Regulation of Tumor Angiogenesis by Dietary Fatty Acids and Eicosanoids

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Abstract: Angiogenesis is a prerequisite for tumor growth and metastasis. Vascular endothelial cell proliferation, migration, and capillary formation are stimulated by angiogenic growth factors, which include the proteins vascular endothelial growth factor, basic fibroblast growth factor, and transforming growth factor-β, and eicosanoids synthesized from n-6 fatty acids. Clinical studies have shown that angiogenesis in solid tumors relates to a poor prognosis and, in premalignant lesions, indicates potential for cancerous transformation. High-fat, n-6 fatty acid-rich diets were associated with a relatively poor prognosis in breast cancer patients; in a nude mouse model the same diet enhanced breast cancer progression, whereas n-3 fatty acids exerted suppressive effects that were associated with impaired angiogenesis. Lipoxygenase and cyclooxygenase products of n-6 fatty acid metabolism are angiogenic in vitro assays. This activity is blocked by pharmacological inhibitors of eicosanoid biosynthesis, and one, indomethacin, suppressed n-6 fatty acid-stimulated murine mammary carcinoma growth and metastasis and tumor vascularization. Review of the experimental data suggests that selective inhibitors of eicosanoid-synthesizing enzymes and dietary intervention with n-3 fatty acids merit clinical evaluation as adjuvant therapy and chemopreventive agents.

Introduction

Angiogenesis, the formation of new blood vessel networks to permit sustained tumor growth, is one of the most rapidly growing fields in basic and applied cancer research, and there has been considerable progress in our understanding of the regulation of angiogenesis by protein growth factors and an emerging delineation of the corresponding signal transduction pathways. However, less appreciated are the accumulating experimental data that indicate the significance of dietary factors in the angiogenesis-dependent growth of tumors and their capacity for local invasion and spread to distant metastatic sites.

In this review, we discuss the influence of dietary fatty acids and the eicosanoids derived from the metabolism of polyunsaturated n-6 fatty acids on the process of angiogenesis. These bioactive lipid factors are involved in all the steps in angiogenesis: degradation of the vascular basement membrane and interstitial matrix by proteolytic enzymes such as the type IV collagenases and urokinase-type plasminogen activator (uPA), endothelial cell proliferation and migration, and formation of capillary loops and three-dimensional tubular networks. Also, with particular emphasis on breast cancer, we propose that manipulation of the absolute levels and relative proportion of n-6 and n-3 fatty acids consumed in the diet and the use of selective pharmacological inhibitors of eicosanoid biosynthesis offer novel approaches to antiangiogenesis and, hence, to cancer prevention and treatment.

Angiogenesis and Vascular Endothelial Growth Factor: Clinical Correlates

The formation of new blood vessels is a prerequisite for solid tumor growth and metastasis (1), and vigorous neovascularization, or angiogenesis, has been associated with a poor prognosis for cancers arising at several primary sites (2–11). In noncancerous proliferative breast disease and ductal carcinoma in situ, active angiogenesis predicts an increased propensity for progression to invasive cancer (12–14), and a similar phenomenon has been reported for prostate intraepithelial neoplasms (15) and latent carcinomas (16). Cervical intraepithelial neoplasia is also a potentially premalignant condition, although it often undergoes spontaneous regression. However, it may progress to invasive squamous cell carcinoma of the cervix, and when it does so, there is a requirement for increased angiogenesis (17).
The process of angiogenesis is required for sustaining tumor growth in the face of an accumulating cell mass and providing access to the systemic circulation so that tumor microemboli can be transported to distant sites and, there, to establish metastatic foci. For many years, it has been recognized that microscopic, dormant metastases may exhibit rapid growth after surgical removal of the primary cancer (18). More recently, experimental studies have attributed this deleterious effect to the elimination of antiangiogenic factors that are secreted by the primary tumor, permitting the proliferation of vascular endothelial cells located around the growth-arrested micrometastases (19,20).

Among the various defined or putative angiogenic factors, vascular endothelial growth factor (VEGF) is generally believed to be a mitogen that is specific for endothelial cells and involved in tumor angiogenesis. Various human epithelial tumor cells express VEGF (17,21–26), and its degree has been positively associated with angiogenesis, as judged by microvessel counts (17,21,23), early relapse in breast cancer (21), and lymph node metastasis in primary lung cancer (27). VEGF may also be an important switch for the establishment of angiogenic activity in ductal breast carcinomas in situ (28) and, hence, critical for progression to invasive breast cancer.

