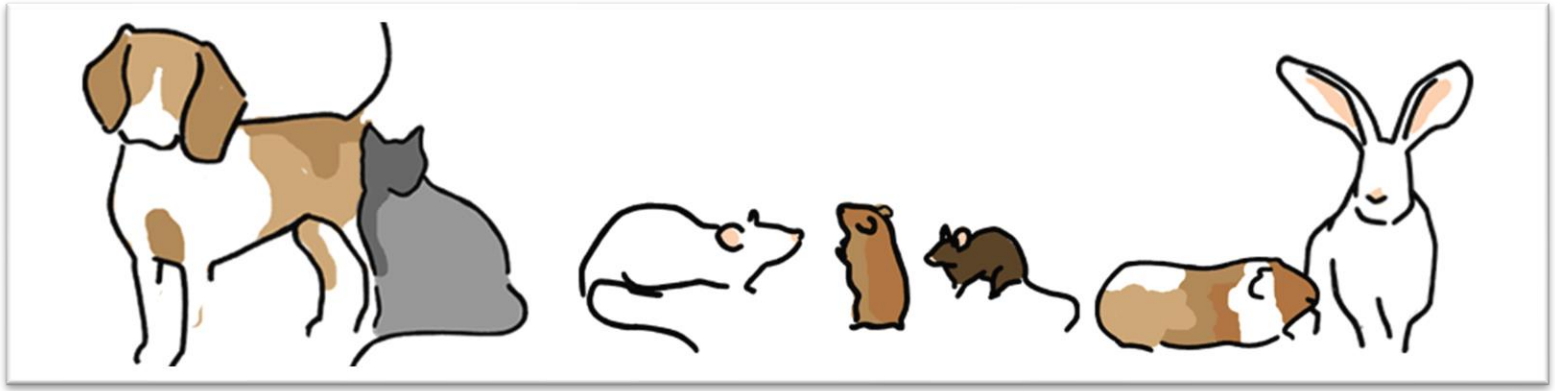
實驗動物照護及使用計畫

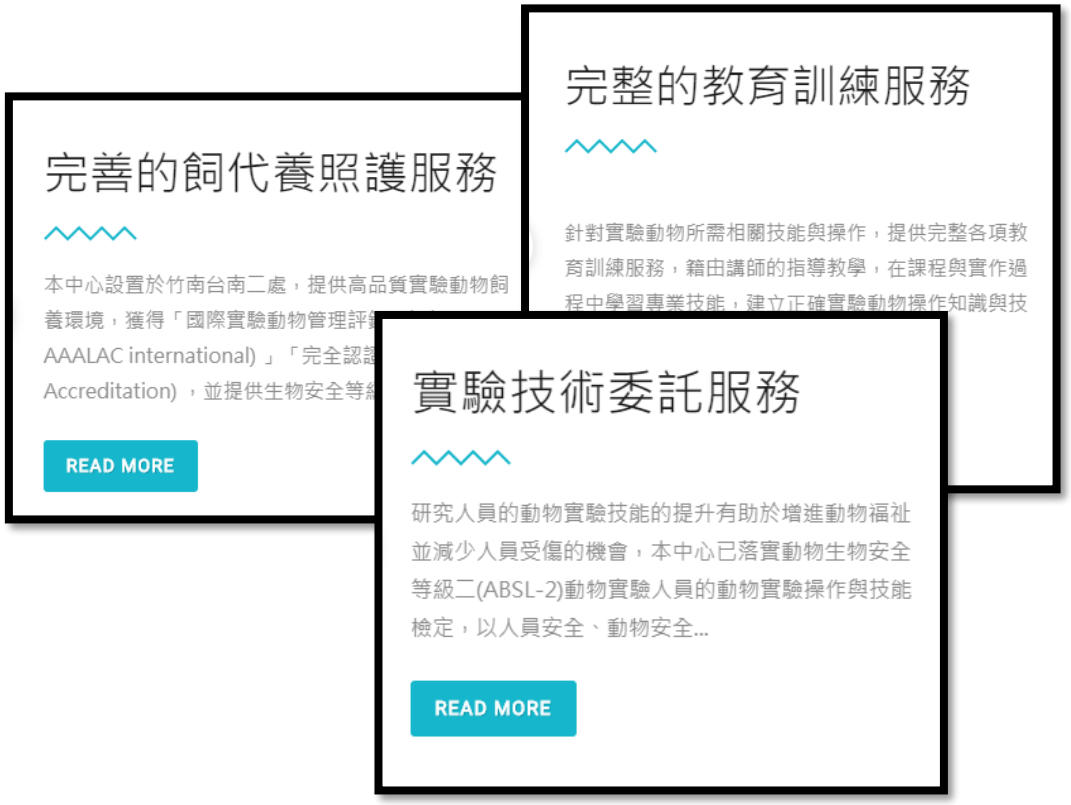
簡旭哲 獸醫師



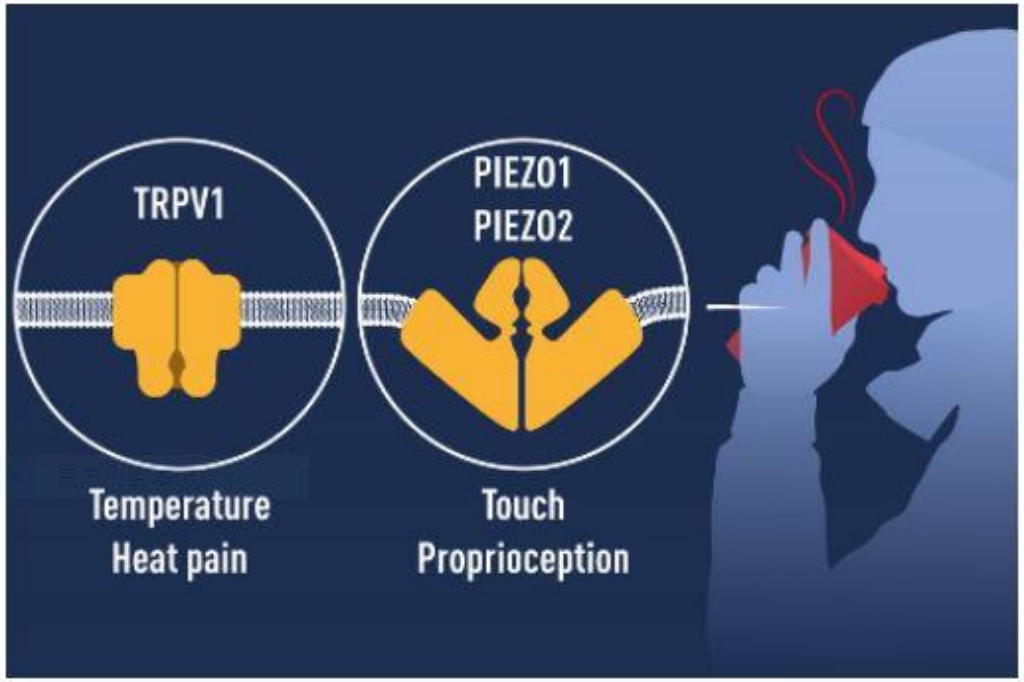
Über LAS interactive

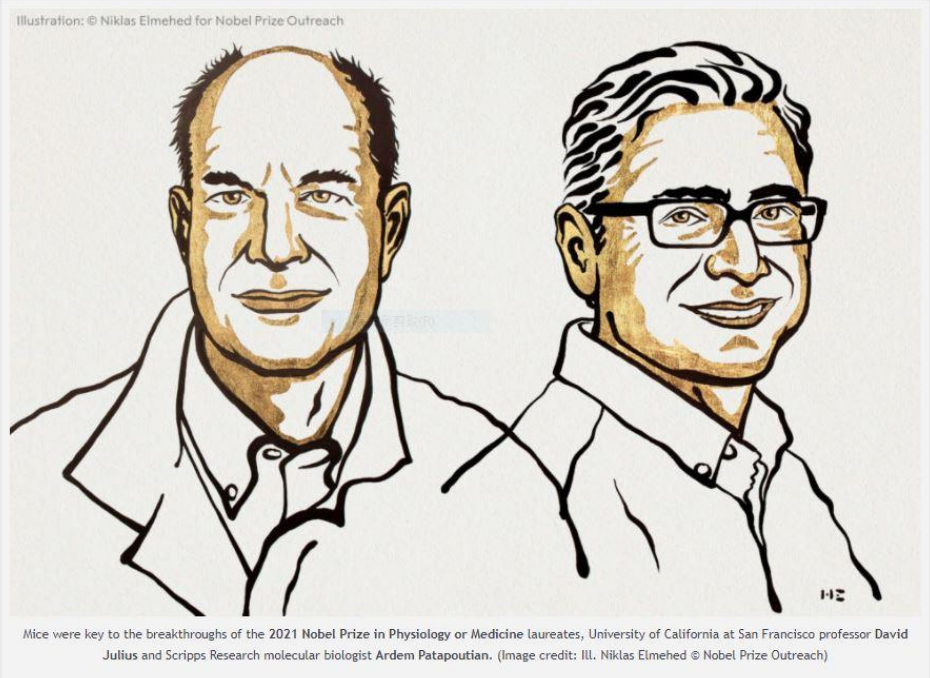
財團法人國家衛生研究院
實驗動物中心
National Health Research Institutes of Laboratory Animal Center

- **營運**：2005 ~ present
- **面積**：2066 (竹南)/ 90 (台南)坪
- **物種**：Mouse、Rat、Hamster 、Rabbit、Dog、Zebrafish
- **ABSL2**：2011正式啟用
- **AAALAC 認證**: 2015通過/2018再次合格

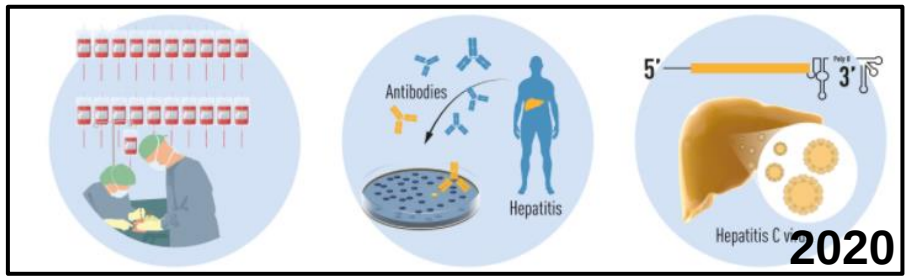


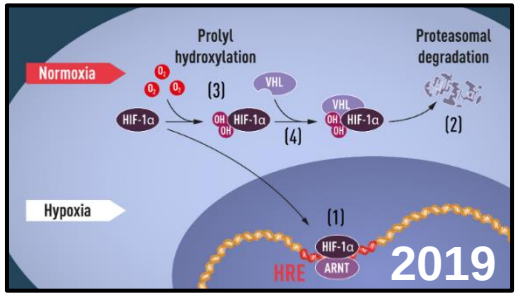
諾貝爾生理醫學獎 (2018-2021)

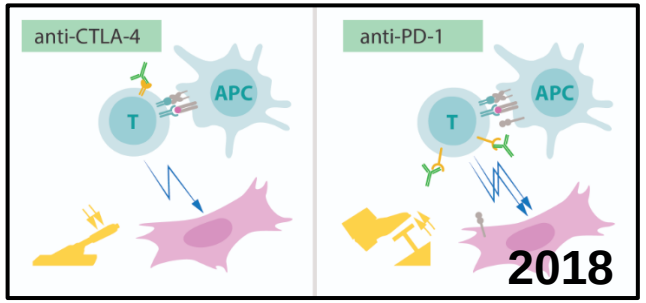




Mice were key to the breakthroughs of the **2021 Nobel Prize in Physiology or Medicine** laureates, University of California at San Francisco professor **David Julius** and Scripps Research molecular biologist **Ardem Patapoutian**. (Image credit: Ill. Niklas Elmehed © Nobel Prize Outreach)







他們的共同點是？

YEAR	NOBEL LAUREATE	ANIMAL MODEL	CONTRIBUTION TO MODERN MEDICINE
2021	David Julius and Ardem Patapoutian	Mice	For their discoveries of receptors for temperature and touch
2020	Harvey J. Alter, Michael Houghton and Charles M. Rice	Chimpanzees	For the discovery of the hepatitis C virus
2019	William G. Kaelin, Jr., MD, Gregg L. Semenza, MD, PhD, and Peter J. Ratcliffe, MD, FRS, FMedSci	Mice	Their groundbreaking research that has led to an understanding of how cells in the body adapt to changing oxygen availability.
2018	Dr. Tasuku Honjo and James P. Allison, PhD	Mice	For their discoveries in cancer therapy via inhibition of negative immune regulation
2017	Dr. Michael Rosbash, PhD, Jeffrey C. Hall, PhD, and Michael W. Young, PhD	Fruit flies	Their discoveries of molecular mechanisms controlling the circadian rhythm
2016	Dr. Yoshinori Ohsumi	Mice	
2015	William C. Campbell and Satoshi Ōmura & Youyou Tu	mice, dogs, shee cattle, chickens, monkeys	discoveries concerning a novel therapy against Malaria
2014	John O'Keefe and May-Britt & Edvard I. Moser	rats	Discoveries of cells that constitute a positioning system in the brain (an inner GPS)
2013	James E. Rothman, PhD and Thomas C. Südhof, PhD	Mice, hamsters	For their discoveries of machinery regulating vesicle traffic, a major transport system in our cells
2012	Sir John B. Gurdon	Frogs, mice	For the discovery that mature cells can be reprogrammed to become pluripotent
2012	Shinya Yamanaka	Frogs, mice	For the discovery that mature cells can be reprogrammed to become pluripotent
2011	Bruce A. Beutler	Mice	Discoveries concerning the activation of innate immunity
2011	Jules A. Hoffmann	Flies	Discoveries concerning the activation of innate immunity
2011	Ralph M. Steinman	Mice	For his discovery of the dendritic cell and its role in adaptive immunity
2010	Robert G. Edwards	Rabbits	The development of in vitro fertilization

188/224 生醫獎得主於研究中使用**實驗動物**

https://fbresearch.org/medical-advances/nobel-prizes/

大綱

I. 概論 （實驗動物照護與使用計畫）

II. 疼痛與緊迫

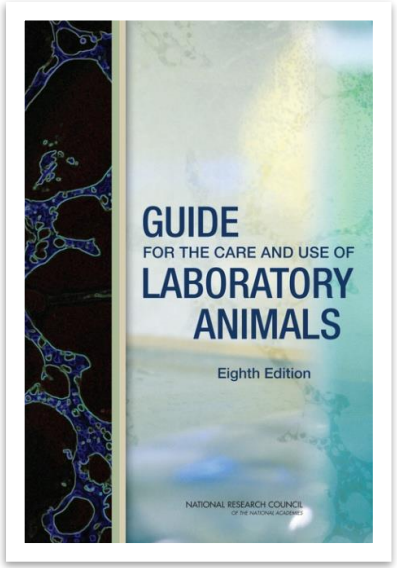
III. 安樂死

概論

實驗動物

為了研究、測試或教學而生產、使用的所有脊椎動物（例如:傳統實驗動物、農業動物、野生動物或水生物種）。

Any vertebrate animal (e.g., traditional laboratory animals, agricultural animals, wildlife, and aquatic species) produced for or used in research, testing, or teaching.

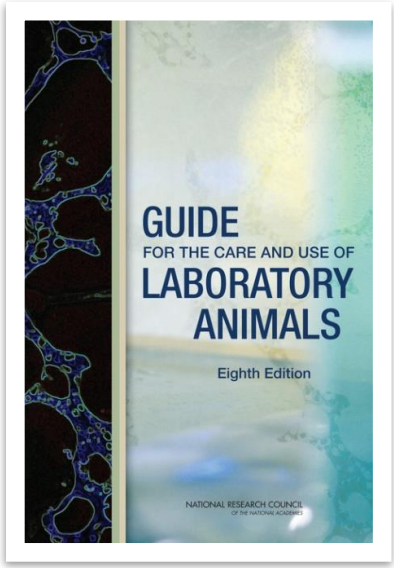


The Guide 8th, 2011

動物使用

為了研究、測試或教學目的而生產或使用的實驗動物應給予適當照護、使用與人道對待。

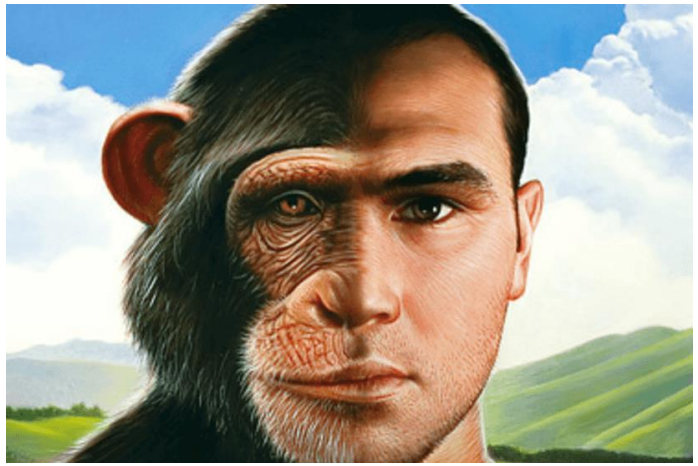
The proper care, use, and humane treatment of laboratory animals produced for or used in research, testing, or teaching.



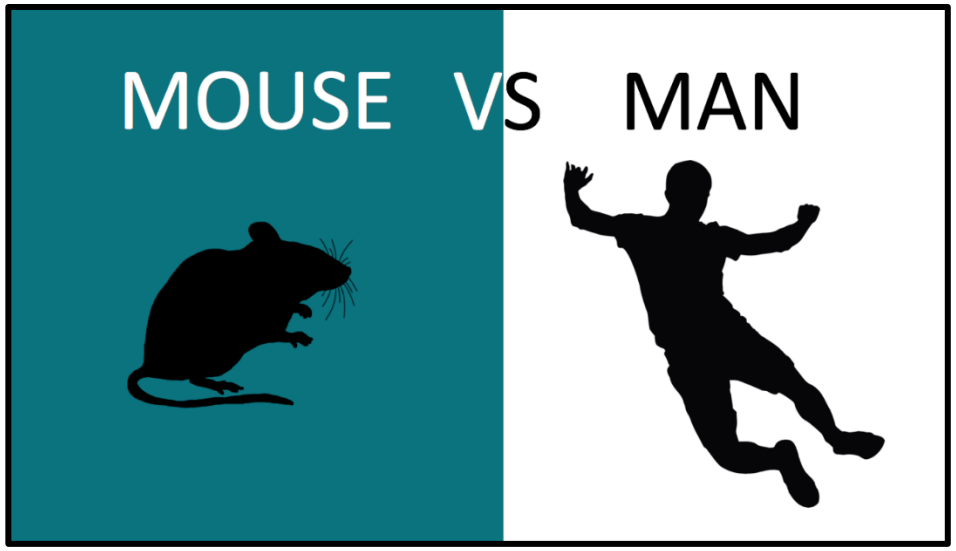
The Guide 8th, 2011

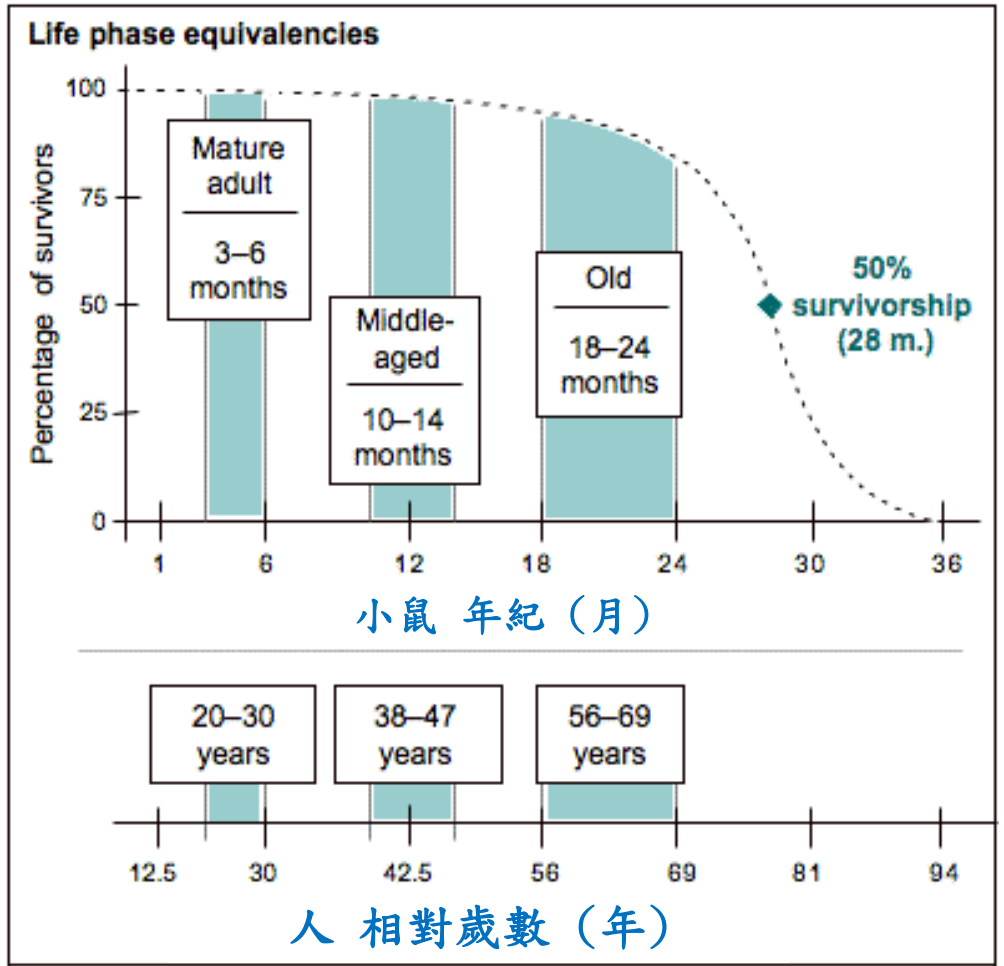
實驗動物的特性

- 特性一致/個體差異小
- 繁殖快速
- 生長快速/疾病進程短
- 模式與人類相似



生長快速/疾病進程短





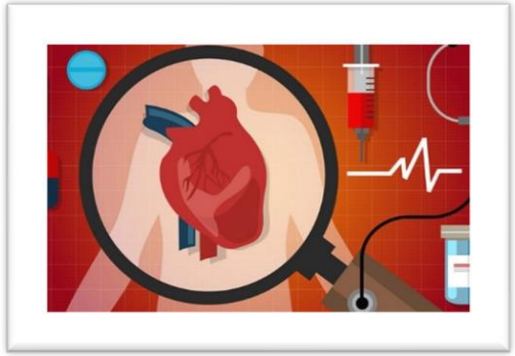
Maturational rate comparisons

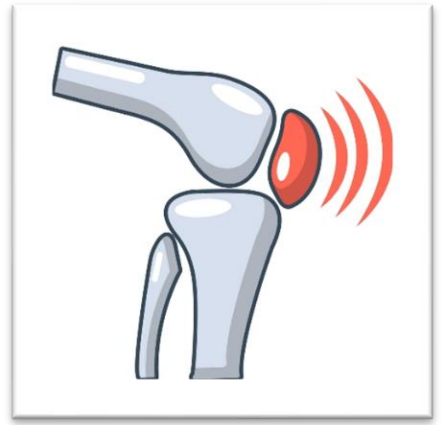
Ages	Mouse vs. human
Birth–1 month	150 times faster
1–6 months	45 times faster
6+ months	25 times faster

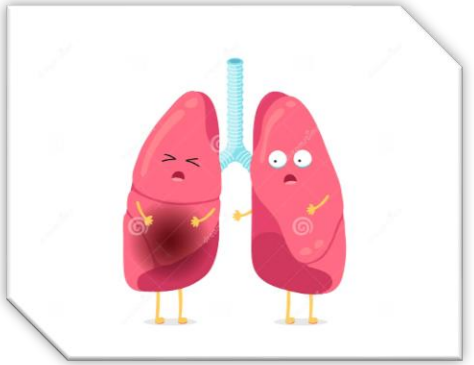
https://www.jax.org/news-and-insights/jax-blog/2017/november/when-are-mice-considered-old

動物模式是什麼?

健康　　疾病







動物模式的定義

- 動物在患有疾病的狀況下，具備與人類**相似**或**同樣**的症狀，可以作為研究人類疾病的基礎。

- 自發性/ 誘導性/ 基因改造性/ 陰性/ **孤兒型**

實驗動物照護及使用管理制度

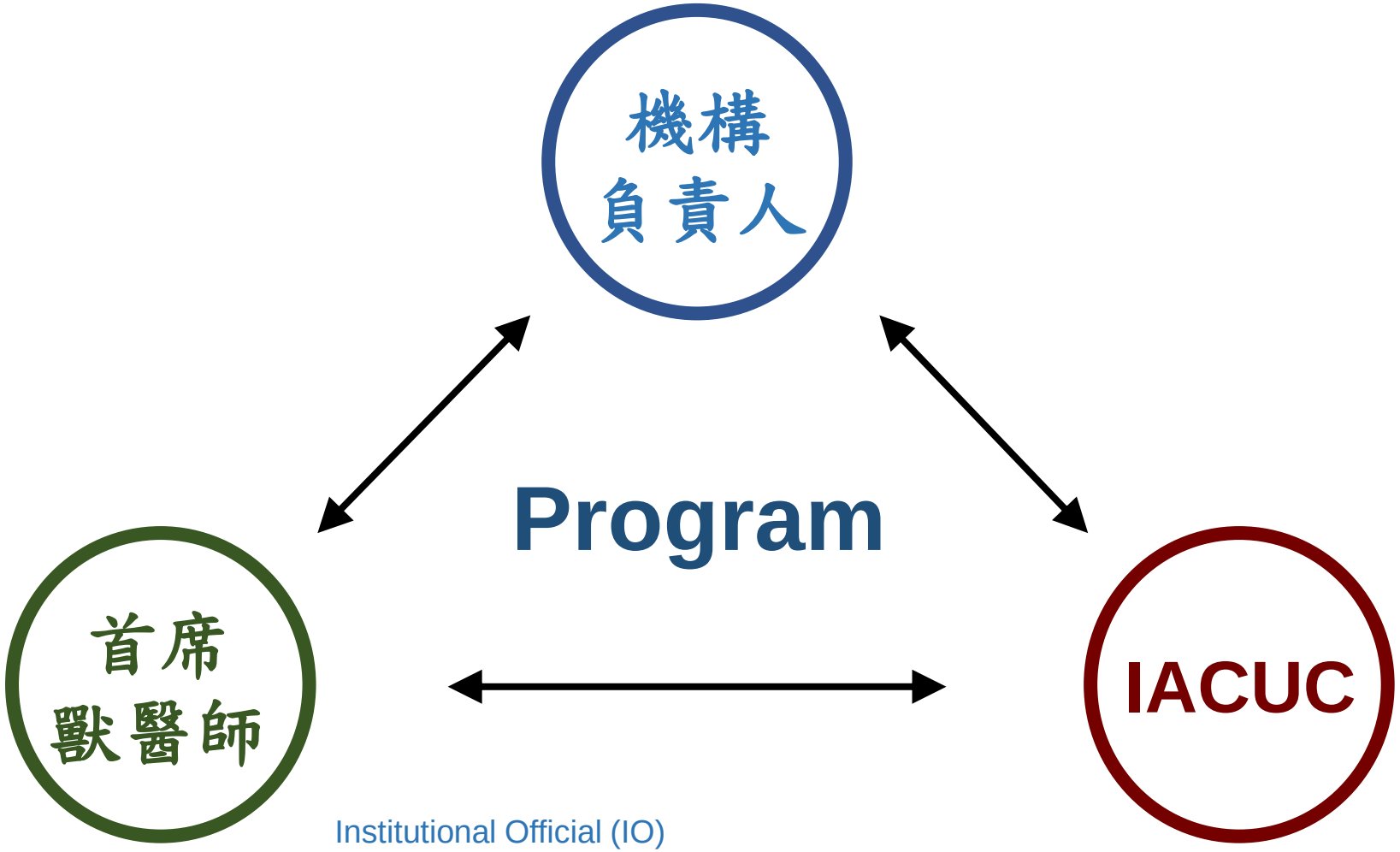
https://www.managementstudyhq.com/what-is-management.html

實驗動物照護及使用管理制度

任何在機構內進行會直接影響動物福祉的**活動/行為**，包含動物與獸醫照護、政策與程序、人員與制度的管理與監督、職業健康與安全、IACUC功能與動物設施設計與管理。

The activities conducted by and at an institution that have a direct **impact on the well-being of animals**, including animal and veterinary care, policies and procedures, personnel and program management and oversight, occupational health and safety, IACUC functions, and animal facility design and management.

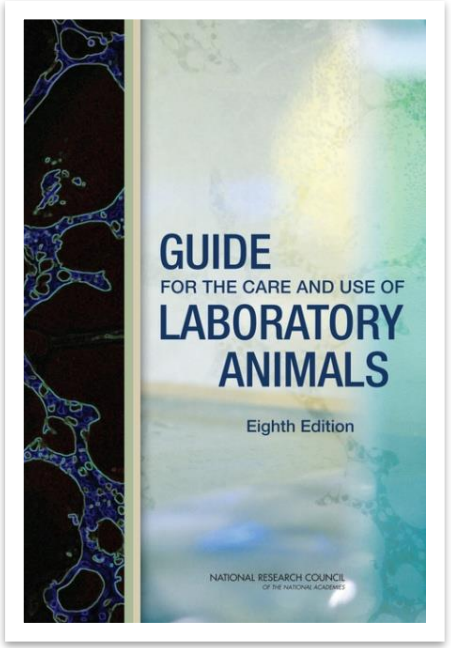
實驗動物照護及使用管理制度

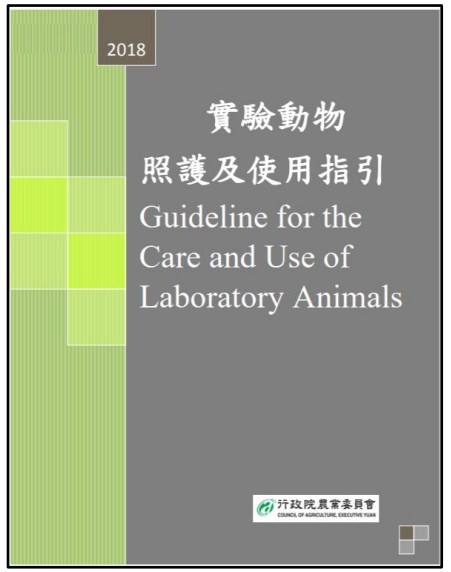


Institutional Official (IO)
Attending Veterinarian (AV)
Institutional Animal Care and Use Committee (IACUC)

規範

- The Guide 8th ed (2011)
- 農委會–實驗動物照護及使用指南(2018)
- 農委會–動物保護法 (2018)
- IACUC
- 動物設施





實驗動物照護及使用管理制度

1. 所有照顧、使用或生產動物以進行研究、測試或教學的人員，都應**對動物福祉負責**。

2. 動物科學應用機構應建立、實施及維持**實驗動物照護及使用管理制度**，並符合相關法規及動物科學應用機構實地查核的要求。

3. **機構負責人**應設置實驗動物照護及使用委員會或小組，並得指派**管理制度負責人**及**獸醫師**協助管理，但仍應承擔管理制度的最終責任。

4. 實驗動物照護及使用委員會或小組、獸醫師及相關成員應將管理制度執行需求**定期**向**機構負責人**與或**管理制度負責人報告**。

實驗動物照護及使用管理制度

5. 機構應賦予獸醫師權責及提供資源，以執行獸醫照護相關作業。若現場無法配置一位全職獸醫師，機構得有諮詢或兼職獸醫師，依據管理制度需求定期至機構巡檢。在此情況下，應指派專人負責每日動物照護使用與設施管理。

6. 所有參與管理制度的人員都應接受必要訓練訓練應有紀錄，並維持最新狀態。

7. 機構應指派專人保存管理制度執行的相關紀錄。

8. 動物房舍得依實驗動物種類設置或諮詢獸醫師、畜牧技師及水產養殖技師等專業人員，執行動物照護管理相關作業。

機構間合作計畫的管理權責

1. 參與動物科學應用**合作計畫之機構應簽署正式書面文件**，明確載明實驗動物照護及使用之權責。

2. 任一合作方的實驗動物照護及使用委員會或小組得決定是否要就合作計畫相關的申請文件進行複審。

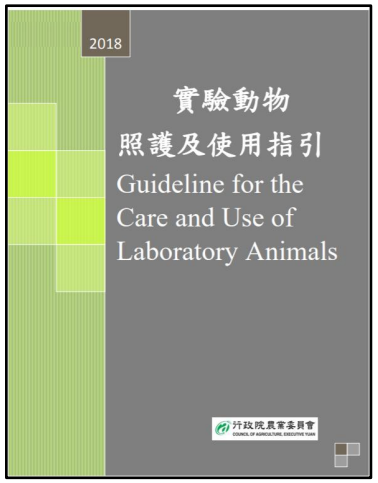
機構職責

- 人員資格與訓練
- 人員執業健康與安全
- 災害規劃與緊急應變措施

IACUC Membership

農委會指引

- 三人以上，且應包含獸醫師及外部委員各一人以上，外部委員應由非動物實驗研究背景者擔任。
- 執行秘書一人，受訓12小時以上且取得證照(有效期3年)，得由委員兼任。
- 成立IACUC於30日內核報縣市主管機關(轉中央)，異動也應申報。



IACUC

監督與定期評估制度:

1. 審核所有實驗動物計畫書

2. 確保符合所有法條、規範、政策

3. 檢視所有動物設施與實驗室

4. 監督訓練與教育計劃

機構負責人 (IO)

- 代表行政高層，對管理制度負責，具備資源規劃與確保制度目標與機構任務一致。

首席獸醫師 (AV)

- 對機構內所有動物健康與福祉負責。
- 機構應提供首席獸醫師足夠權限，包含能夠訪視所有動物與足夠資源來管理獸醫照護計畫。
- 首席獸醫師也需要監督動物照護與使用(照護、飼養)來確保計畫符合the Guide。

動保法

- 法規類別: 行政 > 行政院農業委員會 > 畜牧目
- 1998年立法，16次條文修正(5/19/2021)

第 一 章 總則

第 1 條　1　為尊重動物生命及保護動物、增進動物福利，特制定本法。

　　2　動物之保護，依本法之規定。但其他法律有特別之規定者，適用其他法律之規定。

第 2 條　1　本法所稱主管機關：在中央為行政院農業委員會；在直轄市為直轄市政府；在縣（市）為縣（市）政府。

　　2　直轄市及縣（市）政府應設機關專責動物保護，執行本法各項工作。

動保法

第 3 條 本法用詞，定義如下：

一、動物：指犬、貓及其他人為飼養或管領之脊椎動物，包括經濟動物、**實驗動物**、寵物及其他動物。
二、經濟動物：指為皮毛、肉用、乳用、役用或其他經濟目的而飼養或管領之動物。
三、**實驗動物：指為科學應用目的而飼養或管領之動物。**
四、**科學應用：指為教學訓練、科學試驗、製造生物製劑、試驗商品、藥物、毒物及移植器官等目的所進行之應用行為。**
五、寵物：指犬、貓及其他供玩賞、伴侶之目的而飼養或管領之動物。
六、寵物食品：指為供應經中央主管機關指定之寵物均衡營養之食料及其他物質。
七、**飼主：指動物之所有人或實際管領動物之人。(PI & 動物中心)**
八、寵物繁殖場：指為供商業用途而培育、改良或繁殖寵物之場所。
九、寵物食品業者：指經營寵物食品之製造、加工、分裝、批發、販賣、輸入或輸出之業者。
十、**虐待**：指除飼養、管領或處置目的之必須行為外，以**暴力、不當使用藥品、器物、不作為或其他方法**，致**傷害動物**或**使其無法維持正常生理狀態**之行為。
十一、運送人員：指以運送動物為職業者。
十二、屠宰從業人員：指於屠宰場宰殺經濟動物為職業者。
十三、展演：在公共場所或公眾得出入之場所以動物供展示、表演或與人互動。

動保法

第 5 條 *1*　動物之飼主為自然人者，以成年人為限。未成年人飼養動物者，以其法定代理人或監護人為飼主。

2　飼主對於其管領之動物，應依下列規定辦理：

一、提供適當、乾淨且無害之食物及二十四小時充足、乾淨之飲水。

二、提供安全、乾淨、通風、排水、適當及適量之遮蔽、照明與溫度之生活環境。

三、提供法定動物傳染病之必要防治。

四、避免其遭受騷擾、虐待或傷害。

五、以籠子飼養寵物者，其籠內空間應足供寵物充分伸展，並應提供充分之籠外活動時間。

六、以繩或鍊圈束寵物者，其繩或鍊應長於寵物身形且足供寵物充分伸展、活動，使用安全、舒適、透氣且保持適當鬆緊度之項圈，並應適時提供充分之戶外活動時間。

七、不得以汽、機車牽引寵物。

八、有發生危害之虞時，應將寵物移置安全處，並給予逃生之機會。

九、不得長時間將寵物留置密閉空間內，並應開啟對流孔洞供其呼吸。

十、提供其他妥善之照顧。

十一、除絕育外，不得對寵物施以非必要或不具醫療目的之手術。

飼主飼養之動物，除得交送動物收容處所或直轄市、縣（市）主管機關指定之場所收容處理外，不得棄養。

動保法

第 6 條 任何人不得騷擾、虐待或傷害動物。

第 9 條 *1* 運送動物應注意其食物、飲水、排泄、環境及安全，並避免動物遭受驚嚇、痛苦或傷害。

2 經中央主管機關公告之動物種類，其運送人員應經運送職前講習結業，取得證書，始得執行運送業務。

3 前項運送人員經運送職前講習結業並執行業務後，每二年應接受一次在職講習；其運送人員講習、動物運送工具、方式及其他應遵行事項之辦法，由中央主管機關定之。

第 11 條 *1* 飼主對於受傷或罹病之動物，應給與必要之醫療。

***2* 動物之醫療及手術，應基於動物健康或管理上需要，由獸醫師施行。**
但因緊急狀況或基於科學應用之目的或其他經中央主管機關公告之情形者，不在此限。

動保法

第 12 條 *1* 對動物不得任意宰殺。但有下列情事之一者，不在此限：

一、為肉用、皮毛用，或餵飼其他動物之經濟利用目的。

二、為科學應用目的。

三、為控制動物群體疾病或品種改良之目的。

四、為控制經濟動物數量過賸，並經主管機關許可。

五、為解除動物傷病之痛苦。

六、為避免對人類生命、身體、健康、自由、財產或公共安全有立即危險。

七、收容於動物收容處所或直轄市、縣（市）主管機關指定之場所，經獸醫師檢查患有法定傳染病、重病無法治癒、嚴重影響環境衛生之動物或其他緊急狀況，嚴重影響人畜健康或公共安全。

八、其他依本法規定或經中央主管機關公告之事由。中央主管機關得公告禁止宰殺前項第一款之動物。

任何人不得因第一項第一款所定事由，有下列行為之一：

一、宰殺犬、貓或販賣、購買、食用或持有其屠體、內臟或含有其成分之食品。

二、販賣經中央主管機關公告禁止宰殺動物之屠體。

依第十四條第二項規定准許認領、認養之動物，不包括依第八條公告禁止飼養或輸入之動物。但公告前已飼養或輸入，並依第三十六條第一項辦理登記者，准由原飼主認領。

本法中華民國一百零四年一月二十三日修正之條文施行之日起二年內，收容於動物收容處所或直轄市、縣（市）主管機關指定之場所，經通知或公告超過十二日而無人認領、認養或適當處置之動物，得予以宰殺，不適用第一項規定。

動保法

第 13 條 *1* 依前條第一項所定事由宰殺動物時，應以使動物產生最少痛苦之人道方式為之，並遵行下列規定：

一、除主管機關公告之情況外，不得於公共場所或公眾得出入之場所宰殺動物。

二、為解除寵物傷病之痛苦而宰殺寵物，除緊急情況外，應由獸醫師執行之。

三、宰殺收容於動物收容處所或直轄市、縣（市）主管機關指定場所之動物，應由獸醫師或在獸醫師監督下執行之。

四、宰殺數量過賸之動物，應依主管機關許可之方式為之。

中央主管機關得依實際需要，訂定以人道方式宰殺動物之準則。

經濟動物之屠宰從業人員，每年應接受主管機關辦理或委託辦理之人道屠宰作業講習。

動保法

第 三 章 動物之科學應用

第 15 條 *1* 使用動物進行科學應用，應儘量避免使用活體動物，有使用之必要時，應以最少數目為之，並以使動物產生最少痛苦及傷害之方式為之。（3Rs）

2 中央主管機關得依動物之種類，訂定實驗動物之來源、適用範圍及管理辦法。

第 16 條 *1* 進行動物科學應用之機構，應設置實驗動物照護及使用委員會或小組，以督導該機構進行實驗動物之科學應用。
(IACUC)

2 中央主管機關應遴聘學者、專家、相關機關及立案之民間動物保護團體代表定期監督及管理動物之科學應用；其中至少應含獸醫師及民間動物保護團體代表各一人。

3 實驗動物照護及使用委員會或小組之組成、任務及管理之辦法，由中央主管機關定之。

動保法

第 三 章 動物之科學應用

第 17 條 *1* 科學應用後，應立即檢視實驗動物之狀況，如其已失去部分肢體器官或仍持續承受痛苦，而足以影響其生存品質者，應立即以產生最少痛苦之方式宰殺之。(**Humane endpoints/Euthanasia**)

2 實驗動物經科學應用後，除有科學應用上之需要，應待其完全恢復生理功能後，始得再進行科學應用。(**Reuse**)

