Molecular Pathways: Beta-Adrenergic Signaling in Cancer

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Abstract

Beta-adrenergic signaling has been found to regulate multiple cellular processes that contribute to the initiation and progression of cancer, including inflammation, angiogenesis, apoptosis/anoikis, cell motility and trafficking, activation of tumor-associated viruses, DNA damage repair, cellular immune response, and epithelial–mesenchymal transition. In several experimental cancer models, activation of the sympathetic nervous system promotes the metastasis of solid epithelial tumors and the dissemination of hematopoietic malignancies via β-adrenoreceptor–mediated activation of protein kinase A and exchange protein activated by adenyl cyclase signaling pathways. Within the tumor microenvironment, β-adrenergic receptors on tumor and stromal cells are activated by catecholamines from local sympathetic nerve fibers (norepinephrine) and circulating blood (epinephrine). Tumor-associated macrophages are emerging as key targets of β-adrenergic regulation in several cancer contexts. Sympathetic nervous system regulation of cancer cell biology and the tumor microenvironment has clarified the molecular basis for long-suspected relationships between stress and cancer progression, and now suggests a highly leveraged target for therapeutic intervention. Epidemiologic studies have linked the use of β-blockers to reduced rates of progression for several solid tumors, and preclinical pharmacologic and biomarker studies are now laying the groundwork for translation of β-blockade as a novel adjuvant to existing therapeutic strategies in clinical oncology.

Clin Cancer Res; 18(5); 1201–6. ©2011 AACR.

Background

The β-adrenergic signaling pathway (Fig. 1) mediates sympathetic nervous system (SNS)–induced fight-or-flight stress responses (1, 2). SNS neural fibers innervate most major organ systems and can release micromolar concentrations of the catecholamine neurotransmitter norepinephrine into target tissues in response to physiologic, psychologic, and environmental threats to homeostasis (1–3). Acute SNS activation also elevates catecholamine levels in circulating blood via the release of epinephrine from chromaffin cells of the adrenal medulla and norepinephrine spill-over from vascular neuro-muscular junctions (1–3). Acute stress responses can elevate norepinephrine and epinephrine levels by >10-fold within seconds, but basal levels also fluctuate tonically over time in response to organismic and environmental conditions (1–3). In addition to central nervous system (CNS) control of general SNS neural outflow, local regulatory processes also influence SNS nerve fiber activity and catecholamine release and degradation. As a result, norepinephrine and epinephrine concentrations can differ substantially in solid tissues versus blood, as well as across different tissue environments at the same point in time (1–3).

The biologic effects of norepinephrine and epinephrine are mediated by α1-, α2-, and β-adrenergic receptor families, which show distinct patterns of tissue distribution and signal through distinct biochemical pathways (3, 4). The 3 subtypes of β-adrenergic receptor, β1, β2, and β3, are present at many sites of tumor growth and metastasis, such as the brain, lung, liver, kidney, adrenal gland, breast, ovary, prostate, lymphoid tissues, bone marrow, and vasculature. β-adrenergic signaling regulates the function of several cancer-relevant cell types, including epithelial cells, vascular myocytes and pericytes, adipocytes, fibroblasts, neural and glial cells, and most lymphoid and myeloid immune cells (3, 4). Ligation of β-receptors by norepinephrine and epinephrine activates the Gguanine nucleotide-binding protein to stimulate adenylyl cyclase synthesis of cyclic AMP (cAMP). The resulting transient cAMP flux can regulate a diverse array of cellular processes via 2 major downstream effector systems (Fig. 1).

