Role of n-3 Polyunsaturated Fatty Acids and Exercise in Breast Cancer Prevention: Identifying Common Targets

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ABSTRACT: Diet and exercise are recognized as important lifestyle factors that significantly influence breast cancer risk. In particular, dietary n-3 polyunsaturated fatty acids (PUFAs) have been shown to play an important role in breast cancer prevention. Growing evidence also demonstrates a role for exercise in cancer and chronic disease prevention. However, the potential synergistic effect of n-3 PUFA intake and exercise is yet to be determined. This review explores targets for breast cancer prevention that are common between n-3 PUFA intake and exercise and that may be important study outcomes for future research investigating the combined effect of n-3 PUFA intake and exercise. These lines of evidence highlight potential new avenues for research and strategies for breast cancer prevention.

KEYWORDS: breast cancer, breast cancer risk, n-3 PUFA, exercise

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Introduction

Breast cancer is the most common female malignancy in the world with more than one million new cases diagnosed worldwide every year and more than 400,000 deaths.[1] Out of these diagnosed cases, there are more than 2.8 million breast cancer survivors.[2] In 2002, a quarter of cancer cases worldwide were considered a result of excess body weight and physical inactivity.[3] As such, the European Union estimated that 13,000 breast cancer cases could have been avoided by maintaining a healthy body weight through diet and exercise.[3] The World Health Organization stated that obesity is considered a global epidemic, with an estimated 1.9 billion adults older than 18 years being overweight and 600 million being obese in 2014.[4] Obesity has been recognized as a driver of a number of chronic diseases, including coronary artery disease, type II diabetes mellitus (T2D), dyslipidemia, and certain types of cancers. In breast cancer specifically, obesity is associated with breast cancer development and progression.[5]

Germline mutations in the *BRCA1* gene, responsible for DNA repair, are thought to be largely responsible for inherited breast cancer risk.[6] Lubinski et al[7] found higher incidence of breast cancer in Canadian women who are carriers of *BRCA1* mutation vs. Polish *BRCA1* carriers (72% vs. 49% incidence in Canadian and Polish women, respectively). The authors attributed the significant difference in incidence to multiple potential factors, including difference in genetic backgrounds, difference in intensity of screening (more breast cancer cases were detected through screening in Poland compared to North America), and differences in dietary intake habits.[7] Hence, this study demonstrated the impact of environmental factors even in the case of genetic predisposition. Diet and exercise have been recognized as important lifestyle factors contributing to breast cancer risk. Dietary n-3 polyunsaturated fatty acids (PUFAs) have been shown to play an important role in breast cancer prevention.[8] Furthermore, the role of exercise in cancer and chronic disease prevention is widely recognized.[9]

This review will focus on the role of n-3 PUFA and exercise in modulating some of the modifiable factors contributing to breast cancer initiation, development, or progression, identifying common targets between n-3 PUFA intake and exercise training. These factors will be suitable outcomes for investigation in future studies determining the combined effect of n-3 PUFA and exercise on breast cancer prevention.

Modifiable vs. nonmodifiable breast cancer risk factors. Breast cancer risk is influenced by numerous factors, some of which are modifiable, while some are not related to individual's life choices. Nonmodifiable factors altering risk of breast cancer include sex, age, ethnicity, family history, genetic predisposition, age at menarche, age at menopause, breast tissue density, and personal history.[10] Modifiable factors, contributing to breast cancer risk, include number of births, breast feeding, use of hormonal contraceptives and

hormonal replacement therapy, alcohol intake, obesity, and exercise.[10]

The American Institute for Cancer Research estimates that 33% of breast cancer cases can be prevented through meaningful lifestyle changes.[11] Diet and exercise have been recognized as important factors contributing to breast cancer prevention. The importance of exercise is not only specific to prevention of disease development but also extends to reducing treatment-associated side effects such as reduction in physical fitness, negative changes in body composition, increased fatigue, depression, and anxiety.[2]

Types of breast cancer prevention. To date, studies investigating cancer prevention are primarily focused on chemoprevention of the disease using chemically synthesized drugs such as vaccines (eg, human papilloma virus vaccines to prevent cervical cancers) or established chemotherapeutic agents (eg, tamoxifen for prevention of breast cancer recurrence). Chemoprevention, or the eradication of cells that can result in cancer, includes three levels of prevention, namely, primary, secondary, and tertiary. Primary chemoprevention aims to prevent the development of precancerous lesions, while secondary chemoprevention focuses on the prevention of cancer progression. Tertiary chemoprevention aims at preventing cancer recurrence.[12]

Davis and Wu[13] discussed the current use of chemoprevention and the future challenges. The authors argued the need to optimize chemoprevention by targeting at-risk populations, identifying specific targets of chemoprevention such as disease-driving mutations and the importance of combined treatments to reduce or overcome drug resistance.[13] Similarly, these aims can be approached from a *non-chemo* prevention angle. Diet and exercise can be used as cancer-preventing tools in specific at-risk populations of cancer initiation, progression, or recurrence. The use of diet and exercise can be targeted toward specific modifiable markers of disease risk and can be administered in a combinatory fashion to optimize their beneficial effect. Hence, the remainder of this review will identify some of the populations where n-3 PUFA intake and exercise training have been successful in preventing breast cancer development or in modifying markers of breast cancer risk.

This review will highlight studies pertaining to different stages of prevention, such as primary, secondary, or tertiary. It will explore risk factors that have been reported to be subject to the influence of both exercise and n-3 PUFA intake. By doing so, we identify common targets between exercise and n-3 PUFA intake that should be investigated in future studies exploring the combined effect of those two factors. Due to the young nature of the field of exercise oncology, this review has focused on literature generated during the past decade, for the role of exercise and breast cancer, and the past two decades, for the role of n-3 PUFA intake and breast cancer.

Role of Dietary PUFAs

Breast cancer is more prevalent in Western societies compared to Asian populations, but Asian populations tend to have an increased breast cancer risk after migrating to Western countries, possibly as a result of adopting a Western diet.[14,15] This is thought to be due to differences in dietary and consumption of n-3 vs. n-6 PUFAs. Asian populations tend to consume more foods that are high in the n-3 PUFAs eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), such as fish and other marine species. However, consumption of n-6 polyunsaturated fatty acids (n-6 PUFAs) is higher than that of n-3 PUFA in Western countries, likely due to the increased fortification of foods and increased consumption of vegetable oils in recent years.[16] Higher ratio of n-6 to n-3 PUFA may lead to chronic inflammation, as n-6 PUFAs are generally thought of as pro-inflammatory and n-3 PUFAs are viewed as anti-inflammatory.[16] It is thought that eicosanoids, a product of n-6 PUFA, may play a role in tumor promotion, while n-3 PUFAs inhibit pro-inflammatory eicosanoid production, thereby regulating cell growth.[16] It has been speculated that the true benefits of diets rich in n-3 PUFA may not lie in the specific actions of EPA and DHA themselves, but in the decrease in n-6 PUFA to n-3 PUFA ratio, specifically in regard to their incorporation into cellular membranes.[17] Thus, it is not the increase in n-6 PUFA, but the decrease in n-3 PUFA intake. Indeed, Western diets tend to be higher in n-6 PUFA and lower in n-3 PUFA consumption. Studies have demonstrated that lower ratios of n-3 PUFA to n-6 PUFA in both erythrocytes and breast tissue are inversely associated with breast cancer risk.[18] In addition, a study by Goodstine et al[17] found a 41% reduction in breast cancer risk in premenopausal women with very high n-3 to n-6 PUFA ratios, when compared to those with very low n-3 to n-6 PUFA ratios. However, in postmenopausal women, only 11% reduction in breast cancer risk was seen in women with high n-3 to n-6 PUFA ratios, suggesting that high n-3 to n-6 PUFA ratios may be more strongly associated with reduced breast cancer risk in premenopausal women.[17] In contrast to these reports, Gago-Dominguez et al[19] found that high consumption of n-3 fatty acids, specifically from marine species, was associated with a greater reduction in breast cancer risk in postmenopausal women, when compared to premenopausal women.