第 18 條 高級中等以下學校不得進行主管教育行政機關所定課程綱要以外，足以使動物受傷害或死亡之教學訓練。

動保法

第 五 章 行政監督

第 23 條 *1* 直轄市、縣（市）主管機關應置專任動物保護檢查員，並得甄選義務動物保護員，協助動物保護檢查工作。

2 動物保護檢查員得出入動物比賽、宰殺、繁殖、買賣、寄養、展示及其他營業場所、訓練、**動物科學應用場所**，稽查、取締違反本法規定之有關事項。

3 對於前項稽查、取締，不得規避、妨礙或拒絕。

4 第二項之稽查，直轄市、縣（市）主管機關得委任、委託或委辦其他機關（構），法人、團體或個人辦理。

5 動物保護檢查員於執行職務時，應出示有關執行職務之證明文件或顯示足資辨別之標誌；必要時，得請警察人員協助。

6 直轄市、縣（市）政府警察局協助動物保護檢查員執行本法有關動物保護之工作，應經相關專業訓練。

7 為期本法之有效實施，主管機關應逐年編列預算，推動流浪犬族群控制、多元創新性認領養、工作犬、校園犬計畫及確保收容管理品質等動物保護有關工作。

罰則

第 六 章 罰則

第 25 條 有下列情事之一者，處二年以下有期徒刑或拘役，併科新臺幣二十萬元以上二百萬元以下罰金：

一、違反第五條第二項、第六條或第十二條第一項規定，宰殺、故意傷害或使動物遭受傷害，致動物肢體嚴重殘缺或重要器官功能喪失。

二、違反第十二條第二項或第三項第一款規定，宰殺犬、貓或經中央主管機關公告禁止宰殺之動物。

項、第二項或第
動物死亡情節重
新臺幣五十萬元

第 六 章 罰則

第 27 條 有下列情事之一者，處新臺幣五萬元以上二十五萬元以下罰鍰，並得公布其姓名、照片及違法事實，或限期令其改善；經限期令其改善，屆期未改善者，得按次處罰之：

一、違反第十條第一款規定，驅使動物之間或人與動物搏鬥。
二、違反第十條第一款規定，與動物搏鬥。
三、違反第十條第二款規定，以直接、間接賭博為目的，利用動物進行競技。
四、違反第十條第三款規定，以直接、間接賭博或其他不當目的，進行動物交換與贈與。
五、違反第十條第六款規定，其他有害社會善良風俗之利用動物行為。
六、違反第十二條第三項規定，販賣、購買、食用或持有犬、貓之屠體、內臟或含有其成分之食品或經中央主管機關公告禁止宰殺動物之屠體。
七、寵物繁殖業者違反中央主管機關依第二十二條第二項所定辦法中有關寵物繁殖作業之規定。
八、違反第二十二條第三項規定，未為寵物絕育且未申報及提出繁殖管理說明，或未申報繁殖需求而繁殖寵物。
九、製造、加工、分裝、批發、販賣、輸入、輸出、贈與或意圖販賣而公開陳列有第二十二條之四第一項第一款或第二款情形之一之寵物食品。
十、違反第二十三條之二規定，未於直轄市或縣（市）主管機關所定期限內回收、銷毀或為其他適當處置。

第 29 條 1 有下列情事之一者，處新臺幣三萬元以上十五萬元以下罰鍰：

三、違反第十五條第一項、第十七條或第十八條規定，未依第二十四條規定限期改善或為必要之處置。
四、違反第十六條第一項規定，未成立實驗動物照護及使用委員會或小組。
六、違反第二十三條第三項規定，規避、妨礙或拒絕動物保護檢查員依法執行職務。
第二款或第三款所涉動物，不問屬於何人所有，直轄市或縣（市）主管機關得沒入之。

十五之一:使用動物進行科學應用，應儘量避免使用活體動物，有使用之必要時，應以最少數目為之，並以使動物產生最少痛苦及傷害之方式為之。中央主管機關得依動物之種類，訂定實驗動物之來源、適用範圍及管理辦法。

十七:科學應用後，應立即檢視實驗動物之狀況，如其已失去部分肢體器官或仍持續承受痛苦，而足以影響其生活品質者，應立即以產生最少痛苦之方式宰殺之。實驗動物經科學應用後，除有科學應用上之需要，應待其完全恢復生理功能後，始得再進行科學應用。

二十四:直轄市或縣（市）主管機關對於違反第十五條、第十六條第一項、第十七條或第十八條規定之機構、學校應先通知限期改善或為必要之處置。

第 30 條 1 有下列情事之一者，處新臺幣一萬五千元以上七萬五千元以下罰鍰：

一、違反第五條第二項第一款至第十款各款之一或第六條規定，故意傷害或使動物遭受傷害，而未達動物肢體嚴重殘缺、重要器官功能喪失或死亡，或過失傷害或使動物遭受傷害，致動物肢體嚴重殘缺、重要器官功能喪失或死亡。
三、違反第十一條第一項規定，對於受傷或罹病動物，飼主未給與必要之醫療，經直轄市或縣（市）主管機關通知限期改善，屆期未改善。
違反前項第一款至第八款規定之一，經裁罰處分送達之日起，**五年內故意再次違反**前項第一款至第八款規定之一者，**處二年以下有期徒刑。**

第 30-1 條 有下列情事之一者，處新臺幣三千元以上一萬五千元以下罰鍰，並得按次處罰之：
一、違反第五條第二項第一款至第十款規定，未達動物受傷狀況，經限期令其改善，屆期仍未改善。
二、違反第五條第二項第一款至第十款及第六條規定，過失傷害或使動物遭受傷害，而未達動物肢體嚴重殘缺、重要器官功能喪失或死亡。
三、違反第二十二條第四項，不提供其特定寵物飼養現況及受轉讓飼主資料，經限期令其改善，屆期仍未改善。

第 32 條 1 有下列情事之一者，直轄市或縣（市）主管機關得逕行沒入飼主之動物：

一、飼主違反第五條第二項規定，使其飼養之動物遭受惡意或無故之騷擾、虐待或傷害，情節重大且有致死之虞。
二、違反第五條第三項規定經飼主棄養之動物。
三、違反第七條規定，無故侵害他人之生命或身體，致造成他人生命或身

關得禁止其認養自直
，及不許可其申請經

第 33 條 1 有下列情事之一者，除依本法處罰外，直轄市或縣（市）主管機關應令飼主限期改善；屆期未改善者，得逕行沒入其動物：

一、違反第五條第二項規定，使動物遭受惡意或無故之騷擾、虐待或傷害。
二、違反第十條規定，利用動物。
三、違反第十一條第一項規定，未給與動物必要之醫療。
四、違反第二十條第二項規定，使具攻擊性寵物無成年人伴同或未採取適當防護措施，出入於公共場所或公眾得出入之場所。
2 違反前項各款規定之飼主，直轄市、縣（市）主管機關得禁止其認養自直轄市、縣（市）主管機關管轄之動物收容處之動物，及不許可其申請經營寵物繁殖、買賣或寄養。

第 33-2 條 直轄市或縣（市）主管機關就違反第二十五條至前條規定者，應按季彙整並陳報中央主管機關。
中央主管機關應匯總前項資料，按季提供各直轄市或縣（市）主管機關及動物收容處所，以作為拒絕或同意認養，或依前條第二項規定處罰之依據。
因檢舉而查獲違反本法行為者，直轄市、縣（市）主管機關對於檢舉人身分及有關資料應予保密，並得酌予獎勵。
前項檢舉獎勵辦法，由中央主管機關定之。

動物保護法

第 六 章 罰則

第 34 條 本法所定之罰鍰，由直轄市或縣（市）主管機關處罰之。

第 35 條 依本法所處之罰鍰，經限期繳納，逾期仍不繳納者，移送法院強制執行。

罰則 summary:

- 罰款最多５００萬
- 有期徒刑最高５年
- 沒入動物
- 重大違反，主管機關得公告其姓名、照片、違法事實

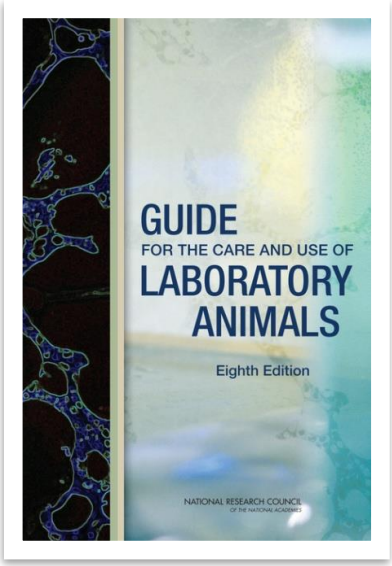
疼痛與緊迫

- 動物福祉 – 歷史/定義
- 那些是疼痛與緊迫的生理表徵?

道德與動物使用

於研究中使用動物是一種社會授予研究團體的特權，期待能夠提供重要的新知或能改善人類/動物福祉。

(McCarthy, 1999; Perry, 2007)



The Guide 8th, 2011

Russell and Burch, 1959

3Rs

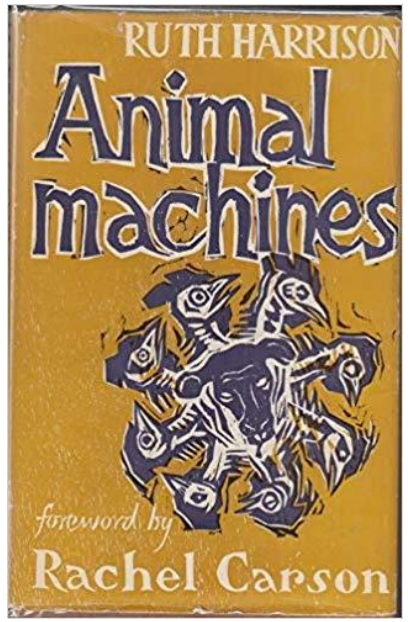
精緻化

減量

替代

五種自由

- 免於飢渴的自由
- 免於不舒服的自由
- 免於疼痛、受傷與疾病的自由
- 能表現正常行為的自由
- 免於恐懼與緊迫的自由

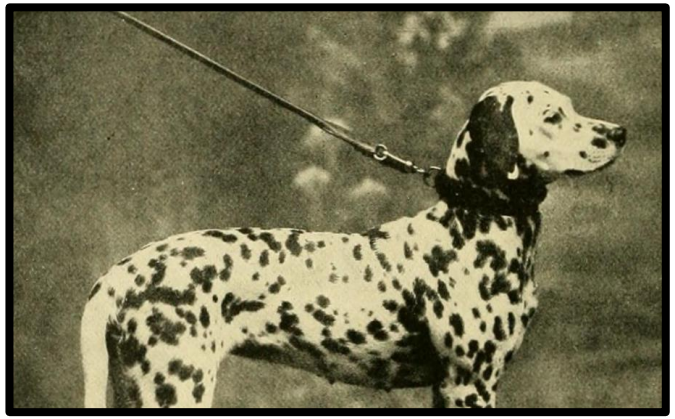


Ruth Harrison, 1964

The Brambell Report, 1965

美國動物保護法

1966



THE LOST PETS THAT STRAY TO THE LABS

Science's need for experimental animals is very real but is often filled by unscrupulous and cruel professional dognappers *by COLES PHINIZY*

It is fence-mending time on Capitol Hill now, and the halls of Congress are deserted—except perhaps for the ghost of a dog named Pepper. Late last June, Pepper, who was a 5-year-old Dalmatian bitch of affectionate disposition, disappeared from the 80-acre farm of Mr. and Mrs. Peter Lakavage in Slatington, Pa. Nine days later she turned up at New York City's Montefiore Hospital, where her body was used in a scientific experiment and then cremated. Because of her untimely end, her ghost soon after appeared to haunt the U.S. House of Representatives in the form of a bill (H.R. No. 9743) that would require anyone dealing in dogs to be licensed by the Federal Government and to keep records of all his transactions.

Whether or not the martyred Pepper will succeed in making a federal case out of dognapping is up to the men who make our nation's laws, but there are two things that the legislative investigation of her death and disappearance have made quite clear: 1) many pet dogs are being stolen from the front lawns and sidewalks of this country, and 2) the thefts in large part are motivated by science's constant and growing need for laboratory animals.

How did Pepper, the prime mover of the pending legislation, get from a Pennsylvania farm to a New York hospital? No one knows exactly, but this is certain: Pepper was last seen by her owner late on the evening of June 22. Like most family dogs, she had too much faith in people. She wagged her tail at strangers, and she liked to ride in automobiles. Probably in the early hours of the 23rd a dog thief simply stopped his car on the road in front of the Lakavage house, opened the door, invited

continued

https://awahistory.nal.usda.gov/node/119

疼痛 & 緊迫

USDA United States Department of Agriculture
Animal and Plant Health Inspection Service

1971

POLICY #11—PAINFUL/DISTRESSFUL PROCEDURES—APRIL 14, 1997

https://www.ncbi.nlm.nih.gov/books/NBK99537/

分類B:沒有疼痛/緊迫
分類C:極小的、短暫的或是沒有疼痛/緊迫
分類D:輕微到嚴重疼痛/緊迫，但可以給予藥物或其它方式緩解
分類E:嚴重疼痛/緊迫且無法以藥物或其它方式緩解

疼痛

一種感官與情緒上不舒服的體驗，與實際或可能的組織傷害有關，或是任何類似的傷害。

International Association for the Study of Pain
IASP
Working together for pain relief

An unpleasant sensory and emotional experience associated with actual or potential **tissue damage**, or described in terms of such damage.

緊迫

一種厭惡的狀態，當多種壓力出現時動物無法應付或調適。

The Guide 8th, 2011

An aversive state in which **an animal fails to cope or adjust** to various stressors with which it is presented.

UCSD

Category C	Category D	Category E
Animals that undergo procedures that cause no pain or distress, or only momentary or slight pain or distress. These procedures DO NOT require the use of pain-relieving drugs.	Animals that undergo procedures that are potentially painful or distressful; AND for which they receive appropriate anesthetics, analgesics and/or tranquilizer drugs.	Animals that undergo procedures that are potentially painful or distressful; AND for which they DO NOT receive anesthetics, analgesics and/or tranquilizer drugs.
Examples	**Examples**	**Examples**
• Holding or weighing animals in teaching, outreach or research activities • Observation of animal behavior • Ear punching of rodents • Tail snips in mice ≤ 21 days old • Peripheral injections and blood collection • Feed studies, which do not result in clinical health problems • Live trapping • Positive reward training or research • Chemical restraint • Exposure to alterations in environmental conditions (not extreme) with appropriate conditioning and microenvironment • Food restriction that reduces the animals weight by no more than 20% of normal age matched controls • AVMA approved euthanasia procedures • Euthanasia of breeding animals or unused offspring • Exsanguination under anesthesia • Transcardial Perfusion • Unknown genetically engineered phenotype • Non-invasive, routine, field procedures (capture, tissue sampling, marking, etc.) Note: Includes marine tissue collection. •Subcutaneous tumors within the size allowed by the Neoplasia Policy (Policy 9.03)	• Survival surgery • Non-survival surgical procedures • Laparoscopy or needle biopsies • Retro-orbital blood collection • Exposure of blood vessels for catheter implantation • Induced infections • Tattooing • Tail snips in mice > 21 days old • Genetically engineered phenotype that causes pain or distress that will be alleviated	• Toxicological or microbiological testing, cancer research or infectious disease research that requires continuation after clinical symptoms are evident without medical relief or require death as an endpoint • Ocular / skin irritancy testing • Application of painful stimuli such as electrical shock that the animal cannot avoid/escape • Any procedures for which needed analgesics, tranquilizers, sedatives, or anesthetics must be withheld for justifiable study purposes • Exposure to extreme environmental conditions • Euthanasia by procedures not approved by the AVMA • Paralysis or immobilization of a conscious animal • Genetically engineered phenotype that causes pain or distress that will not be alleviated •Experimental autoimmune encephalomyelitis (EAE) •Adjuvant arthritis

疼痛及緊迫分類	動物操作	臨床症狀
B.不引起不適或緊迫	僅單純養於人為的飼育環境，無實驗進行	無不良反應
C.極小的不適或緊迫，不需用藥緩解	1.注射(靜脈.皮下.肌肉.腹腔)、口服 2.採血（不包含眼窩採血等動物需鎮靜之方法） 3.短時間禁食或禁水 4.完整的麻醉 5.被核准的安樂死方法	無不良反應
D1.短時間的輕微緊迫或疼痛，需給予適當的藥物緩解	1.麻醉中插管 2.全身麻醉下進行次要存活性手術 3.全身麻醉下進行非存活性手術 4.暴露於不致命性的藥物或化學物下，未對動物造成顯著的物理性變化	動物應無自殘、食慾不振、脫水及過動現象，但休息或睡眠時間增加，喊叫次數增加，攻擊性/防禦性行為增加，或社會化行為退縮及自我孤立
D2.中等至嚴重程度的緊迫或疼痛，需給予適當的藥物緩解	1.在全身麻醉下進行主要存活性手術 2.長時間的物理性保定 3.誘導行為上的緊迫，如：剝奪母親照顧、侵略性行為、掠奪者/誘餌之相互作用 4.誘導解剖學或物理學異常造成的疼痛或緊迫輻射性病痛	1.行為異常 2.不整理皮毛 3.脫水 4.不正常的喊叫 5.長時間的食慾不振 6.循環系統之瓦解 7.極度倦怠或不願移動
	5.藥物或化學物損害動物體的生理系統	8.中等至嚴重程度的局部或全身性感染
E.對神智清醒、未麻醉的動物,造成劇烈疼痛且接近或超過疼痛極限，無法以藥物或其他方式緩解（這些實驗需經 IACUC 及獸醫人員謹慎監督）	1.毒性試驗、微生物試驗或腫瘤試驗於不做治療下導致動物重病或瀕死 2.使用藥物或化學物嚴重損害動物生理系統而造成動物死亡、劇烈疼痛或極度緊迫 3.未麻醉情形下使用麻痺或肌肉鬆弛劑 4.燒燙傷或大規模皮膚創傷 5.任何會造成接近疼痛閾值且無法以止痛劑解除該疼痛的操作步驟（如：關節炎模式、眼睛/皮膚刺激性試驗、強烈炎症反應模式、視覺剝奪、電擊/加熱試驗…等） 6.未經 IACUC 核准的安樂死方法	1.自我孤立 2.社會化行為嚴重退縮 3.休息或睡眠增加 4.嚴重的食慾不振 5.動物外表的顯著改變 6.極度倦怠 7.垂死

D1

D2

農委會

人道

科學

https://thedailyguardian.com/the-balance-of-life/

終止點

研究 vs 人道

研究終止點

當預設的實驗目的條件達成時。

人道終止點

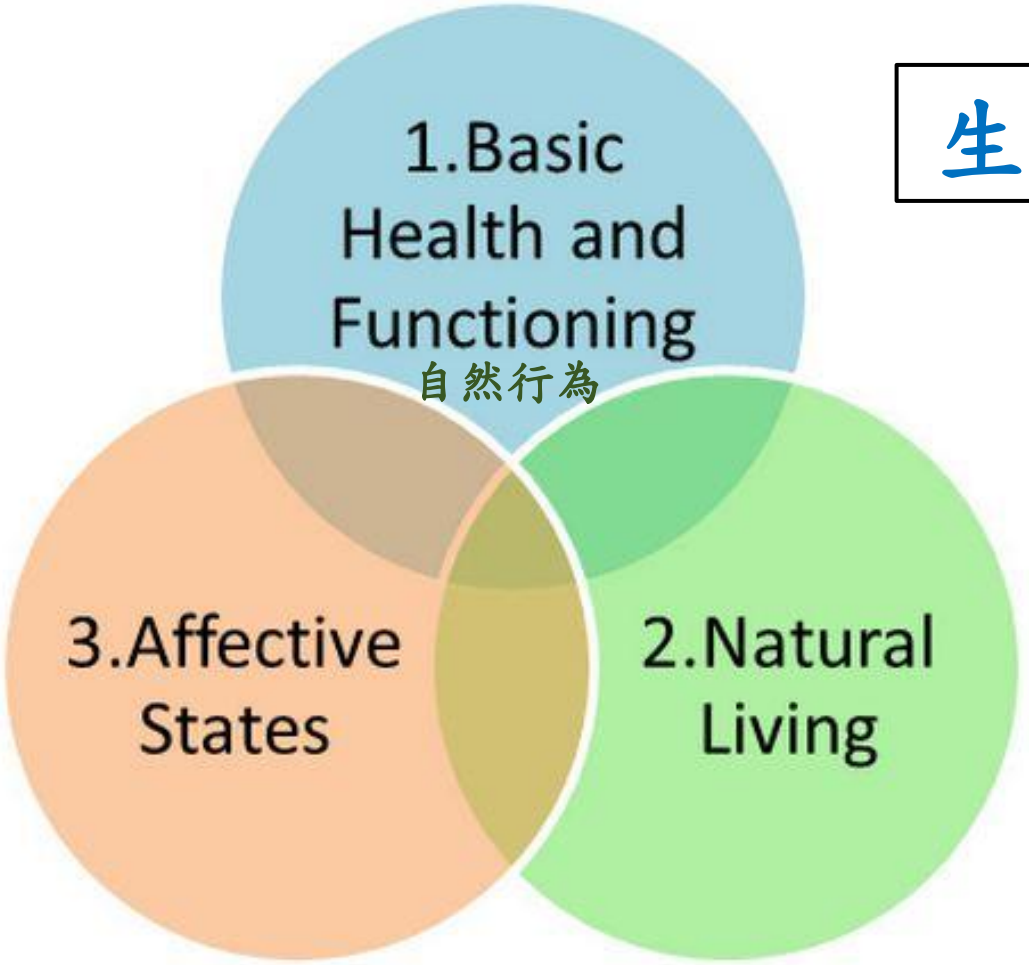
一種精緻化策略，設計來減輕動物於實驗過程中經歷的疼痛、受苦與緊迫。

Hendriksen et al., 2011; OECD definition

Note:

Does not always mean euthanasia – can mean terminating a painful procedure and/or giving treatment to alleviate pain and/or distress.

動物福祉



生理

感受

自然行為

https://www.canr.msu.edu/news/animal_welfare_for_youth_part_3_introducing_the_three_circles_model_to_yout

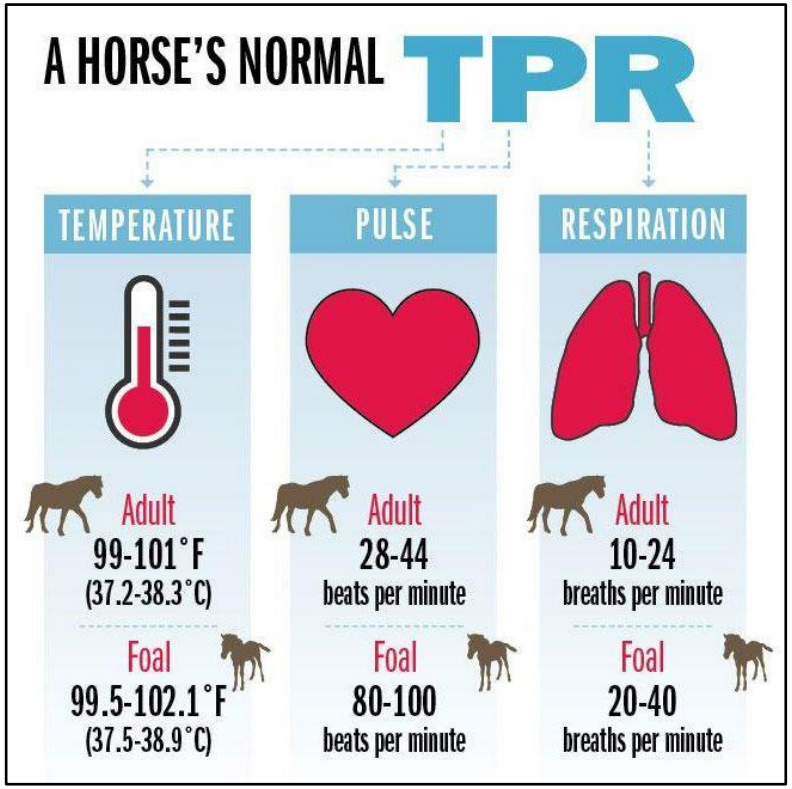
David Fraser, 2008

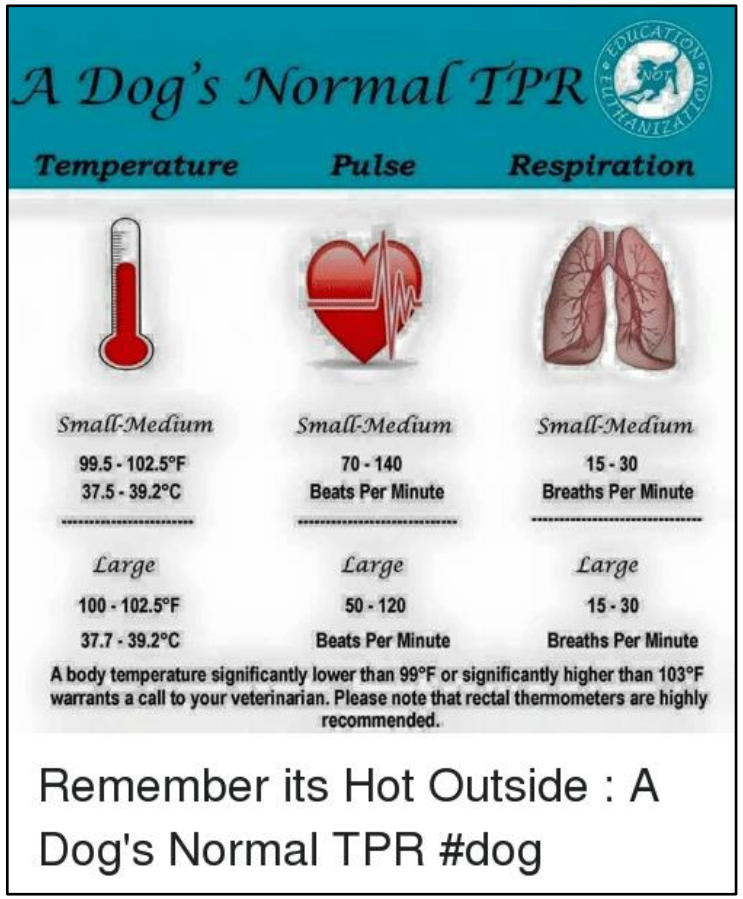
生理

TPR / 生命象徵

- **T**emperature/ 體溫
- **P**ulse/ 脈搏

  (not suitable for small rodents)
- **R**espiratory rate/ 呼吸頻率

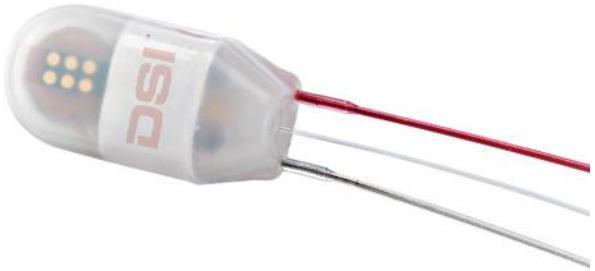




Remember its Hot Outside : A Dog's Normal TPR #dog

小鼠

紅外線溫度計

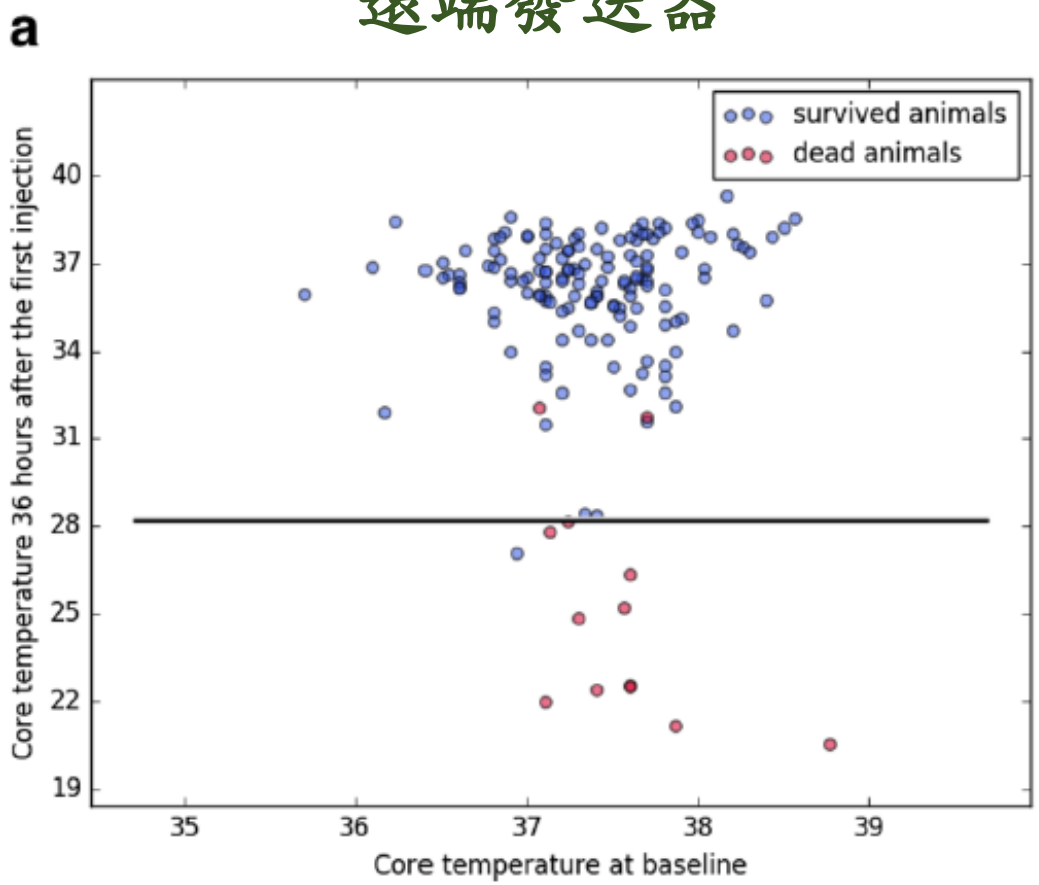


遠端發送器(植入)

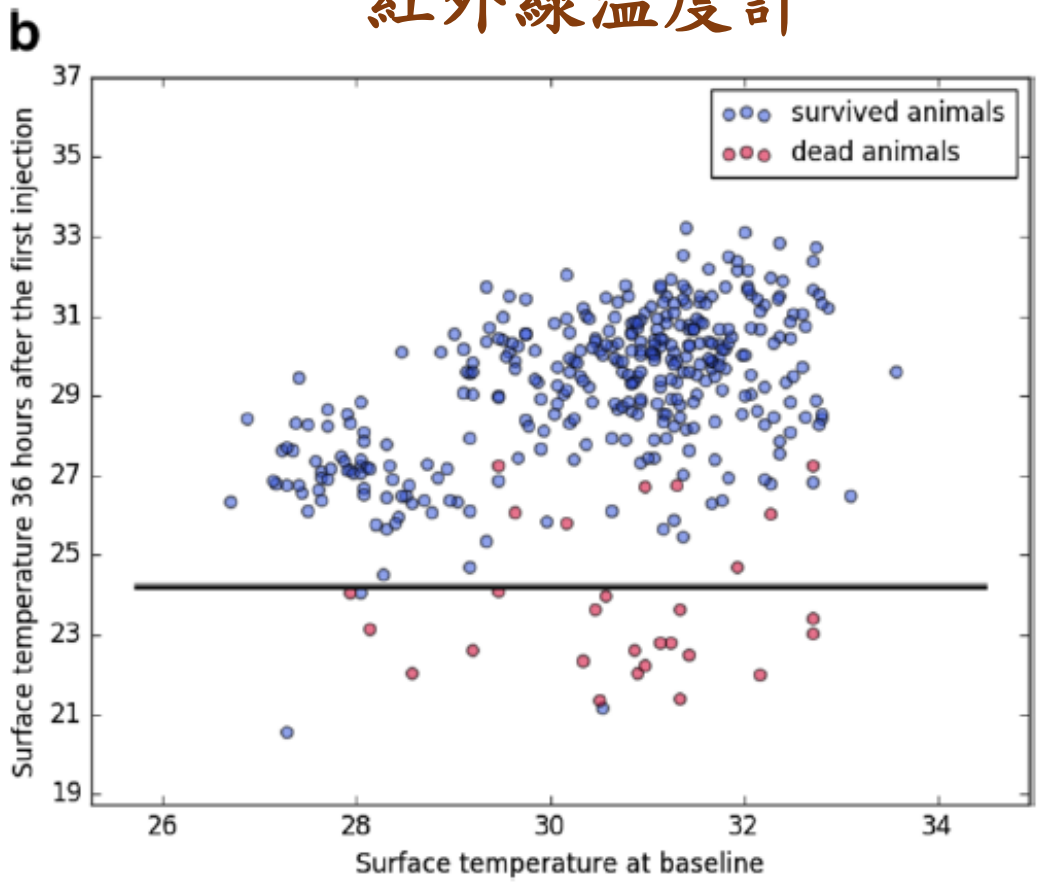
肛溫計

LPS 誘發小鼠體溫過低

遠端發送器



紅外線溫度計



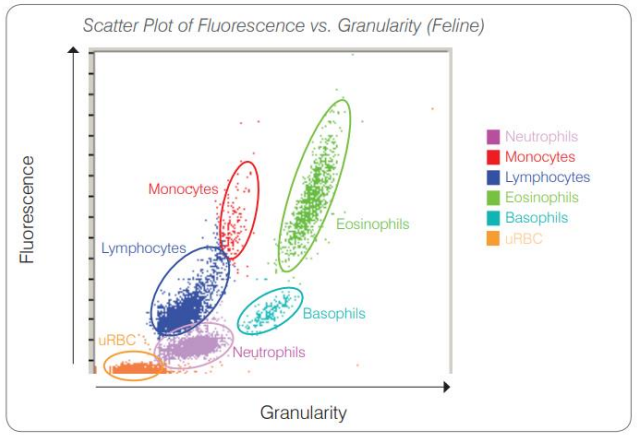
LPS注射後36小時體溫

起始體溫

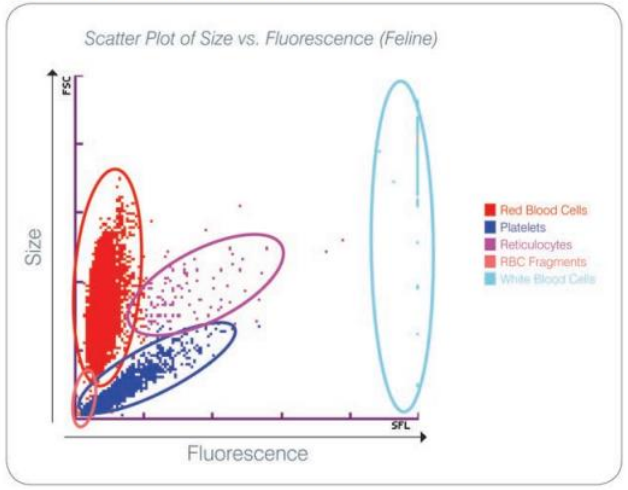
Mei et al., Scientific Reports, 2018

血液生化

White Blood Cell Classification

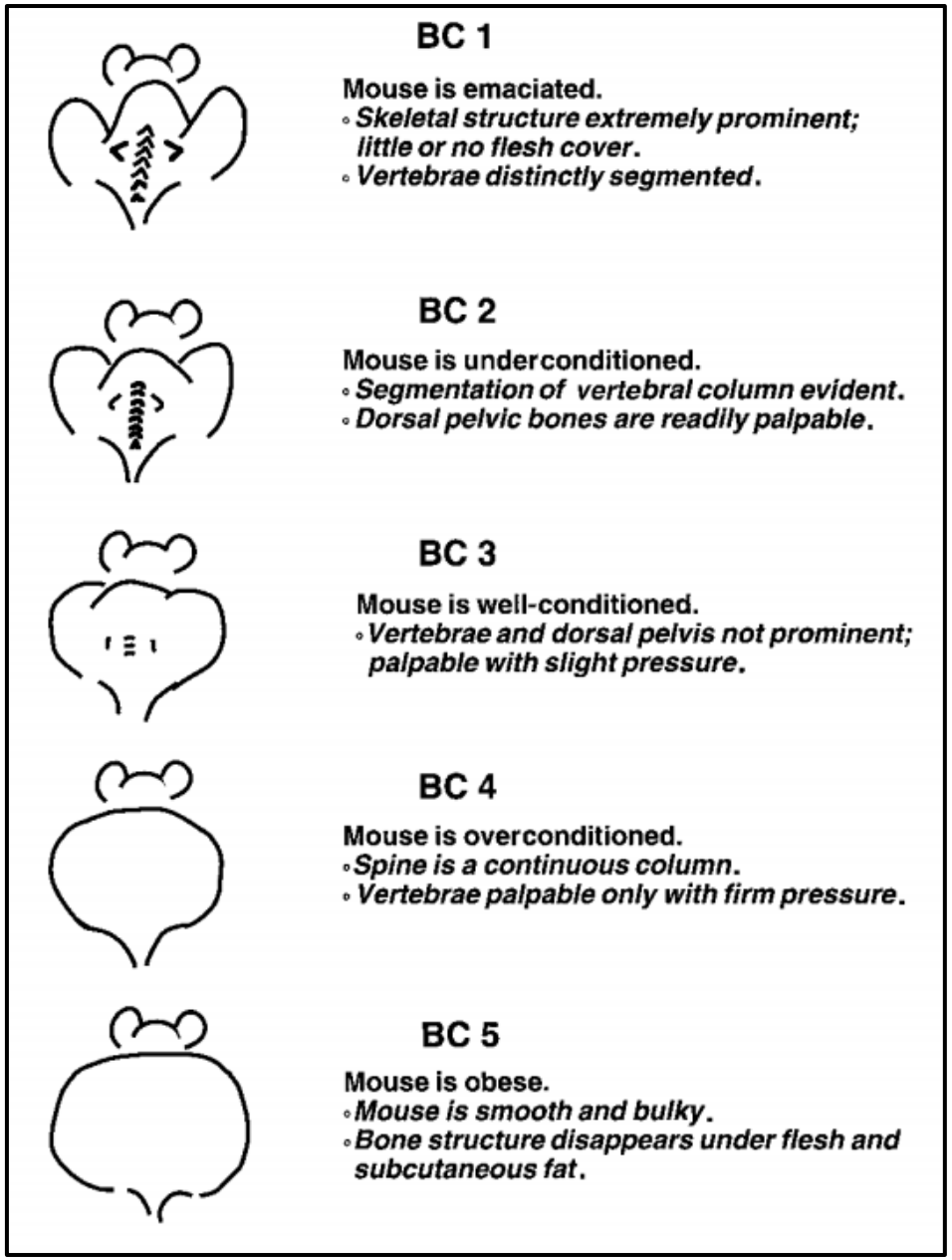


Red Blood Cell Classification



Chemistry	Abbreviation	Chem 17 CLIP	Chem 15 CLIP	Chem 10 CLIP	Equine 15 CLIP	NSAID 6 CLIP	UPC Panel†	Lyte 4 CLIP	SDMA and TT_4 Kit	QC CLIP	Individual Slides
Albumin	ALB	✓	✓	✓	✓					✓	✓
Alkaline Phosphatase	ALKP	✓	✓	✓	✓	✓				✓	✓
Alanine Aminotransferase	ALT	✓	✓	✓		✓				✓	✓
Amylase	AMYL	✓									✓
Aspartate Aminotransferase	AST				✓	✓					✓
Blood Urea Nitrogen	BUN	✓	✓	✓	✓	✓					✓
Calcium	Ca	✓	✓		✓					✓	✓
Cholesterol	CHOL	✓	✓								✓
Creatine Kinase	CK				✓						✓
Creatinine	CREA	✓	✓	✓	✓	✓					✓
Chloride	Cl							✓			
C-Reactive Protein‡	CRP										✓
Fructosamine†	FRU										✓
Gamma-glutamyltransferase	GGT	✓	✓		✓						✓
Glucose	GLU	✓	✓	✓	✓					✓	✓
Potassium	K							✓			
Lactate	LAC										✓
Lactate Dehydrogenase	LDH				✓						✓
Lipase	LIPA	✓									✓
Magnesium	Mg										✓
Sodium	Na							✓			
Ammonia	NH_3									✓	✓
Phenobarbital†	PHBR										✓
Inorganic Phosphate	PHOS	✓	✓								✓
Progesterone	PROG										✓
Symmetric dimethylarginine†	SDMA								✓		✓
Total Bilirubin	TBIL	✓	✓		✓						✓
Total Protein	TP	✓	✓	✓	✓						✓
Total T_4†	TT4								✓		✓
Triglycerides	TRIG										✓
Urine Creatinine	UCRE						✓				

IDEXX LABORATORIES



Body **C**ondition **S**cores (**BCS**) in Mouse

Ullman-Cullere & Foltz, 1999

BCS in Rat

BC 1
Rat is emaciated

- Segmentation of vertebral column prominent if not visible.
- Little or no flesh cover over dorsal pelvis. Pins prominent if not visible.
- Segmentation of caudal vertebrae prominent.

BC 2
Rat is under conditioned

- Segmentation of vertebral column prominent.
- Thin flesh cover over dorsal pelvis, little subcutaneous fat. Pins easily palpable.
- Thin flesh cover over caudal vertebrae, segmentation palpable with slight pressure.

BC 3
Rat is well-conditioned

- Segmentation of vertebral column easily palpable.
- Moderate subcutaneous fat store over pelvis. Pins easily palpable with slight pressure.
- Moderate fat store around tail base, caudal vertebrae may be palpable but not segmented.

BC 4
Rat is overconditioned

- Segmentation of vertebral column palpable with slight pressure.
- Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis palpable with firm pressure.
- Thick fat store over tail base, caudal vertebrae not palpable.

BC 5
Rat is obese

- Segmentation of vertebral column palpable with firm pressure; may be a continuous column.
- Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis not palpable with firm pressure.
- Thick fat store over tail base, caudal vertebrae not palpable.

Hickman & Swan, 2010

BCS

Guinea pig Size-O-Meter

Size-O-Meter Score:		Characteristics:
1	Very Thin More than 20% below ideal body weight	Each individual rib can be felt easily, hips and spine are prominent and extremely visible and can be felt with the slightest touch. Under abdominal curve can be seen. Spine appears hunched.
2	Thin Between 10-20% below ideal body weight	Each rib is easily felt but not prominent. Hips and spine are easily felt with no pressure. Less of an abdominal curve can be seen.
3	Ideal	Ribs are not prominent and cannot be felt individually. Hips and spine are not visible but can be felt. No abdominal curve. Chest narrower then hind end.
4	Overweight 10-15% above ideal body weight	Ribs are harder to distinguish. Hips and spine difficult to feel. Feet not always visible.
5	Obese 15-20% above ideal body weight	Ribs, hips and spine cannot be felt or can with mild pressure. No body shape can be distinguished. Underbelly touching floor when Guinea-pig is in standing position, feet cannot be seen.

