Investigating the anti-inflammatory effect of curcumin and piperine in preventing biomaterial rejection *in vitro*

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Abstract

The inflammatory response to foreign biomaterials is a significant concern in various medical procedures. Chemokines and cytokines recruit monocyte-derived macrophages to the site of the implanted biomaterial, at which they adhere to and attempt isolate it from the body. This typically results in device failure and often requires replacement later in life. Natural products have garnered attention recently as potential therapies that could have anti-inflammatory properties. Curcumin, a yellow hydrophobic polyphenol extract from turmeric, has been shown to have anti-inflammatory characteristics. However, it has a poor bioavailability as it is quickly metabolized upon consumption. Piperine, an alkaloid component in black pepper, has been shown to increase the bioavailability of other nutrients. Through *in vitro* THP-1 cell adhesion assays, our data show that curcumin alone and in combination with piperine inhibit inflammatory cell attachment to polyurethane films, a common biomaterial. Ongoing experiments with an *ex vivo* Chandler Loop apparatus are being conducted to confirm the *in vitro* findings. These results indicate that curcumin and piperine have potential anti-inflammatory properties and warrant further investigation as potential therapies for biomaterial rejection.

Introduction

•The foreign body reaction relates to the implementation of biomaterials and their function when inside the body. Monocyte-derived macrophages respond in the chronic inflammatory pathway in the presence of biomaterials (1). Chemokines and cytokines recruit the macrophages to the site of the implant, and the macrophages adhere and isolate the foreign material.
•Inhibiting this response may reduce the inflammatory response while also enhancing the biocompatibility of the material in the body (1).
•Curcumin, the active molecule present in Curcuma longa, is known for its antioxidant, anti-inflammatory, and antitumor properties (2). Curcumin has been shown to suppress symptoms of certain disorders or diseases, such as rheumatoid arthritis, multiple sclerosis, and Alzheimer's disease (3). However, it has a low bioavailability due to it being rapidly metabolized in the liver and intestinal wall (3).
•Piperine is the major component of black pepper. Although its primary use is for culinary reasons, it also has fundamental uses in its function as a bioenhancer. Research has shown that piperine increases the bioavailability of curcumin when orally administered (4).
•Current research has focused on using THP-1 cells, an established model of monocyte-derived macrophages that models the body's response to biomaterials *in vitro* (5).

Methods

• Commercially available 10 mM curcumin and 10 mM piperine were dissolved in 95% ethanol
•THP-1 cells were cultured using RPMI-1640, 5% FBS, 1X Penicillin/Streptomycin, and 0.05 mM BME
• Cells were incubated at 37 °C and 5% CO_2.
•THP-1 cells were differentiated with 160 nM PMA for THP-1 adhesion assays using polyurethane films, a common polymer used in medical devices.
• THP-1 cells were treated with 1.425% ethanol (control), 50 µM curcumin, 50 µM piperine, and 50 µM curcumin + 50 µM piperine combined.
• Nuclei were stained with 5 µM SYTO 60 and imaged using fluorescence microscopy under 400X total magnification.

Results

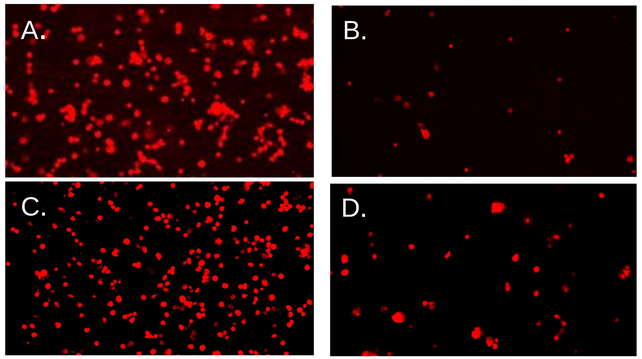

Fig. 1 Representative THP-1 Adhesion Assay Images: THP-1 cells were cultured using complete media containing various treatments: **A.** control (1.425% ethanol), **B.** 50 µM curcumin, **C.** 50 µM piperine, **D.** 50 µM curcumin + 50 µM piperine together. Nuclei were stained with SYTO 60 and imaged using fluorescence microscopy under 400X total magnification.

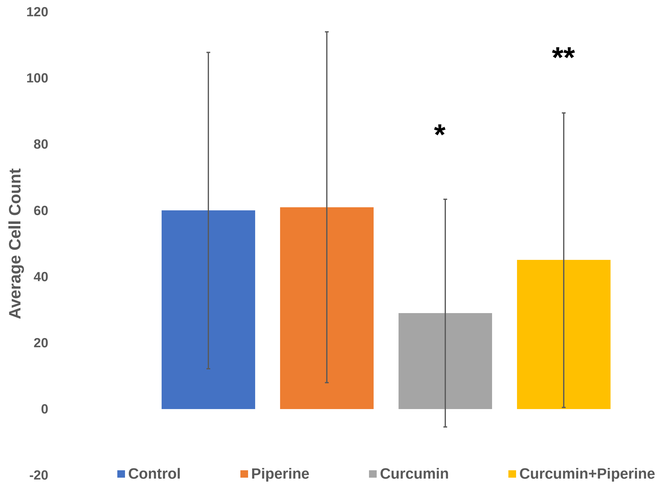

Fig. 2 Quantification of THP-1 Adhesion Assays: The polyurethane films were imaged in nine random fields of view and the adhered THP-1 cells were counted in each view. The number of counts for each treatment was 144 throughout eight trials, with the data being analyzed using an ANOVA test with Tukey post-hoc. Statistical significance relative to the control: * $p < 0.01$ ** $p < 0.05$.

Discussion

•The THP-1 cells were not negatively impacted by the ethanol control as seen in the high number of adhered cells to the polyurethane films.
•The 50 µM curcumin alone treatment decreased the number of adhered cells compared to control, suggesting that curcumin itself had an anti-inflammatory effect on the cells.
•The 50 µM piperine alone treatment did not significantly change the number of adhered ells when compared to the control, suggesting that on its own it did not have a significant anti-inflammatory effect.
•The 50 µM curcumin + 50 µM piperine treatment significantly decreased the number of adhered cells compared to control, suggesting that piperine and curcumin together exhibit anti-inflammatory properties. However, the decrease observed was to an extent less than curcumin alone, suggesting the lack of a synergistic effect.

Future Work

•Future experiments will be done using a Chandler Loop to mimic normal blood flow in the body. The THP-1 cells will be placed into tubing to be spun and examined the same way as in this experiment.

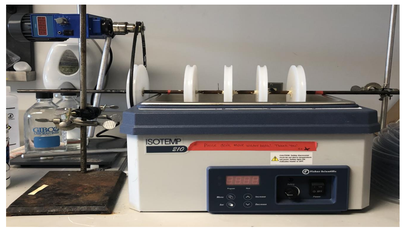

Fig. 3 Chandler Loop Apparatus: Photograph showing the design of the Chandler Loop Apparatus. This will be used to mimic physiological conditions of cell attachment to a biomaterial.

References

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