

Molecular Pathways: Adipose Inflammation as a Mediator of Obesity-Associated Cancer

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Abstract

The increasing rate of obesity worldwide is predicted to be associated with a surge in diseases. Notably, obesity has been linked to approximately 20% of cancer cases in the United States; obesity is associated with both increased risk and worse outcomes after diagnosis. Altered levels of circulating factors are strongly implicated, including insulin, insulin-like growth factor 1, leptin, adiponectin, and interleukin-6 (IL-6). In addition, increasing attention has focused on the consequences of local adipose inflammation. Inflammatory foci characterized by crown-like structures consisting of dead adipocytes encircled by macrophages occur in white adipose depots, including the breast tissue, of most overweight and obese women. Saturated fatty acids, released as a consequence of obesity-associated lipolysis, induce macrophage activation via Toll-like receptor 4, thereby stimulating NF- κ B signaling. This, in turn, activates transcription of proinflammatory genes including *COX-2*, *IL-6*, *IL-1 β* , and *TNF α* . Elevated levels of proinflammatory mediators cause both local and systemic effects. Of particular relevance with regard to breast cancer is increased transcription of the *CYP19* gene encoding aromatase, the rate-limiting enzyme for estrogen synthesis. Notably, this obesity–inflammation–aromatase axis provides a plausible explanation for increased rates of postmenopausal, hormone receptor–positive breast cancer associated with obesity and hence may offer targets for interventions to attenuate risk or improve prognosis. Potential approaches include weight reduction, exercise, and suppression of obesity-driven signaling pathways using pharmaceutical or dietary agents. A key future goal is to identify biomarkers that accurately report adipose inflammation, both for identification of at-risk individuals and to assess the efficacy of interventions. *Clin Cancer Res*; 19(22); 6074–83. ©2013 AACR.

Background

Using the conventional definition for obesity of body mass index (BMI) ≥ 30 kg/m² [(weight in kg)/(height in m)²], it was recently estimated that more than 500 million adults worldwide are obese and almost twice that number are overweight (defined as BMI 25.0–29.9; ref. 1). Recent decades have witnessed a steady increase in both absolute numbers and the proportion of obese individuals. Because obesity is a key driver of diseases such as type II diabetes, cardiovascular disease, and cancer, its increasing incidence has profound clinical implications. Excessive adiposity is specifically associated with increased risk of multiple malignancies including non-Hodgkin lymphoma, esophageal adenocarcinoma, and cancers of the colon, liver, pancreas, gallbladder, kidney, uterine endometrium, and

breast (2, 3). For female breast cancer, differential associations have been identified according to menopausal status. There is clear evidence for increased breast cancer risk as a function of increasing BMI in postmenopausal women, but epidemiologic analyses suggest a reduced overall risk of premenopausal breast cancer associated with obesity (2, 3). However, emerging data indicate a positive association between obesity and triple-negative disease before menopause (4). In addition, obesity is associated with worse prognosis after breast cancer diagnosis (5).

Multiple molecular changes arising as a consequence of increased body mass are likely to contribute to the increased incidence of neoplasia and worse outcomes in obese individuals. These include hyperinsulinemia, elevated insulin-like growth factor I (IGF-I) levels, adipokine imbalances, and increased cytokine and estrogen levels (6). Importantly, obesity is characterized not only by increased adipose burden but also by altered adipose biology. Specifically, white adipose tissues from obese individuals and murine obesity models exhibit inflammation, defined by infiltration of leukocytes, including macrophages, as well as CD8-positive T lymphocytes and mast cells (7). Adipose inflammation is increasingly recognized as a key component of obesity-associated diseases such as type II diabetes. Here, we focus on the proneoplastic consequences of inflamed