In addition to VEGF, several other protein growth factors are involved in the angiogenic process. Prominent among these is basic fibroblast growth factor (bFGF) (29), which acts synergistically with VEGF in stimulating capillary growth (30,31) and is consistently expressed in human primary breast cancer tissues (32). Transforming growth factor-β (TGF-β) is angiogenic in vivo, but it appears to function primarily as an indirect mitogen for vascular endothelial cells, exerting its stimulatory activity by upregulating VEGF (33,34). Insulin-like growth factor I was found to induce VEGF expression in colon cancer cells, an effect that appeared to involve an increase in the transcription of the VEGF gene (35).

### Angiogenesis in Animal Models

Angiogenesis and the expression of VEGF have been studied in human cancer cell lines when these had grown as solid tumors in athymic nude mice (36–41). Transfection of the estrogen-dependent MCF-7 breast cancer cell line (36) and the melanoma SK-MEL-2 cell line (37) so as to over-express biologically active VEGF enhanced angiogenesis and tumor growth. Although VEGF overexpression promoted MCF-7 cell solid tumor growth in nude mice, it had no effect on estrogen dependence or on the sensitivity of the transplanted cells to antiestrogen treatment. The expression of VEGF receptors was also examined in the melanoma solid tumors; KDR/flk-1 mRNA was highly overexpressed in the cells transfected with VEGF cDNA, but this was confined to the endothelial cells lining the tumor microvessels, and none was associated with the melanoma cells. Thus, there was no evidence of an autocrine function for the tumor cell-secreted VEGF. The melanoma cell lines were also evaluated for their ability to form tumorous lung deposits after they had been injected intravenously into nude mice. With this “experimental metastasis” technique, it was shown that VEGF overexpression dramatically increased the formation, size, and vascularization of lung colonies.

### Dietary Fatty Acids and Breast Cancer Progression

Previously, we reviewed the use of human tumor xenografts to study the influence of dietary fatty acids on breast cancer growth and metastasis (42). Experiments were performed which demonstrated that the growth of the MDA-MB-231 and MDA-MB-435 estrogen-independent human breast cancer cell lines in the mammary fat pads of female nude mice was enhanced by feeding a high-fat, linoleic acid (LA)-rich diet (43–46), whereas an n−3 fatty acid-supplemented diet exerted corresponding suppressive effects (46–48). The partial inhibition of MDA-MB-231 cell tumor mass acquisition by the long-chain n−3 fatty acid docosahexaenoic acid (DHA) was shown to result from a combination of decreased cell proliferation and increased apoptotic cell death (46), together with reduced angiogenic activity (49).

LA is an essential fatty acid and is the metabolic precursor of arachidonic acid, which is also an n−6 fatty acid and the substrate for the cyclooxygenase (COX) and lipoxygenase (LOX) families of enzymes. There are two COX proteins. The constitutive isofromm is COX-1, which is expressed in most normal tissues and is responsible for the synthesis of prostaglandins (PGs) required for essential physiological functions (50). In contrast, COX-2 is not detectable in most normal tissues; it is induced by phorbol esters, cytokines, and growth factors (50,51), including TGF-β (52) and bFGF (53), and has been associated with carcinogenesis (54).

In cell culture experiments, LA-stimulated growth of MDA-MB-435 cells was inhibited by the nonsteroidal anti-inflammatory drug indomethacin, but only at high concentrations that inhibited LOX and COX activity (55). However, in the nude mouse model, dietary LA-stimulated MDA-MB-435 cell solid tumor growth and metastasis were suppressed by treatment with indomethacin at a dose that blocked only the production of COX-mediated metabolites (56). These two sets of results indicated that inhibition of COX activity was effective in vivo because of interdiction in host-tumor cell interaction, a conclusion that was consistent with suppression of angiogenesis.

Although the MDA-MB-435 and MDA-MB-231 human breast cancer cell lines grow readily when injected into the mammary fat pads of nude mice, they differ significantly in key biological characteristics. The MDA-MB-231 cell line exhibits a greater capacity for invasion in vitro, but, unlike the MDA-MB-435 cell line, no further increase occurs when LA is added to the assay medium (45). Both breast cancer cell lines express a low constitutive level of COX-1, but MDA-
MB-231 cells also possess a high level of constitutive COX-2, which is further upregulated in response to phorbol ester (57,58). As a consequence of this higher net COX activity in MDA-MB-231 cells, PGE₂ production far exceeds that in the MDA-MB-435 cell line (58). However, except at extremely high concentrations, MDA-MB-231 cell growth in vitro was unaffected by piroxicam (59), which is predominantly a COX-1 inhibitor, or NS-398 [N-(2cyclohexyloxy-4-nitrophenyl) methanesulfonamide], a highly selective COX-2 inhibitor (E. M. Gilhooly and D. P. Rose, unpublished data). On the basis of the results obtained with indomethacin (56), when the experiment is done, it may be found that MDA-MB-231 cell tumor growth in nude mice is suppressed by a COX-2 inhibitor as a result of its antiangiogenic activity.

