The effect of 17β-estradiol on the bio-energetic metabolism of macrophages

Tollemache, F.H. van der Westhuizen, E. Erasmus, and G. Venter

North-West University, Potchefstroom, South Africa

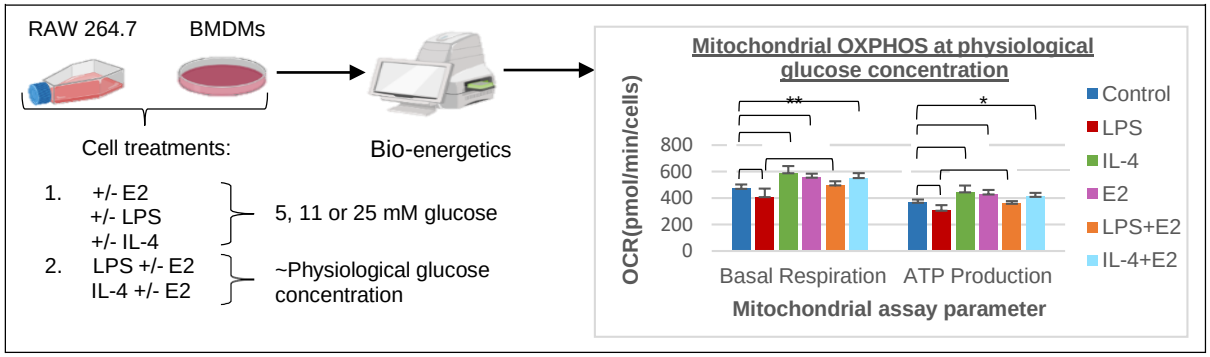

Figure 1: Overview of the experimental procedure and main results (representative of RAW 264.7 cells and BMDMs)

Background: Macrophages are a prime topic in breast cancer studies due to their functional and metabolic plasticity. For example, pro-inflammatory macrophages have cancer-killing abilities, whereas anti-inflammatory macrophages are known to be cancer-promoting[i]. Estrogen is also implicated in cancer initiation. This hormone has an anti-inflammatory effect on macrophages (which can further promote cancer)[ii]. Macrophage polarization is tightly linked to metabolic state[iii], however it is not known how estrogens such as 17β-estradiol (E2) affect the bio-energetic metabolism of naïve, pro-, or anti-inflammatory macrophages.

Materials and methods: RAW 264.7 cells and mouse bone-marrow derived macrophages (BMDMs) were incubated with E2 at different glucose concentrations (5, 11 and 25 mM glucose) and in the presence or absence of the pro- and anti-inflammatory stimulators lipopolysaccharide (LPS) and interleukin 4 (IL-4). The Seahorse extracellular flux analyser was used to determine the oxygen consumption rate (OCR) and extracellular acidification rate (ECAR) as a measure of mitochondrial oxidative phosphorylation (OXPHOS) and glycolytic activity, respectively.

Results: OCR in both RAW 264.7 cells and BMDMs decreased with increased medium glucose concentrations. Bio-energetic differences between naïve, LPS, IL-4, and E2 treated cells were only visible under conditions where the cells neither had limited nor excess glucose. Under these conditions (11 mM for RAW 264.7 and 5 mM for BMDMs), LPS treatment caused a reduction in oxygen consumption, while IL-4 and E2 stimulated a more oxidative metabolism. For both cell types, treatment with IL-4, E2, or IL-4+E2 had similar effects on OCR. However, treatment of the cells with LPS+E2 reversed the metabolic effect of LPS treatment alone and drove the cells towards a more oxidative metabolism.

Conclusion: The effect of LPS, IL-4, and E2 on macrophage metabolism is influenced by the glucose concentration in the culture medium. E2 induces an oxidative metabolism in macrophages, similar to IL-4 treatment. Moreover, E2 causes a shift in the bio-energetic metabolism of LPS-treated cells towards a more anti-inflammatory OXPHOS metabolic profile. Since macrophage polarization and metabolism is reciprocally coupled, this may mean that E2 present in the breast tissue may lead to the conversion of pro-inflammatory, anti-cancer macrophages (present during the early stages of tumor development) to anti-inflammatory tumor promoting macrophages by inducing a metabolic switch. This may favor tumor initiationand progression.

Keywords: Macrophages, metabolism, estrogen, breast cancer

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