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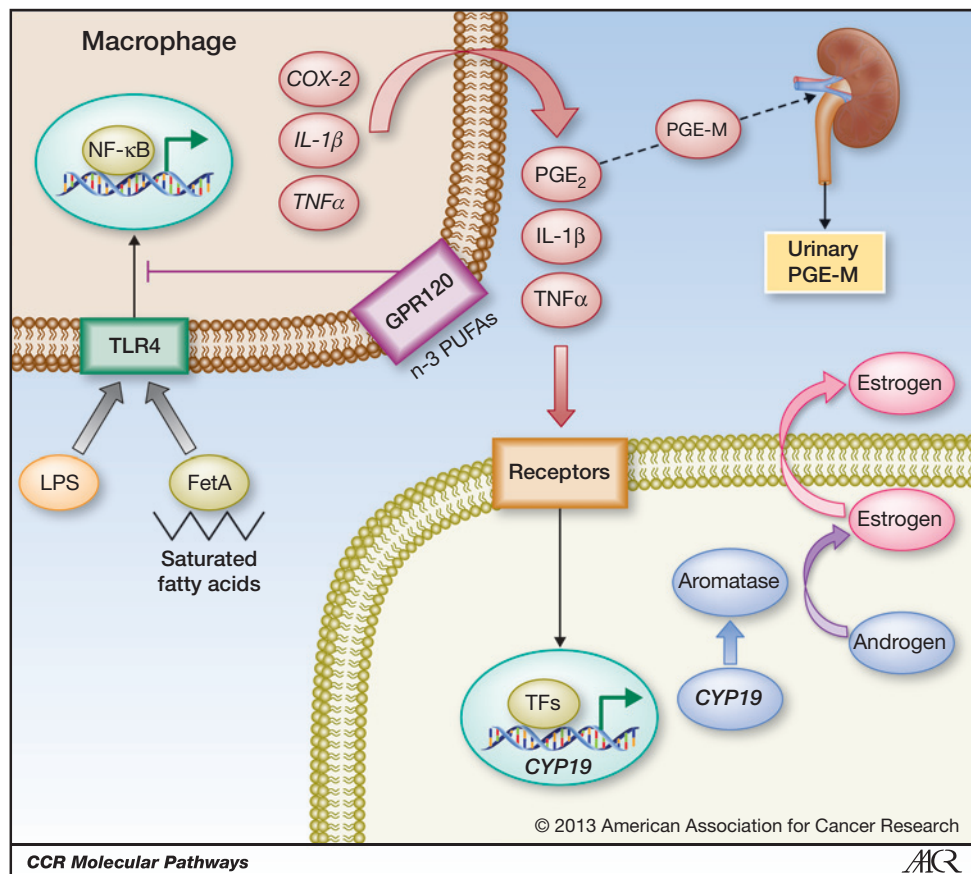


Figure 1. Paracrine interactions between macrophages and other cell types establish an inflammatory milieu in obese breast adipose tissue, resulting in activation of ER α -dependent gene expression. Saturated fatty acids, released from adipocytes as a result of obesity-associated lipolysis, complex with Fetuin A (FetA) and activate TLR4 signaling, resulting in enhanced NF- κ B activity in macrophages. In addition, bacterial endotoxin (LPS) entering systemic circulation as a consequence of obesity-induced impairment of gut mucosal integrity may also elicit NF- κ B activation through TLR4 ligation. NF- κ B induces expression of proinflammatory genes including *COX-2*, *IL-1 β* , and *TNF α* in macrophages. n-3 PUFAs signal via GPR120 and can suppress TLR4 signaling. Cytokines and COX-2-derived PGE₂ activate transcription of the *CYP19* gene encoding aromatase in neighboring cells, including preadipocytes, leading to elevated expression and activity of aromatase. Consequently, estrogen biosynthesis is enhanced, which manifests as increased expression of ER target genes, including. Systemic consequences of adipose inflammation include increased circulating levels of cytokines as well as accumulation of the PGE₂ metabolite PGE-M in urine. Urinary PGE-M levels may therefore provide a valuable biomarker of obesity-related white adipose tissue inflammation.

adipose, delineating paracrine interactions between adipocytes, macrophages, and other cell types (Fig. 1) that are likely to contribute to the elevated cancer incidence in general and worse overall outcomes associated with excess adiposity. These observations also have unique implications for estrogen-driven carcinogenesis.

Adipose tissue macrophages can comprise up to 40% of the cells in obese adipose tissue and represent a rich source of cytokines, which are key mediators of the increased risk of insulin resistance associated with obesity (7). Histologically, this macrophage infiltration manifests as inflammatory foci known as crown-like structures (CLS), which consist of dead adipocytes encircled by macrophages (8–10). Initially identified in visceral and subcutaneous fat, these inflammatory foci were recently observed in breast white adipose tissue from obese women and mice, where they were called CLS-B, with "B" denoting breast (11–13). Strikingly, CLS abundance generally increases as a function of body mass,

both in breast and in other adipose depots (8–12, 14). Consistent with these observations, gene expression analyses have identified selective enrichment of macrophage markers in breast tissue from obese women (14). It is important to emphasize that the relationship between CLS-B presence and obesity is not linear: Some obese individuals lack excess (or any detectable) CLS, whereas a minority of lean individuals exhibit this sign of adipose inflammation. Hence, from the standpoint of targeting the consequences of obesity-associated inflammation, BMI alone is not sufficient for the selection of at-risk patients.

