

Propionate Alters Macrophage Morphology and Migration

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Background

- **Macrophages** are professional phagocytic immune cells. Macrophages can be either classically activated to a pro-inflammatory phenotype to defend against pathogens or alternatively activated to an anti-inflammatory phenotype to aid in healing.
- **Propionate** is a short chain fatty acid produced by our gut microbes. Propionate is known to have anti-inflammatory and antibacterial effects in many cell types, including macrophages.

Main Research Questions

- How does propionate alter morphology of macrophages that have already been classically activated?
- How does exposure to varying levels of propionate alter macrophage phenotype?

Research Methods

- **Length to Width Ratio:** RAW264.7 cells were grown overnight with or without LPS & IFN γ for activation. Overnight media was replaced with fresh media with or without propionate for 3 hours. Images were taken with an inverted phase contrast microscope. Length-to-width ratios were measured using Image J.
- **Nitrite Concentration:** RAW264.7 cells were grown as previously described. Propionate was added either overnight or for 3 hours. Nitrite concentration was measured using a Greiss Reagent.
- **Migration:** RAW264.7 cells were seeded with or without LPS & IFN γ and propionate into one side of a microfluidic device and incubated overnight. Channel entry was quantified.

Results

Three-hour propionate treatment as low as 1 mM significantly increased length-to-width ratios of naïve and classically activated macrophages

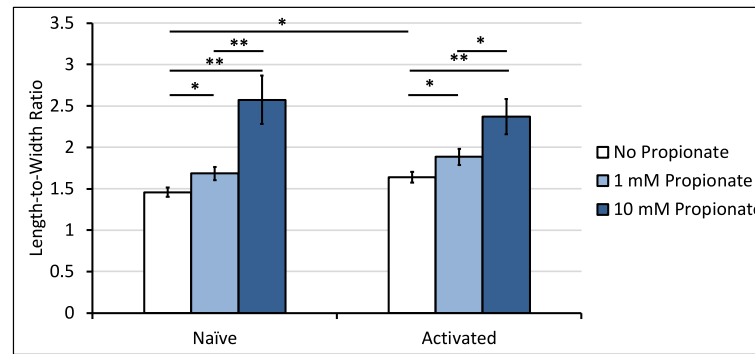


Figure 1. Length-to-width ratios of RAW264.7 macrophages.

Representative images of microfluidic devices used to assess macrophage migration

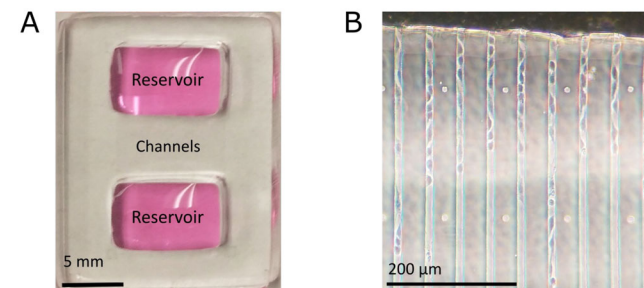


Figure 2. Reservoirs filled with media, one containing cells (LEFT). Microscopic image of cells in the channels (RIGHT). Image from DOI 10.3389/fmicb.2021.721801

Migration of naïve and classically activated macrophages was significantly enhanced by 10 mM propionate treatment

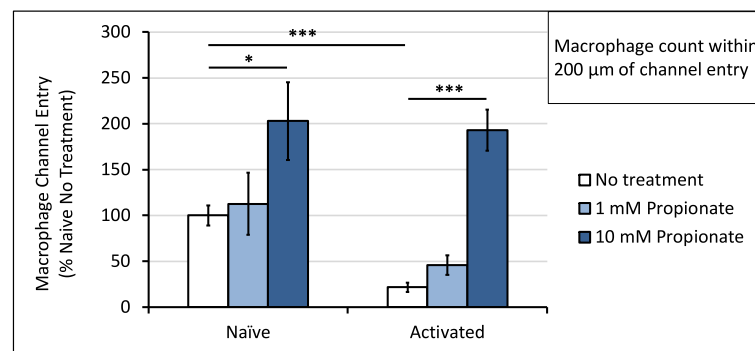


Figure 3. Percent average macrophage channel entry (naïve no treatment set at 100%).

Results

Three-hour propionate treatment is not sufficient to influence nitric oxide production by macrophages

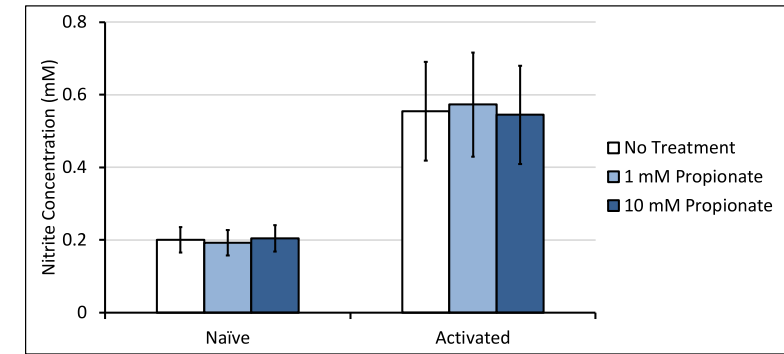


Figure 4. Nitrite concentration of RAW264.7 macrophages treated with propionate for 3 hours.

Overnight propionate treatment as low as 1 mM significantly reduces nitric oxide production by classically activated macrophages

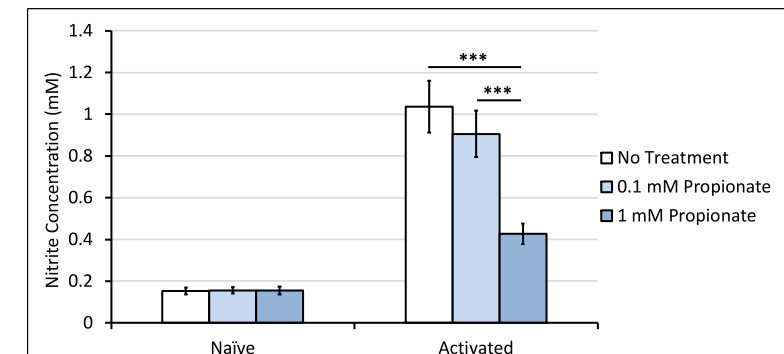


Figure 5. Nitrite concentration of RAW264.7 macrophages treated with propionate overnight.

Conclusions

- Propionate treatment of both naïve and classically activated macrophages results in elongated cell shape and increased motility. → Propionate can potentially promote macrophage migration to infection sites.
- Overnight, but not short (3 hour), propionate treatment of classically activated macrophages reduces nitric oxide production. → Propionate can potentially reduce inflammatory responses by macrophages.



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