The role of saturated and polyunsaturated fatty acids in microglia modulation

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Introduction

Obesity is considered a worldwide pandemic, being responsible for the development of non-communicable diseases, presenting severe health and economical consequences. Several investigations suggested that brain plays a major role in obesity development: saturated fatty acids (SFAs) bind to a specific receptor (TLR4) in the hypothalamus, triggering inflammatory processes resulting in impaired brain processes. Recently, associations between the current western diet, associated with high-fat (mostly SFAs) and fructose consumption, and obesity have been suggested.

In contrast, polyunsaturated fatty acids (PUFAs), such as omega-3 from fish oils were reported as having anti-inflammatory properties in brain and revert the SFAs-induced obesity effects. Such processes involve the inhibition of the NFkB pathway. In addition, some conjugated fatty acids isomers have shown to reduce body fat mass in animal models and possess anti-inflammatory properties.

Objectives

Considering the beneficial role of conjugated linoleic acid (CLA) and conjugated linolenic acid isomers (CLNA) in obesity, namely their anti-inflammatory properties, we hypothesized that they may present similar properties as omega-3 fatty acids in hypothalamus. Since microglia is the main cellular component of the brain innate immune system and a key player in both regulation and protection of the central nervous system homeostasis, we selected a human microglia cell line as our model. Thus, this work aimed to analyze the modulatory potential in a human microglia cell line of a solution mimicking the western pattern diet (palmitic acid, a SFA, and fructose) and the preventive role of different PUFAs (Omega-3 – EPA and DHA-, CLA and CLNA isomers), specifically targeting the NFkB pathway and other important processes such as reactive oxygen species (ROS) production and LynSrc activation.

Methods

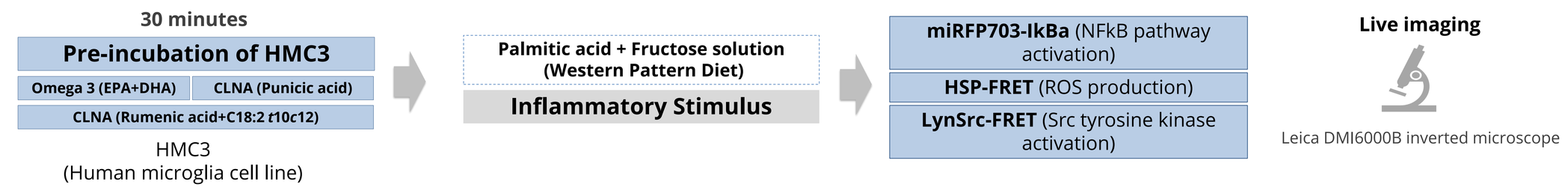

Figure 1. Experimental design schematic representation.