Macrophages can be broadly divided into two functional classes, M1, also referred to as classically activated, and M2, or alternatively activated. Although oversimplified, this binary classification of macrophage polarity has proved useful in understanding the link between inflammation and cancer biology. M1 macrophages secrete proinflammatory mediators including prostaglandin (PG) E₂, interleukin-1 β (IL-1 β),

IL-6, and TNF α , whereas M2 macrophages, triggered by Th2 cytokines, are functionally distinct and associated with tissue remodeling and immunosuppression. Resident macrophages in lean adipose tend to be M2-polarized, whereas it has been suggested that M1 macrophages play an important role in inflamed adipose tissue in the obese (15). The formation of CLS provides a functional structure for macrophage clearance of dead adipocytes but also results in macrophage exposure to saturated fatty acids because of obesity-associated lipolysis (refs. 16, 17; Fig. 1). Saturated fatty acids can activate Toll-like receptor 4 (TLR4) at the macrophage cell surface and thereby induce NF- κ B signaling, which leads to transcriptional activation of proinflammatory genes including cyclooxygenase-2 (COX-2), IL-1 β , and TNF α (18, 19; Fig. 1). Importantly, the liver secretory protein fetuin A was recently found to act as an adaptor protein between free fatty acids and TLR4, providing a link between free fatty acids, white adipose tissue inflammation, and insulin resistance (20). Ingress of bacterial endotoxin [lipopolysaccharide (LPS)] facilitated by obesity-associated defects in gut mucosal integrity has also been suggested to lead to activation of NF- κ B via TLR4 ligation (21, 22). Proinflammatory molecules including TNF α , IL-1 β , and IL-6 released from activated macrophages have both local and systemic actions, which are likely to contribute both to insulin resistance and to the increased cancer risk and worse outcomes associated with obesity (2, 3). Thus, the morphologic entity denoted CLS likely corresponds to a functional unit that contributes to obesity-related disease via increasing both local and systemic exposure to inflammatory mediators.

Using both genetic and diet-induced obesity models, it has recently been shown that NF- κ B signaling and expression of COX-2, TNF α , and IL-1 β are all increased in both mammary glands and visceral fat from obese female mice (12). Increased NF- κ B signaling and cytokine expression is detected in the stromal-vascular fraction of the mammary gland, consistent with upregulation occurring, at least in part, in macrophages. Similar effects were observed in the inflamed breast white adipose tissue of obese women (11, 13). Thus, adipose inflammation, histologic and molecular, occurs in breast tissue in both obese mice and women, validating the mouse as a model of the human disease (11, 12).

Importantly, activation of estrogen receptor (ER) α -dependent gene expression [e.g., progesterone receptor (PR)] as a consequence of adipose inflammation has also been observed in both murine mammary fat pads and human breast. The relevance of this observation is increased by the knowledge that estrogen signaling is likely to be a key contributor to obesity-associated breast cancer (11–13). Approximately two thirds of human breast carcinomas express ER α , and the importance of estrogen and ER signaling in breast neoplasia is clearly illustrated by the protective effects of early menopause and oophorectomy and by the use of selective estrogen receptor modulators (SERM) and aromatase inhibitors in the prevention and treatment of the disease (23, 24). Furthermore, estrogen metabolites have been proposed to have carcinogenic activity indepen-

dent of ER signaling (25). Hence, the demonstration that there is increased expression of an estrogen-regulated gene suggests a critical role for inflammation—associated with obesity—in the pathogenesis of postmenopausal breast cancer.

