**Substance:** PCAP931

**Date:** 160601

**Zebrafish DOB:** 160521 (11 dpf)

**Aim:** Dose response PCAP931/ light

**Volume in wells:** 1 ml + 20 µl

**Stock PCAP931 solution:** 10 mM in DMSO (50 µl)

**Dilute to:** **500 µM** (50 µl 10 mM PCAP931 + 950 µl E3)

**150 µM** (300 µl 500 µM PCAP931 + 700 µl E3)

**50 µM** (100 µl 500 µM PCAP931 + 900 µl E3)

**10 µM PCAP931:** (20 µl 500 µM PCAP931) / **3 µM PCAP931:** (20 µl 150 µM PCAP931) / **1 µM PCAP931:** (20 µl 50 µM PCAP931)

**Control:** 20 µl 5% DMSO/E3 (0.2% in wells)

**Stock Apomorphine solution:** 10 mM in E3 (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 | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM |
| C | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM |
| D | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM |

Plate 2:

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Well | 1 | 2 | 3 | 4 | 5 | 6 |
| E | Control | Control | Control | Control | Control | Control |
| F | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM | PCAP931 1 µM |
| G | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM | PCAP931 3 µM |
| H | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM | PCAP931 10 µM |

File

**Add PCAP931**

**Record during 13x5 min in light**

After 0 min: recording during 5 min 103428

After 5 min: recording during 5 min 103929

After 10 min: recording during 5 min 104431

After 15 min: recording during 5 min 104932

After 20 min: recording during 5 min 105433

After 25 min: recording during 5 min 105935

After 30 min: recording during 5 min 110437

After 35 min: recording during 5 min 110938

After 40 min: recording during 5 min 111440

After 45 min: recording during 5 min 111941

After 50 min: recording during 5 min 112443

After 55 min: recording during 5 min 112945

After 60 min: recording during 5 min 113446

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

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

After 0 min: recording during 5 min 114701

After 5 min: recording during 5 min 115202

After 10 min: recording during 5 min 115704

After 15 min: recording during 5 min 120205

After 20 min: recording during 5 min 120707

After 25 min: recording during 5 min 121209

After 30 min: recording during 5 min 121710

After 35 min: recording during 5 min 122212

After 40 min: recording during 5 min 122714

After 45 min: recording during 5 min 123215

After 50 min: recording during 5 min 123717

After 55 min: recording during 5 min 124219

After 60 min: recording during 5 min 124721

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

Exposure: 1380 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