Impact of Dietary Fatty Acids on Macrophage Lipid Metabolism, Signaling and Function

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ABSTRACT

Leukocytes are potent regulators of adipose tissue biology and whole-body metabolic homeostasis. In lean, non-obese conditions (insulin-sensitive), adipose tissue has innate and adaptive immune cells, including eosinophils, regulatory T cells, invariant NK cells, and M2 macrophages. A vast expansion in adipose tissue occurs in obesity, and this is associated with a marked alteration in the tissue leukocyte profile. There is a marked increase in B cells, CD8+ T cells, NK cells, neutrophils, and M1 macrophages. This condition induces a state of low-grade, chronic inflammation in the adipose tissue, which disrupts whole-body metabolism. Macrophages were the first leukocyte to be discovered in adipose tissue. Due to their proximity to nearby adipocytes, the macrophages are exposed to high levels of fatty acids and other lipids reported in obesity. Lipid uptake by tissue-resident macrophages is essential for their biological actions. Specifically, lipid uptake and metabolism, particularly of long-chain saturated fatty acids, activate inflammatory signaling pathways, potentiating adipose tissue inflammation, and metabolic dysfunction. Obesity exhibits increased fatty acid levels within the adipose tissue microenvironment. The increased lipid accumulation in the resident macrophages reflects the fatty acid composition of the adipocytes. The dietary fatty acid composition of the adipose tissue is determined by the fatty acids indirectly provided by the diet. The composition varies with the acyl chain length, e.g., short-, medium-, or long-chain, and saturated fatty acids. These fatty acids have wide-ranging effects on macrophages. We described herein in detail the impact of the different dietary fatty acids on...
Introducing macrophage functions. Shortly, long-chain saturated fatty acids are pro-inflammatory, whereas medium-chain fatty acids are relatively benign. Long-chain unsaturated fatty acids often antagonize the pro-inflammatory effects of long-chain saturated fatty acids.

**KEYWORDS:** macrophage; inflammatory function; fatty acids

**INTRODUCTION TO MACROPHAGES**

Macrophages play an essential role in the orchestration of both innate and acquired immune responses. They are versatile cells that exert a wide range of biological functions that include not only defending the organism against pathogenic agents but also participating in tissue homeostasis (in both steady state and injury) and remodeling [1,2]. Elie Metchnikoff was the first to describe macrophages in the 1880s. He stated that macrophages play a central role in the inflammatory response due to their phagocytic nature, which was called theory of cellular immunity [3]. After Metchnikoff, the great diversity of macrophage-like cells observed in both vertebrates and invertebrates led to the concept of the mononuclear phagocytic system (MPS) model proposed by Van Furth and Cohn [4] in an attempt to unify diverse populations of cells into one group [1,5]. According to this model, all tissue macrophages originate from blood monocytes, despite their biological, developmental, functional, and molecular diversity [4,5]. The first view of MPS suggests that recruited macrophages play a crucial role during inflammatory processes, whereas tissue-resident ones are important to local homeostasis [6]. However, the considerable variation of morphologic features, biological functions, and anatomic location [7,8], as well as the lack of a unique marker to distinguish recruited from tissue macrophages [5], led to a revision of the traditional MPS framework and the expansion of this model by including the concept of polarization [5,9].

The origin of macrophages has been extensively reviewed [5,6,8,10,11]. Differences in ontogeny are not sufficient to explain the diversity of macrophages, and functional classifications based on inflammatory states. The most recognized macrophage states are activated macrophages (M1) and alternatively activated macrophages (M2) [6,8]. The microenvironment conditions drive the polarization of macrophages to one type or another. The different activation programs lead the macrophages to carry out the appropriate functions according to the inflammatory state [6]. M1 macrophages utilize arginine metabolism for NO production. They exhibit high expression of pro-inflammatory cytokines (e.g., tumor necrosis factor α (TNF-α), interleukin 1β (IL-1β), and interleukin 6 (IL-6)), chemokines (e.g., C-X-C chemokine receptor type 1 (CXCR1) and C-X-C chemokine receptor type 2 (CXCR2)). M1 macrophages also show a high production of reactive oxygen species (ROS). These cells then have enhanced microbicidal, inflammatory, and tumoricidal
M2 macrophages utilize arginine metabolism to drive ornithine and polyamine production. They are associated with high expression of anti-inflammatory cytokines (e.g., interleukin 4 (IL-4), interleukin 10 (IL-10), and transforming growth factor β (TGF-β)), which allows them to promote tissue remodeling and dampen inflammation by exerting immunoregulatory effects [6,12]. However, this binary classification is just a simplified model to describe the functional diversity of macrophages, and many subsets other than M1 and M2 have been identified [6].

Besides macrophage polarization, transcriptomic studies identified considerable diversity in gene expression profiles in mouse macrophages from different sites [13,14]. The Immunological Genome (ImmGen) Project [15] addressed this issue. Peritoneal, red pulp splenic, lung, and brain macrophages (microglia) revealed an enormous diversity among them. Classical antigenic protein markers such as F4/80 (adhesion and phagocytic receptor present in mouse macrophages) [16,17], CD68- (oxidized LDL receptor) [18] and CD115 (receptor for M-CSF) [19], are routinely used to identify macrophages, but problems arise for sub-type discrimination. However, CD14 and CD64 are additional important markers [14], being related to LPS (CD14) and IgG (CD64) binding [20,21].

Different stimuli induce cytokine production by macrophages (e.g., self-danger signal, microbial stimulus, ROS production). The pattern recognition receptors (PRRs) include Toll-like receptors (TLR) [22,23], and some inflammatory receptors, interleukin-18 receptor (IL-18R), and IL-1R, which require the signaling protein myeloid differentiation primary response gene 88 (Myd88), participate in the induction of pro-inflammatory cytokines expression [22,24]. Myd88 is an adapted molecule that recruits IL-1R-associated kinase (IRAK) family [25], which leads to the activation of the mitogen activated protein kinases (MAPK) and nuclear factor κB (NF-κB) pathways [24,25].

NOD-like receptors (NLRs), recognize self and non-self-antigens, are characterized by a central nucleotide-binding and oligomerization (NACHT) domain, which is the only common region among NLR family members [26,27]. NLR family members, named inflammasomes, play an essential role in innate immunity. These receptors are involved in the activation of caspases [28]. The NACHT, LRR, PYD domains-containing protein 3 (NLRP3) inflammasome is one of these complexes that control the activation of (IL)-1 family cytokines: IL-1β and interleukin 18 (IL-18) [28,29] in macrophages.

We described herein the main immunological and functional modifications in macrophages caused by saturated, ω-3, ω-6, ω-9, and ω-7 fatty acid treatments either in vitro or in vivo.

Saturated, ω-3, ω-6, ω-9, and ω-7 families are the main fatty acids found in the diet. ω-3 and ω-6 fatty acids are essential fatty acids because the organism cannot synthesize them, so they have to be obtained in the diet. Safflower and soybean oil are common sources of ω-6 fatty acids. Marine fish are the primary source of ω-3 fatty acids due to their diet rich in...
phytoplankton and zooplankton (the real sources of omega-3 fatty acids) [30]. Saturated fatty acids are naturally present in many foods, such as fatty beef, pork, and dairy products. Rich sources of ω-7 fatty acids are macadamia oil and sea buckthorn oil.

**IMPACT OF SATURATED FATTY ACIDS (SFAs) ON MACROPHAGE FUNCTION**

The main long-chain SFAs (16 or more carbon atoms in the molecule) in mammals are palmitic (16:0) and stearic (18:0) acids. Animal fat products, as whole milk dairy products and fatty meats, are the primary source of this class of fatty acids. Most studies to investigate the effect of SFA on macrophage function used palmitic and stearic acids. Medium-chain fatty acids were also studied, particularly myristic, caprylic, and lauric acids (Table 1). Lokesh and Wrann (1984) [31] reported that treatment with palmitic acid decreases phagocytosis. However, this fatty acid did not change the capacity of mouse macrophages to kill bacteria. Palmitic acid also did not change the production of superoxide after either stimulation with phorbol myristate acetate (PMA) or opsonized zymosan.

In the same way, the authors [31] described that peritoneal macrophages enriched with myristic or palmitic acids display reduced phagocytic capacity and enhanced adhesion to plastic culture dishes. In contrast, enrichment with polyunsaturated fatty acids increases phagocytosis. In human monocyte-derived macrophages treated with stearic acid, an increase in the effector function and decreased colony-forming units (CFU) of *M. tuberculosis* was observed [32].

**Table 1.** Saturated fatty acids (SFAs).

| Ex vivo studies | Fatty acid | Animal | Dose     | Period of treatment | Main findings                                                                 | Reference |
|-----------------|------------|--------|----------|---------------------|--------------------------------------------------------------------------------|----------|
| Palmitic        | B6D2F1 mice | 25 μg/mL | 24 h     | ↔ killing of intracellular bacteria and production of superoxide anion | [31]     |
| Palmitic        | PO mice    | 33 μM  | 48 h     | ↓ phagocytosis rate | [33]     |
| Palmitic        | Human      | 5 and 7 ppm | 120 h | ↓ CFU of *M. tuberculosis* | [32]     |
| Palmitic        | C57BL/6J mice | 0.1 mM | 24 h     | ↑ CD86 and iNOS     | [34]     |
| Palmitic        | C57BL/6J mice | 0.2 mM | 24 h     | ↑ CD86 and TLR4     |          |
| Palmitic        | C57BL/6J mice | 0.4 mM | 24 h     | ↑ CD86 and TLR4, ↓ MHCII and CD36 |          |
| Palmitic        | C57BL/6J mice | 200–500 μM | 24 h | ↑ NLRP3, IL-1β and IL-18 | [35]     |
| Palmitic        | C57BL/6 mice | 250 and 500 μM | 24 h | ↑ NLRP3 and IL-1β | [36]     |
Table 1. Cont.

| Ex vivo studies | Fatty acid | Animal | Dose     | Period of treatment | Main findings                                      | Reference |
|-----------------|-----------|--------|----------|--------------------|----------------------------------------------------|-----------|
|                 | Palmitic  | Human  | 0.1 mM   | 24 h               | ↑TNF-α, IL-6, IL-1β and CD36                        | [37]      |
|                 | Palmitic  | Human  | 0.4 mM   | 24 h               | ↑IL-1β                                             |           |
|                 | Palmitic  | Human  | 200 μM   | 8 h                | ↑IL-1β and NLRP3                                    | [38]      |
|                 | Palmitic  | Human  | 500 μM   | 1 and 3 h          | ↑p38, c-Jun and ERK1/2 phosphorylated               | [39]      |
|                 | Palmitic  | Human  | 500 μM   | 9 h                | ↑p38, c-Jun, TNF-α, IL-6, IL-8 and ERK1/2 phosphorylated↑IL-10|           |
|                 | Stearic   | Human  | 500 μM   | 3 h                | ↑p38 and c-Jun phosphorylated                       |           |
|                 | Stearic   | Human  | 500 μM   | 9 h                | ↑TNF-α, IL-6 and IL-8                              |           |
|                 | Myristic  | Human  | 500 μM   | 9 h                | ↔ cytokines expression                             |           |
|                 | Stearic   | C57BL/6J TLR2−/− and TLR4−/− mice | 90 μM | 24 h | ↑ER stress                                          | [40]      |

| In vitro studies | Fatty acid | Cell line | Dose     | Period of treatment | Main findings                                      | Reference |
|------------------|------------|-----------|----------|--------------------|----------------------------------------------------|-----------|
|                  | Palmitic   | J774      | 1–10 μM  | 48 h               | ↑NO                                                | [41]      |
| Palmitic Stearic | J774       | 50–200 μM | 48 h     | ↑NO                |                                                    |           |
|                  | Palmitic   | J774      | 750 μM   | 8 h                | ↑NO, iNOS, TNF-α, monocyte chemoattractant protein 1 (MCP-1) ↔ CXCL2 | [42]      |
|                  | Palmitic   | THP-1     | 100–400 μM| 48 h               | ↑PPARY, LXR2 and CD206 ↑HLA-DR, CD86, CD36, TLR4, TNF-α, IL-6 and phagocytosis rate | [34]      |
|                  | Palmitic   | THP-1     | 100 μM   | 24 h               | ↑IL-1β, IL-8, TNF-α                                | [43]      |
|                  | Palmitic   | THP-1     | 150 μM   | 12 h               | ↑IL-1β, caspase-1, TLR2                            | [44]      |
|                  | Palmitic   | THP-1     | 100 μM   | 24 h               | ↑IL-1β, ROS and NLRP3                              | [45]      |
|                  | Palmitic   | THP-1     | 100 μM   | 24 h               | ↑IL-1β, IL-8, TNF-α                                | [46]      |
|                  | Palmitic   | J774      | 50 and 100 μM | 24 h | ↑TNF-α                                              |           |
|                  | Palmitic   | BV-2 microglial cell line | 25–100 μM | 24 h | ↑TNF-α, IL-6, IL-1β, NO and ROS                    | [47]      |
Table 1. Cont.

| Fatty acid | Cell line | Dose     | Period of treatment | Main findings                                                                 | Reference |
|------------|-----------|----------|--------------------|------------------------------------------------------------------------------|-----------|
| Palmitic   | THP-1     | 500 μM   | 1–6 h              | ↑c-Jun, p38, pIkBα phosphorylated                                            | [48]      |
| Palmitic   | THP-1     | 500 μM   | 3 h                | ↑TNF-α                                                                       |           |
| Palmitic   | THP-1     | 500 μM   | 6 h                | ↑IL-8                                                                        |           |
| Palmitic   | THP-1     | 350 mM   | 72 h               | ↑TNF-α, IL-6, IL-1β, IL-10, CCL2 and MCP-1                                  | [49]      |
| Palmitic   | RAW 264.7 | 400 μM   | 6 and 24 h         | ↑TNF-α, IL-1β and MCP-1                                                     | [50]      |
| Palmitic   | RAW 264.7 | 400 μM   | 24 h               | ↑NF-κB, c-Jun N-terminal kinases (JNK) and TGF-β-activated kinase (TAK1)       |           |
| Palmitic   | U937      | 150 μM   | 48 h               | ↑IP-10, IL-8, MCP-1 and COX-2                                               | [51]      |
| Lauric     | U937      | 150 μM   | 6–72 h             | ↑IP-10                                                                       |           |
| Lauric     | U937      | 0–150 μM | 48 h               | ↑IP-10                                                                       |           |
| Palmitic   | U937      | 150 μM   | 4 h                | ↑NF-κB activity                                                              |           |
| Palmitic   | U937      | 150 μM   | 4–24 h             | ↑IkBα phosphorylation                                                       |           |
| Lauric     | RAW 264.7 | 75 μM    | 18 h               | ↑NFκB activation, AKT phosphorylation                                        | [52]      |
| Lauric     | RAW 264.7 | 55 μM    | 18 h               | ↑TLR2 and TLR4                                                               | [53]      |
| Palmitic   | RAW 264.7 | 200 μM   | 24 h               | TLR4 and NFκB activation                                                     | [54]      |
| Stearic    | RAW 264.7 | 75 μM    | 11 h               | ↑COX-2 and TLR-4                                                            | [55]      |
| Lauric     | THP-1     | 200 μM   | 3 h                | ↑IkBα phosphorylation                                                       | [56]      |
| Palmitic   | THP-1     | 100 and 200 μM | 8 h           | ↑IL-1β and NLRP3                                                            |           |
| Palmitic   | THP-1     | 200 μM   | 24 h               | ↑NF-κB/AP-1 activity                                                        |           |
| Palmitic   | RAW 264.7 | 500 μM   | 8 h                | ↑TNF-α and MCP-1                                                            | [57]      |
| Palmitic   | RAW 264.7 | 300 μM   | 24 h               | ↑IL-1β, TLR4 and IL-12p40                                                    |           |
| Palmitic   | THP-1     | 250 μM   | 2 h                | ↑COX2 and IL-1β                                                             | [58]      |
| Palmitic   | THP-1     | 250 μM   | 4 h                | ↓NLRP3                                                                       |           |
| Stearic    | THP-1     | 250 μM   | 16 h               | ↑IL-1β                                                                       |           |
| Stearic    | THP-1     | 250 μM   | 2 h                | ↓COX2                                                                       | [59]      |
| Palmitic   | THP-1     | 150 μM   | 6 h                | ↑PPARγ activity                                                             |           |
| Stearic    | THP-1     | 300 μM   | 6 h                | ↑IL-6                                                                        |           |
| Myristic   | THP-1     | 150 μM   | 6 h                | ↑TNF-α                                                                       |           |
Nitric oxide (NO) is an important effector molecule of cytotoxic activated macrophages [60]. Palmitic and stearic acids increase NO production by J774 macrophages at low doses (up to 10 μM); however, at high concentrations (above 50 μM), these fatty acids lead to the opposite effect, including a decrease of NO production [41]. This finding is not a consensus since increased inducible nitric oxide synthase (iNOS) expression and NO production occurred in J744 [61] and THP-1 [34] cell lines treated with high doses of palmitic acid (above 400 μM).

