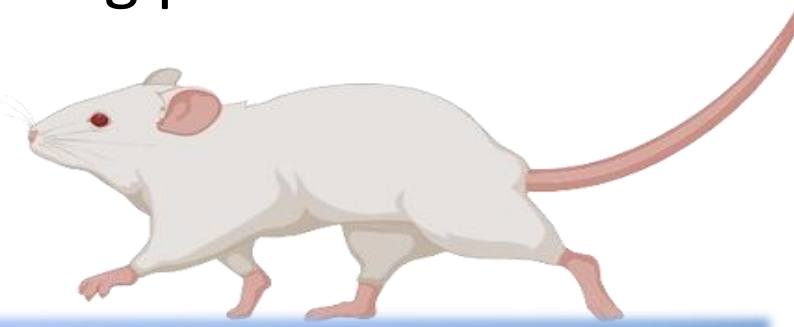


Introduction

Gait Analysis is a robust, non-invasive method for assessing functional recovery after musculoskeletal injury. By evaluating spatiotemporal parameters - such as stride length, step length, paw area ratio, and paw angle - researcher can obtain objective data on weight-bearing symmetry, coordination, and joint stability throughout the healing process.



But do improvements in gait truly reflect tissue regeneration, or simply pain reduction?

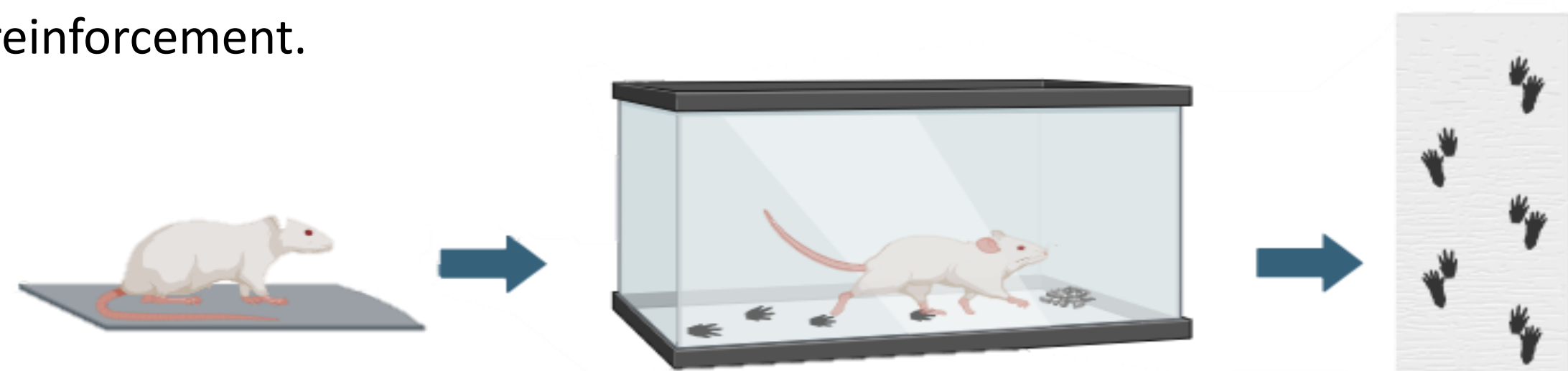
This study addresses this challenge by distinguishing pain-driven functional improvements from regeneration using a time-sensitive gait analysis framework.

