PROTOCOL FOR ANIMAL USE AND CARE

Handwritten forms are not accepted

CRPRC

EH&S USE ONLY

PROTOCOL #__10000_

EXPIRES: _____

	Investigator		Contact
Last Name:		Last Name:	
First:		First:	
Middle:		Middle:	
email:		email:	
Department:		Department:	
Phone / Fax:			
After hrs. #:		After hrs. #:	

Species (common names):	Number:	Source:
Rhesus Monkeys	24	CRPRC

Project Title Pulmonary Effects of Environmental Oxidant Pollutants in Adult Rhesus Monkeys

Overnight housing location:: CRPRC Day use only :

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

Procedures: Provide a one or two sentence layman's description of the procedures employed on the animals in this project. This information will help the animal care staff understand any conditions they may encounter while caring for your animals.

Adult Monkeys that have been sensitized to house dust mite allergen (HDMA) will be exposed to repeated episodes of aerosolized house dust mite allergen and/or the photochemical air pollutant, ozone, in order to determine the role that ozone plays in exacerbating allergic asthma.

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.

Animals must remain free from infectious respiratory disease.

Other instructions for animal care staff: (check applicable entries)

Sick Animals	**Dead Animals**	**Pest Control**
[X] Call Investigator	[X] Call Investigator	[ ] Call Investigator
[X] Clinician to treat	[ ] Save for Investigator	[ ] OK to use pesticides
[ ] Terminate	[ ] Bag for disposal	[X] No Pesticides in animal area
[ ] Necropsy	[ ] Necropsy	

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

Infectious Agents?	[ ] Yes [X] No	Agent(s):	
Radioisotopes?	[ ] Yes [X] No	Agent(s):	
Chemical Carcinogens?	[ ] Yes [X] No	Agent(s):	
Toxic Chemicals?	[ ] Yes [X] No	Agent(s):	

Funding source:	NIH	Previously approved?	[X] Yes [ ] No
Is the project already funded?	[X] Yes [ ] No	Previous protocol number (if any):	8502

What Veterinarian or veterinary clinic will provide care for your animals? (check one)

[ ]	Lab Animal Health Clinic (2-0514)	[X]	California Primate Research Center (2-0447)
[ ]	VMTH Large Animal Field Service (2-0292)	[ ]	Another Veterinarian

If you checked ÏAnother VeterinarianÓ, please provide:

Veterinarian:		Address:	
Day phone:			
Emergency phone:		Email:	

If your veterinarian is not affiliated with one of the three service units listed above, please contact the campus veterinarian, 2-2357 (email pctillman@ucdavis.edu) for current information about training and record keeping requirements.

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.

This series of experiments will test the overall hypothesis that oxidant stress from episodes of exposure to ozone exacerbates the development of asthma when allergen exposure occurs during the portion of the episode where acute inflammation and epithelial cell injury are most pronounced.

Immunological changes and airway responsiveness to inhaled house dust mite allergen (HDMA) and cholinergic antagonist will be evaluated periodically during the exposure regimen. Following the exposure regimen, the monkeys will be euthanized, and the lung and airway tissue will be evaluated by a multidisciplinary group of investigators. This multidisciplinary approach promotes the maximization of information obtained from the animals used.

These experiments are important to further our understanding of how environmental air pollutants exacerbate allergic asthma and are especially timely given the increased incidence of allergic asthma in young children living in polluted urban environments.

