 AM 7/8/2003 -0700, wrote:
I sent these to , but I thought maybe I should have sent it to you instead. These are protocol questions from

Hi,
I have received and pre reviewed the protocol which has been assigned accession number 10677 for future reference. I have attached a copy of the protocol for ease of making revisions to the questions that follow.
For this protocol to be considered on the committee agenda of July 17th, please return your revised protocol to me on or before noon, Tuesday, July 8th.
If you have any questions, please contact me by phone or email.
Thanks in advance,

Protocol 10677 ( )
Note: In the future, one suggestion that has come up in committee meetings as a result of sorting through your previous protocols: to make it easier to revise as well as read, could you please summarize all of the similar procedures to be performed on all of the groups of animals in one paragraph and list what is different for each of the groups in separate paragraphs?
1. On page 1, under special husbandry, you have listed "none"; however, you marked yes for infectious agents. Won't the animals be housed in "infectious housing?" If so, the special husbandry requirements should have the infectious housing as a requirement.
2. In all of the groups, please provide clarification to the following:
a. You mention inoculating multiple sites, but have not listed the sites. Please list the number of sites to be inoculated at each interval.
b. There appears to be text missing after the lymph node biopsies (...)...?? in the middle of each paragraph. There is no period and it appears you intended to include additional information, but it does not appear to exist. Please clarify. This again happens near the end of each of the paragraphs when you repeat the lymph node biopsies sentence. Please clarify both of these sections.
c. What are the time points of the multiple lymph node biopsies? You mention a total of 6, but have not included the time points. Will you be taking 6 biopsies before and 6 after SIV challenge or a total of 6? Please expand to include this information.
d. How often will the animals be sedated? More than once weekly or? Please clarify.
3. In section e, it appears that the last sentence was not completed. Please expand to complete your justification.
4. In section g, you list medazolam and topical anesthetic. Have you consulted with the vet staff on the agents currently used? The vet staff is recommending ketamine or telazol. Please consult with the vet staff for agents, dose route and frequency.
5. In section i, potential adverse effects, there is no mention of infection as a result of lymph node biopsies. Since these animals will be immunocompromised, please address the potential problems with infection secondary to the lymph node biopsies.
Date: Mon, 14 Jul 2003 12:18:12 -0700
To: 
From: 
Subject: Re: Fwd: Protocol 10677

At 10:10 AM 7/14/2003 -0700, you wrote:
Question from     regarding your protocol.

Hi ,

I received the following questions from committee members regarding the protocol 10677 "Mechanisms of Protection with Primate Lentiviral Vaccines". Please include the questions when the clarification is returned. This protocol is being considered on this Thursdays committee agenda.

Thanks in advance,

Protocol 10677 ( )

1. The number of animals on page 1, page 5 section d, and page 6, section e don't match. Are they intending to use an N of 6 or 9 per group?

2. The rationale does not match the number shown. Please provide further clarification.