Fear Conditioning IMPC_FEA_001

Purpose

This procedure is used for measuring aversive learning and memory. A neutral conditioned stimulus (CS) such as steady tone is paired with an aversive unconditioned stimulus (US) such as one or more mild foot shocks. After conditioning, the spatial context or the CS (tone) elicits a central state of fear in the absence of the US (shock) that is expressed as reduced locomotor activity or total lack of movement (freezing). Immobility time is used as a measure of learning/memory performances.

Experimental Design

- Minimum number of animals: 7M + 7F
- Age at test: Week 11
- Sex: We would expect the results of this test to show sexual dimorphism

Equipment

- Fear conditioning chambers
- Arenas and components for cue and conditioning phases
- Computer with recording software
- Olfactory substance

Procedure

1. Day 1: Conditioning
   a. Transport animals to the testing area.
   b. Set up conditioning arenas in the chambers ensuring that shocker, tone delivery and camera/beam break system are working correctly. If using video tracking equipment, ensure camera lenses are free of dust.
   c. Remove the animals from their cages and place in the arenas. Animals are conditioned using the training protocol which consists of a baseline period, audible tone, a single foot shock that co-terminates with the tone, and a period with no stimuli at the end.
   d. Remove animals from arenas and place back in home cage. Clean arenas.
2. Day 2: Context
   a. To be performed 24 hours after conditioning.
   b. Transport animals to the testing area.
   c. Set up conditioning arenas in the chamber as on day 1 except for shocker and tone delivery system which are not used.
   d. Remove animals from their cages and place in the arena. No CS or US stimuli are presented.
3. Day 2: Cue
   a. To be performed 2 - 24 hours after the context protocol.
   b. Set up cue arenas using different floors, walls, olfactory cues and/or lighting from the conditioning context. Ensure that tone delivery and camera/beam break system are working correctly.
   c. Remove animals from their cages and place in the arena. Animals are exposed to a baseline period without any stimulus followed by the presentation of the tone.
   d. Remove animals from arenas and place back in home cage. Clean arenas.

4. At the end of the cue protocol animals are placed back into their holding room and data is analyzed by collecting the number of freezing episodes and the total freezing/immobility time.

## Parameters and Metadata

### Conditioning Baseline Freeze Count

| Description: conditioning_baseline_freeze_count |

### Conditioning Baseline Freezing Time

| Unit Measured: s |

### Conditioning Baseline % Freezing Time
simpleParameter

**Conditioning Baseline Average Motion Index** IMPC_FEA_004_01 | v1.0

**Description:** conditioning_baseline_average_motion_index

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**Conditioning Baseline Minimum Motion Index** IMPC_FEA_005_001 | v1.0

**Description:** conditioning_baseline_minimum_motion_index

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Conditioning Baseline Maximum Motion Index  IMPC_FEA_006_001 | v1.0

simpleParameter


Description: conditioning_baseline_maximum_motion_index

Context Freeze Count  IMPC_FEA_007_001 | v1.0

simpleParameter


Description: context_freeze_count

Context Freezing Time  IMPC_FEA_008_001 | v1.0

simpleParameter


Unit Measured: s

Description: context_freezing_time

Context % Freezing Time  IMPC_FEA_009_001 | v1.1
**Description:** context_freezing_time

**Derivation:** \( \text{mul}(\text{div('IMPC_FEA_008_001', 'IMPC_FEA_049_001')}, 100) \)

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**Context Average Motion Index** IMPC_FEA_010_001 | v1.0

**Description:** context_average_motion_index

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**Context Minimum Motion Index** IMPC_FEA_011_001 | v1.0

**Description:** context_minimum_motion_index

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**Context Maximum Motion Index** IMPC_FEA_012_001 | v1.0

**Description:** context_maximum_motion_index
Description: context\_maximum\_motion\_index

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**Cue Baseline Freeze Count**  IMPC\_FEA\_013\_001  |  v1.0

simpleParameter

Description: cue\_baseline\_freeze\_count

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**Cue Baseline Freezing Time**  IMPC\_FEA\_014\_001  |  v1.0

simpleParameter

Unit Measured: s

Description: cue\_baseline\_freezing\_time

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**Cue Baseline % Freezing Time**  IMPC\_FEA\_015\_001  |  v1.1

simpleParameter

Description: cue\_baseline\_freezing\_time

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Unit Measured: %

Description: cue_baseline_freezing_time

Derivation: \( \text{mul}\left(\text{div}\left('IMPC\_FEA\_014\_001', 'IMPC\_FEA\_057\_001'\right), 100\right) \)