Results

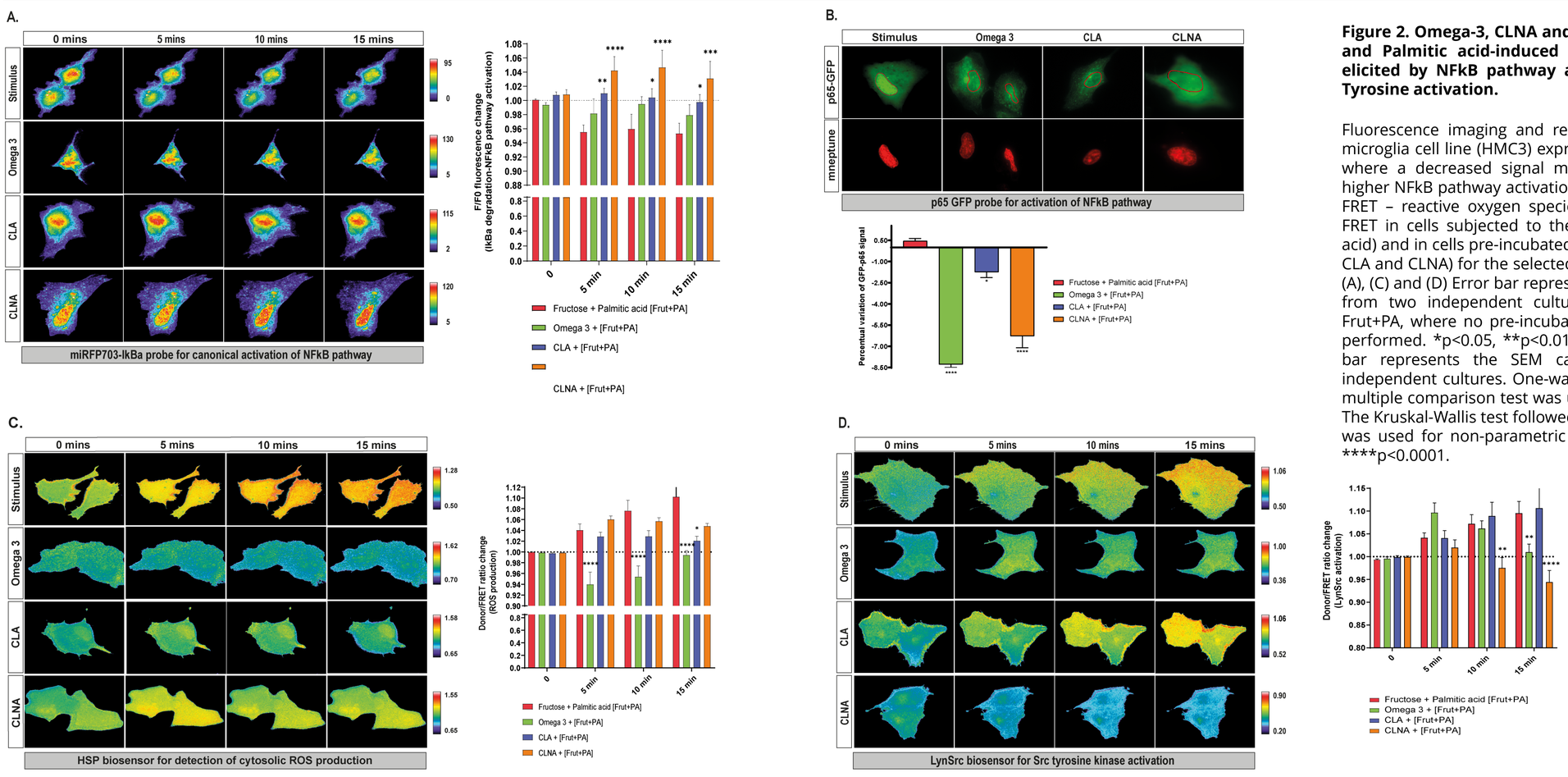

Figure 2. Omega-3, CLNA and CLA fatty acids can revert Fructose and Palmitic acid-induced microglia inflammatory imbalance elicited by NFkB pathway activation, ROS production and Src Tyrosine activation.

Fluorescence imaging and results of the quantification of human microglia cell line (HMC3) expressing the (A) miRFP703-IkBα sensor – where a decreased signal means a bigger IkBα degradation and higher NFkB pathway activation-, (B) GFP-p65 and mneptune, (C) HSP FRET – reactive oxygen species (ROS) production - and (D) LynSrc FRET in cells subjected to the stimulus solution (Fructose+Palmitic acid) and in cells pre-incubated with the studied fatty acids (omega-3, CLA and CLNA) for the selected time points (0, 5, 10 and 15 minutes). (A), (C) and (D) Error bar represents the SEM calculated from >10 cells from two independent cultures. Two-way ANOVA in relation to Frut+PA, where no pre-incubation with the selected fatty acids was performed. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001. (B) Error bar represents the SEM calculated from >10 cells from two independent cultures. One-way ANOVA followed by the Bonferroni's multiple comparison test was used for data with normal distribution. The Kruskal-Wallis test followed by Dunn's multiple comparations test was used for non-parametric data. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

Conclusions

The incubation of the human microglia cell line cultures with the solution mimicking the western pattern diet (Fructose+Palmitic acid) showed an activation of the NFkB pathway (Figure 2A and B) by a significative IkBa degradation (decreased signal) and NFkB p65 subunit nuclear translocation, an increased ROS production (Figure 2C) and LynSrc activation (Figure 2D). We found that pre-incubation of microglia with either CLA or CLNA, significantly prevented the Fructose+PA-induced canonical NFkB pathway activation. Omega-3 and CLA abrogated the Fructose+PA-induced ROS generation in living microglia, suggesting that, under these experimental conditions, these PUFAs displays antioxidant properties. CLNA and omega-3 significantly suppressed the activation of Src triggered by Fructose+PA.

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