Morphine-induced elevation of glymphatic flow and inflammatory microparticles in mice

S.R. Thom, A. Bhat

Repetitive dosing of morphine in mice to induce anti-nociceptive tolerance increases by 10-fold microparticles (MPs, 0.1-1.0 μm extracellular vesicles) in deep cervical lymph nodes (DCLN) draining brain glymphatics[1]. These MPs express proteins specific to microglia, astrocytes, neurons and oligodendrocytes. Similar elevations of DCLN MPs in response to other noxious stimuli reflect neuroinflammation and an increase of glymphatic flow[2]. When these MPs enter the systemic circulation, they trigger neutrophil activation, elevations of blood-borne MPs and prompt further neuroinflammation due to disruption of the blood-brain barrier[3, 4, 5, 6, 7]. As patients entering treatment for opioid use disorder exhibit similar MPs elevations in blood as do tolerant mice, we decided to investigate whether glymphatic function was perturbed by morphine tolerance using a recently developed MRI technique[1, 2].

Methods: Control mice and those rendered tolerant to morphine following published methods were imaged in the University Bruker BioSpec 70/30USR Avance III 7 Tesla horizontal bore MR scanner following published methods[2]. Similarly, DCLN and blood-borne MPs were obtained and analyzed by flow cytometry following published methods[1, 2].

Results: Gadolinium contrast signal intensity, an index of glymphatic flow[2], was significantly higher in brains of morphine tolerant mice. Image results were similar when 60,000 blood MPs, but not MPs from control mice, were injected IV into naïve mice. Elevations of MPs in DLCN and blood were consistent with prior publications, as were increases in inflammatory blood-borne MPs expressing filamentous (F-) actin on the membrane surface. F-actin expressing MPs are known to be lysed by plasma gelsolin[1, 2, 8]. Plasma gelsolin was significantly lower in tolerant mice.

Conclusions: These data provide insight into mechanisms associated with development and dissemination of neuroinflammation after morphine administration. Inflammatory responses outside the CNS appear to play a role in opiate tolerance, but mechanisms are unknown[9, 10]. CNS-derived MPs cause peripheral vascular disorders once liberated to the blood stream, and MPs generated systemically can exacerbate traumatic and other causes of brain injury[1, 2, 5, 8].

References:

1. D. Ruhela et al. Brain, Behavior and Immunity 87, 465-472 (2020).
2. S. R. Thom et al. J Neurophysiol 129, 662-671 (2023).
3. J. Xu et al. Toxicol Appl Pharmacol 273, 410-417 (2013).
4. M. Yang et al. J Appl Physiol 112, 204-211 (2012).
5. M. Yang et al. J Appl Physiol 115, 1481-1486 (2013).
6. A. Kumar et al. J Neuroinflammation 14, 47 (2017).
7. S. R. Thom, et al. J Appl Physiol (1985) 123, 297-302 (2017).
8. V. M. Bhopale et al. J Appl Physiol. 130, 1604-1613 (2021).
9. Y. A. Kolesnikov, et al. J Pharmacol Exp Ther 279, 502-506 (1996).
10. R. R. Ji, et al. Nat Rev Drug Discov 13, 533-548 (2014).

Funding was provided by NIH grant NS122855 and Office of Naval Research N00014-22-1-2818.