Treatment of primary and cell line macrophages with palmitic acid has been reported to increase IL-1β [43–45,50,57] TNF-α [43,46,47,62], IL-6 [47], IL-8 [48], IL-18 [35], monocyte chemoattractant protein-1 (MCP-1) [49,50], interferon gamma-inducible factor (IP-10) [51] chemokine (C-C motif) ligand 2 (CCL2) [49]. Concerning IL-10 production, treatment of cell lines with palmitic acid leads to both increase [49] or decrease [39]. Some authors described a dose-dependent effect of palmitic acid: low doses enhance the production of proinflammatory cytokines, whereas high doses (above 200 μM) reduce the production of TNF-α and IL-6 by THP-1 cells [34]. Stearic acid promotes the same enhancing effect on TNF-α [32,39,46,59], IL-6 [32,39,59], IL-8 [32,39] IL-12, macrophage inflammatory protein-1 alpha and beta (MIP-1α and MIP-1β) [32] production in both primary and cell line macrophages. Pararasa et al. (2016) [59] escribed decreased IL-10 production by THP-1 cells after treatment with stearic acid. In line with palmitic and stearic acids, myristic acid also has a stimulatory effect on the production of TNF-α [39,59], IL-6, and IL-8 [39] by macrophages.

In systemic inflammatory conditions (such as obesity), plasma cytokines can originate from adipocytes and macrophages that migrate into the adipose tissue [63–66]. Infiltrating macrophages stimulated by fatty acids released from adipocytes play a crucial role, not only in the establishment of inflammation in this tissue but also in the development of insulin resistance in skeletal muscle and liver, through production and release of inflammatory cytokines [67].

SFAs induce inflammatory responses of macrophages through activation of members of Toll-like receptors (TLR) family TLR2 and TLR4 that play a vital role in the innate immune response [62,63,68,69]. Palmitic acid activates TLR4-dependent signaling pathways, increasing phosphorylation of TAK1 [50], JNK [39], p38 [38,44], and c-Jun [39,48], which leads to activation of the AP-1 transcription factor. Similarly, palmitic acid increases the phosphorylation of nuclear factor of kappa light peptide gene enhancer in B-cells inhibitor α (IκBα), which leads to phosphorylation and translocation of NF-κB p65 to the nucleus [38,50]. Phosphorylation of p38, c-Jun [39], IκBα and NF-κB p65 [39], endoplasmic reticulum stress [40] is also described after treatment of macrophages with stearic acid. Activation of both MAPK signaling and NF-κB pathway by SFAs [52,55,62] leads to the production of cytokines and activation of cyclooxygenase (COX-2) [58] and iNOS. In addition to binding to TLRs,
palmitic [54,62] and lauric [53,55] acids enhances TLR4 expression in different macrophage cell lines as well as expression of other receptors involved in the immune response, such as CD36 [37] and CD86 [34]. The effect of SFAs on the expression of surface receptors is dose dependent. Xiu et al. (2016)[34] described the decreased expression of TLR-4, MHC (major histocompatibility complex) class II, CD36, and CD86 after treatment with high doses of palmitic acid (above 200 µM).

Besides the activation of TLRs, palmitic acid also increases cellular ceramide content [70,71]. The sphingolipids play an important cell function as bioactive lipids. Sphingolipids, including ceramide, sphingosine, sphingosine-1-phosphate, and ceramide-1-phosphate, regulate inflammation, cell growth, migration, and angiogenesis [72]. Sphingomyelin hydrolysis generates ceramide. This bioactive lipid mediates the effect of saturated fatty acids on proinflammatory gene expression. TLR4 signaling up-regulates the expression and activity of the enzymes involved in ceramide de novo synthesis [70,71]. LPS treatment increases ceramide synthesis by activating TLR4 signaling. Corroborating these data, inhibition of ceramide content decreased IL-6 production induced by palmitic fatty acid in murine macrophages [70].

In addition to TLRs, the free fatty acid receptors (FFARs) are G protein-coupled receptors (GPCRs) activated by free fatty acids (FFAs). The FFARs play essential roles not only as signaling of fatty acid nutritional components but also as signaling molecules in numerous physiological processes [73]. FFARs vary with the chain length of the FFA that activates each FFAR. Medium- or long-chain FFAs activate FFAR1 and GPR120, GPR84 is activated by medium-chain FFAs, whereas short-chain FFAs activate FFAR2 and FFAR3 [73]. Intestine and pancreatic β-cells highly express FFAR1, which induces the incretin and insulin secretion, respectively [74]. FFAR1 or protein-coupled receptor GPR40 was involved in the increase of IL-6 expression in endothelial cells [75] and on beta-cell apoptosis after palmitic acid stimulation [76].

Saturated fatty acids also activate MAPK signaling and NF-κB pathways [52,53,62], leading to the production of proinflammatory cytokines. As summarized above, saturated fatty acids raise NO and cytokine production by activating TLR-4 and NF-κB signaling. Additionally, these molecules activate the NLRP3 inflammasome [35,77]. Activated NLRP3 triggers the transformation of pro-caspase 1 to caspase 1. This enzyme leads to proteolytic cleavage of pro-IL-1β and pro-IL-18 into their active forms [78,79]. Treatment of primary macrophages with both palmitic and stearic acids increases NLRP3 expression in primary macrophages [35,36,56] in an IRE-1α-dependent way [37]. In THP-1 cells, treatment with stearic acid leads to the same enhancing effect [56,58]. However, palmitic acid at low doses (up to 200 µM) increases NLRP3 expression [56], whereas, at high doses (250 µM), NLRP3 expression does not change [58]. The NLRP3 inflammasome is an oligomer of caspase 1, PYCARD (an adaptor protein), NALP (a NOD-like receptor) and caspase 5 (sometimes) [80,81]. The
consequence observed after inflammasome activation is caspase-1, IL-1β, and IL-18 production [81,82].

In summary, SFAs trigger a proinflammatory response in macrophages. TLR4-dependent signaling pathways, as MAPK and NF-κB pathways, activate the NLRP3 inflammasome. The increase in the production of NO, proinflammatory cytokines, and other inflammatory mediators then occur (summarized in Table 1 and Figure 1).

**IMPACT OF OMEGA-3 POLYUNSATURATED FATTY ACIDS (ω-3 PUFAs) ON MACROPHAGE FUNCTION**

ω-3 PUFAs are beneficial in inflammation conditions associated with disease states, including atherosclerosis, autoimmune disorders, tumor malignancy, and sepsis [83–85]. ω-3 PUFAs may impact many macrophage functions in healthy and diseased states such as phagocytosis, respiratory burst, pathogen killing, and cytokine release (Table 2).

**Table 2. Omega-3 polyunsaturated fatty acids (ω-3 PUFAs).**

| Fatty acid | Animal | Dose | Period of treatment | Main findings | Reference |
|-----------|--------|------|---------------------|---------------|-----------|
| Fish, olive and coconut oil-fed | Male Wistar rats | - | 8 weeks | ↓lateral membrane movements | [86] |
| EPA | Male weanling mice | 4.4 g or 2.2 g/100 g | 7 days | ↑phagocytosis rate | [87] |
| EPA DHA | Healthy adults | 3.8 g/day 3.6 g/day | 7 weeks | ↑phagocytosis rate ↔ phagocytosis rate (sn-2 position) | [88] |
| EPA DHA | Healthy adults | 4.7 g/day 4.9 g/day | 4 weeks | ↔ phagocytosis rate | [89] |
| EPA | Adult male Sprague-Dawley rats | 5.2% | 4 days | ↔ respiratory burst ↔ phagocytosis assays ↓LTB4 ↓PGE2 | [90] |
| Fish oil | C57BL/6J | 20% diet | 6 weeks | ↓TNF-α levels ↓PGE2 production | [91] |
| Fish oil | Healthy volunteers | 18 g/day | 6 weeks | ↓TNF-α and IL1-β levels | [92] |
| Fish oil | FVB mice | 35% diet | 35 days | ↓IL-6, TNF-α, CCL2 and IL-10 | [93] |
| Fish Oil | C57BL/6J | 200 g/kg | 6 weeks | ↓TNF-α and IL-1β levels ↓LTB4 | [94] |
| ω-3 PUFA | Female BALB/c (H-2d) | 10% Ropufa (ω-3, EPA content 1.5% and DHA 1.1%) | 5 weeks | ↑pathogen killing | [95] |
| Table 2. Cont. |
|---------------|
| **In vivo studies** | **Fatty acid** | **Animal** | **Dose** | **Period of treatment** | **Main findings** | **Reference** |
| **ω-3 PUFA (EPA + DHA)** | C57BL/6J | 100 mg/kg | 6 weeks, twice a week | ↓NLRP3 inflammasome activation caspase-1 IL-1β secretion | [96] |
| LNA | Male Wistar rats | 0.22 g/kg body weight | 10 days, daily | ↓IL-1β, IL-6 and vascular endothelial growth factor (VEGF)(−LPS) ↓IL-10 (+LPS) ↑ IL-1β(+LPS) | [97] |
| **Ex vivo studies** | **Fatty acids** | **Animal** | **Dose** | **Period of treatment** | **Main findings** | **Reference** |
| **EPA/DHA** | Male PO mice | 33 µM | 48 h | ↑phagocytosis rate | [33] |
| Linseed oil | Sprague-Dawley rats | 10% diet | 6 weeks | ↓prostaglandin E (PGE) | [98] |
| Fish oil | Male MF1 mice | 20% diet | 8 weeks | ↓PGE2 e IL-6 levels | [99] |
| Linseed and fish oil | Male Wistar | 15% diet | 6 weeks | ↓PGE2 and LTB4 | [100] |
| **In vitro studies** | **Fatty acids** | **Cell line** | **Dose** | **Period of treatment** | **Main findings** | **Reference** |
| LNA | P388D1 | 20 µM | 8 days | ↑phagocytosis rate | [101] |
| LNA | RAW 264.7 | 15 µM | 72 h | ↑phagocytosis rate ↔ kill intracellular bacteria | [102] |
| EPA | RAW 264.7 | 15 µM | 72 h | ↑phagocytosis rate ↑kill intracellular bacteria | [102] |
| DHA | RAW 264.7 | 15 µM | 72 h | ↑phagocytosis rate ↑kill intracellular bacteria | [102] |
| EPA | RAW 264.7 | 15 µM | 72 h | ↑reactive nitrogen intermediate (RNI)/reactive oxygen intermediate (ROI) ↔ RNI/ROI (with LPS) | [103] |
| DHA | RAW 264.7 | 15 µM | 72 h | ↑RNI/ROI ↑RNI/ROI (with LPS) | [103] |
| ω-3 PUFA (emulsion) | RAW 264.7 | 12% | 4 h | ↓IkB phosphorylation ↓NF-κB activity ↓TNF-α expression | [104] |
| ω-3 PUFA (emulsion) | RAW 264.7 | 12% | 4 h | ↓p44/42 and JNK/SAPK phosphorylation | [105] |
| EPA | RAW 264.7 | 100 µM | 12 and 24 h | ↓NO production ↓iNOS expression | [106] |
| EPA | RAW 264.7 | 30 µM | 19 h 30 min | ↓NO production ↓pp38 | [107] |
| DHA | RAW 264.7 | 30 µM | 19 h 30 min | ↓NO production ↔ NO production | [107] |
### Table 2. Cont.

