CHEST

Serum Free Fatty Acid Biomarkers of Lung Cancer

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BACKGROUND: Lung cancer is the leading cause of cancer-related mortality. Surgical removal of the tumor at an early stage can be curative. However, lung cancer diagnosis at an early stage remains challenging. There is evidence that free fatty acids play a role in cancer development.

METHODS: Serum samples from 55 patients with lung cancer were propensity matched with samples from 165 similar pulmonary patients without known cancer. Patients were propensity matched on age, sex, smoking history, family history of lung cancer, and chronic diseases that might affect free fatty acid levels.

RESULTS: Free fatty acids arachidonic acid (AA) and linoleic acid (LA) and their metabolites hydroxyeicosatetraenoic acids (HETEs)(5-HETE, 11-HETE, 12-HETE, and 15-HETE) were an estimated 1.8- to 3.3-fold greater in 37 patients with adenocarcinoma vs 111 patients without cancer (all $P < .001$). Areas under the receiver operating characteristic curve were significantly > 0.50, discriminating patients with lung cancer and control subjects for six of eight biomarkers and two of seven phospholipids tested, and ranged between 0.69 and 0.82 (all $P < .001$) for patients with lung cancer vs control subjects. AA, LA, and 15-HETE had observed sensitivity and specificity > 0.70 at the best cutpoint. Concentrations of free fatty acids and their metabolites were similar in 18 patients with squamous cell carcinoma and 54 control subjects without cancer.

CONCLUSIONS: Serum fatty acids and their metabolites demonstrate good sensitivity and specificity for the identification of adenocarcinoma of the lung.

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ABBREVIATIONS: AA = arachidonic acid; AUC = area under the curve; FFA = free fatty acid; HETE = hydroxyeicosatetraenoic acid; HODE = hydroxyoctadecadienoic acid; HODE-PC = hydroxyoctadecadienoylphosphatidylcholine; LA = linoleic acid; lysoPAF = lysophospholipid platelet activating factor; PAF = platelet activating factor

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Lung cancer is among the most common types of cancer. Surgery is curative only at relatively early stages. By the time lung cancer becomes symptomatic, the best chance for surgical cure is usually well past. Consequently, lethality is high, and lung cancer remains the leading cause of cancer deaths in the United States and worldwide.

Chest radiographs are not sensitive for detection of early-stage lung cancer, and yearly chest radiographs do not reduce lung cancer mortality.[1,2] The only currently accepted method of screening for lung cancer is low-dose CT imaging.[3,4] However, CT imaging is expensive and exposes patients to small doses of radiation. Furthermore, approximately 95% of lung nodules identified by CT scan are benign, and it can be challenging for radiologists to identify those that are not.[4] Low-dose CT scan screening for lung cancer could, thus, be optimized with tools that improve risk stratification and nodule management.

Fatty acids and phospholipids are necessary for cancer cell proliferation.[5] Platelet activating factor (PAF), a potent phospholipid, promotes lung cancer growth and metastasis.[6] PAF is hydrolyzed by phospholipase A2 to produce lysophospholipid PAFs (lysoPAFs). Patients with lung cancer have increased expression and activity of phospholipase A2.[7,8] This enzyme catalyzes oxidized phospholipids to form lysoPAFs and free fatty acids (FFAs), which, in turn, play a major role in tumor development, angiogenesis, lymphangiogenesis, progression, and metastasis.[9-13] LysoPAFs show altered expression in cancer cells. They bind to and activate specific cell-surface G protein-coupled receptors that initiate cell growth, proliferation, and survival pathways (Fig 1).[14,15] LysoPAFs (eg, lysophosphatidylcholines) have been proposed as a potential biomarker for ovarian cancer.[16,17] Plasma phospholipids, including phosphatidylcholine and lysophosphatidylcholines, may be potential biomarkers in prostate cancer[18] and lung cancer.[19]

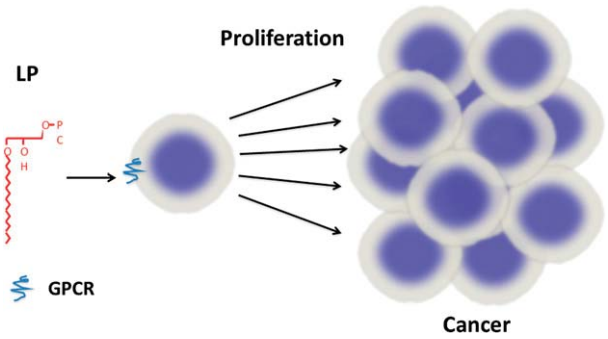

Figure 1 – *The role of LPs in tumor development. LP binds its cognate GPCR and stimulates cell proliferation and migration, which may initiate cancer cell development and metastasis. GPCR = G-protein-coupled receptor; LP = lysophospholipid.*

The theory that elevated FFAs promote cancer is supported by the observation that high dietary linoleic acid (LA) intake promotes breast cancer metastasis in animals.[20,21] The key enzymes for fatty acid synthesis are upregulated in cancer cells, play a critical role in cancer development, and have been chosen as anticancer therapeutic targets.[22-26] The hydroxyeicosatetraenoic acids (HETEs) and hydroxyoctadecadienoic acids (HODEs) are stable oxidative products of arachidonic acid (AA) and LA, respectively. There is considerable basic science evidence that FFAs and the oxidized FFAs HETE and HODE promote tumor development, progression, and metastases in lung cancer using cell culture,[27,28] tissues,[29-31] and animal models.[27,32-34] For example, 5-HETE augments lung cancer cell survival,[30,35] 12-HETE enhances lung cancer cell adhesion,[36-39] and 15-HETE promotes human lung cancer metastasis[40] and induces angiogenesis.[41,42] HODE is involved in many types of cancer, although its specific role remains unclear.[43-46] Studies show that 9-HODE and 13-HODE are ligands of peroxisome proliferator-activated receptor-γ, which promote lung cancer progression and metastasis.[47,48] We, therefore, tested the hypotheses that serum concentrations of phospholipids, FFAs, and their metabolites are greater in patients with known lung cancer than in matched patients apparently without cancer.

Materials and Methods

With Cleveland Clinic Institutional Review Board approval (IRB # 13-306) and informed consent, we retrieved serum samples from 55 patients with lung cancer (cases) from the Cleveland Clinic Lung Biobank. Serum samples were collected from patients with lung cancer at the time of their diagnosis, prior to the initiation of treatment. Pathologic analysis indicated that 37 had lung adenocarcinoma and 18 had squamous cell carcinoma.

Control subjects were selected from a biobank whose inclusion criteria included age 40 to 75 years and at least one of the following criteria: (1) current or ex-smoker with at least a 10 pack-year history, (2) a first-degree relative with a history of lung cancer, or (3) a clinical diagnosis of COPD. Specific matched control subjects (three for each patient with cancer) were chosen separately for patients with adenocarcinoma and squamous cell carcinoma from among all available control patients (see statistical analysis).

Sample Processing

Samples were centrifuged at 3,000 × *g* within 2 h of collection, and the resulting serum was frozen at −70°C until assayed. Biochemical analysis was conducted by an investigator who was strictly blinded to cancer status and patient characteristics.