A growing number of preclinical studies have demonstrated the preventative effects of n-3 PUFA in reducing breast cancer risk. n-3 PUFAs have been shown to exert their effect through multiple mechanisms including modulation of oncogenic protein signaling through disruption of lipid rafts in the plasma membrane.[20] Lipid rafts are rich in sphingolipids and cholesterol and function to enhance protein signaling. They are particularly important for various tyrosine kinase receptors, such as epidermal growth factor receptor (EGFR) and human epidermal growth factor-2 (HEGF-2). These receptors are involved in the upregulation of cell proliferation, thereby contributing to uncontrolled cell proliferation and tumorigenesis.[21,22] It is thought that EPA and DHA may disrupt these lipid raft domains, causing a reduction in the signaling activity of these oncogenic proteins and a decrease

in cell proliferation in both benign and malignant mammary tissues.[21–23] Current preclinical studies are also suggesting that EPA and DHA may also act by increasing the expression of various proteins that are involved in cell cycle control and DNA repair. These proteins include BRCA1, BRCA2, phosphatase and tensin homolog (PTEN), and others.[24] Another mechanism by which n-3 PUFA may exert anticancer effects in breast cancer risk is through lipid peroxidation. Studies suggest that n-3 PUFA incorporation into cell membranes contributes to lipid peroxidation. Accumulated products of lipid peroxidation, in turn, can result in cytostatic or cytotoxic effects that, ultimately, inhibit tumor cell growth.[25–28]

Studies in human breast cancer cell lines have provided mechanistic insight into the anticancer effects of n-3 PUFA and potential insight into its chemopreventive properties. Treatment with EPA and DHA has been found to inhibit the growth of human breast cancer cells by at least 48%.[29,30] The mechanism of this inhibitory effect involves a significant decrease in lipid raft sphingomyelin, cholesterol, and diacylglycerol levels in the presence of EPA and DHA.[29] EPA and DHA have also been found to decrease the expression and increase phosphorylation of EGFR in lipid rafts.[29] Prolonged activation of EGFR by phosphorylation has been associated with apoptosis in human breast cancer cell lines, thereby decreasing net cell proliferation and hindering tumor growth.[29] Similar effects have been observed in other studies demonstrating increased cell death, inhibition of cell growth, and cell proliferation by reducing the n-3 to n-6 PUFA ratio.[30,31]

Studies in rodents have found direct evidence for the anticancer effects of n-3 PUFA in mammary tumorigenesis.[32,33] An aggressive breast cancer model, expressing the neu gene driven by the mouse mammary tumor virus (MMTV) promoter and maintained on diets supplemented with n-3 PUFA, displayed significant reductions in tumor volume and multiplicity.[32,33] Studies also found a dose-dependent effect of n-3 PUFA on mammary tumor outcomes.[33] When the mammary gland tissue of these mice was examined, differential incorporation of EPA and DHA were observed in mammary gland tissue and preferential incorporation of DHA was observed in tumors. Leslie et al[33] examined whether n-3 PUFA intake would reduce mammary gland tumors in female MMTV-neu (ndl)-YD5 mice in a dose-dependent manner. Consuming n-3 PUFA from fish oil for 20 weeks at 3% or 9% w/w resulted in a significant decrease in tumor burden, compared to 10% w/w fat from n-6 PUFA.[33] The study suggested that n-3 PUFA may exert its antitumorigenic effect by incorporating into the cell plasma membrane and ultimately modifying protein–protein interactions.[33] Although the majority of studies conducted on this topic suggest an inverse relationship between serum and breast tissue EPA and DHA with breast cancer risk, there have been a few studies that have reported no effect.[34–37]

Several human clinical studies have examined the effect of diets rich in n-3 PUFA on breast cancer risk. Inverse associations between breast cancer risk and women who consume diets high in n-3 PUFA from marine sources (eg, fatty fish and shellfish) have been reported.[19,38] In addition, several case–control studies in humans have examined the effects of long-term n-3 PUFA consumption.[18,37,39,40] These studies validated n-3 PUFA consumption using food frequency questionnaires and by examining the fatty acid composition of erythrocytes and breast tissue.[18,37,39,40] Elevated erythrocyte EPA and DHA were associated with significantly lower risk and incidence of breast cancer.[39] Similar results have been reported when examining the total fatty acid composition of breast adipose tissue. Increased incorporation of n-3 PUFA into breast adipose was associated with reduced breast cancer risk.[18,37] Shannon et al[40] found an inverse association between total n-3 PUFA and breast cancer risk, but reported a more significant impact for EPA. Recently, the Japan Public Health Center-based prospective study investigated breast cancer incidence in 38,234 Japanese women, aged 45–74 years, during 14.5 years follow-up.[41] The study found a positive association between n-6 PUFA intake and estrogen and progesterone receptor positive (ER+PR+) tumor development and a negative association between EPA intake and ER+PR+ breast cancer incidence.[41] A Swedish women lifestyle and health cohort study found that women in the highest quintile of PUFA intake had decreased breast cancer incidence compared to women in the lowest quintile.[42] However, the effect of PUFA intake was not associated with estrogen receptor (ER) or progesterone receptor (PR) status.[42] Similarly, other studies have found significant risk reduction with increasing DHA concentrations.[43]

In addition to the anticancer effects of EPA and DHA, a case–control study by Maillard et al[18] found that adipose tissue concentrations of alpha-linolenic acid (ALA), a plant-based n-3 PUFA, along with DHA, were associated with decreased risk of breast cancer. To date, the majority of studies have examined the effects of marine n-3 PUFA containing EPA and DHA. Nevertheless, these findings suggest that the effect of plant-based n-3 PUFA on breast cancer risk should also be examined.

