
BILLIAR LAB NEWS

Volume 14, Issue 1

May/June/July 2012

Colony Status: As of 7/23/12

Established Colonies

iNos KO
Trifl^{PS-2}
CD14 KO
Myd88 KO*
Myd88 WT*
TLR9^{CpG1/CpG1} (formerly known as CpG1 KO)
J α 281^{-/-} (formerly known as NKT KO)
HC TLR4 KO
TLR4 flox
TLR4 KO(global)
Het TLR4KO/TLR4flox
RAGE KO
C3 KO
TLR2 KO
Caspase1KO*
DC TLR4 KO
TLR4 KO/TL9^{CpG1/CpG1}
Nalp 3 KO*
TLR4 KO/RAGE KO
Lyz Cre TLR4 KO
Adipose TLR4KO
HMGB1 flox
HC HMGB1 KO
NFkB luc
NFkB WT
TLR4 KO x TLR4 flox Small quantities
AIM2 KO shipped from Jax after genotyped
Pf4creTLR4 KO small quantities

Colonies in Development (Estimated release date)

Pf4creHMGB1flox
ATF3 KO On order/to be quarantined

Planned Double KO (Estimated release date)

TLR4 KO x TLR2 KO (Set up; no pups yet)

Strains Available from Collaborators

CAV KO (Michael Bauer)
IFNabR KO (M. Bauer)
IRF-2 KO (David Geller)
eNos GCHtg (A. Chen)
TSP-1 KO (A. Chen)
TSP-2 KO (A. Chen)
CD36 KO (A. Chen)
hph-1 (A. Chen)
GDF-15 KO (Yoram Vodovotz)

Rederivation Strains

Cryopreserved Strains

eNos tg
CD36 KO
TLR2 KO
AIM2 KO
EGR1 KO
IFNabR KO

Stud Colonies

TLR4 KO x CD11c
TLR4 KO x Albumin Cre
HMGB1 flox x Albumin Cre
TLR4 x Fab cre (for AdiposeTLR4KO)
TLR4 KO x Lyz Cre
HMGB1 flox x Pf4-cre
TLR4 KO x Pf4-Cre

To reserve mice, contact Deb Williams (williamsd8@upmc.edu). Some colonies may need adjusted to meet your needs.

Lab Announcements

Please **DO NOT TAMPER** with core equipment (Cryostat) if there is a problem with the equipment see Rick.

There are new policies to follow in rooms NW615 and NW66 for housing animals overnight—including animals that have been brought over from the facility to be used the following day or later in the week for an experiment. There are two log sheets posted on the wall by the door of 615 and on the hood in 636. Please make sure to fill these forms out properly and to note the number of cages and health of the animals. John and Lauryn will take care of doing this during the week for room 636, but if you have mice in the room over the weekend, it is your responsibility to fill out. Always make sure all animals have food, water, a cage card, and a surgery card if necessary and never leave animals that are not fully awake unattended.

In order to replace Amanda Chipman and Amanda Teeple starting in August Alicia Frank will focus on hepatocyte harvests, liver I/R, and running the IVIS and will no longer be performing hemorrhagic shock.

Please be patient with hemorrhagic shock experimental request backups over the next couple of months as a new technician is found and trained to replace those that are leaving.

Please remember to always request hepatocytes in number of dishes and plates NOT number of animals. If you would like the any extra cells from the harvests ask for them, but a target number is needed.

Email Deb with suggestions for updates to equipment and computer software.

Everyone needs to inventory your samples in the dewers. Please discard any old samples and consolidate what you are keeping. Please provide Kathy with an updated list of your samples and where they are stored for our records. Do this as soon as possible.

Environmental Health and Safety recently completed their inspection of the Montefiore lab space. Please check your email for the results.

The mandatory ceiling tile replacement in Montefiore was completed the week of July 23rd. Thank you to everyone for your co-operation and patience during this process.

Tenth Annual Department of Surgery Research Day

Please join me in congratulating the following individuals who are receiving prizes for their presentation at the Tenth Annual Richard L. Simmons lecture and Department of Surgery Research Day.

Best Oral Presentation – Matthew Neal MD

Best Oral Presentations Runner- Up – Eileen Bauer PhD and Ibrahim Nassour MD

Best Poster Presentation – Meihong Deng MD, Raymond Eid MD, and Deepa Magge MD

I want to thank all the participants for presenting their work

Best Wishes,

Timothy R. Billiar, MD

Chair, Department of Surgery

George Vance Foster Endowed Professor

Distinguished Professor of Surgery

Acute Lung Injury Conference

The Call for Abstracts will close on **August 31, 2012**.

Be sure to register for the conference also by clicking on announcement below!