Rabbit Size-O-Meter

Size-O-Meter Score:		Characteristics:
1	Very Thin More than 20% below ideal body weight	• Hip bones, ribs and spine are very sharp to the touch • Loss of muscle and no fat cover • The rump area curves in
2	Thin Between 10-20% below ideal body weight	• Hip bones, ribs and spine are easily felt • Loss of muscle and very little fat cover • Rump area is flat
3	Ideal	• Hip bones, ribs and spine easily felt but are rounded, not sharp – Ribs feel like a pocket full of pens! • No abdominal bulge • Rump area is flat
4	Overweight 10-15% above ideal body weight	• Pressure is needed to feel the ribs, spine and hip bones • Some fat layers • The rump is rounded
5	Obese More than 15% above ideal body weight	• Very hard to feel the spine and hip bones – Ribs can't be felt! • Tummy sags with obvious fat padding • Rump bulges out

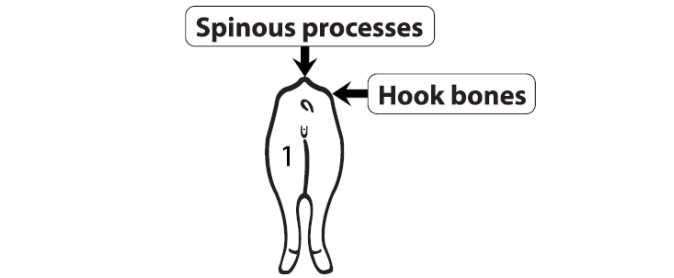
Dog Size-O-Meter

Size-O-Meter Score:		Characteristics:
1	Very Thin More than 20% below ideal body weight	• Ribs, spine and hip bones are very easily seen (in short haired pets) • Obvious loss of muscle bulk • No fat can be felt under the skin
2	Thin Between 10-20% below ideal body weight	• Ribs, spine and hip bones easily seen • Obvious waist and abdominal tuck • Very little fat can be felt under the skin
3	Ideal	• Ribs, spine and hip bones easily felt • Visible waist with an abdominal tuck • A small amount of fat can be felt
4	Overweight 10-15% above ideal body weight	• Ribs, spine and hip bones are hard to feel • Waist barely visible with a broad back • Layer of fat on belly and at base of tail
5	Obese More than 15% above ideal body weight	• Ribs, spine and hip bones extremely difficult to feel under a thick layer of fat • No waist can be seen and belly may droop significantly • Heavy fat pads on lower back and at the base of the tail

Cat Size-O-Meter

Size-O-Meter Score:		Characteristics:
1	Very Thin More than 20% below ideal body weight	• Ribs, spine and hip bones are very easily seen (in short haired pets) • Pronounced waist • Obvious loss of muscle mass with no belly fat
2	Thin Between 10-20% below ideal body weight	• Ribs, spine and hip bones easily seen • Obvious waist • Very little belly fat
3	Ideal	• Ribs, spine and hip bones easily felt • Visible waist • A small amount of belly fat
4	Overweight 10-15% above ideal body weight	• Ribs, spine and hip bones are hard to feel • No defined waist • Slightly sagging belly
5	Obese More than 15% above ideal body weight	• Ribs, spine and hip bones extremely difficult to feel under a padding of fat • No waist can be seen • Heavy fat pads on lower back and an obvious sagging belly – skin rolls may sway from side to side when walking

Body Condition Score- Swine



1. Emaciated
Landmark bones are prominent even without palpation. Considered unfit to travel.

2. Thin
Bones can be easily felt with slight pressure.

3. Ideal
The pig's bones are barely felt when palpating with firm pressure.

4. Fat
Bones of the pig are undetectable with palpation.

5. Overly Fat
A body score of 5 has the same palpation characteristics as a body score of 4. However, this animal is excessively overweight.

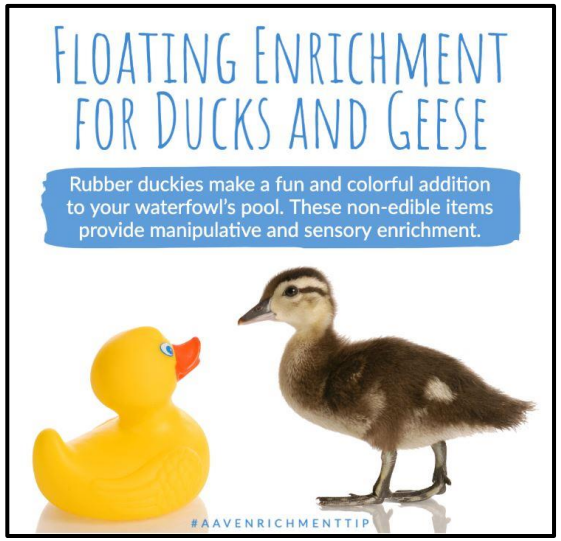
1. NVAP Module 21: Animals' Fitness to Travel, July 2011
2. www.pfma.org.uk

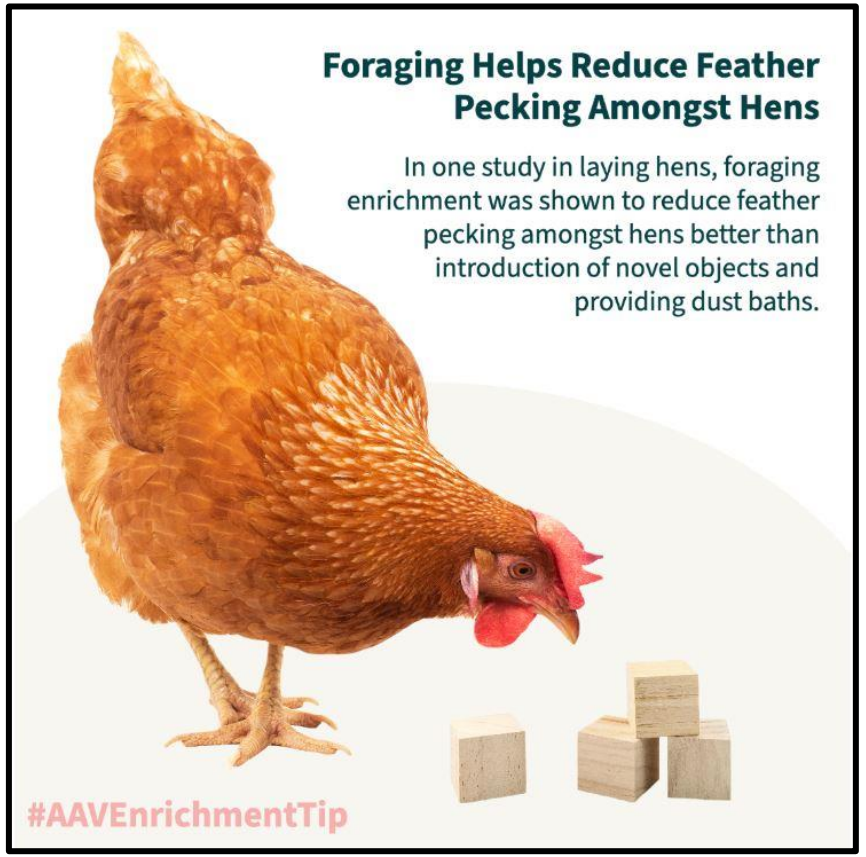
自然行為

自然行為

- ✓ 挖掘
- ✓ 覓食
- ✓ 啃咬
- ✓ 築巢
- ✓ 社交

環境豐富化物質





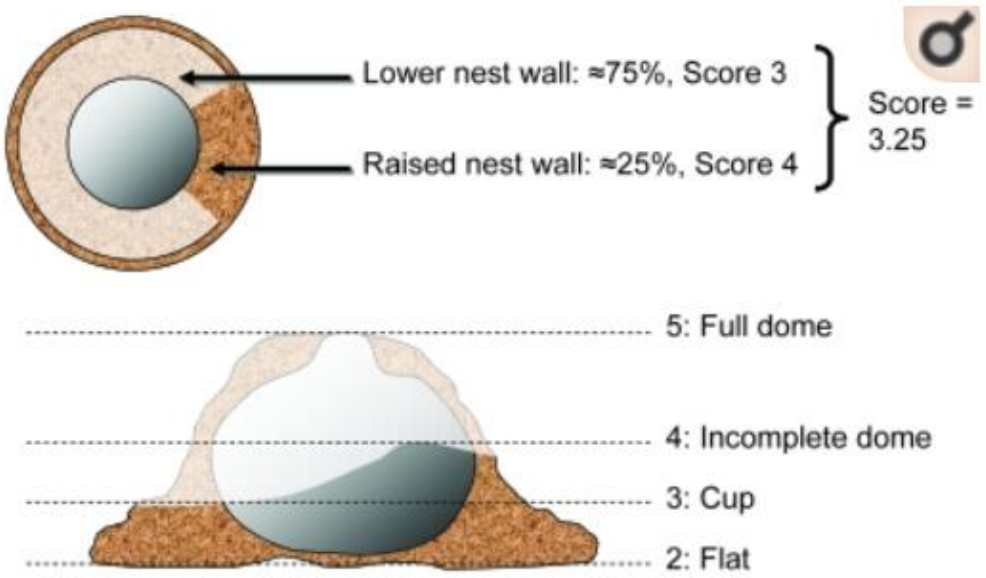
https://www.aav.org/

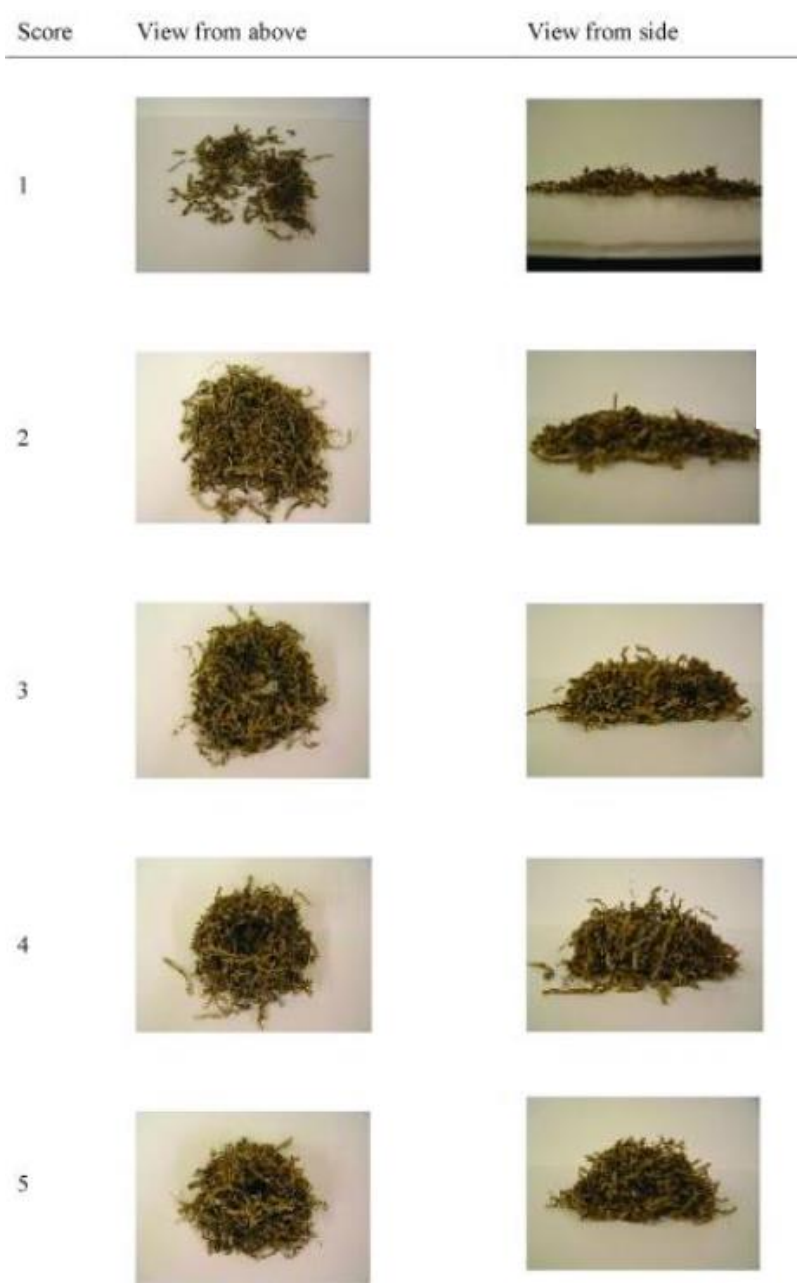
社交動物

https://www.na3rsc.org/pigs/

https://www.nc3rs.org.uk/3rs-resources/housing-and-husbandry-rabbit

小鼠築巢-自然行為

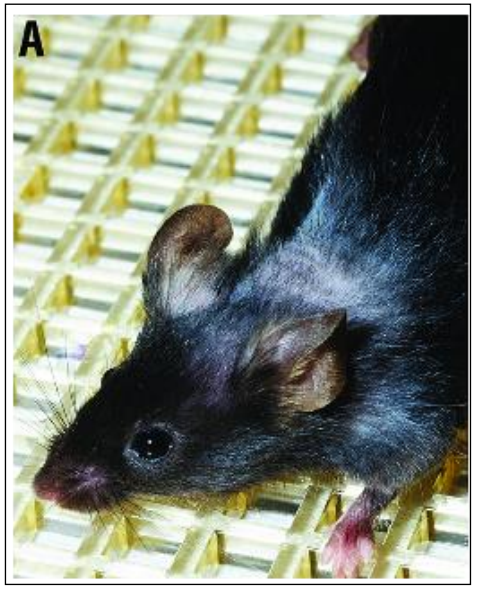




Hess et al., JAALAS., 2008

刻板行為 – 緊迫

（重複行為）



過度理毛

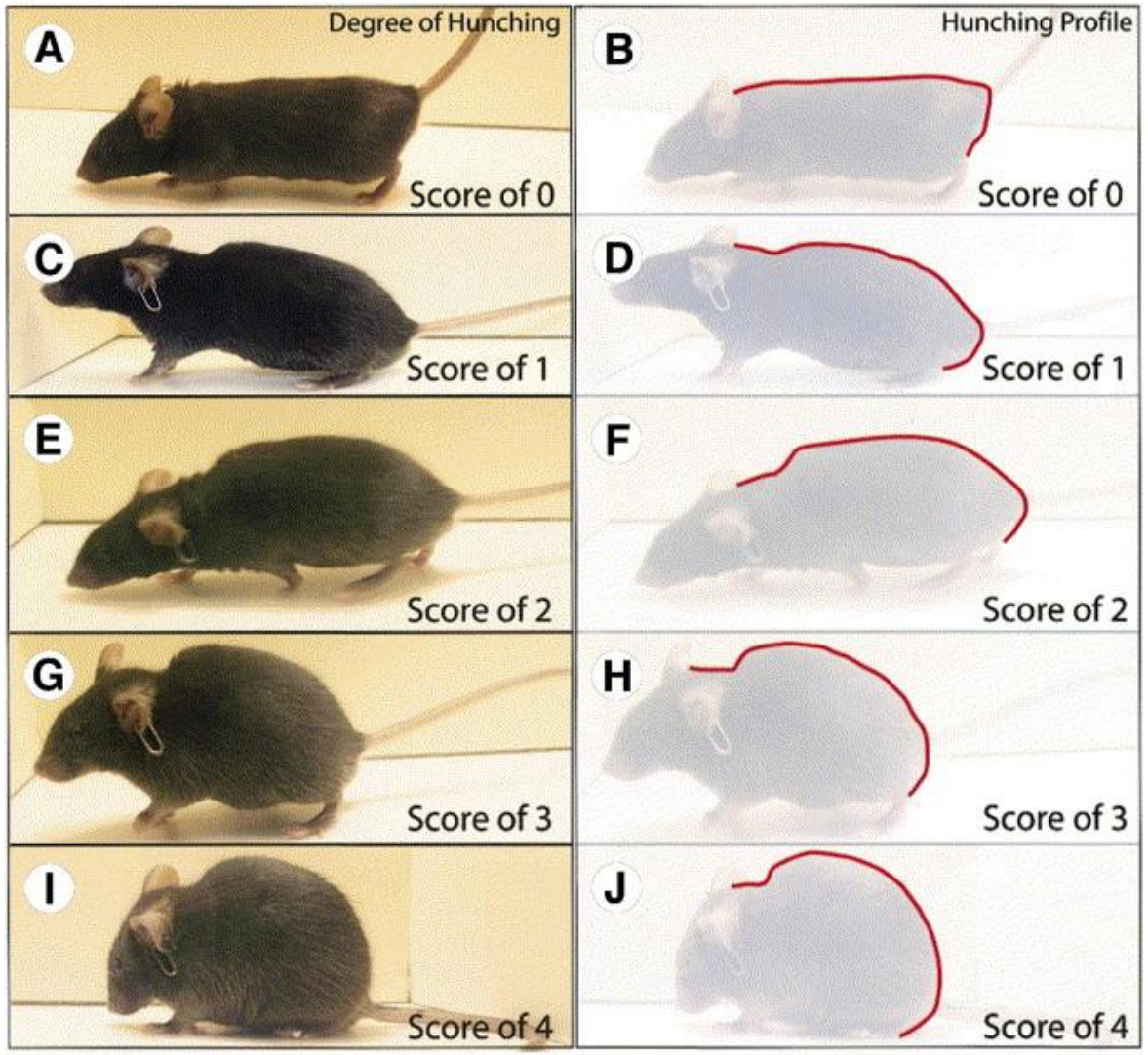
（自殘/被霸凌）

咬槽攝氣癖

吸吮拇指

感受

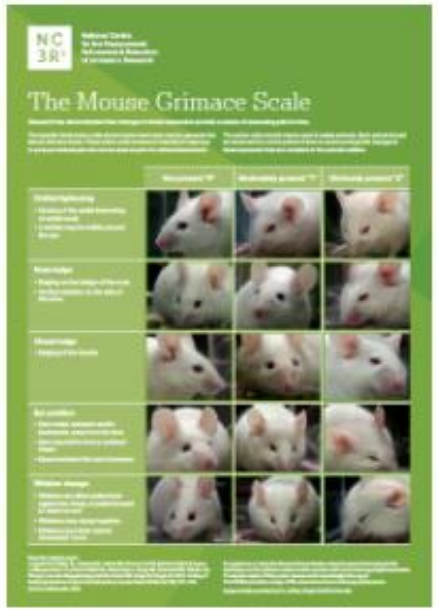
拱背(疼痛)

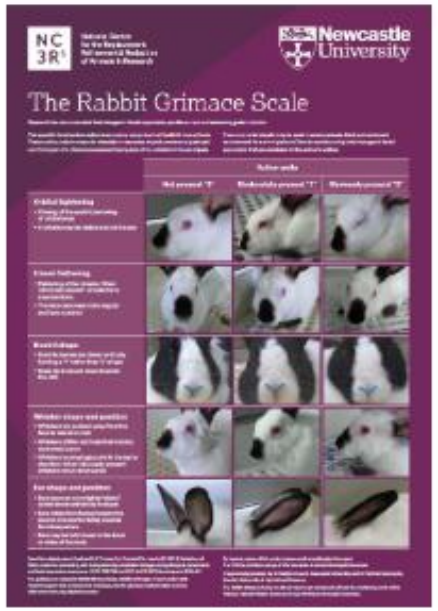


Sevcik et al., 2006

Grimace Scales - Pain

鬼臉評分表　　疼痛





Action Uints (AU)

AU is not present	=	0
AU moderately visible	=	1
AU severe	=	2

https://www.nc3rs.org.uk/grimacescales


"0"


"1"


"2"

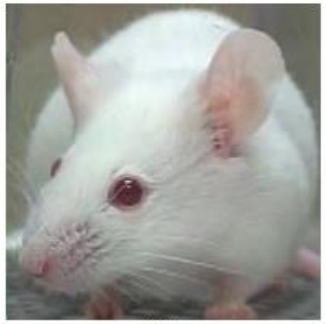
瞇眼


"0"


"1"


"2"

臉頰鼓起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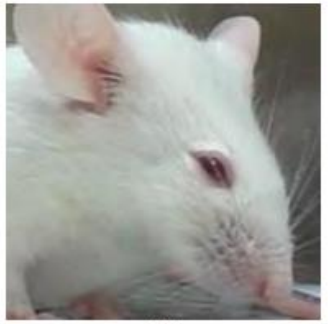
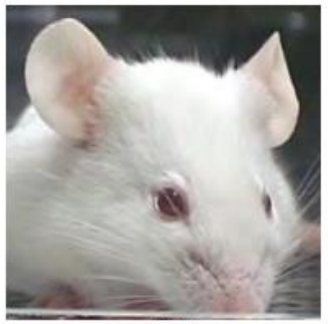

"1"


"2"

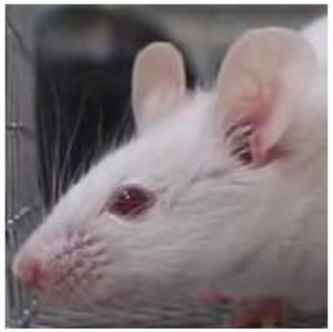
觸鬚改變


"0"


"1"


"2"

鼻吻部鼓起


"0"


"1"


"2"

耳朵位置

Nat Methods. 2010 Jun;7(6):447-9. doi: 10.1038/nmeth.1455. Epub 2010 May 9.

Coding of facial expressions of pain in the laboratory mouse.

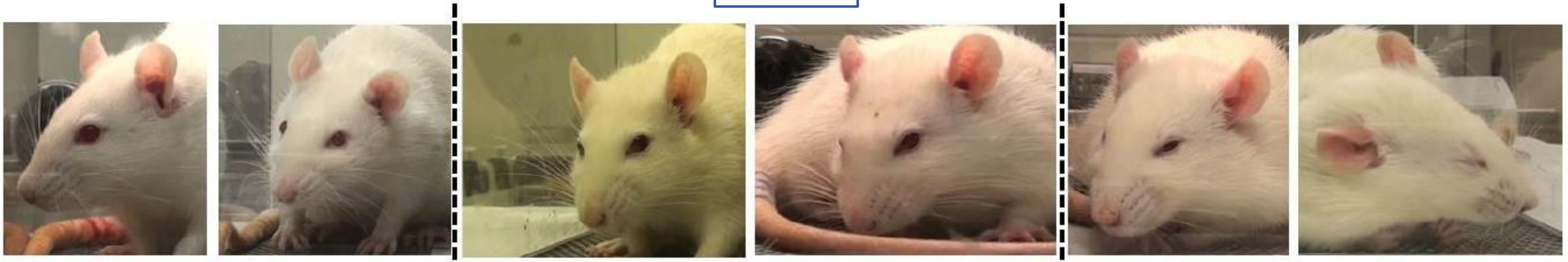
Langford DJ[1], Bailey AL, Chanda ML, Clarke SE, Drummond TE, Echols S, Glick S, Ingrao J, Klassen-Ross T, Lacroix-Fralish ML, Matsumiya L, Sorge RE, Sotocinal SG, Tabaka JM, Wong D, van den Maagdenberg AM, Ferrari MD, Craig KD, Mogil JS.

Author information

1 Department of Psychology and Alan Edwards Centre for Research on Pain, McGill University, Montreal, Quebec, Canada.

Rat

瞇眼



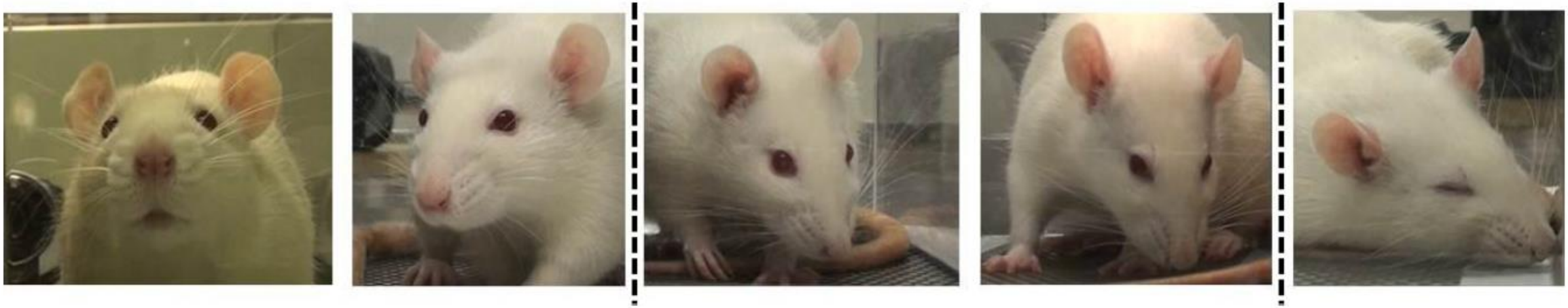
"0" "1" "2"

觸鬚改變

"0" "1" "2"

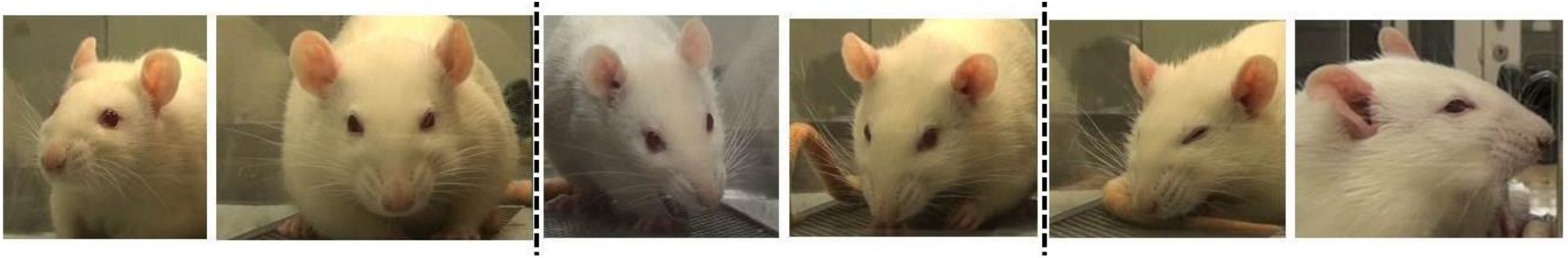
Rat

耳朵位置 - 捲曲變尖/向前/夾角變大45度



"0" "1" "2"

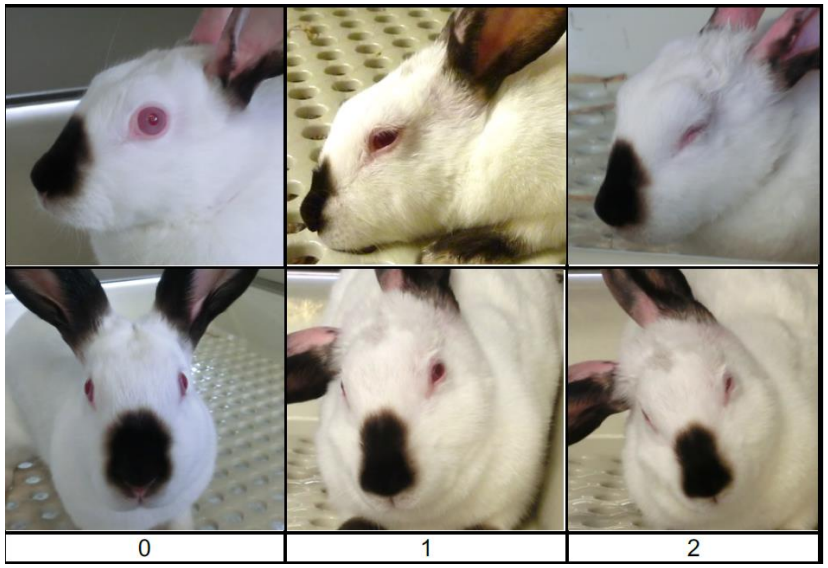
鼻子 & 臉頰變平



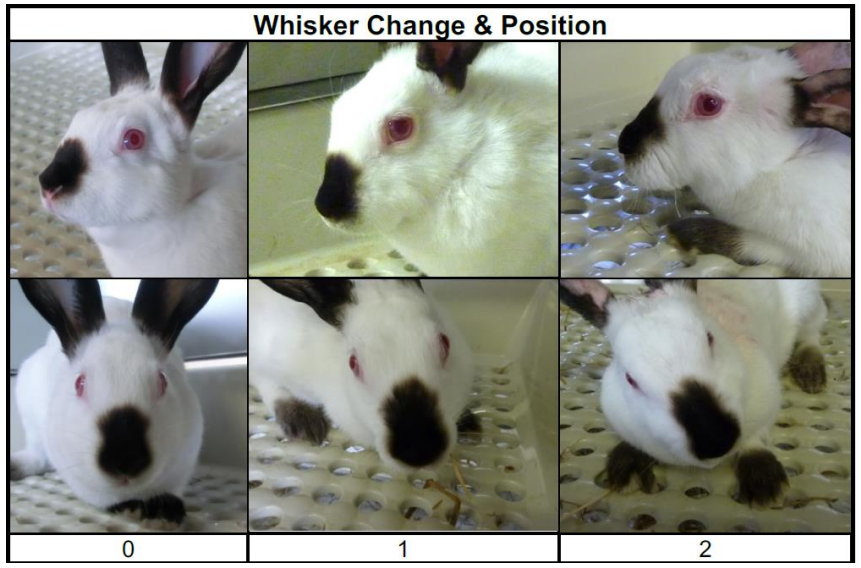
"0" "1" "2"

Rabbit

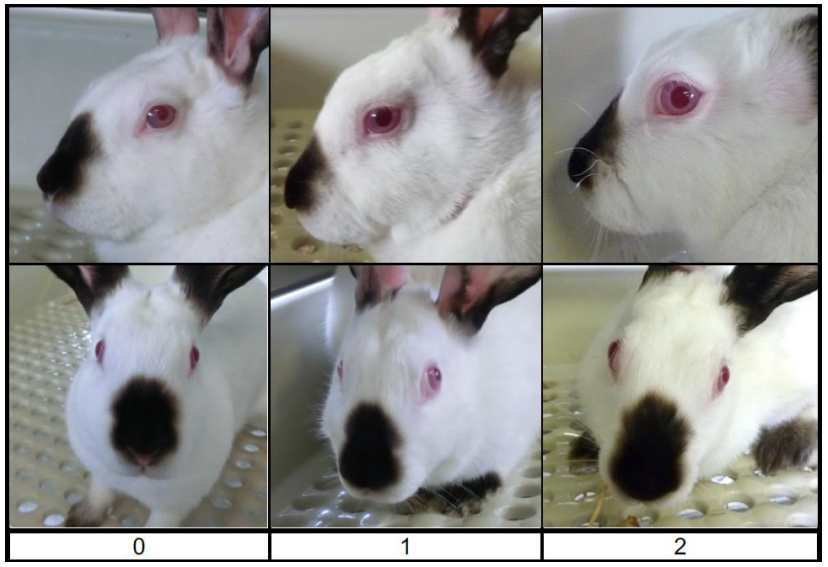
瞇眼



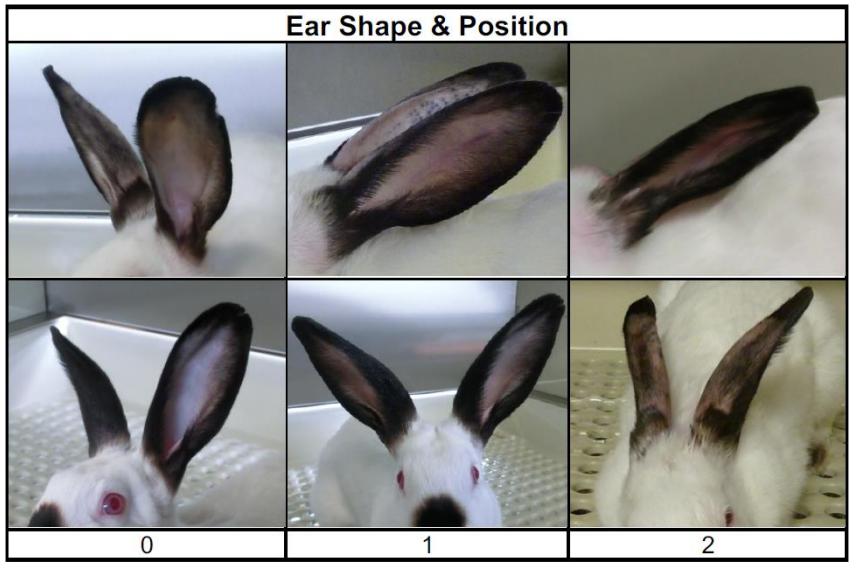
觸鬚改變



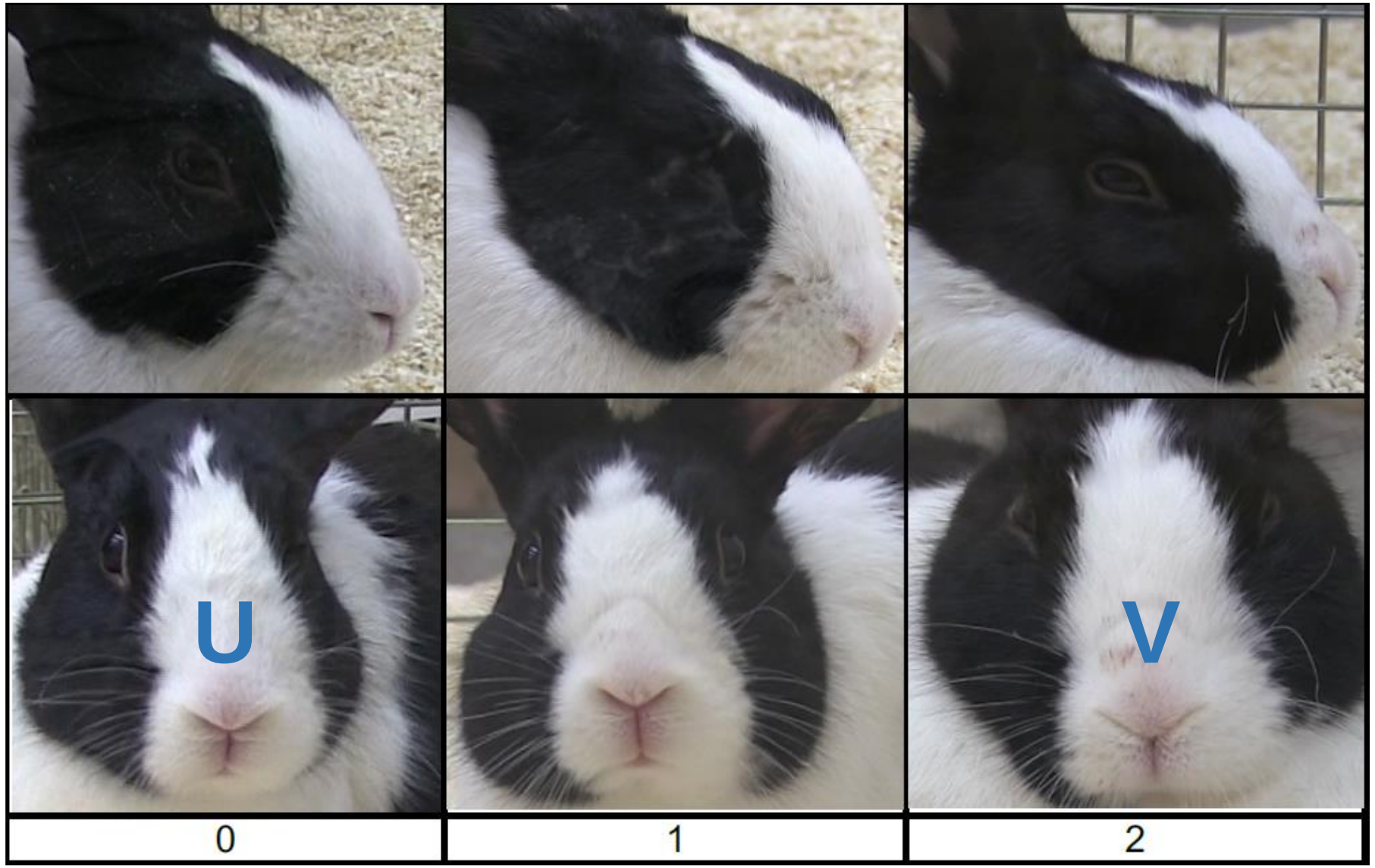
臉頰變平

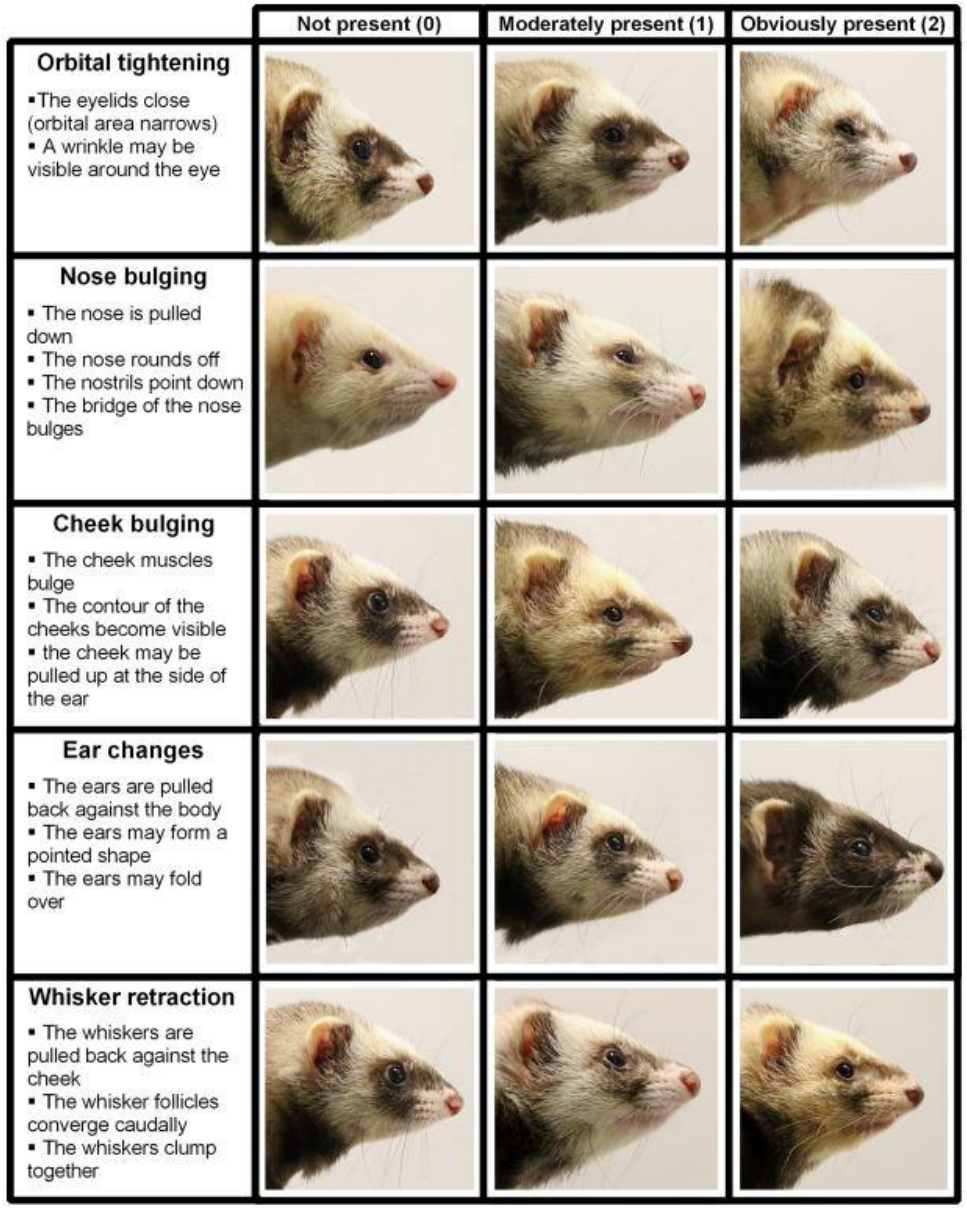


耳朵位置

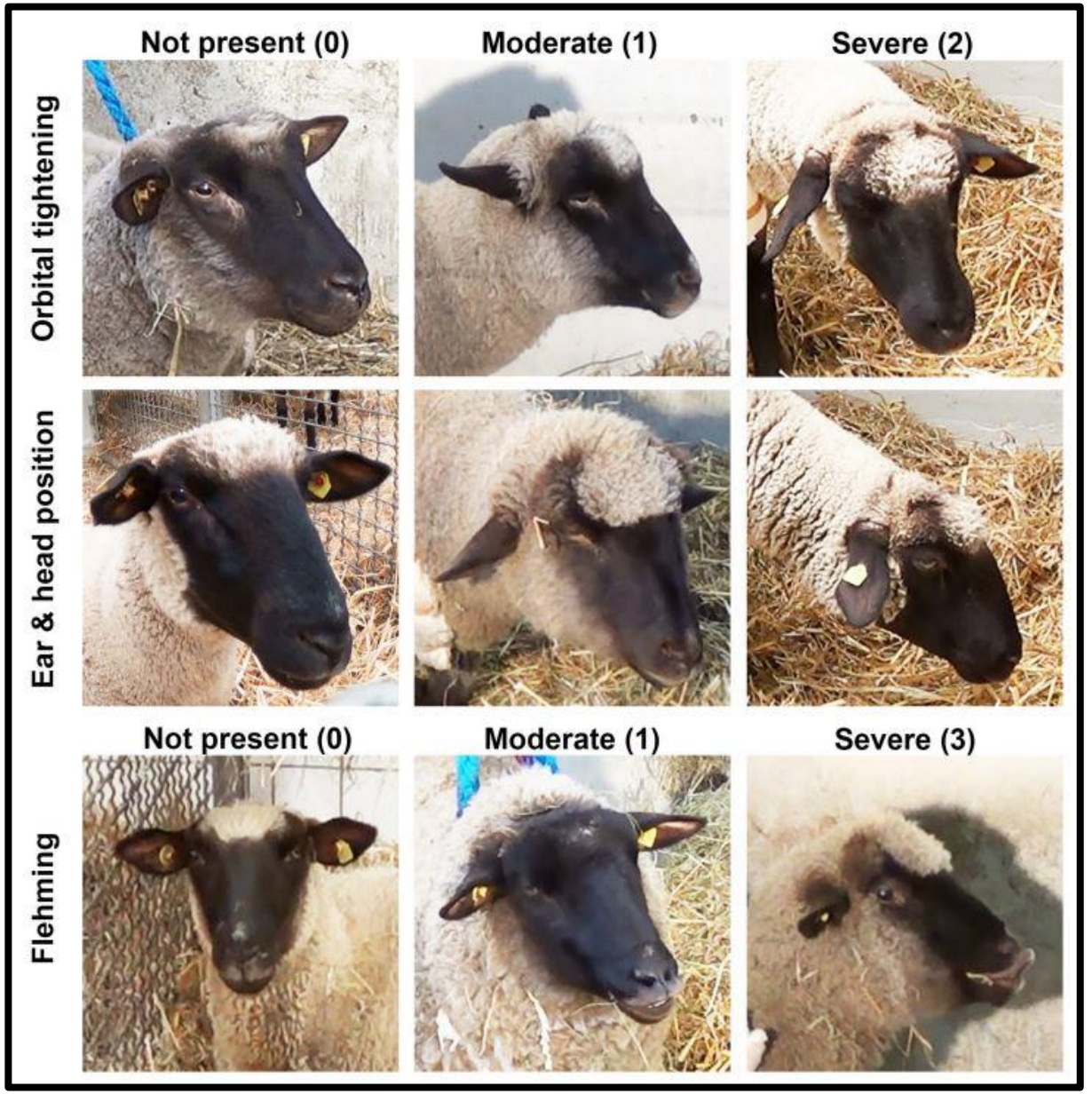


鼻子形狀
U → V





Reijgwart et al., 2017



Häger et al., 2017

Stiffly backwards ears

Not present (0) | Moderately present (1) | Obviously present (2)

The ears are held stiffly and turned backwards. As a result, the space between the ears may appear wider relative to baseline.

