Technology in Cancer Research & Treatment
Volume 20: 1-8

DOI: 10.1177/15330338211041472
journals.sagepub.com/home/tct
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Roles of Lipid Profiles in Human Non-Small Cell Lung Cancer

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Abstract

Aims: This review aims to identify lipid biomarkers of non-small cell lung cancer (NSCLC) in human tissue samples and discuss the roles of lipids in tissue molecular identification, the discovery of potential biomarkers, and surgical margin assessment. **Methods:** A review of the literature focused on lipid-related research using mass spectrometry (MS) techniques in human NSCLC tissues from January 1, 2015, to November 20, 2020, was conducted. The quality of included studies was assessed using the QUADAS-2 tool. **Results:** Twelve studies met the inclusion criteria and were included in the review. The risk of bias was unclear in the majority of the studies. The contents of lipids including fatty acids, phosphatidyl choline, phosphatidyl ethanolamine, phosphatidyl inositol, cardiolipin, phosphatidyl serine, phosphatidyl glycerol, ceramide, lysophosphatidylethanolamine, lysophosphatidylcholine, and lysophosphatidylglycerol differed significantly between cancer and healthy tissues. The sensitivity or specificity of the discrimination model was reported in 8 studies, and the sensitivity and specificity varied among the reported methods. The lipid profiles differed between adenocarcinoma and squamous cell carcinoma NSCLC subtypes. **Conclusion:** In preclinical studies, MS analysis and multiple discrimination models can be combined to distinguish NSCLC tissues from healthy tissues based on lipid profiles, which provides a new opportunity to evaluate the surgical margin and cancer subtype intraoperatively. Future studies should provide guidance for selecting patients and discrimination models to develop an improved method for clinical application.

Keywords

non-small cell lung cancer, lipids, mass spectrometry, discrimination model, cancer subtype

Abbreviations

AC, adenocarcinoma; AFADESI-MSI, air flow-assisted desorption electrospray ionization mass spectrometry imaging; Cer, ceramide; CL, cardiolipin; DESI-MSI, desorption electrospray ionization mass spectrometry imaging; DG, diacylglycerol; DOPC, oleoyl phosphatidylcholine; DPPC, dipalmitoyl phosphatidylcholine; ESI-MS, electrospray ionization mass

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spectrometry; FA, fatty acids; FFA, free fatty acids; LC-MS, liquid chromatography mass spectrometry; LE-ESSI-MSI, liquid extraction electrosonic spray ionization mass spectrometry imaging; LPC, lysophosphatidylcholines; LPE, lysophosphatidylethanolamine; LPG, lysophosphatidylglycerol; MALDI-MS, matrix-assisted laser desorption/ionization mass spectrometry; nanoESI, nanoelectrospray ionization; OPLS-DA, orthogonal projections to latent structures discriminant analysis/orthogonal partial least squares discriminant analysis; PC, phosphatidyl choline; PCA, principal component analysis; PCA-LDA, principal component analysis and linear discriminant analysis; PE, phosphatidyl ethanolamine; PG, phosphatidyl glycerol; PI, phosphatidyl inositol; PLS-DA, partial least squares-discriminant analysis; PLS-LDA, partial least squares linear discriminant analysis; POPC, phosphatidylcholine; PS, phosphatidyl serine; SAPC, arachidonic acid stearoyl phosphatidylcholine; SCC, squamous cell carcinoma; large, large-cell lung carcinomas; SM, sphingomyelin; TG, triglyceride; TSI-MS, tissue spray ionization mass spectrometry; iEESI-MS, internal extractive electrospray ionization mass spectrometry.

Received: March 10, 2021; Revised: June 22, 2021; Accepted: August 5, 2021.

Introduction

Non-small cell lung cancer (NSCLC) is the leading cause of cancer death worldwide.[1] NSCLC, which includes adenocarcinoma (AC), squamous cell carcinoma (SCC), large-cell lung carcinomas, and adenosquamous carcinoma, accounts for approximately 85% of all lung cancers.[2] Currently, chest radiography, computerized tomography, bronchoscopy, endoscopic ultrasound, endobronchial ultrasound, and pathological diagnosis are the main diagnostic strategies for lung cancer.[3–5] The treatment options and overall survival rate of NSCLC depend on the stage. Surgical resection is the gold-standard treatment for early-stage NSCLC.[6–8] Unfortunately, 3.6% to 60% of patients with NSCLC suffer recurrence after surgery.[9–13] Furthermore, the recurrence rate of stage IB NSCLC patients with negative pathological margins was reported to be 12.7% to 13.79%.[14] This suggests that the lung cancer surgical margin determined by conventional histologic pathological diagnosis may not provide reliable information from onco-biology, and emerging evidence suggests that current onco-surgical techniques are frequently inadequate.[15–18] Therefore, there is an urgent need to develop a novel technique or device for accurately assessing the molecular margin based on tumor biology.