Method

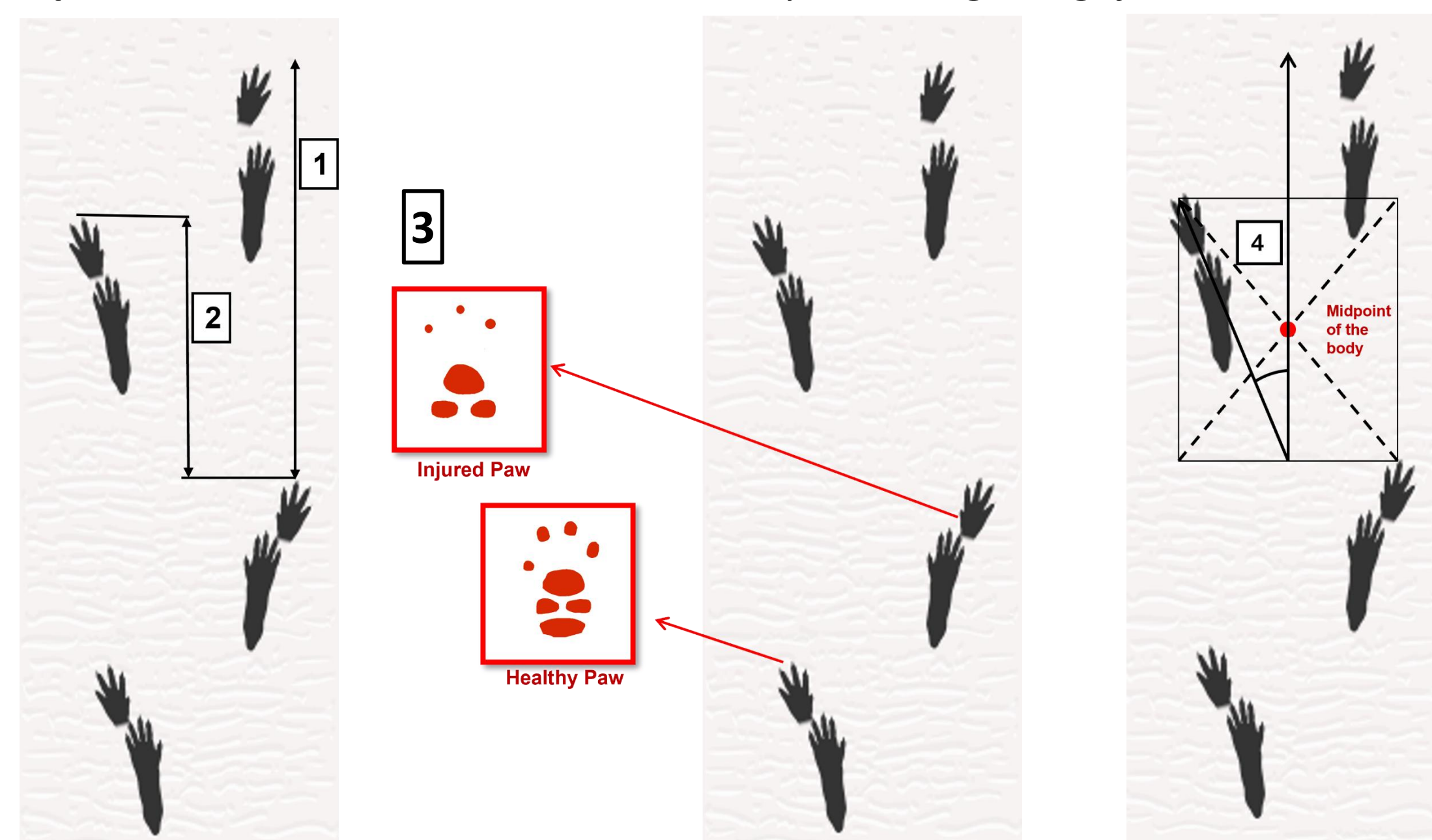
Treatment Groups:

1. **Healthy** (Positive Control)
2. **Suture** (Negative Control)
3. **Platelet Derived Growth Factor (PDGF) Adhesive** (Regenerative Treatment)
4. **Cromolyn** (Mast Cell Stabilizer – Anti-Inflammatory Treatment)

To collect gait data, the animals were placed on an ink pad and encouraged to walk across a custom-built gait analysis platform, with treats used as positive reinforcement.



Gait parameters were measured and analyzed using imagej.



1. **Stride Length:** Injured paw to the next injured paw. Represents the functional range of motion.
2. **Step Length:** Injured paw to healthy paw. Reflects limb coordination.
3. **Paw Area Ratio:** Injured paw to Healthy paw ratio. Assesses how much pressure the rat is putting on the injured paw.
4. **Paw Angle Variation:** Difference in the angles at which the rat places its injured paw and healthy paw. Indicates compensatory behavior.