Role of Exercise

It is estimated that 7.9% of Canadian cancer cases (breast, colon, endometrium, prostate, lung, and ovarian) was associated with physical inactivity, demonstrating that thousands of cancer cases can be prevented by following a healthy lifestyle.[44] Gonçalves et al[45] analyzed 14 case–control and 7 cohort studies investigating breast cancer prevention via exercise. With the exception of one study that did not find a significant association, physical activity was associated with reduced breast cancer incidence in postmenopausal women.[45] This was attributed to exercise modulating metabolic and sex hormone status, growth factors, and adiposity.[45] One study examined the effects of exercise in 1504 women and found that women in the third quartile of exercise experienced a

30% reduction in risk.[46] This is consistent with previous studies reporting an average of 25% breast cancer risk reduction resulting from exercise.[46,47] These results are likely due to the positive impact exercise has on energy balance and obesity-related implications, including insulin resistance and chronic inflammation.[45,47] Lipid peroxidation and stress-induced apoptosis are suggested to be among the mechanisms by which exercise protect against breast cancer.[19] In addition, exercise also aids in assuaging symptoms of menopause and improving quality of life.[45]

To date, several reviews have discussed the benefits of exercise after breast cancer diagnosis and treatment.[2,48] Benefits of exercise in breast cancer survivors include improvements in peak oxygen consumption, functional capacity, muscle strength, lean mass, cardiovascular risk factors, and bone health.[48] Decreased peak oxygen consumption has been associated with decreased overall health and increased mortality. There is also evidence of a relationship between peak oxygen consumption and risk of breast cancer-related death.[49] Before and during treatment for breast cancer, peak oxygen consumption decreases significantly and remains low following the completion of treatment when compared to sedentary controls.[49] During and following breast cancer treatment, aerobic exercise training is an effective way to maintain or improve peak oxygen consumption, helping to decrease overall risk of mortality.[50] Also in relation to cardiorespiratory risk factors associated with breast cancer, breast cancer diagnosis has been found to correlate with higher total cholesterol, triglyceride, and low density lipoprotein (LDL) levels, resulting in increased risk of cardiovascular disease as a result.[48] However, the effects of exercise on these risk factors associated with breast cancer have been shown to be conflicting, and more research is needed to determine the exact effects of exercise on these breast cancer-related outcomes.[51] The types of exercise regimes studied include moderate level aerobic training, interval cardio training, resistance exercise, and yoga.[52]

Obesity

In modern society, obesity, due in part to physical inactivity and sedentary lifestyles, continues to increase, but is a readily modifiable risk factor of breast cancer.[53] Obesity results in hypertrophy and hyperplasia of adipocytes, causing hypoxia.[47] It also results in an increase in macrophage infiltration, leading to whole body inflammation and irregular adipokine secretion.[47] Obesity-accelerated breast cancer progression may be mediated through a number of mechanisms including increased subcutaneous mammary adipose tissue inflammation, resulting in inflammatory tumor microenvironment, adipokine secretion and AKT/mammalian target of rapamycin (mTOR) activation, hyperinsulinemia, and upregulated estrogen signaling, and increase in local estrogen production.[54–56]

Obesity is an important factor contributing to breast cancer development and progression, as a result of inflammatory conditions and hormonal irregularities. Exercise and n-3 PUFA have been implicated in modulating both inflammatory responses and hormones. The effects of n-3 PUFA intake and exercise on some obesity-associated breast cancer risk factors, such as excess body weight, hormones, and inflammation, are discussed in the following sections.

Excess weight. The Canadian Study of Diet, Lifestyle, and Health determined that weight gain in adulthood is positively correlated with risk of postmenopausal breast cancer with a 6% increase for every 5 kg of weight gained after the age of 20 years.[53] Weight gain, however, is common in postmenopausal women, as basal metabolic rate (BMR) and lean body mass decrease with age.

Weight gain is prevalent following diagnosis of breast cancer during chemotherapy, radiation, and hormonal therapy.[57–59] Weight gain has been attributed to a decrease in energy expenditure (EE), as most studies report women being less active after diagnosis, and energy intake may actually decrease during the first year due to psychological factors.[57] Average weight gain in women with breast cancer following treatment is 1–5 kg and is further influenced by age, menopausal status, and comorbidities.[57] Excessive weight gain after diagnosis has been linked with a poorer prognosis and increased mortality rates.[57]

Role of PUFA in weight gain. In human subjects, studies have found that serum biomarkers of inflammation and breast cancer risk, including hormones, adipokines, and cytokines, appear to be unaffected by supplementation with n-3 PUFA.[60–62] Recently, a preclinical animal study found that a high-fat diet containing n-3 PUFA compared to high-fat diet containing lard resulted in lower weight, lipid gain, and energy efficiency in the n-3 PUFA-fed group.[63] PUFA intake also resulted in improved insulin signaling and modulation of mitochondrial function.[63]

Interesting and conflicting results have been noted in regard to the interaction between obesity and EPA and DHA on breast cancer risk.[62,64] Yee et al[62] reported that increased body mass index (BMI) attenuated the dose–response benefits of EPA and DHA supplementation, as seen in serum EPA and DHA levels, as well as breast adipose DHA levels. Contrary to this, a study conducted by Sandhu et al[64] reported that only participants with BMI > 29 experienced an increase in plasma DHA, which was associated with a decrease in absolute breast density, thereby contributing to decreased breast cancer risk. Krishnan and Cooper[65] compared human studies investigating diet-induced thermogenesis (DIT), EE, or fat oxidation (FOx) after a high-fat meal. The authors found that monounsaturated fatty acids and PUFA are more metabolically beneficial by inducing greater EE, DIT, and FOx compared to saturated fats.[65]

Role of exercise in weight gain. It has been observed that postmenopausal women gain body weight and total body fat 3 years after breast cancer diagnosis.[66] Although the exact mechanisms are not confirmed, physical inactivity and age-related factors are likely major contributors to this increase

in body weight.[66] This observation may be indicative of an increased risk in recurrence of breast cancer in survivors. The Yale exercise and survivorship (YES) study targeted this finding and created a exercise intervention where 80% of the women reached the goal of 120 minutes/week at the end of the trial.[66] Women showed better adherence to exercise as they noticed improvements in BMI and a decrease in waist circumference.[66] A Canadian cohort study of 3320 women found a significant positive association between weight gain as an adult and postmenopausal breast cancer risk (6% for every 5 kg gained since the age of 20 years).[53] The study also found a 21% decrease in breast cancer risk in women exercising 30.9 metabolic equivalent task (MET) hours per week compared to women exercising 3.0 MET hours/week.[53] The muscle mass, omega-3, diet, exercise and lifestyle (MODEL) study noted that the loss of lean body mass and gain of fat mass, even in the absence of weight gain, can put breast cancer women at risk for cardiovascular and metabolic diseases.[67]

Interaction of diet and exercise in weight gain. A relationship between diet and exercise in weight gain with regard to breast cancer risk has also been observed in previous studies. McDonald et al[67] discussed the interaction between long-chain n-3 PUFA and anabolic resistance training. Long-chain n-3 PUFAs have been thought to preserve lean body mass by favoring protein synthesis, reducing anabolic resistance, and stimulate nerve activation to working skeletal muscle, along with their role as an anti-inflammatory mediator.[67] Cantarero-Villanueva et al[68] analyzed subcutaneous adipose tissue biopsies in 45 women during a six-month diet and exercise intervention. Mean weight loss was 7.9 kg, which resulted in decreased leptin levels and increased insulin-like growth factor-binding protein 3.[68] This study determined that diet and exercise intervention positively influenced steroid hormone metabolism, as well as, leptin and insulin signaling, which are two pathways that may mechanistically link obesity with cancer.[68] The sex hormones and physical exercise-2 (SHAPE-2) trial tested the effect of diet (calorie restriction) and exercise on weight loss and sex hormones in a total of 243 women (randomized to diet, exercise, or control) who were both overweight and insufficiently active.[69] The authors reported significant weight loss by both diet and exercise groups; however, exercise treatment resulted in a greater beneficial effect, compared to diet, on estradiol, free estradiol, sex hormone-binding globulin, and testosterone.[69]