Lipidomics is a new field that focuses on lipids and their interactions in objects, tissues, and cells.[19] Lipids including fatty acids, glycerolipids, glycerophospholipids, sphingolipids, sterol lipids, prenol lipids, saccharolipids, and polyketides are currently understood as being relevant to tumor biology. Phospholipids including sphingolipids and phosphoglycerides are the major structural molecules of cell membranes and have important roles in maintaining barrier function and fluidity.[20] Phospholipids also act as the second messenger in intercellular and intracellular signal transduction.[21] Sphingolipids including sphingomyelin and glycosphingolipid regulate cancer cell death and survival by controlling cancer cell signal transduction.[22] Phosphoglycerides including phosphatidyl choline (PC), cardiolipin (CL), phosphatidyl ethanolamine (PE), phosphatidyl serine (PS), phosphatidyl glycerol (PG), and phosphatidyl inositol (PI) play important roles in activating cells, maintaining metabolism, and enhancing human immunity and regeneration.[20,23] In cancer cells, fatty acids (FAs) play crucial roles in membrane formation, energy storage, and signaling.[24]

Tumor molecular analysis has attracted considerable attention as it can provide additional information to incorporate cancer-related biomarkers into clinical decision-making.[25–27] The majority of analytical methods for determining lipid profiles are based on mass spectrometry (MS). Ambient MS analysis allows the extensive and rapid analysis of metabolites, resulting in multiparameter datasets containing quantitative information on a range of metabolites.[28,29] Recently, based on the direct profiling of molecules from various biological samples, MS methods for identifying tumor tissues have advanced rapidly.[5,30,31] In particular, the lipid profiling of biological tissues by MS has been developed for the molecular diagnosis of cancers.[5,30] This review focuses on recent applications of MS for lipid profiling in human NSCLC tissues and discusses the roles of lipids in discriminating cancer from healthy tissue, evaluating tumor stage and subtype, and assessing disease-specific biomarkers of NSCLC. Specifically, this review summarizes the applications of MS for the molecular diagnosis of NSCLC, including tissue molecular identification, the discovery of potential biomarkers, and surgical margin assessment. Finally, this review discusses the challenges and future perspectives for the use of MS in the precise clinical molecular diagnosis of NSCLC.

Methods

A literature search of papers published in English was performed using *Web of Science* and *PubMed* (*Medline*). The search period was from January 1, 2015, to November 20, 2020, and the search focused on studies of lipid profiling in human NSCLC tissue. The search strategy employed a combination of MeSH terms: “mass spectrometry” [All Fields] AND (“lung neoplasms” [MeSH Terms] OR (“lung” [All Fields] AND “neoplasms” [All Fields]) OR “lung neoplasms” [All Fields] OR (“lung” [All Fields] AND “cancer” [All Fields]) OR “lung cancer” [All Fields]) AND (“lipids” [All Fields] OR “lipidate” [All Fields] OR “lipidated” [All Fields] OR “lipidates” [All Fields] OR “lipidation” [All Fields] OR “lipidations” [All Fields] OR “lipide” [All Fields] OR “lipides” [All

Fields] OR "lipidic" [All Fields] OR "lipids" [MeSH Terms] OR "lipids" [All Fields] OR "lipid" [All Fields]). Two reviewers independently screened the potentially relevant studies by title and abstract via electronic search. The references of the returned papers were also searched to find additional potentially relevant articles. We included studies on the lipid profiles for human cancer and matched normal tissues from patients with NSCLC. Only studies that included MS as an analytical platform were included. The following studies were excluded: studies on biological samples other than tissue; cell line studies; animal studies; and studies without a control group. The following data were independently extracted from the selected studies by 2 reviewers: primary author; year of publication; study country/region; cancer subtype(s); analytical platform(s); discriminant model(s); sensitivity and specificity (if reported); and significantly different metabolites between the cancer and control groups. The main outcome measure was lipid abundance that was determined to be significantly different between cancer and normal samples. The quality of each included study was assessed using the Quality Assessment of Diagnostic Accuracy Studies-2 (QUADAS-2) tool. The risk of bias was unclear in the majority of studies.

Results

Study Characteristics

Twelve studies met the inclusion criteria and were selected for review. The search results are shown in Supplemental Figure 1. The analytical platforms used for lipid detection included liquid chromatography–MS,[32–35] electrospray ionization–MS,[36] nanoelectrospray ionization,[37] air flow-assisted desorption electrospray ionization MS imaging,[38,39] liquid extraction electrosonic spray ionization MS imaging,[40] matrix-assisted laser desorption/ionization MS (MALDI-MS),[35,36] MasSpec Pen,[16] desorption electrospray ionization,[16] tissue spray ionization MS,[41] and internal extractive electrospray ionization MS.[17] The cancer types included AC, SCC, large-cell lung carcinoma, and others (Table 1).