The growth of MDA-MB-435 and MDA-MB-231 cell mammary fat pad solid tumors is stimulated by feeding a high-fat, LA-rich diet to nude mice, but we have observed an instructive difference in the growth curves (45). Figure 1 shows that whereas the MDA-MB-435 cell tumors exhibited an immediate growth response, there was a delay in the formation of palpable tumors from the injected MDA-MB-231 cells. However, after a lag time of five to six weeks, their growth rate was relatively rapid, and by the end of the experiment, tumor size greatly exceeded that of the MDA-MB-435 cell tumors. We postulated that there was an initial avascular phase, which was followed by a period of intense angiogenic activity, mediated by LA-derived eicosanoids, and then a vascular phase of rapid LA-stimulated tumor growth. The eicosanoids produced by MDA-MB-231 tumor cells from LA as a result of COX-2 expression may be important in this vascularization process.

**Eicosanoids as Angiogenic Factors**

**Prostanoids**

The prostanoids are autacoids derived from arachidonic acid via COX-mediated metabolism and include the PGs prostacyclin (PGI₂) and thromboxane (Tx) (Figure 2). A role for arachidonic acid-derived PGs in the process of angiogenesis was proposed by Ben Ezra in 1978 (60) and later established directly by angiogenic bioassays (61,62). Ziche and colleagues (61) used a rabbit corneal assay to demonstrate the angiogenic activity of PGE₁ and PGE₂ and that stimulation of neovascularization by mouse fibroblasts was blocked by indomethacin, a nonselective COX inhibitor. They also showed that fluid secreted by a transplantable rat mammary carcinoma contained PGE₁ and/or PGE₂ and that it was angiogenic in the corneal assay. The angiogenicity of PGE₂, at concentrations that were considered to be physiologically relevant, was also demonstrated by Form and Auerbach (62) in a chick embryo chorioallantoic membrane assay.

Several different experimental approaches have shed light on the steps in the angiogenic process that are influenced by the PGs. In general, these COX products appear to stimulate vascular endothelial cell migration and tube formation, rather than enhance endothelial cell proliferation (63–65). A key study by Tsujii and co-workers (65) used human colon cancer cell lines that constitutively expressed a high level of COX-2 or had been genetically engineered to produce a 10-fold increase in constitutive COX-2 mRNA and protein, with a corresponding elevation in PGE₂ synthesis. Overexpression of COX-2 by colon cancer cells resulted in stimulation of vascular endothelial cell migration and the formation of capillary-like tubes in coculture experiments, which was not associated with altered endothelial cell proliferation. These steps in angiogenesis were inhibited by the addition of the selective COX-2 inhibitor NS-398.

Vascular endothelial cells are not only targets for tumor epithelial cell-secreted eicosanoids, but they also produce

![Figure 1. Growth pattern of MDA-MB-231 (filled triangles) and MDA-MB-435 (filled circles) human breast cancer cells in 2 groups of athymic nude mice fed a 23% (wt/wt) fat diet with 12% linoleic acid.](image)

![Figure 2. Biosynthesis of prostanoids from arachidonic acid, an n–6 fatty acid. Cox, cyclooxygenase; PG, prostaglandin: PGI₂, prostacyclin; TX, thromboxane.](image)
these bioactive compounds from arachidonate metabolism. Gerritsen (66) reviewed the results of her own studies and those of others which showed that the endothelium of microvessels of various anatomic origins secretes PGE2. This COX product modulates vascular reactivity by reducing the response to vasoconstrictors such as norepinephrine (67) and may contribute to physiological neovascularization as well as tumor-associated angiogenesis.

PGL2 is also indirectly a product of COX activity, being formed from PGH2 under the influence of prostacyclin synthase (Figure 2). This prostanooid is synthesized in endothelial cells, although it is principally a major product of arachidonic metabolism in large vessel endothelium, rather than the capillary endothelial cells involved in angiogenesis (66,68). Gerritsen (66) emphasized the involvement of PGE2, rather than PGI2, in microvessel-derived angiogenesis, but a more recent study has demonstrated that the increase in the vascular permeability of microvascular endothelial cells, a likely VEGF-mediated prerequisite for angiogenesis (69), involves PGI2 (70).