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

1) Skin Testing Protocol: Prior to inclusion in a study and following the sensitization protocol, skin testing will be conducted. Monkeys to be used in these studies will be skin test positive. Monkeys will be sedated using Ketamine hydrochloride (5-10 mg/kg, IM). Under sedation each monkey will be shaved on the lateral thorax. Three intradermal injections consisting of 0.1 ml of HDMA, histamine HCl and phosphate buffered saline (diluent) will be made into the shaved area. The injection sites will be observed for wheal development for 30 minutes. At 20 minutes post-injection, each site will be measured and the measurement will be recorded. Skin testing will be performed initially during routine animal health screening and selection for the study. It will be repeated after sensitization and near day 35 of the exposure regimen. If necessary, skin testing might be repeated near days 56 and/or 140 of the exposure regimen. Monkeys will be monitored for any adverse responses over the next 2 hours as they recover from sedation. In addition, blood (9 ml) will be obtained from each monkey from a venous site to be determined by the attending veterinarian. These samples will be drawn after induction of anesthesia and prior to skin testing and used as sample for HDMA specific IgE and IgG ELISA as well as on total IgE ELISA.

2) Allergen Sensitization and Aerosol Exposure Protocol: Monkeys will be primed with an injection of *Dermatophygoides farinae*, house dust mite extract, precipitated in aluminum hydroxide. Injections consist of 12.5 micrograms HDMA extract precipitated in 10 mg alum in 1 ml given subcutaneously (SQ) on the back of the animal and 2.5 X 10^{11} killed *Bortadella pertussis* cells in 0.25 ml by the intramuscular route (IM). At 14 days after the initial injections, each monkey will be sedated with Ketamine hydrochloride (5-10 mg/kg, IM), and 0.25 ml of 0.6 mg/ml HDMA solution will be instilled into each nostril. After two days, intranasal instillation of HDMA will be repeated. A final SQ injection of HDMA in alum will be administered 21 days following the initial injections.

Further sensitization will consist of exposure by inhalation to an aerosol of HDMA dissolved in phosphate buffered saline. The aerosol will be administered within an exposure chamber for about 2 hours per day on days 3, 4, and 5 of every 14 days for up to 11, 14 day cycles, a total period of 154 days. (See attached table describing the episodic exposure regimen.) To make this practical, we wish to expose paired monkeys in a single standard cage for monkeys less than 10 kg so that four monkeys at a time can be exposed to the allergen in a large chamber. Observation of the animals by an animal health technician, attending veterinarian and/or exposure personnel will be continuous during exposure. The concentration of HDMA protein in the chamber air will be 500 $\mu g/m^3$ or less. A small serum sample (4 ml of whole blood) will be obtained from awake animals using the arm-pull technique, with blood being drawn from a cephalic vein weekly for CBC and antibody analysis. On day 40 monkeys will be skin tested again as described above for the selection process. If monkeys are not skin test positive by day 42, each will receive another injection of HDMA antigen in alum. If needed, skin testing may be repeated on days 56 and/or 140 to establish or to confirm reactivity.

3) Ozone Exposure and Animal housing: Monkeys will be housed during the study at the Inhalation Exposure Facility at the CRPRC in specially designed inhalation exposure chambers suitable for long-term housing. Monkeys are housed as compatible pairs. The caging meets or exceeds space requirements in the ILAR Guide. On the days of ozone exposure (see attached protocol time line) monkeys will inhale selected concentrations of 0.1 to 0.5 ppm ozone for 6 or 8 hours per day for 5 days every 14 days repeated for 11 cycles. (See attached table describing the episodic exposure regimen.) At

all other times the chamber atmospheres will be filtered air. For HDMA exposure the monkeys will be moved from these chambers to an identical chamber set-up for allergen aerosol exposure as described in section 3 above.

4) Pulmonary Function Testing, Static Lung Mechanics and Aerosol Challenges to Evaluate Airway Responsiveness: Pulmonary function testing will be performed on each animal to obtain baseline values before the regimen begins; then testing will be repeated near the midpoint of exposure around day 75; and finally near the end of the regimen at about day 145; however, depending on the results, it could be necessary to assess pulmonary function up to six times during the course of the regimen. CRPRC veterinary and therapeutics staff will provide sedation and animal monitoring during all procedures. Before testing, monkeys will be sedated using Ketamine hydrochloride (5-10 mg/kg, IM). The monkeys will be anesthetized using Diprivan (0.1 to 0.2 mg/kg/min, IV) with the dose adjusted as deemed necessary by the attending veterinarian. Monkeys will be intubated with an appropriate sized cuffed endotracheal tube (4.5-5.0 mm). The maximum period needed to complete the tests described below is about two hours.