Lipid Profiling in Human NSCLC

FAs and phospholipids have been extensively studied in human NSCLC. The profiles of FAs were found to be significantly different between the cancer and control samples in 4 studies on NSCLC. There is a general trend of FA upregulation in cancer compared to normal across the majority of studies. However, Fan et al[32] reported that the abundance of free fatty acids (FFAs) was lower in lung cancer tissue compared to distal noncancerous tissue. Sphingomyelin (SM) was identified as a potential biomarker in 4 studies. In most of the studies, SM was downregulated in tumor tissue compared to normal tissue. However, Kahyo et al found that SM (d35:1) was increased in adenocarcinoma tissue compared to normal from the recurrent group patients.[33] The major structural lipids in membranes are glycerophospholipids including PC, PE, PI, CL, PS, and PG, which have been extensively examined in NSCLC research. Messenger lipids such as lysophosphatidylglycerol (LPG) and ceramide (Cer) were upregulated in tumor tissue compared to normal tissue. One study found that the level of Cer was lower in tumor tissue than in normal tissue due to the conversion of Cer into its derivatives.[32] Another 2 studies found that lysophosphatidylethanolamine, lysophosphatidylcholine (LPC), and LPG were upregulated in tumor tissue compared to in healthy tissue (Table 1).[32,34]

Sensitivity and Specificity of the Discrimination Model in Human NSCLC

The sensitivity and specificity of the diagnostic discrimination model are critical factors. The reviewed studies used the following models to differentiate between tumor tissue and normal tissue (Table 1): partial least squares discriminant analysis (PLS-DA; $n = 2$); principal component analysis (PCA; $n = 3$); orthogonal projections to latent structures discriminant analysis/orthogonal partial least-squares discriminant analysis (OPLS-DA; $n = 5$); PCA and linear discriminant analysis (PCA-LDA; $n = 1$); and partial least squares linear discriminant analysis (PLS-LDA; $n = 1$). The sensitivity or specificity of the discrimination model was reported in 8 studies (Table 2). The sensitivity or specificity varied among these studies. In 3 studies, the sensitivity of the discrimination model reached 100%,[17,33,36] while only one study reported a specificity of 100% (for the OPLS-DA in Li et al[39]). Interestingly, Zhang et al reported sensitivities of 85.2% for AC and 82.1% for SCC using the OPLS-DA model.[38]

Lipid Profiling in Human NSCLC Subtypes

Abundant evidence has shown that the lipid profile is closely related to the NSCLC subtype. Due to differences in research design, analysis methods, discrimination models, and so on, the lipid profiles reported for human NSCLC subtypes show great variations among studies. In 4 of the reviewed studies, the lipid profiles of patients with AC and SCC were analyzed (Table 3). Fan et al reported that PE(20:0_18:1), DG(18:0_20:3), DG(18:2_18:2), PE(22:0_20:4), PE(18:0_20:4), DG(18:1_18:2), DG(36:2), PC(18:1_20:4), DG(16:0_18:2), and Cer(d18:1/26:0) were increased in AC patients compared to SCC patients.[32] Another study found that $[PC(34:0) + Na]^+$, [phosphorylcholine $+ K]^+$, and [phosphorylcholine $+ Na]^+$ were upregulated in AC patients compared to SCC patients.[38] In contrast, You et al[34] found that the FFA22:1, FFA24:1, LPC20:1, and PC38:2 were downregulated in AC compared to SCC.

Discussion

Lipids Profile Research and Surgical Margin Assessment in Human NSCLC

Lipids are involved in energy storage, are components of the cell membrane, and serve as messengers in signal transduction

Table 1. Summary of Included Studies.

Author, country/ region and year of study	Analysis platform	Cancer type	Lipids of cancer compared to control	
			Down-regulated	Up-regulated
Yingying Fan (China 2020)	UHPLC-MS	AC, SCC	FFA, CL, PI, SM, Cer	LPG, PG, TG, PEs, PCs
Yusuke Takanashi (Japan 2020)	LC-MS/MS	AC	TG (15:0_14:0_14:0)	SM (d35:1), Cer (d42:0)
Wenbo Cao (China 2020)	nanoESI-MS	NSCLC	C = C location PC(34:1), C = C location PC(36:1), C = C location PC(36:2)	sn-position isomers PC(34:1), sn-position isomers PC(38:5)
Haiyan Lu (China 2019)	iEESI-MS/MS	Lung cancer	PC(32:0)	PC(34:1)
Lei You (China 2020)	LC-MS	AC, SCC, others	–	LPC(20:1), PC(38:2), LPE(16:0), PE(34:3)
Min Zhang (China 2019)	AFADESI-MSI	AC, SCC	–	FA(20:5); FA(20:3); FA(20:2); FA(22:6); FA(22:5); FA(22:4); FA(22:2); FA(24:4); PC(16:0/18:1); PC(34:1)
Eyra Marien (Belgium 2015)	ESI-MS, MALDI-MS	AC, SCC, Large	SM(40:1),SM(42:1), SM(36:1)	PI(38:3), PI(40:3), PI(38:2)
Lei Guo (China 2020)	LE-ESSI-MSI	AC, SCC, Large, NSCC	SM(34:1), SM(42:2)	FA(20:3), FA(20:4), FA(22:4), FA(22:5), FA(22:6), PG(36:4), PI(34:1), PI(36:3), PI(36:4), PI(38:3), PI(38:4), PI(40:4), PI(40:5), PC(34:1), PC(34:3), PC(34:4), PC(36:3), PC(36:4), PC(38:3)
Jialing Zhang (America 2017)	MasSpec Pen, DESI-MSI	AC, SCC, others	PI(38:4), PE(36:1)	PI(36:1), PG(36:2), PG(34:1), FA(18:1)
Yusuke Muranishi (Japan 2019)	MALDI-IMS, LC-MS	AC	PC(16:0/16:0), SM (42:2)	–
Yiping Wei (China 2015)	TSI-MS	SCC	DPPC, POPC	DOPC, SAPC
Tiegang Li (China 2015)	AFADESI-MSI	AC, SCC	–	PC(16:0/18:2), PC(34:1)