Inflammation. It is well accepted that chronic low-grade inflammation is a result of excess adipose tissue and plays a role in the development of metabolic diseases and breast cancer development and progression. In a preclinical model, using the MMTV-polyoma middle T oncoprotein (PyMT) murine model, Cowen et al[70] demonstrated that high-fat, high-calorie intake results in mammary adipose tissue inflammation. The MMTV-PyMT murine model carries the PyMT, along with the MMTV promoter, which drives mammary tissue self-expression and tumor formation.[70,71] Inflammation was associated with increased macrophage infiltration, elevated plasma levels of monocyte chemoattractant protein-1, and increased leptin and pro-inflammatory cytokine concentrations.[70]

Chronic overnutrition in overweight and obese individuals leads to insulin resistance, pro-inflammatory macrophages, and organelle dysfunction causing unfolded protein response (UPR).[72] UPR induction by tumor necrosis factor alpha (TNF-α), via production of reactive oxygen species, leads to endoplasmic reticulum stress, which is the site for protein synthesis and lipid droplet formation.[72] TNF-α has been extensively studied and plays an important role in insulin resistance by promoting serine phosphorylation of insulin receptor substrate (IRS)-1, thereby blocking insulin signaling.[72] TNF-α also inhibits adiponectin, which is responsible for decreasing inflammation, improving insulin sensitivity, and increasing FOx.[72] Another marker of inflammation is cyclooxygenase-2 (COX-2) expression. COX-2 expression has been associated with increased tumor size and other factors in aggressive breast cancer cases.[73] Rodent studies have found that moderate-to-high COX-2 expression is linked to mammary tumorigenesis.[73] The mechanism for this link likely involves dysregulation of breast tissue apoptosis.[73]

Nuclear factor peroxisome proliferator-activated receptor gamma (PPAR-γ) plays a role in breast cancer development through its anti-inflammatory properties. Studies have reported conflicting results regarding the role of PPAR-γ in breast cancer progression.[74,75] Nakles et al[74] reported that increased activation of PPAR-γ is related to tumor suppression in noninvasive cancers. This is also supported by studies in cultured breast cancer cells, where PPAR-γ function was suppressed.[75] Contrary to these reports, studies have found that activation of PPAR-γ inhibited cell proliferation, induced apoptosis, and promoted differentiation.[75]

Chronic inflammation has been shown to play a key role in breast cancer development and progression by assisting tumor growth and metastasis.[76] Park and Kang[76] measured interleukin-6 (IL-6), interferon-γ (IFN-γ), and C-reactive protein linked upregulation of inflammatory cytokines with increased breast cancer incidence, more advanced stages, and increased mortality rate. In support of these findings, Irahara et al[77] found increased expression of TNF-α, IL-6, and COX-2 in breast tumor tissue. Their study demonstrated that cytokines play a role in the upregulation of aromatase to enhance estrogen biosynthesis, causing an increase in ER+ breast cancer tumors.[77]

Role of PUFA in inflammation. EPA and DHA are of particular interest in the context of inflammation as they are involved in the production of anti-inflammatory eicosanoids and inflammation-resolving mediators, namely, resolvins. Through the reduction in pro-inflammatory eicosanoids and cytokines, these n-3 PUFAs are able to reduce inflammation and thereby decrease overall breast cancer risk. In rats, it has been found that COX-2 protein levels were approximately three times higher than COX-1 levels in mammary tumors.[78] Rats fed a diet high in n-3 PUFA showed significant suppression of both COX-1 and COX-2 protein levels in mammary tissue.[78]

Another key finding in cultured breast cancer cells treated with EPA or DHA involves the expression and activation of nuclear factor PPAR-γ. Rovito et al[79] found that treatment of cells with EPA and DHA lead to the enhanced protein and mRNA expression of PPAR-γ. Enhanced PPAR-γ expression resulted in upregulation of autophagy and reduction in cell proliferation.[79,80] Decreasing cell proliferation is a key mechanism by which n-3 PUFA reduces breast cancer risk. n-3 PUFA activates G-protein coupled receptor 120 and PPAR-γ, which downregulates nuclear factor κB (NFκB) signaling and translocation and, in turn, results in a decrease in cell proliferation.[79]

Role of exercise in inflammation. The immunomodulatory role of exercise, in cancer initiation and progression, has been extensively reviewed by Koelwyn et al.[81] Briefly, studies investigating the effect of exercise on pro-inflammatory effectors in cancer found that exercise intervention in cancer patients with advanced disease resulted in decreased circulating IL-1b, IL-2, IL-4, macrophage inflammatory protein-1b (MIP-1b), and TNF-α.[81–83] Studies examining the role of exercise in innate immune surveillance, orchestrated by natural killer cells (NK cells), found that exercise intervention increased NK cell cytotoxic activity in postmenopausal breast cancer patients who completed primary adjuvant therapy.[81,84] However, findings pertaining to the effects of exercise on NK cell function were not consistent between studies possibly because of differences in exercise dose and heterogeneous patient populations as well as differences in methodological assays used to determine NK cell function.[85] Studies examining the effect of exercise on intratumoral T-cell composition are few. Preclinical studies demonstrated that exercise intervention resulted in increased production of INF-γ, IL-2, IL-12, and TNF-α and decreased expression of IL-4, IL-10, and transforming growth factor-β (TGF-β).[81,86] In patients with solid tumors, exercise interventions resulted in changes in circulating T-lymphocyte populations.[81,87,88] In a preclinical mouse model, a combination of tamoxifen and interval exercise training resulted in a significant reduction of tumor IL-6, NFκB and signal transducer and activator of transcription 3 (STAT3) expression and upregulation of tropomyosin 1 (TPM1), and programmed cell death 4 (PDCD4) expressions through a mechanism involving microRNA, miR-21.[89]

Rogers et al[90] conducted an exercise intervention in breast cancer survivors and examined pro-inflammatory cytokine levels. Cardiorespiratory fitness, muscle strength, body composition, and fatigue significantly improved; however, reduction in systemic inflammation was small or insignificant. IL-6 slightly increased after the intervention, which may be imparted due to IL-6 stimulating AMP-activated protein kinase (AMPK) during exercise to promote FOx and hepatic glucose production. However, increased IL-6 in a rested state in obese individuals activates the Janus kinase/signal transducer and activator of transcription pathway (JAK/STAT pathway), impairing insulin signaling.[90] More recently, a randomized controlled trial (RCT) with breast cancer survivors found that supervised resistance training performed three times a week resulted in significant reductions in numbers of NK cell and NK T-cell expression of TNF-α. In addition, significant improvements were achieved in regard to measurements of strength.[91]

Hormones. Breast cancer risk factors that are modifiable through diet and exercise include circulating insulin, estrogen, leptin, and adiponectin levels.[92] Obese individuals may become insulin, leptin, and adiponectin resistant, which cause inflammation, insulin resistance, and decreased FOx.[47] However, these processes are reversible through exercise, which can improve insulin-stimulated glucose uptake and leptin-stimulated FOx.[47] The following section discusses studies demonstrating the effects of exercise and n-3 PUFA intake on insulin, estrogen, and leptin levels. As factors that influence breast cancer risk, insulin, estrogen, and leptin levels may be potential common targets through which exercise and n-3 PUFA may influence breast cancer risk. More studies are needed to determine the combinatory effect of exercise and n-3 PUFA intake on those markers in relation to breast cancer development or progression.