Each monkey will then be placed in a whole body plethysmograph and the intubation tube attached to a pneumatic four-way valve/pneumotachograph assembly. The monkey will spontaneously breathe through the valve/pneumotachograph to measure frequency and tidal volume. If needed supplemental O_2 will be bled into the valve to maintain adequate arterial O_2 saturation levels. The monkeyís lungs will then be inflated to 30 cm H_2O and allowed to deflate to functional residual capacity (FRC) to measure total lung capacity. The lungs will then be deflated to -10 cm H_2O to determine residual volume. The lungs will then be re-inflated to 30 cm H_2O then rapidly deflated to -10 cm H_2O to determine residual volume. The monkey will then be allowed to spontaneously breathe against a closed valve. Measurement of both mouth pressure and box pressure will make it possible to calculate quasi-static lung compliance.

After the static lung mechanics have been evaluated, the monkey will be moved to a head-out body plethysmograph (body box) and the intubation tube attached to a pneumatic four-way valve/pneumotachograph assembly. All challenges will be administered as aerosols at a set inflation pressure and breathing frequency (10.0 cm H_2O and 30.0 bpm) using a compressed air (Vortran, Inc., Miniheart Model) in series with a positive pressure ventilator (Bird Mark 7A respirator). The first aerosol challenge will be saline followed by doubling concentrations of histamine, phenyl biguanide (PBG) or methacholine with the initial concentration being 0.0625 mg/ml and ending at 64.0 mg/ml. The final concentration of aerosol to be delivered will be the concentration that doubles Raw (EC200Raw) or causes arterial O2 saturation to fall below 75%. Allergen challenge will be completed using a set concentration of house dust mite allergen (0.02 mg protein/ml) delivered at repeated five minute intervals separated by 60 second data collection periods. Allergen challenge will be terminated when airway resistance (Raw) doubles or arterial O2 saturation falls below 75%. Data will be expressed as the cumulative dose of allergen (mass concentration of allergen in mg protein/ml x tidal volume in ml x number of breaths) that doubles Raw (CDA200Raw). The CDA200Raw and EC200Raw will be determined by linear interpolation on the log-log plot of the dose response curve with the response being expressed as the percent of baseline Raw.

For the evaluation of responsiveness (see above) pulmonary mechanics will be measured using a transfer impedance method. Briefly, the monkey breathes spontaneously through the pneumotachograph (Fleisch no. 2) while the thorax of the monkey is vibrated using a pseudo-random noise waveform encompassing frequencies of 2 to 128 Hz by two speakers mounted in the walls of the head-out plethysmograph (Pulmetrics Group, Boston, MA). The small changes in flow produced at the mouth are measured along with the changes in pressure inside the plethsmograph using a Microswitch transducer (model 743PC). This

technique allows the monkey to breathe spontaneously while making pulmonary mechanics measurements at 4-second intervals. Concurrently, changes in lung compliance will be assessed by placing either a fluid or air-filled balloon catheter in the esophagus at the level of the heart to measure transpulmonary pressure. Pressure/flow signals will be collected and processed using a digital data acquisition system (PO-NE-MAH, Gould Instruments Inc) and lung compliance calculated for each dose of either allergen or histamine/PBG/methacholine.

During the evaluation of responsiveness (see above) tidal volume (VT) and breathing frequency will be recorded on a breath-by-breath basis by integrating the output of the pneumotachograph using a digital data acquisition system (PO-NE-MAH, Inc). Arterial oxygen saturation (O_2Sat%) will be recorded at the beginning and ending of each data collection period with a pulse oximeter.