Orbital tightening

Not present (0) | Moderately present (1) | Obviously present (2)

The eyelid is partially or completely closed. Any eyelid closure that reduces the eye size by more than half should be coded as "obviously present" or "2".

Tension above the eye area

Not present (0) | Moderately present (1) | Obviously present (2)

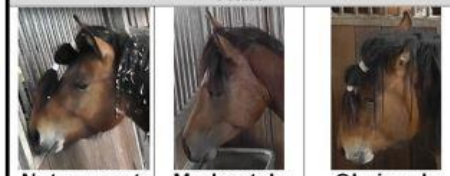
The contraction of the muscles in the area above the eye causes the increased visibility of the underlying bone surfaces. If temporal crest bone is clearly visible should be coded as "obviously present" or "2".

Prominent strained chewing muscles

Not present (0) | Moderately present (1) | Obviously present (2)

Straining chewing muscles are clearly visible as an increase tension above the mouth. If chewing muscles are clearly prominent and recognizable the score should be coded as "obviously present" or "2".

Mouth strained and pronounced chin



Not present (0) | Moderately present (1) | Obviously present (2)

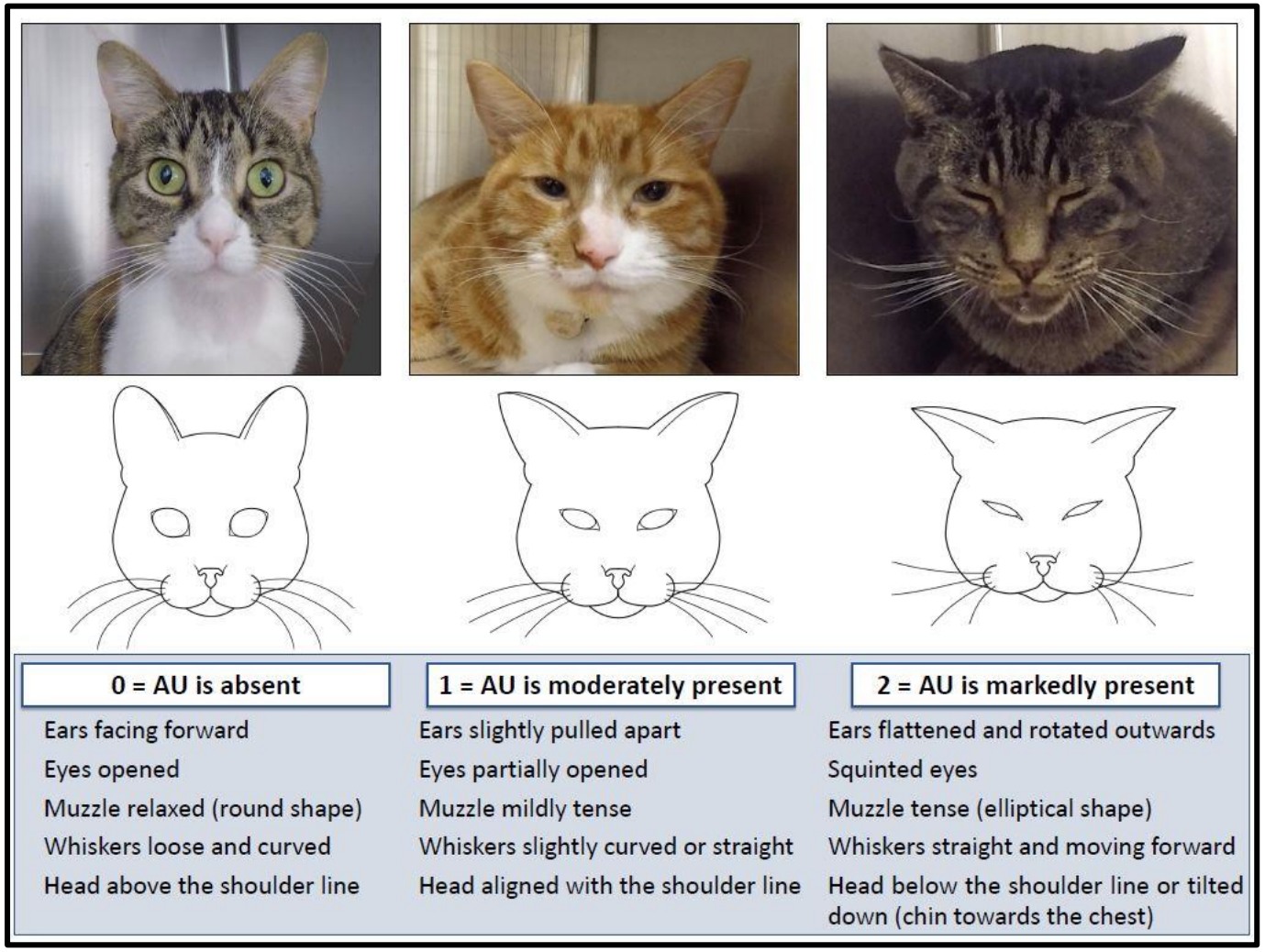
Strained mouth is clearly visible when upper lip is drawn back and lower lip causes a pronounced "chin".

Strained nostrils and flattening of the profile

Not present (0) | Moderately present (1) | Obviously present (2)

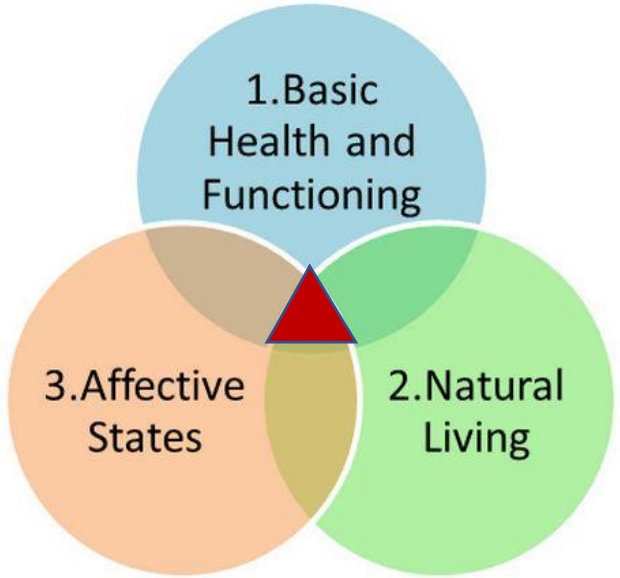
Nostrils look strained and slightly dilated, the profile of the nose flattens and lips elongate.

Costa et al., 2014



Evangelista et al., 2020

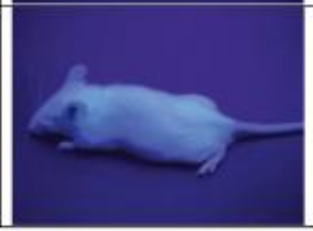
手術



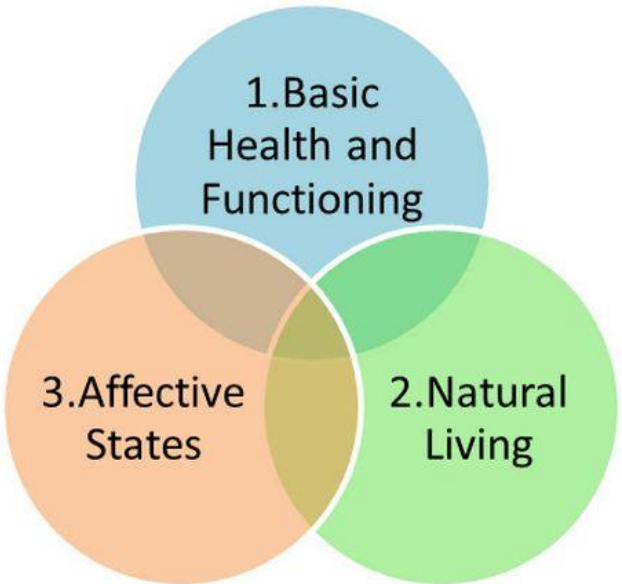
疼痛

理毛

- 開腹
- 螢光油
- 測量/評分理毛時間

Score	Description	Example Image	
		CD1	C57BL6
1	A strong fluorescent signal is present at the application site on the forehead between the ears		
2	Fluorescence present at the application site as well as the front and/or rear nails		
3	Fluorescence present at the application site and the ears. Front and/or rear nails may also fluoresce		
4	Fluorescence is absent from the nails and ears but remains present in trace amounts at the application site		
5	Fluorescence is no longer detected		

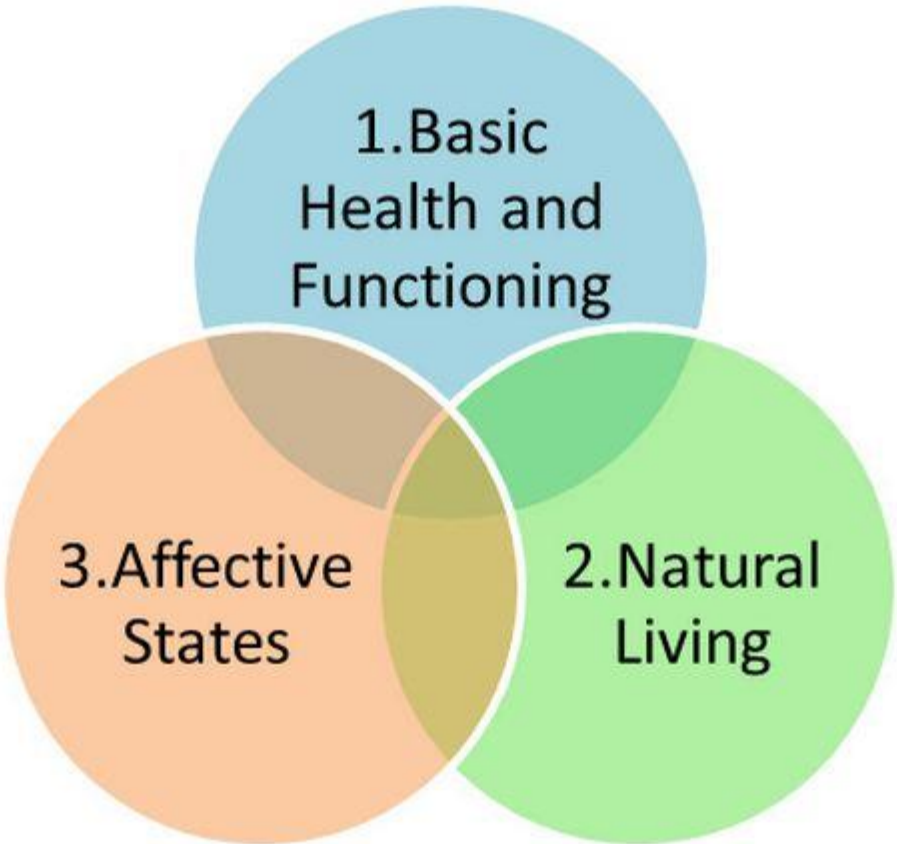
Oliver et al., J Am Assoc Lab Anim Sci., 2018



Pain Score	Example	Psychological & Behavioral	Response to Palpation	Body Tension
0		☐ **Comfortable** when resting ☐ **Happy, content** ☐ Not bothering wound or surgery site ☐ Interested in or curious about surroundings	☐ **Nontender** to palpation of wound or surgery site, or to palpation elsewhere	Minimal
1		☐ **Content to slightly unsettled** or restless ☐ **Distracted easily** by surroundings	☐ **Reacts to palpation** of wound, surgery site, or other body part by **looking around**, **flinching**, or **whimpering**	Mild
2		☐ Looks **uncomfortable** when resting ☐ May **whimper** or cry and may **lick or rub wound** or surgery site when unattended ☐ Droopy ears, **worried facial expression** (arched eye brows, darting eyes) ☐ **Reluctant to respond** when beckoned ☐ **Not eager to interact** with people or surroundings but will look around to see what is going on	☐ Flinches, whimpers cries, or guards/pulls away	Mild to Moderate **Reassess analgesic plan**
3		☐ **Unsettled, crying, groaning, biting or chewing** wound when unattended ☐ **Guards or protects** wound or surgery site by altering weight distribution (i.e., limping, shifting body position) ☐ **May be unwilling to move** all or part of body	☐ May be **subtle** (shifting eyes or increased respiratory rate) if dog is too painful to move or is stoic ☐ May be **dramatic**, such as a sharp cry, growl, bite or bite threat, and/or pulling away	Moderate **Reassess analgesic plan**
4		☐ **Constantly groaning or screaming** when unattended ☐ May bite or chew at wound, but unlikely to move ☐ **Potentially unresponsive** to surroundings ☐ **Difficult to distract** from pain	☐ **Cries at non-painful palpation** (may be experiencing allodynia, wind-up, or fearful that pain could be made worse) ☐ May react aggressively to palpation	Moderate to Severe **May be rigid to avoid painful movement** **Reassess analgesic plan**

COLORADO STATE UNIVERSITY | COLLEGE OF VETERINARY MEDICINE AND BIOMEDICAL SCIENCES

例外 ???


1.Basic Health and Functioning
3.Affective States
2.Natural Living

瀕死

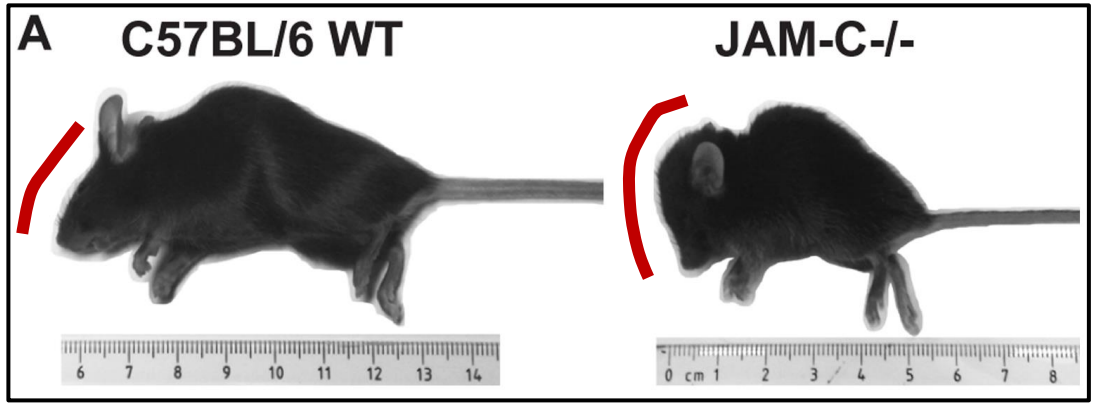
NIH ARAC 定義 (2011) :

1. 對人為操作刺激無反應
2. 無法移動
3. 無法自行飲食或飲水

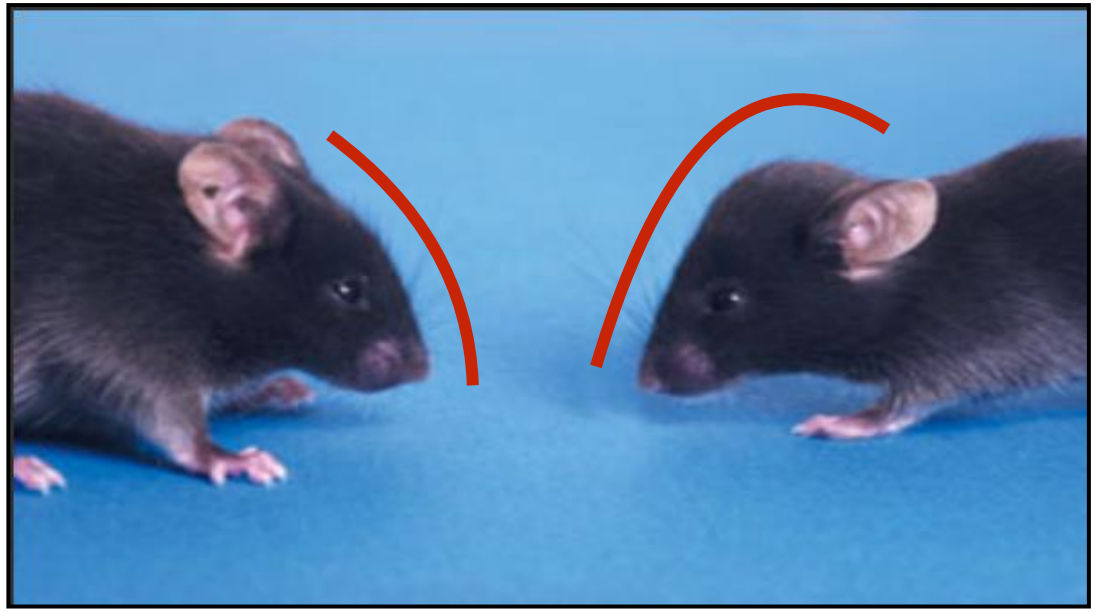
例外：有科學性理由且IACUC同意，才能將瀕死當作實驗中止點。

Burkholder et al., Curr Protoc Mouse Biol., 2012

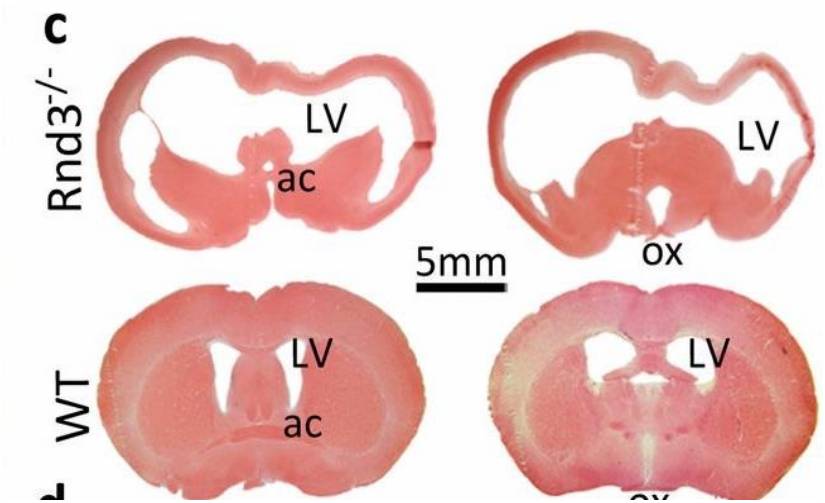
水腦

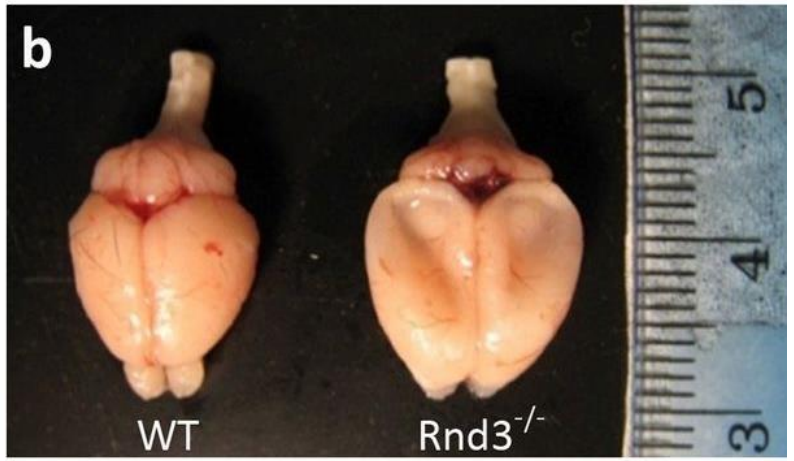


Wyss *et al*., PLoS One., 2012



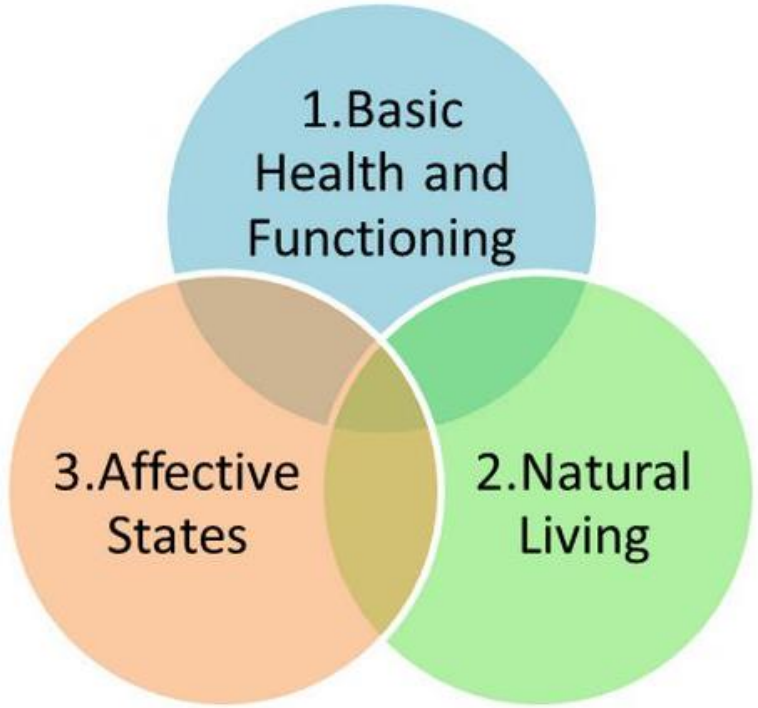
http://www.the-scientist.com/?articles.view/articleNo/15081/title/Hydrocephalus-and-the-Accidental-Transgene--Redox-RNA--To-Be-a-Bee--but-He-or-She-/

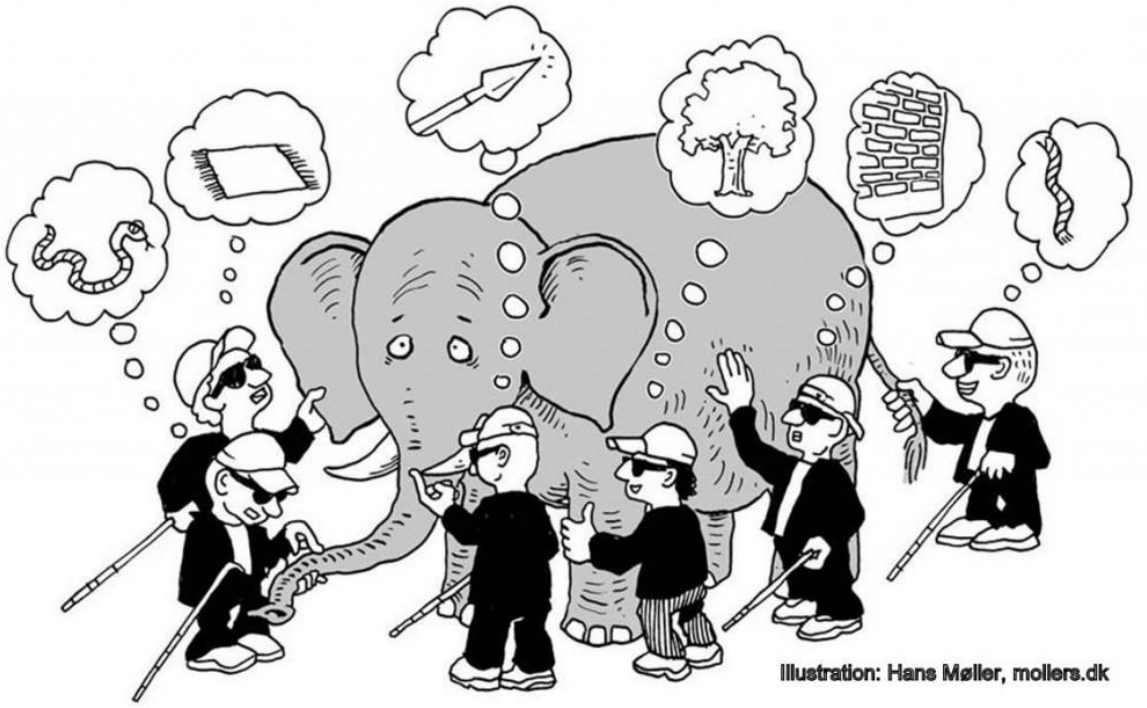




Lin et al., PNAS, 2013

評估動物福祉時不能僅僅單靠一項來做決定!!!





如何建立實驗動物的人道終止點?

科學

動物福祉

Clinical Signs (NIH, ARAC, 2011)	
1	Rapid or progressive weight loss
2	Debilitating diarrhea
3	Dehydration/reduced skin turgor
4	Edema
5	Sizable abdominal enlargement or ascites
6	Progressive dermatitis
7	Rough hair coat
8	Hunched posture
9	Lethargy or persistent recumbency
10	Coughing, labored breathing, nasal discharge
11	Jaundice, cyanosis, and/or pallor/anemia
12	Neurological signs
13	Bleeding from any orifice
14	Self-induced trauma
15	Any condition interfering with daily activities (e.g. eating or drinking, ambulation, or elimination)
16	Excessive or prolonged hyperthermia or hypothermia
17	For aquatic species additional signs can include scoliosis, emaciation, significant skin lesions, and/or exposure of muscle or other tissues

Clinical Signs and Evaluation Criteria Used to Determine Humane Endpoints

Burkholder et al., Curr Protoc Mouse Biol., 2012

Cancer Research

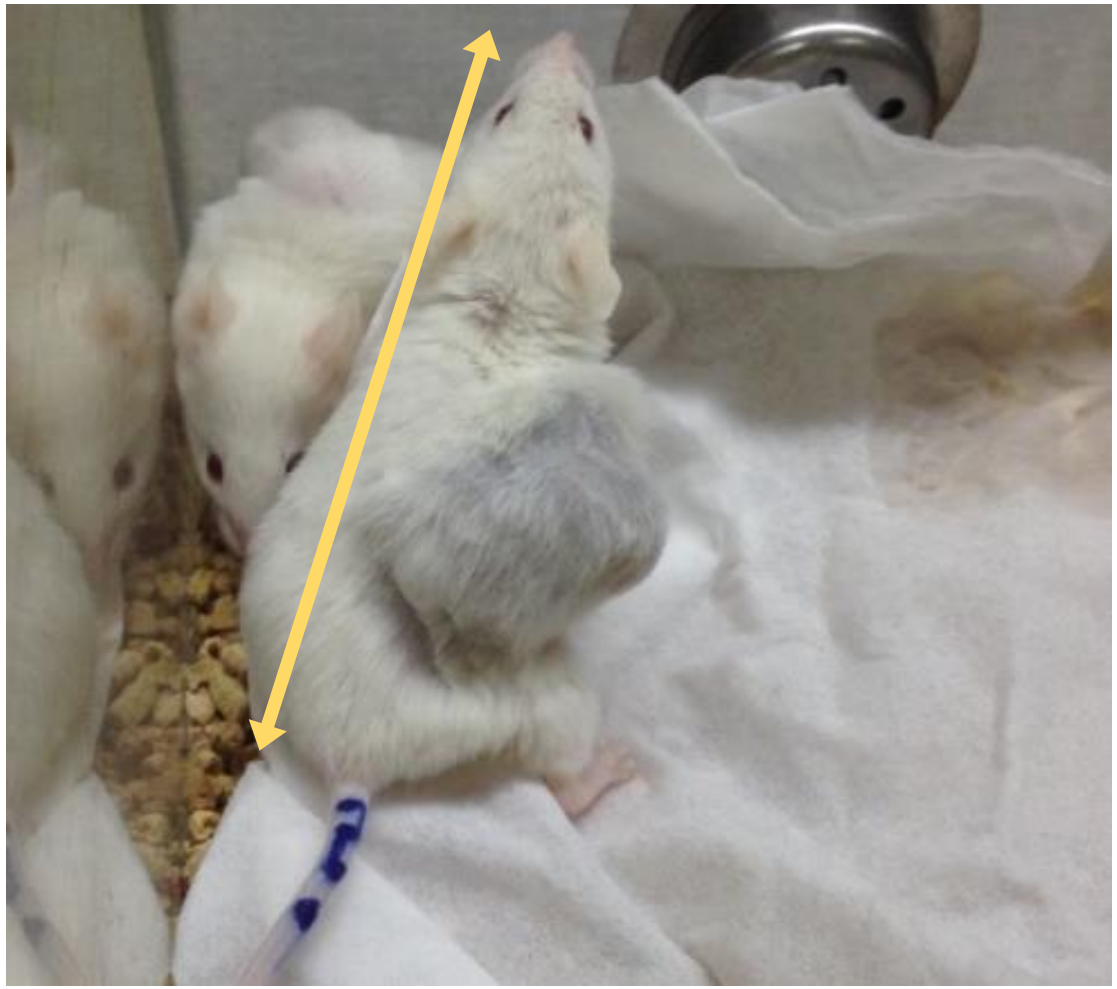
Neoplasia Endpoints (NIH, ARAC, 2011)	
1	A tumor burden greater than 10% body weight. In an adult mouse, a tumor should not exceed 20 mm in any one dimension; in an adult rat, a tumor should not exceed 40 mm in any one dimension
2	Tumors that ulcerate, become necrotic, or infected
3	Tumors that interfere with eating or impair ambulation

Humane Endpoints:

- Tissue distension - Pain & disability
- Paraneoplastic syndromes - Cachexia
- Ulceration - Infection, necrosis, loss of body fluid
- Lethality - Aggressive tumors

Burkholder et al., Curr Protoc Mouse Biol., 2012; James Wallace, ILAR J, 2000

腫瘤大小是否洽當?



3000 mm^3

腫瘤體積 = **(寬)^2 x 長 / 2**

Tumor size (Subcutaneous/ visible) - 2015

University	Diameter	
	Mouse	Rat
UMDNJ	1 cm <	3 cm <
UC Berkeley	1.5 cm <	2.5 cm <
IOWA State Univ.	1.6 cm < (25g)	3.6 cm < (250g)
UPenn, URI, UCSF Thomas Jefferson Univ.	2 cm <	4 cm <
Tumor size	1700~2000mm^3	3400~5000mm^3

Tumor Size (Subcutaneous/ visible) - 2020

University	Tumor size is gone Diameter in any direction	
	Mouse	Rat
Johns Hopkins	2cm <	4cm <
North Carolina at Chapel Hill	2cm <	4cm <
UCSF	2cm <	4cm <
UC Berkeley	1.5cm <	2.5cm <
IOWA State	1.5cm < ($2000mm^3$)	2.1cm < ($5000mm^3$)

in vivo *31*: 1073-1080 (2017)
doi:10.21873/invivo.11172

大鼠膀胱癌

Implementation of Humane Endpoints in a Urinary Bladder Carcinogenesis Study in Rats

MÓNICA OLIVEIRA[1], ELISABETE NASCIMENTO-GONÇALVES[2], JESSICA SILVA[2], PAULA A. OLIVEIRA[2,3], RITA FERREIRA[4], LUÍS ANTUNES[2,3], REGINA ARANTES-RODRIGUES[3,5] and ANA I. FAUSTINO-ROCHA[3,6]

Chemical Induced Bladder Carcinoma in Wistar Rats

		Response to manipulation
	0	Normal
	1	Stress response to manipulation (signs of discomfort, vocalization)
Behavior	2	Absence of response (lethargic animal)
		Breathing
	0	Normal
	1	Tachypnea
		Hydration status (skin pinch test)
	0	Normal
	1	Abnormal
Clinical signs		**Body temperature**
	0	Normal (35.6- 38.9°C)
	1	Hyperthermia (> 38.9°C)
	2	Hypothermia (< 35.6°C)
		Urine color
	0	Normal
	1	Hematuria (+)
	2	Hematuria (++)
Urine	3	Hematuria (+++)
	4	Hematuria (++++)
	5	Hematuria (+++++)
		Urine volume
	0	Normal
	1	Oliguria
	2	Anuria

血尿程度

尿液量

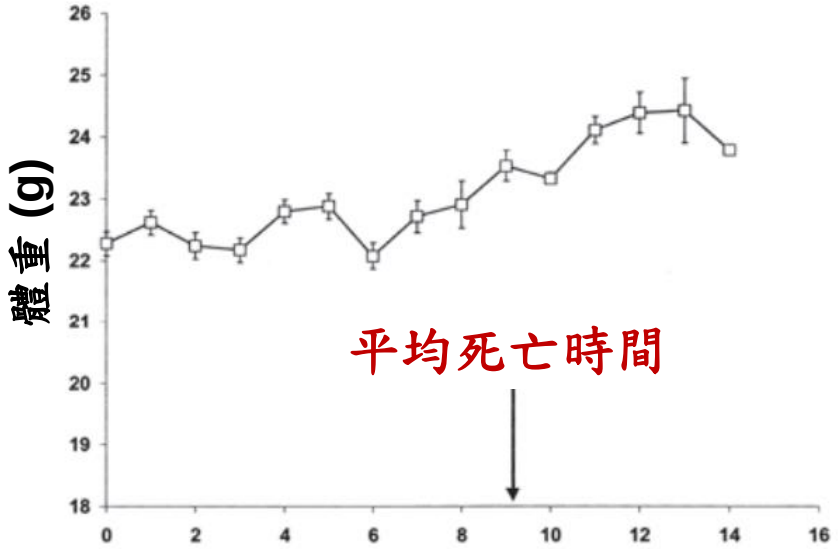
Oliveira et al., In Vivo, 2017

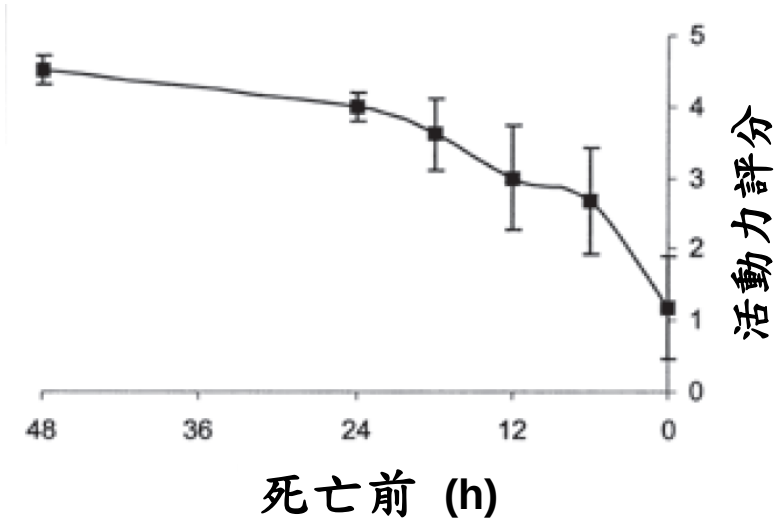
Determination of a Humane Endpoint in the L1210 Model of Murine Leukemia 小鼠白血病

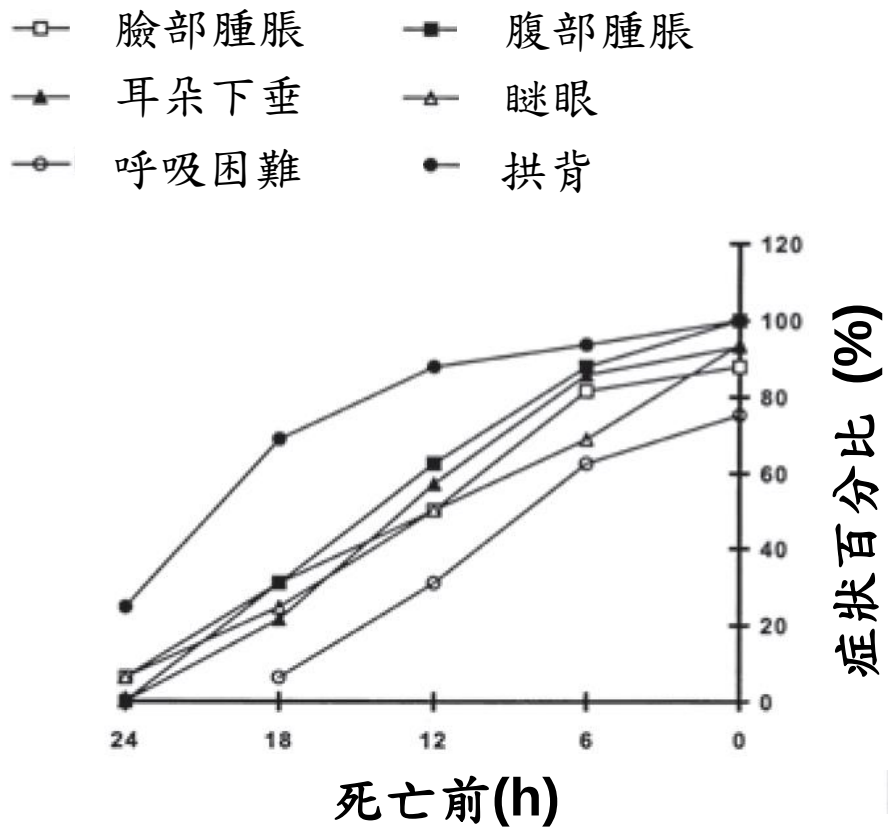
ANDREW J. ALDRED, BSC, MING C. CHA, PHD, AND KELLY A. MECKLING-GILL, PHD*

Contemp Top Lab Anim Sci., 2002

Murine Leukemia in B6D2F1/CrlB Mice







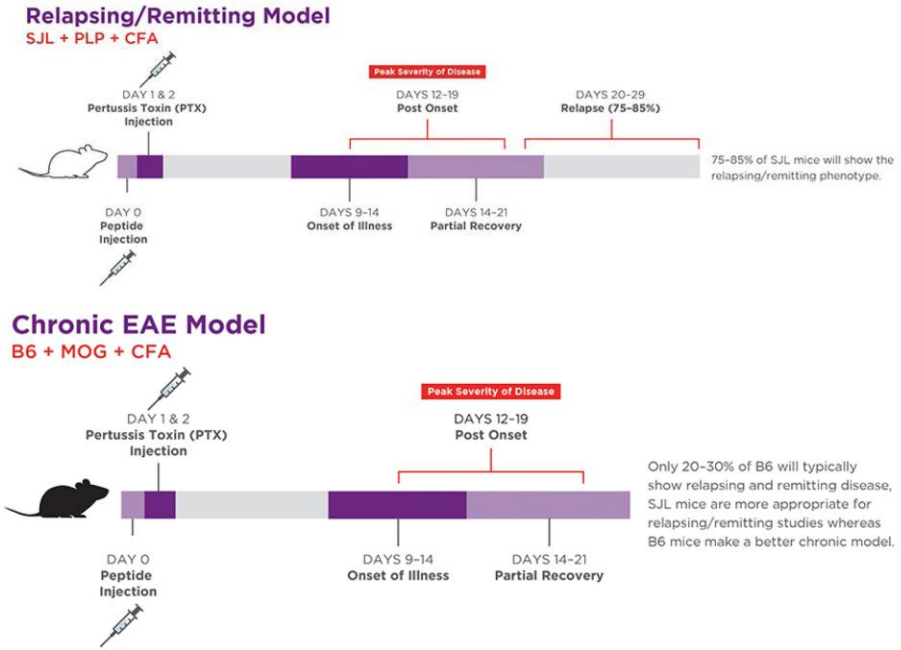
Aldred et al., Contemp Top Lab Anim Sci, 2002

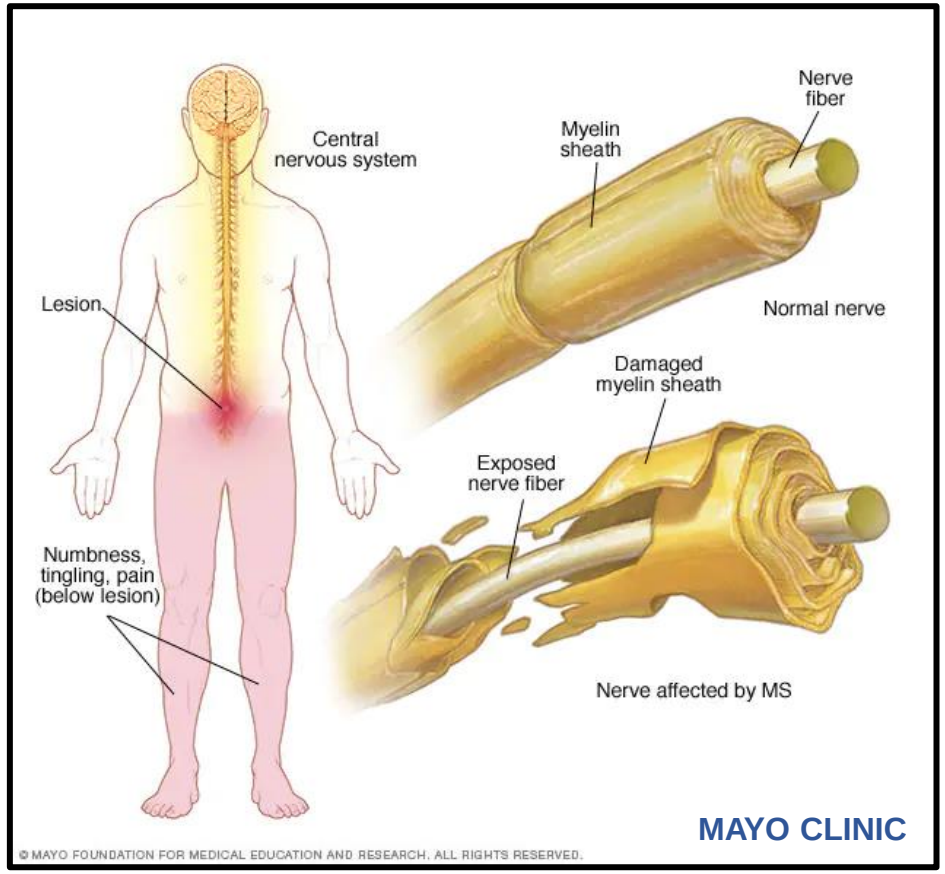
EAE Model for 多發性硬化症

Know your model !!!