One cAMP effector involves activation of protein kinase A (PKA), which subsequently phosphorylates serine or threonine residues on target proteins that bear PKA-receptive amino acid motifs [e.g., R-R-X-(S/T)-Y, in which R = arginine, S = serine, T = threonine, X = any amino acid, and Y = hydrophobic amino acid]. PKA regulates a wide variety of cellular processes ranging from general...
metabolism and growth to cell-specific processes, such as differentiation, morphology, motility, secretion, neurotransmission, and gene transcription. Gene expression effects are mediated by PKA-induced phosphorylation of transcription factors such as the cAMP-responsive element binding protein/activating transcription factor (CREB/ATF) family, which collectively engages approximately 20% of human genes (5, 6). PKA-induced transcriptional alterations often promote cell differentiation at the expense of proliferation and coordinate
transcriptome-wide responses to stress or homeostatic perturbation (5). PKA also activates the β-adrenergic receptor kinase (BARK), which subsequently induces β-arrestin to transiently desensitize further β-receptor signaling and activate the Src/Ras/mitogen-activated protein kinase (MAPK) pathway (7). Recent work also shows that PKA can directly activate Src (A.K. Sood; unpublished data).

A second major cAMP effector involves the guanine nucleotide exchange protein activated by adenyl cyclase (EPAC; ref. 8). EPAC activates the Ras-like guanine triphosphatase Rap1A, which in turn stimulates downstream effectors B-Raf, MAP/extracellular signal-regulated kinase (ERK) 1/2, and ERK1/2. In addition to the well-known effects of the MAPK pathway on cell growth and proliferation, EPAC signaling accounts for many cAMP-induced effects on cell morphology, motility, and secretion dynamics. The effects of EPAC can be distinguished from those of PKA through pharmacologic agonists and molecular manipulations (e.g., target-specific siRNA). Although some overlaps occur, β-adrenergic influences on inflammation, angiogenesis, and invasion seem to be mediated predominantly by PKA induction of genes encoding cytokines and growth factors, whereas EPAC induces complementary but distinct effects on cell morphology and motility.

**Beta-adrenergic regulation of tumor biology**

Studies of β-adrenergic influence on tumor biology were motivated by epidemiologic observations associating stressful life circumstances with accelerated progression of incident cancers (9, 10) and studies linking the use of β-adrenergic antagonists (“β-blockers”) with reduced disease progression (11–15). Epidemiologic studies reveal the most consistent relationships between stressful conditions and progression of already-incident tumors, and relatively little data suggest that stress affects the initial incidence of cancer (9, 10). In vivo laboratory models also show the most consistent effects of experimentally imposed stress on xenograft and syngeneic tumor models (i.e., already initiated tumors; refs. 16, 17), whereas effects on spontaneous incidence or primary tumor growth are less common (but do occur occasionally; refs. 18–21). In mouse models of breast (22) and prostate carcinomas (16, 17), as well as malignant melanoma (23, 24) and leukemia (25, 26), β-adrenergic agonists have been found to block stress-induced enhancement of tumor progression and/or metastasis without affecting primary tumor growth in vivo or tumor cell proliferation in vitro. β-adrenergic agonists have also been found to accelerate in vivo tumor progression and metastasis in the absence of stress (16, 17, 22, 27).

Several cellular and molecular processes have been found to mediate β-adrenergic influences on tumor progression (Fig. 1), including recruitment of macrophages into the primary tumor (22), increased expression of proinflammatory cytokines such as interleukin-6 (IL-6) and IL-8 by tumor cells (28–30) and immune cells (29), VEGF-mediated increases in angiogenesis (27, 31, 32), matrix metalloproteinase (MMP)–related increases in tissue invasion (31, 33, 34), tumor cell mobilization and motility (17, 35, 36), focal adhesion kinase (FAK)–mediated resistance to anoikis apoptosis (37), and BAD-mediated resistance to chemotherapy-induced apoptosis (16, 38), β-antagonists alone or in conjunction with nonsteroidal antiinflammatory agents (NSAID) have also been found to inhibit surgery-induced metastasis in animal models (24, 39, 40). Some evidence suggests that β-adrenergic signaling can also inhibit p53-mediated DNA repair (41), suppress cytotoxic T-lymphocyte and natural killer cell responses (26), inhibit expression of type I IFNs (22, 42), upregulate the Her2-signaling pathway (43, 44), stimulate arachadonic acid signaling (45), activate gene expression by tumor-promoting viruses (9, 46), and upregulate the SNAI2 transcription factor regulating epithelial–mesenchymal transition (S.W. Cole, S.K. Lutgendorf, and A.K. Sood; unpublished data). Each of the latter dynamics may contribute to SNS-induced tumor progression in vivo, but has not yet been confirmed to do so in direct inhibitor studies of mediation. However, it is clear that SNS activation can regulate a wide range of cancer-related molecular pathways via both direct regulation of β-receptor–bearing tumor cells and regulation of other β-receptor–bearing cells present in the tumor microenvironment, such as macrophages and vascular cells.

