**Substance:** NDMC

**Date:** 160609

**Zebrafish DOB:** 160530 (10 dpf)

**Aim:** Dose response NDMC/ light

**Volume in wells:** 0.5 ml + 0.5 ml

**Stock NDMC solution:** 50 mM in DMSO

**Dilute to:** **200 µM** (80 µl 50 mM NDMC + 19.920 ml E3)

**100 µM** (5 ml 200 µM NDMC + 5 ml E3)

**50 µM** (2.5 ml 200 µM NDMC + 7.5 ml E3)

**100 µM NDMC:** (0.5 ml 200 µM NDMC) / **50 µM NDMC:** (0.5 ml 100 µM NDMC) / **25 µM NDMC:** (0.5 ml 50 µM NDMC)

**Control:** 0.5 ml 0.4% DMSO/E3 (0.2% in wells)

**Stock Apomorphine solution:** 10 mM in DMSO (50 µl)

**Dilute to: 500 µM** (50 µl 10 mM Apomorphine + 950 µl E3)

**25 µM** (60 µl 500 µM Apomorphine + 1140 µl E3)

Plate 1:

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Well | 1 | 2 | 3 | 4 | 5 | 6 |
| A | Control | Control | Control | Control | Control | Control |
| B | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM |
| C | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM |
| D | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM |

Plate 2:

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Well | 1 | 2 | 3 | 4 | 5 | 6 |
| E | Control | Control | Control | Control | Control | Control |
| F | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM | NDMC 25 µM |
| G | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM | NDMC 50 µM |
| H | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM | NDMC 100 µM |

File

**Add NDMC**

**Record during 13x5 min in light**

After 0 min: recording during 5 min 145944

After 5 min: recording during 5 min 150502

After 10 min: recording during 5 min 151015

After 15 min: recording during 5 min 151541

After 20 min: recording during 5 min 152100

After 25 min: recording during 5 min 152622

After 30 min: recording during 5 min 153147

After 35 min: recording during 5 min 153711

After 40 min: recording during 5 min 154231

After 45 min: recording during 5 min 154744

After 50 min: recording during 5 min 155304

After 55 min: recording during 5 min 155827

After 60 min: recording during 5 min 160347

**Add Apomorphine (0.5 µM) to all wells**

**Record during 13x5 min in total darkness**

After 0 min: recording during 5 min 161319

After 5 min: recording during 5 min 161858

After 10 min: recording during 5 min 162413

After 15 min: recording during 5 min 162919

After 20 min: recording during 5 min 163505

After 25 min: recording during 5 min 164044

After 30 min: recording during 5 min 164625

After 35 min: recording during 5 min 165208

After 40 min: recording during 5 min 165755

After 45 min: recording during 5 min 170336

After 50 min: recording during 5 min 170919

After 55 min: recording during 5 min 171504

After 60 min: recording during 5 min 172054

**Other conditions:** Cover blanket, light on

Exposure: 1310 µsec, 300 s per recording

Thermostat: 31.5°C

Water temperature: in bath: 30.5°C in wells: 28.0°C

After experiment: all larvae terminated on ice