Characterization of released exosomes from satellite glial cells under normal and inflammatory conditions

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Aims: Satellite glial cells (SGCs) are non-neuronal cells that entirely surround neurons within sensory ganglia. This unique structure allows SGC-neuron interactions. Altered cross-talk following nerve injury or inflammation is thought to contribute to the pathogenesis of chronic pain. Release of extracellular vesicles in form of exosomes has been found to play a key role in cell-cell communication. However, release of exosomes from SGCs and their potential role in modulating pain remain unknown. Hence, this study aimed at identifying and characterizing exosomes from SGCs under normal and inflammatory conditions.

Methods: Fresh primary cultures of rat trigeminal ganglia (TG) were prepared from adult male Sprague–Dawley rats. Danish Animal Inspectorate approved the study protocol. Primary SGCs were kept in culture up to 21 days and were characterized by morphology and immunohistochemistry. Cultured SGCs were monitored under normal and LPS (50 ng/mL) treatment. Collection of conditioned media was performed over time and exosomes were isolated. Particle size distribution and total protein were determined by NTA and LC–MS/MS, respectively.

Results: SGCs formed small clusters, spread outwards to areas devoid of cells but remained spindle-like in appearance with larger cell bodies. The primary cultures of SGCs were clearly GS positive with a low expression of GFAP. LPS treatment led to higher GFAP expression. Particle size distribution showed that two thirds of the particles were in the exosomal size range. Upon LPS-stimulation, four proteins (histone H2B, ubiquitin–60S ribosomal, myosin–9, elongation factor 1-alpha) were found exclusively expressed compared to normal treated SGCs.

Conclusions: For the first time it was demonstrated that SGCs shed extracellular vesicles in exosomal size range. Myosin-9 was identified as a possible novel marker of SGCs activation under inflammatory conditions. This protein plays a role in cell-cell adhesion and possibly contributes to SGC–SGC cross-talk upon inflammation which may consequently influence the excitability of nearby neurons.

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Cell-based platform for studying trigeminal satellite glial cells under normal and inflammatory conditions

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Aims: Satellite glial cells (SGCs) in sensory ganglia contribute to the pathogenesis of chronic pain. In vitro, providing enough fresh primary SGCs poses some practical limitations; hence, frozen stocks of primary cells for culture could be an attractive alternative for cell-based studies or drug screening. This study was designed to investigate the morphology and marker expression of frozen and freshly isolated trigeminal SGCs under normal and inflammatory conditions.

Methods: SGCs from trigeminal ganglia of three male Sprague–Dawley rats and three frozen (sub cultured and passaged) batches of stored primary SGCs were cultured. Their morphology was observed by phase microscopy and the phenotype was characterized by immunocytochemistry of glutamine synthetase (GS) and glial fibrillary acidic protein (GFAP). Lipopolysaccharide (LPS) was used to simulate a state of neurogenic inflammation in vivo. A pilot test was performed to determine the optimal concentration of LPS to activate SGCs based on GFAP expression. A long-term activation of the SGCs with 50 ng/mL LPS was chosen for further characterization.

Results: The fresh and frozen primary SGCs elicited similar phenotypes based on GS marker expression. However, frozen primary SGCs differed in terms of size and morphology. GFAP was constantly expressed in frozen primary SGCs regardless of LPS stimulation. Activation of primary fresh SGCs with LPS spread the GFAP expression from around the cell body throughout the longer processes and activation was only seen in the LPS treatment.

Conclusions: The phenotypic marker, GS was independent of culture conditions. There was no difference in upregulation of GFAP in thawed SGCs regardless of LPS stimulation. This indicates that freeze-thawing might activate SGCs and therefore frozen and passaged cells cannot be suitable for use in cell-based models for inflammation. Fresh primary cells are therefore optimal for studying SGCs under normal and inflammatory conditions.

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Tramadol in postoperative pain – 1 mg/ml IV gave no pain reduction but more side effects in third molar surgery

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Aims: Does pre-emptive single dose intravenous tramadol produce a safe and effective postoperative analgesia?

Methods: Randomized, placebo controlled, single blinded clinical trial of pre-emptive intravenous tramadol 1 mg/kg in combination with IV midazolam in patients with dental fear. A “Pain diary” evaluates the efficacy. The safety is evaluated perioperatively monitoring (SpO2 and BP).

