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TRACKING LIPID CHANGES IN BLOOD TO PREDICT ONSET OF CANCER

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ABSTRACT

Cancer the dreaded disease continues to evoke fear despite great advances in medical research. The passion that drives cancer research is to find an early harbinger of the disease. Early detection in the form of a biomarker can be used to predict onset of, identify the presence of a tumor, and determine its stage, subtype, and ability to respond to therapy. Biomarkers are therefore an invaluable tool. Protein based biomarkers have been identified but have fallen short of their expectations. Gene based markers are in their infancy. Alterations in lipid metabolism have been shown to be positively related to onset of various cancers. To establish the role of lipids in cancer, studies have focused on the enzymes, metabolites and other biomolecules related to lipids. In this review an attempt has been made to highlight the progress in research done in this area.

KEY-WORDS: **Metabolism; Lipidomics; Cancer; Biomarker**

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INTRODUCTION

Lipid is an important group of biomolecules with diverse roles. Lipids are the main source of energy for the cell and they also act as key molecules for maintenance of membrane structure. Lipid anabolism and catabolism helps as a molecular integrator of energy homeostasis, membrane structure and function. Attempts to use lipids as biomarkers of cancer have been made. [1] The National Institute of Health (NIH) defines a biological marker (biomarker) as a biological molecule found in blood, other body fluids, or tissues that is an objective indicator of normal or abnormal process, or of a condition or disease. [2] Proteins were the first to be tried and a host of protein molecules have been identified as biomarkers; some of which have stood the test of time and others have been discarded[3]. In contrast to proteins, blood lipid levels are amenable to variations depending on diet. Changes in lipid profile have been very well correlated with coronary diseases. With this premise it is hypothesized that a disease like cancer could also arise from changes in lipid metabolism. The most important criterion for a biomarker is the ability for assessment in a non invasive fashion. Lipid changes can easily be monitored in blood. Over the years researchers have shown a positive correlation between alterations in lipid profile with cancer. This review aims to summarize the research in the field of lipids and cancer.

1. Lipid and Cancer

The most obvious role that lipids have is its function as a store of chemical energy, constituent of cell membrane and signaling. These cellular processes are of critical relevance to cells, which undergo transformation, cancer progression and metastasis. Thus, it is likely that lipids have a determinate role in cancer onset, progression and outcome. Certain classes of lipids are reflective of the cell's physiology, this is more so in cancer cells and tissue. Breast cancer accounts for the largest number of newly diagnosed cases in female cancer patients. Although mammography is a powerful screening tool, about 20% of breast cancer cases cannot be detected by this method. New diagnostic biomarkers for breast cancer are necessary. Recent studies show that serum proteome pattern could be used as a diagnostic tool for early stage of breast cancer. As a result of these studies markers like BRCA-1, BRCA-2, CA 15.3, BR27.29, tissue specific Ag, HER-2, BC-1, BC-2, BC-3 etc have been identified and are used in clinical practice.[2] However, using protein based biomarkers could be very expensive and time consuming process over and above being less specific and sensitive. Apart from this, these markers can be detected only once the cancer has established itself in the breast. Lipid based molecules have gained centre stage in breast cancer research. The abnormal metabolism of lipids is closely related to formation of many human diseases.[5] Cellular biomembrane is composed of 50% lipids and protein. Membrane fluidity, non lamellar phase propensity and formation of micro domains depend upon lipid–lipid and lipid-protein interactions. Ligand receptor interaction, endocytosis and antigen presentation are also controlled by membrane fluidity and specialized membrane micro domains.[6] Lipids play a very important role in regulation of various cellular functions by acting as signaling molecules or as precursors for secondary messenger. These functions include cell proliferation, inflammation, immunity and apoptosis among others.[7] These processes have been shown to be accelerated in cancerous cells leading to uncontrolled growth. As a consequence, due to tight molecular coupling it can be argued that lipid could serve as an excellent source of information on these cellular effects in cancerous growth. There are unique lipid molecules that have been identified during cancerous growth like PIPs, sphingosine-1-phosphate (S1P) and lysophospholipids.[8] S1P functions as an effector molecule which is involved in regulation of various aspects of cancer pathogenesis and therapy.[8] Some tumor regulating functions might be controlled through interaction of sphingolipid with protein.[1, 10, 11, 12] Ceramide transfer protein (CERT) is involved in cancer.[13] The total lipid composition of a cell or organ is defined as the "lipidome" and the study of lipids and their interacting partners is termed "lipidomics". With the rapid development of lipidomics,