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**Cue Baseline Average Motion Index** IMPC\_FEA\_016\_001 | v1.0

simpleParameter


Description: cue_baseline_average_motion_index

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**Cue Baseline Minimum Motion Index** IMPC\_FEA\_017\_001 | v1.0

simpleParameter


Description: cue_baseline_minimum_motion_index

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**Cue Baseline Maximum Motion Index** IMPC\_FEA\_018\_001 | v1.0

simpleParameter

Description: cue_baseline_maximum_motion_index

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**Cue Tone Freeze Count** IMPC_FEA_019_001 | v1.0

*seriesParameter*


Description: cue_tone_freeze_count

Increments: Minimum 1

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**Cue Tone Freezing Time** IMPC_FEA_020_001 | v1.0

*seriesParameter*


Unit Measured: s

Description: cue_tone_freezing_time

Increments: Minimum 1

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**Cue Tone % Freezing Time** IMPC_FEA_021_001 | v1.1

*simpleParameter*

Unit Measured: %

Description: cue_tone_freezing_time

Derivation:
\[ \text{mul}(\text{div}((\text{sumOfIncrements('IMPC_FEA_020_001',1)}, 'IMPC_FEA_060_001'), 100) \]

Cue Tone Average Motion Index  IMPC_FEA_022_001  | v1.0

Description: cue_tone_average_motion_index

Increments: Minimum 1

Cue Tone Minimum Motion Index  IMPC_FEA_023_001  | v1.0

Description: cue_tone_minimum_motion_index

Increments: Minimum 1

Cue Tone Maximum Motion Index  IMPC_FEA_024_001  | v1.0
Description: cue_tone_maximum_motion_index

Increments: Minimum 1

Conditioning: Date and time IMPC_FEA_025_001 | v1.0

Description: conditioning_date_and_time

Conditioning: Room acclimation time IMPC_FEA_026_001 | v1.0

Unit Measured: min

Description: conditioning_room_acclimation_time

Options: 30, 60,
**Conditioning: White house light level in arena** IMPC_FEA_027_001 | v1.0

**procedureMetadata**

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

**Unit Measured:** Lux

**Description:** conditioning_white_house_light_level_in_arena

**Options:** 26, 100, 200, 40,

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**Conditioning: Olfactory cue** IMPC_FEA_028_001 | v1.0

**procedureMetadata**

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

**Description:** conditioning_olfactory_cue

**Options:** Alcohol, None,

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**Conditioning: Mouse arena shape** IMPC_FEA_029_001 | v1.0

**procedureMetadata**

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

**Description:** conditioning_mouse_arena_shape

**Options:** Rectangle, Square,
Conditioning: Mouse arena dimension IMPC_FEA_030_001 | v1.0

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

Unit Measured: cm

Description: conditioning_mouse_arena_dimension

Options: 30.5 L X 24.1 W X 24.5 H cm, 16.6 L X 17 W X 25 H cm, 25 L x 21 W x 19 H cm, 25 x 25 x 19 cm, 17 x 17 x 25 cm, 30.5 cm L X 24.1 cm W X 21 cm H,

Conditioning: Mouse arena colour IMPC_FEA_031_001 | v1.0

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

Description: conditioning_mouse_arena_colour

Options: Metallic, White and Clear, Clear, White, Metallic, clear,

Conditioning: Mouse arena floor IMPC_FEA_032_001 | v1.0

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

Options: Metal grill, Clean saw dust, Steel rods,

Conditioning: Length of time baseline  IMPC_FEA_033_001 | v1.0


Unit Measured: s

Description: conditioning_length_of_time_baseline

Options: 240, 150, 120,

Conditioning: Tone stimulus intensity ( dB; conditioned stimulus)  IMPC_FEA_034_001 | v1.0


Unit Measured: dB

Description: conditioning_tone_stimulus_intensity_db_conditioned_stimulus

Options: 85, 80, 75, 77, 70,
**Conditioning: Tone stimulus frequency (Hz; conditioned stimulus)**  
IMPC_FEA_035_001 | v1.0

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

**Unit Measured:** kHz

**Description:** conditioning_tone_stimulus_frequency_hz_conditioned_stimulus

**Options:** 10, 2.8, 4, 2,

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**Conditioning: Length of time tone stimulus**  
IMPC_FEA_036_001 | v1.0

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

**Unit Measured:** s

**Description:** conditioning_length_of_time_tone_stimulus

**Options:** 20, 30,

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**Conditioning: Shock stimulus intensity (unconditioned stimulus)**  
IMPC_FEA_037_001 | v1.1