You are cordially invited to attend the 2012 Pittsburgh International Lung Conference, the ninth in a series of educational conferences for increasing awareness among medical practitioners of the medical and scientific advances being made in treating lung disease. This year's conference will focus on **ACUTE LUNG INJURY: New Mechanisms, Future Therapies, and the Translation to Clinical Care** and will be held at the University Club on October 5-6, 2012 in Pittsburgh, Pennsylvania.

This exciting conference aims to provide a forum for the exchange of new and cutting-edge information about ARDS by bringing together leading regional, national, and international experts in the field.

Learn more details about the Pittsburgh International Lung Conference and view videos from the two Activity Directors: Mark T. Gladwin, MD and Rama K. Mallampalli, MD, University of Pittsburgh.

DLAR News

Avoid sending DLAR and IACUC emails to quarantine

To avoid the potential of DLAR and IACUC emails being captured as SPAM, add their email addresses to the **Approved Senders list**.

Instructions for Pitt E-Mail Users:

Log into **my.pitt.edu**

Click on "**Spam/Virus Message Center**" along the right hand side of the screen

On the top right corner of the screen click on "**My Settings**"

In the Junk Settings section click on "**Approve Senders**"

Add: **IACUC@pitt.edu** and **DLAR@pitt.edu** in the Approved Senders box

Click on "**Update Approved Senders**" to save.

Change to Procedures when weaning and separating rodents

Recently the DLAR has experienced an increase in the number of animal boxes being discovered without food and/or water after animals have been separated into new housing boxes, resulting in poor health or death. It is the responsibility of every individual working with animals to ensure food and water are provided when creating new mouse boxes, and to verify adequate food and water is present between weekly cage changes.

In an effort to maintain a healthy environment and animal welfare, the DLAR is immediately implementing the following changes which will alert our husbandry and veterinary services personnel to a newly created box, and allowing them to assess the housing environment and the health status of the animals prior to beginning other operational activities within the housing room.

1. Provide food and water to newly created mouse boxes and ensure the initial box contains adequate food and water.



2. Place a **RECENTLY WEANED** tag on the newly created mouse box.
3. Complete all columns on the room's Breeding/Weaning Log sheet, inclusive of the column indicating food and water was provided to the newly separated box.

The Recently Weaned tag and the Breeding/Weaning Log will serve as indicators to DLAR personnel, ensuring our surveillance of these animals.

Do not hesitate to contact your facility's Husbandry Supervisor if you have questions.

Change in Garbing Requirements

The DLAR, in collaboration with EH&S and approved by the IACUC, has revised the required PPE (garb) for working within the various species within this University's Animal Care and Use Program. New signage will be posted within each facility, indicating the required PPE for working with your animals.

EBST Animal Housing Room 907

Helicobacter spp.: During the 2012 first quarterly sentinel testing, designated *Helicobacter*-free room 907 tested confirmed, PCR positive for *Helicobacter hepaticus* and several other *Helicobacter* spp. Husbandry and veterinary disease management plans were developed in consultation with affected PIs. After partial depopulation (*Helicobacter* positive and suspect cages), the remaining mouse population twice tested negative for *Helicobacter* in PCR testing. Room 907 is now considered cleared of *Helicobacter* and repopulation with *Helicobacter* free mice commenced after room decontamination.

Continued on page 5 (DLAR)

IACUC News

Summer 2012 IACUC Newsletter

The Summer 2012 edition of the *IACUC Newsletter* is now available. Download the PDF version from the following link: [Summer 2012 Newsletter](#).

IACUC Policy on the Use of Analgesics in Animal Surgery

Given the varied professional and educational background of the investigative community at the University of Pittsburgh, each research investigator's approach to analgesia is equally diverse. With this in mind, a veterinary working group was created to produce a policy covering the post procedural and post operative administration of analgesics to vertebrate species used in research.

This policy was established to safeguard animal welfare with respect to pain and thereby improve the overall outcomes of research involving invasive manipulations.

As always, it must be remembered that every animal must be diligently monitored to ensure the efficacy of the analgesics used. If analgesics are not going to be used to relieve pain and discomfort, scientific justification MUST be provided in an approved IACUC protocol. Finally, when deciding on the appropriate use of analgesics, consultation with a DLAR veterinarian is strongly recommended.

A table describing surgical procedures and required analgesics can be downloaded from: <https://www.iacuc2.pitt.edu/sop/PolicyChartAnalgesicsSurgery.pdf>

Sodium Pentobarbital for Injection is Available Commercially Again

Last September, investigators were apprised that the manufacturer of Nembutal (SODIUM PENTOBARBITAL FOR INJECTION) limited the distribution of the drug to hospitals, and authorized its use solely for the treatment of epilepsy. As of December 22, 2011, Nembutal was sold to Oak Pharmaceuticals, which is again offering the drug for veterinary purposes. Nembutal can again be purchased through Butler-Schein, the supplier that University of Pittsburgh investigators must use to obtain controlled substances, in both 20 ml (043698

\$952.28) and 50 ml (043305 **\$1135.78**) volumes. Consequently, Butler-Schein should be the supplier for future orders of Nembutal.

