The Development of a High-Speed Rat Pain Ethogram

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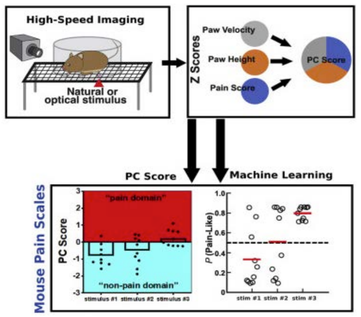
Background

- Pain is the largest public health crisis in the United States. Up to 635 billion dollars are spent each year in costs associated for chronic pain patients,[1] affecting more than 100 million patients; more than heart disease, cancer, and diabetes combined[2]. A major issue that has risen from this epidemic is the lack of treatment options available, with the use of a common chronic pain treatment of opioids becoming a dangerous treatment. Over 91 people die every day from an opioid overdose[3], with almost half of these deaths involving a prescription opioid[3], there must be a safer way to treat chronic pain. However, in order to find a safer alternative, there must be a better way to assess pain in order to understand if a treatment works.

- In humans, a pain scale of 1-10 is normally used, but in mice there are currently 3 different behavior assays: reflex-based assays with paw and or tail, operant-based assays, or spontaneous pain behaviors. While most preclinical studies use these reflex–based assays[4], it is not the best indicator of pain to have a mouse responding to a stimuli at an increased frequency. Only 11% of pain therapeutics entering phase 1 become approved by the FDA[5], showing that the current assays to measure pain are not accurate enough for rodents.

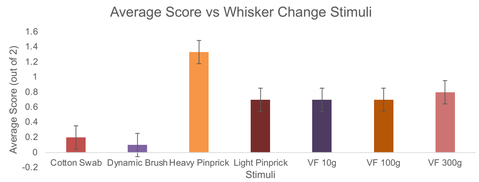
- In 2019, Dr. Fried published a paper detailing a way to objectively assess pain sensation in mice by using high-speed videography to capture sub-second behavioral features following hind paw stimulation involving both noxious and innocuous stimuli[6]. The result of this study was the creation of a "mouse pain scale" that allows users to assess pain sensation in a graded manner for each withdrawl[6].

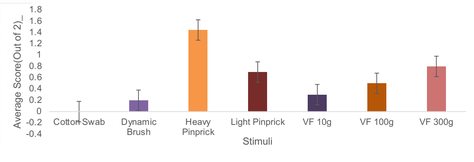
- While working in Dr. Fried's lab during the summer, I helped score various videos using this "mouse pain scale" and I noticed something interesting. I felt that more behaviors could be tested in addition to the ones currently being used by Dr. Fried's lab (height, velocity, shaking, guarding, looking first, orbital tightening, jumping). After reviewing the rat and mouse grimace scale created by Jeffrey Mogil's lab[7], I wanted to see if this could be applied in addition to the current pain behaviors being looked at by Dr. Fried's lab in capturing sub-second behavioral features.

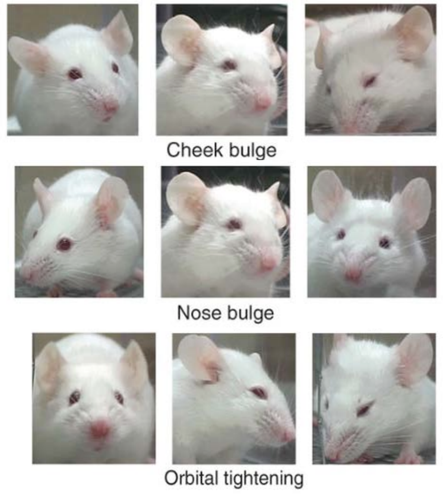


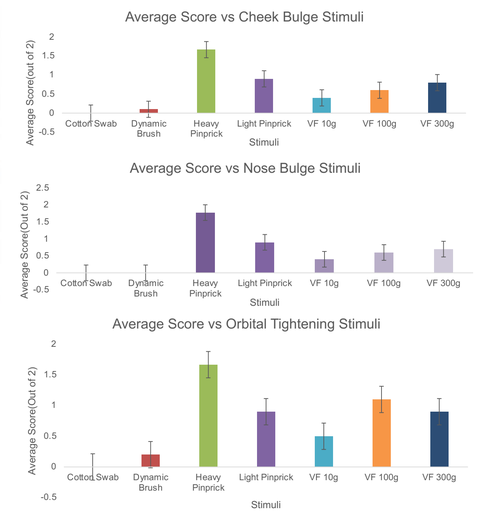
Methodology and Analysis

- The Rat and Mouse Grimace scale studies 5 different behaviors: Whisker Change, Ear Position, Cheek Bulge, Nose Bulge, and Orbital Tightening[7]. I used the Rat Grimace Scale (RGS) to look at sample videos studying saline sired rats that undergo various stimuli. The stimuli studied included Cotton Swab, Dynamic Brush, Heavy Pinprick, Light Pinprick, Von Frey 10g, Von Frey 100g, and Von Frey 300g. I scored each video using the RGS on a scale from 0(not present) to 2 (Severely present)[7] and looked at the difference in score among each stimuli and the behaviors studied.









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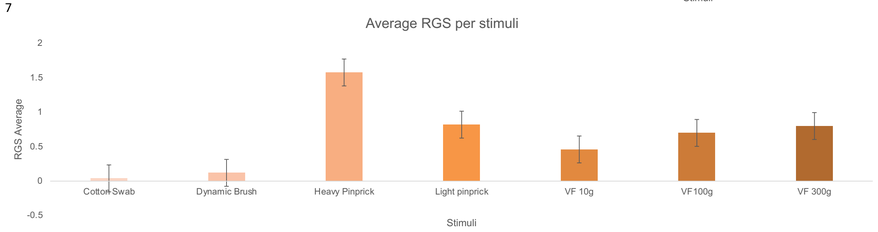

Discussion

- After scoring the videos I found that in the RGS averages that it can differentiate the various stimuli between painful and nonpainful movements. The use of the RGS could be a beneficial addition to the current scale used for sub-second imaging by Dr. Fried's lab.

- In addition, the idea of using grimace to look at spontaneous, sub-second pain response or evoked assays have not been looked at before and could be a useful tool for the future after further study. Another point to look at is if the Mouse Grimace Scale (MGS) could work for sub second pain response in mice as well.

- While the data is promising, there are several points of concern. The data is not blinded so any pre-determined bias (heavy pinprick will hurt more than cotton swab) may show without realization of it. Also, the skill level in scoring these videos is rather juvenile, by only having the actual scale as a reference several features may be confused with others. More training will be needed to accurately score these videos, but the initial testing is promising.

Conclusions and Directions for Future Research

- In conclusion, the results of this analysis indicates a possible use of the RGS in sub-second imaging of noxious stimuli. There is great potential in the RGS/MGS for future use of using withdrawal reflex assays to assess pain sensation[6], but more testing is needed to determine accuracy of data.

- In the future, I hope to be able to fine tune scoring abilities to be able to properly run accurate tests in a blinded setting that will allow for an unbiased look at the stimuli. If proven true, this could be a vital tool in looking at spontaneous pain response on a sub second level.

References and Acknowledgments

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