TxA2 is an alternative product of PGH2 metabolism (Figure 2) and is synthesized by vascular endothelial cells (71,72). Daniel and colleagues (72) found that when COX-2 expression was induced in renal microvascular endothelial cells by tetradecanoylphorbol acetate (TPA), the principal expression was induced in renal microvascular endothelial cells by tetradecanoylphorbol acetate (TPA), the principal prostanooids secreted were PGE2, PGI2, and TxA2, a metabolite of TxA2, and an index of its biosynthesis. Endothelial cell migration was stimulated by TPA, a response that was inhibited by selective inhibitors of COX-2, but virtually recovered in the presence of a TxA2 mimetic, but not PGE2, PGI2, or a PGI2 analog. Vascular endothelial cells also possess TxA2 receptors, and Daniel and co-workers showed that a receptor antagonist blocked TPA-stimulated endothelial cell migration in vitro and bFGF-stimulated angiogenesis in a corneal assay; a COX-2 inhibitor had a similar effect in this model. The authors postulated that the levels of TxA2 required to enhance angiogenesis in vivo were likely to be obtained from alternative or additional sources to the vascular endothelium, including platelets in tumor-associated microthrombi.

The expression of VEGF was shown to be induced by PGE2 in a nonneoplastic preosteoblastic cell line (73,74) and in synovial fibroblasts (75), and VEGF and bFGF mRNA expression was induced by PGE2 in a rat retinal glial cell line (76). This regulatory mechanism also applies to at least some transformed cells. In the experiments reported by Tsujii and colleagues (65), COX-2 expression in colon cancer cell lines stimulated upregulation of mRNA not only for VEGF, but also for bFGF, TGF-β, and platelet-derived growth factor, and increased synthesis of the corresponding growth factor proteins. Again, angiogenic growth factor production was suppressed by pharmacological inhibition of tumor cell COX-2 activity.

A mutated ras gene is the most common oncogene detected in human cancers and occurs with a high frequency in colon cancer (77). The expression of mutant ras oncogenes has been associated with elevated levels of VEGF mRNA and secreted VEGF protein in transformed rodent intestinal cell lines and human colon cancer cell lines (78,79). Oncogenic ras also induced VEGF expression in NIH 3T3 cells (80) and primary murine endothelial cells (81), and the level of activated H-ras expression was found to correlate with VEGF upregulation through the different steps in mouse skin tumor progression (82). Lovastatin, a drug that interferes with ras protein function, suppressed VEGF production in an Ha-ras-transformed 3T3 cell model and potentiated the antiangiogenesis and antitumor effects of tumor necrosis factor-α (83). In another study, pharmacological inhibition of the ras signaling pathway with a farnesyl transferase inhibitor suppressed the upregulation of VEGF expression in a mutated ras-transfected skin epithelial cell line (84).

Astrocytoma is a brain tumor that does not contain a mutated ras, but VEGF secretion by astrocytoma cell lines was found to be regulated by functional activation of the ras signaling pathway (85). It may be relevant to the present discussion that although ras mutations are not commonly found in breast cancer, one study demonstrated overexpression of unmuted ras gene mRNA in 67% of 27 primary human breast cancer tissues (86). In this context, any consideration of ras has to include the role of its signaling pathway in the regulation of c-myc expression. Not only is c-Myc protein formation essential for cell proliferation (87), but oncogenic myc and ras can cooperate in the transformation of cells (88).

The presence of a mutated ras in human non-small-cell lung cancer cell lines is associated with overexpression of COX-2, together with cytosolic phospholipase A2, and a high level of PGE2 secretion (89). Treatment with a farnesyl transferase inhibitor decreased the expression of both enzymes and reduced PGE2 synthesis. A similar relationship between oncogenic ras and COX-2 expression was observed in human breast cancer cell lines (57,58), a mouse mammary cell line transfected with the V-Ha-ras oncogene (54), and a human breast epithelial cell line transfected with the mutated T-24 H-ras gene (58).

Despite these reports that clearly associate ras gene mutations with elevated VEGF expression and enhanced angiogenesis, on the one hand, and overexpression of COX-2, on the other, we are not aware of any reported studies that have attempted to demonstrate a causal relationship between the overexpression of COX-2, VEGF, and the ras gene or the presence of a mutated ras oncogene.