- **Req. Analysis:** true  
- **Req. Upload:** true  
- **Is Annotated:** false
Unit Measured: mA

Description: conditioning_shock_stimulus_intensity_unconditioned_stimulus

Options: 0.4, 0.5, 0.75,

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**Conditioning: Length of time shock stimulus**  IMPC_FEA_038_001  v1.0

**Procedure Metadata**


Unit Measured: s

Description: conditioning_length_of_time_shock_stimulus

Options: 0.5, 1, 2,

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**Conditioning: Length of time no stimulus**  IMPC_FEA_039_001  v1.0

**Procedure Metadata**


Unit Measured: s

Description: conditioning_length_of_time_no_stimulus

Options: 120, 150,
**Conditioning: Total time**  IMPC_FEA_040_001 | v1.0

*procedureMetadata*

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

**Unit Measured:** s

**Description:** conditioning_total_time

**Options:** 380, 300,

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**Context: Date and time**  IMPC_FEA_041_001 | v1.0

*procedureMetadata*

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

**Description:** context_date_and_time

**Options:** Conditioning +24h,

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**Context: Room acclimation time**  IMPC_FEA_042_001 | v1.0

*procedureMetadata*

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

**Unit Measured:** min
Description: context_room_acclimation_time
Options: 30, 60,

Context: White house light level in arena IMPC_FEA_043_001 | v1.2


Unit Measured: Lux

Description: context_white_house_light_level_in_arena
Options: 26, 100, 200, 40,

Context: Olfactory cue IMPC_FEA_044_001 | v1.0


Description: context_olfactory_cue
Options: Alcohol, None,

Context: Mouse arena shape IMPC_FEA_045_001 | v1.0
procedureMetadata

**Description:** context_mouse_arena_shape

**Options:** Rectangle, Square,

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**Context:** Mouse arena dimension  IMPC_FEA_046_001 | v1.0

**Unit Measured:** cm

**Description:** context_mouse_arena_dimension

**Options:** 30.5 cm L X 24.1 cm W X 24.5 cm H, 16.6 cm L X 17 cm W X 25 cm H, 25 L x 21 W x 19 H cm, 25 x 25 x 19 cm, 17 x 17 x 25 cm, 30.5 cm L X 24.1 cm W X 21 cm H,

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**Context:** Mouse arena colour  IMPC_FEA_047_001 | v1.0

**Description:** context_mouse_arena_colour

**Options:** Metallic, White and Clear, Clear, White, Metallic, clear,
**Context: Mouse arena floor** IMPC_FEA_048_001 | v1.0

**Description:** context_mouse_arena_floor

**Options:** Metal grill, Steel rods,

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**Context: Total time** IMPC_FEA_049_001 | v1.0

**Description:** context_total_time

**Options:** 360, 300,

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**Cue: Date and time** IMPC_FEA_050_001 | v1.0

**Description:** cue_date_and_time
**Options:** Context + 2-4h, Context + 4h, Context + 24h,

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**Cue: White house light level in arena**  IMPC_FEA_051_001 | v1.1

*procedureMetadata*

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

**Unit Measured:** Lux

**Description:** cue_white_house_light_level_in_arena

**Options:** 0, 16, 14-18,

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**Cue: Olfactory cue**  IMPC_FEA_052_001 | v1.0

*procedureMetadata*

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

**Description:** cue_olfactory_cue

**Options:** 5% acetic acid, Vanillin, Cinnamon,

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**Cue: Mouse arena shape**  IMPC_FEA_053_001 | v1.0

*procedureMetadata*

- **Req. Analysis:** false
- **Req. Upload:** true
- **Is Annotated:** false
Description: cue_mouse_arena_shape

Options: A frame ceiling, Cylinder, Rectangle,

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Cue: Mouse arena dimension IMPC_FEA_054_001 | v1.0

procedureMetadata


Unit Measured: cm

Description: cue_mouse_arena_dimension

Options: 30.5 cm L X 24.1 cm W X 24.5 cm H, 20 cm dia X 30 cm H, 25 L x 21 W x 19 H cm, 25 x 25 x 19 cm, 17 x 17 x 25 cm, 30.5 cm L X 24.1 cm W X 21 cm H,