#### In vitro studies

| Fatty acids | Cell line | Dose               | Period of treatment | Main findings                                                                 | Reference |
|-------------|-----------|--------------------|---------------------|-------------------------------------------------------------------------------|-----------|
| LNA         | RAW 264.7 | 10–40 mg/mL        | 24 h                | ↓NO production                                                               | [108]     |
|             |           |                    |                     | ↓PGE production                                                              |           |
|             |           |                    |                     | ↓COX-2 expression                                                             |           |
|             |           |                    |                     | ↓iNOS expression                                                              |           |
| DHA         | RAW 264.7 | 15, 30, 60 or 100 μM | 48 h, 6 h         | ↓NO, IL-1β and IL-6 levels                                                   | [109]     |
|             |           |                    |                     | ↑heme oxygenase-1 (HO-1) mRNA                                                |           |
|             |           |                    |                     | ↓phospho-STAT1                                                                |           |
| DHA         | J774      | 1–10 μM            | 48 h                | ↑NO production                                                               | [41]      |
| EPA         | J774      | 1–10 μM            | 48 h                | ↑NO production                                                               | [41]      |
| DHA         | J774      | 50–200 μM          | 48 h                | ↑NO production                                                               | [41]      |
| EPA         | J774      | 50–200 μM          | 48 h                | ↑NO production                                                               | [41]      |
| EPA         | RAW 264.7 | 114 μM             | 2 h                 | ↑TNF-α levels                                                                | [110]     |
| EPA (omegaven) | RAW 264.7 | 7 μM               | 4 h                 | ↑TNF-α levels                                                                | [111]     |
| ω-3 PUFA    | RAW 264.7 | 15 μM              | 24 h                | ↑TNF-α, IL-1β and IL-6 levels                                               | [112]     |
| LNA         | RAW 264.7 | 2 or 15 μM (combined with 15 or 2 μM LA, respectively) | 72 h                | ↓NADPH oxidase activity                                                      | [113]     |
|             |           |                    |                     | ↓TNF-α and IL-6 levels                                                       |           |
| EPA         | J774      | 50 μM              | 5 days (pretreat) 48 h | ↑TNF-α, IL-1β and IL-6 levels                                               | [114]     |
| DHA         | J774      | 50 μM              | 5 days (pretreat) 48 h | ↑TNF-α, IL-1β and IL-6 levels                                               | [114]     |
| EPA         | THP-1     | 100 μM             | 48 h                | ↑TNF-α, IL-1β and IL-6 levels                                               | [115]     |
| EPA DHA     | THP-1     | 25 μM              | 6 h                 | ↑TNF-α, IL-1β and IL-6 levels                                               | [116]     |
|             | human macrophages | 100 μM       | 24 h                | ↑TNF-α and IL-6 levels                                                      | [117]     |
| DHA         | THP-1     | 100 μM             | 48 h                | ↑TNF-α, IL-1β and IL-6 levels                                               | [115]     |
|             |           |                    |                     | ↑NFKB expression                                                             |           |
|             |           |                    |                     | ↑cytoplasmic IkappaBalpha expression                                          |           |
| EPA         | THP-1     | 50 μM              | 24 h                | ↑MAPK, AKT1, NFKB and NOS2 expression                                        | [118]     |
| DHA         | THP-1     | 10–20 μg/mL        | -                   | ↑IL-1β and TNF-α expression                                                 | [119]     |
| EPA and DHA | THP-1     | 25 μM              | 24 h                | ↑NO, IL-1β and IL-6 levels and expression                                     | [115]     |
Table 2. Cont.

| Fatty acids | Cell line | Dose   | Period of treatment | Main findings                                                                 | Reference  |
|-------------|-----------|--------|---------------------|--------------------------------------------------------------------------------|------------|
| EPA         | J774      | 15 μM  | 3 h                 | ↓TNF-α ↓bacteria survival                                                      | [95]       |
| DHA         | J774      | 50 μM  | 24 h                | ↓oxidative metabolism ↓phagolysosome maturation ↓TNF-α, IL-6, MCP-1 ↓co-stimulatory molecules (CD40 and CD86) | [120]      |
| DHA         | RAW 264.7 | 20 μM  | 3 + 8 h (stimulation)| ↑activation of TLR2 and 4                                                      | [121]      |
| DHA         | RAW 264.7 | 20 μM  | 1 + 8 h (stimulation)| ↑activation of TLR3, 4, 5, 9 ↓TLR2 dimerized with TLR1 or TLR6                | [53]       |
| DHA         | THP-1 cells | 10 μM | 3 h                 | ↑caspase-1 inflammasome ↓IL-1β secretion                                       | [44]       |
| DHA         | human THP-1 monocyte-derived macrophages | 50 and 100 μM | 24 h | ↑NLRP3 inflammasome ↑IL-1β secretion ↑translocation of NF-kB to the nucleus | [122]      |

Peritoneal macrophages from rodents fed a commercial laboratory chow contain 15–20% of PUFAs. The main PUFAs are arachidonic acid, eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA) [33]. Peritoneal macrophages from rats fed purified diets containing linseed oil (rich in ω-3 alpha-linolenic acid) for 6 weeks have increased the percentage of ω-3 PUFAs and decreased of ω-6 PUFAs in their plasma membrane. Reduced synthesis of prostaglandins (PG) [98,99] and leukotriene B4 [123] from arachidonic acid occurs under the latter condition.

Macrophage phagocytic activity is a crucial step in the inflammatory response. Changes in plasma membrane phospholipid composition and fluidity determine the efficiency of macrophages to engulf and kill microorganisms [86]. Both structural organization and fluidity vary with the fatty acid composition of the membrane phospholipids [103]. ω-3 PUFAs increase macrophage phagocytosis activity [33,87,88,101,102], either in vitro or in vivo experiments.

The enrichment of RAW264.7 with ω-3 PUFAs results in a marked increase of phagocytosis activity [101,102]. However, some authors reported the phagocytic capacity of monocytes from healthy human [88,89] and mouse [90] remains unchanged after ω-3 PUFA
supplementation. A controversy then continues on ω-3 PUFA effects on macrophage phagocytosis.

Evidence suggests that ω-3 PUFAs impair NF-κB signal transduction [104] and mitogen-activated protein kinase (MAPK) pathway, inhibiting pro-inflammatory cytokine and NO production [105] by macrophages. ω-3 PUFAs inactivate the NF-κB signal transduction pathway through inhibition of IκB phosphorylation [104].

Macrophages treated with ω-3 PUFAs (EPA, DHA, and alpha-linolenic acid) produce significantly less NO due to a decrease in iNOS activity [106–109,124]. On the other hand, in J774 cells, treatment with low concentrations of EPA and DHA increases NO production, whereas at high levels, NO production is decreased [41].

Treatment of lipopolysaccharide (LPS)-stimulated macrophages with EPA, DHA, and alpha-linolenic acid (LNA) decreases the production of TNF-α [110–113,124], IL-1β and IL-6 [109,110,112–116]. EPA also reduces TNF-α and IL-6 expression by macrophages derived from blood mononuclear cells and stimulated with LPS and prostaglandin E2-PGE2 [117].

Decreased cytokine production caused by the addition of ω-3 PUFAs associated with impairment in the NF-κB signaling pathway. Both EPA and DHA down-regulate LPS-induced NF-κB/DNA binding in THP-1 macrophages [115]. However, DHA promotes a more pronounced decrease of macrophage nuclear NF-κB p65 [118] and IL-1β [115–119] expression as compared to EPA. DHA is also more effective than EPA to attenuate LPS-induced pro-inflammatory cytokine production by macrophages [114,115]. Reduction of both nuclear translocation and binding capacity to DNA of p65 subunit, as well as augment of NF-κB inhibitor IκBα by ω-3 PUFAs, impair the production of inflammatory mediators [115].

ω-3 PUFAs facilitate bacterial growth due to their effects on the inhibition of pro-inflammatory cytokines. In fact, in vitro treatment of *Mycobacterium tuberculosis*-infected murine macrophages with EPA [95] and DHA [120] enhances the units of mycobacteria due to inhibition of TNF-α production.

T helper 1 cytokines (interferon γ (IFN-γ), TNF-α, IL-6) and T helper 2 cells (IL-4 and IL-13) have a crucial role in macrophage polarization and activation [125,126] during homeostasis and inflammatory diseases [127,128]. Since 1989 it was observed that ω-3 supplementation decreased the production of IL-1β and TNF-α of mononuclear cells [92].

Mice with androgen-sensitive prostate cancer fed with a diet containing ω-3 PUFAs during 35 days had decreased expression of M1 (F4/80, iNOS) M2 (ARG1) markers and IL-6, TNF-α, IL-10 tumor-infiltrating macrophages [93]. Some trials have been showing that ω-3 supplementation during six weeks before prostatectomy surgery, decrease Ki-67 (proliferation marker), prostate cancer development, and progression [129,130].

ω-3 PUFAs decrease the production of pro-inflammatory as mentioned above. However, there are controversial results in primary or cell line macrophages in different states of activation, which lead to variations in
the production of inflammatory mediators. Inflammatory macrophages from fish oil-fed mice had lowered the output of TNF-α and IL-1β compared to control mice. In contrast, resident macrophages from fish oil-fed mice had increased the production of cytokines. These findings support the proposition that dietary ω-3 PUFAs may exert opposite effects depending on the activation status of macrophages [94].

The anti-inflammatory effects of ω-3 PUFAs in NF-κB signaling activation involve several molecular mechanisms [131]. GPR120 (G protein-coupled receptor for long-chain unsaturated fatty acids), recently renamed free fatty acid receptor 4 (FFAR4), is an n-3 PUFA receptor and has anti-inflammatory roles [132,133] in macrophages. Activation of GPR120 by ω-3 PUFAs leads to the internalization of this receptor and its binding to β-arrestin 2. The complex formed by GPR120 and β-arrestin 2 sequesters TAB1 (TGFβ1 activated kinase 1), impairs NF-κB pathway, and inhibits subsequent inflammatory events [96].

Treatment with ω-3 PUFAs limits inflammasome and TLR activation in macrophages. DHA also suppresses NF-κB activation and COX-2 expression induced by TLR-2 agonist [121] and by the agonist for TLR3, 4, 5, 6, or 9 [53] in a cell line macrophage. DHA inhibits NLRP3 inflammasome and subsequent caspase-1 activation [44,96] and IL-1β secretion [96,122].

In summary, dietary ω-3 PUFAs have anti-inflammatory properties, in part, due to competitive inhibition of macrophage arachidonic acid (20:4 ω-6) metabolism [134]. ω-3 PUFAs improve macrophage phagocytic activity (or remains unchanged). however, they impair their capacity to produce cytokines and NO by alleviating NF-κB signaling, TLRs, and NLRP3 inflammasome activation (please see Table 2 and Figure 1).

IMPACT OF OMEGA-6 POLYUNSATURATED FATTY ACIDS (ω-6 PUFAs) ON MACROPHAGE FUNCTION

Dietary fat was considered only as a source of energy until 1929 when George and Mildred Burr reported that dietary fatty acid is required to prevent a deficiency disease that occurred in rats fed a fat-free diet [135]. They concluded that some fatty acids were essential nutrients, such as linoleic acid, which prevented the disease. They named this as an essential fatty acid [136,137].

Arachidonic (AA, 20:4 ω-6) and linoleic (LA, 18:2 ω-6) acids are the primary fatty acids of the ω-6 family. The major sources of linoleic acid are vegetable oils, nuts, seeds, meats, and eggs, except for coconut, cocoa, and palm [138]. AA is found predominantly in grain-fed animals, dairy, and eggs [139]. Human breast milk has high levels of ω-6 PUFAs [140]. These fatty acids are essential during early human life development to maintain the structure and function of cell membranes, tissues, and organs [141,142].

Linoleic acid is a precursor of arachidonic acid and bioactive eicosanoids [139,143]. Dietary linoleic acid is associated with tissue arachidonic acid content. Therefore, eicosanoid formation and
subsequently enhance the inflammatory state intensity related to acute and chronic diseases [138].

Macrophages can present inflammatory profiles when exposed to metabolic stimuli, such as modified lipoproteins in atherosclerosis and diabetes mellitus. The microenvironment lipid content controls the activation state of macrophages to a pro-inflammatory state [144]. Besides the important role of lipid tissue in modifying macrophage profile, macrophages also accumulate in adipose tissue with increasing body weight and contribute to inflammation state in obesity [145].

Metabolic changes in the environment may stimulate non-classical inflammatory macrophages due to different forms of polarization and activation [126]. Once present in adipose tissues, macrophages exhibit levels of heterogeneity display in their activities and functions, reflecting the local metabolic and immune microenvironment [144,146–148].

Resident macrophages from rats fed with linoleic acid abundant diet have decreased the production of IL-1β, IL-6, and VEGF. However, in LPS-stimulated macrophages, linoleic acid promotes an increase of IL-1β release and reduces IL-10 production [97]. Despite LA ingestion not having modified the TNF-α, PGE2 and LTB4 production by peritoneal macrophages [98], the addition of gamma-linolenic acid (GLA) by gastroduodenal feeding catheter decreased PGE2 and LTB4 production in alveolar macrophages after LPS stimulation [90].

The mouse infections can be accompanied by an increase of ω-6 level in plasma, spleen, and liver [149], contributing to an inflammatory state. Mice treated with ω-6 PUFAs presented less control in the dissemination of *Mycobacterium tuberculosis* [95], but after AA treatment in vitro, the infection of J744 macrophages by *M. tuberculosis* decreased [95,150].

In the context of infections caused by pathogens in vitro (Rhodococcus equi and Pseudomonas aeruginosa), ω-6 PUFA family down-regulated inflammation process, decreasing IL-6, TNF-α, and IL-1β levels, and CD86 expression [112]. In the same way, treatment with ω-6 PUFA family (without infection) also decreased IL-6, TNF-α levels, and NADPH oxidase activity [113].

Treatment of LPS-stimulated RAW 264.7 macrophages with eicosadienoic acid (EDA) leads to an increase of TNF-α and PGE2 production and decreased NO synthesis. Macrophages rapidly incorporate EDA and increase the composition of AA. The percentages of both EDA and AA increase in cellular phospholipids in a dose-dependent manner. Modulation of PGE2 and NO production involves a change in the expression of COX-2 and iNOS [151]. Increased production of prostaglandins occurs when murine macrophages are treated with dihomo-γ-linolenic acid (DGLA) [152]. Besides LA, gamma-linolenic acid (GLA) treatment also attenuated the inflammatory state after LPS stimulation in RAW 264.7 macrophages, GLA decreased nuclear factor-κB (NF-κB), activator protein-1 (AP-1) and iNOS expression [153].
In addition to cytokine and NO production, ω-6 PUFAs alter respiratory burst and production of ROS. LA and AA treatment in vitro increased ROS [154] and NO production by RAW 264.7 macrophages [103]. However, LA and AA treatment did not affect respiratory burst in LPS-stimulated macrophages [103]. LA and AA increased (low concentrations) and decreased (high concentrations) NO production by murine cell line J774 [41]. On the other hand, supplementation with LA for ten days did not change the release of the superoxide anion, hydrogen peroxide, and nitrite by peritoneal macrophages [97].

Only a few studies reported LA induced changes in the production of cytokines. LA ingestion for 8 weeks did not affect the production of IL-6, TNF-α, IFN-γ, and IL-10 by peripheral blood mononuclear cells (PBMCs) even after fish oil ingestion [155]. AA increases the phagocytosis capacity of PBMCs isolated from healthy donors and cultivated during 7 days with 10% autologous serum [156]. LA treatment of RAW 264.7 macrophages [102] and mouse monocyte/macrophage line P388D1 [101] in vitro increased phagocytosis capacity.

Excessive levels of ω-6 PUFAs or low levels of ω-3 PUFAs results in an unhealthy ω-6/ω-3 ratio of 20:1, instead of 1:1 that is optimal for humans [157]. LA is the major ω-6, which generates arachidonic acid (AA) and subsequently PGE2, and LTB4 leads to increased production of IL-6, TNF-α, IL-1β, and NF-κB expression [158].

ω-6 fatty acid family reduces and alters the inflammatory grade, particularly after pathogen exposure. They increased the phagocytosis rate, bacterial killing, and decreased NOS and ROS. These findings remain controversial because each treatment depends on the period of treatment and stimulation conditions (please see Table 3 and Figure 1).