These observations may resolve a longstanding clinical paradox, which is that increased rates of hormone-dependent breast cancer are seen in the decade after menopause when circulating estradiol is known to decrease and become noncyclical. Perhaps the explanation is that local tissue concentrations of estrogen are actually increased in some patients because of obesity and localized inflammation. The primary site of estrogen biosynthesis in premenopausal women is the ovary but, post climacteric, peripheral sources assume increased relative importance in estrogen synthesis. In particular, adipose tissue as well as breast cancer epithelium express the estrogen synthase aromatase, encoded by the *CYP19* gene, and produce estrogen (26–30). On the basis of importance of adipose as an estrogen source in postmenopausal women, it has long been assumed that increased adiposity is associated with elevated circulating estrogen and indeed BMI is an established determinant of serum estradiol in postmenopausal women (31–33). However, recent studies identify increased aromatase expression in inflamed adipose tissue of obese women and mice due to upregulation by proinflammatory mediators released, at least in part, from CLS-associated macrophages (11–13). *CYP19* transcription can be induced by the interaction of PGE₂, TNF α , and IL-1 β with their cognate receptors (refs. 12, 34–39.; Fig. 1). For example, PGE₂ stimulates *CYP19* transcription via a signaling cascade involving cAMP, protein kinase A (PKA), and phosphorylation of cAMP response element-binding protein (CREB) to induce aromatase (34–38, 40). Notably, a switch in *CYP19* promoter usage occurs in breast cancers and cancer proximal stromal tissue, involving selective use of cAMP-sensitive promoters (41–44). Positive correlations between COX and aromatase expression have been identified in human breast cancers (45–47), and evidence for a causal basis for these findings is provided by transgenic and knockout mouse studies (48). Thus, on the basis of the observation of increased levels of PGE₂, TNF α , and IL-1 β in breast tissue from obese women and mice, with corresponding increases in the expression of aromatase and ER α target genes (11–13), it is likely that paracrine interactions between macrophages and other cell types establish an inflammatory milieu, resulting in elevated estrogen biosynthesis and signaling, as depicted in Fig. 1. In support of this pathway, striking correlations are evident between the extent of inflammation in human breast tissue (i.e., CLS-B) and levels of components of the signaling pathway—COX-2 protein, PGE₂, aromatase expression and activity, and PR protein levels (13). In addition, correlations are observed between inflammation and cAMP levels and PKA activity (13), consistent with the ability of PGE₂ to induce cAMP production (49).

In the aggregate, these data establish that obesity can drive adipose inflammation, leading to induction of aromatase

and increased estrogen signaling in the breast and other adipose depots. White adipose inflammation may also be associated with estrogen-independent activation of ER α signaling induced by covalent modification of ER α in response to elevated levels of growth factor or proinflammatory mediators. Delineation of the link between obesity-related white adipose tissue inflammation and induction of ER α -dependent gene expression provides mechanistic insight into the observed correlation between obesity and postmenopausal breast cancer risk and may also explain the increasing proportion of ER α -positive breast cancer observed as a function of age (and despite cessation of ovarian estrogen production), given that aging is associated with inflammation (5). Notably, elevated cytokine levels may also impact carcinogenesis independent of estrogen biosynthesis. Multiple proneoplastic consequences of cytokine overproduction have been propounded (50). Similarly, COX/PG signaling is strongly implicated in neoplasia, and obesity-driven local prostanoïd overproduction may drive tumorigenesis via pleiotropic mechanisms (51–53). Importantly, obesity-dependent increases in proinflammatory mediators not only exert proneoplastic effects locally but also have systemic consequences, as discussed below.

Local versus Systemic Effects of White Adipose Tissue Inflammation

As detailed above, in-breast white adipose tissue inflammation is likely to help explain the link between obesity and postmenopausal, hormone-driven breast cancer. Importantly, multiple lines of evidence suggest that obesity-related changes in levels of circulating factors including insulin, IGF-I, adipokines (leptin, adiponectin), and proinflammatory mediators also play a significant role in the pathogenesis of breast cancer (54, 55). Increased circulating levels of TNF α and IL-6 are found in obese women and have been associated with breast cancer development and progression (56–58). The relative importance of local white adipose tissue inflammation versus altered levels of circulating factors in the development and progression of breast cancer remains to be elucidated. It should be stressed, however, that the local and systemic effects of obesity are interrelated processes. If a woman has breast white adipose tissue inflammation, it is highly likely that inflammation will be present in other fat depots including within the abdomen. Obesity has already been shown to be associated with adipose inflammation in both mammary and visceral fat depots in murine models of obesity (12), and studies are under way in women to assess the relationship between breast white adipose tissue inflammation and inflammation in other fat depots.