Should an investigator require a custom formulation of Nembutal that is not commercially available, use of Heiber's Compounding Pharmacy may be permitted. In such a case, an investigator must obtain a prescription in order for the drug to be compounded by Heiber's pharmacy.

Please note that Nembutal is a schedule II controlled substance, and must be obtained, stored, dispensed, and administered in accordance with the University's controlled substance guidelines (see: <http://www.rcco.pitt.edu/ControlledDrugs/>). Further information about Nembutal can be obtained from: http://akorn.com/prod_detail.php?ndc=67386-501-52.

Contacts for more information:

University Controlled Substances
Guidelines: <http://www.rcco.pitt.edu/ControlledDrugs/>

Veterinary Consultation: 412-648-8950
dlar@pitt.edu

IACUC Office: 412-383-2008, iacuc@pitt.edu

Improvements to ARO (Animal Research Online)

Since January, University of Pittsburgh investigators have submitted all new IACUC protocols and three-year renewals through ARO (Animal Research Online). The ARO development team is continuing to improve the system, and has implemented an abbreviated modification function for making simple administrative changes to protocols. This function is called "Modification Light" (MOD LIGHT).

Protocol changes that can be made through MOD LIGHT include:

- Personnel Changes
- Protocol Administrator or Editor Changes
- Title Changes
- Emergency Contact Changes

MOD LITE allows users to make these kinds of changes on one form, without having to subsequently edit a copy of the protocol.

Continued on page 6 (IACUC)

Room 907 is once again a Helicobacter free Room
You must have a re-tour of the animal facility before you are aloud to retrieve animals from 907.

EBST Room Order:

935/931- Immunocompromised and Helicobacter Free

931/934/907- Helicobacter Free

932/904/922- Immunocompromised

930/924/925/933/920/921/923/914/913/912/911/910 /909/908/985E/906/903/902- SPF Status

905/915- Rat Housing

940-BSL2 Housing

1501A (Rats)

1501B (BSL2 Housing)

Change in Animal Fasting Requests for all Non-Rodent Species.

Effective Tuesday, July 24, 2012, a DLAR designed web-based animal fasting request form and web submission process will be activated on the DLAR website, <http://www.dlar.pitt.edu/>. This web-based fasting request is applicable to ALL species except rodents; replacing the NHP Fasting Request Form, and is required for all non-rodent fasting activities throughout the program. A fasting request is to be submitted when non-rodent species require food removal, as per IACUC protocol approval or routine standards of preoperative preparation. Please note, DLAR Veterinary Services does not recommend the fasting of rodents, and to do so, requires rodent fasting to be listed in an approved IACUC protocol.

This new one page request process is user friendly, prohibits form submission unless all fields are complete, automatically generates an email confirmation that contains the completed request form to the submitter for their records, and distributes the request to DLAR personnel in the facility were the animal is housed.

This process replaces the word document and email fasting request process. The previous email submission process will remain active for two weeks

to avoid missing a fasting request. Following this transition period, any fasting requests submitted via email will be returned to the sender, informing them to submit the request online for implementation to occur.

This communication documents the primary steps to request how all large animal preoperative or procedural fasting requests are submitted. In the event investigative staff will implement and carry out the fasting process with the animal, completion of the form is still required and serves as communication to the DLAR staff that this activity is occurring.

Instructions for Completing a Fasting Request

1. To access the Large Animal and Non-Rodent Fasting Request Form, go to the DLAR website <http://www.dlar.pitt.edu/>, log in via PI & Researcher Login using your HS Connect login user name and password, click on the Forms tab at the top of the page, and click on the Large Animal and Non-Rodent Fasting Request Form hyperlink.
2. Read the forms instructions, complete all requested information and hit submit. The form will automatically be distributed to the correct site team.
3. If lead time for fasting is less than 48 hours, please contact the Site Supervisor in addition to submitting the form.
4. If fasting will be carried out by investigative staff, indicate this information and who will be responsible in the special instructions section of the form.
5. An email including all information on the request will be sent to the individual submitting the form as confirmation that the request was submitted.
6. If investigative staff returns the animal to the cage after the procedure, they must:
 - Flip over the "NPO IN PROGRESS" sign on the housing room door,
 - Replace the feeder,
 - Remove the Do Not Feed tag from the cage,
 - Provide food to the animal if it is returned after the DLAR afternoon feeding time of 2:00PM.