Insulin. Higher breast cancer incidence and mortality have been observed in individuals with insulin resistance and T2D.[3] Studies suggested that 16%–20% of women with breast cancer have been diagnosed with T2D.[93] Some studies have linked insulin to increased breast cancer cell proliferation or decreased apoptosis; however, the mechanisms by which insulin exerts pro-cancerous effects are unclear.[3] Elevated insulin levels resulted in hyperinsulinemia, upregulated synthesis of sex hormones, and downregulation of sex hormone-binding proteins, which resulted in increased circulating sex hormones and increased breast cancer risk.[3,94,95] Sieri et al[94] also noted that hyperglycemia and hyperinsulinemia increased tumor cell growth and that fasting glucose levels were positively associated with breast cancer in pre- and postmenopausal women.

The health, eating, activity, and lifestyle (HEAL) study examined insulin resistance and adiponectin levels in 527 women diagnosed with stage I–IIIA breast cancer.[95] High homeostasis model assessment (HOMA) scores and low levels of adiponectin were associated with obesity, as well as breast cancer mortality.[95] The women's healthy eating and living (WHEL) study examined 3003 early-stage breast cancer survivors and the association with T2D.[93] Women with hyperglycemia (>6.5% HbA1C) were more likely to be obese and have more advanced breast cancer. Women with >7.0% HbA1C had a 26% increased rate of breast cancer progression than women with <6.5% HbA1C.[93] A case–control study found that insulin resistance did not have an effect on breast cancer risk; however, increased HbA1C levels were associated with increased breast cancer risk.[96] Collectively, these findings demonstrated the relevance of targeting insulin signaling and function as markers of breast cancer risk.

Role of n-3 PUFA in insulin function. The effect of n-3 PUFA on insulin levels or function in breast cancer patients

or in relation to breast cancer risk has not been extensively investigated. A recent preclinical study found that feeding mice with high-fat diet containing fish oil compared to high-fat diets containing soybean oil, oleic sunflower oil, or flaxseed oil diet resulted in increased adiponectin and decreased leptin concentrations in plasma and adipose tissue.[97] Although the study found no effect on insulin levels, it demonstrated a beneficial effect of n-3 PUFA on insulin resistance.[97] A three-month randomized placebo-controlled trial investigated the effect of calorie-restriction with or without n-3 PUFA intake in 48 obese subjects.[98] The study found that caloric restriction with EPA–DHA supplementation had a beneficial effect on insulin resistance.[98]

Role of exercise in insulin function. The effect of exercise on insulin levels or function and cancer is also not yet clear. Löf et al[99] conducted a systemic review of 12 RCTs on the effect of exercise on multiple biomarkers of breast cancer risk including insulin. The authors found that although five RCTs reported a significant effect of exercise on insulin and insulin growth factors, the results were inconsistent.[99]

It has been well documented that high HbA1C, insulin resistance, and T2D are associated with breast cancer risk and a poorer prognosis. It is also known that these metabolic conditions are linked to obesity, but can be reversed by diet and exercise. Studies report that weight gain after diagnosis is typically not excessive but may lead to metabolic dysfunction.[57] Guinan et al[58] identified increased fasting glucose levels and HbA1c from baseline to follow-up in women undergoing adjuvant breast cancer treatment. These women were more likely to develop insulin resistance and metabolic dysfunction.[58] Whether increased body mass is present or not, breast cancer patients are prone to developing sarcopenic obesity, whereby fat mass is increased and lean body mass is decreased. In addition, the increase in fat mass is predominately visceral, resulting in an increased risk for metabolic complications.[57] Since exercise may play a role in insulin signaling and function, and based on the aforementioned studies on susceptibility of breast cancer patients to metabolic complications, future research should investigate the effect of exercise and n-3 PUFA intake on insulin function.

Estrogen. Obesity results in increased adipose tissue mass and volume, which is a major site for sex hormone production. Weight gain is more prevalent in postmenopausal women, leading to an increase in visceral adipose tissue and an increased ability for sudden estrogen production. Increased levels of circulating estrogen have been linked to increased breast cancer risk, which is thought to accelerate breast tumor cell growth.[77,100] Testosterone and other androgens are converted to estrogen in breast cancer tissue by aromatase, and upregulation of aromatase mRNA led to increased production of estrogen in breast cancer tumor tissue.[77] Irahara et al[77] measured aromatase mRNA levels in breast tumor tissue. Aromatase mRNA levels were found to be upregulated in breast cancer patients, when compared to the control.[77]

Mirtavoos-Mahyari et al[100] measured ER status in breast tumor cells and its role in activating tyrosine kinase human epidermal growth factor receptor 2 (HER2). A total of 67% of breast cancer subjects had ER+ tumors and 31% had tumors overexpressing HER2.[100] Similarly, Rodrigue et al[101] found that 61% of the breast cancer subjects had ER+ tumors. They also measured PR status and found the majority of breast cancer subjects to be ER+/PR+.[101] These studies attributed hormone receptor-positive breast cancer, which led to higher steroid hormone responsiveness, higher BMI, and increased body fat,[100,101] while ER–/PR– breast cancer was attributed to having a family history of breast cancer.[101] Overall, more than 50% of the subjects were ER+, which is an avoidable subtype of breast cancer. The high prevalence of ER+ breast cancer and the association of this subtype with higher body fat indicate that maintaining a healthy body weight can significantly decrease the incidence of breast cancer globally.