The SNS can potentially regulate tumor β-adrenergic signaling both via circulating norepinephrine/epinephrine and via local norepinephrine release from SNS nerve fibers. However, growing evidence suggests the latter dynamic plays a dominant role. Analyses of catecholamine levels in human ovarian carcinomas document substantially higher norepinephrine levels in tumor tissue than in blood, and they find no detectable epinephrine in tumor tissue (as would be expected if blood were the primary source of tumor catecholamines; refs. 47, 48). Intratumor norepinephrine levels also correlate with patient psychosocial risk factors and with tumor gene expression profiles, but blood levels of norepinephrine/epinephrine do not (47, 48). Both observations suggest a primary role for local nerve fiber-derived norepinephrine in driving β-adrenergic effects on tumor biology. Histologic analyses of catecholaminergic fibers within human breast and ovarian carcinomas show extensive perivascular innervation and occasional radiation of nerve fibers into the tumor parenchyma (E.K. Sloan; unpublished data; ref. 29). This pattern of SNS innervation is similar to that observed in other solid tissues (e.g., lymph nodes; ref. 49) and provides a source of norepinephrine to directly regulate β-adrenergic receptors on both tumor cells and stromal cells (particularly tumor-associated macrophages; ref. 22). Interestingly, data from the lymph node setting have shown that chronic stress can increase the density of SNS nerve fibers within parenchymal tissue (49). Activated macrophages may also synthesize catecholamines (50), but no evidence has yet shown this to occur within tumors. An additional pathway by which SNS activity may regulate cancer biology, both within the primary tumor microenvironment and systemically at metastatic target sites, involves β-adrenergic regulation of myelopoesis (51–55) and its effects on monocyte and/or...
macrophage trafficking and gene expression (22, 52, 55). This pathway implies that some β-adrenergic influences on tumor biology may originate outside the tumor, via SNS innervation of the bone marrow hematopoietic environment or catecholamine "conditioning" of trafficking monocytes that are ultimately recruited into the tumor microenvironment (22). Such dynamics would complicate the targeting of therapeutic interventions based on tumor tissue β-receptor expression, but they also imply that adjuvant therapy with β-antagonists may suppress systemic support for tumor progression.

**Clinical–Translational Advances**

Because β-adrenergic signaling modulates tumor progression via multiple downstream molecular pathways, β-antagonists may provide a highly leveraged adjuvant therapy strategy with pleiotropic impacts on the primary tumor, its surrounding microenvironment, and metastatic target sites. The biologic appeal of this concept is enhanced by the widespread availability of safe, inexpensive, and well-understood β-antagonists (4). However, several issues need to be resolved to establish the translational potential of β-blockers as adjuvant therapy for cancer.