Table 3. Omega-6 polyunsaturated fatty acids (ω-6 PUFAs).

| Fatty acid       | Animal                 | Dose      | Period of treatment | Main findings                        | Reference |
|------------------|------------------------|-----------|---------------------|--------------------------------------|-----------|
| Linoleic         | Wistar rats            | 0.22 g/kg | 10 days             | ↓IL-1β, IL-6 and VEGF                 | [97]      |
| Linoleic         | Sprague-Dawley rats    | 59%       | 4 days              | ↔respiratory burst ↔phagocytosis assays | [90]      |
| EPA+gama-linolenic (GLA) | Sprague-Dawley rats | 5%        | 4 days              | ↔respiratory burst ↔phagocytosis assays ↓LTB4 ↓PGE2 | [90] |
| Linoleic         | BALB/c (H-2d)          | 7.5%      | 5 weeks             | ↑bacterial growth                     | [95]      |
### Table 3. Cont.

#### Ex vivo studies

| Treatment | Animal species | Dose | Period of treatment | Main findings | Reference |
|-----------|----------------|------|---------------------|---------------|-----------|
| DGLA (dihomo-γ-linolenic acid) | BALB/c | 25 or 50 μM | 24 h | ↑prostaglandins | [152] |
| LA | Healthy men | Sunflower oil or Becel 60 | 8 weeks | ↔ IL-6, TNF-α, IFN-γ and IL-10 levels | [155] |
| AA | Healthy men | Autologous serum | 7 days | ↑phagocytosis ability | [156] |

#### In vitro studies

| Fatty acid | Cell line | Dose | Period of treatment | Main findings | Reference |
|-----------|-----------|------|---------------------|---------------|-----------|
| AA | J774 | 125 μM | 3 h | ↑bacterial killing | [95] |
| AA | J774 | 125 μM | 6 or 24 h | ↑bacterial killing | [150] |
| Linoleic | RAW 264.7 | 15 μM | 72 h | ↓IL-1β, IL-6 and TNF-α levels ↓CD86 expression | [112] |
| Linoleic | RAW 264.7 | 2 or 15 μM | 72 h | ↓NADPH oxidase activity ↓TNF-α and IL-6 levels | [113] |
| EDA (eicosadienoic acid) | RAW 264.7 | 25, 50 or 100 μM | 24 h | ↓NO ↑IL-1β and IL-6 levels ↑COX-2 expression | [151] |
| GLA (gamalinolenic) | RAW 264.7 | 50, 100 and 200 μM | 18 h | ↓iNOS expression ↓pro-IL-1β expression ↓COX-2 expression ↓NO production ↓NF-κB and AP-1 | [153] |
| LA (linoleic) | RAW 264.7 | 200 μM | 24 h | ↓iNOS expression ↓pro-IL-1β expression ↓COX-2 expression ↓NO production | [153] |
| LA | J744 | 50 or 100 μg/mL | 18 h | ↑ROS | [154] |
| Linoleic (LA) | RAW 264.7 | 15 μM | 72 h | ↑phagocytosis rate ↑kill intracellular bacteria | [102] |
| Arachidonic (AA) | RAW 264.7 | 15 μM | 72 h | ↑phagocytosis rate ↑kill intracellular bacteria | [102] |
| Linoleic (LA) | RAW 264.7 | 15 μM | 72 h | ↑ROS/NO ↑ROS/NO (with LPS) | [103] |
Table 3. Cont.

| Fatty acid       | Cell line | Dose    | Period of treatment | Main findings                                      | Reference |
|------------------|-----------|---------|---------------------|----------------------------------------------------|-----------|
| Arachidonic (AA)| RAW 264.7 | 15 μM   | 72 h                | ↑ROS/NO → ROS/NO (with LPS)                         | [103]     |
| Linoleic         | J774      | 1–10 μM | 48 h                | ↑NO production                                      | [41]      |
| Arachidonic      | J774      | 1–10 μM | 48 h                | ↑NO production                                      |           |
| Linoleic         | J774      | 50–200 μM| 48 h                | ↑NO production                                      | [41]      |
| Arachidonic      | J774      | 50–200 μM| 48 h                | ↑NO production                                      |           |
| LA/LNA           | P388D1    | 20 μM   | 7 days              | ↑phagocytosis rate ↓oxidative burst                 | [101]     |

IMPACT OF MONOUNSATURATED FATTY ACIDS (MUFAs) ON MACROPHAGE FUNCTION

OMEGA-9

Some ω-9 MUFAs are abundant in animal fats and vegetable oils. The most abundant ω-9 MUFAs is oleic acid (OA, 9-octadecenoic acid, 18:1, ω-9), which is the main component of olive oil [158].

A high membrane phospholipid composition of ω-9 MUFAs modulates fluidity and micro-domain structures that affect phagocytosis and respiratory burst [113,159,160]. Macrophages treated with OA have decreased adhesion capacity, which may be related to alterations in membrane fluidity and production of adhesion molecules [33,161]. In line with this finding, Lokesh and Wrann (1984) [31] reported impairment of sheep red blood cell phagocytosis by macrophages enriched with OA. Treatment of primary or cell line macrophages with OA increases phagocytosis [33,162], and fungicidal activity [33,163].

ω-9 MUFAs (more specifically, OA) have anti-inflammatory properties by decreasing the production of some mediators—such as cytokines. Inhibition of tumor necrosis factor-α (TNF-α) secretion in many cell types [164] occurs as a consequence of OA treatment. The effects of ω-9 MUFAs on cytokine production depend on the concentration, period of treatment, cell type (whether it is primary or cell line), and activation state of macrophages. In vitro treatment with OA does not lead to alteration or reduction of steady-state production of interleukin-1β (IL-1β) [56,165], TNF-α [46,59,161,165–167], interleukin-6 (IL-6) [59,161,165,167] monocyte chemokine-activating protein-1 (MCP-1) [165,168], interleukin-10 (IL-10) [59,165], leukotriene B4 (LTB4), prostaglandin E2 (PGE2), prostaglandin D2 (PGD2), prostaglandin F2α (PGF2α), thromboxane B2 (TBX2), 12-hydroxyeicosatetraenoic acid (12-HETE) and 15-hydroxyeicosatetraenoic acid (15-HETE) [165]. On the other hand, in some conditions, OA promotes upregulation in the production of TNF-α [46,161] and IL-10 [59]. Ex vivo treatment with OA also leads to reduction or no alteration of steady-
state production of TNF-α [165,169], IL-1β, IL-6 and MCP-1 [165], but increases the production of IL-10 [165].

High intake of ω-9 MUFAs correlates with a lower risk of cardiovascular disease [170] by decreasing the production of inflammatory mediators. This beneficial effect of ω-9 MUFAs still requires confirmation. Supplementation with ω-9 MUFAs does not lead to alteration of plasminogen activator inhibitor-1 (PAI-1) production [171], reduction of PDGF-A and PDGF-B (both platelet-derived growth factors), MCP-1 [168,172], TNF-α [97,173], vascular endothelial growth factor (VEGF), PGE2, LTB4 [94,97] and 15-HETE [97] or increase of IL-10 [59,97,172]. On the other hand, specific supplementation protocols result in decreased production of IL-1β [66,94,97,162,174], cytokine-induced neutrophil chemoattractant-2αβ (CINC-2αβ) [97], and IL-6 [97,174]. The effect of OA on IL-6, LTB4, TNF-α, PGE2, and PGD2 production remains controversial since supplementation with this fatty acid increases IL-6 and LTB4 [173] and reduces TNF-α [91,94,174] production. OA can also lead to both decrease [91,94] and increase [175] of PGD2 as well as increase of PGE2 [175], depending on the activation state of macrophages. Tappia [173] reported a time-dependent pattern of OA actions: after 4-week supplementation, mice macrophages present decreased production of IL-1β without alteration in PGE2, whereas after 8-week supplementation there is a decrease of PGE2 and an increase of IL-1β.

The effect of OA on NO production depends on its concentration, macrophage cell type, and stimulation state. Treatment of RAW 264.7 macrophages with OA leads to a reduction of iNOS expression [165], with decreased [176] or no change in NO release [177] in LPS-stimulated cells. However, in non-stimulated J774 cells, low doses of OA (up to 50 μM) have been reported to increase both iNOS expression and NO release [41,46]. In primary macrophages, Magdalon [97] reported no change in NO production. The action of OA appears to be time-dependent: mice supplemented with OA for a short period display increased NO production [99] whereas mice supplemented for an extended period display decreased NO production [91,174].

Production of inflammatory mediators is closely related to the macrophage phenotype. The most recognized phenotypes are M1 and M2: in general, the first is proinflammatory, and the last is anti-inflammatory. ω-9 MUFAs alter the production of some mediators by macrophages, but the effect of ω-9 MUFAs on macrophage populations remains unknown. Few studies have reported that OA treatment increases the expression of CD206 [166], macrophage galactose-type lectin-1 (Mgl1), and arginase-1 (Arg1) [165,166], which drive macrophages to M2 phenotype. These cells display an anti-inflammatory feature that is characterized by increased production of anti-inflammatory mediators and decreased production of proinflammatory mediators. The impairment of cytokine and NO production by OA may be related to inhibition of NF-κB activation through the suppression of the TLR4 signaling pathway [121,161]. Although ROS
and cytokine production is associated with NF-κB activation, there is no direct relationship between the modulation of ROS and cytokine release by fatty acids in the same way [41,161]. The effect of OA on TLR4 is not apparent, since both increase [177] and decrease [169] of its expression, as well as the rise of TLR4 migration into lipid rafts [178], have been reported. Concerning the TLR4-activated signaling pathway, OA decreases IkBα phosphorylation [56], which impairs NF-κB translocation [169] and activation [56]. There are reports on increased [41,178] or unaltered [179] NF-κB activity after OA treatment. Augment of PPARγ activity due to decreased acetylation [59] could also be involved in the reduction of NF-κB activation caused by OA. Despite the impairment of NF-κB signaling pathway and the increase of PPARγ activity, OA increases the expression of CD36. This receptor binds modified lipoproteins, necessary for the foam cell formation [180,181], and inflammatory mediators such as chemokine (C-X-C motif) ligand 1 (CXCL1), C-C motif chemokine 22 (CCL22) [166], and COX2 [182]. OA also reduces NLRP3 activation, possibly by impairing mechanisms upstream inflammasome assemblies, such as decreasing caspase-1 activation and pro-IL-1β cleavage [56]. The action of OA is specific for NLRP3, since other inflammasomes (NLRC4, NLRP7, and AIM2) are not affected by this fatty acid [56].

OA does not alter ROS production by both primary and cell line macrophages [99,179]. However, in a short-period treatment, this fatty acid increases superoxide production [161]. The augment of superoxide production may be due to the activation of protein kinase C, leading to an increase of NADPH oxidase activity [161,183]. In the presence of LPS, superoxide may be released to extracellular medium or degraded by antioxidant enzymes. One of these latter enzymes is superoxide dismutase (SOD) that converts superoxide anion to $H_2O_2$. This molecule is a more stable oxygen species associated with a potent microbicidal activity [161]. Treatment with OA increases $H_2O_2$ production in a dose-dependent manner, and this effect increases in the presence of LPS [161]. The increased $H_2O_2$ in extracellular medium occurs concomitantly with a decrease of intracellular superoxide anion. This fact supports the proposition that, in the presence of LPS or after a long period of OA treatment, superoxide anion produced inside the cell is converted by SOD to $H_2O_2$ and released into extracellular medium [161]. In line with these results, diets rich in $\omega$-9 MUFAs increase SOD and glutathione peroxidase activities [184,185]. The effect of OA on ROS production in J774 macrophages may be related to the increased fungicidal activity. Diets enriched with OA may be beneficial to patients with diseases that require pathogen elimination [161]. However, increased ROS production due to dietary supplementation with OA is controversial. There are reports on both decreased $H_2O_2$ production [174] and no alteration of ROS release (both superoxide anion and $H_2O_2$) [31,97,99] due to the effect of OA.

OA alters macrophage percentage in different tissues, which contributes to modulate local immune response [186,187]. C3H/Hen mice
supplemented with a high oleate sunflower oil (ω-9-rich diet) and challenged with *Listeria monocytogenes* present an increased percentage of peritoneal macrophages [186]. In mesenteric adipose tissue (MAT) of OA-supplemented mice, Camell [166] reported an increase of F4/80* cells (macrophages). However, Shirakawa [171] did not observe the alteration of the total number of macrophages in epididymal adipose tissue (EAT). Therefore, the effect of OA seems to depend on the adipose tissue depot. None of these studies reported alteration in the percentage of CD11c* macrophages (M1 macrophages) [166,171]. Also, Cardoso [188] reported that topical treatment with OA decreases the number of macrophages infiltrated into skin wounds, which leads to improvement of the wound healing process.

ω-9 MUFA actions vary according to treatment or supplementation (whether OA or olive oil). The dose used in the procedure, exposure period, cell type (whether it is primary or cell line macrophages), and activation state of macrophages, which explains the great diversity of findings reported in the literature (described in Table 4 and Figure 1).

**Table 4. Omega-9 monounsaturated fatty acids (ω-9 MUFAs).**

| In vivo studies | Fatty acid | Animal | Dose | Period of treatment | Main findings | Reference |
|----------------|------------|--------|------|---------------------|---------------|-----------|
|                | Oleic acid | CH3/Hen mice | 20% in the diet | 6 weeks | ↑macrophages in peritoneal cavity and spleen | [186] |
|                | Olive oil  | BALB/c mice | 15% in the diet | 5, 15, 30, 60 and 90 days | ↑phagocytosis (90 days) ↑IL-1β (except 30 days) | [162] |
|                | Oleic acid | Gck*−/− mice | 72.3 g/kg diet | 25 weeks | ↔total and CD11c* macrophages in epididymal adipose tissue ↔PAI-1 | [171] |
|                | Oleic acid | Wistar rats | 0.22 g/kg body weight | 10 days | ↔ROS, NO, TNF-α, VEGF, PGE2, LTB4 and 15(S)-HETE ↑IL-1β and CINC-2αβ | [97] |
|                | Oleic acid | C57BL/6J mice | 0.5 μmol/kg | 3 h | ↑TLR-4 | [178] |
|                | Olive oil  | C57BL/6J mice | 20 g/100 g diet | 6 weeks | ↑PGE2, TNF-α and NO (with LPS) | [91] |
| Olive oil (60% oleic acid) | C57BL/6J mice | 20 g/100 g diet | 6 weeks | ↑TNF-α and PGE2 (inflammatory macrophages with LPS) ↓LTB4 and IL-1β (inflammatory macrophages with LPS) ↑TNF-α and IL-1β (resident macrophages with LPS) ↓PGE2 and LTB4 (resident macrophages with LPS) | [94] |
### Table 4. Cont.