Abbreviations: LC-MS, liquid chromatography mass spectrometry; ESI-MS, electrospray ionization mass spectrometry; nanoESI, nanoelectrospray ionization; AFADESI-MSI, air flow-assisted desorption electrospray ionization mass spectrometry imaging; LE-ESSI-MSI, liquid extraction electrosonic spray ionization mass spectrometry imaging; MALDI-MS, matrix-assisted laser desorption/ionization mass spectrometry; DESI-MSI, desorption electrospray ionization mass spectrometry imaging; TSI-MS, Tissue spray ionization mass spectrometry, iEESI-MS, internal extractive electrospray ionization mass spectrometry; AC, adenocarcinomas; SCC, squamous cell carcinomas; large, large cell lung carcinomas; PC, phosphatidyl choline; PE, phosphatidyl ethanolamine; PI, phosphatidyl inositol; CL, cardiolipin; PS, phosphatidyl serine; PG, phosphatidyl glycerol; LPE, Lysophosphatidylethanolamine; LPC, lysophosphatidylcholines; LPG, lysophosphatidylglycerol; FA, fatty acids; FFA, free fatty acids; SM, sphingomyelin; Cer, ceramide; TG, Triglyceride; DPPC, dipalmitoyl phosphatidylcholine; POPC, phosphatidylcholine; DOPC, oleoyl phosphatidylcholine; SAPC, arachidonic acid stearoyl phosphatidylcholine.

and molecular recognition processes. In cancer cells, lipid metabolism is disordered, which can affect numerous cellular processes such as cell growth, proliferation, differentiation, and motility. Lipids are a major component of cell membranes, and increasing evidence suggests that lipids play a crucial role in the occurrence and development of NSCLC.[30] Since lung cancer is the leading cause of cancer-related death among both men and women, more and more research is focusing on the role of lipid metabolism in lung cancer.[42–45] Increasing evidence suggests that disordered lipid metabolism is important when evaluating tumor stage and metastasis, determining the effectiveness of treatment, and developing new therapeutic targets and disease-specific biomarkers.[46–48] Various biological samples including tissues, surgical aerosols, and sera have been used in studies on lipid metabolism in lung cancer.[5,30] The serum-based lipidomic metabolism of NSCLC has been used to identify serum lipid biomarkers for the early screening and detection and NSCLC and the assessment of NSCLC subtypes.[32,49,50] Interestingly, differences in lipid metabolism in surgical aerosols have been used to determine the boundary of lung cancer intraoperatively based on rapid evaporative ionization MS; using this approach, a sensitivity of 97.7% and a specificity of 96.5% were achieved for the binary classifications of cancer and healthy tissue.[51] In studies on human NSCLC tissue, excellent models for the discrimination of cancer and healthy tissues have been established based on lipids. These methods provide a new opportunity for the real-time, in situ evaluation of surgical margins during surgery based on molecular characterization.

In previous studies, type II alveolar epithelial cells, a type of lung stem cell, were transformed into growing monoclonal lung tumors during active KRAS mutation.[52] Pulmonary surfactant, a complex mixture of phospholipids (85% phosphatidylcholine) and surfactant proteins, was synthesized and secreted by type II epithelial cells.[53] Sin et al suggested that pro-surfactant protein

Table 2. The sensibility and Specificity of Discriminant Model of Included Studies.

Author, country/region and year of study	Discriminant model	Cancer type	Sensibility (%)	Specificity (%)
Yingying Fana (China 2020)	PLS-DA	AC, SCC	94	67
Yusuke Takanashi (Japan 2020)	PCA	AC	100	80
Haiyan Lu (China 2019)	OPLS-DA	Lung cancer	100	51
Lei You (China 2020)	PLS-DA	AC, SCC, others	94.10	84.40
Min Zhang (China 2019)	OPLS-DA	AC, SCC	85.2% of AC, 82.1% of SCC	–
Eyra Marien (Belgium2015)	PCA-LDA	AC, SC, Large	100	96.70
Jialing Zhang (America 2017)	PCA	AC, SCC, others	97.90	95.70
Tiegang Li (China 2015)	OPLS-DA	AC, SCC	93.50	100

Abbreviations: PLS-DA, partial least squares-discriminant analysis; PCA, principal component analysis; OPLS-DA, orthogonal projections to latent structures discriminant analysis/orthogonal partial least squares discriminant analysis; PCA-LDA, principal component analysis and linear discriminant analysis; PLS-LDA, partial least squares linear discriminant analysis; AC, adenocarcinoma; SCC, squamous cell carcinoma; large, large-cell lung carcinomas.

Table 3. Lipids profiling research in human NSCLC Subtype.