The most pressing need involves direct assessment of β-antagonists’ clinical efficacy in randomized phase II trials. Conflicting results from currently available nonrandomized observational studies (11–15, 56) suggest that further observational studies are not likely to definitively establish the clinical utility of β-blockers in cancer because of methodologic difficulties such as (i) confounding by indication (e.g., the primary historic indication for β-blockade, cardiovascular disease, shares common pathophysiologic drivers with cancer progression such as smoking, adiposity, and systemic inflammation); (ii) confounding with other pharmacologic exposures that may affect cancer progression (e.g., angiotensin-converting-enzyme inhibitors); (iii) absence of information on influential risk factors and treatment parameters (e.g., cardiovascular data sets provide limited information on cancer progression and/or mortality risk factors, and cancer-related data sets provide limited measures of β-blocker agents and/or utilization); and, (iv) time- and practice pattern-related confounding of cancer survival trends with β-blocker utilization trends (particularly for nonselective β-antagonists that are most likely to be efficacious, as outlined below). Randomized controlled trials provide the only certain way to overcome such biases and definitively assess the protective effects of β-antagonists’ on clinical cancer progression. The availability of preclinical data and approved, safe, and inexpensive β-antagonists with well-understood pharmacology and minimal side effects provides a favorable risk–benefit profile for initial phase II proof-of-concept trials in clinical oncology.

Clinical trial initiation will require selection of optimal disease settings and treatment regimens for assessing clinical impact. Preclinical laboratory models and human pharmacologic epidemiologic studies both suggest that β-antagonists are likely to be most effective in inhibiting the micrometa-
efficacy. Several reasons why tumor β-receptor expression might not provide an accurate predictive biomarker include the fact that receptor expression does not assess the amount of SNS norepinephrine/epinephrine ligand impinging upon the receptor and potential adrenergic effects at extratumoral sites, such as metastatic target tissues or the bone marrow hematopoietic generation of subsequently infiltrating macrophages and lymphocytes (22, 60).

Although a variety of translational parameters remain to be optimized, a growing body of preclinical and pharmacoepidemiologic data suggests that β-adrenergic antagonists hold considerable promise for inhibiting the pleiotropic effects of SNS activation on tumor progression and metastasis. Over the next few years, we can expect further data expanding the range of tumor types examined, identifying additional mechanisms of β-adrenergic effects on tumor progression, and initial randomized clinical trials assessing the efficacy of β-blockade as an adjuvant therapy in clinical oncology.

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were disclosed.

Acknowledgments
Space constraints have necessitated the omission of many relevant references.

Grant Support
Preparation of this review was supported by NIH grants CA116778 and CA109298.