more and more attention has been paid to lipids because of their important roles in tumor diagnosis and therapy.[13, 14] Anti-lipid autoantibody is routinely used in the diagnosis of autoimmune diseases, but their potential for cancer diagnosis has not been explored. Dysregulation of cellular signaling in cancer cells would be expected to lead to irregular metabolism of many lipids, which could be sensed by the immune system and cause the production of auto antibodies. Discovery of anti-lipid antibodies could be used as biomarkers for early breast cancer diagnosis.[15] During primary nodule formation in breast cancer the homeostasis of vicinity cells changes tremendously, as a result of disturbed redox reactions. The lipid oxides and lipid peroxide produced in these cells are potentially able to deliver stress to other nodules. In addition lipid peroxides are precursors for the numerous prostaglandins and signaling molecules they are potentially able to activate and enhances expression of numerous lipases. Thus it is very likely that lipid associated changes could be the forerunner in breast cancer and that this could be picked up as a marker. Some of the lipid based biomarkers identified are Apo lipoprotein A1 (ApoA1c), Pro-apolipoprotein A1 (proApoA1a), Apo lipoprotein A4 (ApoA4) and Apo lipoprotein H (ApoH)b2.[4] These however have not yet been commercially exploited. Most work reported with Indian population deals with looking at gross lipid profile changes in breast cancer and non cancer population.[16] Similar studies have also been done on breast cancer patients post treatment with Tamoxifen.[17] To prove a link between lipid metabolism and cancer studies have been done with obese patients. Current estimations are that 20% of all tumors and 50% of endometrial and oesophageal cancers can be attributed to obesity.[18] Obesity contributes to increased cancer risk mainly by causing acquired insulin resistance. Alteration in lipid metabolism can also be a consequence of cancer development a condition known as cancer cachexia. The changes in various classes of lipids like fatty acids, triglycerides, cholesterol, phosphoglycerides, sphingomyelins, and ceramide in cancer has been studied and some of these are listed below.

1.1. Phosphoglycerides

Phosphoglycerides are the main component of cell membrane. They play an important role in membrane function. Phosphoglycerides are of four types, Phosphatidylcholine Phosphatidylethanolamine, Phosphatidylserine, Phosphatidyinsitol. During cancer the levels of all were shown to be altered and this could be taken as a hallmark of cancer. Phosphatidylcholine Phosphatidylethanolamine, Phosphatidylserine, Phosphatidyinsitol levels were found to be decreased in colorectal cancer.[19] In colon cancer, only Phosphatidylcholine level increased.[20] In leukemiaPhosphatidylcholine Phosphatidylethanolamine, and Phosphatidyinsitol level were decreased.[21] Phosphatidylserine, Phosphatidyinsitol and Phosphatidylcholine levels are significantly altered in esophageal cancer.[22] The main role for phosphoinositides is in cell signaling. During cancer these functions are disturbed due to alterations in their levels. This is reported in breast, ovarian, lung, prostate and gastric cancer.[8] In Human gliomas PC/PE ratio is increased during malignancy.[23] In malignant neoplasm cell with metastases high PC/PE ratio was observed as compared to non-metastatsic malignant neoplasm cell.[23] Phosphorylation of Phosphatidyinsitol by PI3 kinase has been shown to be hyper activated in some cancers. Downstream signaling via Akt which is activated by phosphorylated PIs is also dysregulated in hepatic, ovarian, thyroid and prostate cancer tissues.[25, 26] Thus, PIs and their immediate upstream and downstream metabolites (e.g. DAG and PA) which are the first line indicators for cellular signaling may contribute to the onset of cancer pathology.

1.2 Sphingolipids

Bioactive sphingolipids such as ceramide, sphingosine-1-phophate (S1P), sphingosine, and glucosyl ceramide, act as effector molecules in cancer pathogenesis.[27] Sphingomyelins are most abundant sphingolipids in various cellular membranes and are also substrates for sphingomyelinases. Its main function is to maintain structure of cell membrane and in cell signaling. Sphingomyelin levels are increased in breast and lung cancer but

decrease in leukemia and human meningiomas. S1P is considered to be a 'prosurvival' factor, as it is involved in malignant transformation, cancer proliferation, inflammation and vasculogenesis.[1,28] Increased generation of S1P triggers signaling pathways that mediate these pro-survival processes mainly by engaging with S1P receptors 1–5 (S1PR), members of the family G-protein coupled receptors. Increased S1P levels promote proliferation and survival in human glioma and breast cancer cells. [29, 30] S-1-P levels are elevated in ovarian cancer and S-1-PR1 has also been shown to be a requirement for tumor angiogenesis *in vivo*.[31]

1.3 Ceramides

Ceramide is a product of sphingosine. Sphingomyelinases liberate choline from the sphingomyelin headgroup thus generating ceramides in the membrane. Some tumor regulating functions might be controlled through interactions of sphingolipids with proteins. Ceramides have some important function in cellular process like cell growth, mitotic maturation, senescence, and apoptosis. Targets of ceramide include protein phosphatases and kinases that regulate important signaling pathways in cancer. Ceramide level are decreased in ovary and glail tumours. Ceramide is metabolically closely linked with glucosyl ceramide via activity of glucosyl ceramide synthase (GCS) which is highly expressed in some cancer cells.[32] They have been assigned a key role in tumor progression, ceramide analogues/mimetics, and inhibitors of sphingosine kinase or enzymes of ceramide clearance could be exploited for use as cancer biomarkers.[9, 33]