Author, country/ region and year of study	Analysis platform	Discriminant model	Differential molecules	Lipids of AC compared with SCC	
				Down-regulated in AC	Up-regulated in AC
Yingying Fan (China 2020)	UHPLC-MS	PLS-DA	PE(20:0_18:1), DG(18:0_20:3), PE-O 39:4, DG(18:2_18:2), PE(22:0_20:4), PE(18:0_20:4), DG(18:1_18:2), DG 36:2, PC(18:1_20:4), DG(16:0_18:2), Cer(d18:1/ 26:0)	–	PE(20:0_18:1), DG(18:0_20:3), DG(18:2_18:2), PE(22:0_20:4), PE(18:0_20:4), DG(18:1_18:2), DG(36:2), PC(18:1_20:4), DG(16:0_18:2), Cer(d18:1/ 26:0)
Lei You (China 2020)	LC-MS	PLS-DA	FFA22:1, FFA24:1, LPC 20:1, PC 38:2, LPE 16:0, PE 34:3, SM 35:2	FFA22:1, FFA24:1, LPC 20:1, PC 38:2,	LPE 16:0, PE 34:3, SM 35:2
Min Zhang (China 2019)	AFADESI-MSI	OPLS-DA	$[PC(34:0)+Na]^+$, $[phosphorylcholine+K]^+$, $[phosphorylcholine+Na]^+$	–	$[PC(34:0)+Na]^+$, $[phosphorylcholine+K]^+$, $[phosphorylcholine+Na]^+$
Eyra Marien (Belgium2015)	ESI-MS MALDI-MS	PCA-LDA	PI36:4, PS40:8, PS42:9, PC40:2, PE44:5, PI36:3, PE42:2, SM36:2	–	–

Abbreviations: LC-MS, liquid chromatography mass spectrometry; ESI-MS, electrospray ionization mass spectrometry; AFADESI-MSI, air flow-assisted desorption electrospray ionization mass spectrometry imaging; MALDI-MS, matrix-assisted laser desorption/ionization mass spectrometry; AC, adenocarcinoma; SCC, squamous cell carcinoma; PC, phosphatidyl choline; PE, phosphatidyl ethanolamine; PI, phosphatidyl inositol; PS, phosphatidyl serine; LPE, lysophosphatidylethanolamine; LPC, lysophosphatidylcholines; FFA, free fatty acid; SM, sphingomyelin; Cer, ceramide; DG, diacylglycerol; PLS-DA, partial least squares discriminant analysis; PCA, principal component analysis; OPLS-DA, orthogonal projections to latent structures discriminant analysis/orthogonal partial least squares discriminant analysis; PCA-LDA, principal component analysis and linear discriminant analysis; PLS-LDA, partial least squares linear discriminant analysis.

B can serve as a biomarker to predict lung cancer.[54] Furthermore, Umeda et al[55] reported that higher levels of serum surfactant protein D were correlated with a lower number of distant metastases and prolonged overall survival and progression-free survival. The phospholipids in pulmonary surfactant are mainly in the form of dipalmitoylphosphatidylcholine (DPPC). In a previous study, we found that in SCC patients, cancer tissue contained less DPPC than healthy tissue.[41] These findings suggest that abnormal pulmonary surfactant and the destruction of type II cell homeostasis are closely related to the occurrence of lung cancer.

Lipid Profiles for NSCLC Screening

Previous studies suggest that plasma phospholipids including phosphatidylcholine and LPC are potential biomarkers of lung cancer.[56] Louis et al reported that the concentrations of SM and phosphatidylcholine were decreased in the blood plasma of lung cancer patients compared to normal.[57] Increasing research is focusing on the analysis of serum lipid profiles and the identification of potential serum phospholipids as biomarkers for the diagnosis and screening of human NSCLC.[49,50] Serum phospholipid may be suitable as biomarkers for convenient lung cancer

screening without the need for radiation. Due to the complex composition of blood plasma, the specificity and sensitivity of reported detection methods vary, making these methods difficult to apply in clinical diagnosis.

Lipid Profiling for the Evaluation of Human NSCLC Stage and Subtype

The need for clinical treatment for lung cancer is decided based on the tumor stage and subtype. Lipid metabolism of NSCLC subtypes is critical for understanding the mechanisms, identifying potential diagnostic biomarkers, and targeted therapy. The differences in the characteristics of lipids between human AC and SCC tissues have been studied. In 2012, the lipid profiles of AC and ACC tissues were detected by MALDI-MS,[58] providing a strategy to discriminate the 2 major histologic types of NSCLC. Subsequently, in 2015, Marien et al[36] revealed that the lipids in AC and SCC tissues were different. Increasing evidence suggests that lipids are potential biomarkers of AC and SCC.[32,34,36,38] Interestingly, Takanashi et al[33] found that SM (d35:1) can be used as a predictor for lung AC recurrence after surgery. The different lipid profiles of AC and SCC tissues provide a way to evaluate tumor stage and prognosis and guide treatment decisions for different tumor subtypes.