SJL mice: 症狀一開始會很嚴重，但會趨於緩和。

B6 mice: 隨著時間漸進性變嚴重。





Hickman, OLAW, Sep. 20, 2018

0-5 0-6 0-10

Table 1. Examples of experimental autoimmune encephalomyelitis (EAE) scoring systems.

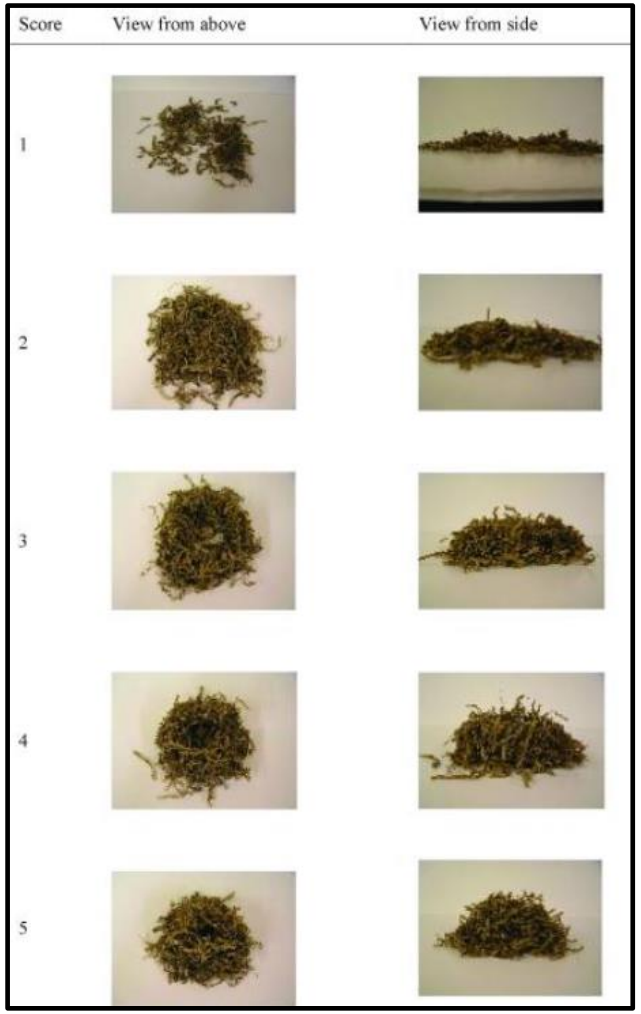
	Miller (2007)[16]	Bachmann (1999)[17]	Axial-rotatory EAE[20]		Bebo (1998)[34]	Bittner (2014)[35]	Expanded disability status scale (MS in patients)[3]
0	No clinical signs	No clinical signs	No clinical signs	0	No clinical signs	No clinical signs	No clinical signs
0.5		Distal limp tail		1	Minimal hind limb weakness	Partial limp tail	No impairment
1	Limp tail or hind limb weakness	Limp tail	Mild tilting of the head	2	Moderate hind limb weakness or mild ataxia	Paralysed tail	Minimal impairment
1.5		Limp tail and hind limb weakness		3	Moderate severe hind limb weakness	Hind limb paresis	Moderate impairment
2	Both limp tail and limb weakness	Unilateral partial hind limb paralysis	Marked tilting of the head	4	Severe hind limb weakness or mild forelimb weakness or moderate ataxia	Hind limb paraplegia	Severe impairment
2.5		Bilateral partial hind limb paralysis		5	Paraplegia with moderate forelimb weakness	Both hind limbs paralysed	Walking restricted to <200 m
3	Partial hind limb paralysis	Complete bilateral hind limb paralysis	Tilting of the body	6	Paraplegia with severe forelimb weakness or severe ataxia	Quadriparesis	Constant assistance
3.5		Complete bilateral hind limb paralysis and partial forelimb paralysis		7		1 forelimb paralysed	Wheelchair bound
4	Complete hind limb paralysis	Total paralysis of hind and forelimbs	Continuous axial rotation	8		Quadriplegia	Bed bound
4.5		Moribund		9		Moribund	Helpless bed patient
5	Death	Death	Death	10		Death	Death

MS: multiple sclerosis.

Palle et al., Lab Anim., 2016

Humane Endpoints for EAE Model

- 體重 & BCS
- 水和 – 皮膚回彈時間 & 血液學檢查
- 行為學 – 理毛 & 築巢



Hess et al., J Am Assoc Lab Anim Sci., 2008

什麼是疼痛反應?

除非相反的結論被證實，我們應該考量任何**會造成人疼痛的過程也會對其它動物造成疼痛**。

IRAC 1985 & The Guide 8th

Interagency Research Animal Committee

Unless the contrary is known or established, it should be considered that procedures that **cause pain in humans may also cause pain in other animals.**

人道終止點提示

1. 文獻探討/搜尋 https://www.humane-endpoints.info/en#
   (標準會改變!)

2. 熟悉動物疼痛與緊迫的症狀

3. 預測在你實驗動物模式上可能發生的各種併發症

4. 評分&累計系統

5. 諮詢獸醫師

6. 如果對人道中止點有歧見，IACUC、研究人員與獸醫師應該一同參予討論

安樂死

http://thedailycorgi.com/bereavement-suppor

Euthanasia

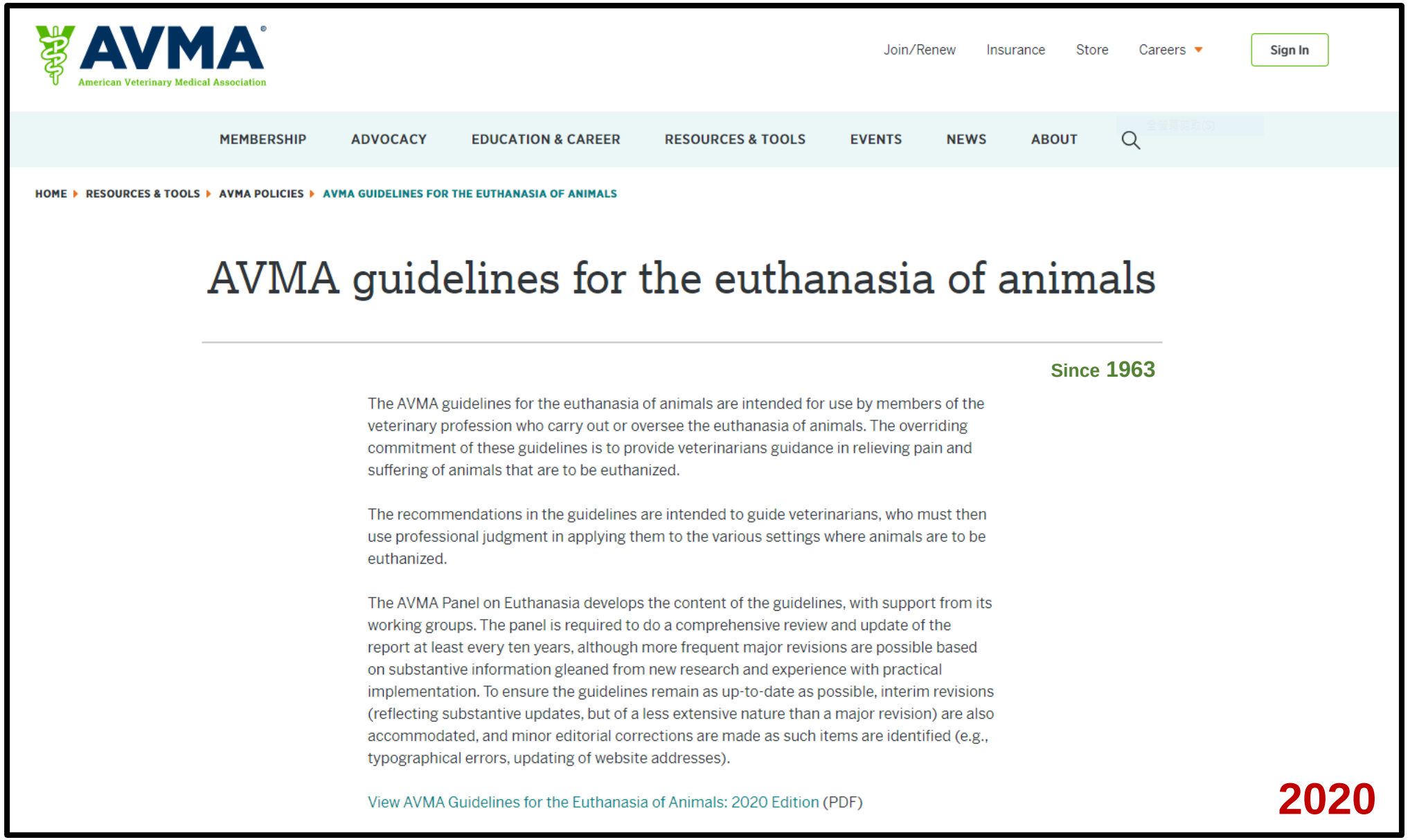
εὐθανασία

動物何時應該安樂死?

Brown University

- IACUC
- 研究人員
- 獸醫師

- 疼痛與緊迫臨床症狀
- 人道中止點
- 不可逆症狀 (瀕死 & 水腦)



https://www.avma.org/resources-tools/avma-policies/avma-guidelines-euthanasia-animals

安樂死方法評估

- 誘發失去意識與死亡時產生最輕微疼痛與緊迫的能力
- 造成意識喪失的時間
- 可靠性
- 人員安全
- 不可逆
- 與動物使用與目的相容
- 紀錄對觀察者或操作者情感上的影響
- 對後續評估、檢驗或組織使用的相容性
- 藥物取得與是否有人類濫用的可能性
- 對物種、年齡與健康狀態的相容性
- 設備能維持正常運作的能力
- 若動物屍體會被掠食者或清除者食用的安全性
- 法律要求
- 方法或動物屍體廢棄對環境的影響

人類行為

- **尊重人與動物之間的關係**
- 適合執行安樂死的地方
- 讓參予者知道過程中會觀察到哪些狀況
- 執行者有足夠技巧
- 情感支持 & 哀傷輔導
- 研究機構 – 動物照護員、獸醫師、研究人員
- **IACUC** – 應制定政策且確保訓練完善

https://www.avma.org/one-health/human-animal-bond

安樂死的機制

1. 直接抑制生命功能必須的神經元 (Vocalization, muscle contraction)

2. 缺氧 (Motor activity, convulsions)

3. 物理性阻斷大腦功能 (Motor activity, convulsions)

理想狀態下，安樂死的方法應該要快速造成意識喪失，接著是心肺功能停止，最後則是大腦功能停止。

安樂死方法分類

可接受：單獨使用時穩定性高。

To produce a humane death consistently when used as the sole means of euthanasia.

有條件可接受：非完美需要其他條件輔助。

It may require certain conditions to be met to consistence humane death, e.g., greater potential for operator error or safety hazard, not well documented in the scientific literature, require a secondary method to ensure death.

不可接受：任何狀況下都無法達成人道犧牲。

Methods **deemed inhumane under any conditions** or that the POE found **posed a substantial risk to the human applying the technique**

安樂死方法

吸入型: 氣體麻醉劑, CO, N_2, Ar, CO_2

非吸入型: Barbituric Acid Derivatives, Pentobarbital Combinations, MS 222 … etc.

物理性: 頸椎脫臼, 斷頭, 電擊 ...etc.

(二)、適用之安樂死方法

1.陸生動物-嚙齒類、兔、狗、貓、猿猴

安樂死方法	小於 200 g 嚙齒動物	200 g~ 1 kg 嚙齒動物/兔	1 kg~ 5 kg 嚙齒動物/兔	狗	貓	猿猴
一、化學性方法						
二氧化碳	○	○	×	×	×	×
鎮定後二氧化碳	○	○	○	×	×	×
Barbiturate 注射液，靜脈注射（麻醉劑量的 3 倍劑量）	○	○	○	○	○	○
Barbiturate 注射液，腹腔注射（麻醉劑量的 3 倍劑量）	○	○	○	×	○	×
深度麻醉後採血(放血)致死	○	○	○	○	○	○
深度麻醉後靜脈注射 KCl (1-2 meq/kg)或神經肌肉阻斷劑	○	○	○	○	○	○

10-30% of the chamber volume/min

農委會-實驗動物照護及使用指南(2018)

(二)、適用之安樂死方法

1.陸生動物-嚙齒類、兔、狗、貓、猿猴

安樂死方法	小於 200 g 嚙齒動物	200 g~ 1 kg 嚙齒動物/兔	1 kg~ 5 kg 嚙齒動物/兔	狗	貓	猿猴
二、物理性方法						
深度麻醉後斷頭	○	○	×	×	×	×
深度麻醉後頸椎脫臼	○	○	×	×	×	×
動物清醒中直接斷頭	★	★	×	×	×	×
動物清醒中直接頸椎脫臼	★	×	×	×	×	×
電昏後放血致死	×	×	×	×	×	×

說明：1. ○：建議使用的方法；×：不得使用的方法。★：一般情況不推薦使用，除非實驗需要（需說明於動物實驗申請表，由 IACUC 審核通過並現場確認操作人員之技能）。

2.Barbiturate 巴比妥鹽類最常使用藥劑為 pentobarbital 注射劑，屬管制藥品，需事先洽行政院衛生福利部食品藥物管理署申請核可後購買使用。

農委會-實驗動物照護及使用指南(2018)

AVMA (2020)

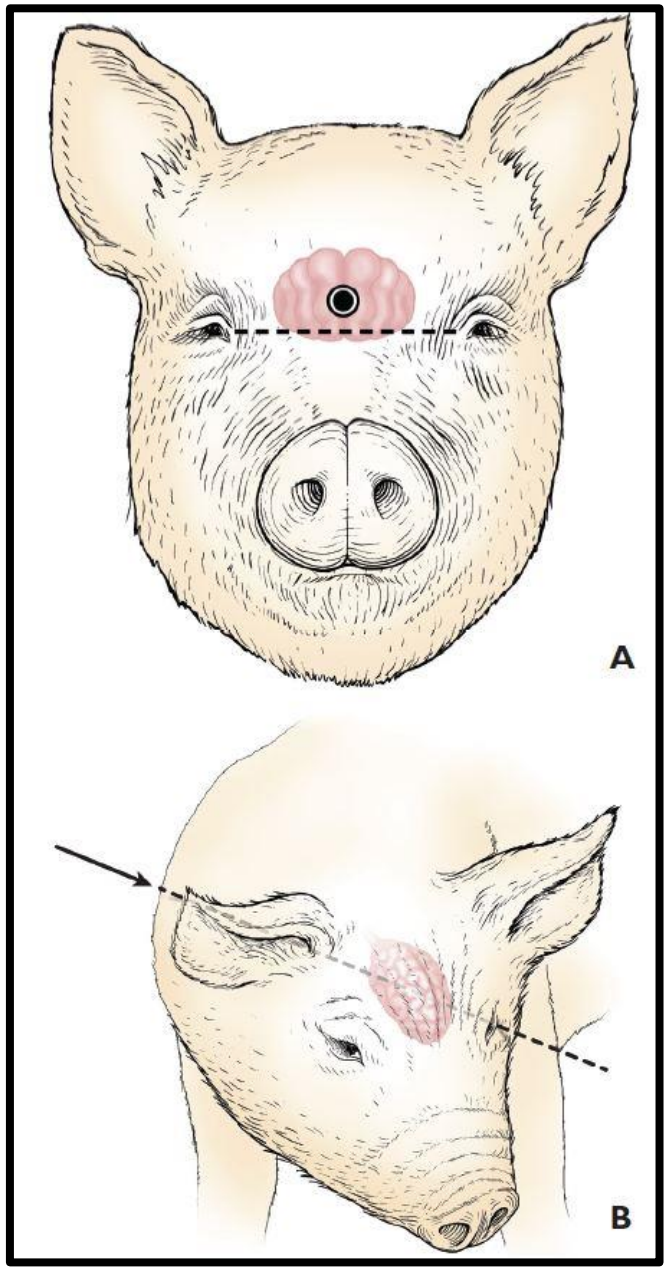
安樂死方式		
動物別	可接受	有條件可接受
小型嚙齒類	1. Barbiturate & barbituric acid derivatives: 麻醉劑量 3 倍 2. Dissociative agent combinations: Ketamine + Xylazine or diazepam	1. Isoflurane 2. CO_2: 成年 30-70%體積置換率/分鐘，未成年耐受性強 深度昏迷後併用頸椎脫臼或斷頭 3. 頸椎脫臼: 避免化學藥物汙染組織，且體重<200g 4. 斷頭: 避免化學藥物汙染組織 5. 低溫: 僅適用於小於 10 日齡之小鼠與大鼠，不可直接接觸低溫源，且應有二次犧牲法確認，如斷頭與頸椎脫臼 6. 急速冷凍(液態氮): 僅適用於小於 5 日齡之小鼠與大鼠
兔子	Barbiturate & barbituric acid derivatives: 可先行鎮靜	1. Isoflurane 2. CO_2: 50-60%體積置換率/分鐘，建議先行給予鎮靜。

AVMA (2020)

安樂死方法	可接受	有條件可接受	輔助
仔豬	Barbiturates & barbituric acid derivatives	1. CO_2 2. NPCB 3. Manually applied blunt force trauma	NA
保育豬 （小於32kg）		1. CO_2 alone or combined with N_2/Ar 2. NPCB 3. PCB 4. Electrocution	NA
生長–肥育豬		1. CO_2, N_2, NO, Ar 2. Gunshot 3. PCB 4. Electrocution	1. Exsanguination 2. Pithing

Non-Penetrating Captive Bolt Device (NPCB)

Walsh et al., Animals(Basel), 2017



AVMA (2020), p119

AVMA (2020)

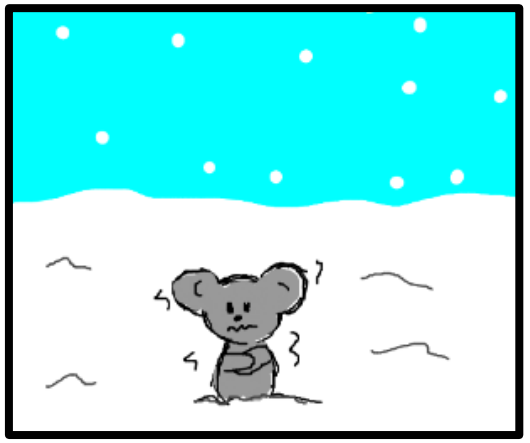
安樂死方法	可接受	有條件可接受	輔助
禽類 雞、火雞、鵪鶉、雉、鴨、雁	Barbiturates & barbituric acid derivatives	1. CO_2, CO, N_2, Ar 2. Reduction of atmospheric pressure 3. Cervical dislocation, 4. Decapitation 5. Manually applied blunt force trauma (≤ 16Kg) 6. Electrocution 7. Gunshot 8. PCB and NPCB	1. KCl or $MgSO_4$ (IV/Intracardiac) 2. Exsanguination
新生動物		1. Decapitation 2. Cervical dislocation 3. Cranial compression 4. Maceration (less than 72hr of age old)	NA
蛋		1. CO_2 for 20min 2. 4hr at 40°F or freezing (before 80% of incubation) 3. Egg addling 4. Maceration	NA

死亡確認

丟棄任何屍體前必須(應)確認動物已死亡。

2nd犧牲法 – 嚙齒類

1. 頸椎脫臼
2. 雙側性開胸(Rat ≥ 200g)



https://drawception.com/game/eMLcDpEeSW/cold-mouse/

其它動物:

無脈搏、呼吸、角膜反側、對捏腳趾無反應;透過聽診器無法聽到呼吸音與心跳; 粘膜面變白與屍僵。僅有**屍僵**能當作唯一死亡確認。

CO_2使用注意事項

- 開每分鐘體積置換率: **10-30%** (農委會)
  30-70% (AVMA guideline)
- **不要**預先充氣
- 原飼養籠比較好，緊迫產生的較少
- 二次犧牲法
- **5000 ppm** 對房內操作者有危害

附件四 常見實驗動物的最小建議空間需求

表 4.1 常見的實驗用齧齒類動物在群飼狀態下的最小建議空間需求*

動物類別	體重，公克	底面積/隻，[a] 平方英吋.(平方公分)	高度，[b] 英吋.(公分)	附註說明
群飼小鼠 [c]	<10 至 15 至 25 >25	6 (38.7) 8 (51.6) 12 (77.4) ≥15 (≥96.7)	5 (12.7) 5 (12.7) 5 (12.7) 5 (12.7)	體型較大的動物可能需要更大的空間，以符合成效標準要求。
帶仔小鼠		51 (330) (群飼狀態下的空間需求)	5 (12.7)	其他的飼養規格可能需要較大的空間，空間需求調整將依成鼠及仔鼠的隻數、及仔鼠的體型與年齡而定。[d]
群飼大鼠 [c]	<100 至 200 至 300 至 400 至 500 >500	17 (109.6) 23 (148.35) 29 (187.05) 40 (258.0) 60 (387.0) ≥70 (≥451.5)	7 (17.8) 7 (17.8) 7 (17.8) 7 (17.8) 7 (17.8) 7 (17.8)	體型較大的動物可能需要更大的空間，以符合成效標準要求。
帶仔大鼠		124 (800) (群飼狀態下的空間需求)	7 (17.8)	其他的飼養規格可能需要較大的空間，空間需求調整將依成鼠及仔鼠的隻數、及仔鼠的體型與年齡而定。[d]
倉鼠 [c]	<60 至 80 至 100 >100	10 (64.5) 13 (83.8) 16 (103.2) ≥19 (≥122.5)	6 (15.2) 6 (15.2) 6 (15.2) 6 (15.2)	體型較大的動物可能需要更大的空間，以符合成效標準要求。
天竺鼠 [c]	至 350 >350	60 (387.0) ≥101 (≥651.5)	7 (17.8) 7 (17.8)	體型較大的動物可能需要更大的空間，以符合成效標準要求。

*在應用解讀此表格時，應該將本指引所提及的成效指標一併列入考量。

[a] 單獨或小族群飼養的個體，其單隻所需的空間需求可能會大於由群居建議值換算出的數值。

[b] 從盒底到蓋子的高度。

[c] 應該將各品系或品種動物的生長特性、性別等因素列入考慮。對於增重較快的動物，可能需以

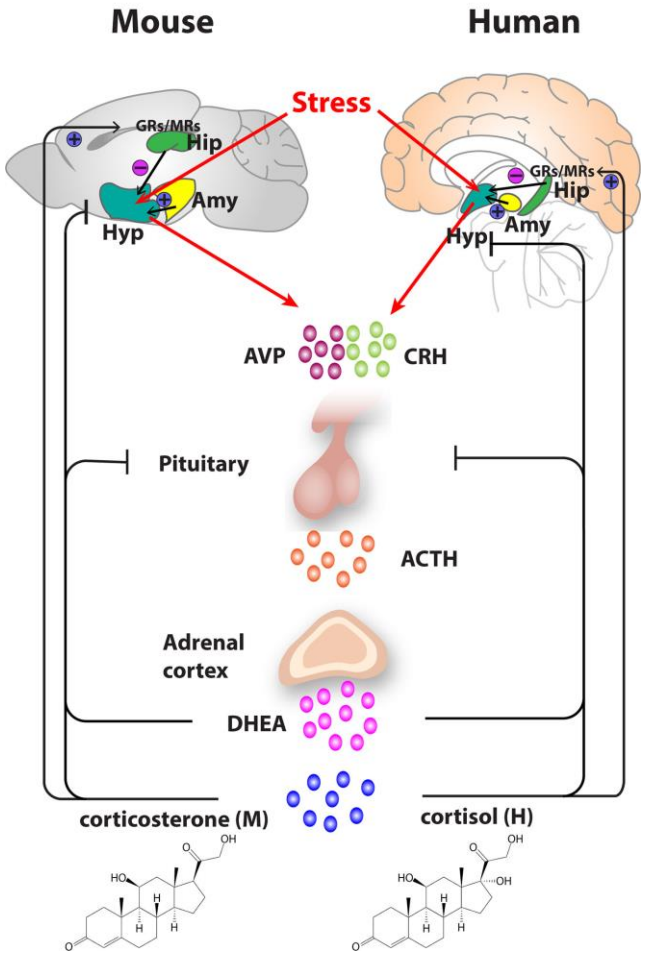
密度過高

焦慮&壓力!!!

不要在動物飼養房內犧牲動物或是在存活動物前犧牲動物

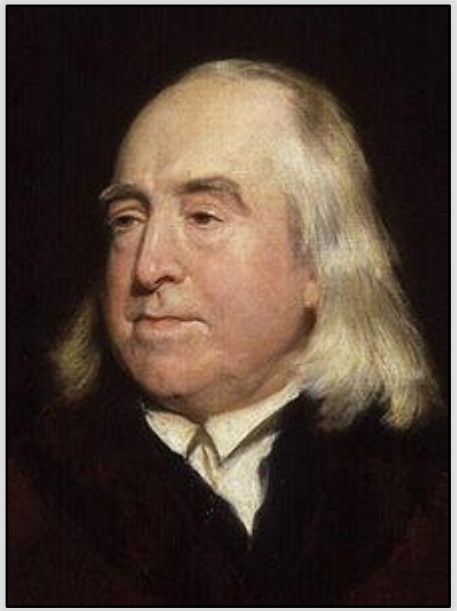
壓力：
- 超音波(超過人類可聽見音頻)
- 費洛蒙(人類聞不到)

結果：
- 改變生理數值 (實驗誤差)
- 超影響動物福祉



Nandam et al., Front. Psychiatry, 2020

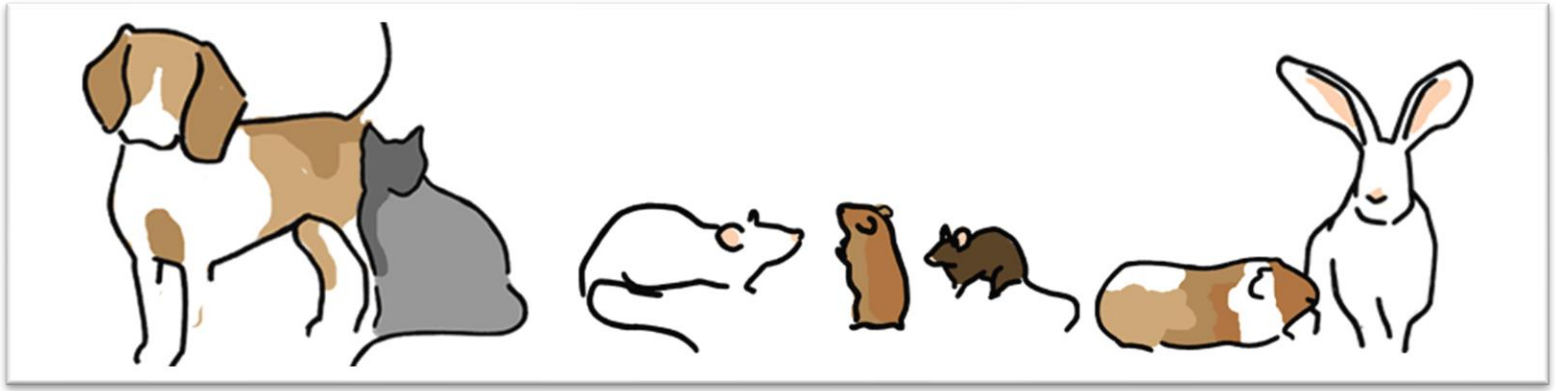
"The question is not, Can they reason? nor Can they talk? But can they suffer?"



Jeremy Bentham
1748-1832

IACUC審查常見缺失案例分享

簡旭哲 獸醫師

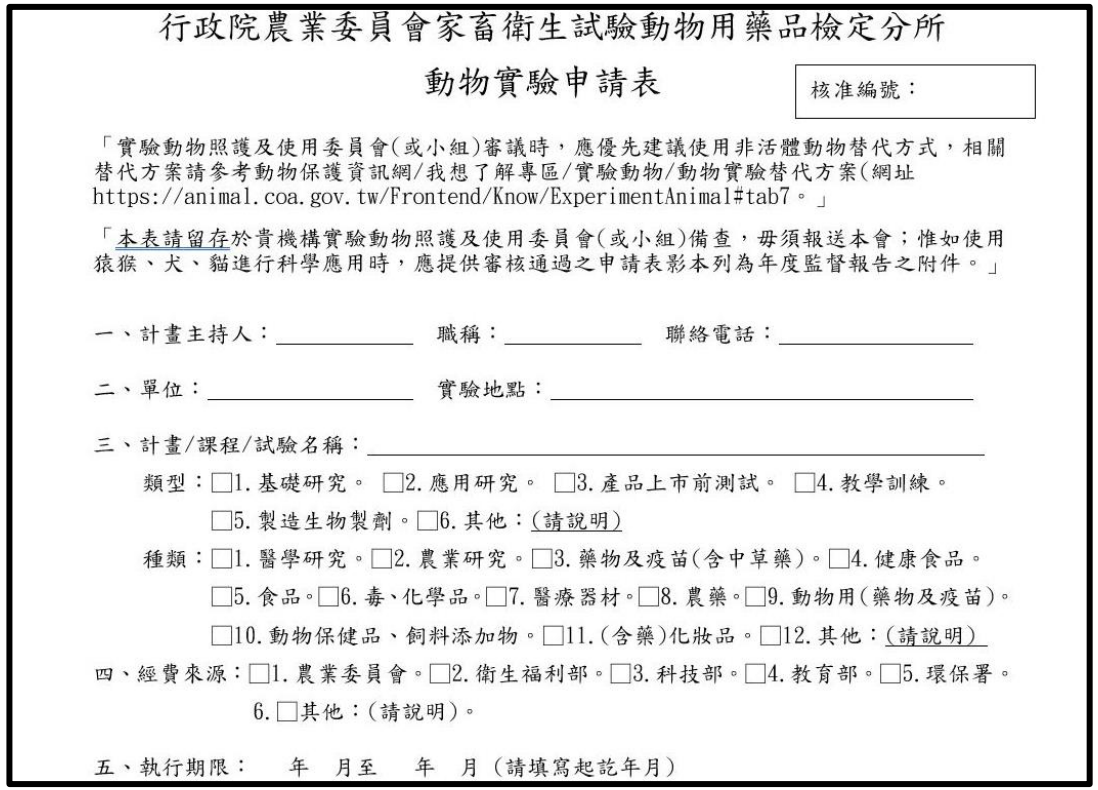


Über LAS interactive

大綱

1. 規範

2. 常見審查問題



行政院農業委員會家畜衛生試驗動物用藥品檢定分所

動物實驗申請表　　核准編號：

「實驗動物照護及使用委員會(或小組)審議時，應優先建議使用非活體動物替代方式，相關替代方案請參考動物保護資訊網/我想了解專區/實驗動物/動物實驗替代方案(網址 https://animal.coa.gov.tw/Frontend/Know/ExperimentAnimal#tab7。」

「本表請留存於貴機構實驗動物照護及使用委員會(或小組)備查，毋須報送本會；惟如使用猿猴、犬、貓進行科學應用時，應提供審核通過之申請表影本列為年度監督報告之附件。」

一、計畫主持人：＿＿＿＿　職稱：＿＿＿＿　聯絡電話：＿＿＿＿

二、單位：＿＿＿＿　實驗地點：＿＿＿＿

三、計畫/課程/試驗名稱：＿＿＿＿

類型：□1.基礎研究。□2.應用研究。□3.產品上市前測試。□4.教學訓練。
□5.製造生物製劑。□6.其他：(請說明)

種類：□1.醫學研究。□2.農業研究。□3.藥物及疫苗(含中草藥)。□4.健康食品。
□5.食品。□6.毒、化學品。□7.醫療器材。□8.農藥。□9.動物用(藥物及疫苗)。
□10.動物保健品、飼料添加物。□11.(含藥)化妝品。□12.其他：(請說明)

四、經費來源：□1.農業委員會。□2.衛生福利部。□3.科技部。□4.教育部。□5.環保署。
6.□其他：(請說明)。

五、執行期限：　年　月至　年　月（請填寫起訖年月）

IACUC委員計畫審核的職責與特殊考量

1. 使用動物進行研究應事先申請，內容包含計畫名稱、主持人、使用物種、品種、數量、實驗設計、執行期限、負責實驗人員、3Rs評估說明;經IACUC核可始得執行，變更亦同。

2. IACUC審議時應優先建議使用非活體，由一位以上具實驗動物專業或動物福祉背景，且非隸屬於該機構。

3. IACUC應提供關於動物實驗設計諮詢、訓練計畫、及動物相關SOP與飼養設施改善建議。

4. 申請案若與委員有利益衝突，應迴避該計畫審核。

IACUC委員計畫審核的職責與特殊考量

5. 使用犬、貓、猿猴核可計畫書，年度監督報告應列為附件。

6. 重覆多次存活手術應由IACUC核可。

7. 使用犬、貓、猿猴， IACUC應制定再應用、退休、康復、認養、安置之政策與監督機制。

8. 應使用醫藥級化學品，若使用非醫藥級需經IACUC核准。

9. 任何例外應由IACUC明確定義與評估。

IACUC委員計畫審核的職責與特殊考量

10. 對小型嚙齒類使用剪腳趾標示，應由IACUC核准。

11. 腫瘤模式、感染性實驗、疫苗攻毒、疼痛模式、創傷、單株抗體製備、毒理試驗、器官或系統衰竭及心血管休克模式等侵入性實驗，IACUC應仔細審查實驗中止與人道中止時機。

12. 保定裝置得在尺寸、設計及操作上有所考量，以減低動物不適、疼痛、或緊迫的感受，甚至對動物與人員傷害。

IACUC委員計畫書核定後監督職責(PAM)

1. 依動物保護法規，每半年應施行內部稽核1次，查核結果呈報IO；查核結果應列為年度監督報告附件，年度結束後三個月內報中央主管機關備查，副知所屬縣市機關。應保存結果6年。

2. 依指引督導機構。

3. 機構應建立動物福祉通報與調查制度。所有案件應予以紀錄、追蹤。

4. 內部查核至少包含: 軟體查核 (機構政策與職責、動物健康與照護及飼養管理)、硬體查核(動物飼養區、供應區、儀器與設備、動物手術或實驗場所) 。

IACUC委員計畫書核定後監督職責(PAM)

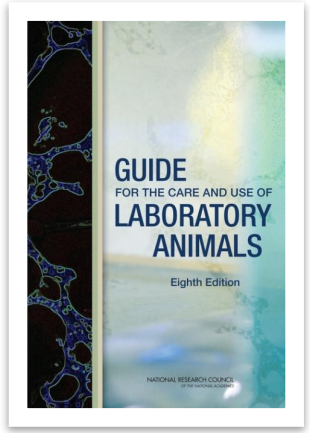
5. 應配合主管機關進行外部查核，IACUC招集人於現場引導說明，備妥文件: IACUC成立、異動、流程規章、5年內動物申請及審核紀錄、動物飼養管理SOPs、動物房設與坪數一覽表、5年內監督報告、5年內內外查核表。

6. 監督內容應包含機構動物取得、飼養、管理及是否依審核結果進行實驗。

7. 計畫核定後監督方式得包括: 持續性計畫審閱、實驗室稽核、由IACUC委員、動物照護者、獸醫師進行，或由外部管理部門進行查核與評鑑。

The Mission of IACUC

1. Protocol review
2. Experimental and humane endpoints
3. Unexpected outcomes
4. Physical restrain
5. Multiple survival surgical procedures
6. Food and fluid regulation
7. Use of non-pharmaceutical-grade chemicals and other substances
8. Field investigations
9. Agricultural animals



The Guide 8th

- ☐ Post-approval monitoring/PAM (Program)
- ☐ Disaster planning and emergency preparedness (Institution & IO)

Protocol review-1

- **Rationale and purpose** of the proposed use of animals

- A clear and concise sequential **description of the procedures** involving the use of animals that is **easily understood** by all members of the committee

- Availability or appropriateness of the use of less invasive procedures, other species, isolated organ preparation, cell or tissue culture, or computer simulation.

- **Justification of the species and number** of animals proposed; whenever possible, the number of animals and experimental group sizes should be statistically justified (e.g., provision of a power analysis)

- **Unnecessary duplication of experiments**

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Protocol review-2

- Nonstandard housing and husbandry requirements
- Impact of the proposed procedures on the animals' **well-being**
- Appropriate sedation, analgesia, and anesthesia (indices of pain or invasiveness might aid in the preparation and review of protocols)
- Conduct of surgical procedures, including multiple operative procedures
- Postprocedural care and observation (e.g., inclusion of post-treatment or postsurgical animal assessment forms)
- Description and rationale for anticipated or selected endpoints

The Guide 8th

Protocol review-3

- Criteria and process for timely intervention, removal of animals from a study, or euthanasia if painful or stressful outcomes are anticipated

- Method of euthanasia or disposition of animals, including planning for care of long-lived species after study completion

- Adequacy of training and experience of personnel in the procedures used, and roles and responsibilities of the personnel involved

- Use of hazardous materials and provision of a safe working environment.

常見審查問題

- **Nomenclature (命名學) – 嚙齒類動物**
- **使用動物數量**
- **實驗內容 – 投予&採樣、手術、飼料**
- **動物運輸**
- **危害物質 – turn on/of gene, chemotherapy, …..**
- **Genotyping or marking – tail snip/ toe clip/ ear punch**
- **疼痛與緊迫分類**
- **人道終止點**

Nomenclature (命名學)

B6, C57BL/6 or 小黑鼠

BALB/c or 小白鼠

七、實驗所需之動物：

	動物別/品系 a	使用量/年	動物來源 b	動物飼養場所 c	是否需要繁殖 d
1			例如：國內繁殖場(國家實驗動物中心)		
2					
3					

國動常見品系

	縮寫	全名
小鼠	BALB/c	BALB/cByJNarl
	B6	C57BL/6JNarl
	BALB/c Nude	BALB/cAnN.Cg-*Foxn1*^{nu}/CrlNarl
	NOD SCID	NOD.CB17-*Prkdc*^{scid}/CrlNarl
	ASID	NOD.Cg-*Prkdc*^{scid}/*Il2rg*^{tm1Wjl}/YckNarl

NARLabs 國家實驗研究院
國家實驗動物中心
National Laboratory Animal Center

樂斯科常見品系

	縮寫	全名
小鼠	BALB/c	BALB/cAnNCrlBltw
	B6	C57BL/6NCrlBltw
	BALB/c Nude	CAnN.Cg-*Foxn1*^{nu}/CrlBltw
	NOD SCID	NOD.CB17-*Prkdc*^{scid}/NcrCrlBltw
	Nu/Nu	Bltw:NU-*Foxn1*^{nu} (Outbred)

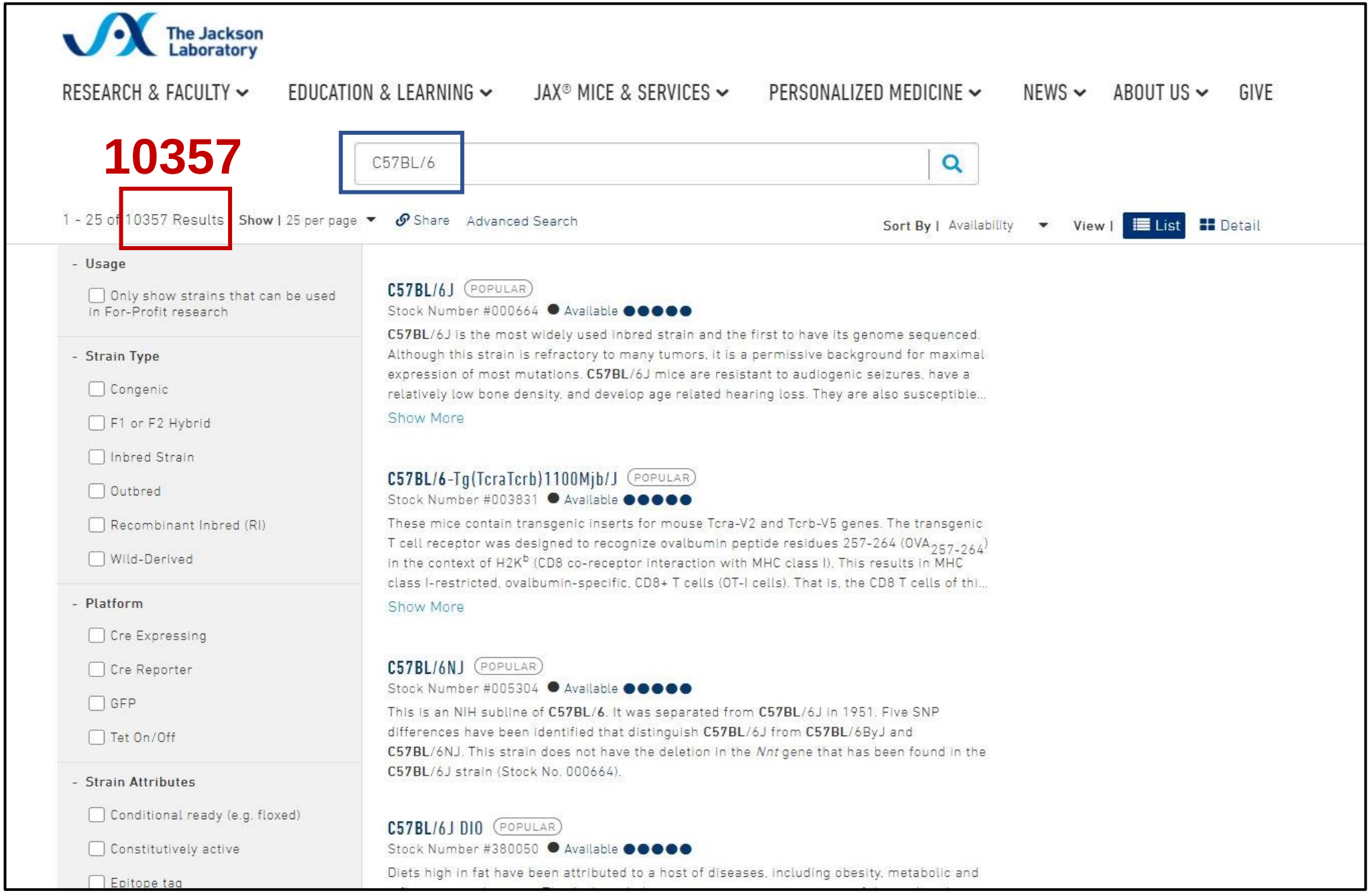
LASCO 樂斯科生物科技

C57BL/6J
C57BL/6NJ
C57BL/6JEiJ
C57BL/6HaJ

C57BL/6NHsd
C57BL6/JOlaHsd
C57BL/6JRccHsd

C57BL/6NCrl

C57BL/6NTac


The Jackson Laboratory
RESEARCH & FACULTY
EDUCATION & LEARNING
JAX® MICE & SERVICES
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NEWS
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C57BL/6
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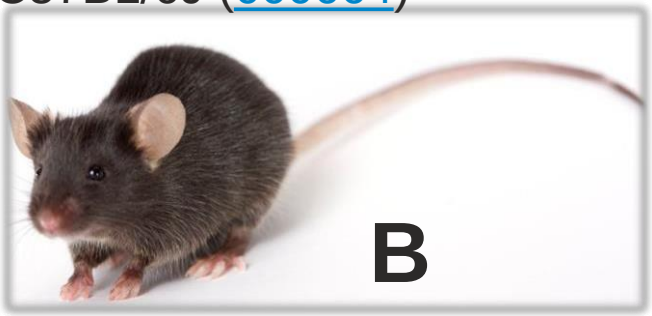
- Usage
Only show strains that can be used in For-Profit research
- Strain Type
Congenic
F1 or F2 Hybrid
Inbred Strain
Outbred
Recombinant Inbred (RI)
Wild-Derived
- Platform
Cre Expressing
Cre Reporter
GFP
Tet On/Off
- Strain Attributes
Conditional ready (e.g. floxed)
Constitutively active
Epitope tag
C57BL/6J POPULAR
Stock Number #000664 Available
C57BL/6J is the most widely used inbred strain and the first to have its genome sequenced. Although this strain is refractory to many tumors, it is a permissive background for maximal expression of most mutations. C57BL/6J mice are resistant to audiogenic seizures, have a relatively low bone density, and develop age related hearing loss. They are also susceptible...
Show More
C57BL/6-Tg(TcraTcrb)1100Mjb/J POPULAR
Stock Number #003831 Available
These mice contain transgenic inserts for mouse Tcra-V2 and Tcrb-V5 genes. The transgenic T cell receptor was designed to recognize ovalbumin peptide residues 257-264 ($OVA_{257-264}$) in the context of $H2K^b$ (CD8 co-receptor interaction with MHC class I). This results in MHC class I-restricted, ovalbumin-specific, CD8+ T cells (OT-I cells). That is, the CD8 T cells of thi...
Show More
C57BL/6NJ POPULAR
Stock Number #005304 Available
This is an NIH subline of C57BL/6. It was separated from C57BL/6J in 1951. Five SNP differences have been identified that distinguish C57BL/6J from C57BL/6ByJ and C57BL/6NJ. This strain does not have the deletion in the Nnt gene that has been found in the C57BL/6J strain (Stock No. 000664).
C57BL/6J DIO POPULAR
Stock Number #380050 Available
Diets high in fat have been attributed to a host of diseases, including obesity, metabolic and

How well do you know your B6 mice?