**Lipoxygenase Products**

The 12-LOX product of arachidonic acid metabolism, 12-hydroxyeicosatetraenoic acid (12-HETE) (Figure 3), has been shown to promote tumor angiogenesis. In experiments of similar design, Connolly and Rose (90) and Nie and associates (91) demonstrated enhanced angiogenesis in nude mouse solid tumors formed from 12-LOX cDNA-trans-
fected MCF-7 human breast cancer and PC-3 human prostate cancer cell lines, respectively.

This angiogenic activity accelerates several steps in the neovascularization process. Nie and associates (91) showed that their PC-3 cell transfectant had an enhanced capacity for stimulating vascular endothelial cell migration in an in vitro system, but, unlike the PGE$_2$ produced by the colon cancer cell lines studied by Tsujii and colleagues (65), 12-HETE also possesses mitogenic activity for fetal bovine aortic endothelial cells (92) and microvascular endothelial cells (93).

As in the case of the PGs, the effect of products of LOX activity on angiogenesis occurs in collaboration with the angiogenic protein growth factors. Our knowledge of the mechanisms involved is incomplete, but it has been shown that interaction occurs with bFGF. Using selective LOX inhibitors, including one with high specificity for 12-LOX, Dethlefsen and co-workers (94) were able to suppress serum- and bFGF-stimulated bovine capillary endothelial cell growth without causing cytotoxicity. It is also noteworthy that COX inhibition with indomethacin did not affect endothelial cell proliferation in this study.

Critical to the migration of endothelial cells, which has much in common with local tissue invasion by tumor cells (Table 1), is their ability to adhere to the extracellular matrix (95,96) and to initiate its degradation (97,98). The adhesion process is facilitated by integrin $\alpha_\beta_3$, one of the family of integrin adhesion molecules (95,96), and in a series of studies, Honn and colleagues showed that 12-HETE increased microvascular endothelial cell adhesion and the expression of cell surface integrin $\alpha_\beta_3$ (99) by a mechanism that involved protein kinase C (PKC) activation (99,100).

uPA and the type IV collagenases are proteolytic enzymes that are secreted by vascular endothelial and tumor epithelial cells. The induction of uPA expression by bFGF and VEGF forms part of the early response to an angiogenic stimulus (97,101), and, given the role of 12-HETE as a second messenger in the angiogenic function of bFGF (94), it may well be that this or another arachidonic acid-derived eicosanoid is involved in uPA expression.

Matrix metalloproteinase-9 is a type IV collagenase that is expressed in microvascular endothelial cells, where it is inducible by TPA-mediated PKC activity (102). Whether 12-HETE is involved in this process is not known, but matrix metalloproteinase-9 expression in the MDA-MB-435 breast cancer cell line is induced by 12-HETE (103).

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**Eicosanoid Synthesis Inhibitors as Antiangiogenesis Agents**

The development and clinical evaluation of pharmacological and natural antiangiogenic agents is one of the most active areas of cancer research (104–106). Suramin, one of the earliest but more toxic agents to enter clinical trial, has been shown to exert multiple antiangiogenic effects, including inhibition of vascular endothelial cell uPA expression and bFGF binding capacity and suppression of endothelial cell proliferation and migration (107). Flavonoids, which occur in a variety of plants and are present at biologically relevant concentrations in some human diets, have antiangiogenic properties: they block angiogenesis in in vitro assays, suppressing bFGF/VEGF-induced invasion of microvascular endothelial cells by mechanisms that include the inhibition of PKC (108).

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### Table 1. Tumor Cell Invasion and Angiogenesis: Two Parallel Processes$^a$

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<tr>
<th>Tumor Invasion</th>
<th>Angiogenesis</th>
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<tr>
<td>Tumor cell proliferation</td>
<td>Vascular endothelial cell proliferation</td>
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<tr>
<td>Proteolytic enzyme production</td>
<td>Proteolytic enzyme production</td>
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<tr>
<td>Cell migration</td>
<td>Cell migration</td>
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<tr>
<td>Growth factor involvement: EGF, TGF-$\alpha$, etc.</td>
<td>Growth factor involvement: VEGF, FGF, TGF-$\beta$, etc.</td>
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<td>Eicosanoids as 2nd messengers: cyclooxygenase and lipoxygenase products</td>
<td>Eicosanoids as 2nd messengers: cyclooxygenase and lipoxygenase products</td>
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<td>PKC activation</td>
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$^a$: Abbreviations are as follows: EGF, epidermal growth factor; VEGF, vascular endothelial growth factor; TGF, transforming growth factor; FGF, fibroblast growth factor family, including basic FGF; PKC, protein kinase C.
We previously remarked on the inhibition of angiogenesis in vitro by COX inhibitors (61,65) and of bFGF-stimulated capillary endothelial cell proliferation by a selective inhibitor of 12-HETE biosynthesis (94). In similar experiments, Ito and colleagues (109) cocultured human omental microvascular endothelial cells and a human esophageal cancer cell line to demonstrate the antiangiogenic properties of two inhibitors of COX and LOX activity. These novel compounds, a natural lignan and a synthetic derivative, blocked the cancer cell-stimulated endothelial tube formation and the subsequent formation of a vascular network.