It is easy to envision the importance of local white adipose tissue inflammation in the pathogenesis of breast cancer because the breast epithelium is surrounded by adipose tissue. Furthermore, visceral fat, which also exhibits obesity-associated inflammation, is contiguous with internal organs, including the colon, pancreas, and kidney, for

which obesity is associated with increased cancer risk (2, 3). In contrast, for some other tumor types, systemic consequences of obesity-related white adipose tissue inflammation may outweigh local effects. For example, white adipose tissue inflammation contributes to insulin resistance and consequent hyperinsulinemia, which has been suggested to contribute to both the development and progression of tumors (59). The relative importance of circulating factors in altering the development and progression of cancer due to effects on epithelial versus stromal cells is uncertain and the subject of ongoing research. These effects do not necessarily relate to the specific changes in *CYP19* expression and estrogen production discussed above, but collectively, these findings imply that developing interventions to attenuate obesity-related white adipose tissue inflammation or its consequences offers promise both as a risk reduction strategy and to improve the outcomes for a variety of malignancies including breast cancer. Of course, interrupting this systemic pathophysiology also promises to attenuate nononcologic risks such as cardiovascular disease.

Clinical–Translational Advances

Identification of the obesity–inflammation axis depicted in Fig. 1 provides a framework for designing rational interventions to reduce obesity-associated breast cancer risk and improve prognosis. Importantly, interventions that reduce breast white adipose tissue inflammation are also likely to have beneficial effects on other adipose depots and malignancies. Potential strategies can broadly be organized into three groups: those that aim to ameliorate adipose inflammation, those targeting key components of the dysregulated inflammatory signaling pathways, and those that seek to remediate the downstream consequences of adipose inflammation on tumor biology. Little is known about the potential use of these different strategies to reduce the risk of obesity-related cancers or improve prognosis. Each of these potential strategies is considered below.

Reducing adipose inflammation

In terms of strategies to reduce obesity-related adipose inflammation, the most obvious candidate approach is weight loss, as attenuation of obesity is likely to be associated with amelioration of inflammation. Important proof-of-principle for this approach was provided by a clinical study in which weight loss achieved by feeding obese subjects a very low calorie diet for 28 days was associated with an improved inflammatory gene expression profile in subcutaneous fat, with effects most evident in the stromal vascular fraction (60). Furthermore, reductions in risk of recurrence were noted in a large randomized adjuvant treatment trial in which a low-fat diet, associated with weight loss, was tested, although the subjects were not obese (61). Consistent data were obtained in a recent animal study of caloric restriction (CR); 30% CR for 7 or 14 weeks in a mouse diet-induced obesity (DIO) model arrested the weight gain induced by feeding a high-fat diet and was associated with a profound reduction in breast

inflammation (i.e., CLS-B multiplicity) and with normalization of levels of proinflammatory mediators, aromatase, and PR (62). Importantly, several clinical studies have established improvements in circulating biomarkers of inflammation associated with weight loss, dietary modification, and/or exercise (63–67). Together, these datasets suggest the potential use of altered energy balance as a strategy to resolve inflammation and potentially reduce the risk of breast cancer or improve outcomes in survivors. On the basis of this constellation of findings, a clinical trial to evaluate the efficacy of weight loss and exercise in improving the outcomes specifically of obese women with early-stage breast cancer is needed (68). Nevertheless, concerns regarding the limited long-term success achieved clinically with diet and exercise in the at-risk population provide the impetus for parallel searches for effective pharmacologic and/or surgical interventions.

Several weight loss drugs are currently available. Sympathomimetic drugs (e.g., phentermine) act as appetite suppressants. A combination formulation of phentermine with the antiseizure medication topiramate was recently approved by the U.S. Food and Drug Administration. Lorcaserin, also approved in 2012, functions as an agonist for the 5-HT-2C receptor, thereby increasing the sense of satiety, whereas the pancreatic lipase inhibitor orlistat reduces absorption of dietary fat. However, the extent of weight loss achieved with each of these drugs is modest and reversible upon drug cessation. To our knowledge, the potential use of existing weight loss drugs for reducing obesity-related white adipose tissue inflammation or modulating the development or progression of cancer is uncertain. Thus, the potential of effective weight control agents to modulate obesity-related cancer risk or improve outcomes should be considered along with their more widely perceived benefits in terms of diabetes and cardiovascular risk.