Advantages and Applications of Lipid Profiles to Diagnosis Human Non-Small Cell Lung Cancer

In situ and real-time identification of tumor margins are essential for tumor surgery to obtain reliable curative resection and accurate prognosis, as well as to minimize losses of healthy tissue.[41] Frozen section histology is the gold-standard method to accurately determining tumor margin during surgery, but this approach suffers from shortcomings, such as time-consuming (mean turnaround time of 30-40 min), subjective interpretation of the results (diagnosis results depend on the experience of the pathologist), increased risk of surgery (during this time the patient remains under anesthesia), and so on.[41,51,59] Lipids are one of the important units for the human, and it plays a key role in activating cells, maintaining metabolism, enhancing human immunity and regeneration, maintaining cell barrier function, controlling cell signal transduction and so on.[20–24] Most importantly, lipids constitute the outermost structure of cells, which is beneficial for the diagnosis of diseases. MS analysis is increasingly applied to rapid and accurate lipids diagnosis of cancer, based on the advantages of real-time and direct analysis of lipids information from biological samples without or minimal sample pretreatment.[5] As mass spectrometry can provide rich lipid information on and in the tissue which can be correlated with pathology. In situ and real-time lipid profiles analysis by MS may be an alternative to standard frozen-section histology. Due to no/minimal sample pretreatment, numerous MS techniques allow the direct lipid profiling from tissues, showing potential in the real-time diagnosis of lung cancer.[16,17,32–41] The application of lipid biomarkers and multiple discrimination models to lung cancer detection has only been demonstrated in laboratory-based studies. Unfortunately, the risk of bias for patient selection was unclear in the reviewed studies. It is unclear whether consecutive patient data were included or whether highly selected samples were chosen. The application of lipid biomarkers and multiple discrimination models to lung cancer detection has only been demonstrated in laboratory-based studies. Farther, there is a lack of standards, including study designs, models, objects, methods, and analyses, which might lead the data was chosen to report. Future researches should design random, double-blind, multicenter, use a unified distinction model to evaluate the feasibility of lipid profile for diagnosis NSCLC compare with the immunohistochemistry results.

Conclusion and Perspectives

The review demonstrates the variations in the relative abundances of the metabolites of phospholipids and FAs in tissue samples from patients with NSCLC. MS can be combined with different discrimination models to distinguish cancer tissue from healthy tissue based on differences in lipid composition, providing a new way to evaluate surgical margins in situ and in real time in intraoperative settings. In situ and real-time lipid profiles analysis by MS may be an alternative to standard frozen-section histology. Future studies should provide details on how the patients and discrimination model(s) were selected and develop a suitable analysis method for clinical application.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Ethical Approval

Not applicable, because this article does not contain any studies with human or animal subjects.

Funding

This work was supported by the National Nature Science Foundation of China grant (NSFC nos. 82060390, 32070764, 81860379, 81800087); Science and Technology Support Program of Sichuan province Nos. 2017SZ0129; "1.3.5 project for disciplines of excellence, West China Hospital, Sichuan University (ZYJC18021)."

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Supplemental Material

Supplemental material for this article is available online.

References

1. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2020. *CA Cancer J Clin*. 2020;70(1):7-30.