Role of n-3 PUFA in estrogen levels. Falavigna et al[92] determined that an increased BMI can double the risk of breast cancer recurrence within 5 years and result in a 60% increase in breast cancer mortality within 10 years. However, this negative effect seems to be reversible with lifestyle interventions where it was noted that a restriction of dietary fat to 18%–25% of total caloric intake can result in a reduction in serum estrogen levels in both pre- and postmenopausal women.[92]

In preclinical breast cancer models, EPA and DHA have been shown to inhibit the growth of estrogen-dependent MCF-7 breast cancer cells, by influencing the effects of estrogen on breast cancer development.[30] This suggests that n-3 PUFA may act differently in women with different estrogen levels, ie, pre- vs. postmenopausal women. EPA and DHA treatment of MCF-7 cells shifted estrogen's pro-survival effect to a pro-apoptotic effect by altering estrogen signaling response.[102] Hilakivi-Clarke et al[103] investigated whether n-3 PUFA intake during pregnancy alters pregnancy estrogen levels and breast cancer incidence in offspring. The authors found that n-3 PUFA intake during pregnancy increased circulating estrogen levels in pregnant rats and reduced development of carcinogen-induced tumors in offspring.[103] A population-based case–control study found that n-3 PUFA intake was inversely associated with breast cancer incidence in obese Mexican women, but neither in normal weight nor in overweight women.[104] The authors suggested that the effects may be a result of n-3 PUFA altering inflammatory status as well as adipokine and estrogen levels in women's adipose tissue.[104]

Role of exercise in estrogen levels. A systemic review of 23 RCTs (total of 3239 subjects) reported that exercise resulted in statistically significant reduction in both total estradiol and free estradiol.[105] The Breast Cancer and Exercise Trial in Alberta randomized 400 inactive women, aged 50–74 years with BMI 22–40 kg/m^2, to 12 months of high (300 minutes/week) and moderate (150 minutes/week) volumes of exercise.[106] Per-protocol analysis found a small, but significant, decrease in

total and free estrogen in the high volume group.[107] A 12-week diet and exercise intervention pilot study in 7 healthy obese postmenopausal women found reductions, compared to baseline, in serum levels of estradiol (–25%), leptin (–36%), estrone sulfate (–10%), and IL-6 (–33%).[108] Estrogen levels were found to be inversely associated with exercise measured by accelerometers in 37 obese Latina adolescents (15.7 ± 1.1 years).[109] Thus, the beneficial effects of exercise on estrogen levels may extend beyond postmenopausal women to younger women. Few studies have examined the combinatory effect of diet and exercise. A blind 12-month randomized controlled trial RCT from 2005 to 2009 investigated the individual and combinatory effects of calorie-restricted diet and moderate-to-vigorous exercise on hormone levels of overweight and obese postmenopausal women (439 subjects aged 50–75 years). Greater estrogen reduction was achieved by diet and exercise as a result of greater weight loss.[110]

Leptin. Leptin is secreted from adipose tissue and plays an important role in food intake and EE; hence, it is an important regulator of adiposity as well as breast cancer development and progression.[111,112] Leptin appears to play an important role in mammary carcinogenesis by contributing to the proinflammatory microenvironment in obese patients.[1] Using a preclinical rat model of breast cancer progression driven by diet-induced obesity, Chang et al[113] demonstrated a transcriptional role for leptin in obesity-induced breast cancer progression. The authors found that leptin regulated a transcriptional pathway involving STAT3 and G9a histone methyltransferase, which promotes the formation of breast cancer stem cell-like cells through the epigentic silencing of miR-200c.

Role of n-3 PUFA in leptin levels. The role of PUFA in leptin production and signaling has been recently reviewed by Monk et al.[54] Briefly, n-3 PUFAs were shown to downregulate leptin receptor gene expression,[114] and disrupt lipid raft composition,[115] to which leptin receptors localize and induce leptin-mediated proliferative signaling.[116] Mice consuming high fish oil diet had lower leptin concentrations in plasma and adipose tissue.[97]

Role of exercise in leptin levels. The role of exercise on leptin secretion, signaling, and breast cancer progression is not clear and has been recently reviewed by Schmidt et al.[117] C57BL6 male mice, following a 12-week voluntary exercise training program, had significantly enhanced metabolic symptoms associated with high-fat diet and improved hypothalamic leptin signaling.[118] Recently, the SHAPE-2 trial, a 16-week RCT of 243 females, investigated the effect of weight loss by hypocaloric diet or exercise on serum high-sensitivity C-reactive protein (hsCRP) and leptin levels.[119] Weight loss through diet or exercise resulted in lower serum hsCRP and leptin.[119]

Epigenetics

Epigenetic alterations have been recognized as a key player in carcinogenesis.[120] Epigenetic changes include methylation of CpG islands in promoter regions or acetylation of chromatin and histones, leading to changes in gene expression.[121] Epigenetic alterations also include the posttranscriptional modification of gene expression through the action of microRNA and RNA interference (RNAi).[122] These epigenetic modifications of DNA can result in silencing of tumor suppressor genes or activation of oncogenes,[123] hence altering susceptibility to cancer. Thus, epigenetic changes as potential markers of cancer risk and prognosis represent novel targets for further study.[124]

Role of n-3 PUFA in epigenetics. Studies examining the effect of n-3 PUFA on epigenetic changes are in their infancy. Nonetheless, studies have demonstrated that n-3 PUFA intake can result in changes in DNA methylation status, which suggests that n-3 PUFA intake may influence cancer risk through modifications of the epigenome.[125] Several preclinical animal studies have demonstrated the role of maternal n-3 PUFA intake during pregnancy and lactation and its role in offspring susceptibility to breast cancer.[32,33,126,127] These cancer-protective effects of n-3 PUFA are very likely to be driven by changes at the epigenetic level. Emerging evidence has associated n-3 PUFA with epigenetic modification in colorectal cancer.[128–131] However, the exact mechanisms by which n-3 PUFA epigenetically modifies susceptibility to breast cancer are not yet known. A cross-sectional study in 69 Greek preadolescents found that dietary fat intake, including intake of PUFA, significantly correlated with CpG island methylation levels.[132] The study found that some of the pathways influenced by changes in methylation included the leptin pathway.[132] Building on this notion, another study in obese mice found an increase in binding of methyl-CpG-binding domain protein 2 and DNA methyltransferases at the leptin promoter and a decrease in RNA polymerase II.[133] In addition to these findings, the authors determined that lysine 4 of histone H3 was hypomethylated, resulting in increased binding of histone deacetylases 1, 2, and 6 at the leptin promoter in these mice.[133] These findings suggest that breast cancer risk may be epigenetically modified through the leptin pathway, which is thought to contribute to mammary carcinogenesis. A recent review identified a potential link between n-3 PUFA and epigenetic changes related to obesity, mediated through altered methylation of genes involved in lipid metabolism and RNAi.[134] The role of n-3 PUFA intake in breast cancer-related changes to the epigenome is still an untapped area of research that could potentially shed light on an important mechanism by which n-3 PUFA protects against breast cancer.

Role of exercise in epigenetics. Exercise has also been shown to modulate epigenetic changes. Mechanisms modulating exercise effect on epigenetic status include DNA promoter methylation, histone-postranslational modifications, and microRNA expression.[135] A review by Voisin et al[136] discussed the reported literature pertaining to exercise and its epigenetic influence and concluded that both acute and chronic exercise significantly alter DNA methylation in both tissue- and gene-specific manner.