The most effective treatment for significant and sustained weight loss is bariatric surgery, including Roux-en-Y gastric bypass and gastric banding. Gastric bypass surgery is frequently associated with complete remission of type II diabetes, although the rapid timeframe of improved insulin sensitivity frequently precedes significant weight loss (69, 70). Strikingly, reduced adipose macrophage density and decreased stromal vascular fraction expression of factors responsible for macrophage recruitment have been observed in subcutaneous white adipose tissue of patients 3 months after gastric bypass surgery (8). These findings suggest bariatric surgery as a potential route to achieve durable weight loss and resolution of inflammation. Notably, significant reductions in cancer incidence and mortality have been identified in gastric bypass patients compared with matched obese control subjects in both retrospective and prospective studies (71, 72). Reduced cancer incidence was observed in women but not in men (71).

Targeting inflammatory signaling

In addition to attempting to reduce adipose inflammation, targeting key components of the dysregulated inflammatory signaling pathways shown in Fig. 1 may be bene-

ficial. In obesity, both saturated fatty acids and endotoxin have been suggested to stimulate TLR4 signaling resulting in activation of NF- κ B (73). Increased circulating levels of endotoxin are believed to be a consequence of obesity-related increases in gut permeability, and this may provide a signaling source that adds to or amplifies the effect of free saturated fatty acids released as a consequence of obesity-associated lipolysis (21, 22). In preclinical studies, modulating the gut microbiota can reverse high-fat diet-induced metabolic disorders including fat mass gain, endotoxemia, and adipose tissue inflammation (74). Possibly, therapies that modulate gut microbiota will reduce endotoxemia and thereby prevent this source of activation of TLR4 signaling and secondarily suppress levels of inflammatory mediators in white adipose tissue depots including the breast. Targeting cross-talk between the host and gut microbiota to reduce obesity-related cancers is an exciting possibility that warrants investigation. In addition to attempting to reduce levels of TLR4 agonists, for example, endotoxin, as a therapeutic strategy, we note that TLR4 antagonists have been developed. Although these agents possess anti-inflammatory effects (75), it is not known whether they can modulate obesity-related carcinogenesis.

Certain types of lipids possess anti-inflammatory activity. n-3 Polyunsaturated fatty acids (PUFA), including docosahexanoic acid (DHA) and eicosapentaenoic acid (EPA), can inhibit inflammation. Recently, the G-protein-coupled receptor GPR120 on macrophages was identified as a sensor for n-3 fatty acids, activating a β -arrestin/TAB1 signaling system that results in inhibition of TLR4 signaling (73; Fig. 1). n-3 Fatty acids, constituents of fish oil, can also activate PPAR γ and block NF- κ B-mediated induction of proinflammatory mediators (76). Consistent with these effects, dietary n-3 PUFA administration suppresses adipose inflammation and hyperinsulinemia in mouse obesity models (77, 78). Epidemiologic analyses of dietary n-3 PUFAs and human breast cancer incidence have yielded equivocal results (79). Intriguingly however, one recent report identified a protective effect of n-3 PUFA consumption selectively in obese women in a Mexican case-control study (80). Together, these data suggest a potential application of n-3 PUFAs for reducing obesity-associated inflammation and consequent neoplastic risk.

The so-called calorie restriction mimetics are also potentially useful for blocking the activation of NF- κ B and suppressing inflammation. These agents are believed to favorably modulate metabolic and stress response pathways regulated by calorie restriction without actually lowering caloric intake. Not surprisingly, these compounds (e.g., resveratrol, rapamycin, metformin) target pathways involved in inflammation, growth factor signaling (especially insulin/IGF-I), oxidative stress, and nutrient metabolism (81, 82). Resveratrol, a polyphenolic compound abundant in grapes and some berries, has pleiotropic activities, including activation of sirtuin-1 (SIRT-1) and suppression of NF- κ B signaling, which likely contribute to its ability to attenuate high-fat diet-induced adipose inflammation in mice (83).