5) Flexible Fiberoptic Bronchoscopy, including bronchoalveolar lavage and airway brushing: Animals will undergo at most 1-2 bronchoscopy procedures when they are already anesthetized for the pulmonary function testing described above. These procedures will add a maximum of 45 minutes to the period needed for pulmonary function testing. Procedures will follow the CRPRC SOP for bronchoscopy; CRPRC Veterinary staff will provide sedation and animal monitoring. This will allow us to gather serial information about the development of airway inflammation in these rhesus monkeys and obtain cells and lavage fluid for analysis of inflammatory mediators. Results to date have demonstrated that this is a good model for the development of asthma, likely to be very relevant to the development of disease in humans. We now have the ability to elucidate the steps in this process through bronchoscopic sampling of the lower airway. This should allow us to increase the amount of information obtained from this study without increasing the number of animals.

6) Computed Tomography Scan: Animals will be transferred to the Radiology department at the VMTH for the procedure, but will be attended to by CRPRC veterinary or therapeutics staff. This is a relatively new procedure in our studies, but we estimate that each monkey would be tested no more than three times. The animals will be sedated with a cocktail of Ketamine (5-10mg/kg) and Medetomidine (30-50ug/kg) IM. Animals will be intubated using CRPRC SOP for intubation. The monkeys will be placed into the CT scanner and then given contrast medium (Conray 400, 2ml/kg) for vascular contrast. Animals will spontaneously breathe until the time of the actual scan, then the lungs will be step-wise inflated to 5, 10, 15, and 20 cm H_2O pressure, utilizing an Ambu bag (fitted with a pop-off valve set to the desired pressure) to insure a constant volume. The airway pressure will be maintained for 30-60 seconds while the scan is completed. The scan may be repeated several times to insure high quality images. Following the procedure, the Medetomidine will be reversed with an equivalent dose of Atipamazole, also given IM.

7) Administration of Bromodeoxxyuridine (BrdU): In order to identify lung cells undergoing DNA synthesis and replication all monkeys are to receive a single pulse label of BrdU at a dose of 50 mg/kg IP one hour before sacrifice and after sedation with Ketamine (5-10mg/kg). BrdU is a detectable DNA nucleotide analog that is incorporated into the DNA of dividing cells.

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.

Group	Procedures / Drugs	Number of Animals	Category
1	NOT HDMA Sensitized/NOT exposed to HDMA Aerosol or ozone.	4	2

1S	HDMA Sensitized/NOT exposed to HDMA Aerosol or ozone.	4	2
2	HDMA Sensitized/exposed to HDMA Aerosol, but NOT exposed to ozone.	4	2
3	NOT HDMA Sensitized/exposed to ozone, but NOT exposed to HDMA aerosol.	4	2
3S	HDMA Sensitized/exposed to ozone, but NOT exposed to HDMA aerosol.	4	2
4	HDMA Sensitized/exposed to HDMA Aerosol and ozone.	4	2-3

Categories of invasiveness

Category	Description
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 skillful 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?

Though several other animal models of asthma have been used, including guinea pigs, rats and mice; they are all quite limited in that each one of these species differ greatly from humans in their immunology, airway neural control and airway morphology. We have chosen the rhesus monkey because the immunological, structural and cellular aspects of its respiratory system are very similar to those in the human. The results of our initial pilot studies established that rhesus monkeys sensitized to HDMA represent a relevant model for human asthma with respect to antigen challenge and airway responsiveness.

Like humans rhesus monkeys show a wide range of responsiveness to HDMA sensitization and challenge. We begin a regimen with four animals per group. If trends are detected in any of the multitude of parameters under investigation, a group or groups may be repeated with four additional animals. The effects seen to date are fairly profound, so that even with high variability between animals, significant results can be found comparing eight per group. The previous protocol was approved for nine per group, four groups, 36 animals total over the three year period. This number of animals was approximated using power analysis in which the mean baseline responsiveness to methylcholine of monkeys studied in our pilot work along with the calculated standard deviation and assuming a desired responsiveness shift of one order of magnitude was used.