A & B are most similar!

B6(Cg)-*Tyr*$^{c-2J}$/J (000058)

C57BL/6J (000664)



C57BL/6NJ (005304)

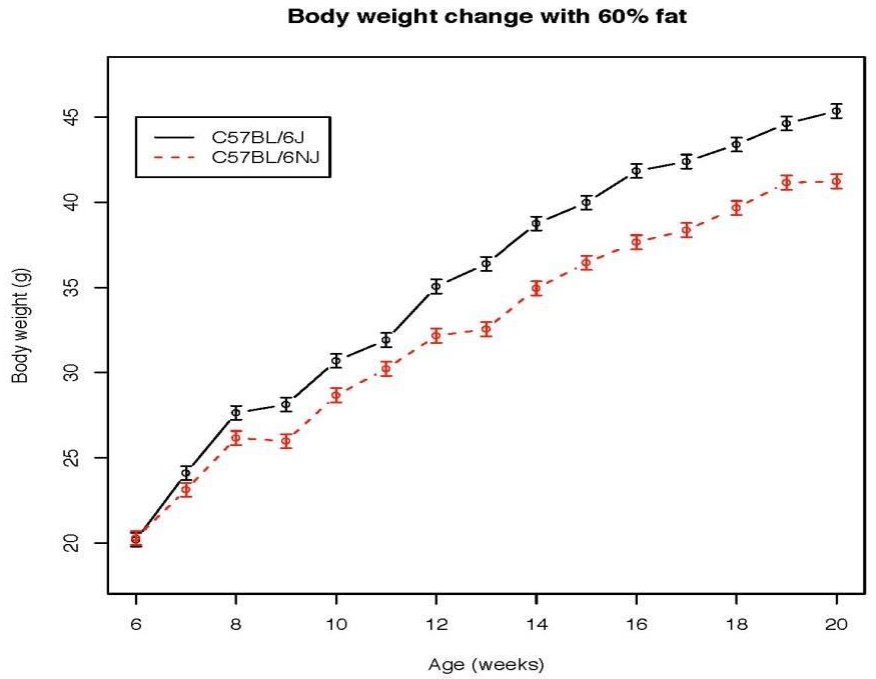
- A & B differ by a single allele (*Tyr*$^{c-2J}$)
- B & C differ in multiple alleles
  - Metabolism
  - Neurobiology
  - Immunology
  - Vision & hearing
  - Behavior

Metabolic Differences (DIO)

B6J gains more weight than B6NJ on high fat diet (HFD)

C57BL/6J (000664) vs C57BL/6NJ (005304)

- Mice fed a 60 kcal% high fat diet
  - Beginning at 6 weeks of age



Nicholson, A et al. 2010. *Obesity* 18(10): 1902-1905. PMID: 20057372

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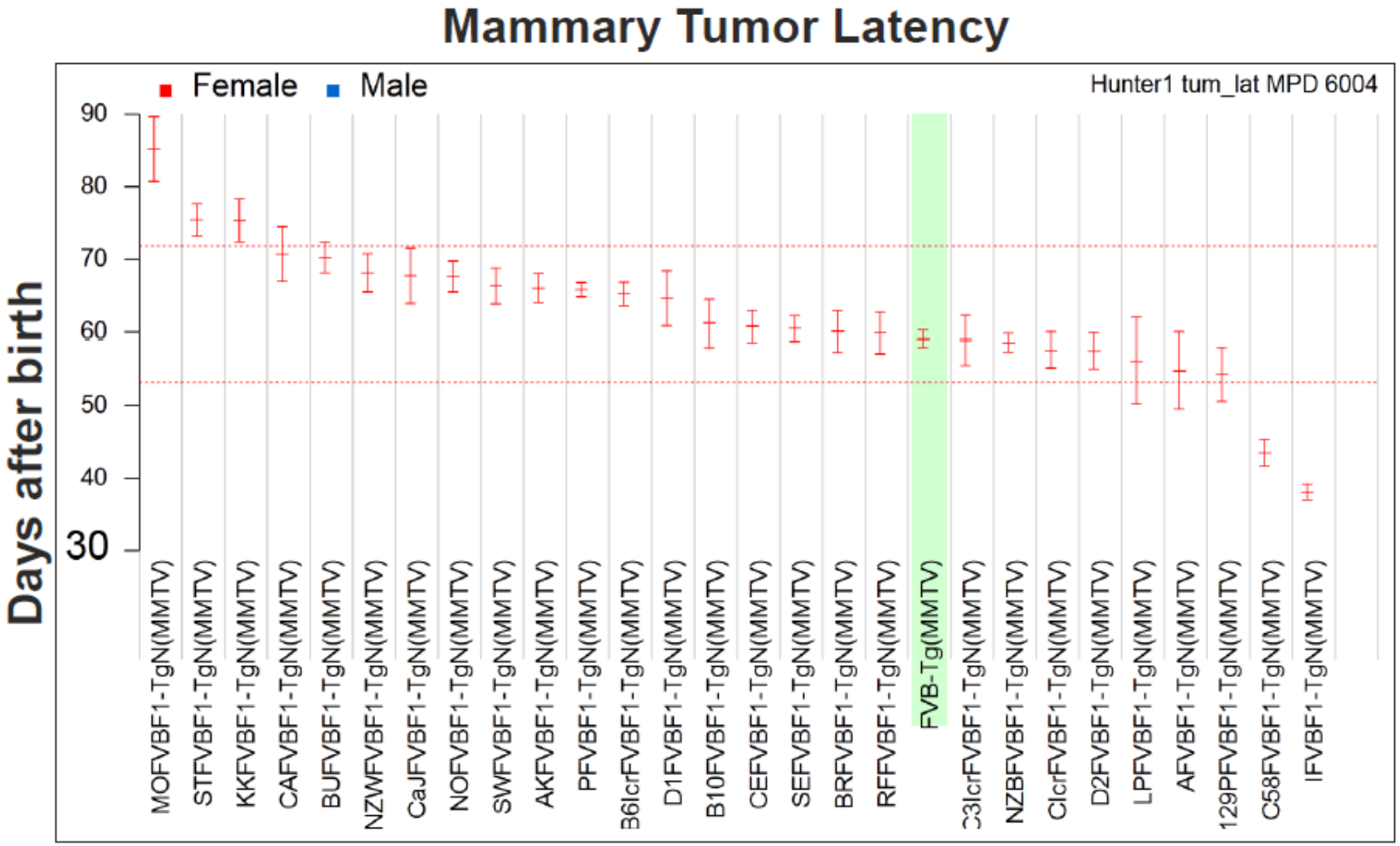
Genetic Background Has an Influence on Phenotype

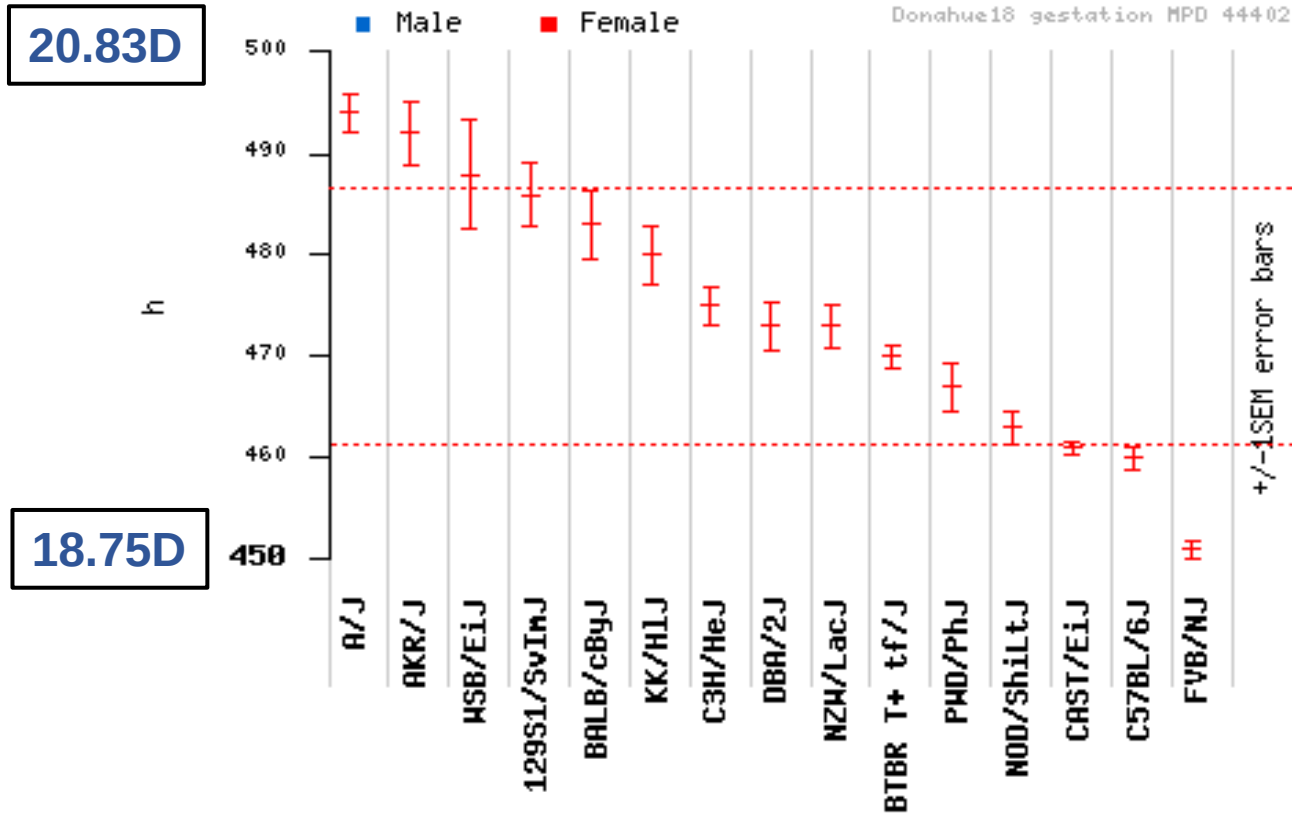


Mouse Phenome Database, Hunter1 dataset

JAX lab

Genetic Background Effects

Inbred strain gestation duration



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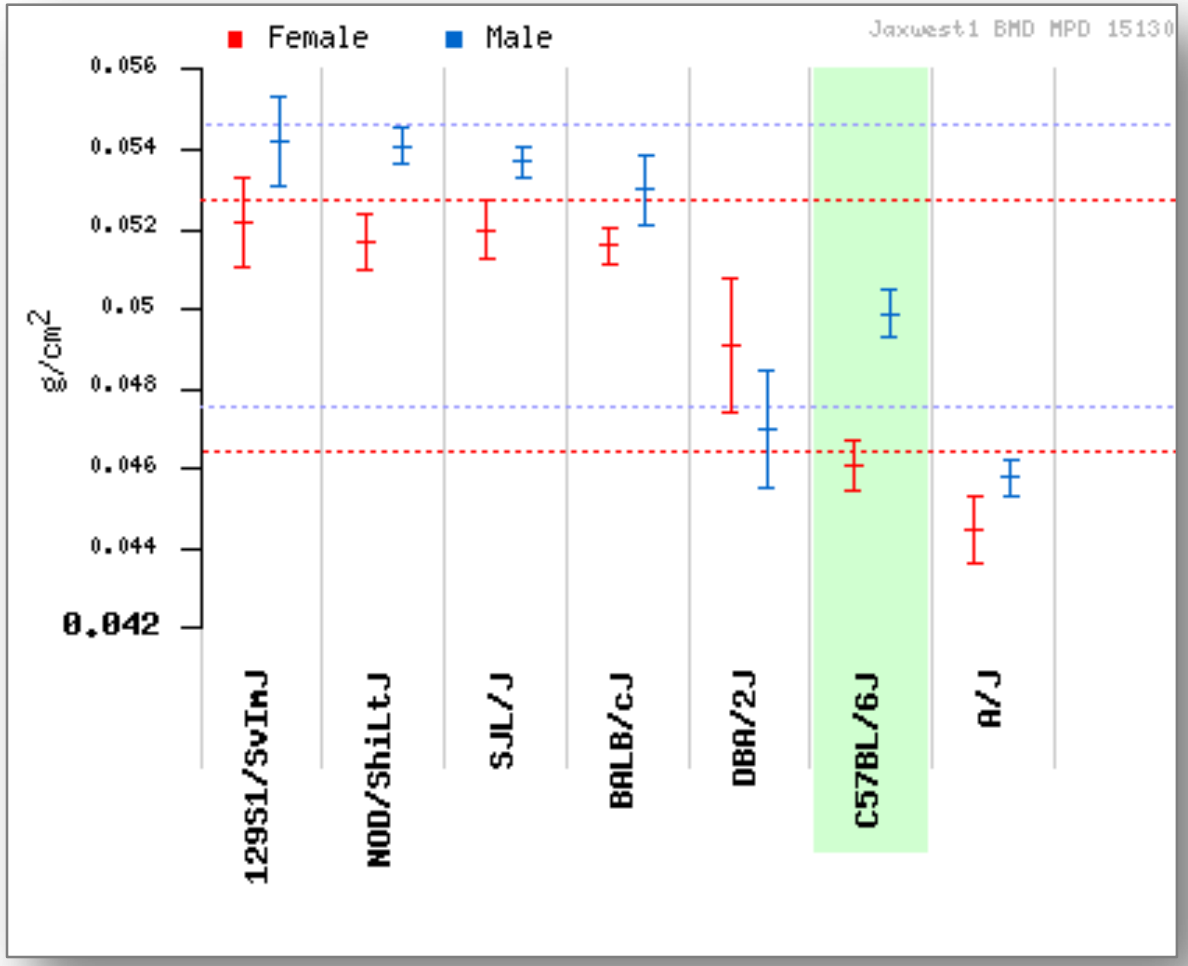
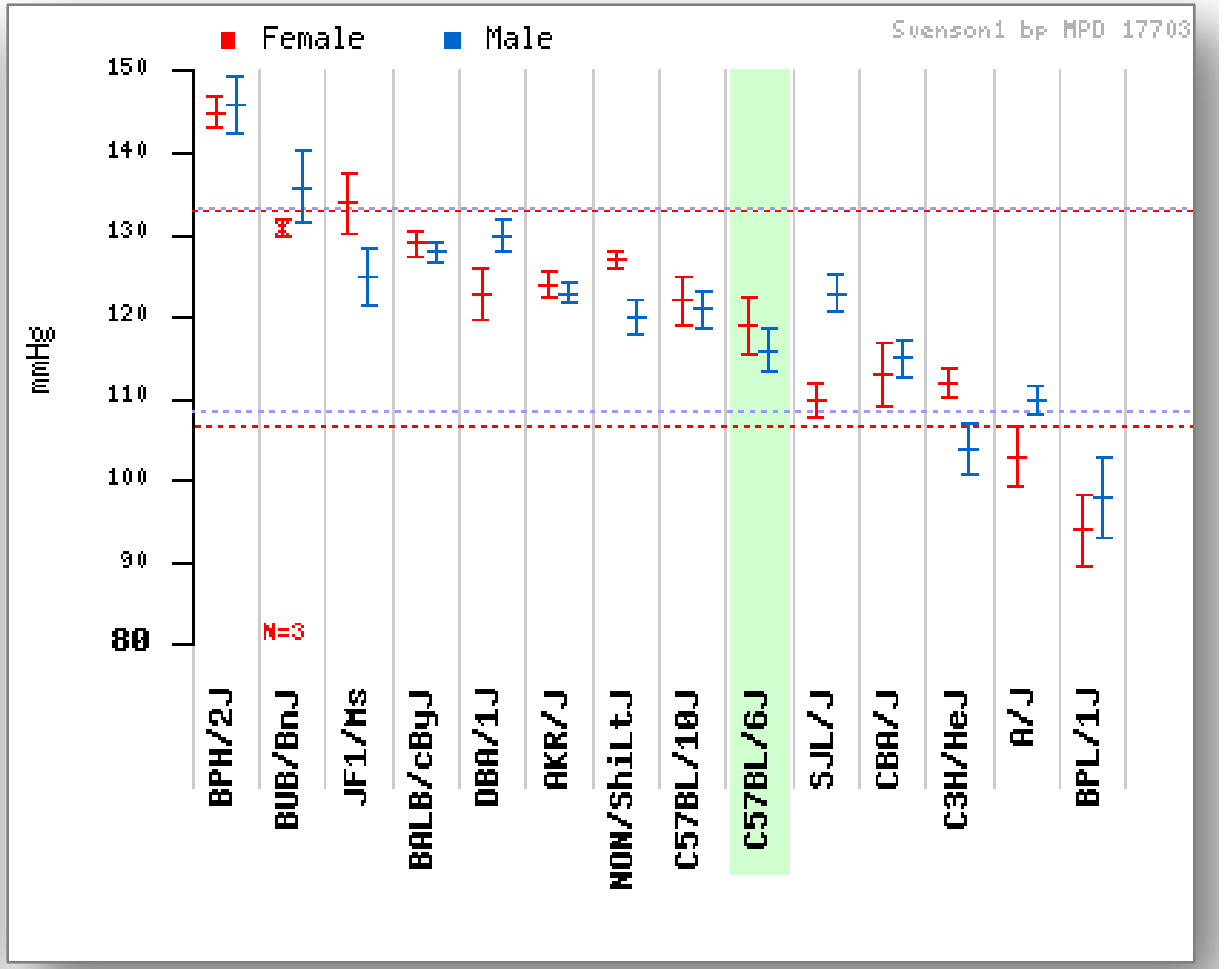
Bone Mineral Density



Blood Pressure



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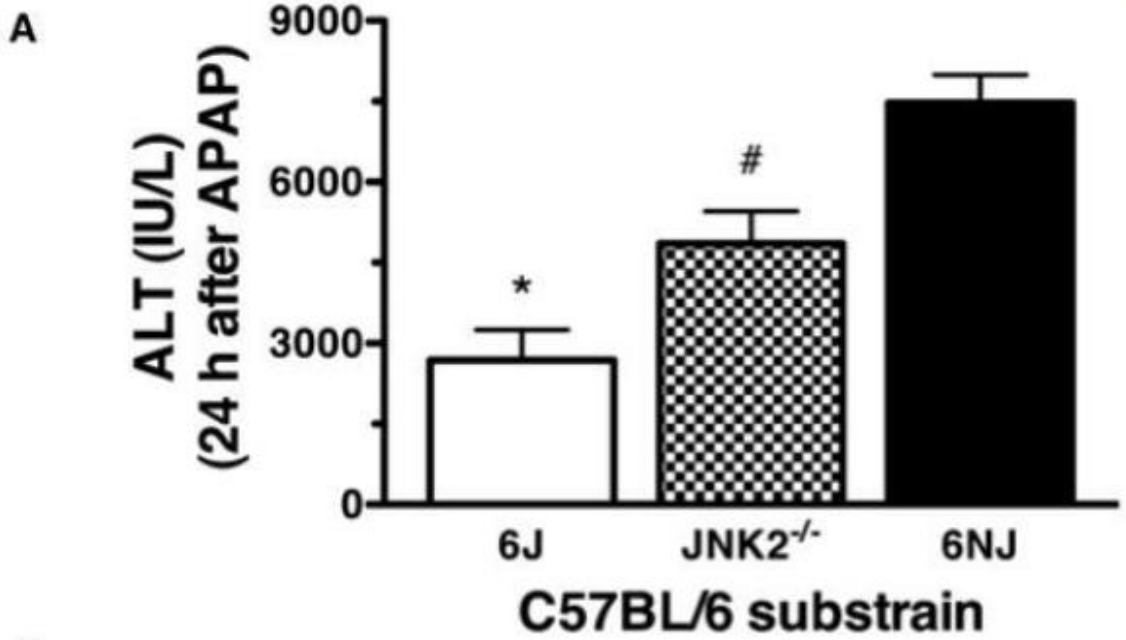
NSG variants

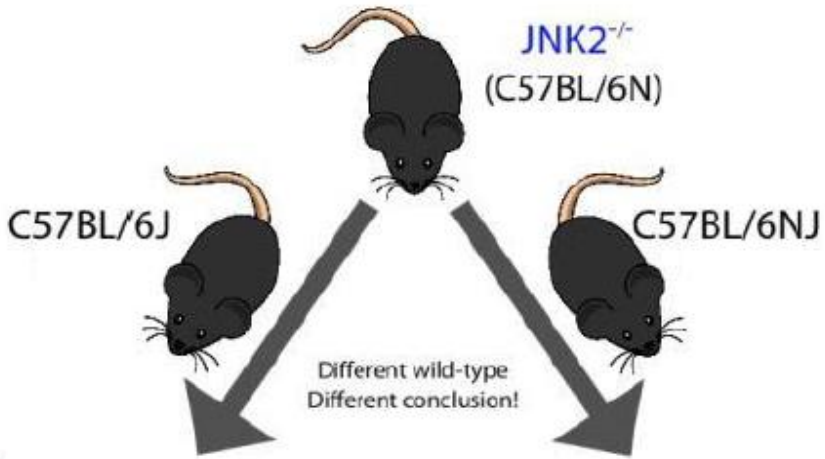
Name & Stock Number	NOD.*Cg-Prkdc*^{scid} *Il2rg*^{tm1Wjl}/SzJ (005557) **NSG**	NOD.*Cg-Prkdc*^{scid} *Il2rg*^{tm1Wjl} Tg(CMV-IL3,CSF2,KITLG)1Eav/MloySzJ (013062) **NSGS**	NOD.*Cg-Rag1*^{tm1Mom} *Il2rg*^{tm1Wjl}/SzJ (007799) **NRG**
Common name	NSG™, NOD *scid* gamma	NSGS, NOD *scid* gamma Il3- GM-SF (NSG-SGM3)	NRG, NOD *Rag* gamma
Mature B cells	Absent	Absent	Absent
Mature T cells	Absent	Absent	Absent
Dendritic cells	Defective	Defective	Defective
Macrophages	Defective	Defective	Defective
Natural killer cells	Absent	Absent	Absent
Complement	Absent	Absent	Absent
Leakiness	Very Low	Absent	Absent
Irradiation tolerance	Low	Low	High
Lymphoma incidence	Low	Low	Low

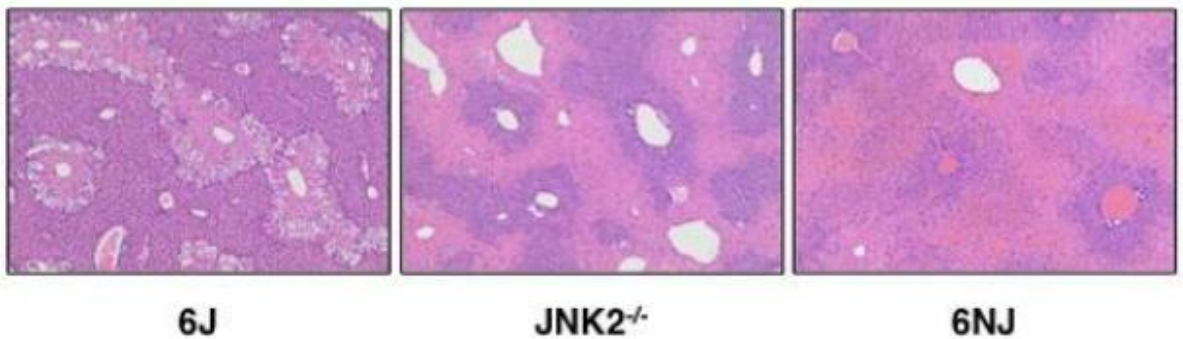
JAX lab

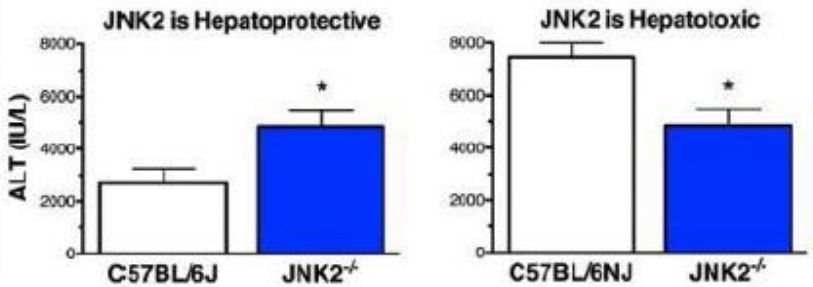
Selecting the Proper C57BL/6 Substrain Control

Effects of *Mapk9* (*Jnk2*) on acetaminophen-induced liver injury (AILI)









Bourdi M et al. 2011. *Chem Res Toxicol* 24: 794-6. PMID:21557537

Inbred and Hybrid

Inbred*

C57BL/6J = B6

129S1/SvImJ = 129S

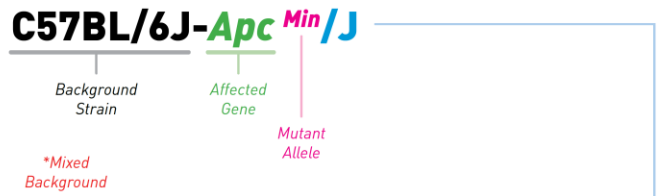
F1Hybrid*

B6129SF1/J

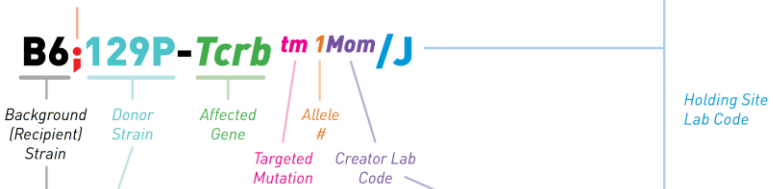
♀ ♂

*Standard strain abbreviations. Learn more at jax.org/nomenclature.

Spontaneous or Induced Mutation

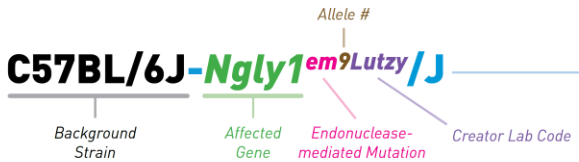


Knock-out, Knock-in or Floxed



Transgenic (Tg)

Endonuclease-mediated



*Mixed Strain Background = semicolon (;)
Backcrossed to recipient inbred strain < 5 generations

** Congenic or Incipient Congenic = period (.)
Backcrossed to recipient inbred strain ≥ 5 generations

1-800-422-6423
(US, Canada & Puerto Rico)
1-207-288-5845
(from any location)
jax.org/nomenclature

The Jackson Laboratory

LT0018 US 02 09/2018

Nomenclature links

Mouse Genome Informatics

http://www.informatics.jax.org/mgihome/nomen/index.shtml

JAX Lab

http://jax.org/nomenclature

使用動物數量

十、請以動物實驗應用3Rs之替代及減量原則，說明動物實驗試驗設計、實驗動物需求、動物種別及數量之必要性：

(五) 說明動物實驗試驗設計(動物分組方法、每組使用動物數量等)：

使用動物數量

- **Power analysis**
- **Pilot study results**
- **Publications**

Examples:

1. 需控制與實驗組。每組至少5隻老鼠符合基本生物統計標準，必要時，將實驗重複一次，增加實驗n值。

2. 需控制與實驗組。每組至少40隻老鼠符合基本生物統計標準，必要時，將實驗重複一次，增加實驗n值。

IACUC of Boston University: Sample size calculations

投予 & 採樣

十一、請以實驗動物應用3Rs之精緻化原則，說明實驗中所進行之動物實驗內容：

(一) 實驗物質之投予、採樣方法及其頻率：

採血規範

Table 3. Circulating blood volume in laboratory animals

Species	Blood volume (ml kg^{-1})	
	Recommended mean[a]	Range of means
Mouse	72	63–80
Rat	64	58–70
Rabbit	56	44–70
Dog (Beagle)	85	79–90
Macaque (Rhesus)	56	44–67
Macaque (Cynomolgus)	65	55–75
Marmoset	70	58–82
Minipig	65	61–68

[a]The recommended mean corresponds to the mid-point of the range of means.

Table 4. Limit volumes and recovery periods

Single sampling (e.g. toxicity study)		Multiple sampling (e.g. toxicokinetic study)	
% Circulatory blood volume removed	Approximate recovery period	% Circulatory blood volume removed in 24 h	Approximate recovery period
7.5%	1 week	7.5%	1 week
10%	2 weeks	10–15%	2 weeks
15%	4 weeks	20%	3 weeks

Side effects : Anemia, Altered physiologic parameters, Hypovolemic shock, Death.

Diehl et al., J Appl Toxicol., 2001

Guidelines for Blood Collection in Mice and Rats

Table 1: Approximate Blood Sample Volumes for a Range of Body Weights				
Body weight (g)	*CBV(ml)	1% CBV *every 24 hrs†*	7.5% CBV *every 7 days†*	10% CBV *every 2 - 4wks†*
20	1.10 - 1.40	11 - 14 µl	90 - 105 µl	110 - 140 µl
25	1.37 - 1.75	14 - 18 µl	102 - 131 µl	140 - 180 µl
30	1.65 - 2.10	17 - 21 µl	124 - 158 µl	170 - 210 µl
35	1.93 - 2.45	19 - 25 µl	145 - 184 µl	190 - 250 µl
40	2.20 - 2.80	22 - 28 µl	165 - 210 µl	220 - 280 µl
125	6.88 - 8.75	69 - 88 µl	516 - 656µl	690 - 880 µl
150	8.25 - 10.50	82 - 105 µl	619 - 788 µl	820 - 1000 µl
200	11.00 - 14.00	110 - 140 µl	825 – 1050 µl	1.1 - 1.4 ml
250	13.75 - 17.50	138 - 175 µl	1.0 – 1.3 ml	1.4 - 1.8 ml
300	16.50 - 21.00	165 - 210 µl	1.2 – 1.6 ml	1.7 - 2.1 ml
350	19.25 - 24.50	193 - 245 µl	1.4 – 1.8 ml	1.9 - 2.5 ml
*Circulating blood volume (1ml = 1000µl)		†Maximum sample volume for that sampling frequency		

Blood Collection

SPECIES	*Mean blood volume	AVERAGE WEIGHT	+MAXIMUM VOLUME IN MILLILITERS FOR SINGLE SAMPLING Based on Recovery Period		
			Weekly (7.5% of blood volume removed)	Every 14 days (10.0% of blood volume removed)	Every 30 days (15.0% of blood volume removed)
Mouse/Dormice (Based on mean blood volume)	72 ml/kg	20 g	0.10	0.15	0.20
		30 g	0.16	0.23	0.32
		40 g	0.20	0.30	0.45
Rat/Cotton Rat (Based on mean blood volume)	64 ml/kg	250 g	1.20	1.60	2.40
		500 g	2.40	3.20	4.80

Blood Collection Guidelines

Guidelines for Rodents

Table 1: Approximate Blood Sample Volumes for a Range of Body Weights

Body weight (g)	***CBV(ml)** **Circulating Blood Volume**	**1% CBV (ml)** every 24 hrs†	**7.5% CBV (ml)** every 7 days†	**10% CBV (ml)** every 2 wks†
20	1.10 – 1.40	.011 – .014	.082 – .105	.11 – .14
25	1.37 – 1.75	.014 – .018	.10 – .13	.14 – .18
30	1.65 – 2.10	.017 – .021	.12 – .16	.17 – .21
35	1.93 – 2.45	.019 – .025	.14 – .18	.19 – .25
40	2.20 – 2.80	.022 – .028	.16 – .21	.22 – .28
125	6.88 – 8.75	.069 – .088	.52 – .66	.69 – .88
150	8.25 – 10.50	.082 – .105	.62 – .79	.82 – 1.0
200	11.00 – 14.00	.11 – .14	.82 – 1.05	1.1 – 1.4
250	13.75 – 17.50	.14 – .18	1.0 – 1.3	1.4 – 1.8
300	16.50 – 21.00	.17 – .21	1.2 – 1.6	1.7 – 2.1
350	19.25 – 24.50	.19 – .25	1.4 – 1.8	1.9 – 2.5

IACUC, Boston Univ.

投予規範

Table 1. Administration volumes considered good practice (and possible maximal dose volumes)[a]

Species	Route and volumes (ml kg^{-1})					
	Oral	s.c.	i.p.	i.m.	i.v. (bolus)	i.v. (slow inj.)
Mouse	10 (50)	10 (40)	20 (80)	0.05[b] (0.1)[b]	5	(25)
Rat	10 (40)	5 (10)	10 (20)	0.1[b] (0.2)[b]	5	(20)
Rabbit	10 (15)	1 (2)	5 (20)	0.25 (0.5)	2	(10)
Dog	5 (15)	1 (2)	1 (20)	0.25 (0.5)	2.5	(5)
Macaque	5 (15)	2 (5)	[c] (10)	0.25 (0.5)	2	[c]
Marmoset	10 (15)	2 (5)	[c] (20)	0.25 (0.5)	2.5	(10)
Minipig	10 (15)	1 (2)	1 (20)	0.25 (0.5)	2.5	(5)

[a]For non-aqueous injectates, consideration must be given to time of absorption before re-dosing. No more than two intramuscular sites should be used per day. Subcutaneous sites should be limited to two or three sites per day. The subcutaneous site does *not* include Freund's adjuvant administration.
[b]Values in millilitres per site.
[c]Data not available.

Diehl et al., J Appl Toxicol., 2001

Dose Volumes in Laboratory Animals

Table 1: Maximum Volumes to be used for dosing of species by route of administration (All volumes are ml/kg unless otherwise noted)

Species	PO gavage/in tragastric	IV* (bolus)	IV** (ml/kg/h)	IP@	SC@	IM***@	IN*** IT***	ID***
Mouse	**10** (20)	**5** (10)	**1** (4)	**10** (20)	**5** (20)	**0.03** (0.05)$	**0.05**	**0.05** (0.1)
Rat	**10** (20)	**5** (10)	**1 or 2** (4)	**10** (20)	**5** (10)	**0.1** (0.2)$	**0.1**	**0.05** (0.1)
Hamster	**10** (20)	**5** (10)	--	**10** (20)	**1** (10)	**0.05** (0.1)$	**0.05**	**0.05** (0.1)
Guinea pig	**10** (20)	**1** (5)	--	**10** (20)	**5** (10)	**0.1** (0.2)	**0.1** (0.2)	**0.05** (0.1)
Rabbit	**2** (10)	**1** (5)	**1**	Not recommended	**2.5** (10)	**0.25** (0.5)	**0.2** (0.5)	**0.05** (0.1)

Ideal maximum dose volumes are bolded, absolute maximum dose volumes are (smaller in parenthesis)

*An IV bolus injection is typically dosed in less than 1 min.

**Continuous IV infusions are typically dosed over 3-10 minutes. Solution properties such as tonicity, pH, etc. must be taken into account when approaching the volume limits or determining the volume to be infused IV. The recommended working range for pH is 4.5-8.0. The order of degree of tolerance of pH for different dosing routes is oral>intravenous>intramuscular>subcutaneous>intraperitoneal. Animal health must also be taken into consideration, such as kidney function and cardiovascular function. These systems must be normal to handle increased fluid volume.

***The values listed in this column are the total volume in ml per site, total of 2 sites/day.

@When administering a solution IP, SC, or IM, the viscosity, concentration, tonicity and pH of the solution need to be taken into account.

$IM dosing in mice, rats, hamsters and voles is **NOT** recommended.

Drug and Chemical Administration

Table 1. Drug Administration Routes and Volumes (and maximal dose volumes) [D]

Species	PO* (ml/kg)	IV bolus* (ml/kg)	IV drip* (ml/kg/hr)	IP* (ml/kg)	SC* (ml/kg)	ID* (ml/inj)	IM* (ml/kg/site)	IN* (ml/inj)	Gavage (ml/kg)
Mouse	10	1-5	2-4	1-20	1-40	0.05-0.1	0.05[B] (0.1)[D]	0.03-0.05	1-20
Rat	10	1-5	2-4	1-10	1-10	0.05-0.1	0.1[B] (0.2)[D]	0.03-0.05	1-20
Guinea Pig	5	1-5	2-4	1-10	1-5	0.05-0.1	0.05[B] (0.1)[D]	0.03-0.05	1-20
Hamster	5	1-5	2-4	1-10	1-5	0.05-0.1	0.05[B] (0.1)[D]	0.03-0.05	1-20
Rabbit	10	1-5	2-4	1-5	1-5	0.05-0.1	0.05-0.1[C]	0.2-0.5	1-10
Dog	5	1-5	2-4	1	1-5	0.05-0.1	0.05-0.1[C]	0.2-0.5	1-20
Cat	5-20	1-5	2-4	1-10	1-5	0.05-0.1	0.05-0.1[C]	0.2-0.5	1-20
Swine	5-20	1-5	2-4	1-10	1-5	0.05-0.1	0.05-0.1[C]	0.2-0.5	1-20
Sheep/ Ruminant	5-20	1-5	2-4	1-10	1-5	0.05-0.1	0.05-0.1[C]	0.2-0.5	1-20

[A] *For non-aqueous injectates, consideration must be given to time of absorption before re-dosing. No more than two intramuscular sites should be used per day. Subcutaneous sites should be limited to two or three sites per day.*

[B] *Values in ml/site for adult animals and large muscle bodies. Adjust for size*

[C] *Values in ml/kg/site*

[D] *Maximum volume in ml/site in adult animal and large muscle bodies. Lower volume for smaller sizes*

IACUC, WSU

手術

I. 定義

II. Pre-Op., Op., Post-Op.

（三）本實驗中動物之使用涉及(請勾選，未涉項目請於後說明中填「無」)

☐ 鎮靜，其方法、劑量、投藥方法為：

☐ 無外科手術之麻醉，其方法、劑量、投藥方法與麻醉的照護為：

☐ 有外科手術之麻醉，其方法、劑量、投藥方法與手術後的照護：

定義

1. 存活 vs 非存活

2. 重要(Major) vs 非重要(Minor) 存活手術

Major – To penetrate and expose a body cavity, to produce substantial impairment of physical or physiologic functions, or to involve extensive tissue dissection or transection.
(**3Days**)

Minor - Does not expose a body cavity and causes little or no physical impairment.