2. Herbst RS, Morgensztern D, Boshoff C. The biology and management of non-small cell lung cancer. *Nature*. 2018;553(7689): 446-454.
3. National Lung Screening Trial Research T, Aberle DR, Adams AM, et al. Reduced lung-cancer mortality with low-dose computed tomographic screening. *N Engl J Med*. 2011;365(5): 395-409.
4. Alexander M, Kim SY, Cheng HY. Update 2020: management of non-small cell lung cancer. *Lung*. 2020. 198(6):897-907.
5. Lu H, Zhang H, Wei Y, Chen H. Ambient mass spectrometry for the molecular diagnosis of lung cancer. *Analyst*. 2020; 145(2):313-320.
6. McDonald F, De Waele M, Hendriks LEL, Faivre-Finn C, Dingemans AMC, Van Schil PE. Management of stage I and II nonsmall cell lung cancer. *Eur Respir J*. 2017;49(1).
7. Yang HX, Woo KM, Sima CS, et al. Long-term survival based on the surgical approach to lobectomy for clinical stage i nonsmall cell lung cancer: comparison of robotic, video-assisted thoracic surgery, and thoracotomy lobectomy. *Ann Surg*. 2017; 265(2):431-437.
8. Huber RM, De Ruysscher D, Hoffmann H, Reu S, Tufman A. Interdisciplinary multimodality management of stage III nonsmall cell lung cancer. *Eur Respir Rev*. 2019;28(152).
9. Uramoto H, Tanaka F. Recurrence after surgery in patients with NSCLC. *Transl Lung Cancer Res*. 2014;3(4):242-249.
10. Kamigaichi A, Tsutani Y, Fujiwara M, Mimae T, Miyata Y, Okada M. Postoperative recurrence and survival after segmentectomy for clinical stage 0 or IA lung cancer. *Clin Lung Cancer*. 2019;20(5):397-403. e391.
11. Conforti F, Pala L, Pagan E, et al. Effectiveness of intensive clinical and radiological follow-up in patients with surgically resected NSCLC. Analysis of 2661 patients from the prospective MAGRIT trial. *Eur J Cancer*. 2020;125:94-103.
12. Brown LM, Louie BE, Jackson N, Farivar AS, Aye RW, Vallieres E. Recurrence and survival after segmentectomy in patients with prior lung resection for early-stage non-small cell lung cancer. *Ann Thorac Surg*. 2016;102(4):1110-1118.
13. Uramoto H, Tanaka F. Prediction of recurrence after complete resection in patients with NSCLC. *Anticancer Res*. 2012; 32(9):3953-3960.
14. Fang Z, He J, Fang W, Ruan L, Fang F. Long-term outcomes of thoracoscopic anatomic resections and systematic lymphadenectomy for elderly high-risk patients with stage IB non-small-cell lung cancer. *Heart Lung Circ*. 2016;25(4):392-397.
15. Pirro V, Alfaro CM, Jarmusch AK, Hattab EM, Cohen-Gadol AA, Cooks RG. Intraoperative assessment of tumor margins during glioma resection by desorption electrospray ionization-mass spectrometry. *Proc Natl Acad Sci USA*. 2017;114(26):6700-6705.
16. Zhang JL, Rector J, Lin JQ, et al. Nondestructive tissue analysis for ex vivo and in vivo cancer diagnosis using a handheld mass spectrometry system. *Sci Trans Med*. 2017;9(406).
17. Lu HY, Zhang H, Chingin K, et al. Sequential detection of lipids, metabolites, and proteins in one tissue for improved cancer differentiation accuracy. *Anal Chem*. 2019;91(16):10532-10540.
18. Sun CL, Li TG, Song XW, et al. Spatially resolved metabolomics to discover tumor-associated metabolic alterations. *Proc Natl Acad Sci USA*. 2019;116(1):52-57.
19. Brugger B. Lipidomics: analysis of the lipid composition of cells and subcellular organelles by electrospray ionization mass spectrometry. *Annu Rev Biochem*. 2014;83:79-98.
20. van Meer G, Voelker DR, Feigenson GW. Membrane lipids: where they are and how they behave. *Nat Rev Mol Cell Bio*. 2008;9(2):112-124.
21. Spiegel S, Milstien S. The outs and the ins of sphingosine-1-phosphate in immunity. *Nat Rev Immunol*. 2011;11(6):403-415.
22. Ogretmen B. Sphingolipid metabolism in cancer signalling and therapy. *Nat Rev Cancer*. 2018;18(1):33-50.
23. Huang CF, Freter C. Lipid metabolism, apoptosis and cancer therapy. *Int J Mol Sci*. 2015;16(1):924-949.
24. Currie E, Schulze A, Zechner R, Walther TC, Farese RV Jr. Cellular fatty acid metabolism and cancer. *Cell Metab*. 2013; 18(2):153-161.
25. Mertins P, Tang LC, Krug K, et al. Reproducible workflow for multiplexed deep-scale proteome and phosphoproteome analysis of tumor tissues by liquid chromatography-mass spectrometry. *Nat Protoc*. 2018;13(7):1632-1661.
26. Chen FJ, Chandrashekar DS, Varambally S, Creighton CJ. Pan-cancer molecular subtypes revealed by mass-spectrometry-based proteomic characterization of more than 500 human cancers. *Nat Commun*. 2019;10.
27. Zhang JL, Li SQ, Lin JQ, Yu WD, Eberlin LS. Mass spectrometry imaging enables discrimination of renal oncocytoma from renal cell cancer subtypes and normal kidney tissues. *Cancer Res*. 2020;80(4):689-698.
28. Cooks RG, Ouyang Z, Takats Z, Wiseman JM. Ambient mass spectrometry. *Science*. 2006;311(5767):1566-1570.
29. Takats Z, Wiseman JM, Gologan B, Cooks RG. Mass spectrometry sampling under ambient conditions with desorption electrospray ionization. *Science*. 2004;306(5695):471-473.
30. Zhang L, Zhu B, Zeng Y, Shen H, Zhang J, Wang X. Clinical lipidomics in understanding of lung cancer: opportunity and challenge. *Cancer Lett*. 2020;470:75-83.
31. Ferrarini A, Di Poto C, He SS, et al. Metabolomic analysis of liver tissues for characterization of hepatocellular carcinoma. *J Proteome Res*. 2019;18(8):3067-3076.
32. Fan Y, Noreldeen HAA, You L, et al. Lipid alterations and subtyping maker discovery of lung cancer based on nontargeted tissue lipidomics using liquid chromatography-mass spectrometry. *J Pharm Biomed Anal*. 2020;190:113520.
33. Takanashi Y, Funai K, Sato S, et al. Sphingomyelin(d35:1) as a novel predictor for lung adenocarcinoma recurrence after a radical surgery: a case-control study. *Bmc Cancer*. 2020;20(1).
34. You L, Fan Y, Liu X, et al. Liquid chromatography-mass spectrometry-based tissue metabolic profiling reveals major metabolic pathway alterations and potential biomarkers of lung cancer. *J Proteome Res*. 2020;19(9):3750-3760.
35. Muranishi Y, Sato T, Ito S, et al. The ratios of monounsaturated to saturated phosphatidylcholines in lung adenocarcinoma microenvironment analyzed by liquid chromatography-mass