#### In vivo studies

| Fatty acid | Animal | Dose | Period of treatment | Main findings | Reference |
|------------|--------|------|---------------------|---------------|-----------|
| Oleic acid | C57BL/6J mice | 0.6 g/day | 3 days | ↑F4/80, CD206, Mgl1, CXCL 1, IGF-1 and CCL22 in MAT ↓CD11c in MAT | [166] |
| Olive oil  | Swiss mice | 15 g/100 g diet (77.8% of oleic acid in olive oil) | 8 weeks | ↓H₂O₂ (with PMA) ↓NO, PGE₂, TNF-α, IL-1β and IL-6 (with LPS) | [174] |
| Olive oil  | Wistar rats | 74 g/kg diet | 4 weeks | ↓IL-1β, ↓IL-6 and LTB₄ (with TNF-α) ↓PGE₂ (with TNF-α) | [173] |
| Olive oil  | MF1 mice | 20 g/kg diet (69.9 g/100 g of olive oil) | 8 weeks | ↔superoxide and H₂O₂ (with PMA) ↓NO (with LPS) | [99] |
| Olive oil  | Sprague-Dawley rats | 2 mL/kg | 2 h | ↔NF-κB activity | [179] |

#### Ex vivo studies

| Fatty acid | Animal | Dose | Period of treatment | Main findings | Reference |
|------------|--------|------|---------------------|---------------|-----------|
| Oleic acid | C57Bl/6J mice | 24 μg/mL | 4 days | ↑phagocytosis | [163] |
| Oleic acid | Human (PBMC) | 100 and 300 μM | 4 and 24 h | ↑CD36 | [180] |
| Oleic acid | Human (PBMC) | 10 and 50 μM | 24 h | ↑CD36 | [181] |
| Oleic acid | C57BL/6J mice | 2 μM | 6 h | ↑NF-κB activity (with LPS) | [178] |
| Oleic acid | C57BL/6J mice | 500 μM | 20 h | ↑Arg1 ↓TNF-α | [166] |
| Oleic acid | B6D2F1 mice | 25 μg/mL | 6 h | ↑phagocytosis ↔superoxide | [31] |
| Oleic acid | Human (PBMC) | 200 μM | 8 h | ↑IL-1β | [56] |
| Oleic acid | C57BL/J6 mice | 750 μM | 24 h | ↔TNF-α, IL-6, IL-1β and MCP-1 ↑IL-10, Mcr1, Mgl1 and Arg1 | [165] |
| Oleic acid | Human (PBMC) | 30 μM | 1 h | ↔NF-κB translocation | [169] |
| Oleic acid | Human (PBMC) | 30 μM | 44 h | ↑TLR4, TNF-α and total ERK1/2 | |
## Table 4. Cont.

### Ex vivo studies

| Fatty acid | Animal | Dose | Period of treatment | Main findings | Reference |
|------------|--------|------|---------------------|---------------|-----------|
| Oleic acid | Sprague-Dawley rats (Kupfer cells) | 100 μM | 1 h | ↔ Superoxide | [179] |
| Oleic acid | Peritoneal macrophages (no information about animal species) | 50 μM | 24 h | ↔ TNF-α and IL-6 | [86] |
| Oleic acid | Peritoneal macrophages (no information about animal species) | 100 μM | 24 h | ↔ TNF-α and IL-6 | |}

### In vitro studies

| Fatty acid | Cell line | Dose | Period of treatment | Main findings | Reference |
|------------|-----------|------|---------------------|---------------|-----------|
| Oleic acid | RAW 264.7 | Up to 100 μM | 6, 12 and 24 h | ↔ NO | [177] |
| Oleic acid | J774 | 5 μM | 48 h | ↑NO and iNOS | [41] |
| Oleic acid | J774 | 5 μM | 6, 12 and 24 h | ↑iNOS and NF-κB activity (12 h) | | |
| Oleic acid | RAW 264.7 | 200 μM | 48 h | ↓NO | [176] |
| Oleic acid | J774 | 50 μM | 1, 6 and 12 h | ↔ TFN-α | [46] |
| Oleic acid | J774 | 50 μM | 24 h | ↑TNF-α | | |
| Oleic acid | J774 | 100 μM | 24 h | ↑TNF-α (with LPS) | | |
| Oleic acid | J774 | 25 μM | 24 h | ↑H₂O₂ | [161] |
| Oleic acid | J774 | 50 μM | 24 h | ↑H₂O₂ (with and without LPS), fungicidal activity and TNF-α | | |
| Oleic acid | J774 | 100 μM | 1 h | ↑ROS | | |
| Oleic acid | J774 | 100 μM | 24 h | ↑H₂O₂ (with and without LPS) and fungicidal activity | | |
| Oleic acid | THP-1 | 5, 10 and 100 μM | 24 h | ↑CD36 | [181] |
| Oleic acid | THP-1 | 50 μM | 6, 12 and 24 h | ↑CD36 | | |
| Oleic acid | THP-1 | 50 μM | 24 h | ↑PGD₂ and PGE₂ | [175] |
| Oleic acid | THP-1 | 75 μM | 24 h | ↑PGD₂ and PGE₂ | | |
| Oleic acid | RAW 264.7 | 2 μM | 5 min | ↑TLR4 migration to lipid rafts | [178] |
**Table 4. Cont.**

| Fatty acid | Cell line | Dose | Period of treatment | Main findings | Reference |
|------------|-----------|------|---------------------|---------------|-----------|
| Oleic acid | RAW 264.7 | 500 μM | 20 h | ↑Arg1, CD206 and Mgl1, ↓TNF-α, ↔CD11c | [166] |
| Oleic acid | THP-1     | 500 μM | 24 h | ↔MCP-1 | [168] |
| Oleic acid | THP-1     | 200 μM | 3 h | ↓Ixβα phosphorylation | [56] |
| Oleic acid | THP-1     | 200 μM | 8 h | ↑pro-IL-1β cleavage, caspase-1 activation, NF-κB/AP1 activation | |
| Oleic acid | THP-1     | 200 μM | 24 h | ↑NLRP3 | |
| Oleic acid | THP-1     | 300 μM | 6 h | ↑TNF-α, IL-6 and PPARγ acetylation | |
|           |           |      |         | ↑IL-10 (with LPS) | |
|           |           |      |         | ↔IL-10 (without PMA) | |
| Oleic acid | THP-1     | 50, 150 and 300 μM | 6 h | ↑TNF-α (without LPS) | [165] |
| Oleic acid | RAW 264.7 | 750 μM | 24 h | ↑TNF-α, IL-6, IL-1β, MCP-1, ↓iNOS, LTB4, PGE2, PGD2, PGF2α, TBXB2, 11-HETE, 12-HETE and 15-HETE | |
|           |           |      |         | ↔IL-10, Mcr1 and Mgl1 | |
|           |           |      |         | ↑Arg1 | |
| Oleic acid | THP-1     | 1.8 mM | 4 days | ↑COX2 | [182] |

**OMEGA-7**

The primary fatty acid of this class is the monounsaturated palmitoleic acid (9-hexadecenoic acid) (16:1, ω-7). They are in ester form in triacylglycerol, phospholipids, and other glycerolipids of plants [189]. Macadamia seed and sea buckthorn oil [190–192] contain palmitoleic acid in the ester form. Palmitoleic acid is available from dietary sources, but it is also endogenously produced by adipocytes [193]. This fatty acid increases the fluidity of the cell membrane, inhibits the expression of oncogenes, and reduces inflammation associated with diabetes and the risk of heart diseases [194–197]. However, there are only a few studies regarding the effects of palmitoleic acid on the inflammatory response and, more specifically, on macrophage function.

The phagocytic activity of macrophages is closely associated with the structure of the cell membrane and to the fluidity determined by its lipid composition. The increase of macrophage phagocytosis activity depends on the degree of fatty acid unsaturation; the more unsaturated, the higher the phagocytosis activity. It could be explained by a decrease in the viscosity of the cell membrane, allowing rapid movements and
diffusibility. Schroit & Gallily [163] reported that palmitoleic acid increases peritoneal macrophage phagocytic activity. However, the effects of linoleic acid (18:2, ω-6), α-linolenic acid (18:3, ω-3), and arachidonic acid (20:4, ω-6) are more pronounced.

Guo [193] reported that C57BL/6J mice fed a low-fat diet (LFD) or a high-fat diet (HFD) and supplemented with palmitoleic acid have a lower inflammatory response with a decrease in the number of macrophages/Kupffer cells in the liver [193]. Since macrophages/Kupffer cells play a critical role in the inflammatory response of the liver [198], suppression of their function may contribute to the anti-inflammatory effects of palmitoleic acid. This fatty acid has anti-inflammatory actions on macrophages. Guo [193] and Liu et al. [176] investigated the effect of palmitoleic acid at 50 or 200 μM on RAW 264.7 macrophages. The authors reported a decreased expression of proinflammatory cytokines such as TNF-α [176,193] and IL-6 [193]. There was also a reduction in the phosphorylation of NF-κB at serine 468 of p65 subunit [193]. In line with these studies, treatment of both primary and cell line macrophages with palmitoleic acid reduces iNOS expression and NO release [42,199] and productions of TNF-α [42,199], IL-6 [199] and MCP-1 [42]. Treatment of macrophages with palmitoleic acid also increased the expression of M2 genes as Mrc1, Tgfb1, Il-10, and Mgl2 [199]. These anti-inflammatory effects are mediated by reduction of IκBα degradation and NF-κB p65 nuclear translocation, both dependent on AMPKα [199].

Table 5. Omega-7 monounsaturated fatty acids (ω-7 MUFAs).

| In vivo studies                                      |
|-----------------------------------------------------|
| Fatty acid  | Animal       | Dose   | Period of treatment | Main findings                                      | Reference |
|------------|--------------|--------|---------------------|---------------------------------------------------|-----------|
| Palmitoleic| C57Bl/6j mice| 600 mg/day/kg | 4 weeks             | ↓macrophages infiltration in the liver             | [193]     |

| Ex vivo studies                                      |
|-----------------------------------------------------|
| Fatty acid  | Animal       | Dose   | Period of treatment | Main findings                                      | Reference |
|------------|--------------|--------|---------------------|---------------------------------------------------|-----------|
| Palmitoleic| C57BL/6J mice| 24 μg/mL | 4 days              | ↑phagocytosis rate                                | [163]     |
| Palmitoleic| C57BL/6J mice| 0.5 mM    | 6 h                 | ↓iNOS, NO, IL-6, TNF-α, IκBα degradation and NF-κB translocation ↑Mcr1, Tgfb1, Il-10, Mgl2 and AMPKα | [199]     |

| In vitro studies                                      |
|-----------------------------------------------------|
| Fatty acid  | Cell line    | Dose   | Period of treatment | Main findings                                      | Reference |
|------------|--------------|--------|---------------------|---------------------------------------------------|-----------|
| Palmitoleic| RAW 264.7    | 50 and 200 μM | -                  | ↔TNF-α                                             | [176]     |
| Palmitoleic| J774         | 0.75 mM     | 8 h                 | ↓iNOS, NO, TNF-α and MCP-1 ↔CXCL2                 | [42]      |

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Interest in the effects of ω-7 MUFAs has grown since they seem to invoke anti-inflammatory effects and, thus, might be helpful in the treatment of associated inflammatory diseases. However, they are still under-investigated (see Table 5 and Figure 1 for a summary of effects), and more studies are necessary to unravel their actions on leukocytes, and more specifically, on macrophages.

![Pro- and anti-inflammatory effects of the main dietary fatty acids on macrophage functions. TNF: tumor necrosis factor; IL: interleukin; p: phospho; Akt: protein kinase B, PKB; NF-κB: nuclear factor kappa B; TLR: toll-like receptor; NO: nitric oxide; CD: cluster of differentiation; PG: prostaglandin; LT: leukotriene; H₂O₂: hydrogen peroxide; ROS: reactive oxygen species; NLRP3: NACHT, LRR and PYD domains-containing protein 3.](image)

**CONCLUDING REMARKS**

A summary of the effects of the fatty acid classes on the macrophage inflammatory state herein reported is in Figure 1. The changes in cytokine production, prostaglandin production, phagocytosis capacity, NO production, NF-κB activation, and NLRP3 activation induced by each fatty acid class are in Figure 1. The changes are indicated by increase, decrease, or conflicting. This latter term means that the literature remains undecided about the findings reported. The fatty acid classes were ordered, considering the inflammatory properties. From the pro- to anti-inflammatory fatty acids saturated, ω-9 monounsaturated, ω-7...
monounsaturated, ω-6 polyunsaturated, and ω-3 polyunsaturated. The opposite sequence applies to the anti-inflammatory properties of the fatty acid classes. Some discrepancies may exist between the macrophage inflammatory state and the systemic inflammatory properties of the fatty acid classes. The reported pro-inflammatory effects of the saturated fatty acids and the anti-inflammatory properties of the ω-3 polyunsaturated fatty acids are widely reported in a variety of experimental models and disease conditions.

**AUTHOR CONTRIBUTIONS**

MRD and ARC performed the bibliographical search, and design and generated all the tables. MRD, ARC, PN, and RC wrote this review. GM designed and generated Figure 1. All authors have seen and approved the final version of the manuscript.

**CONFLICTS OF INTEREST**

The authors declare that they have no conflict of interest.