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Cue: Mouse arena colour IMPC_FEA_055_001 | v1.0

procedureMetadata


Description: cue_mouse_arena_colour

Options: Black and white, Red, Clear, Beige, black and white,

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Cue: Mouse arena floor IMPC_FEA_056_001 | v1.0

procedureMetadata
Cue: Length of time baseline  IMPC_FEA_057_001 | v1.0

Requirements:
- Analysis: true
- Upload: true
- Annotated: false

Description: cue_length_of_time_baseline

Options: 120,

Unit Measured: s

Cue: Tone stimulus intensity (conditioned stimulus)  IMPC_FEA_058_001 | v1.1

Requirements:
- Analysis: true
- Upload: true
- Annotated: false

Description: cue_tone_stimulus_intensity_conditioned_stimulus

Options: 85, 80, 75, 77, 70,
Cue: Tone stimulus frequency (Hz; conditioned stimulus)  
MPC_FEA_059_001 | v1.0


Unit Measured: kHz

Description: cue_tone_stimulus_frequency_hz_conditioned_stimulus

Options: 10, 2.8, 4, 2,

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Cue: tone response measurement time  IMPC_FEA_060_001 | v1.0


Unit Measured: s

Options: 120, 180,

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Cue: Length of time no stimulus  IMPC_FEA_061_001 | v1.0


Unit Measured: s
Description: cue_length_of_time_no_stimulus
Options: 120, 0, 220,

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**Cue: Total time**  IMPC_FEA_062_001 | v1.0
procedureMetadata


Unit Measured: s
Description: cue_total_time
Options: 480, 300, 420, 240,

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**Mouse arenaID**  IMPC_FEA_063_001 | v1.0
procedureMetadata


Description: mouse_arenaid
Options: Arena 1, Arena 2, Arena 3, Arena 4, Arena 5, Arena 6, Arena 7, Arena 8,

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**Equipment ID**  IMPC_FEA_064_001 | v1.0
procedureMetadata
Description: equipment_id

Options: Machine A, Machine B, Machine 1, Machine 2,

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**Equipment Manufacturer** IMPC_FEA_065_001 | v1.0

Description: equipment_manufacturer

Options: Med Assoicates, Ugo Basile, San Diego Instruments,

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**Equipment Model** IMPC_FEA_066_001 | v1.0

Description: equipment_model

Options: MED-FVC-SCT-M, Freeze monitor, FearCS Mouse-46152, MED-VFC2-SCT-M, 46000, MED-VFC-NIR-M SOP #1, MED-VFC-NIR-M SOP #3, MED-VFC-NIR-M SOP #2,

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**Tracking Method** IMPC_FEA_067_001 | v1.0
procedureMetadata


Description: tracking_method

Options: Video tracking, Beam break,

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Software  IMPC_FEA_068_001 | v1.0

procedureMetadata


Description: software

Options: Med Associates Video Freeze, Anymaze, Freeze Monitor,

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Sound generator manufacturer  IMPC_FEA_069_001 | v1.0

procedureMetadata


Description: sound_generator_manufacturer

Options: Med Associates, Ugo Basile, San Diego Instruments,
Sound generator model IMPC_FEA_070_001 | v1.0

Description: sound_generator_model
Options: VFC-100, other, 46000-165,
**Experimenter ID day 2** IMPC_FEA_073_001 | v1.0

**Description:** experimenter_id_day_2

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**Experimenter ID day 2 cue** IMPC_FEA_074_001 | v1.0

**Description:** experimenter_id_day_2_cue

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**Date equipment last calibrated (shock)** IMPC_FEA_075_001 | v1.0

**Description:** date_equipment_last_calibrated_shock

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**Date equipment last calibrated (tone)** IMPC_FEA_076_001 | v1.0

**Description:** date_equipment_last_calibrated_tone
Description: date_equipment_last_calibrated_tone

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**Freeze definition** IMPC_FEA_077_001 | v1.1


Unit Measured: s

Description: freeze_definition

Options: 1, 2, 0.25,

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**Conditioning: Near infra red light arena** IMPC_FEA_078_001 | v1.0


Description: conditioning_near_infra_red_light_arena

Options: On,

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**Context: Near infra red light arena** IMPC_FEA_079_001 | v1.0

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Cue: Near infra red light arena  IMPC_FEA_080_001 | v1.0

Conditioning: Start time of baseline  IMPC_FEA_081_001 | v1.0

Conditioning: Start time of each tone stimulus  IMPC_FEA_082_001 | v1.0
procedureMetadata

**Conditioning: Start time of each tone stimulus**  IMPC_FEA_0

83_001 | v1.0

procedureMetadata

**Conditioning: Start time of each shock stimulus**  IMPC_FEA_0

83_001 | v1.0

procedureMetadata

**Conditioning: Start time of no stimulus**  IMPC_FEA_084_001 | v1.0
**Cue: Start time of baseline**  IMPC_FEA_085_001 | v1.0