Results

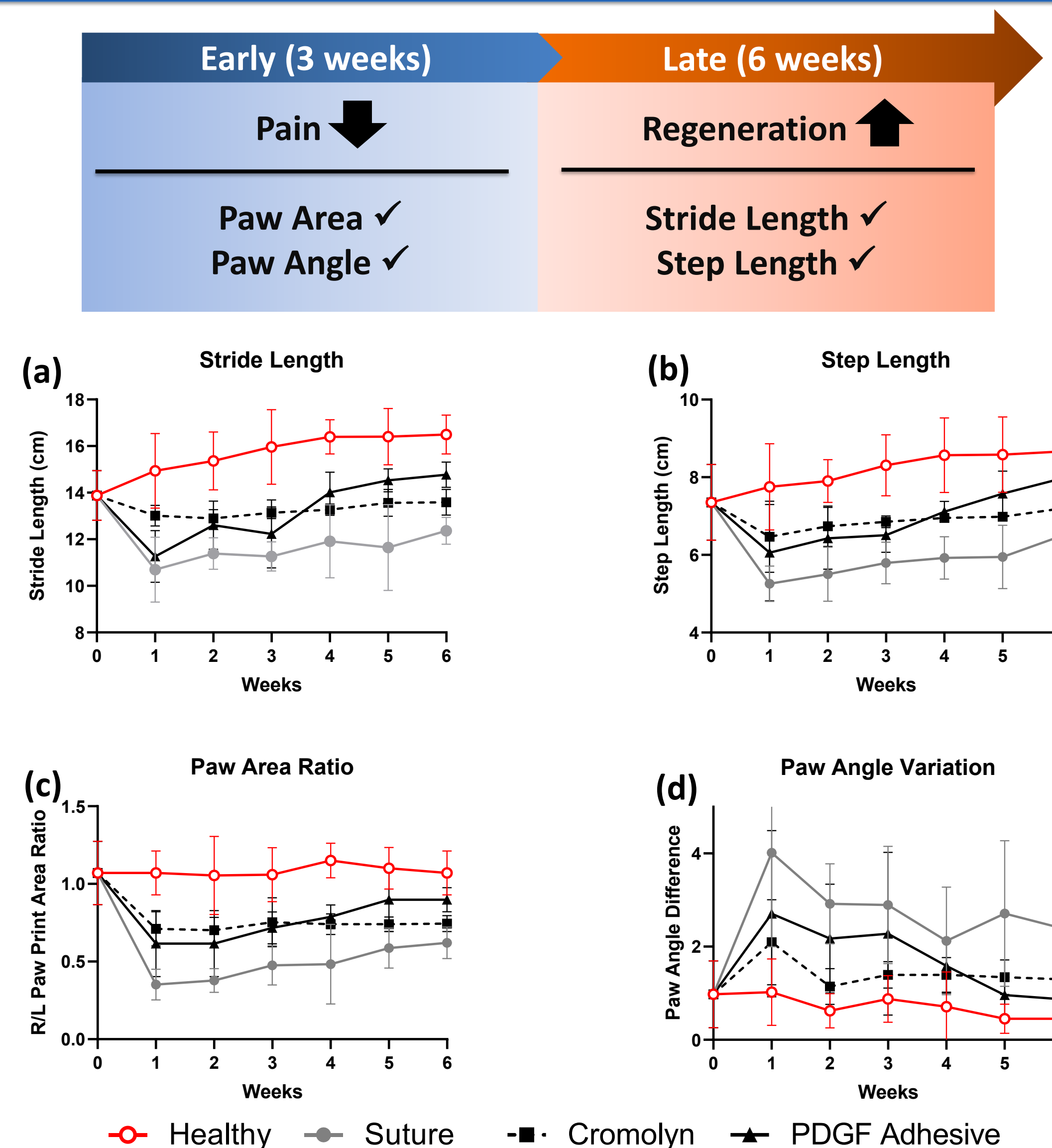


Figure 1: Early improvements reflect pain reduction, best captured by Paw Area Ratio (c) and Paw Angle Variation (d), while later improvements reflect regeneration, best captured by Stride Length (a) and Step Length (b).

Paw Area Ratio and Paw Angle show strong correlation with pain related biological markers.