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**REFERENCES**

1. Gordon S, Plüddemann A, Martinez Estrada F. Macrophage heterogeneity in tissues: phenotypic diversity and functions. Immunol Rev. 2014;262(1):36-55.
2. Gosselin D, Link VM, Romanoski CE, Fonseca GJ, Eichenfield DZ, Spann NJ, et al. Environment drives selection and function of enhancers controlling tissue-specific macrophage identities. Cell. 2014;159(6):1327-40.
3. Metchnikoff E. Lectures on the Comparative Pathology of Inflammation. New York (US): Dover Publication; 1968.
4. van Furth R, Cohn ZA, Hirsch JG, Humphrey JH, Spector WG, Langevoort HL. The mononuclear phagocyte system: a new classification of macrophages, monocytes, and their precursor cells. Bull World Health Organ. 1972;46(6):845-52.
5. Geissmann F, Mass E. A stratified myeloid system, the challenge of understanding macrophage diversity. Semin Immunol. 2015;27(6):353-6.
6. Italiani P, Boraschi D. From Monocytes to M1/M2 Macrophages: Phenotypical vs. Functional Differentiation. Front Immunol. 2014;5:514.
7. Hume DA, MacDonald KP. Therapeutic applications of macrophage colony-stimulating factor-1 (CSF-1) and antagonists of CSF-1 receptor (CSF-1R) signaling. Blood. 2012;119(8):1810-20.
8. Wynn TA, Chawla A, Pollard JW. Macrophage biology in development, homeostasis and disease. Nature. 2013;496(7446):445-55.
9. Murray PJ, Allen JE, Biswas SK, Fisher EA, Gilroy DW, Goerdt S, et al. Macrophage activation and polarization: nomenclature and experimental guidelines. Immunity. 2014;41(1):14-20.
10. Perdiguerro EG, Geissmann F. The development and maintenance of resident macrophages. Nat Immunol. 2016;17(1):2-8.
11. Kierdorf K, Prinz M, Geissmann F, Gomez Perdiguerro E. Development and function of tissue resident macrophages in mice. Semin Immunol. 2015;27(6):369-78.
12. Patel U, Rajasingh S, Samanta S, Cao T, Dawn B, Rajasingh J. Macrophage polarization in response to epigenetic modifiers during infection and inflammation. Drug Discov Today. 2017;22(1):186-93.
13. Joic V, Shay T, Sylvia K, Zuk O, Sun X, Kang J, et al. Identification of transcriptional regulators in the mouse immune system. Nat Immunol. 2013;14(6):633-43.
14. Gautier EL, Shay T, Miller J, Greter M, Jakubzick C, Ivanov S, et al. Gene-expression profiles and transcriptional regulatory pathways that underlie the identity and diversity of mouse tissue macrophages. Nat Immunol. 2012;13(11):1118-28.
15. Heng TS, Painter MW, Consortium IGP. The Immunological Genome Project: networks of gene expression in immune cells. Nat Immunol. 2008;9(10):1091-4.
16. Gordon S, Hamann J, Lin HH, Stacey M. F4/80 and the related adhesion-GPCRs. Eur J Immunol. 2011;41(9):2472-6.
17. Austyn JM, Gordon S. F4/80, a monoclonal antibody directed specifically against the mouse macrophage. Eur J Immunol. 1981;11(10):805-15.
18. Chistiakov DA, Killingsworth MC, Myasoedova VA, Orekhov AN, Bobryshev YV. CD68/macrosialin: not just a histochemical marker. Lab Invest. 2017;97(1):4-13.
19. Chitu V, Stanley ER. Regulation of Embryonic and Postnatal Development by the CSF-1 Receptor. Curr Top Dev Biol. 2017;123:229-75.
20. Wu Z, Zhang Z, Lei Z, Lei P. CD14: Biology and role in the pathogenesis of disease. Cytokine Growth Factor Rev. 2019;48:24-31.
21. Akinrinmade OA, Chetty S, Daramola AK, Islam MU, Thepen T, Barth S. CD64: An Attractive Immunotherapeutic Target for M1-type Macrophage Mediated Chronic Inflammatory Diseases. Biomedicines. 2017;5(3):E56. doi: 10.3390/biomedicines5030056
22. Chen CJ, Kono H, Golenbock D, Reed G, Akira S, Rock KL. Identification of a key pathway required for the sterile inflammatory response triggered by dying cells. Nat Med. 2007;13(7):851-6.
23. Asehnoune K, Strassheim D, Mitra S, Kim JY, Abraham E. Involvement of reactive oxygen species in Toll-like receptor 4-dependent activation of NF-kappa B. J Immunol. 2004;172(4):2522-9.
24. Wesche H, Henzel WJ, Shillinglaw W, Li S, Cao Z. MyD88: an adapter that recruits IRAK to the IL-1 receptor complex. Immunity. 1997;7(6):837-47.
25. Kawagoe T, Sato S, Matsushita K, Kato H, Matsui K, Kumagai Y, et al. Sequential control of Toll-like receptor-dependent responses by IRAK1 and IRAK2. Nat Immunol. 2008;9(6):684-91.
26. Chang FY, Shaio MF. Decreased cell-mediated immunity in patients with non-insulin-dependent diabetes mellitus. Diabetes Res Clin Pract. 1995;28(2):137-46.
27. Tschopp J, Martinon F, Burns K. NALPs: a novel protein family involved in inflammation. Nat Rev Mol Cell Biol. 2003;4(2):95-104.
28. Martinon F, Burns K, Tschopp J. The inflammasome: a molecular platform triggering activation of inflammatory caspases and processing of proIL-beta. Mol Cell. 2002;10(2):417-26.
29. Bauernfeind FG, Horvath G, Stutz A, Alnemri ES, MacDonald K, Speert D, et al. Cutting edge: NF-kappaB activating pattern recognition and cytokine receptors license NLRP3 inflammasome activation by regulating NLRP3 expression. J Immunol. 2009;183(2):787-91.
30. Saini RK, Keum YS. Omega-3 and omega-6 polyunsaturated fatty acids: Dietary sources, metabolism, and significance—A review. Life Sci. 2018;203:255-67.
31. Lokesh BR, Wrann M. Incorporation of palmitic acid or oleic acid into macrophage membrane lipids exerts differential effects on the function of normal mouse peritoneal macrophages. Biochim Biophys Acta. 1984;792(2):141-8.
32. Mosquera-Restrepo SF, Caro AC, Peláez-Jaramillo CA, Rojas M. Mononuclear phagocyte accumulates a stearic acid derivative during differentiation into macrophages. Effects of stearic acid on macrophage differentiation and Mycobacterium tuberculosis control. Cell Immunol. 2016;303:24-33.
33. Calder PC, Bond JA, Harvey DJ, Gordon S, Newsholme EA. Uptake and incorporation of saturated and unsaturated fatty acids into macrophage lipids and their effect upon macrophage adhesion and phagocytosis. Biochem J. 1990;269(3):807-14.
34. Xiu F, Diao L, Qi P, Catapano M, Jeschke MG. Palmitate differentially regulates the polarization of differentiating and differentiated macrophages. Immunology. 2016;147(1):82-96.
35. Wen H, Gris D, Lei Y, Jha S, Zhang L, Huang MT, et al. Fatty acid-induced NLRP3-ASC inflammasome activation interferes with insulin signaling. Nat Immunol. 2011;12(5):408-15.
36. Robblee MM, Kim CC, Porter Abate J, Valdearcos M, Sandlund KL, Shenoy MK, et al. Saturated Fatty Acids Engage an IRE1α-Dependent Pathway to Activate the NLRP3 Inflammasome in Myeloid Cells. Cell Rep. 2016;14(11):2611-23.
37. Kratz M, Coats BR, Hisert KB, Hagman D, Mutskov V, Peris E, et al. Metabolic dysfunction drives a mechanistically distinct proinflammatory phenotype in adipose tissue macrophages. Cell Metab. 2014;20(4):614-25.
38. Hillyer L, Dao B, Niemiec P, Lee S, Doidge M, Bemben I, et al. Elevated bioactivity of the tolerogenic cytokines, interleukin-10 and transforming
growth factor-beta, in the blood of acutely malnourished weanling mice. Exp Biol Med (Maywood). 2006;231(8):1439-47.

39. Snodgrass RG, Boß M, Zezina E, Weigert A, Dehne N, Fleming I, et al. Hypoxia Potentiates Palmitate-induced Pro-inflammatory Activation of Primary Human Macrophages. J Biol Chem. 2016;291(1):413-24.

40. Anderson EK, Hill AA, Hasty AH. Stearic acid accumulation in macrophages induces toll-like receptor 4/2-independent inflammation leading to endoplasmic reticulum stress-mediated apoptosis. Arterioscler Thromb Vasc Biol. 2012;32(7):1687-95.

41. de Lima TM, de Sa Lima L, Scavone C, Curi R. Fatty acid control of nitric oxide production by macrophages. FEBS Lett. 2006;580(13):3287-95.

42. Talbot NA, Wheeler-Jones CP, Cleasby ME. Palmitoleic acid prevents palmitic acid-induced macrophage activation and consequent p38 MAPK-mediated skeletal muscle insulin resistance. Mol Cell Endocrinol. 2014;393(1-2):129-42.

43. Haversen L, Danielsson KN, Fogelstrand L, Wiklund O. Induction of proinflammatory cytokines by long-chain saturated fatty acids in human macrophages. Atherosclerosis. 2009;202(2):382-93.

44. Snodgrass RG, Huang S, Choi IW, Rutledge JC, Hwang DH. Inflammasome-mediated secretion of IL-1β in human monocytes through TLR2 activation; modulation by dietary fatty acids. J Immunol. 2013;191(8):4337-47.

45. Fu L, Zhou F, Wang X, Lu F. [Effect of free fatty acid on NALP3 inflammasome signaling pathway in THP-1 macrophages]. Zhong Nan Da Xue Xue Bao Yi Xue Ban. 2014;39(8):811-7.

46. de Lima-Salgado TM, Alba-Loureiro TC, do Nascimento CS, Nunes MT, Curi R. Molecular mechanisms by which saturated fatty acids modulate TNF-alpha expression in mouse macrophage lineage. Cell Biochem Biophys. 2011;59(2):89-97.

47. Wang Z, Liu D, Wang F, Liu S, Zhao S, Ling EA, et al. Saturated fatty acids activate microglia via Toll-like receptor 4/NF-kappaB signalling. Br J Nutr. 2011;107(2):229-41.

48. Namgaladze D, Lips S, Leiker TJ, Murphy RC, Ekroos K, Ferreiros N, et al. Inhibition of macrophage fatty acid β-oxidation exacerbates palmitate-induced inflammatory and endoplasmic reticulum stress responses. Diabetologia. 2014;57(5):1067-77.

49. Caspar-Bauguil S, Kolditz CI, Lefort C, Vila I, Mouisel E, Beuzelin D, et al. Fatty acids from fat cell lipolysis do not activate an inflammatory response but are stored as triacylglycerols in adipose tissue macrophages. Diabetologia. 2015;58(11):2627-36.

50. Chen H, Liu Y, Li D, Song J, Xia M. PGC-1β suppresses saturated fatty acid-induced macrophage inflammation by inhibiting TAK1 activation. IUBMB Life. 2016;68(2):145-55.

51. Laine PS, Schwartz EA, Wang Y, Zhang WY, Karnik SK, Musi N, et al. Palmitic acid induces IP-10 expression in human macrophages via NF-kappaB activation. Biochem Biophys Res Commun. 2007;358(1):150-5.

52. Lee JY, Ye J, Gao Z, Youn HS, Lee WH, Zhao L, et al. Reciprocal modulation of Toll-like receptor-4 signaling pathways involving MyD88 and
phosphatidylinositol 3-kinase/AKT by saturated and polyunsaturated fatty acids. J Biol Chem. 2003;278(39):37041-51.

53. Lee JY, Zhao L, Youn HS, Weatherill AR, Tapping R, Feng L, et al. Saturated fatty acid activates but polyunsaturated fatty acid inhibits Toll-like receptor 2 dimerized with Toll-like receptor 6 or 1. J Biol Chem. 2004;279(17):16971-9.

54. Suganami T, Tanimoto-Koyama K, Nishida J, Itoh M, Yuan X, Mizuarai S, et al. Role of the Toll-like receptor 4/NF-kappaB pathway in saturated fatty acid-induced inflammatory changes in the interaction between adipocytes and macrophages. Arterioscler Thromb Vasc Biol. 2007;27(1):84-91.

55. Lee JY, Sohn KH, Rhee SH, Hwang D. Saturated fatty acids, but not unsaturated fatty acids, induce the expression of cyclooxygenase-2 mediated through Toll-like receptor 4. J Biol Chem. 2001;276(20):16683-9.

56. L’homme L, Esser N, Riva L, Scheen A, Paquot N, Piette J, et al. Unsaturated fatty acids prevent activation of NLRP3 inflammasome in human monocytes/macrophages. J Lipid Res. 2013;54(11):2998-3008.

57. Malandrino MI, Fucho R, Weber M, Calderon-Dominguez M, Mir JF, Valcarcel L, et al. Enhanced fatty acid oxidation in adipocytes and macrophages reduces lipid-induced triglyceride accumulation and inflammation. Am J Physiol Endocrinol Metab. 2015;308(9):E756-69.

58. Martínez-Micaelo N, González-Abuin N, Pinent M, Ardévol A, Blay M. Dietary fatty acid composition is sensed by the NLRP3 inflammasome: omega-3 fatty acid (DHA) prevents NLRP3 activation in human macrophages. Food Funct. 2016;7(8):3480-7.

59. Pararasa C, Ikwuobe J, Shigdar S, Boukouvalas A, Nabney IT, Brown JE, et al. Age-associated changes in long-chain fatty acid profile during healthy aging promote pro-inflammatory monocyte polarization via PPARγ. Aging Cell. 2016;15(1):128-39.

60. Hibbs JB, Taintor RR, Vavrin Z, Rachlin EM. Nitric oxide: a cytotoxic activated macrophage effector molecule. Biochem Biophys Res Commun. 1988;157(1):87-94.

61. Walrand S, Chambon-Savanovitch C, Felgines C, Chassagne J, Raul F, Normand B, et al. Aging: a barrier to renutrition? Nutritional and immunologic evidence in rats. Am J Clin Nutr. 2000;72(3):816-24.

62. Liu SP, Li XY, Li Z, He LN, Xiao Y, Yan K, et al. Octanoylated Ghrelin Inhibits the Activation of the Palmitic Acid-Induced TLR4/NF-κB Signaling Pathway in THP-1 Macrophages. ISRN Endocrinol. 2012;2012:237613.

63. Kamei N, Tobe K, Suzuki R, Ohsugi M, Watanabe T, Kubota N, et al. Overexpression of monocyte chemoattractant protein-1 in adipose tissues causes macrophage recruitment and insulin resistance. J Biol Chem. 2006;281(36):26602-14.

64. Sartipy P, Loskutoff DJ. Monocyte chemoattractant protein 1 in obesity and insulin resistance. Proc Natl Acad Sci U S A. 2003;100(12):7265-70.

65. Weisberg SP, McCann D, Desai M, Rosenbaum M, Leibel RL, Ferrante AW. Obesity is associated with macrophage accumulation in adipose tissue. J Clin Invest. 2003;112(12):1796-808.
66. Xu H, Barnes GT, Yang Q, Tan G, Yang D, Chou CJ, et al. Chronic inflammation in fat plays a crucial role in the development of obesity-related insulin resistance. J Clin Invest. 2003;112(12):1821-30.

67. Olefsky JM, Glass CK. Macrophages, inflammation, and insulin resistance. Annu Rev Physiol. 2010;72:219-46.

68. Nguyen MT, Faveluykis S, Nguyen AK, Reichart D, Scott PA, Jenn A, et al. A subpopulation of macrophages infiltrates hypertrophic adipose tissue and is activated by free fatty acids via Toll-like receptors 2 and 4 and JNK-dependent pathways. J Biol Chem. 2007;282(48):35279-92.

69. Senn JJ. Toll-like receptor-2 is essential for the development of palmitate-induced insulin resistance in myotubes. J Biol Chem. 2006;281(37):26865-75.

70. Jin J, Lu Z, Li Y, Cowart LA, Lopes-Virella MF, Huang Y. Docosahexaenoic acid antagonizes the boosting effect of palmitic acid on LPS inflammatory signaling by inhibiting gene transcription and ceramide synthesis. PLoS One. 2018;13(2):e0193343.

71. Schilling JD, Machkovech HM, He L, Sidhu R, Fujiwara H, Weber K, et al. Palmitate and lipopolysaccharide trigger synergistic ceramide production in primary macrophages. J Biol Chem. 2013;288(5):2923-32.

72. Hannun YA, Obeid LM. Principles of bioactive lipid signalling: lessons from sphingolipids. Nat Rev Mol Cell Biol. 2008;9(2):139-50.

73. Hara T, Kimura I, Inoue D, Ichimura A, Hirasawa A. Free fatty acid receptors and their role in regulation of energy metabolism. Rev Physiol Biochem Pharmacol. 2013;164:77-116.

74. Hara T. Ligands at Free Fatty Acid Receptor 1 (GPR40). Handb Exp Pharmacol. 2017;236:1-16.

75. Lu Z, Li Y, Jin J, Zhang X, Hannun YA, Huang Y. GPR40/FFA1 and neutral sphingomyelinase are involved in palmitate-boosted inflammatory response of microvascular endothelial cells to LPS. Atherosclerosis. 2015;240(1):163-73.