*procedureMetadata*

**Unit Measured:** s

**Description:** cue_start_time_of_baseline

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**Cue: Start time of each tone stimulus**  IMPC_FEA_086_001 | v1.0

*procedureMetadata*

**Unit Measured:** s

**Description:** cue_start_time_of_each_tone_stimulus

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**Cue: Start time of no stimulus**  IMPC_FEA_087_001 | v1.0

*procedureMetadata*

**Unit Measured:** s

**Description:** cue_start_time_of_no_stimulus
Cue: arena rotation  IMPC_FEA_088_001  | v1.0

procedureMetadata


Description: cue_arena_rotation

Options: 90 degrees,
**Conditioning Tone % Freezing Time**  IMPC_FEA_091_001 | v1.1

simpleParameter

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

**Unit Measured:** %

**Description:** conditioning_tone_freezing_time

**Derivation:** \( \text{mul}(\text{div}('IMPC_FEA_090_001', 'IMPC_FEA_036_001'), 100) \)

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**Conditioning Tone Average Motion Index**  IMPC_FEA_092_001 | v1.0

simpleParameter

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

**Description:** conditioning_tone_average_motion_index

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**Conditioning Tone Minimum Motion Index**  IMPC_FEA_093_001 | v1.0

simpleParameter

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

**Description:** conditioning_tone_minimum_motion_index
Conditioning Tone Maximum Motion Index  IMPC_FEA_094_001 | v1.0

Description: conditioning_tone_maximum_motion_index

Conditioning Shock Average Motion Index  IMPC_FEA_095_001 | v1.0

Description: conditioning_shock_average_motion_index

Conditioning Shock Minimum Motion Index  IMPC_FEA_096_001 | v1.0

Description: conditioning_shock_minimum_motion_index
Conditioning Shock Maximum Motion Index  IMPC_FEA_097_001  | v1.0

simpleParameter


Description: conditioning_shock_maximum_motion_index

Conditioning Post-shock Freeze Count  IMPC_FEA_098_001  | v1.0

simpleParameter


Description: conditioning_post_shock_freeze_count

Conditioning Post-shock Freezing Time  IMPC_FEA_099_001  | v1.0

simpleParameter


Unit Measured: s

Description: conditioning_post_shock_freezing_time
Conditioning Post-shock % Freezing Time  IMPC_FEA_100_001 | v1.1

simpleParameter


Unit Measured: %

Description: conditioning_post_shock_freezing_time

Derivation: mul(div('IMPC_FEA_099_001', 'IMPC_FEA_039_001'), 100)

Conditioning Post-shock Average Motion Index  IMPC_FEA_101_001 | v1.0

simpleParameter


Description: conditioning_post_shock_average_motion_index

Conditioning Post-shock Minimum Motion Index  IMPC_FEA_102_001 | v1.0

simpleParameter


Description: conditioning_post_shock_minimum_motion_index
**Conditioning Post-shock Maximum Motion Index**  IMPC_FEA_103_001 | v1.0

**simpleParameter**


**Description:** conditioning_post_shock_maximum_motion_index

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**Freeze definition (minimum motion index)**  IMPC_FEA_104_001 | v1.0

**procedureMetadata**


**Description:** freeze_definition_minimum_motion_index

**Options:** 18, 30,

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**Difference in Context and Conditioning Baseline % Freezing**  IMPC_FEA_105_001 | v1.0

**simpleParameter**

Derivation:
sub(mul(div('IMPC_FEA_008_001', 'IMPC_FEA_049_001'), 100), mul(div('IMPC_FEA_002_001', 'IMPC_FEA_033_001'), 100))

Difference in Cue Tone and Cue Baseline % Freezing IMPC_FEA_106_001 | v1.0

simpleParameter


Derivation:
sub(mul(div('IMPC_FEA_020_001', 'IMPC_FEA_060_001'), 100), mul(div('IMPC_FEA_014_001', 'IMPC_FEA_057_001'), 100))

Difference in Conditioning Post-shock and Conditioning Baseline % Freezing IMPC_FEA_107_001 | v1.0

simpleParameter


Derivation:
sub(mul(div('IMPC_FEA_099_001', 'IMPC_FEA_039_001'), 100), mul(div('IMPC_FEA_002_001', 'IMPC_FEA_033_001'), 100))

Cue: Length of time tone stimulus IMPC_FEA_108_001 | v1.0
procedureMetadata


Unit Measured: s

Options: 20, 120,