1.4. Lysophospholipids

Lysophosphatidic acids (LPAs), which are structurally similar to sphingosine-1-phosphate, have also been implicated for their role in metastasis. These lipids are released from cells and show both autocrine and paracrine activity by binding to G-protein-coupled receptors (GPCRs). Levels of LPAs are increased in serum of patients with ovarian tumors and are being now considered as potential biomarkers for ovarian cancer[33]. The role of LPA inactivating PI3K and phospholipase C resulting in cell proliferation and cell survival has been elucidated. LPA signaling regulates cancer cell proliferation, invasion, angiogenesis, and biochemical resistance to chemotherapy and radiotherapy-induced apoptosis Tumor cell invasion due to LPA is as a result of the receptor mediated activation of the Rho and Rac GTPase pathways, known to be essential for the regulation of the actin cytoskeleton and cell motility.[35, 36]

1.5. Fatty Acids

Fatty acids include free fatty acids, fatty acid amides, prostanoids, hydroxyl and hydro peroxy eicosaenoic acids, leukotrienes and epoxy eicosatrienoic acids. Polyunsaturated free fatty acids are particularly involved in intracellular signaling. The level of fatty acids has been reported to change in the process of many cancers. For example, it has been shown that in hepatocellular carcinoma (HCC) patients, the plasma free fatty acids levels were slightly decreased.[36] By comparing human meningiomas and normal leptomeninges, it was found that unsaturated fatty acids increased, while saturated fatty acids decreased in meningiomas.[37] In colorectal cancer, a decrease in the amount of linoleic acid (LA) and alpha linolenic acid (ALA) was shown, while arachidonic and oleic acids were increased.[38] Omega-3 fatty acids which have always had a pride of place as protective molecules, however they were shown to have detrimental effects in a recent study.[39]

1.6. Triglycerides

Triglyceride levels in plasma are associated with various types of cancer. In a study of lipid profiles of breast and ovary cancer patients, it was found that in breast cancer patients there is a moderate increase in the plasma levels of triglycerides (18%); in ovarian cancer patients, there is a high decrease in the plasma levels of triglycerides (31%); in gynecologic cancers other than breast and ovarian cancer, there is a moderate decrease in plasma levels of triglycerides (25%).[40] In addition, plasma levels of triglycerides could discriminate between patients with benign breast disease and breast cancer patients, as higher triglycerides levels were associated with increased breast cancer risk.[41] Plasma lipid profiles could also change in HCC and in the

majority of reports about HCC, plasma levels of triglycerides were slightly to significantly decreased, while in certain cases plasma levels of triglycerides might be increased.[35] The plasma triglycerides levels were also significantly lower in untreated head and neck cancer patients compared to patients with oral precancerous conditions (OPC) and controls.[42] On the contrary, in colorectal carcinoma the serum triglyceride levels tend to increase.

1.7. Cholesterol

Cholesterol is one of the most essential components which is required for normal function of membrane. It plays important role in membrane fluidity. Cholesterol also serves as precursor for many steroid hormones. It also plays important function in intracellular transport, cell signaling and nerve conduction. Its levels are found to be altered in many disease conditions, like diabetics, heart disease and cancer. For e.g. in breast cancer patients there is moderate increase in the plasma levels of cholesterol (21%) and a high increase in LDL-cholesterol (43%), while there is a decrease in HDL-cholesterol levels (30%) when compared to normal subjects.[40] In benign breast disease, total cholesterol was also significantly lower compared to controls.[41] Similarly, in ovarian cancer patients, there is a decrease in the plasma levels of cholesterol when compared to control group.[43] In untreated head and neck cancer patients, a significant decrease in plasma total cholesterol and HDL-cholesterol was observed.[42] In gynecologic cancers other than breast and ovarian cancer, there is a moderate decrease in plasma levels of the cholesterol (21%) and HDL-cholesterol levels (27%), while a non significant decrease in LDL-cholesterol (6.2%) on comparison with control group.[40] Plasma cholesterol levels also decreased in HCC and decreased serum levels of cholesterol may indicate a poor prognosis.[36] In non diabetics prostate carcinoma HDL cholesterol was significantly decreased whereas LDL cholesterol was higher as compared to control.[44]

CONCLUSION

Lipids seem to have a much greater role to play in the progression and formation of cancer. Advent of newer techniques and availability of human blood sample will help identify new markers for cancer. Our laboratory is looking at the changes in lipid in blood in breast cancer patients.

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