Nine per group within three years was perhaps too ambitious for completing

the very detailed multidisciplinary evaluations performed, and to date we have completed only four per group for 16 total adult monkeys under the previous protocol. Therefore, we need to supplement four groups with four more animals each. In response to scientific reviewers comments, we must also add two groups of four that receive the initial sensitization and either filtered air only or ozone. This should be a sufficient number in each group to detect differences, should any exist, between the filtered air and ozone groups that were not initially sensitized.

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

Building: Room:

Who will be the surgeon?

g) Anesthetics, Analgesics, Tranquilizers, Neuromuscular blocking agents:

Post procedural analgesics should be given whenever there is possibility of pain or discomfort that is more than slight or momentary. If postoperative analgesics are not to be given, justify the practice under part (i) below.

Provide the following information about any of these drugs that you intend to use in this project.

Species	Drug	Dose (mg/kg)	Route	When and how often will it be given?
Rhesus Monkey	Ketamine HCl	10	IM/IV	Maintenance of sedation
Rhesus Monkey	Diprivan	0.1-0.2 mg/kg/min	IV	Adjusted as deemed necessary by attending veterinarian
Rhesus Monkey	Diazepam	0.1-1.0	IM/IV	Maintenance of sedation and muscle relaxation if needed.
Rhesus Monkey	Medetomidine	0.02-0.05	IM	Once, if deemed needed by attending veterinarian.
Rhesus Monkey	Atipamazole	0.02-0.05	IM	Once, if deemed needed by attending veterinarian.
Rhesus Monkey	Albuterol	1 mg/ml of saline	Inhaled Aerosol	If necessary as determined by attending veterinarian.

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?

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

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

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)

Ozone exposure may cause transient respiratory discomfort including substernal pain and cough. Repeated HDMA aerosol inhalation in HDMA sensitized animals may result in respiratory symptoms of bronchoconstriction, wheezing and cough.

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.

Discomfort of ozone inhalation is transient lasting 2-4 hours. If monkeys develop severe acute airway obstruction due to exposure to HDMA, the attending veterinarian will deliver an aerosol of the bronchodialator, albuterol, to the monkey.

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:

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

What was the date on which you conducted this search? 2/15/02

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

Database Name	Years Covered	Keywords / Search Strategy
Melvyl	1972 to present	asthma, air pollution; asthma, monkey
Current Contents	1990 to present	asthma, air pollution; asthma, monkey
PubMed	1986 to present; 1967 to present	asthma, air pollution, lung; asthma, monkey

What were your findings with respect to alternative methodologies?

Though several other animal models of asthma have been utilized, including guinea pigs, rats and mice, they are all limited in that each one of these species differ greatly from humans in their immunology, airway neural control and airway morphology. Whole animal models are required to study asthma because of the multiple cell populations that are involved, including inflammatory and immune cells, epithelial cells, airway smooth muscle and airway sensory and efferent nerves.

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

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

This is a continuation of a previous protocol. Also, two additional groups have been added in response to scientific reviewersí critique.

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

After defined exposure and post-exposure periods, all animals will be euthanized for detailed study of the respiratory tract.

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.

Species	Method	Drug	Dose (mg/kg)	route
Monkeys	Overdose	Sodium pentobarbital	60 mg/kg	IV

m) Surplus animals: What will you do with any animals not euthanized at the conclusion of the project?

None

n) 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.

Last Name	First Name	Middle Name	UC ID Number or SSN	Email Address

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://clueless.ucdavis.edu/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. Autotutorials are also available on the world wide web at ***http://clueless.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.

Principal Investigator	*Rank / Title*	*Date*

Committee Use Only Below

**** 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.

Campus Veterinarian	*Date*