Results: Pain scored by VAS showed no differences between the groups. It took longer time to first rescue pill in tramadol vs. control group (157 vs. 110 min, p = 0.049). Desaturation (SpO2 < 90%) was more commonly found in tramadol vs. placebo and control
Neuroplasticity and Pain (CNAP), SMI

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Exteroceptive system using laser heat and mechanical touch stimulations. NRS increased both with line length and distance between the stimulations. For the mechanical stimulation for both line and 2-point stimulation (34.5 mm). NRS was significantly higher for laser stimulations than for the mechanical stimulation (67.9 mm) was higher than for the mechanical stimulation. Distal and proximal directed stimuli was 68.5 mm and 70.2 mm for distal and proximal directed stimuli, respectively.

Methods: A series of skin locations on the right arm using one cold (−20 °C) and one warm thermode (−40 °C). The two stimulus locations had identical physical distance on the skin. However, the distance between the cold and heat projection signals in the spinal cord varied across three conditions. Cold and warm inputs were delivered (1) within the same dermatome (e.g., C5–C6); (2) across the dermatome boundary of two adjacent spinal segments (e.g., C5–C6); (3) across the dermatome boundary of two non-adjacent spinal segments (e.g., C5–T1). In two experiments, we obtained an estimate of the strength of the TGI by asking 32 healthy participants to complete a temperature matching task.

Results: Participants overestimated the actual average temperature of the two thermodes (Exp. 1) and the cold temperature of one of the two thermodes (Exp. 2). However, this effect was significantly larger when cold and heat stimulations were delivered within the same dermatome (+6.57 ± 3.99 °C and +9.88 ± 5.60 °C) or between dermatomes projecting to adjacent spinal segments (+6.26 ± 4.44 °C and +9.48 ± 5.83 °C), compared to when cold and heat stimulations projected to non-adjacent spinal segments (+3.46 ± 4.46 °C and +4.80 ± 3.21 °C).

Conclusions: These results demonstrate that the strength of the illusion is modulated by the segmental distance between cold and heat spinal signals, and show that the perceived quality and intensity of thermal stimuli depends upon low-level spatial summation mechanisms in the spinal cord.

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Effect of cocoa on endorphin levels and craniofacial muscle sensitivity in healthy individuals


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Aims: Migraine headache is a recurrent incapacitating neurovascular disorder. A combination of events occurring within the trigeminovascular system has been suggested to underlie migraine pathogenesis, including release of inflammatory neuropeptides with subsequent effects on meningeal vessels and sensory transmission. Increased craniofacial muscle tenderness is also present in patients with migraine and a lower β-endorphin concentration has been observed in these patients. Several external factors have been identified, which can modulate migraine. One such factor is cocoa, though controversies still exist whether this substance exerts pro-algesic or analgesic effects. The present study evaluated

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The encoding of the thermal grill illusion in the human spinal cord

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Aims: The spatial alternation of innocuous cold and warm stimuli on the skin can paradoxically provoke a hot, burning sensation, known as the thermal-grill illusion (TGI). Whether the TGI depends on spinal or supraspinal integration mechanisms is still debated. To assess whether the TGI can be accounted for by integration of cold and warm afferent signals in the spinal cord, we leveraged anatomical knowledge on the spatial arrangement of dermatomes and spinal segmental projections.

Methods: We stimulated a series of skin locations on the right arm using one cold (−20 °C) and one warm thermode (−40 °C). The two stimulus locations had identical physical distance on the skin. However, the distance between the cold and heat projection signals in the spinal cord varied across three conditions. Cold and warm inputs were delivered (1) within the same dermatome (e.g., C5–C6); (2) across the dermatome boundary of two adjacent spinal segments (e.g., C5–C6); (3) across the dermatome boundary of two non-adjacent spinal segments (e.g., C5–T1). In two experiments, we obtained an estimate of the strength of the TGI by asking 32 healthy participants to complete a temperature matching task.

Results: Participants overestimated the actual average temperature of the two thermodes (Exp. 1) and the cold temperature of one of the two thermodes (Exp. 2). However, this effect was significantly larger when cold and heat stimulations were delivered within the same dermatome (+6.57 ± 3.99 °C and +9.88 ± 5.60 °C) or between dermatomes projecting to adjacent spinal segments (+6.26 ± 4.44 °C and +9.48 ± 5.83 °C), compared to when cold and heat stimulations projected to non-adjacent spinal segments (+3.46 ± 4.46 °C and +4.80 ± 3.21 °C).

Conclusions: These results demonstrate that the strength of the illusion is modulated by the segmental distance between cold and heat spinal signals, and show that the perceived quality and intensity of thermal stimuli depends upon low-level spatial summation mechanisms in the spinal cord.

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