76. Natalicchio A, Labarbuta R, Tortosa F, Biondi G, Marrano N, Peschechera A, et al. Exendin-4 protects pancreatic beta cells from palmitate-induced apoptosis by interfering with GPR40 and the MKK4/7 stress kinase signalling pathway. Diabetologia. 2013;56(11):2456-66.

77. Reynolds CM, McGillicuddy FC, Harford KA, Finucane OM, Mills KH, Roche HM. Dietary saturated fatty acids prime the NLRP3 inflammasome via TLR4 in dendritic cells-implications for diet-induced insulin resistance. Mol Nutr Food Res. 2012;56(8):1212-22.

78. Ozaki E, Campbell M, Doyle SL. Targeting the NLRP3 inflammasome in chronic inflammatory diseases: current perspectives. J Inflamm Res. 2015;8:15-27.

79. Gross O, Thomas CJ, Guarda G, Tschopp J. The inflammasome: an integrated view. Immunol Rev. 2011;243(1):136-51.

80. Jin C, Flavell RA. Molecular mechanism of NLRP3 inflammasome activation. J Clin Immunol. 2010;30(5):628-31.

81. Choi AJ, Ryter SW. Inflammasomes: molecular regulation and implications for metabolic and cognitive diseases. Mol Cells. 2014;37(6):441-8.
82. Lamkanfi M, Dixit VM. Inflammasomes and their roles in health and disease. Annu Rev Cell Dev Biol. 2012;28:137-61.
83. Kremer JM, Robinson DR. Studies of dietary supplementation with omega 3 fatty acids in patients with rheumatoid arthritis. World Rev Nutr Diet. 1991;66:367-82.
84. Kromhout D, Bosschieter EB, de Lezenne Coulander C. The inverse relation between fish consumption and 20-year mortality from coronary heart disease. N Engl J Med. 1985;312(19):1205-9.
85. Beck SA, Smith KL, Tisdale MJ. Anticachectic and antitumor effect of eicosapentaenoic acid and its effect on protein turnover. Cancer Res. 1991;51(22):6089-93.
86. Tappia PS, Ladha S, Clark DC, Grimble RF. The influence of membrane fluidity, TNF receptor binding, cAMP production and GTPase activity on macrophage cytokine production in rats fed a variety of fat diets. Mol Cell Biochem. 1997;166(1-2):135-43.
87. Kew S, Gibbons ES, Thies F, McNeill GP, Quinlan PT, Calder PC. The effect of feeding structured triacylglycerols enriched in eicosapentaenoic or docosahexaenoic acids on murine splenocyte fatty acid composition and leucocyte phagocytosis. Br J Nutr. 2003;90(6):1071-80.
88. Halvorsen DS, Hansen JB, Grimsgaard S, Bonaa KH, Kierulf P, Nordoy A. The effect of highly purified eicosapentaenoic and docosahexaenoic acids on monocyte phagocytosis in man. Lipids. 1997;32(9):935-42.
89. Kew S, Mesa MD, Tricorn S, Buckley R, Minihane AM, Yaqoob P. Effects of oils rich in eicosapentaenoic and docosahexaenoic acids on immune cell composition and function in healthy humans. Am J Clin Nutr. 2004;79(4):674-81.
90. Palombo JD, DeMichele SJ, Boyce PJ, Lydon EE, Liu JW, Huang YS, et al. Effect of short-term enteral feeding with eicosapentaenoic and gamma-linolenic acids on alveolar macrophage eicosanoid synthesis and bactericidal function in rats. Crit Care Med. 1999;27(9):1908-15.
91. Wallace FA, Neely SJ, Miles EA, Calder PC. Dietary fats affect macrophage-mediated cytotoxicity towards tumour cells. Immunol Cell Biol. 2000;78(1):40-8.
92. Endres S, Ghorbani R, Kelley VE, Georgilis K, Lonennam G, van der Meer JW, et al. The effect of dietary supplementation with n-3 polyunsaturated fatty acids on the synthesis of interleukin-1 and tumor necrosis factor by mononuclear cells. N Engl J Med. 1989;320(5):265-71.
93. Liang P, Henning SM, Schokrpur S, Wu L, Doan N, Said J, et al. Effect of dietary omega-3 fatty acids on tumor-associated macrophages and prostate cancer progression. Prostate. 2016;76(14):1293-302. doi: 10.1002/pros.23218
94. Wallace FA, Miles EA, Calder PC. Activation state alters the effect of dietary fatty acids on pro-inflammatory mediator production by murine macrophages. Cytokine. 2000;12(9):1374-9.
95. Jordao L, Lengeling A, Bordat Y, Boudou F, Gicquel B, Neyrolles O, et al. Effects of omega-3 and -6 fatty acids on Mycobacterium tuberculosis in macrophages and in mice. Microbes Infect. 2008;10(12-13):1379-86.
96. Yan Y, Jiang W, Spinetti T, Tardivel A, Castillo R, Bourquin C, et al. Omega-3 fatty acids prevent inflammation and metabolic disorder through inhibition of NLRP3 inflammasome activation. Immunity. 2013;38(6):1154-63.

97. Magdalon J, Vinolo MA, Rodrigues HG, Paschoal VA, Torres RP, Mancini-Filho J, et al. Oral administration of oleic or linoleic acids modulates the production of inflammatory mediators by rat macrophages. Lipids. 2012;47(8):803-12.

98. Magrum LJ, Johnston PV. Modulation of prostaglandin synthesis in rat peritoneal macrophages with omega-3 fatty acids. Lipids. 1983;18(8):514-21.

99. Yaqoob P, Calder P. Effects of dietary lipid manipulation upon inflammatory mediator production by murine macrophages. Cell Immunol. 1995;163(1):120-8.

100. Dinleyici EC, Kilic Z, Buyukkaragoz B, Ucar B, Alatas O, Aydogdu SD, et al. Serum IGF-1, IGFBP-3 and growth hormone levels in children with congenital heart disease: relationship with nutritional status, cyanosis and left ventricular functions. Neuro Endocrinol Lett. 2007;28(3):279-83.

101. Fuhrmann H, Miles EA, West AL, Calder PC. Membrane fatty acids, oxidative burst and phagocytosis after enrichment of P388D1 monocyte/macrophages with essential 18-carbon fatty acids. Ann Nutr Metab. 2007;51(2):155-62.

102. Adolph S, Fuhrmann H, Schumann J. Unsaturated fatty acids promote the phagocytosis of P. aeruginosa and R. equi by RAW264.7 macrophages. Curr Microbiol. 2012;65(6):649-55.

103. Adolph S, Schoeniger A, Fuhrmann H, Schumann J. Unsaturated fatty acids as modulators of macrophage respiratory burst in the immune response against Rhodococcus equi and Pseudomonas aeruginosa. Free Radic Biol Med. 2012;52(11-12):2246-53.

104. Novak TE, Babcock TA, Jho DH, Helton WS, Espat NJ. NF-kappa B inhibition by omega-3 fatty acids modulates LPS-stimulated macrophage TNF-alpha transcription. Am J Physiol Lung Cell Mol Physiol. 2003;284(1):L84-9.

105. Babcock TA, Kurland A, Helton W, Rahman A, Anwar KN, Espat NJ. Inhibition of activator protein-1 transcription factor activation by omega-3 fatty acid modulation of mitogen-activated protein kinase signaling kinases. JPEN J Parenter Enteral Nutr. 2003;27(3):176-80; discussion 81.

106. Aldridge C, Razzak A, Babcock TA, Helton WS, Espat NJ. Lipopolysaccharide-stimulated RAW 264.7 macrophage inducible nitric oxide synthase and nitric oxide production is decreased by an omega-3 fatty acid lipid emulsion. J Surg Res. 2008;149(2):296-302.

107. Pallares V, Calay D, Cedo L, Castell-Auvi A, Raes M, Pinent M, et al. Enhanced anti-inflammatory effect of resveratrol and EPA in treated endotoxin-activated RAW 264.7 macrophages. Br J Nutr. 2012;108(9):1562-73.

108. Ren J, Han EJ, Chung SH. In vivo and in vitro anti-inflammatory activities of alpha-linolenic acid isolated from Actinidia polygama fruits. Arch Pharm Res. 2007;30(6):708-14.

109. Choi EY, Jin JY, Choi JJ, Choi IS, Kim SJ. DHA suppresses Prevotella intermedia lipopolysaccharide-induced production of proinflammatory mediators in murine macrophages. Br J Nutr. 2014;1-10.
110. Lo CJ, Chiu KC, Fu M, Lo R, Helton S. Fish oil decreases macrophage tumor necrosis factor gene transcription by altering the NF kappa B activity. J Surg Res. 1999;82(2):216-21.

111. Babcock TA, Helton WS, Hong D, Espat NJ. Omega-3 fatty acid lipid emulsion reduces LPS-stimulated macrophage TNF-alpha production. Surg Infect (Larchmt). 2002;3(2):145-9.

112. Schoeniger A, Adolph S, Fuhrmann H, Schumann J. The impact of membrane lipid composition on macrophage activation in the immune defense against Rhodococcus equi and Pseudomonas aeruginosa. Int J Mol Sci. 2011;12(11):7510-28.

113. Walloschke B, Fuhrmann H, Schumann J. Enrichment of RAW264.7 macrophages with essential 18-carbon fatty acids affects both respiratory burst and production of immune modulating cytokines. J Nutr Biochem. 2010;21(6):556-60.

114. Oliver E, McGillicuddy FC, Harford KA, Reynolds CM, Phillips CM, Ferguson JF, et al. Docosahexaenoic acid attenuates macrophage-induced inflammation and improves insulin sensitivity in adipocytes-specific differential effects between LC n-3 PUFAs. J Nutr Biochem. 2012;23(9):1192-200.

115. Weldon SM, Mullen AC, Loscher CE, Hurley LA, Roche HM. Docosahexaenoic acid induces an anti-inflammatory profile in lipopolysaccharide-stimulated human THP-1 macrophages more effectively than eicosapentaenoic acid. J Nutr Biochem. 2007;18(4):250-8.

116. Mullen A, Loscher CE, Roche HM. Anti-inflammatory effects of EPA and DHA are dependent upon time and dose-response elements associated with LPS stimulation in THP-1-derived macrophages. J Nutr Biochem. 2010;21(5):444-50.

117. Hao W, Wong OY, Liu X, Lee P, Chen Y, Wong KK. omega-3 fatty acids suppress inflammatory cytokine production by macrophages and hepatocytes. J Pediatr Surg. 2010;45(12):2412-8.

118. Allam-Ndoul B, Guénard F, Barbier O, Vohl MC. Effect of n-3 fatty acids on the expression of inflammatory genes in THP-1 macrophages. Lipids Health Dis. 2016;15(1):69.

119. Solanki P, Aminoshariae A, Jin G, Montagnese TA, Mickel A. The effect of docosahexaenoic acid (DHA) on expression of IL-1ß, IL-6, IL-8, and TNF-α in normal and lipopolysaccharide (LPS)-stimulated macrophages. Quintessence Int. 2013;44(6):393.

120. Bonilla DL, Ly LH, Fan YY, Chapkin RS, McMurray DN. Incorporation of a dietary omega 3 fatty acid impairs murine macrophage responses to Mycobacterium tuberculosis. PLoS One. 2010;5(5):e10878.

121. Lee JY, Plakidas A, Lee WH, Helkkinen A, Chanmugam P, Bray G, et al. Differential modulation of Toll-like receptors by fatty acids: preferential inhibition by n-3 polyunsaturated fatty acids. J Lipid Res. 2003;44(3):479-86.

122. Williams-Bey Y, Boularan C, Vural A, Huang NN, Hwang IY, Shan-Shi C, et al. Omega-3 free fatty acids suppress macrophage inflammasome activation by inhibiting NF-kappaB activation and enhancing autophagy. PLoS One. 2014;9(6):e97957.
123. Brouard C, Pascaud M. Effects of moderate dietary supplementations with n-3 fatty acids on macrophage and lymphocyte phospholipids and macrophage eicosanoid synthesis in the rat. Biochim Biophys Acta. 1990;1047(1):19-28.

124. Ren J, Chung SH. Anti-inflammatory effect of alpha-linolenic acid and its mode of action through the inhibition of nitric oxide production and inducible nitric oxide synthase gene expression via NF-kappaB and mitogen-activated protein kinase pathways. J Agric Food Chem. 2007;55(13):5073-80.

125. Gordon S, Martinez FO. Alternative activation of macrophages: mechanism and functions. Immunity. 2010;32(5):593-604.

126. Mantovani A, Sica A, Sozzani S, Allavena P, Vecchi A, Locati M. The chemokine system in diverse forms of macrophage activation and polarization. Trends Immunol. 2004;25(12):677-86.

127. Cassetta L, Cassel E, Poli G. Macrophage polarization in health and disease. ScientificWorldJournal. 2011;11:2391-402.

128. Sica A, Erreni M, Allavena P, Porta C. Macrophage polarization in pathology. Cell Mol Life Sci. 2015;72(21):4111-26.

129. Aronson WJ, Kobayashi N, Barnard Rj, Henning S, Huang M, Jardack PM, et al. Phase II prospective randomized trial of a low-fat diet with fish oil supplementation in men undergoing radical prostatectomy. Cancer Prev Res (Phila). 2011;4(12):2062-71.

130. Galet C, Gollapudi K, Stepanian S, Byrd JB, Henning SM, Grogan T, et al. Effect of a low-fat fish oil diet on proinflammatory eicosanoids and cell-cycle progression score in men undergoing radical prostatectomy. Cancer Prev Res (Phila). 2014;7(1):97-104.

131. Im DS. Functions of omega-3 fatty acids and FFA4 (GPR120) in macrophages. Eur J Pharmacol. 2016;785:36-43.

132. Hirasawa A, Tsumaya K, AwaJI T, Katsuma S, Adachi T, Yamada M, et al. Free fatty acids regulate gut incretin glucagon-like peptide-1 secretion through GPR120. Nat Med. 2005;11(1):90-4.

133. Oh DY, Talukdar S, Bae EJ, Imamura T, Morinaga H, Fan W, et al. GPR120 is an omega-3 fatty acid receptor mediating potent anti-inflammatory and insulin-sensitizing effects. Cell. 2010;142(5):687-98.

134. Calder PC. Omega-3 fatty acids and inflammatory processes. Nutrients. 2010;2(3):355-74.

135. Burr GO, Burr MM. Nutrition classics from The Journal of Biological Chemistry 82:345-67, 1929. A new deficiency disease produced by the rigid exclusion of fat from the diet. Nutr Rev. 1973;31(8):248-9.

136. Smith W, Mukhopadhyay R. Essential fatty acids: the work of George and Mildred Burr. J Biol Chem. 2012;287(42):35439-41.