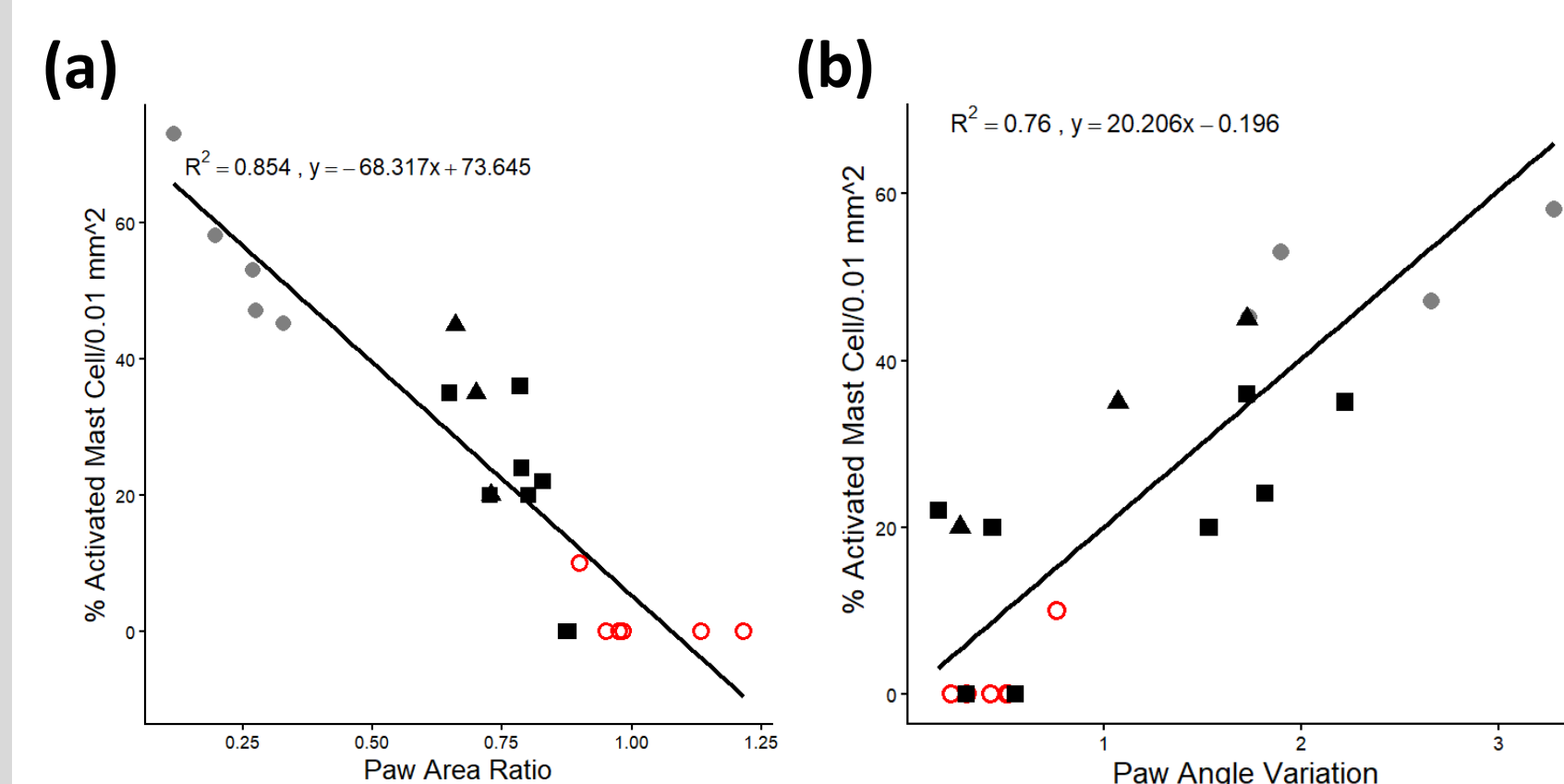


Figure 3: Paw Area Ratio (a) and Paw Angle Variation (b) demonstrate the strongest associations with inflammatory markers.

Stride Length and Step Length show strong correlation with regeneration related biological markers.