Continued from Page 4 (IACUC)

To use the MOD LITE function go to your protocol's home page, then click the CREATE MOD-LITE link under Activities. Doing so will launch the MOD LITE smart form. Please read the instructions carefully, complete the first page, make any necessary changes on the second page (which is a copy of your protocol's Protocol Identification page), then submit it as you would a normal modification request. When the MOD LITE is approved, the requested changes will be integrated into your protocol.

Detailed instructions will be added to the [ARO Users Guide](#) shortly. In the meantime, if you have any questions about MOD LITE or ARO in general, contact Michael Kessler at kesslermc@upmc.edu.

Important Notes

The second page of the MOD LITE smart form will replace your protocol's Protocol Identification page upon approval, so be careful when making your changes. Be sure only to delete those who will no longer be involved with the work described in the protocol.

The MOD LITE function can only be used for simple changes to the Protocol Identification page. All other changes require the submission of a standard modification request.

IACUC Sponsored Training Opportunities

In the fall of 2011, the IACUC sponsored a two-day training webinar for University animal based research groups. Both days, seating for this webinar quickly filled to capacity and the feedback received from those in attendance indicated that the information



presented was very helpful. As this was a successful training tool, the IACUC would like to offer further training opportunities to individuals working with animals in research.

The IACUC is soliciting ideas from you and your research team for training topics you would like to see addressed.

Solicitation for Training Suggestions

- Selected topic proposals will be funded by the IACUC and made available to all animal researchers who wish to attend. Topics must be relevant to animal research and should be applicable on a programmatic level.
- Proposals for topics should include a description of the topic and of how the topic will benefit your research and the institutional program as a whole.
- Topics should be specific and include both species and procedures or methodologies to be discussed. Some topic examples are:
 - Rodent survival surgery, recovery, and post-operative care
 - Analgesia and pain management in porcine surgical models
 - Pain recognition in mice and rats
 - Managing rodent breeding colonies
 - Blood draw techniques in small animal models
 - Protocol writing, alternatives search, and procedural justifications
 - Large animal enrichment
 - Any proposal identified as having a conflict of interest will not be accepted

Proposals must be received by August 31st.

Submissions of Topics

Please send your proposals, or any questions or comments you may have, to Jeremy DeRicco at dericcojs@upmc.edu.



IMPORTANT DATES:

DR. BILLIAR'S MEETINGS – TUES 12:15, F1275 PUH

AUG 2 – MEIHONG DENG

AUG 16 – NING DING

AUG 30 – NABIL AZHAR

DR. SIMMONS' RESEARCH CONFERENCE – TUES 8:00, THUR 8:30, F1275 PUH

AUG 2 – ADRIAN MORELLI

AUG 14 – KENT ZETTEL

AUG 16 – MELANIE SCOTT

AUG 28 – DR. EDWIN DEITCH – GUEST SPEAKER

AUG 30 – RUBEN ZAMORA

DANGER SIGNALS MEETINGS – TUES 2:00PM-3:00PM, F1275 PUH

JULY 10

AUGUST 14

SEPTEMBER 11

OCTOBER 9

NOVEMBER 13

DECEMBER 11

JOURNAL CLUB – TUES 12:15, F1275 PUH

BIOHAZARDOUS WASTE PICK-UP (8:45AM-9:30AM, OUTSIDE NW617 MUH)

AUGUST 9

SEPTEMBER 20

NOVEMBER 1

DECEMBER 13

MONTEFIORE STAFF MEETING (1:00PM, N727 MUH)

AUGUST 10

New Faces

Please welcome the following new people into our labs:

Zhixia Chen (Nichole) – Visiting Scholar (Billiar)

Ramealle How – Resident (Geller)

Benjamin Kauteza – (Zuckerbraun)

Kerry-Ann McDonald – Resident (Allen Tsung)

Ankur Shukla – Resident (Tzeng)

Chris Washington – Resident (Zeng)

Juan Carlos – Summer Student (Zuckerbraun)

Tao Ma – (Billiar)

Guo-Qiang Chen – (Billiar)

Good Luck

We'd like to extend our best wishes to Marcus Hoffman, Sodam Kim, Faez Ayoob, Tiffany Lee, Ibrahim Nassour, Amanda Chipman, and Amanda Teeple. Thank you all for everything you have done for the lab and please stay in touch!

Congratulations!

Congratulations to Meihua Bo and her family on the arrival of baby boy Xiaoshu Wang on May 14th.



If you have any questions regarding anything in this newsletter, please contact Deb Williams

(williamsd8@upmc.edu)

IF YOU KNOW OF ANYTHING EXCITING OR NEWSWORTHY GOING ON IN THE LAB THAT YOU WANT TO SHARE WITH OTHERS EMAIL ALICIA FRANK (FRANKA@UPMC.EDU) WITH THE INFORMATION AND SHE'LL BE SURE TO INCLUDE IT IN THE NEXT ISSUE OF THE BILLIAR LAB NEWSLETTER.