**Hargreaves Test UCD_HRG_001**

**Purpose**

To assess thermal hyperalgesia in the mouse paw.

**Experimental Design**

Minimum number of mutant animals: 7 males + 7 females

Age at test: 16-17 weeks

Sexual dimorphism:

**Procedure**

1. Baseline measurement
   1. Transfer mouse to the testing arena and allow to acclimate to the test environment.
   2. Ensure that the intensity and cut-off timer of the radiant heat source are set correctly.
   3. Position the light source below the platform and directed at the correct part of the paw.
   4. Turn the light source on and record the latency to paw withdrawal. The stimulus is terminated once a response is registered, or if the maximum exposure time is reached before the animal responds.
   5. Repeat the test so that each mouse is tested three times with the same paw.
2. Challenge
   1. The challenge is administered after the baseline measurement has been completed.
   2. Anaesthetise the mouse and administer the challenge injection.
3. Test 1
   1. After the specified length of time since the challenge injection, the mouse is re-tested using the same procedure as described for the baseline measurement.
4. Test 2
   1. After the specified length of time since the challenge injection, the mouse is re-tested again using the same procedure as described for the baseline measurement.

**Notes**

This procedure is a pilot study from the Pain Phenotyping Pilot

**Parameters and Metadata**
**Radiant heat light source manufacturer** UCD_HRG_032_001 | v1.0

** Procedure Metadata **

* Options: IITC, *

** Site of challenge injection** UCD_HRG_013_001 | v1.0

** Procedure Metadata **

* Options: Plantar surface of right hind paw, *

** Testing chamber dimensions** UCD_HRG_029_001 | v1.0

** Procedure Metadata **

* Unit Measured: cm *

* Options: 12.7 cm H x 10.16 cm W x 10.16 cm L, 12.5 cm H x 10 cm W x 10 cm L, *

** Disinfectant** UCD_HRG_035_001 | v1.0

** Procedure Metadata **
Test 2: average latency to withdrawal  UCD_HRG_006_001 | v1.0

Simple Parameter

Unit Measured: s

Derivation: meanOfIncrements('UCD_HRG_005_001',1)

Time between challenge and test 2  UCD_HRG_017_001 | v1.0

Procedure Metadata

Unit Measured: Hours

Options: 144, 48,

Experimenter ID  UCD_HRG_034_001 | v1.0

Procedure Metadata
**Test 1: average latency to withdrawal**  
*UCD_HRG_004_001 | v1.0*

simpleParameter

- Unit Measured: s
- Derivation: $\text{meanOfIncrements('UCD_HRG_003_001',1)}$

**Anaesthetic for challenge injection**  
*UCD_HRG_014_001 | v1.0*

procedureMetadata

- Options: Isoflurane,

**Lid colour/opacity**  
*UCD_HRG_028_001 | v1.0*

procedureMetadata

- Options: Clear,
Glass base heated during acclimatisation

Options: Yes,

Baseline: average latency to withdrawal

Unit Measured: s

Derivation: meanOfIncrements('UCD_HRG_001_001',1)

Length of acclimatisation period

Unit Measured: min

Options: 60, 30, 30 - 60,
**Test intensity**  
UCD_HRG_021_001 | v1.0

Procedure Metadata

Req. Analysis: false  
Req. Upload: true  
Is Annotated: false

Unit Measured: %

Options: 100, 25, 30, 15,

**Glass base manufacturer**  
UCD_HRG_030_001 | v1.0

Procedure Metadata

Req. Analysis: false  
Req. Upload: true  
Is Annotated: false

Options: IITC,

**Idle intensity**  
UCD_HRG_020_001 | v1.0

Procedure Metadata

Req. Analysis: false  
Req. Upload: true  
Is Annotated: false

Unit Measured: %

Options: 2, 3, 10,
**Tetrad colour/opacity** UCD_HRG_024_001 | v1.0

procedureMetadata


Options: Clear,

**Tetrad manufacturer** UCD_HRG_022_001 | v1.0

procedureMetadata


Options: IITC,

**Time between challenge and test 1** UCD_HRG_016_001 | v1.0

procedureMetadata


Unit Measured: Hours

Options: 24,
**Minimum time between stimulus presentation**  
UCD_HRG_011_001 | v1.0

Unit Measured: min

Options: 2,

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**Tetrad material**  
UCD_HRG_023_001 | v1.0

Options: Acrylic,

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**Baseline: latency to withdrawal**  
UCD_HRG_001_001 | v1.0

Unit Measured: s

Increments: 1, 2, 3,
**Glass base temperature: settings**  UCD_HRG_018_001 | v1.0

*procedureMetadata*

**Unit Measured:** C

**Options:** 34, 30, 32,

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**Inset colour/opacity**  UCD_HRG_026_001 | v1.0

*procedureMetadata*

**Options:** White opaque, Clear,

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**Radiant heat light source model**  UCD_HRG_033_001 | v1.0

*procedureMetadata*

**Options:** 390 Plantar test head (Item #300TH),

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**Time between baseline measurement and challenge injection**  UCD_HRG_015_001 | v1.0
Is Annotated: false  
Req. Upload: true  
Unit Measured: Hours

Options: 0.5-1, 2, 24,

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**Challenge** UCD_HRG_012_001 | v1.0

Options: CFA,

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**Test 2: latency to withdrawal** UCD_HRG_005_001 | v1.0

Unit Measured: s

Increments: 1, 2, 3,

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**Inset material** UCD_HRG_025_001 | v1.0
Options: Acrylic,

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**Lid material** UCD_HRG_027_001 | v1.0

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**Test 1: latency to withdrawal** UCD_HRG_003_001 | v1.0

Unit Measured: s

Increments: 2, 1, 3,

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**Glass base model** UCD_HRG_031_001 | v1.0
Options: Model 400 Heated Base (Item #400G),

Number of repeats per paw UCD_HRG_010_001 | v1.0


Options: Right hind paw tested 3 times,

Maximum exposure time UCD_HRG_009_001 | v1.0


Unit Measured: s

Options: 30, 20,

Glass base temperature: measured UCD_HRG_019_001 | v1.0


Unit Measured: C
Options: 29-32,