Received October 23, 2011; revised December 2, 2011; accepted December 2, 2011; published OnlineFirst December 20, 2011.
29. Cole SW, Arevalo JM, Takahashi R, Sloan Ek, Lutgendorf SK, Sood
AK, et al. Computational identification of gene-social environment
30. Shahzad MM, Arevalo JM, Armaiz-Pena GN, Lu C, Stone RL, Moreno-
Smith M, et al. Stress effects on FoxO and interleukin-8 (IL8)-driven
Norepinephrine up-regulates the expression of vascular endothelial
growth factor, matrix metalloproteinase (MMP)-2, and MMP-9 in nasa-
32. Chakroborty D, Sarkar C, Basu S, Dasgupta PS, Basu S. Catechola-
33. Landen CN Jr, Lin YG, Armaiz-Pena GN, Das PD, Arevalo JM, Kamat AA,
et al. Neuroendocrine modulation of signal transducer and activator of
Stress hormone-mediated invasion of ovarian cancer cells. Clin Cancer
35. Lang K, Drell TL 4th, Lindecke A, Niggemann B, Kaltschmidt C,
Zaenker KS, et al. Induction of a metastatogenic tumor cell type by
neurotransmitters and its pharmacological inhibition by established
C. Effects of neurotransmitters on the chemokinesis and chemotaxis of
MDA-MB-468 human breast carcinoma cells. Breast Cancer Res Treat
2004;84:169–78.
Adrenergic modulation of focal adhesion kinase protects human
38. Sastry KS, Karpova Y, Prokopovich S, Smith JH, Essau B, Gersappe A,
et al. Epinephrine protects cancer cells from apoptosis via activation of
cAMP-dependent protein kinase and BAD phosphorylation. J Biol
Improving survival rates in two models of spontaneous postoperative
metastasis in mice by combined adrenergic agonist and a coxycyto-
40. Lee JW, Shahzad MM, Lin YG, Armaiz-Pena G, Mangala LS, Han HD,
et al. Surgical stress promotes tumor growth in ovarian carcinoma. Clin
41. Hara MR, Kovacs JJ, Whalen EJ, Rajagopal S, Strachan RT, Grant W,
et al. A stress response pathway regulates DNA damage through b2-
42. Collado-Hidalgo A, Sung C, Cole S. Adrenergic inhibition of innate anti-
viral response: PKA blockade of Type I interferon gene transcription
mediates catecholamine support for HIV-1 replication. Brain Behav
receptor and Her2 comprise a positive feedback loop in human breast
44. Gu L, Lau SK, Loera S, Somlo G, Kane SE. Protein kinase A activation
45. Caky Y, Plummer HK 3rd, Tlhotlir PK, Schuller HM. Beta-adrenergic and
arachidonic acid-mediated growth regulation of human breast cancer
T, et al. Beta-adrenergoreceptors reactivates Kaposi’s sarcoma-associ-
ated herpesvirus lytic replication via PKA-dependent control of viral
47. Lutgendorf SK, Degeest K, Dahmoush L, Farley D, Peredo F,
Bender D, et al. Social isolation is associated with elevated tumor
norepinephrine modulates myelopoiesis after experimental
48. Lutgendorf SK, DeGeest K, Sung CY, Arevalo JM, Peredo F, Lucci J
3rd, et al. Depression, social support, and beta-adrenergic transcrip-
tion control in human ovarian cancer. Brain Behav Immun 2009;23:
175–83.
49. Sloan Ek, Capitanio JP, Tarara RP, Mendoza SP, Mason WA, Cole SW.
Social stress enhances sympathetic innervation of primate lymph
nodes: mechanisms and implications for viral pathogenesis. J Neu-
50. Fieri MA, Rittirsch D, Nadeau BA, Chen AJ, Sarma JV, Zetoune FS,
et al. Phagocyte-derived catecholamines enhance acute inflammatory
SB. Bone marrow norepinephrine mediates development of function-
ally different macrophages after thermal injury and sepsis. Ann Surg
52. Engler H, Bailey MT, Engler A, Sheridan JF. Effects of repeated social
stress on leukocyte distribution in bone marrow, peripheral blood and
53. Laakko T, Fraker P. Rapid changes in the lymphopoietic and granu-
locytic compartments of the marrow caused by stress levels of
Norepinephrine mediates myelopoiesis after experimental thermal
55. Cole SW. Beta-adrenergic regulation of gene expression in cancer. In:
Proceedings of the 102nd Annual Meeting of the American Association
for Cancer Research; 2011 Apr 2–6; Orlando, Florida; Philadelphia
(PA): AACR; 2011.
56. Shah SM, Carey IM, Owen CG, Harris T, Dewilde S, Cook DG. Does
beta-adrenergoreceptor blocker therapy improve cancer survival? Findings
from a population-based retrospective cohort study, Br J Clin Phar-
and beta-adrenergic receptor (AR) protein expression is associ-
ated with poor clinical outcome in breast cancer: an immunohisto-
58. Antoni MH, Lutgendorf SK, Blomberg B, Stagi J, Carver CS, Lechner S,
et al. Transcriptional modulation of human leukocytes by cognitive-
behavioral stress management in women undergoing treatment for
59. Lutgendorf SK, Sood AK, Antoni MH. Host factors and cancer pro-
60. Lutgendorf SK, Lamkin DM, Jennings NB, Arevalo JM, Peredo F, DeGeest K, et al. Biobehavioral influences on matrix metalloproteinase