There is little published work on the antiangiogenic effects of inhibitors of eicosanoid synthesis in animal tumor models. One exception is the report by Lala and co-workers (110). As part of an investigation into the chemopreventive and chemotherapeutic effects of indomethacin in a spontaneous mouse mammary carcinoma model, they described reduced vascularization in hematoxylin-and-eosin-stained sections of the primary tumors that did develop in the indomethacin-treated animals. Although quantitative analysis of microvessel density by immunohistochemical staining was not performed, this apparent suppression of angiogenesis was associated with twofold reductions in the incidence of lung metastases and in the regression of initially emerging primary tumors. Certainly, the present review indicates that suppressed neovascularization is likely to be a major component in the well-described inhibitory activities of nonsteroidal anti-inflammatory drugs, including indomethacin, on carcinogenesis and tumor progression.

When n–3 fatty acid-enriched diets are fed to nude mice bearing human breast cancer xenografts, the uptake of DHA and/or eicosapentaenoic acid into the tumor cell membranes results in the displacement of arachidonic from the constituent phospholipids. The loss of potentially available substrate, together with reduced conversion of LA to arachidonic acid, and n–3 fatty acid-mediated inhibition of COX and LOX, produces dramatic reductions in tumor eicosanoid production, including proangiogenic PGE₂ and 12-HETE (47,49). Furthermore, in addition to this reduction in enzyme substrate, feeding an n–3 fatty acid-containing diet produced a loss of COX-2 mRNA expression in rat mammary glands (111) and a significant reduction of COX-2 and COX-1 immunoreactive protein in N-nitrosomethylurea-induced rat mammary tumors (112). Data such as these suggested that antiangiogenic effects contributed to the suppression of human breast cancer cell growth and metastasis in the nude mouse model (48,113). The antiangiogenic activity of DHA has now been demonstrated; partial suppression of the growth of the MDA-MB-231 human breast cancer cell line as solid tumors in nude mice fed the n–3 fatty acid was associated with reduced microvessel formation within the tumor mass, an inhibition of cell proliferation, and increased apoptosis (49).

**Commentary**

The COX and LOX products of n–6 fatty acid metabolism may exert stimulatory effects on cancer progression at several levels, and in this review we have presented published data in support of a role in tumor-mediated angiogenesis. In addition, these eicosanoids can modulate tumor cell growth and invasion directly and promote the intravascular steps of the metastatic cascade (42,55,56,103). Thus the multiple events contributing to aggressive tumor behavior may be enhanced indirectly by n–6 fatty acids and directly by the eicosanoids derived from them and suppressed by the n–3 fatty acids and pharmacological inhibitors of eicosanoid biosynthesis.

Experimentally, dietary n–3 fatty acids inhibit the growth of preexisting breast cancer micrometastases when used as adjuvant nutritional therapy after excision of the primary tumor, and most likely the suppression of angiogenesis contributes to this therapeutic effect (48). Elsewhere, we have discussed in detail the potential for dietary n–3 fatty acid supplementation as an adjunct to surgery and conventional combination chemotherapy and for cancer prevention (113,114). Overall, a review of the published experimental studies indicates that selective pharmacological inhibitors of COX and LOX activity and dietary n–3 fatty acid supplementation should be included in future clinical trials and that the biological rationale rests, in part, on their antiangiogenic effects.

In addition to its involvement in cancer progression, COX-2-mediated angiogenesis most likely has a critical role in the progression of preneoplastic lesions to the invasive phenotype. Thus the angiogenic process itself (12–16) and COX-2 expression (54,114,115) have been associated with carcinogenesis, indicating that pharmacological inhibitors of the isozyme may exert their cancer chemopreventive effects by mechanisms that include antiangiogenic activity.

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