Effects of the bacterially derived immunosuppressant rapamycin are best understood in terms of its ability to inhibit mTOR complex 1 (mTORC1), a central regulator of cell growth responsible for integrating growth factor signaling, nutrient, energy, and oxygen availability, and translating the net signal to provide the appropriate level of translational activity within the cell. mTOR inhibition attenuates insulin/IGF-I signaling, a major dysregulated pathway in obesity. Furthermore, rapamycin-mediated mTOR inhibition can also block activation of NF- κ B signaling (84).

The biguanide metformin is currently generating considerable excitement as a potential anticancer drug (85, 86). Metformin is widely prescribed for the treatment of type II diabetes and appears to increase whole-body insulin sensitivity by reducing hepatic gluconeogenesis and enhancing glucose uptake by skeletal muscle. Retrospective population studies have identified reduced cancer incidence associated with metformin use, although the anticancer effect has yet to be confirmed in prospective trials (85, 86). Nevertheless, several potential antineoplastic mechanisms have been ascribed to metformin ranging from systemic reductions in insulin signaling to local effects at the cancer cell level on key regulators of energy balance. Metformin directly affects mitochondrial electron transport and stimulates AMP-activated protein kinase (AMPK), including in human adipose tissue (87). There is emerging evidence that AMPK can suppress the activation of NF- κ B via its downstream mediators SIRT1, FoxO, and PGC-1 α (88). Metformin-mediated AMPK activation may also result in suppression of aromatase expression in breast adipose cells (89). Currently, metformin is being prospectively evaluated in patients with early-stage breast cancer in a randomized, placebo-controlled trial with disease-free survival as the primary endpoint (MA.32, NCT01101438). Result stratification according to BMI is expected to be informative with respect to understanding any selective benefit of metformin in obese patients.

White adipose tissue inflammation is associated with elevated levels of COX-2 and PGE₂ (Fig. 1). COX inhibitors, prototypic inhibitors of PGE₂ synthesis, have been widely evaluated for cancer chemoprevention, although with little specific emphasis on obesity-associated disease. Protective effects have been documented for aspirin, conventional COX-inhibiting nonsteroidal anti-inflammatory drugs (NSAID) and selective COX-2 inhibitors in animal models, epidemiologic analyses, and clinical trials (51, 90). Importantly, these drugs can have pleiotropic actions in addition to COX inhibition, including NF- κ B antagonism (91–96) and stimulation of AMPK (97, 98), which may be particularly important in the context of obesity. To date, little clinical data exist addressing obesity or its associated inflammation as a modifier of, or sensitizer to, NSAID action in human neoplasia (99). Reductions in colon cancer risk associated with aspirin use were not found to be modified by BMI in two large-scale observational studies, the Health Professionals Follow-Up Study and the Nurses' Health Study (100). In contrast, results from the Aspirin/Folate Polyp Prevention Study suggested that aspirin may be more

effective in preventing colorectal adenomas in patients with higher BMI (101). These latter data suggest a selective protective effect of aspirin with respect to colon neoplasia in obese individuals and offer a possible path forward for amelioration of elevated cancer risk in association with obesity through the use of aspirin or other COX inhibitors. Comparable data are not yet available for breast cancer. Of note, however, some studies have identified correlations between the use of COX-inhibiting NSAIDs and reduced levels of serum estradiol in postmenopausal women (102, 103), consistent with the notion that PGE₂ is a significant determinant of aromatase expression *in vivo* and potentially indicative of use in suppressing obesity-associated aromatase upregulation. Testing this hypothesis will require selection of the at-risk group using a validated biomarker for localized tissue inflammation. Elevated BMI alone is unlikely to be sufficient for the identification of patients who are most likely to benefit.

Obesity and cancer therapy

It seems likely that local white adipose tissue inflammation in the breast or related changes in circulating factors, for example, increased insulin, will affect tumor biology and reduce the efficacy of treatment by multiple mechanisms. In addition to attempting to reduce adipose inflammation or targeting inflammatory signaling pathways, specific pathways and processes within tumors may need to be targeted in obese individuals. In one recent preclinical study, the IKK β /mTOR/VEGF signaling pathway was activated by proinflammatory TNF α in the mammary tumors of obese mice (104). Interestingly, inhibition of this pathway with clinically available drugs, including aspirin, rapamycin, and bevacizumab, reduced obesity-mediated tumorigenesis. Whether such an approach would work in women is unknown. AMPK activity is decreased in obesity and the metabolic syndrome (88). As mentioned above, there is extensive preclinical evidence that activation of AMPK may be a useful antitumor approach and this has led to a clinical trial to evaluate the use of metformin in patients with early-stage breast cancer.