In a preclinical study, Rossi et al[137] achieved diet-induced obesity in overiectomized female C57BL/6 mice that were subsequently switched to a control diet, resulting in mice losing their obese phenotype and returning to normal weight. After injection of mice with MMTV-Wnt-1 mouse mammary tumor cells, the authors found that tumor volume, IL-6 serum levels, pro-inflammatory genes in the mammary fat pad, and mammary DNA hypermethylation was similar in formerly obese and diet-induced obese mice.[137] The authors concluded that weight loss may not adequately reverse epigenetic changes in tumor microenvironment.[137] Thus, based on findings from this particular study, exercise may not directly result in epigenetic changes that are important for secondary or tertiary prevention; however, it is plausible that exercise may induce epigenetic changes that aid in primary prevention. In support of this hypothesis, researchers using (C57BL/6) female mice explored the effect of exercise (swimming) before and during pregnancy on offspring susceptibility to obesity. In addition to gaining less weight, the offspring had increased adiponectin expression in skeletal muscle, decreased leptin levels, and increased insulin sensitivity.[138] However, it should be noted that depending on exercise protocol used, animals may experience different levels of stress that may differentially influence study outcomes. Thus, maternal exercise may influence many factors that contribute to offspring breast cancer risk. Interestingly, using male C57BL6 mice, a study investigated the effect of paternal exercise on offspring's susceptibility to insulin resistance.[139] The study found that subjecting fathers to 12-week wheel running resulted in offspring that were more at risk of weight gain and adiposity, impaired glucose tolerance, and increased insulin levels.[139] These inconsistent findings may suggest that the source of exercise (maternal vs. paternal) may play a role in defining the direction of effect (beneficial vs. detrimental); however, a common exercise protocol that produces consistent findings must be established before any concrete conclusions can be drawn. Although the two aforementioned studies did not directly link the effects of maternal and paternal exercise to epigenetic changes, the role of epigenetics cannot be excluded as an underlying mechanism by which exercise alter susceptibility to chronic disease and cancer in offspring. In that regard, the effect of a three-month exercise intervention on epigenetic changes on human male reproductive organ resulted in genome-wide changes in sperm DNA methylation.[140] The influence of exercise on epigenetic changes has been found in other tissues as well. A six-month exercise intervention in 23 healthy men with a history of low-level exercise undercovered a genome-wide pattern of DNA methylation in subjects' adipose tissue.[141]

Challenges and Considerations

Although there is growing evidence supporting the use of dietary n-3 PUFA and exercise for human breast cancer prevention, there are caveats that require further scrutiny. The question of dose and duration of n-3 PUFA intake or exercise training warrants more research to specify and define these parameters. The stage at which prevention is applied adds another layer of complexity, as dosage of exercise or n-3 PUFA intake will likely differ for primary vs. secondary or tertiary breast cancer prevention. So, what is the optimum dose for n-3 PUFA intake for maximum effect? What is the best period and optimum duration of intake for cancer prevention? Should intake commence during *in utero* development, childhood, puberty, premenopause, or postmenopause? Preclinical studies have demonstrated the importance of n-3 PUFA intake during extensive mammary gland modeling and remodeling periods, which include in utero, puberty, and pregnancy, for breast cancer prevention.[32,33,127,142–144] However, the optimum time and length of n-3 PUFA intake in humans are still not clear. In terms of dosage, human intervention studies have shown promising results in both pre- and postmenopausal women. Studies supplementing with as little as 0.84 g/day EPA + DHA and up to 7.56 g/day EPA + DHA have been well tolerated, leading to increased serum and breast adipose tissue EPA and DHA.[60,62,64] However, more studies are warranted to identify optimum daily intake values for maximum breast cancer risk reduction for each stage of prevention.

Similarly, optimum duration and dose of exercise training for cancer prevention are yet to be determined. The Breast Cancer and Exercise Trial was a 1-year long trial of 400 inactive postmenopausal women, with BMI between 22 kg/m^2 and 40 kg/m^2, placed in 150 min/wk (moderate-volume) or 300 min/wk (high volume) aerobic exercise programs.[107] High-volume exercise was shown to be superior to moderate exercise in reducing total fat and subcutaneous fat especially in obese women.[107] In concordance, a 150-minute moderate-intensity aerobic exercise intervention per week did not result in significant improvement in physical functioning in women with metastatic cancer.[145] The discrepancy between studies demonstrates the differential effect of exercise at different levels of intensity, as well as prevention stage, which underscores the need for determining the optimum dose, duration of exercise, and timing for breast cancer prevention.

Moreover, exercise training is met with an additional set of challenges in secondary and tertiary prevention as patients are often suffering from treatment side effects, including fatigue. A large subset (50%) of breast cancer survivors taking aromatase inhibitors (AIs), as part of the Hormones and Physical Exercise (HOPE) Study, suffer from AI-associated joint pain that negatively affect participants' exercise adherence.[146] A recent report found that specific exercise regiments can improve AI-associated arthralgia in breast cancer survivors.[146] Another report found that low-minority recruitment to cancer trials is possibly due to self-reported barriers such as fatigue, family responsibilities, illness, transportation, and negative perception of exercise and diet.[147] The issue of minority inclusion in exercise and treatment trials is of special importance as understanding disease development or prevention requires a strong grasp of ethnically specific molecular, physiological,

and pathological factors. In a recent commentary of the literature, Coughlin and Smith[148] found that increased body fat may be associated with increased breast cancer risk as a result of insulinemia and changes in adipokine and estrogen. The authors note the importance of including women from minority groups (eg, African-American and Hispanic), as these subsets of the population are not well represented in a previous study.[148] Inclusion of ethnically diverse subjects is needed to better define parameters of exercise use to mitigate breast cancer risk.

The challenge of subjects' participation extends beyond clinical studies to patients own exercise training, and the reason may be lack of consistent messaging from medical specialists. A recent report by Nyrop et al[149] found that despite the high number of clinical visits that oncology providers have per month (361 visits per 55 providers) and national guidelines recommending that cancer patients engage in regular exercise, only 35% of these encounters included communications about exercise. The authors concluded that although exercise communication by oncology providers is feasible, the frequency is variable between providers, and thus, strategies are needed to make exercise recommendations by oncologist to patients more frequent and consistent.[149]

Adoption of n-3 PUFA intake and exercise training in daily lifestyle for the prevention of cancer is challenging as discussed above. However, as more studies are conducted, more information is attained to aid in the resolution of the aforementioned concerns. As such, some studies demonstrate the potential for vulnerable populations to adopt exercise training after the correct interventions. For instance, the Strength Through Education, Physical fitness and Support (STEPS) study conducted an exercise intervention targeted at 139 Appalachian women, 40 years or older, consisting of strength, flexibility and balance exercises for 12 weeks.[150] The study reported improved physical health and breast cancer awareness in these women.[150] Hartman et al[151] created an individually tailored exercise intervention for sedentary women with a family history of breast cancer. The women were able to increase the minutes of exercise per week after the 12-week period.

Taken together, the utilization of n-3 PUFA and exercise for breast cancer prevention is faced with numerous challenges; however, studies continue to advance our understanding of the specific parameters to be considered for potentially achieving the optimum beneficial effect of n-3 PUFA intake and exercise. More studies are warranted to reach optimum exercise protocols and n-3 PUFA dosage that are specific for different stages of breast cancer prevention.