137. Burr GO. The essential fatty acids fifty years ago. Prog Lipid Res. 1981;20:xxviii-xxix.

138. Whelan J. The health implications of changing linoleic acid intakes. Prostaglandins Leukot Essent Fatty Acids. 2008;79(3-5):165-7.

139. Simopoulos AP. An Increase in the Omega-6/Omega-3 Fatty Acid Ratio Increases the Risk for Obesity. Nutrients. 2016;8(3):128.
140. Brenna JT, Varamini B, Jensen RG, Diersen-Schade DA, Boettcher JA, Arterburn LM. Docosahexaenoic and arachidonic acid concentrations in human breast milk worldwide. Am J Clin Nutr. 2007;85(6):1457-64.

141. Martinez M. Tissue levels of polyunsaturated fatty acids during early human development. J Pediatr. 1992;120(4 Pt 2):S129-38.

142. Koletzko B, Carlson SE, van Goudoever JB. Should Infant Formula Provide Both Omega-3 DHA and Omega-6 Arachidonic Acid? Ann Nutr Metab. 2015;66(2-3):137-8.

143. Simopoulos AP. The importance of the omega-6/omega-3 fatty acid ratio in cardiovascular disease and other chronic diseases. Exp Biol Med (Maywood). 2008;233(6):674-88.

144. Gordon S. Macrophage heterogeneity and tissue lipids. J Clin Invest. 2007;117(1):89-93.

145. Shoelson SE, Herrero L, Naaz A. Obesity, inflammation, and insulin resistance. Gastroenterology. 2007;132(6):2169-80.

146. Lumeng CN, Bodzin JL, Saltiel AR. Obesity induces a phenotypic switch in adipose tissue macrophage polarization. J Clin Invest. 2007;117(1):175-84.

147. Lumeng CN, Deyoung SM, Bodzin JL, Saltiel AR. Increased inflammatory properties of adipose tissue macrophages recruited during diet-induced obesity. Diabetes. 2007;56(1):16-23.

148. Heemskerk MM, Giera M, Bouazzaoui FE, Lips MA, Pijl H, van Dijk KW, et al. Increased PUFA Content and 5-Lipoxygenase Pathway Expression Are Associated with Subcutaneous Adipose Tissue Inflammation in Obese Women with Type 2 Diabetes. Nutrients. 2015;7(9):7676-90.

149. Lachance C, Segura M, Dominguez-Punaro MC, Wojewodka G, De Sanctis JB, Radzioch D, et al. Deregulated balance of omega-6 and omega-3 polyunsaturated fatty acids following infection by the zoonotic pathogen Streptococcus suis. Infect Immun. 2014;82(5):1778-85.

150. Anes E, Kühnel MP, Bos E, Moniz-Pereira J, Habermann A, Griffiths G. Selected lipids activate phagosome actin assembly and maturation resulting in killing of pathogenic mycobacteria. Nat Cell Biol. 2003;5(9):793-802.

151. Huang YS, Huang WC, Li CW, Chuang LT. Eicosadienoic acid differentially modulates production of pro-inflammatory modulators in murine macrophages. Mol Cell Biochem. 2011;358(1-2):85-94.

152. Kakutani S, Kawashima H, Tanaka T, Shiraishi-Tateishi A, Kiso Y. Uptake of dihomo-gamma-linolenic acid by murine macrophages increases series-1 prostaglandin release following lipopolysaccharide treatment. Prostaglandins Leukot Essent Fatty Acids. 2010;83(1):23-9.

153. Chang CS, Sun HL, Lii CK, Chen HW, Chen PY, Liu KL. Gamma-linolenic acid inhibits inflammatory responses by regulating NF-kappaB and AP-1 activation in lipopolysaccharide-induced RAW 264.7 macrophages. Inflammation. 2010;33(1):46-57.

154. Rom O, Jeries H, Hayek T, Aviram M. Supplementation with linoleic acid-rich soybean oil stimulates macrophage foam cell formation via increased oxidative stress and diacylglycerol acyltransferase1-mediated triglyceride biosynthesis. Biofactors. 2017;43(1):100-16. doi: 10.1002/biof.1319
155. Damsgaard CT, Lauritzen L, Calder PC, Kjaer TR, Frøkiaer H. Reduced ex vivo interleukin-6 production by dietary fish oil is not modified by linoleic acid intake in healthy men. J Nutr. 2009;139(7):1410-4.

156. Gutowska I, Baśkiewicz M, Machaliński B, Chlubek D, Stachowska E. Blood arachidonic acid and HDL cholesterol influence the phagocytic abilities of human monocytes/macrophages. Ann Nutr Metab. 2010;57(2):143-9.

157. Simopoulos AP. Evolutionary aspects of diet and essential fatty acids. World Rev Nutr Diet. 2001;88:18-27.

158. James MJ, Gibson RA, Cleland LG. Dietary polyunsaturated fatty acids and inflammatory mediator production. Am J Clin Nutr. 2000;71(1 Suppl):343S-8S.

159. Stulnig TM, Huber J, Leitinger N, Imre EM, Angelisova P, Nowotny P, et al. Polyunsaturated eicosapentaenoic acid displaces proteins from membrane rafts by altering raft lipid composition. J Biol Chem. 2001;276(40):37335-40.

160. Padovese R, Curi R. Modulation of rat neutrophil function in vitro by cis- and trans-MUFA. Br J Nutr. 2009;101(9):1351-9.

161. Martins de Lima-Salgado T, Coccuzzo Sampaio S, Cury-Boaventura MF, Curi R. Modulatory effect of fatty acids on fungicidal activity, respiratory burst and TNF-α and IL-6 production in J774 murine macrophages. Br J Nutr. 2011;105(8):1173-9.

162. de Pablo MA, Ortega E, Gallego AM, Alvarez C, Pancorbo PL, Alvarez de Cienfuegos G. The effect of dietary fatty acid manipulation on phagocytic activity and cytokine production by peritoneal cells from Balb/c mice. J Nutr Sci Vitaminol (Tokyo). 1998;44(1):57-67.

163. Schroit AJ, Gallily R. Macrophage fatty acid composition and phagocytosis: effect of unsaturation on cellular phagocytic activity. Immunology. 1979;36(2):199-205.

164. Yaqoob P. Monounsaturated fatty acids and immune function. Eur J Clin Nutr. 2002;56(Suppl 3):S9-13.

165. Pardo V, González-Rodríguez Á, Guijas C, Balsinde J, Valverde Á. Opposite cross-talk by oleate and palmitate on insulin signaling in hepatocytes through macrophage activation. J Biol Chem. 2015;290(18):11663-77.

166. Camell C, Smith CW. Dietary oleic acid increases m2 macrophages in the mesenteric adipose tissue. PLoS One. 2013;8(9):e75147.

167. Tappia PS, Man WJ, Grimble RF. Influence of unsaturated fatty acids on the production of tumour necrosis factor and interleukin-6 by rat peritoneal macrophages. Mol Cell Biochem. 1995;143(2):89-98.

168. Cullberg KB, Larsen J, Pedersen SB, Richelsen B. Effects of LPS and dietary free fatty acids on MCP-1 in 3T3-L1 adipocytes and macrophages in vitro. Nutr Diabetes. 2014;4:e113.

169. Zacherl JR, Tourkova I, St Croix CM, Robinson LJ, Peck Palmer OM, Mihalik SJ, et al. Elaidate, an 18-carbon trans-monoenoic fatty acid, but not physiological fatty acids increases intracellular Zn(2+) in human macrophages. J Cell Biochem. 2015;116(4):524-32.

170. Renaud S, de Lorgeril M, Delaye J, Guidollet J, Jacquard F, Mamelle N, et al. Cretan Mediterranean diet for prevention of coronary heart disease. Am J Clin Nutr. 1995;61(Suppl):1360S-7S.
171. Shirakawa J, Fujii H, Ohnuma K, Sato K, Ito Y, Kaji M, et al. Diet-induced adipose tissue inflammation and liver steatosis are prevented by DPP-4 inhibition in diabetic mice. Diabetes. 2011;60(4):1246-57.

172. Baumann KH, Hessel F, Larass I, Müller T, Angerer P, Kiefl R, et al. Dietary omega-3, omega-6, and omega-9 unsaturated fatty acids and growth factor and cytokine gene expression in unstimulated and stimulated monocytes. A randomized volunteer study. Arterioscler Thromb Vasc Biol. 1999;19(1):59-66.

173. Tappia PS, Grimble RF. The relationship between altered membrane composition, eicosanoids and TNF-induced IL1 and IL6 production in macrophages of rats fed fats of different unsaturated fatty acid composition. Mol Cell Biochem. 1996;165(2):135-43.

174. de la Puerta R, Marquez-Martin A, Fernandez-Arche A, Ruiz-Gutierrez V. Influence of dietary fat on oxidative stress and inflammation in murine macrophages. Nutrition. 2009;25(5):548-54.

175. Vanhorn J, Altenburg JD, Harvey KA, Xu Z, Kovacs RJ, Siddiqui RA. Attenuation of niacin-induced prostaglandin D(2) generation by omega-3 fatty acids in THP-1 macrophages and Langerhans dendritic cells. J Inflamm Res. 2012;5:37-50.

176. Liu X, Miyazaki M, Flowers MT, Sampath H, Zhao M, Chu K, et al. Loss of Stearoyl-CoA desaturase-1 attenuates adipocyte inflammation: effects of adipocyte-derived oleate. Arterioscler Thromb Vasc Biol. 2010;30(1):31-8.

177. Ohata T, Fukuda K, Takahashi M, Sugimura T, Wakabayashi K. Suppression of nitric oxide production in lipopolysaccharide-stimulated macrophage cells by omega 3 polyunsaturated fatty acids. Jpn J Cancer Res. 1997;88(3):234-7.

178. Villacorta L, Chang L, Salvatore SR, Ichikawa T, Zhang J, Petrovic-Djergovic D, et al. Electrophilic nitro-fatty acids inhibit vascular inflammation by disrupting LPS-dependent TLR4 signalling in lipid rafts. Cardiovasc Res. 2013;98(1):116-24. doi: 10.1093/cvr/cvt002

179. Rusyn I, Bradham CA, Cohn L, Schoonhoven R, Swenberg JA, Brenner DA, et al. Corn oil rapidly activates nuclear factor-kappaB in hepatic Kupffer cells by oxidant-dependent mechanisms. Carcinogenesis. 1999;20(11):2095-100.

180. Svensson L, Camejo G, Cabré A, Vallvé JC, Pedreño J, Norén K, et al. Fatty acids modulate the effect of darglitazone on macrophage CD36 expression. Eur J Clin Invest. 2003;33(6):464-71.

181. Vallvé JC, Uliaque K, Girona J, Cabré A, Ribalta J, Heras M, et al. Unsaturated fatty acids and their oxidation products stimulate CD36 gene expression in human macrophages. Atherosclerosis. 2002;164(1):45-56.

182. Lloyd EE, Gaubatz JW, Burns AR, Pownall HJ. Sustained elevations in NEFA induce cyclooxygenase-2 activity and potentiate THP-1 macrophage foam cell formation. Atherosclerosis. 2007;192(1):49-55.

183. Huang ZH, Hii CS, Rathjen DA, Poulos A, Murray AW, Ferrante A. N-6 and n-3 polyunsaturated fatty acids stimulate translocation of protein kinase Calpha, -betaI, -betaII and -epsilon and enhance agonist-induced NADPH oxidase in macrophages. Biochem J. 1997;325(Pt 2):553-7.

184. Avula CP, Fernandes G. Modulation of antioxidant enzymes and apoptosis in mice by dietary lipids and treadmill exercise. J Clin Immunol. 1999;19(1):35-44.
185. El Seweidy MM, El-Swefy SE, Abdallah FR, Hashem RM. Dietary fatty acid unsaturation levels, lipoprotein oxidation and circulating chemokine in experimentally induced atherosclerotic rats. J Pharm Pharmacol. 2005;57(11):1467-74.

186. Huang SC, Misfeldt ML, Fritsche KL. Dietary fat influences Ia antigen expression and immune cell populations in the murine peritoneum and spleen. J Nutr. 1992;122(6):1219-31.

187. Vitetta ES, Fernandez-Botran R, Myers CD, Sanders VM. Cellular interactions in the humoral immune response. Adv Immunol. 1989;45:1-105.

188. Cardoso CR, Favoreto S, Oliveira LL, Vancim JO, Barban GB, Ferraz DB, et al. Oleic acid modulation of the immune response in wound healing: a new approach for skin repair. Immunobiology. 2011;216(3):409-15.

189. Wu Y, Li R, Hildebrand DF. Biosynthesis and metabolic engineering of palmitoleate production, an important contributor to human health and sustainable industry. Prog Lipid Res. 2012;51(4):340-9.

190. Nestel P, Clifton P, Noakes M. Effects of increasing dietary palmitoleic acid compared with palmitic and oleic acids on plasma lipids of hypercholesterolemic men. J Lipid Res. 1994;35(4):656-62.

191. Yang ZH, Miyahara H, Hatanaka A. Chronic administration of palmitoleic acid reduces insulin resistance and hepatic lipid accumulation in KK-Ay Mice with genetic type 2 diabetes. Lipids Health Dis. 2011;10:120.

192. Yang B, Kallio HP. Fatty acid composition of lipids in sea buckthorn (*Hippophaë rhamnoides* L.) berries of different origins. J Agric Food Chem. 2001;49(4):1939-47.

193. Guo X, Li H, Xu H, Halim V, Zhang W, Wang H, et al. Palmitoleate induces hepatic steatosis but suppresses liver inflammatory response in mice. PLoS One. 2012;7(6):e39286.

194. Welters HJ, Diakogiannaki E, Mordue JM, Tadayyon M, Smith SA, Morgan NG. Differential protective effects of palmitoleic acid and cAMP on caspase activation and cell viability in pancreatic beta-cells exposed to palmitate. Apoptosis. 2006;11(7):1231-8.

195. Hayatsu H, Arimoto S, Negishi T. Dietary inhibitors of mutagenesis and carcinogenesis. Mutat Res. 1988;202(2):429-46.

196. Maedler K, Oberholzer J, Bucher P, Spinas GA, Donath MY. Monounsaturated fatty acids prevent the deleterious effects of palmitate and high glucose on human pancreatic beta-cell turnover and function. Diabetes. 2003;52(3):726-33.

197. Georgel P, Crozat K, Lauth X, Makrantonaki E, Seltmann H, Sovath S, et al. A toll-like receptor 2-responsive lipid effector pathway protects mammals against skin infections with gram-positive bacteria. Infect Immun. 2005;73(8):4512-21.

198. Deng ZB, Liu Y, Liu C, Xiang X, Wang J, Cheng Z, et al. Immature myeloid cells induced by a high-fat diet contribute to liver inflammation. Hepatology. 2009;50(5):1412-20.

199. Chan KL, Pillon NJ, Sivaloganathan DM, Costford SR, Liu Z, Théret M, et al. Palmitoleate Reverses High Fat-induced Proinflammatory Macrophage
Polarization via AMP-activated Protein Kinase (AMPK). J Biol Chem. 2015;290(27):16979-88.