3. 單次 vs 多次

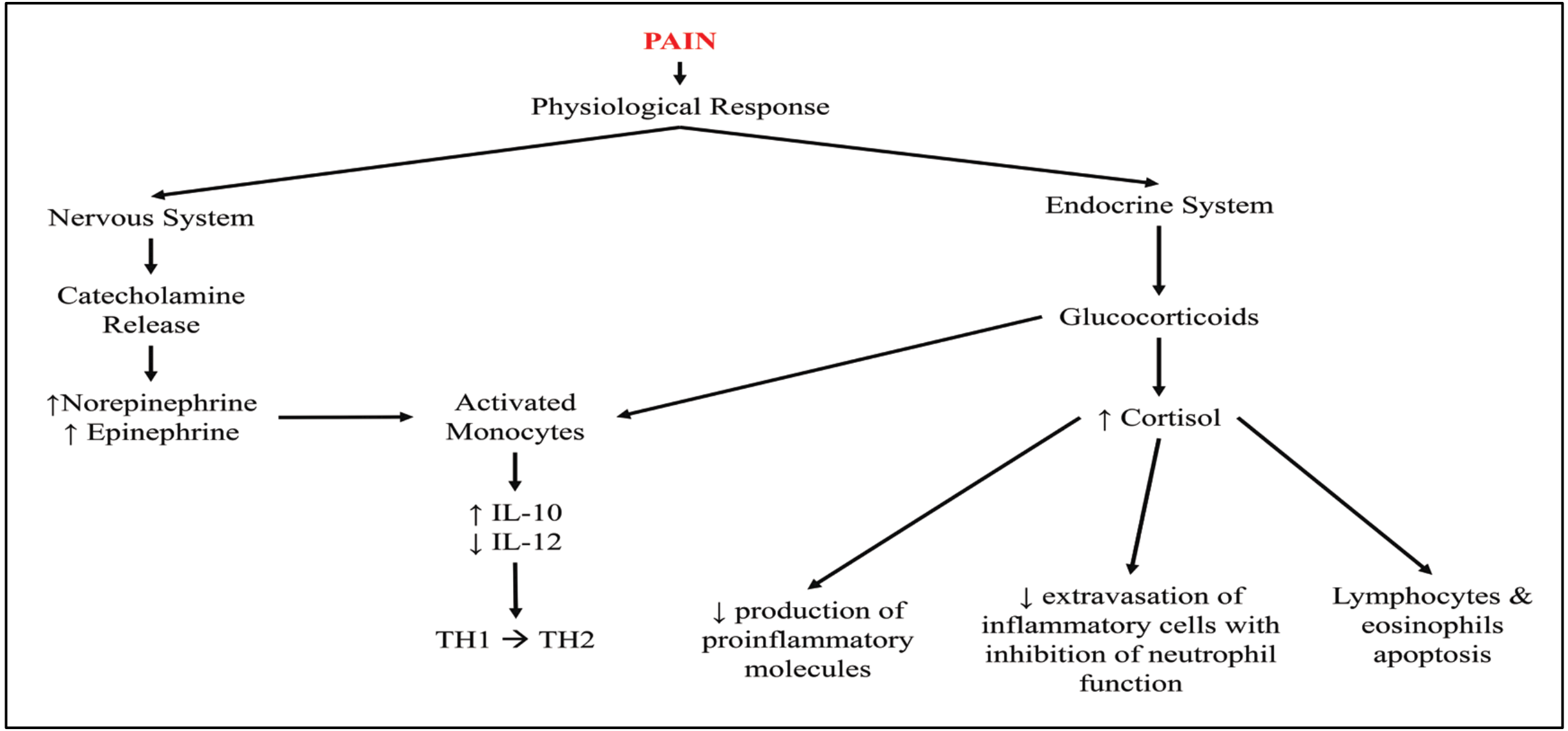
Single – Single surgical procedure is conducted in one animal.

Multiple - Multiple surgical procedures on a single animal should be evaluated to determine their impact on the animal's wellbeing (IACUC).

麻醉 ≠ 止痛

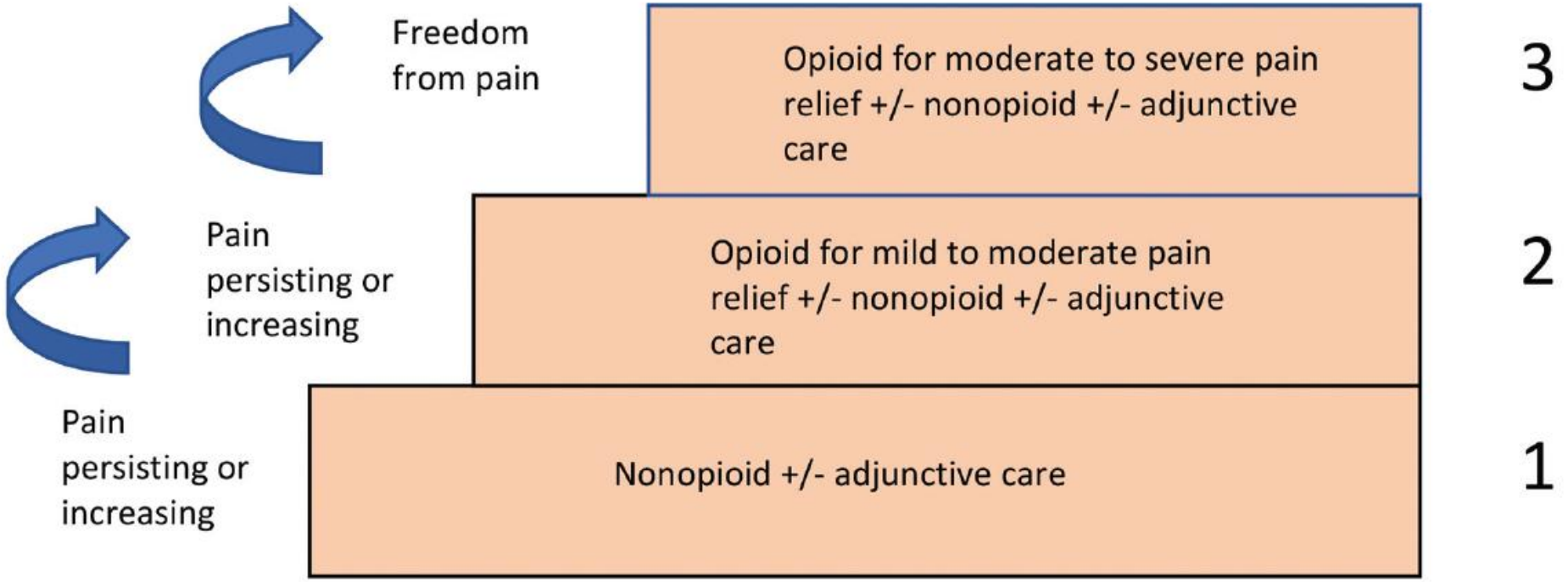

Isoflurane, USP
250 mL
A nonflammable, nonexplosive inhalation anesthetic.
Piramal | Healthcare
WARNING: Not for use in humans


Use Only Isoflurane

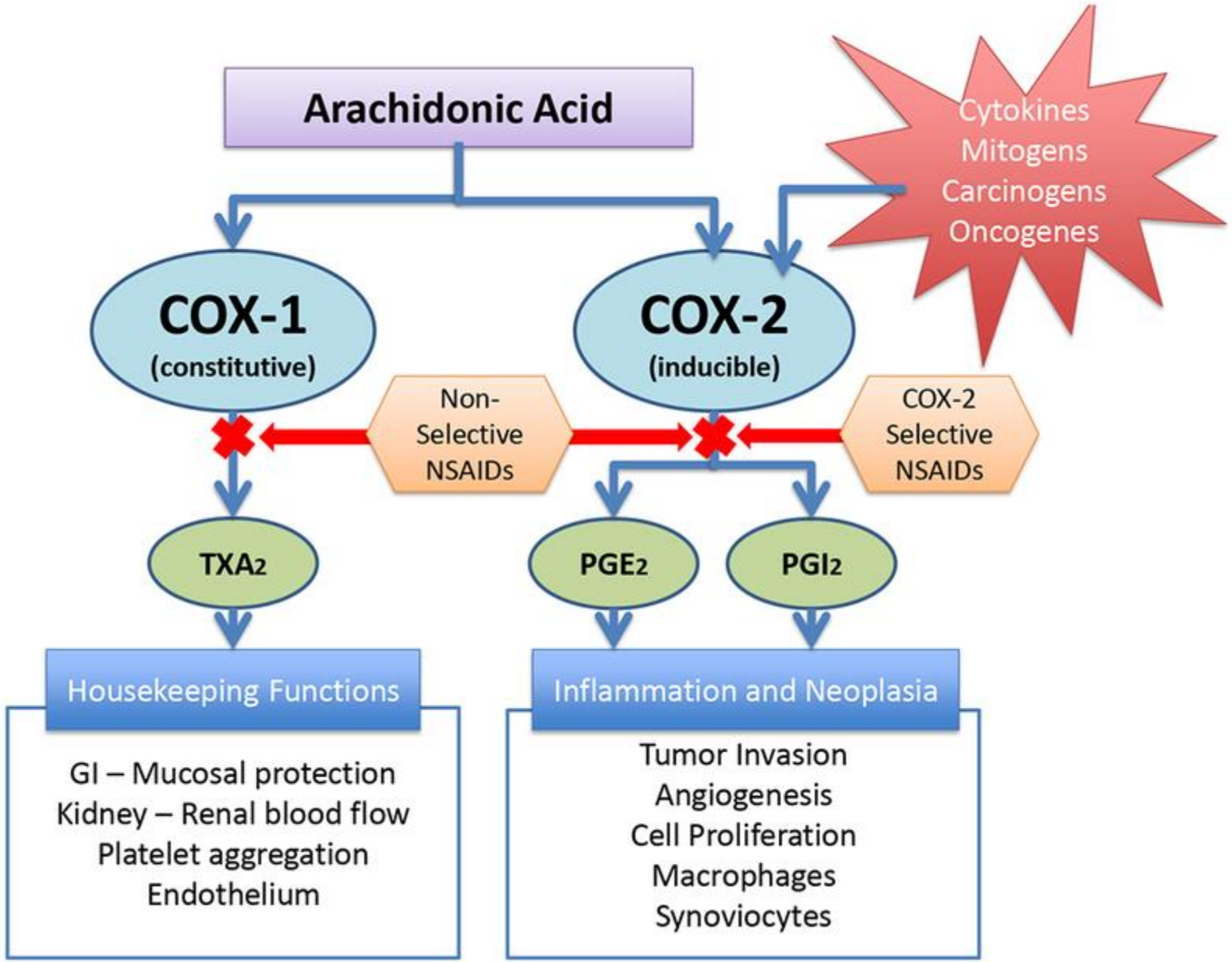


Norman, Comp Med., 2017

Pain Relief Ladder for Patient Management (WHO)

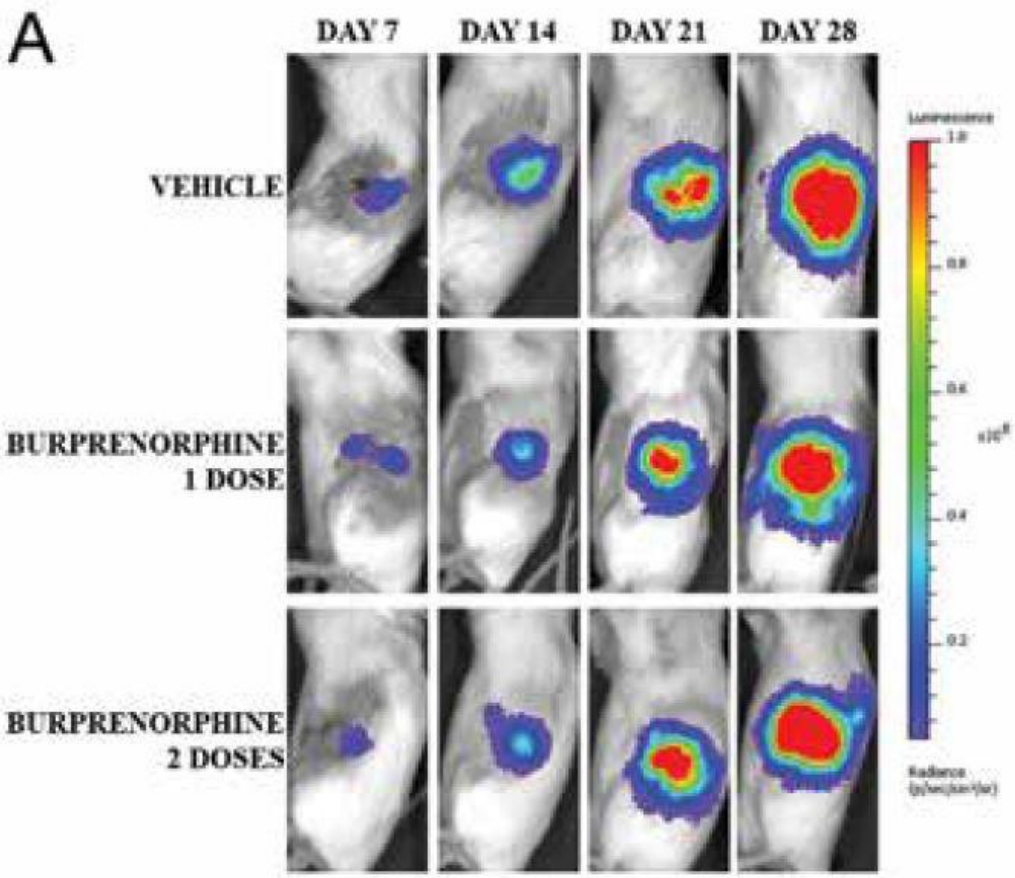


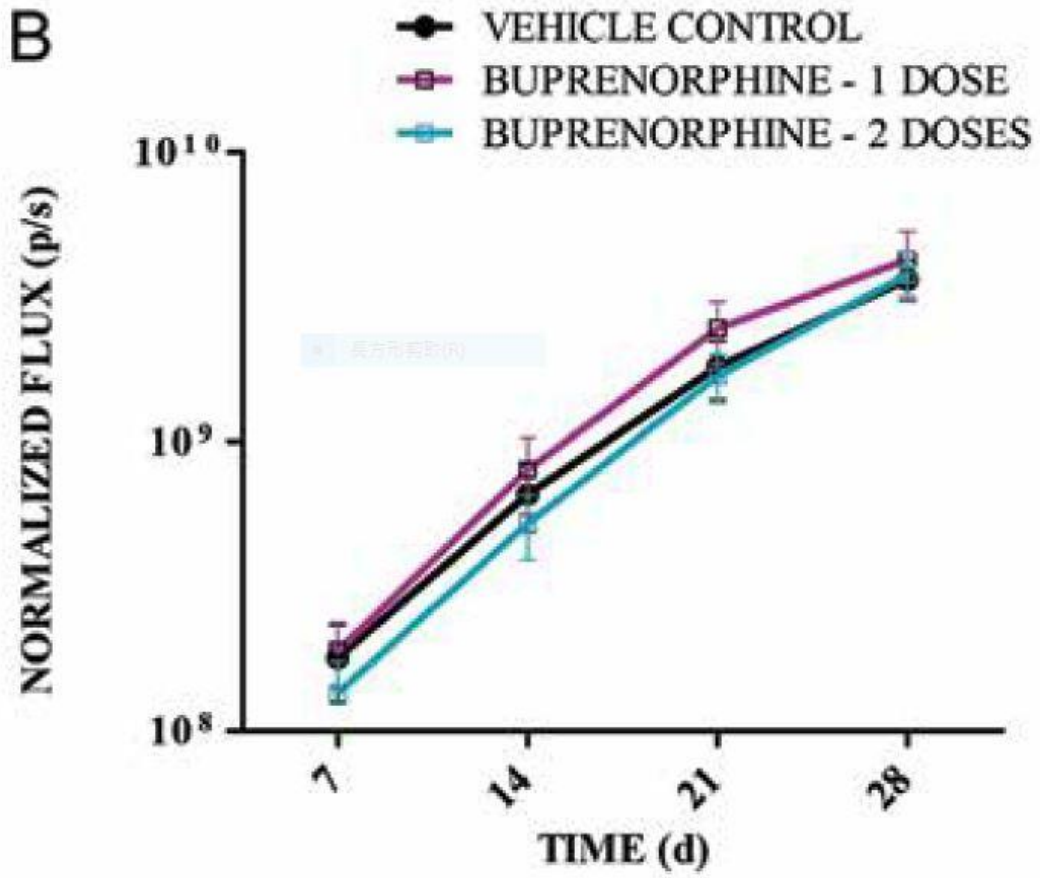
Foley *et al.*, Comp Med., 2019



Desai et al., Nutr Cancer, 2018

Effects of Buprenorphine in a Preclinical Orthotopic Tumor Model of Ovarian Carcinoma in Female CB17 SCID Mice





Bratcher er al., JAALAS, 2019

Selection of Analgesics

Procedures	Pain category	Analgesia selections
• Skin incision only • Subcutaneous implant • Catheter placement	**Mild** (analgesia to last at least 8hr)	Option 1: Local analgesia Option 2: 1 dose of buprenorphine Option 3: NASID
• Castration • Thyroidectomy	**Moderate** (analgesia to last at last 12-24hr) **Multimodal analgesia**	Option 1: Local analgesia + NASID Option 2: Buprenorphine + NASID Option 3: Buprenorphine-SR (lasts 72hr)
• Laparotomy (e.g. C-section) • Craniotomy • Nerve surgery	**Severe** (analgesia to last 24-48hr) **Multimodal analgesia**	Option 1: Local analgesia + Buprenorphine +/- NADID Option 2: Buprenorphine + NASID Option 3: Buprenorphine-SR +/- NASID

Commonly Used Systemic Analgesics in Mice and Rats

	Mouse	Rat	Class
Buprenorphine*	0.05-0.1 mg/kg SC q8-12h	0.01-0.05 mg/kg SC q8-12h	Opioid
Oxymorphone	0.2-0.5 mg/kg SC q4h	0.2-0.5 mg/kg SC q4h	Opioid
Morphine	1-2.5 mg/kg SC q2-6h	1-2.5 mg/kg SC q2-6h	Opioid
Ketoprofen	2-5 mg/kg SC q24h	2-5 mg/kg SC q24h	NSAID
Carprofen	2-5 mg/kg SC q12-24h	2-5 mg/kg SC q12-24h	NSAID
Meloxicam	1-2 mg/kg SC, PO q12h	1-2 mg/kg SC, PO q12h	NSAID
Flunixin	2.5 mg/kg SC q12h	1.1-2.5 mg/kg SC q12h	

*Buprenorphine is the only opioid with long duration effect in rodents.

NSAID: Non-steroidal anti-inflammatory drug.

Table 13. Updated analgesic dosing recommendations

Species	Agent	Dose (mg/kg)	Route	Frequency
Mouse	Buprenorphine	0.1-0.5	SC	4-6 h
	Buprenorphine SR[zp]	0.6	SC	48 h
	Tramadol	80	SC	24 h
	Carprofen	5	SC	12 h
		20	SC	24 h
	Meloxicam	5-10	SC	8-12 h
	Ketoprofen	20	SC	24 h
Rats	Buprenorphine	0.05-0.0.1	SC	6-8 h
		0.5-0.6	PO	24 h
	Buprenorphine SR[zp]	1.2	SC	48 h
	Tramadol	20-40	PO	24 h
		5	SC	24 h
	Carprofen	5	SC	24 h
	Meloxicam	1	SC	12-24 h
	Ketoprofen	5	SC	24 h
Guinea pig	Buprenorphine	0.05	SC	6 h
	Buprenorphine SR[zp,ag]	0.3-0.48	SC	48 h
	Carprofen	4	SC	12-24 h
	Meloxicam	0.2	SC	12-24 h

Modified from Flecknell 2018.[61]
SR = sustained release; # - provided in food treat, should be observed ingesting
z*P* = manufactured by Zoopharm, Windsor, CO; ag= manufactured by Animalgesics Laboratories, Millersville, MD.
Note: caution should be taken with higher doses of NSAIDs. Multimodal analgesia recommended to allow effective use of lower doses.

Foley *et al*., Comp Med., 2019

Table 14. Published multimodal analgesic efficacy studies

Species	Multimodal analgesics	Dose (mg/kg)	Route	Model	Comments	Reference
Mouse	Buprenorphine Carprofen	0.1 30	SC PO-W	Lap	Buprenorphine dosed q12h, carprofen medicated water provided for 72 h. Improved analgesia for 2-8 h postoperative	164
	Gabapentin	3-100	IP	TF, HP, FT	Reduced ED50 for each analgesic	153
	Tramadol	3-100	IP			
	Tramadol	10-100	IP	TF, HP, FT	ED50 reduced with Keto	152
	Ketoprofen	30-250	IP			
	Buprenorphine	0.05	SC	Lap	Buprenorphine dosed once preoperative. Melox was given 24 h postoperative	148
	Meloxicam	5	SC			
	Meloxicam	5	SC	Lap	No effect	149
	Acetaminophen	50	IP			
	Ibuprofen	200	IP	TF	Opioids enhanced latency	217
	Tramadol		SC	WT, HP	Opioids reduced ED50	59,175
Rat	Buprenorphine	0.03	SC	PW	Similar effect to buprenorphine alone	133
	Meloxicam	2	SC			
	Buprenorphine	0.05	SC	SX	Buprenorphine dosed q8-12h, meloxicam daily. No effect; 8 h dosing resulted in pica	183
	Meloxicam	2	SC			
	Acetaminophen	20-1000	PO	HP, VF	ED50 reduced of each	188
	Tramadol	3-30	PO			
	Carprofen	5	SC	Lap	Dosed preoperative and 4 and 24 h postoperative. Increased activity with tramadol	23
	Tramadol	12.5	IP			
	Gabapentin	5-20	IP	HP, VF	Potentiates opioids	81,146,162,167
	Tramadol	10	SC	HP	Tramadol dosed q12h for 60 h, gabapentin dosed daily. Minimal effect	141
	Gabapentin	80	SC			
	Tramadol	10	SC	SX	Tramadol dosed q8-12h and gabapentin dosed daily for 120 h, No effect	27
	Gabapentin	80	SC			
	Levobupivacaine	0.3% 50 µL	SC	SX	Enhanced with ibuprofen and epinephrine	122
	Ibuprofen	2 mg/mL 50 µL	SC			
	Lidocaine	22.6 mmol/kg	SC	VF	Increased threshold	31
	Naloxone	43.2 mmol/kg				
Guinea pig	Meloxicam	0.2	SC	Lap	No effect	52
	Bupivacaine	1	SC			
	Lidocaine	1	SC			
	Buprenorphine SR[ag]	0.48	SC	Lap	Improved analgesia compared with carprofen alone	163
	Carprofen	4	SC			

PO-W = Oral by water
ag= manufactured by Animalgesics Laboratories, Millersville, MD

Multimodal Analgesics

Foley *et al*., Comp Med., 2019

預先給予止痛劑是管理術
後疼痛最好的辦法!!!

麻醉

VS

Inhalation Anesthetics

Injectable Anesthetics

Injectable Anesthetic Agents

COMMONLY USED INJECTABLE ANESTHETIC AGENTS

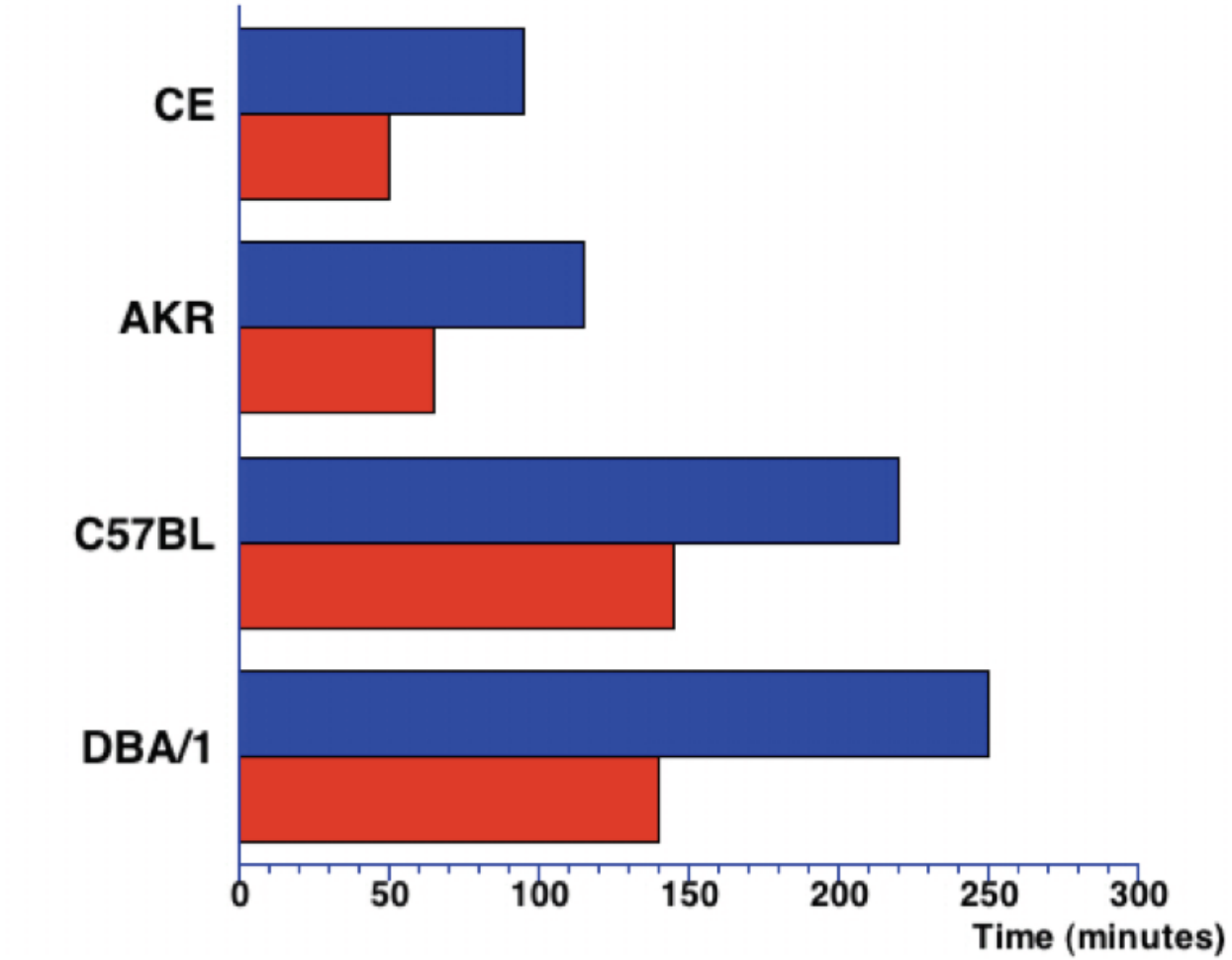
MOUSE

Agent	Dosage	Duration of anesthesia
Ketamine/xylazine*	ketamine 80-100 mg/kg IP xylazine 10-12.5 mg/kg IP	20-30 minutes
Ketamine/xylazine cocktail*	KX mouse cocktail 0.1mL/20g mouse wt. IP Contains: 87.5 mg/kg Ketamine 12.5 mg/kg Xylazine	20-30 minutes
Ketamine/xylazine/acepromazine	ketamine 60-100 mg/kg IP xylazine 10-15 mg/kg IP acepromazine 2-5 mg/kg IP	60-90 minutes
Pentobarbital	50 mg/kg IP	20-40 minutes
Avertin‡ See warning below	240 mg/kg IP	30 minutes

RAT

Agent	Dosage	Duration of anesthesia
Ketamine/xylazine	ketamine 40-100 mg/kg IP xylazine 5-13 mg/kg IP	60-80 minutes
Ketamine/xylazine cocktail*	KX rat cocktail 0.1 mL/100g rat wt. IP Contains: 91 mg/kg Ketamine 9.1 mg/kg Xylazine	60-80 minutes
Ketamine/xylazine/acepromazine	ketamine 20-50 mg/kg IP xylazine 2-10 mg/kg IP acepromazine 0.5-1.5 mg/kg IP	60-120 minutes
Pentobarbital	30-50 mg/kg IP	90-120 minutes

Mice Strain & Male/Female Sleep Times



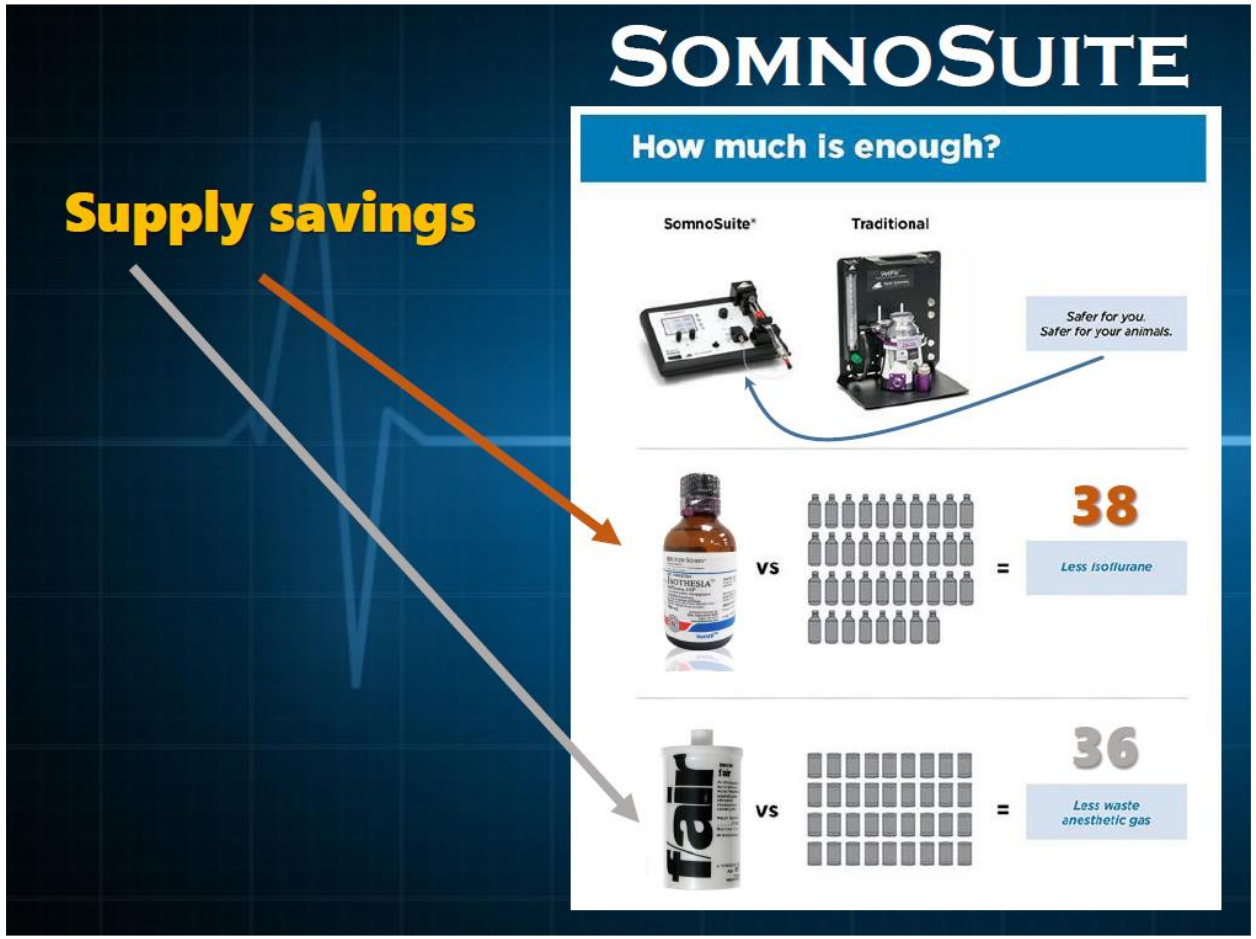
Pentobarbital

Isoflurane

- Scavenging system (Abortion & Hepatoxicity)
- Induction: 4-5%, gradually increased
- Maintenance: 2-4%

SomnoSuite®

Low Flow Anesthesia System
for Mice and Rats



Pre – OP.

Disinfection vs Sterilization

Disinfection

The chemical or physical process that involves the destruction of pathogenic organisms. All disinfectants are effective against vegetative forms of organisms, but **not necessarily spores**.

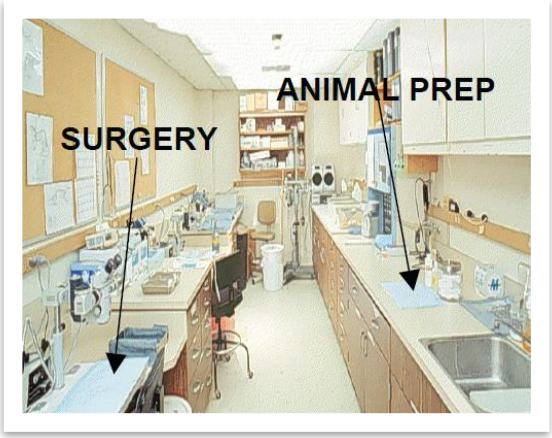
Sterilization

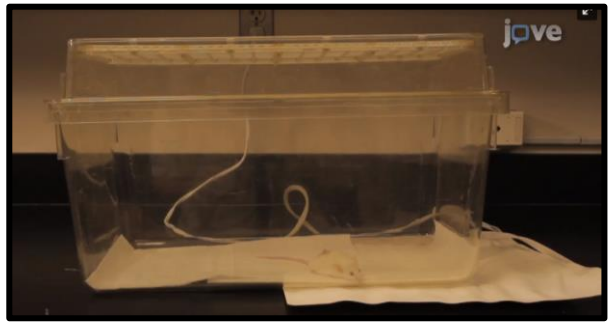
The process whereby **all** viable microorganisms are **eliminated or destroyed**. The criterion for adequate sterilization is the failure of organisms to grow if a growth-supporting medium is supplied.

Sterilization

- **Steam sterilization is the best!!!!**
  - ☐Autoclave
- **Glass bead sterilizer** (≧250°F，10-15 sec)
- **Hydrogen peroxide vapor** (Gas)
- **Chemical reagents–not recommend**
  （70~75% Ethanol）

Prep, Surgical & Recover Areas





1. Cleanliness
2. Working flow
3. Pos. pressure or Lamina flow/BSC
4. Recover area – quite、dark、comfy


This is where you clip hair, away from surgery table to avoid debris from falling in the surgical wound
Prep area
Surgery table

Avoid Eye Drying Out

Comparative Medicine
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by the American Association for Laboratory Animal Science

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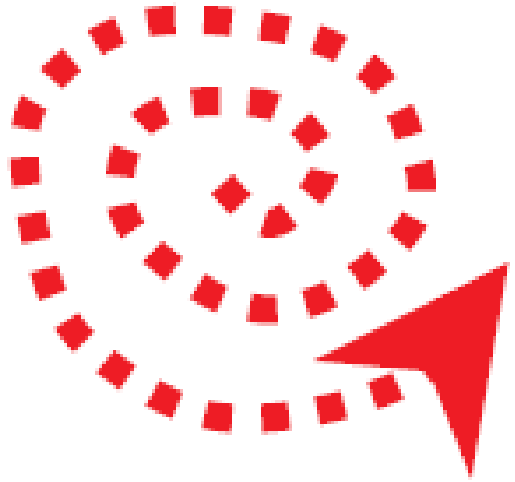
Pages 175-182

Susceptibility of Rats to Corneal Lesions After Injectable Anesthesia

Patricia V. Turner, DVM, DVSc[1,*] and Mudher A. Albassam, DVM, PhD[2]

Eye ointment

Skin Disinfection



- 由手術部位中心向外為擴展，消毒**至少3次**，最後讓**消毒劑*自然乾燥***。
- 消毒區域須涵蓋整個切口和鄰近部位，並須足夠大到供手術者工作，以避免污染。(Ex. **0.5cm** in mice)

常用消毒劑組合:

- 優碘+70%酒精
- Chlorhexidine gluconate+70%酒精

Contents lists available at ScienceDirect

International Journal of Surgery Open

journal homepage: www.elsevier.com/locate/ijso

Review Article

Chlorhexidine-alcohol versus povidone-iodine for pre-operative skin preparation: A systematic review and meta-analysis

Firas Ayoub [a,*], Michael Quirke [a], Ronan Conroy [b], Arnold Hill [a]

[a] Department of Surgery, Beaumont Hospital, Dublin, Ireland
[b] Department of Epidemiology & Public Health Medicine, Royal College of Surgeons, Dublin, Ireland

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ABSTRACT

Background: Surgical site infection (SSI) is a dreaded postoperative complication. Although preoperative skin cleansing in order to prevent surgical site infection (SSI) is standard surgical practice, there is clinical equipoise concerning whether povidone iodine (PI) or chlorhexidine alcohol (CHA) is the antiseptic agent of choice.

Objectives: To determine whether CHA or PI is the preferred preoperative skin preparation for reducing SSI in clean, clean-contaminated and contaminated surgery.

Search methods: PubMed, Embase, and gray literature sources were searched for randomized controlled trials (RCTs) comparing both CHA and PI between 1980 and 2014. Comparative RCTs of preoperative CHA versus PI studying SSI in clean, clean-contaminated and contaminated surgery were included. Risk of bias was assessed using Cochrane risk of bias.

Main result: We identified six eligible studies with an overall 2484 participants. The overall rate of SSI was 6.8% in the CHA group versus 11.0% in the PI group ($P < 0.0002$). CHA was superior to PI in the prevention of SSI with a pooled RR of 0.62 (95% CI, 0.48–0.81).

Conclusions: Preoperative surgical skin preparation with CHA is more effective than PI in preventing SSI across clean and clean-contaminated surgery. Further studies should evaluate the effectiveness of CHA versus PI in contaminated surgery.

Avoid Hypothermia

- Heating lamp – proper distance/ avoid overheating
- Heating blanket – avoid overheating/ don’t contact animals directly
- Isothermal pads
- Circulatory water blanket
- Far Infrared Homeothermic Pad

Supplementary Therapy

- ≈37°C Saline or Ringer’s solution, 3-5%of body weight, SC (0.3-0.5ml/10g.BW).

Hydration + **Normothermia**

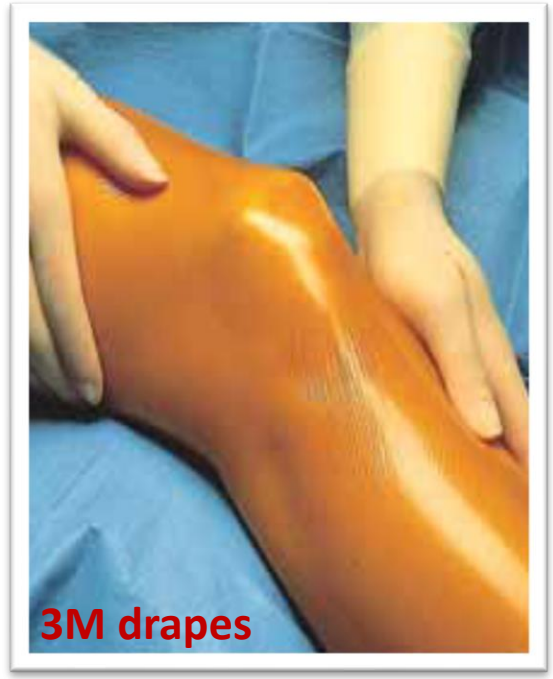
=

Morbidity & Mortality ⬇

Draping

- 利用無菌材料附蓋動物的軀體，避免無菌器材或手術人員的雙手碰到動物的體毛而污染

- 全身覆蓋vs露出口鼻

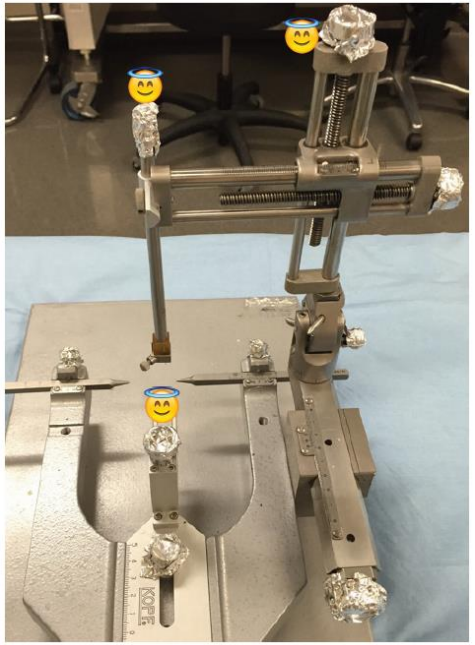


Draping

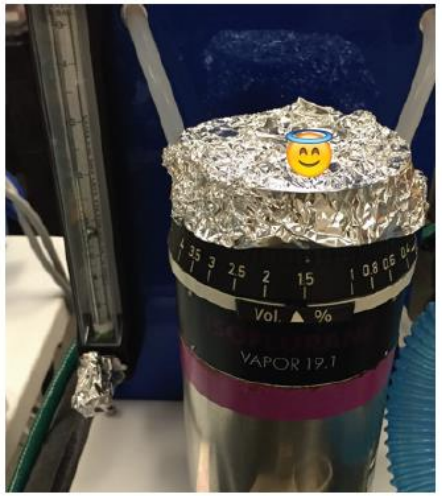

Draping with cotton material leads to wicking of fluids and bacteria into the surgical wound

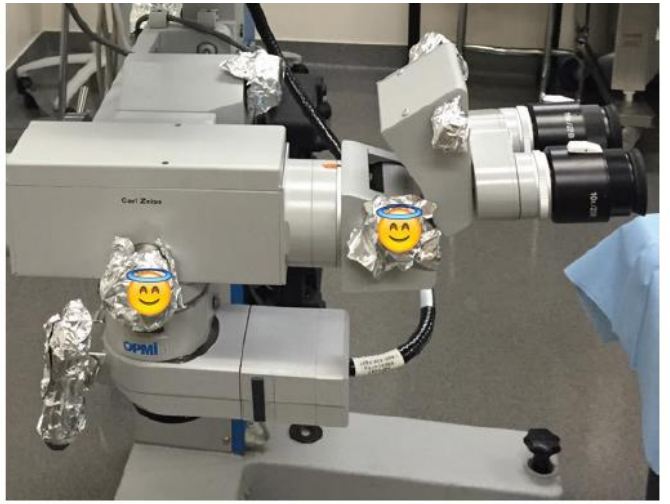

Impermeable material such as Press'n Seal as seen on this rat makes for an ideal drape
GLAD
Press'n Seal

What Can Draping & Foil Do ?