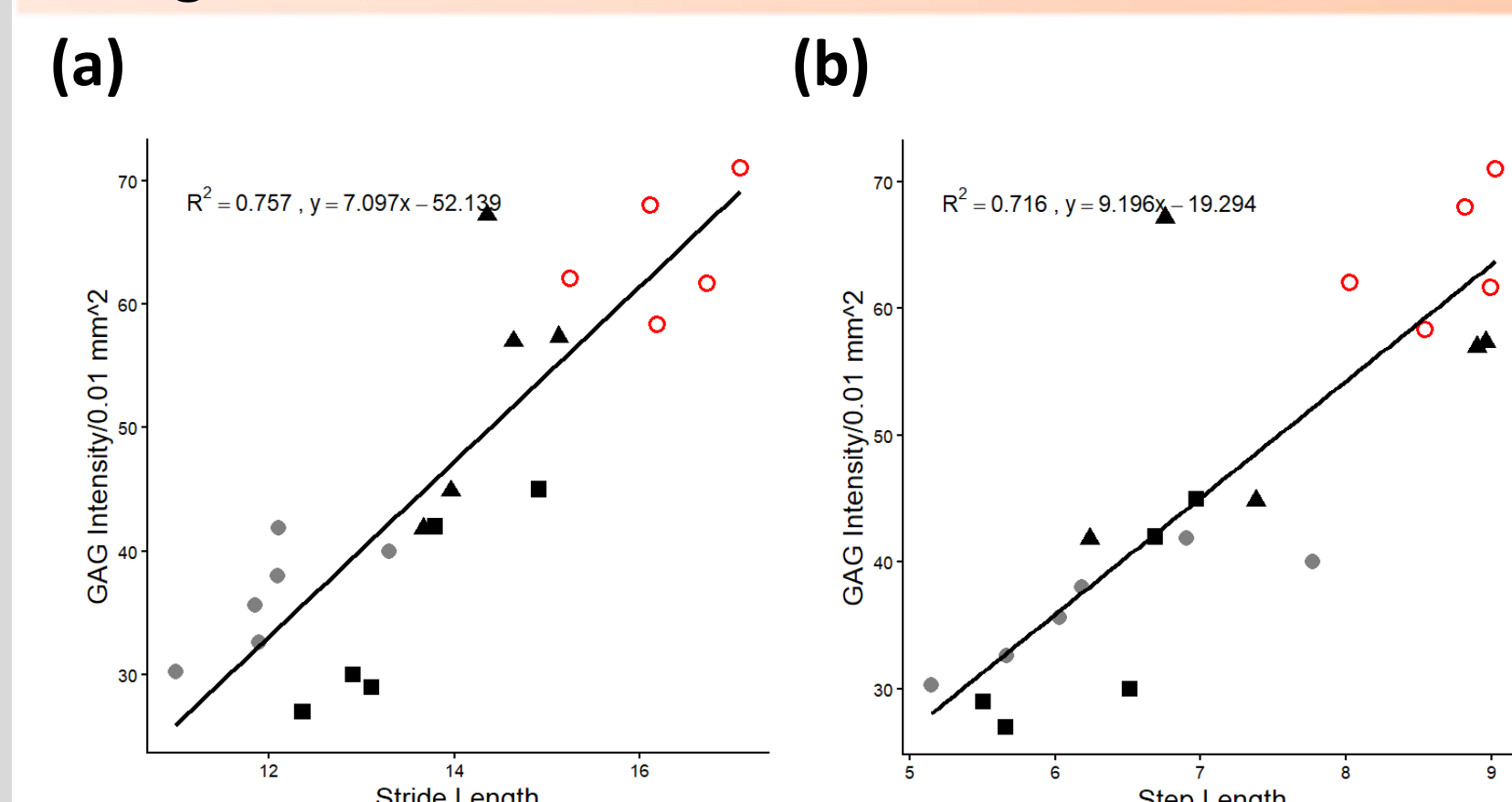


Figure 3: Stride Length (a) and Step Length (b) show the strongest correlations with regenerative markers.

Gait Scoring System

At 3 weeks, inflammation score shows pain relief (Cromolyn improves most).

At 6 weeks, regeneration score shows true healing (PDGF Adhesive performs best).