spectrometry and imaging mass spectrometry. *Sci Rep*. 2019; 9(1):8916-8916.
36. Marien E, Meister M, Muley T, et al. Non-small cell lung cancer is characterized by dramatic changes in phospholipid profiles. *Int J Cancer*. 2015;137(7):1539-1548.
37. Cao WB, Cheng SM, Yang J, et al. Large-scale lipid analysis with C=C location and sn-position isomer resolving power. *Nat Commun*. 2020;11(1):375.
38. Zhang M, He J, Li T, et al. Accurate classification of non-small cell lung cancer (NSCLC) pathology and mapping of EGFR mutation spatial distribution by ambient mass spectrometry imaging. *Front Oncol*. 2019;9:804.
39. Li TG, He JM, Mao XX, et al. In situ biomarker discovery and label-free molecular histopathological diagnosis of lung cancer by ambient mass spectrometry imaging. *Sci Rep*. 2015;5.
40. Guo L, Lai ZZ, Wang YM, Li ZL. In situ probing changes in fatty-acyl chain length and desaturation of lipids in cancerous areas using mass spectrometry imaging. *J Mass Spectrom*. 2020.
41. Wei YP, Chen LR, Zhou W, et al. Tissue spray ionization mass spectrometry for rapid recognition of human lung squamous cell carcinoma. *Sci Rep*. 2015;5.
42. Salvador MM, de Cedron MG, Rubio JM, et al. Lipid metabolism and lung cancer. *Crit Rev Oncol Hemat*. 2017;112:31-40.
43. Merino M, Fernandez LP, Moyano MS, et al. Analysis of lipid metabolism genes in advanced small cell lung cancer. *J Thorac Oncol*. 2019;14(10):S431-S431.
44. Cristea S, Coles GL, Hornburg D, et al. The MEK5-ERK5 kinase axis controls lipid metabolism in small-cell lung cancer. *Cancer Res*. 2020;80(6):1293-1303.
45. Hall Z, Ament Z, Wilson CH, et al. Myc expression drives aberrant lipid metabolism in lung cancer. *Cancer Res*. 2016;76(16): 4608-4618.
46. Corbet C, Feron O. Emerging roles of lipid metabolism in cancer progression. *Curr Opin Clin Nutr Metab Care*. 2017;20(4): 254-260.
47. Cao Y. Adipocyte and lipid metabolism in cancer drug resistance. *J Clin Invest*. 2019;129(8):3006-3017.
48. Luo X, Cheng C, Tan Z, et al. Emerging roles of lipid metabolism in cancer metastasis. *Mol Cancer*. 2017;16(1):76.
49. Noreldeen HAA, Du LJ, Li W, Liu XY, Wang YF, Xu GW. Serum lipidomic biomarkers for non-small cell lung cancer in non-smoking female patients. *J Pharmaceut Biomed*. 2020;185.
50. Cheng F, Wen ZF, Feng XD, Wang XM, Chen YJ. A serum lipidomic strategy revealed potential lipid biomarkers for early-stage cervical cancer. *Life Sci*. 2020;260.
51. Balog J, Sasi-Szabo L, Kinross J, et al. Intraoperative tissue identification using rapid evaporative ionization mass spectrometry. *Sci Transl Med*. 2013;5(194).
52. Desai TJ, Brownfield DG, Krasnow MA. Alveolar progenitor and stem cells in lung development, renewal and cancer. *Nature*. 2014;507(7491):190–+.
53. Whitsett JA, Wert SE, Weaver TE. Alveolar surfactant homeostasis and the pathogenesis of pulmonary disease. *Annu Rev Med*. 2010;61:105-119.
54. Sin DD, Tammemagi CM, Lam S, et al. Pro-surfactant protein B As a biomarker for lung cancer prediction. *J Clin Oncol*. 2013; 31(36):4536–+.
55. Umeda Y, Hasegawa Y, Otsuka M, et al. Surfactant protein D inhibits activation of non-small cell lung cancer-associated mutant EGFR and affects clinical outcomes of patients. *Oncogene*. 2017;36(46):6432-6445.
56. Klupczynska A, Plewa S, Kasprzyk M, Dyszkiewicz W, Kokot ZJ, Matysiak J. Serum lipidome screening in patients with stage I non-small cell lung cancer. *Clin Exp Med*. 2019;19(4):505-513.
57. Louis E, Adriaensens P, Guedens W, et al. Detection of lung cancer through metabolic changes measured in blood plasma. *J Thorac Oncol*. 2016;11(4):516-523.
58. Lee GK, Lee HS, Park YS, et al. Lipid MALDI profile classifies non-small cell lung cancers according to the histologic type. *Lung Cancer*. 2012;76(2):197-203.
59. Jianyong Z, Jianjun X, Yongzhong O, et al. Rapid discrimination of human oesophageal squamous cell carcinoma by mass spectrometry based on differences in amino acid metabolism. *Sci Rep*. 2017;7(1):3738.