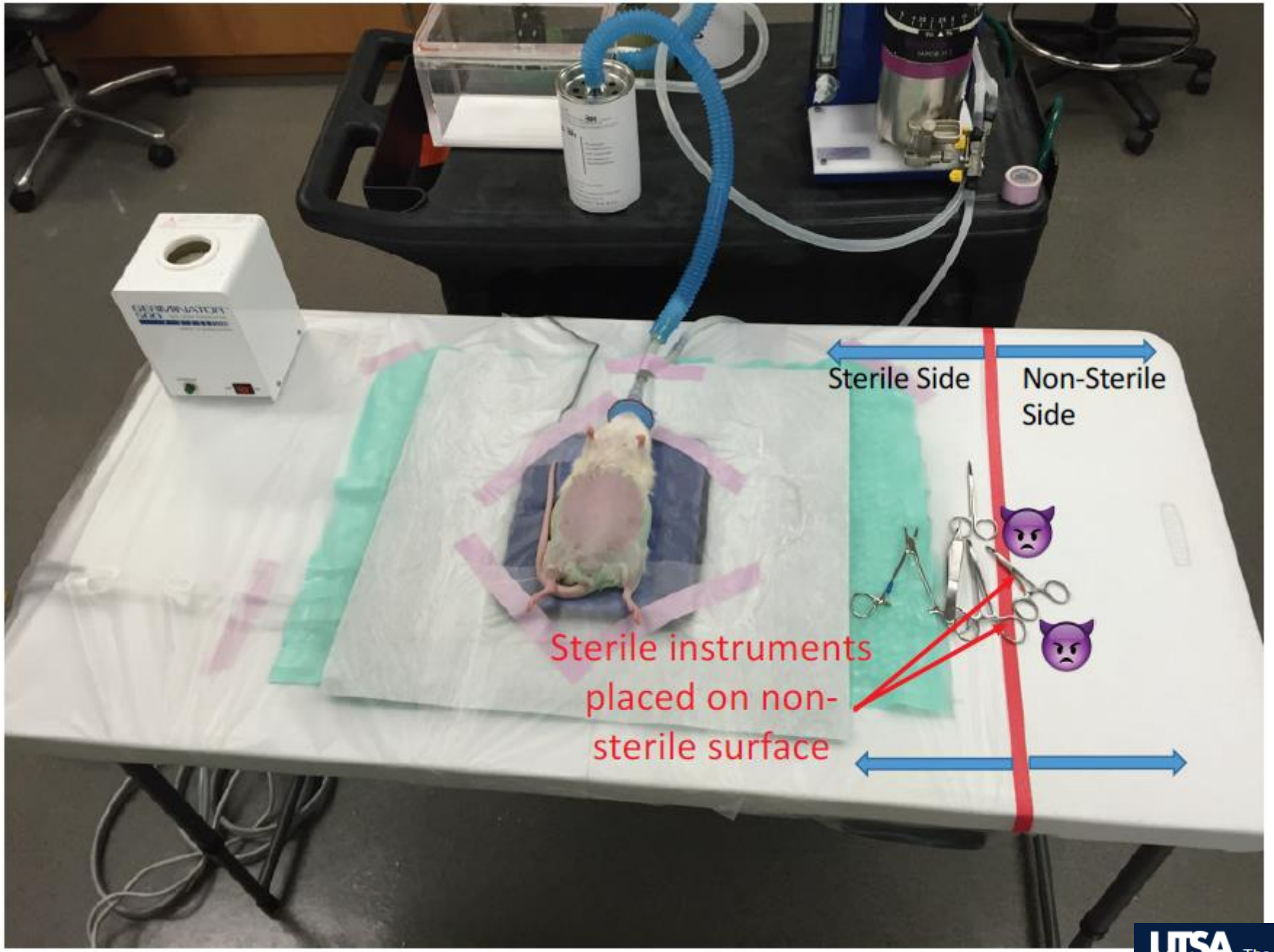


Covering the anesthesia machine dials with Press'n Seal or autoclaved aluminum foil prevents cross-contamination of the surgical wound





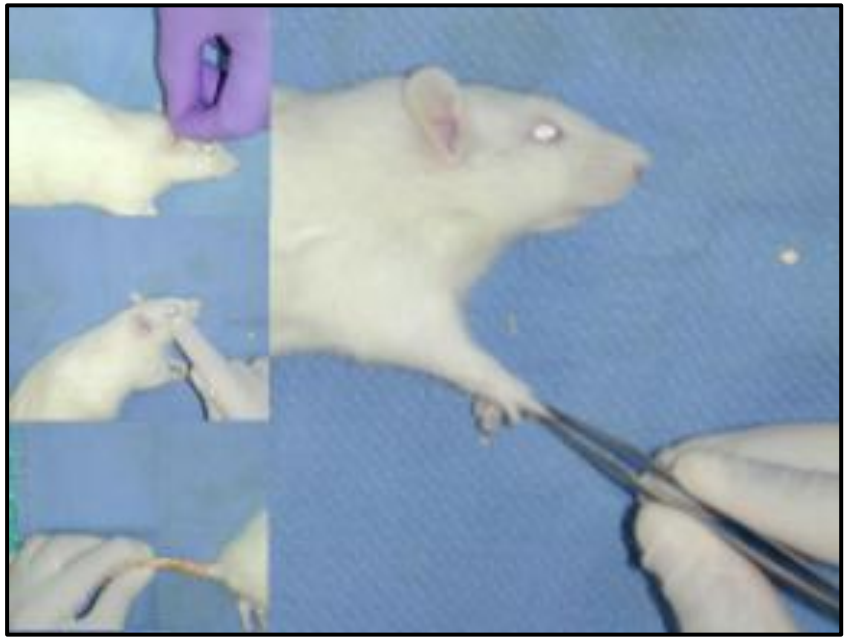
UTSA The University of Texas at San Antonio


Sterile Side
Non-Sterile Side
Sterile instruments placed on non-sterile surface

OP.

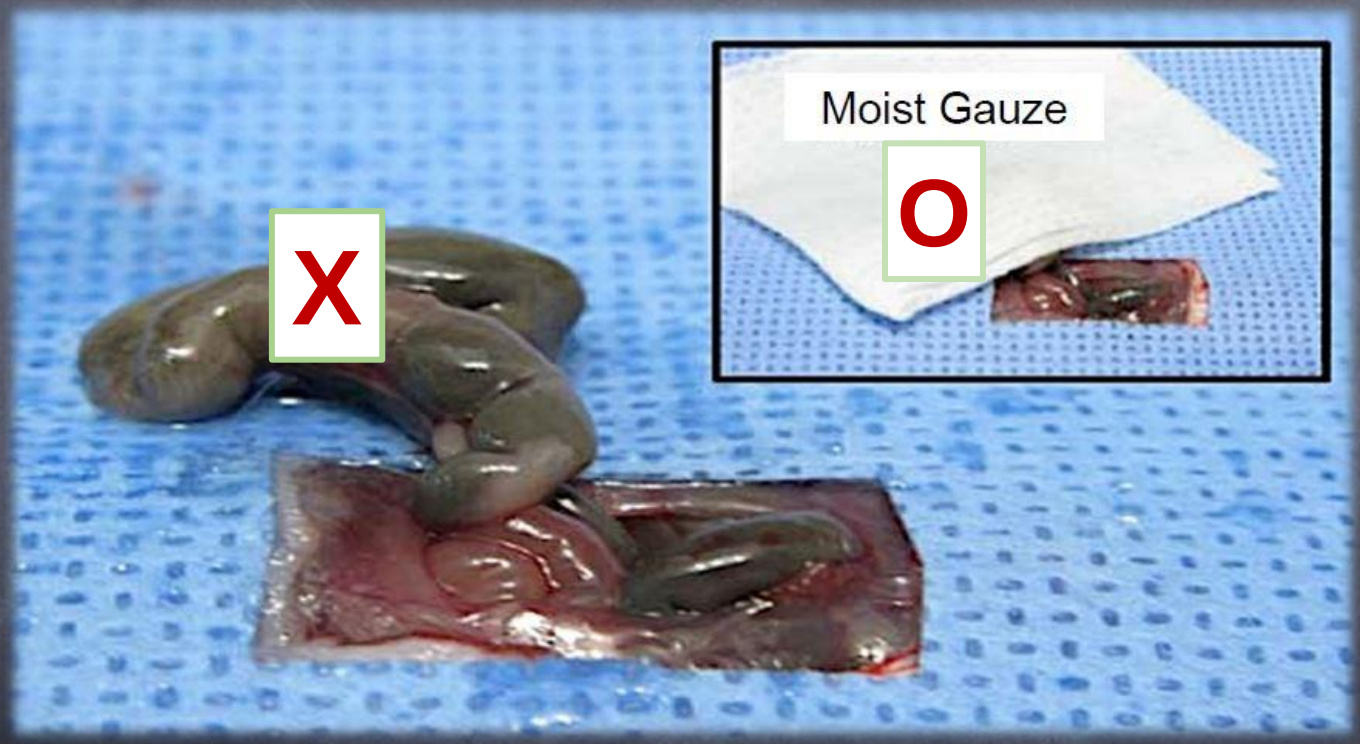
麻醉深度監控

- ✓評估刺激接受與反射（角膜、腳趾、尾巴、耳朵）
- ✓觀察胸腔起伏
- ✓鼻吻部、耳朵、舌頭、腳趾或黏膜面顏色
- ✓體溫
- ✓脈搏、心跳或直接/間接血壓(Cuff or Doppler)
- ✓輔助儀器如:血氧飽和儀、二氧化碳儀



Make sure it's Surgical stage before cutting !!!

Tissue protection



"Wet tissues = Happy tissues"

Aseptic technique (無菌操作)

- **目的**: 減少手術過程中**汙染**的機率(如果沒有感染，**無須給予抗生素**)。

- 所有實驗動物的存活手術應以無菌操作方式進行。
  (嚙齒動物手術替代方式：**尖端無菌/Sterile tip**)

- Tips:
  1. 所有手術器械應滅菌
  2. PPE
  3. 區分手術區域
  4. 良好操作技巧

Aseptic technique increases success of ovarian transplants in mice & speeds return to post-op normal (Cunliffe-Beamer 1972-73; Cunliffe-Beamer 1990)

Contamination activates macrophages (Bancroft, Schreiber et al. 1989), and leads to changes in cytokines & B cells levels (Abbas, Lichtman et al. 1991)

SUBCLINICAL infections induce physiological changes (Committee on Infectious Diseases of Laboratory Rats and Mice 1992)

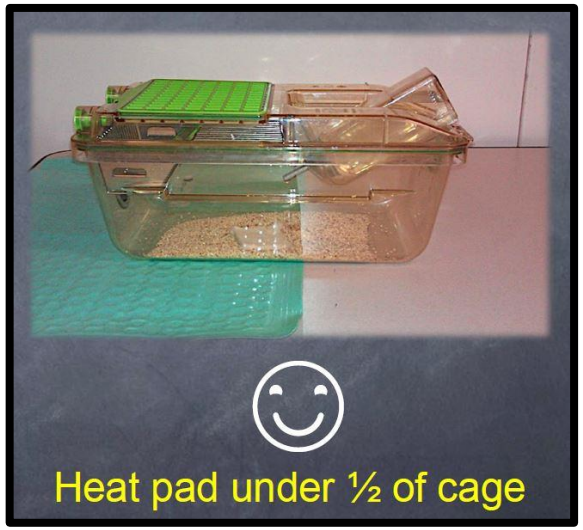
Although NO CLINICAL SIGNS observed, experimentally inoculated rats (10^8 *S. aureus* or *P. aeruginosa*) had significant alterations in plasma fibrinogen, serum glucose, total white blood cell counts, and wound histology scores (Bradfield, John et al. 1992)

Poor Aseptic Rodent Surgery leads to :

- Poor experimental results
- Delay to post-op normality
- Physiological changes
- Alterations in fibrinogen, glucose, leukocytes, histology, cytokines, B cells, etc.
- Questionable data

Post-OP. Care

- 恢復區: 乾淨/安靜/舒適/溫暖



- 時時注意生命表徵
- Hydration (≈37°C Ringer's solution or saline, 3-5% BW, SC or IP)。
- 注射型麻醉劑: 給予拮抗劑 加速恢復

Yohimbine – Xylazine
Atipamezol – Dexmedetomidine, Medetomidine

Tips

- 讓動物快點醒來可以**增加存活率。**
- **正位反射**或**具備活動能力**是知覺恢復的象徵。

Leaving when animals are fully conscious!!!

手術紀錄

- 生理數值
- 藥物紀錄
- 器械/儀器紀錄
- 術後疼痛反應或是體重變化
- 副作用、個體差異

Mouse Anesthesia/Surgery/Post-Operative Record - Example

PI (PI name)		**Protocol #** AN123456		
Personnel (Surgeon's name)		**Surgery Type** Survival		
Date 1/1/2018		**Species** Mouse		
Procedure name Osmotic pump implantation		**Experimental agents administered** Saline		
Anesthetics used	(1) Isoflurane	**Dose + Route (mg/kg)**	(1) 1%-5% to effect	(1) Inhalation
Analgesics used (All must be used)	(1) Lidocaine -OR- Bupivacaine (2) Buprenorphine (3) Meloxicam -OR- Carprofen	**Dose + Route (mg/kg)**	(1) < 7-8 (2) 0.05-0.1 (3) 5-10 (Mel) -OR- ~5 (Car)	(1) Local (2) SC/IP (3) SC/IP/PO

- ☑ Ophthalmic ointment administered in eyes of all anesthetized animals
- ☑ Heat provided during surgery and recovery
- ☑ Pedal reflex (pinch at both foot pads) checked periodically during surgery to evaluate depth of anesthesia
- ☑ Green surgery tag affixed to cage card or onto cage

Circle Animal ID Cage ID	1	2	3		
Body weight (grams)	25g	23g	27g		
Anesthesia start time	10:00am	10:30am	11:00am		
Analgesics Name	**Time and volume administered**				
Lidocaine -OR- Bupivacaine	10:05am/ 1 drop	10:35am/ 1 drop	11:05am/ 1 drop		
Buprenorphine	10:05am/ 0.1ml SC	10:35am/ 0.1ml SC	11:05am/ 0.1ml SC		
Meloxicam -OR- Carprofen	10:05am/ 0.1ml SC	10:35am/ 0.1ml SC	11:05am/ 0.1ml SC		
Other Agents Name	**Time and dosage/volume administered**				
LRS (Lactated Ringers Solution)	1ml IP	1ml IP	1ml IP		
Anesthesia end time	10:20am	10:50am	11:20am		
Complications? Y/N	N	N	Y		

Note complications here and on back of page if needed:
#3 had slow recovery from anesthesia. Kept animal in recovery cage on heat until ambulatory. Eating/active at 12:15pm.

POST-OPERATIVE

- Record dates and times of post-op observations or analgesic administrations.
- Checkboxes below indicate required doses of analgesia. If an NSOP checkbox is also included in the same details section, observe animals for signs of pain. If pain is observed, administer analgesic(s) and re-evaluate at time point when next analgesic administration is due. If no pain is observed, mark the NSOP checkbox.
- Once post-surgical pain assessment has concluded, all other monitoring will continue as approved per protocol.

Animal or Cage ID #	1	2	3		
Date/Time	1/1/18 5:00pm	1/1/18 5:00pm	1/1/18 5:00pm		
NSOP or Analgesic details	NSOP	NSOP	NSOP		
Date/Time	1/2/18 9:00am	1/2/18 9:00am	1/2/18 9:00am		
NSOP or Analgesic details	Meloxicam 0.1ml SC	Meloxicam 0.1ml SC	Meloxicam 0.1ml SC		
Date/Time	1/2/18 5:00pm	1/2/18 5:00pm	1/2/18 5:00pm		
NSOP or Analgesic details	NSOP	NSOP	NSOP		

http://larc.ucsf.edu/forms

飼料

https://dataedo.com/cartoon/the-right-tool

Rodent chow:

- Open, **Closed,** Fixed formula
- Purified, **Grain-based**, Custom designed
  (Natural ingredient)

Common brands:

- US – LabDiet, Research Diet, Dyets
- GRE – Altromin
- JP – Oriental Yest

TABLE 1 Typical sources of nutrients and non-nutrients in rodent purified diets and grain-based diets[1]

	Typical sources	
	Purified-ingredient diet	**Grain-based diet**
Nutrients		
Protein	Casein	Soybean meal, ground corn, wheat, and oats, whey, alfalfa, fish meal, meat meal
Fat	Soybean oil, corn oil	Porcine animal fat, fish meal, meat meal
Carbohydrate	Corn starch, maltodextrin, sucrose	Ground corn, wheat, and oats, wheat middlings
Fiber	Refined cellulose	Ground corn, wheat, and oats, dried beet pulp, alfalfa, wheat middlings
Micronutrients	Mainly vitamin and mineral premixes	Most ingredients, extra micronutrients added
Possible non-nutrients/contaminants		
Phytoestrogens	Absent[2]	Soybean meal (genistein, daidzein), alfalfa meal (coumestrol)
Heavy metals	Trace/not detectable	Grains and animal byproducts (arsenic, lead, cadmium, cobalt)
Pollutants, pesticides, mycotoxins, nitrosamines, endotoxins[3]	Trace/not detectable	Grains (pollutants, mycotoxins) and animal byproducts (pollutants, nitrosamines)

[1] Table adapted from reference 17 with slight modifications.
[2] Unless soy protein isolate is used.
[3] Endotoxin source unknown, but high in grain-based diets (14, 23).

Pellizzon & Ricci, Curr Dev Nutr., 2020

TABLE 2 Questions to ask when considering the diet choice for rodent studies

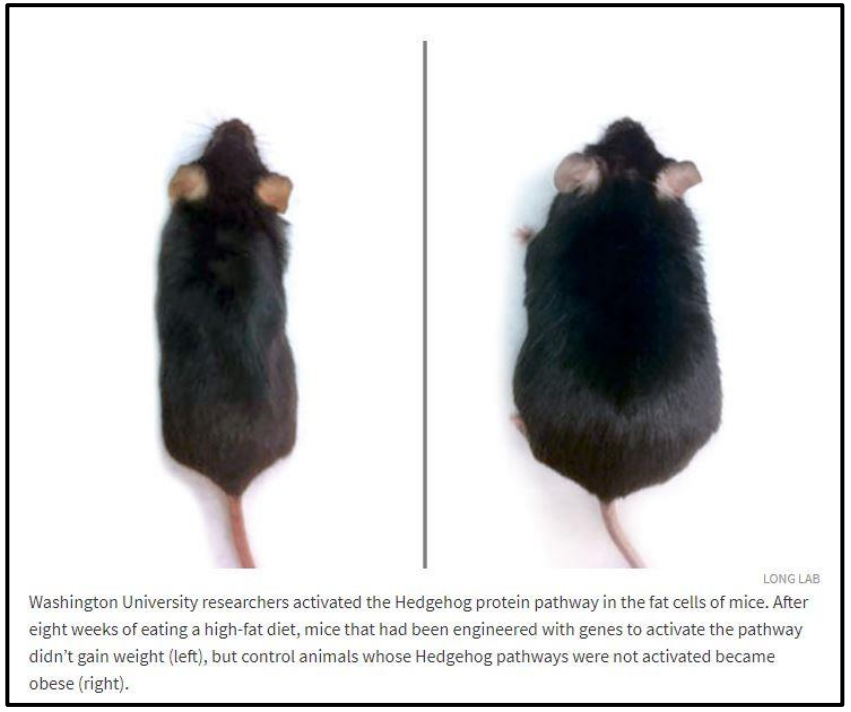
	Purified-ingredient diets	Grain-based diets
Open formulas?	Yes	No[1]
Defined/consistent ingredients?	Yes	No
Can modify 1 nutrient at a time?	Yes	No
Non-nutrient chemical entities?	No	Yes
High/diverse fiber?	No[2]	Yes

[1] Only a select few are open.
[2] Can modify by adding more total fiber and different fiber sources.

Pellizzon & Ricci, Curr Dev Nutr., 2020

Which One is the Properly Matched Control Diet for DIO Model ?

Diet induced obesity (DIO)



Washington University researchers activated the Hedgehog protein pathway in the fat cells of mice. After eight weeks of eating a high-fat diet, mice that had been engineered with genes to activate the pathway didn't gain weight (left), but control animals whose Hedgehog pathways were not activated became obese (right).

https://medicine.wustl.edu/news/obesity-prevented-mice-fed-high-fat-diet/

High fat diet

vs

??? Control diet ???

1. Purified/ Open formula
2. Grain-based/ Closed formula

RESEARCH DIETS INC.
www.ResearchDiets.com

Purified Diet Control vs. Grain-Based Chow

	Purified High Fat Diet	Purified Low Fat Control Diet	Grain-Based Chow	
	Ingredient*	Matched	NOT Matched	Reason
Fat	Lard, soybean oil	✓	X	Variable Sources
Protein	Casein	✓	X	Variable Sources
Carbohydrate	Corn starch, sucrose, maltodextrin	✓	X	Variable Sources
Fiber	Cellulose, insoluble	✓	X	Variable Sources/ 4X Higher
Micronutrients	Vitamins, minerals	✓	X	Variable Level
Phytoestrogens	NONE	✓	X	Variable Level
Heavy Metals	NONE	✓	X	Variable Level

*Ingredients typical of a purified diet, though other purified sources can be used.

FIGURE 1 | Purified diet control versus grain-based chow.

https://www.nature.com/articles/laban.786

動物運輸

2.3.1 動物採購和運輸指導原則：

1. 所有動物應合法取得，接收動物的機構得確信與動物取得有關的所有程序都有依法律規範執行。

2. 動物使用及獲取應依IACUC核准之動物使用方式及數量為之，並得評估供應者的動物品質。

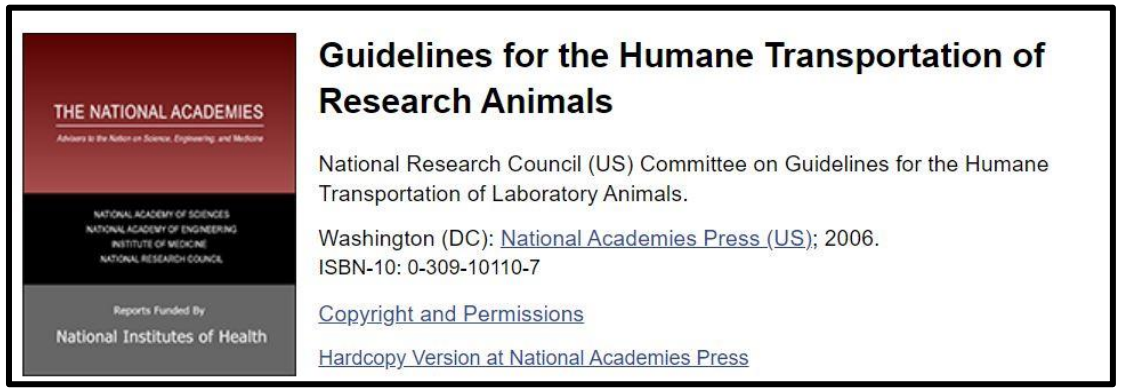
3. 動物運輸機構應遵守國內、外動物運輸相關法規。

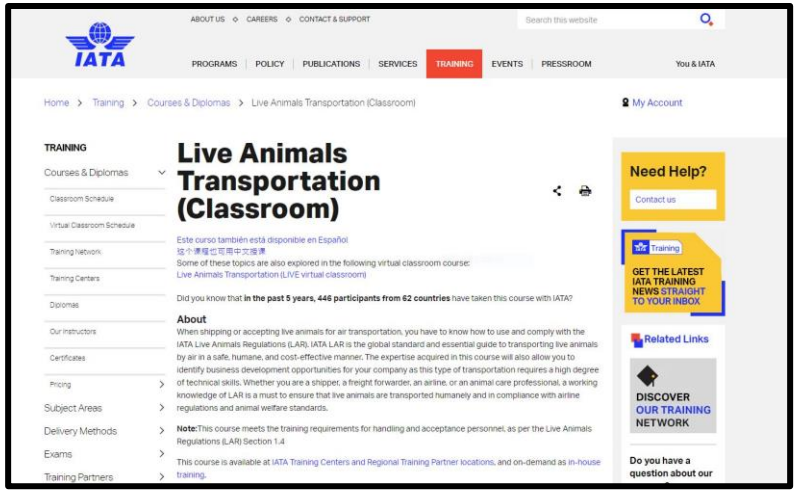
4. 在取得動物之前，研究人員得先確認有足夠的設施及專業人員來飼養和管理所取得的動物。
5. 當機構為研究人員取得動物後，相關的紀錄及表格文件都得妥善保存。得將使用之動物族群數量加以考量。繁殖族群得依需求及動物減量原則(例如凍胚)管理。
6. 得評估動物供應商所提供的動物品質。一般而言，專門繁殖販售實驗用動物的供應商會定期提供所飼養之動物族群或個體的遺傳與健康監測報告，或相關臨床醫療紀錄（例如疫苗施打及驅蟲紀錄）。

7. 動物運輸得仔細規劃，以確保動物的安全和福祉。運輸過程中得提供適當等級的動物生物保全措施。為移動的安全性，得提供適當的裝載及卸載設施以維護動物福祉及人員安全。

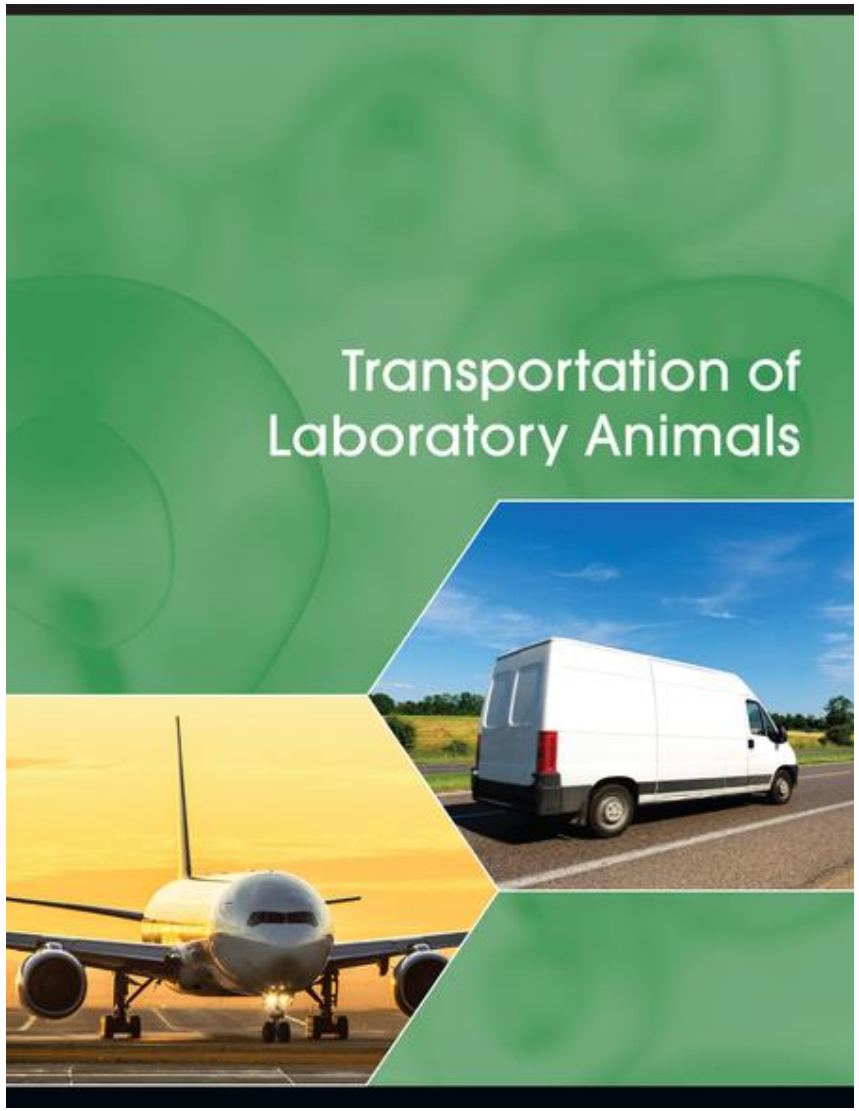
8. 進行不同場所或機構間動物移動作業時，得由雙方機構具備受訓資格的人員執行規劃及協調的工作，以縮短運輸時間或避免發生接收延誤的狀況。動物運輸得協調儘量在上班時間送抵，若要於非上班時間送抵，要安排接收人員。動物運輸時得隨附相關文件，以減少運送及接收程序延誤。

9. 相關設施及作業程序得事先備妥，以協助確認運輸作業環境無任何會對動物福祉或人員安全構成威脅的狀況。





The Guide 8th



https://www.nap.edu/read/21734/chapter/2

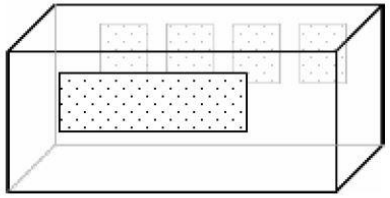
Vehicle

Ideally, all animals should be transported in **environmentally controlled vehicles**. (The Guide – temp. & humidity)

When animals cannot be transported in environmentally-controlled vehicles, the committee **recommends frequent visual inspection** of the animals when practical, as signs of thermogenesis or heat loss may indicate that the animal's thermal environment should be adjusted.

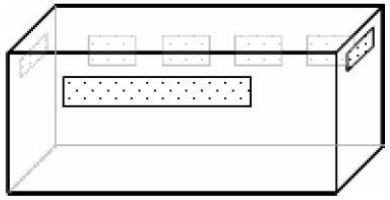
Enclosure

Option A: Two ventilation openings



Surface area of each opening is at least 16% of surface area of that wall.
One-third of surface area of openings must be in lower half of enclosure, and at least one-third of surface area of openings must be on upper half of enclosure.

Option B: Four ventilation openings

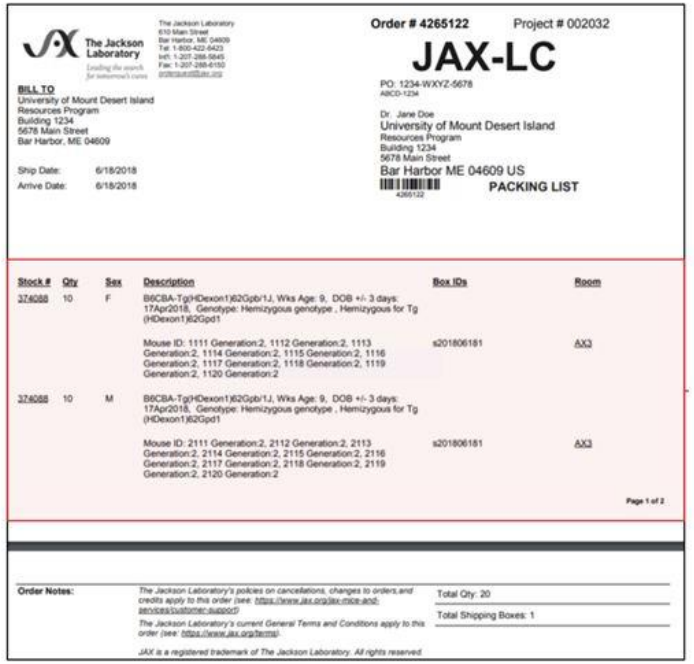


Surface area of each opening is at least 8% of surface area of that wall.
One-third of surface area of openings must be in lower half of enclosure, and at least one-third of surface area of openings must be on upper half of enclosure.

- **Housing density**, following the Guide.
- Projecting rims must be on the exterior of walls that contain ventilation openings and provide a minimal air circulation space of **0.75 in (≅ 2 cm)**.
- Unless the enclosure is permanently affixed to the conveyance, it must be marked on top and on at least one side with the words "**Live Animals**" or "Wild Animals" in letters at least 1 in high and with arrows indicating the correct upright position of the enclosure.

GUINEA PIGS AND HAMSTERS, RABBITS, AND OTHER ANIMALS
(9 CFR 3.35-3.41, 9 CFR 3.60-3.65, 9 CFR 3.136 – 3.142)

Shipping Containers



Charles River

Shipping Containers

Rodents OUTSIDE Small Non-rodents

Univ. of IOWA

https://www.artstation.com/artwork/Vd6X8N

https://vishnutraders.in/new/wide-range-of-water-bottle/

Homemade – Agar, 吉利丁, 洋菜膠（1-5%）, autoclaved

其它注意事項

Records

- Basic information – Animals, Shipper, Receiver, Route planning, Vehicle, Time, Temp., Humidity
- Emergency plan

Container for ABSL-2 , chemical agents, toxins or radioisotopes

- A secondary container is required with the property of leak-proof & shatter-proof
- Decontamination plan

ABSA INTERNATIONAL
The Association for Biosafety and Biosecurity

Robber band

Tape

ABSL-2

https://absa.org/absl2vid/

其它注意事項

Acclamation

- Animals need to take a rest after transportation, e.g., **3 days** for rodents
- Return to normal/ homeostasis

Will you bring animals back to the original facility?

- Biosecurity
- Holding area ?

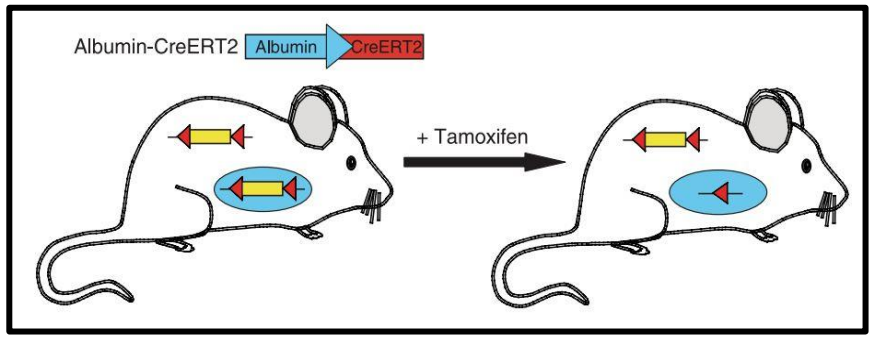
危害物質

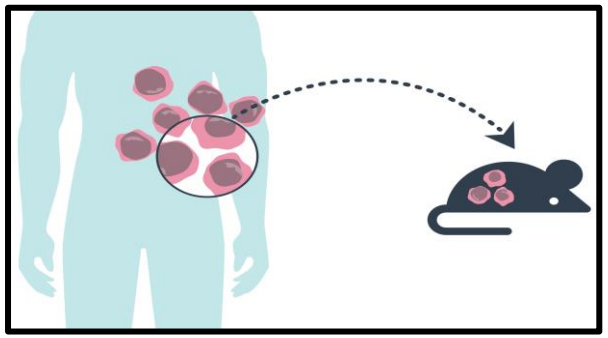
十三、有無進行危險性實驗，如生物危險（含感染性物質、致癌藥物）、放射線及化學危險（含毒物）實驗？ □ 無 □有

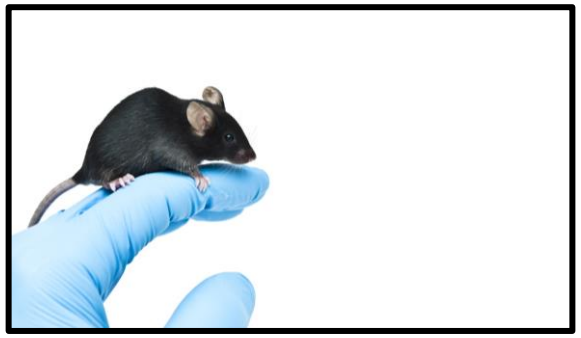
危害物質

- 生物性 - Infectious, Biotoxin, Recomb. DNA, Human cell-lines, Human fluids
- 化學性 - Mutagens, Carcinogens, Teratogens, Immunosuppression
- 物理性 - Radiation

Gene modifications	Anti-cancer drugs	Chemicals
Tamoxifen **Diphtheria toxin**	**Cisplatin** **Paclitaxel**	**STZ** **BrdU**







https://www.creative-biolabs.com/drug-discovery/therapeutics/inducible-gene-knock-out.htm
https://www.cancer.gov/news-events/cancer-currents-blog/2021/cancer-pdx-mouse-models-retain-genetics-human-tumor
https://www.fda.gov/drugs/news-events-human-drugs/spotlight-cder-science-novel-mouse-model-offers-insights-about-hypersensitivity-hiv-drug

標記 & 基因分型

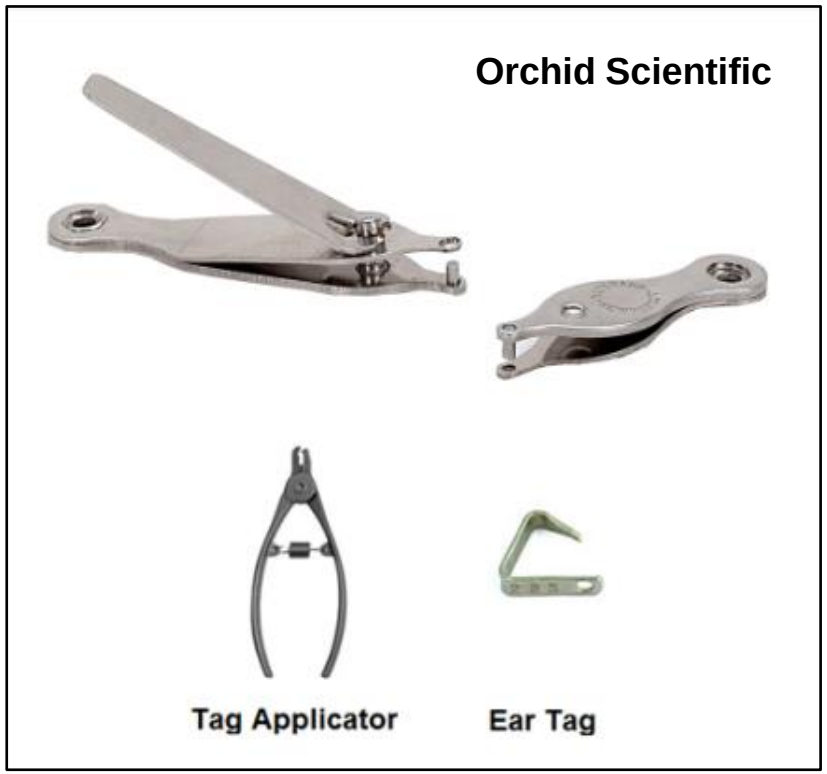
附錄一、實驗動物繁殖表

六、是否為基因改造動物?

(三)請說明篩選基因用採樣方法與採樣時間：

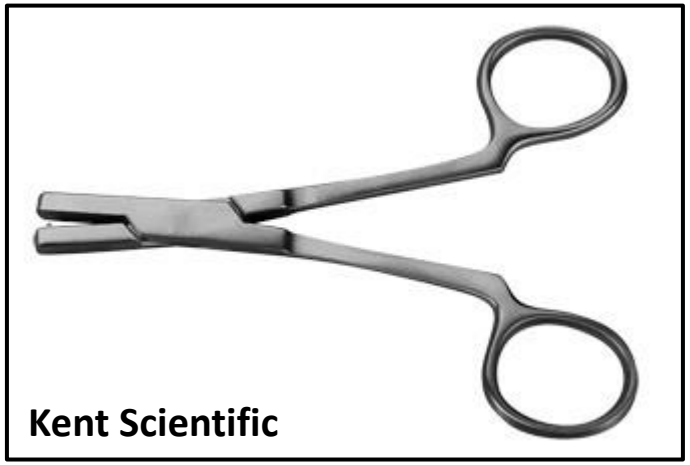
標記


Stoelting


Orchid Scientific
Tag Applicator
Ear Tag

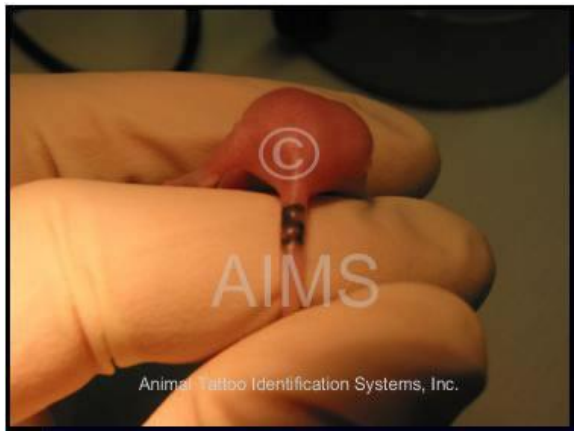

Stoelting


Braintree Scientific


Kent Scientific

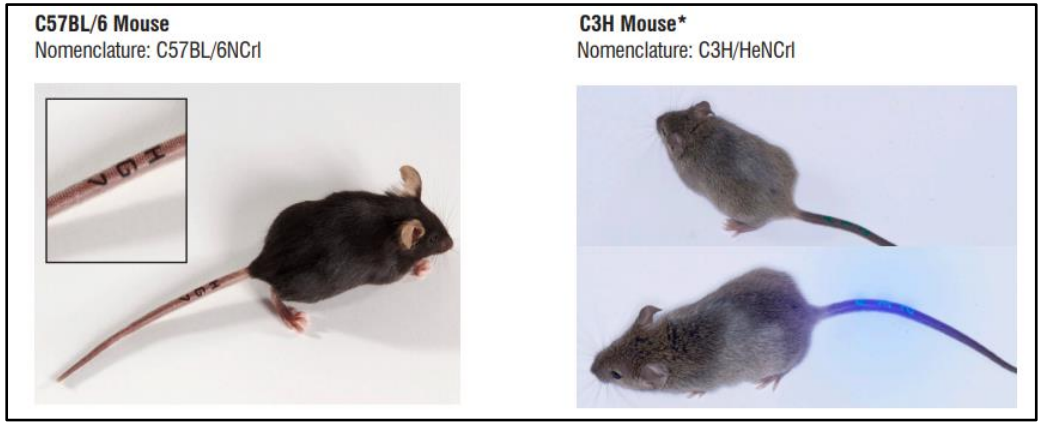

AIMS
© Animal Tattoo Identification Systems, Inc.


AIMS
Animal Tattoo Identification Systems, Inc.


AIMS
Animal Tattoo Identification Systems, Inc.


Chip


Somark - Labstamp
SensaLab
LABSTAMP
START


C57BL/6 Mouse
Nomenclature: C57BL/6NCrl
C3H Mouse*
Nomenclature: C3H/HeNCrl

Tail Snap for Genotyping in Mice

- Do it before tail ossification, which means before **21d** of age
- 3~5mm is enough
- Analgesia
- **Alternative** – ear biopsy

Toe clip ≤ 7d of age in mice

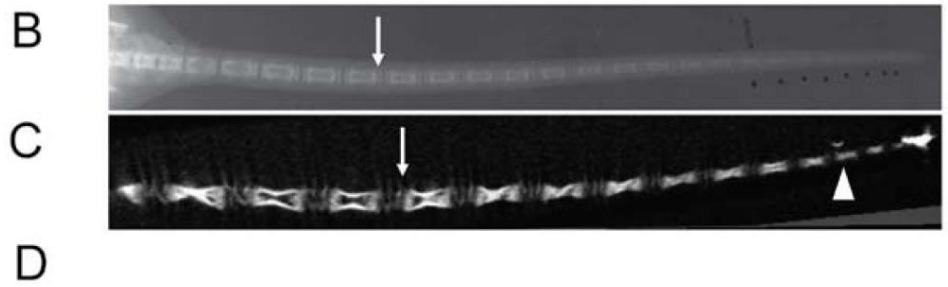


Table 1. Differences in total coccygeal vertebrae based on genetic background

Strain	Age (d)		
	3	21	42
129	16	26	27
BALB/c	21	27	28
B6	15	26	27
C3H	18	25	28
FVB	16	27	28
SW	18	27	29

Values represent the mean vertebral number from 6 mice at each age using microRad.

Hankenson et al., JAALAS, 2008

https://www.quora.com/What-are-some-other-popular-Q-A-websites