Carrageenan-induced inflammation and thermal hyperalgesia in rats

The protocol is designed to detect the inflammatory/anti-inflammatory potential of compounds in rats and to investigate the role of histamine receptors in the acute inflammation and pain: indomethacin 20 mg/kg s.c. is the positive control.

1. Animal preparation

- House male Wistar rats (180-250 g) under controlled standard conditions (23±1°C, 55±10% humidity and a 12-h light/dark cycle). Provide food and water ad libitum.
- Weigh each rat using an animal balance accurate to 1 g.
- Divide rats to receive randomly single doses of test compounds or the vehicle.

2. Evaluation of carrageenan-induced acute paw edema

- Dissolve test compounds in the appropriate vehicle and administer by subcutaneous (s.c.), intraperitoneal (i.p.) or intragastric (i.g.) route in a volume of 1, 2 or 5 ml/kg, respectively.
- Administer vehicles or compounds under study, at increasing single doses, immediately before the induction of paw edema. Alternatively, when compound half-life is short, repeat the administration of test compound after subplantar injection of carrageenan.
- Induce paw edema by subplantar injection of 0.1 ml of carrageenan (suspended in 1% carboxymethylcellulose) into the left hind paw.
- In vehicle- or treated-rats, measure the paw volume with a plethysmometer (Basile, Comerio, Italy) immediately prior to the injection of carrageenan and thereafter at 2, 4 and 6 hours.
- For each animal, express edema as % increase in paw volume after carrageenan injection relative to the pre-injection value, considered as 100%.
- Express data as the means ± SEM from 6-8 rats per group.

3. Evaluation of thermal-induced nociception

- Dissolve and administer test compounds as reported above (step 2).
- Induce paw edema by subplantar injection of 0.1 ml of carrageenan (suspended in 1% carboxymethylcellulose) into the left hind paw.
- Determine the hyperalgesic response to thermal stimuli by using a plantar test apparatus (Ugo Basile, Comerio, Italy). Position the radiant heat under the chamber floor directly beneath the hind paw. The latency to paw withdrawal is automatically recorded by a photocell and an electronic timer.
- In order to avoid tissue damage, adjust the intensity of the radiant heat to achieve baseline latencies of 10-15 sec and a cut-off time of 30 sec.
- Discard unresponsive animals after 30 sec (cut-off time).
- For each animal, consider two subsequent applications of heating stimulus, separated by 1- to 2-min intervals, and the mean of the two measures; record paw withdrawal latencies before carrageenan administration and 2, 4, 6 hours afterwards; express responses to carrageenan as % values relative to the pre-injection value, considered as 100%.
- Express data as the means ± SEM from 6-8 rats per group.

4. Materials and reagents

- Male Wistar rats (180-250 g)
- Animal balance accurate to 1 g (e.g. Sartorius)
- Analytical balance accurate to 0.1 mg
- 50-100 ml glass beakers
- 200-1000-5000 µl pipettes (e.g. Gilson)
- 1- and 2-ml plastic syringes
- Magnetic stirrers
- Vortex mixers
- 1% carboxymethylcellulose
- Test compound solutions in the appropriate vehicle
- Prepare each compound immediately before use

References

CONTRIBUTED BY: Maristella Adami (maristella.adami@unipr.it)
LAST MODIFIED: 2012-12-10