Delineation of the obesity–inflammation–aromatase axis may also refocus existing treatment strategies based on a new appreciation of the increase in estrogen biosynthesis that is likely to occur in association with adipose inflammation (Fig. 1). Strikingly, in the phase III Arimidex, Tamoxifen, Alone or in Combination (ATAC) trial, proportionately worse outcomes were seen in obese versus lean women treated with the aromatase inhibitor anastrozole but not with the SERM tamoxifen (105), suggesting that efficacy of aromatase inhibition and its relative advantage decreases with increasing weight (although it was never inferior to tamoxifen). This finding may be attributable to the increased expression of aromatase in inflamed adipose (Fig. 1) and could suggest a need for increased dosing to achieve comparable efficacy in the setting of obesity. Alternatively, covalent modification of ER α in response to circulating factors such as insulin/IGF-I could result in

estrogen-independent ER α signaling, which would be predicted to be refractory to increased doses of an aromatase inhibitor. Weight reduction, exercise, or pharmacologic strategies that reduce adipose inflammation might improve the efficacy of aromatase inhibitors in this population. Clearly, we are only at the beginning of developing personalized evidence-based approaches that will be useful for the prevention and treatment of obesity-related cancers.

Biomarker development

Noninvasive biomarkers of white adipose tissue inflammation would be useful and will be needed to enable the identification of individuals who may be at increased risk of cancer. Biomarkers that accurately report on the presence of white adipose tissue inflammation would also be valuable for assessing the efficacy of therapeutic interventions that aim to attenuate inflammation. The need for biomarkers is highlighted by the observation that not all overweight and obese individuals exhibit adipose inflammation or related molecular changes while some lean individuals do (11).

The molecular pathway shown in Fig. 1 suggests that quantifying levels of PGE₂ could inform on the presence of white adipose tissue inflammation. Urinary PGE-M is a stable end metabolite of PGE₂ that reflects systemic PGE₂ levels (106). Notably, two recent reports found increased levels of PGE-M in the urine of obese women (107, 108). Interestingly, an association between high levels of urinary PGE-M and increased risk of postmenopausal breast cancer was observed in women who did not use NSAIDs regularly (107). Additional studies are needed to assess the use of this biomarker of inflammation. Whether it will prove superior to other biomarkers of inflammation such as C-reactive protein is unknown. Studies of serum and plasma are under way in an effort to develop a biomarker signature that will report on the presence of white adipose tissue inflammation. An algorithm incorporating multiple blood-based biomarkers may prove to be useful for inflammation-associated risk evaluation in both obese and lean patients.

Conclusions and Future Directions

Chronic inflammation has been linked to the development of numerous epithelial malignancies (109). It seems likely, therefore, that the recent discovery of breast white adipose tissue inflammation with its associated molecular changes will prove important for understanding the increased risk of breast cancer among obese postmenopausal women. Nonetheless, additional studies are needed to evaluate the significance of CLS-B as a determinant of both breast

cancer risk and prognosis. In addition to the macrophages found in CLS, obesity causes numerous other immunologic changes that could also be highly relevant in the pathogenesis of breast cancer and warrant investigation (7). Another significant question concerns the relevance of white adipose tissue inflammation for other tumor types. Will CLS be present at other organ sites and be associated with either increased risk or poor prognosis? If so, it will be important to establish the relative importance of local versus systemic effects of white adipose tissue inflammation on tumor biology. Certainly, given the growing incidence of overweight and obesity, there is a pressing need to both better understand the molecular mechanisms underlying white adipose tissue inflammation and to develop effective interventions that can be tested and applied to the at-risk subpopulation (s). Currently, we know that obesity is associated with poor prognosis for numerous malignancies, but we do not know whether either improved energy balance or alternate (dietary, pharmacologic) strategies that attenuate white adipose tissue inflammation or related molecular changes will improve outcomes. Prospective clinical trials are needed in patients with cancer to address this important question. In the meantime, we already know that improved energy balance can be helpful in managing common obesity-related comorbidities such as type II diabetes and cardiovascular disease. Taken together, these findings make it seem reasonable for more attention and effort to be made to control energy balance in the obese cancer patient with the goal of improving outcome.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Authors' Contributions

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Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): C.A. Hudis

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