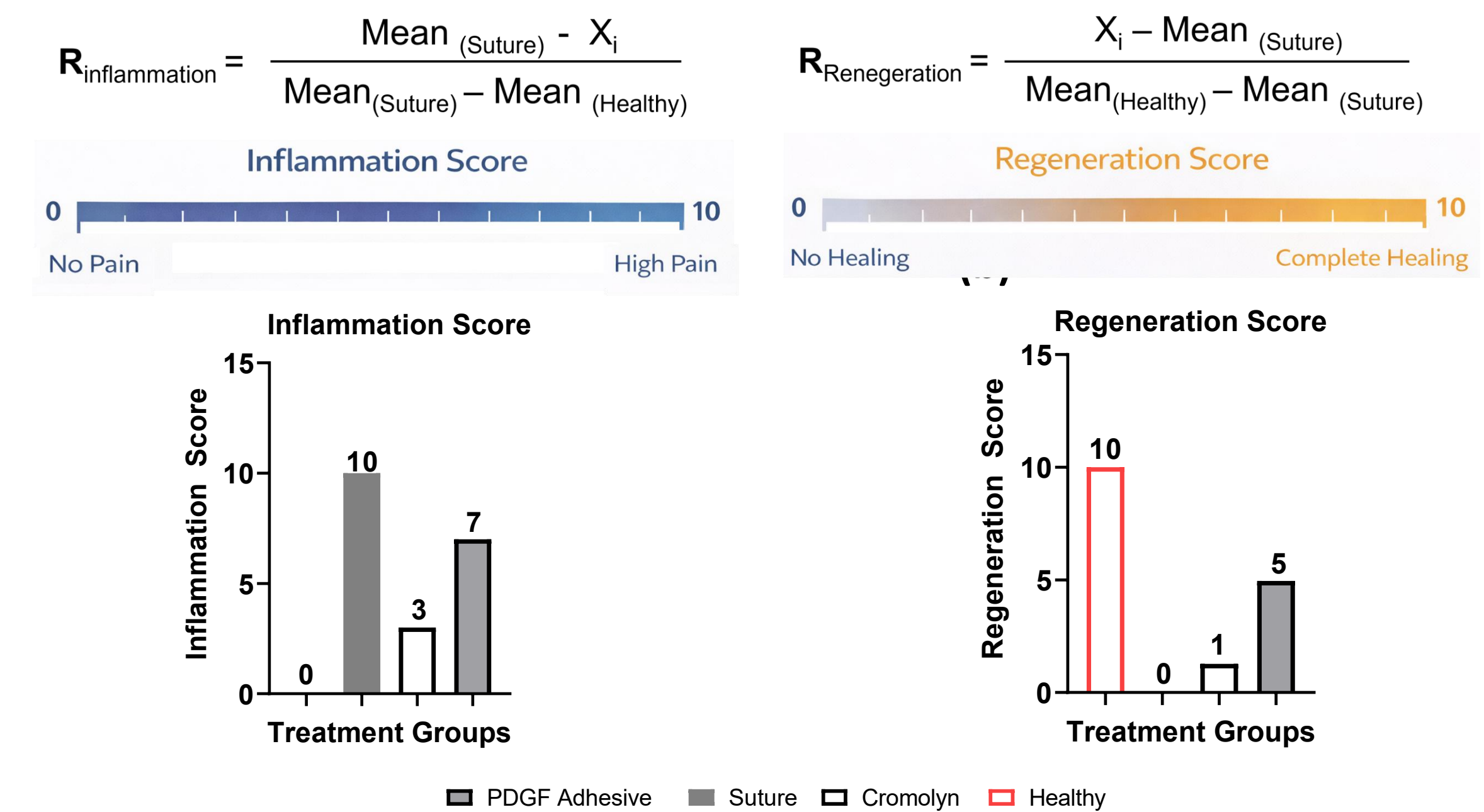


Figure 4: (a) Cromolyn (Pain Reduction) treatment has the highest inflammation score. (b) PDGF Adhesive (Healing) Treatment has the highest regeneration score.

Summary of Findings

- ✓ Cromolyn improves early function via pain reduction.
- ✓ PDGF Adhesive promotes long-term recovery via tissue regeneration/healing.
- ✓ Early (3 weeks) improvements in gait parameters reflect reduced pain, not healing.
- ✓ Later (6 weeks) improvements in gait parameters reflect tissue regeneration (healing).
- ✓ Paw-based metrics, paw area ratio and paw angle variation, capture the response to pain best at 3 weeks.
- ✓ Stride and step length reflect regeneration/healing better at 6 weeks.

Future Implications

- Increase sample size.
- Study the effects of sex on gait parameters.
- Analyze 3-Dimensional gait parameters.

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