Conclusion

Weight gain, inflammation, hormones, and epigenetic modifications that are discussed in this review are important biomarkers of breast cancer risk. We have discussed available literature pertaining to the role of exercise and n-3 PUFA on levels of these biomarkers to demonstrate their relevance as targets of exercise and n-3 PUFA intake. The lack of studies directly linking exercise or n-3 PUFA intake to some of these factors and breast cancer risk demonstrates a gap in the understanding of the role of exercise and n-3 PUFA intake in breast cancer prevention. By identifying this gap, we identify potential areas where future research is warranted. Despite the increasing focus on the cancer-preventative roles of n-3 PUFA and exercise training, a combinatory effect of those two factors on breast cancer prevention has not yet been investigated. In this review, we have identified potential common targets of n-3 PUFA intake and exercise training (Fig. 1). Additional studies are warranted to determine whether a combination of n-3 PUFA intake and exercise can provide a new strategy for preventing breast cancer initiation, development, progression, or recurrence.

Abbreviations

T2D, type II diabetes mellitus; BRCA1, breast cancer susceptibility gene 1; BRCA2, breast cancer susceptibility gene 2; PUFA, polyunsaturated fatty acid; EPA, eicosapentaenoic acid; DHA, docosahexaenoic acid; EGFR, epidermal growth factor receptor; HEGF-2, human epidermal growth factor-2; PTEN, phosphatase and tensin homolog; MMTV, mouse mammary tumor virus; ER+, estrogen receptor positive; PR+, progesterone receptor positive; ALA, alpha-linolenic acid; mTOR, mammalian target of rapamycin; BMR, basal metabolism rate; BMI, body mass index; DIT, diet-induced thermogenesis; EE, energy expenditure; FOx, fat oxidation; MET, metabolic equivalent task; PyMT, polyoma middle T oncoprotein; UPR, unfolded protein response; TNF-α, tumor necrosis factor alpha; COX-2, cyclooxygenase-2; IRS, insulin receptor substrate; PPAR-γ, peroxisome proliferator-activated receptor gamma; NFκB, nuclear factor κB; IL-6, interleukin-6; IFN-γ, interferon-γ; MIP-1, macrophage inflammatory protein-1; NK cells, natural killer cells; TGF-β, transforming growth factor-β; STAT3, signal transducer and activator of transcription 3; TPM1, tropomyosin 1; PDCD4, programmed cell death 4; AMPK, AMP-activated protein kinase; JAK/STAT pathway, Janus kinase/signal transducer and activator of transcription pathway; HOMA, homeostasis model assessment; ER, estrogen receptor; PR, progesterone receptor; hsCRP, high-sensitivity C-reactive protein; CpG, 5′-C-phosphate-G-3′sequence; RNAi, RNA interference; AIs, aromatase inhibitors.

Author Contributions

Wrote the first draft of the manuscript: SAA. Contributed to writing the manuscript: SAA, JLM, SMJ, DWLM. Agree with manuscript results and conclusions: SAA, JLM, DWLM. Jointly developed the structure and arguments for the paper: SAA, JLM, DWLM. Made critical revisions and approved final version: DWLM. All authors reviewed and approved the final manuscript.

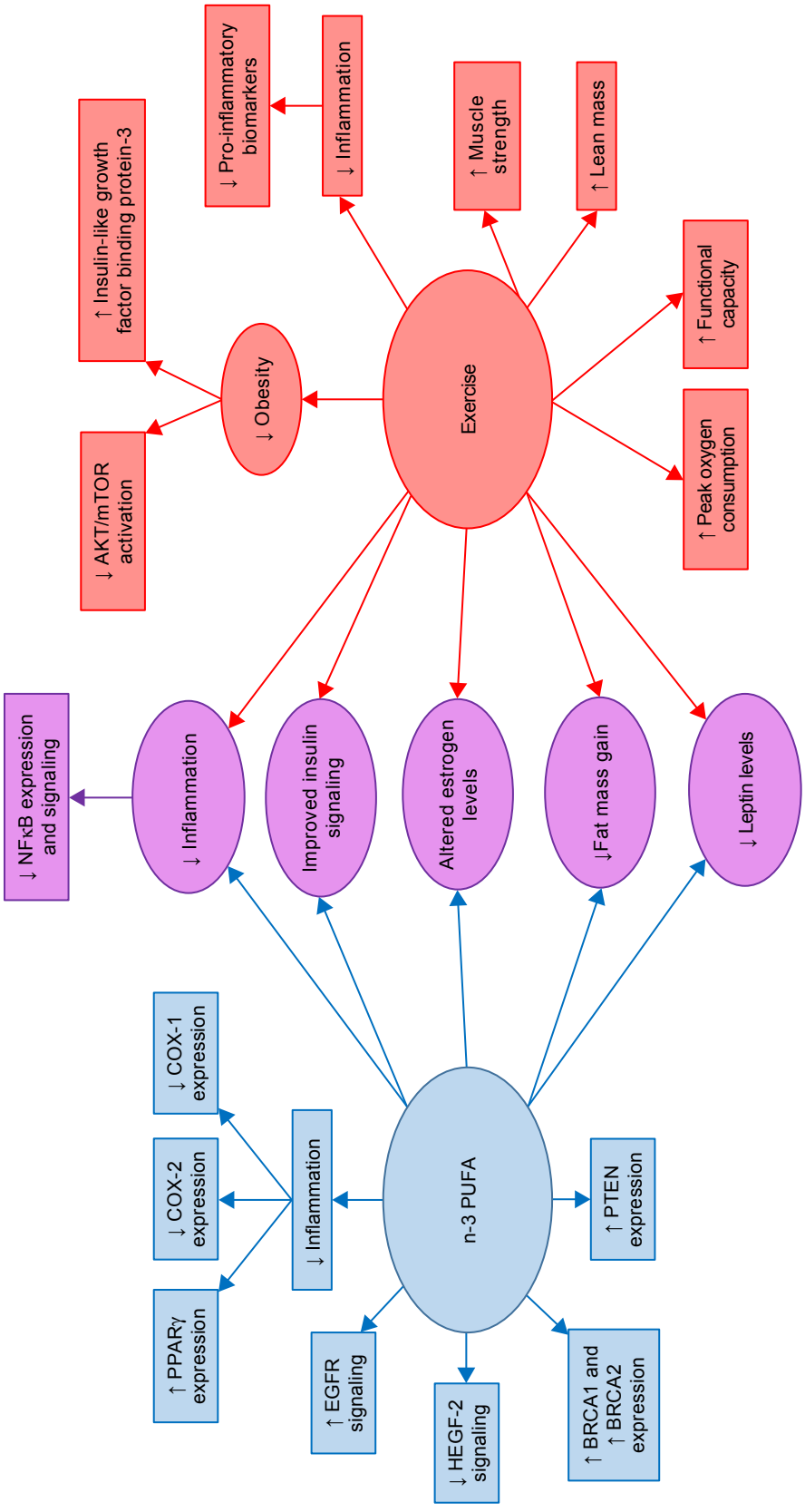

Figure 1. Illustrative figure summarizing common targets of n-3 PUFA and exercise that may contribute to reduced breast cancer risk.

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