

## Review Article

# The emerging lncRNAs as novel regulators in lung cancer

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**Abstract:** Following the development of genomic microarrays and high-throughput genome and transcriptome sequencing technologies, it has been discovered that non-coding RNAs without open reading frame are pervasively transcribed in human genome. Of which, long non-coding RNAs (lncRNAs) have been recently recognized to be a novel class of RNA molecules exerting regulatory function in several biological processes. Moreover, the altered expression of lncRNAs plays an oncogenic or tumor suppressive role in prevalent cancer types including lung cancer, which has dramatically replenished our knowledge about the biology of cancer. Though researchers make efforts to illuminate the effect of dysregulated lncRNAs on the carcinogenesis and progression of diverse cancers, the molecular mechanisms by which lncRNAs function remain to be further clarified. In this review, we will concentrate on their aberrant expression and potential functions in improving lung cancer prognosis, diagnosis and therapy.

**Keywords:** lncRNA, lung cancer, biomarker, therapeutic target

## Introduction

Lung cancer with the nearly highest incidence ranks the leading cause of death related to cancers worldwide [1]. Non-small cell lung cancer (NSCLC) and small-cell lung cancer (SCLC) are the two major types. The former type is mainly comprised of adenocarcinoma, the most common subtype, and squamous carcinoma. NSCLC accounts for about 85% of diagnosed lung cancer [2]. There are a plenty of patients at advanced stage when they were diagnosed. The lack of biomarkers for early diagnosis renders them to miss an optimal chance for surgical resection, the only and effective therapy for the early lung cancer [3, 4]. Although the progressive strategies of radiotherapy and chemotherapy have been implemented in the clinical application, they may not show a satisfactory result [5], which may be attributable to primary or acquired drugs resistance [6]. Nowadays, modified chemotherapy combined with immunotherapy or tyrosine kinase inhibitors has received positive feedback. However, it seems that the efficacy of these therapeutics has

reach the plateau. In addition, cancer cells metastasis and recurrence may be critical problems challenging lung cancer therapy [7]. The prognosis for patients with NSCLC has not been notably improved over years, and the overall 5-year survival rate of 15% remains to be increased [8]. Therefore, a deep understanding of the underlying molecular and pathological mechanisms prompting NSCLC is of paramount importance for recognizing novel diagnostic biomarkers, developing more effective therapeutic strategies and improving the prognosis of lung cancer patients.

The studies with advanced genomic technologies have revealed that the large majority of transcriptome sequences are non-coding RNAs lacking ability to code for protein, including small and long non-coding RNAs [9, 10]. The initial transcriptional noise, non-coding RNA, recently has been considered as not only a pivotal biological molecule in stem cell pluripotency, cellular development and metabolism but also a newly emerging regulator in a great diversity of diseases, particularly in cancers [9, 11].

MiRNAs have been determined to be involved in posttranscriptional regulation of gene expression, whereas few lncRNAs are well characterized in detail [12].

LncRNAs, the focus of this review, are mRNA-like transcripts longer than 200nt in size [9], which are loosely grouped into five classes based on their genomic location relative to protein-coding genes: sense, antisense, intronic, intergenic and bidirectional lncRNAs [13, 14]. As a novel member of RNA field, lncRNAs play essential roles in regulating gene expression at transcriptional, posttranscriptional and epigenetic level through interfering transcription, modulating alternative splicing patterns and inducing chromatin and histone modifications [14, 15]. Accumulating researches have exposed the networks of relationship among the members of non-coding RNA family are complex and ubiquitous. LncRNAs as molecular sponges could sequester miRNAs from target genes through competing binding sites of downstream transcripts with miRNAs, which generates the achievement of gene expression control [10, 13].

Dysregulation of lncRNA was thought of an essential factor setting foot in almost every step during tumorigenesis and cancer progression by influencing tumor cell proliferation, suppressing cell death, and inducing angiogenesis, etc [16]. Though more and more lncRNAs have been identified over the past decades, the detailed researches on functional mechanisms in cancers are still sparse. More evidence has unraveled that the aberrant expression of lncRNAs usually happens in lung cancer. This review will highlight the effect of their aberrant expression on lung cancer development, and their potential applications as diagnostic biomarkers and therapeutic targets will be also discussed.

### Oncogenic lncRNAs

#### *MALAT1: metastasis-associated lung adenocarcinoma transcript 1*

MALAT1, also termed as nuclear-enriched abundant transcript 2 (NEAT2) [17], is a highly conserved lncRNA among mammals. This lncRNA extensively exists in multiple tissues and cells besides cancer cells [18]. After the RNases cleavage, MALAT1 is divided into small (mas-

cRNA) and long fragment. The latter selectively localizes to nuclear speckles and is involved in alternative splicing of pre-mRNA through the regulating splicing factors (SFs). MALAT1 is always functionally dysregulated in numerous of human cancers and interacts with transcription factors (TFs) and microRNAs [19]. It acts as an important role in tumor progression and metastasis by targeting metastasis-associated gene expression [17]. Meta-analysis determined its value as an independent and powerful prognostic factor for OS and DFS of cancer patients [20, 21]. Previous studies had reported that MALAT1 transcriptionally and/or post-transcriptionally regulated the expression levels of metastasis-associated genes such as CTHRC1, CCT4, HMMR, ROD1 and CXCL5 to enhance cell mobility and metastasis development in lung cancer [22, 23].

MALAT1 is upregulated in lung cancer via DNA methylation reduction of CpG in the promoter [23]. In order to detect the functional roles of MALAT1, researchers built a MALAT1 knockout model in A549 cell lines using zinc finger nucleases to generate the integration between RNA-destabilizing elements and human genome. They implemented dataset permutations using six different cell lines, three MALAT1 WT samples (A549, WT, GFP) and three MALAT1 knockout samples (KO1-3). Through comparison among the six sample groups, they found that the lncRNA MALAT1 actually modulated gene expression rather than alternative splicing in human lung cancer cells. In addition, scratch assays *in vitro* identified that MALAT1 affected cell motility and migration but not proliferation. Furthermore, A549 and EBC-1 xenograft mouse model, via intravenous and subcutaneous injection respectively, strongly demonstrated for the first time that MALAT1 was correlated with metastasis phenotype of lung cancer [17]. It was confirmed that MALAT1 enhancement promoted cell migratory potential while RNAi-mediated MALAT1 knockdown reduced the invasion of A549 cells and tumor growth [24]. Moreover, MALAT1 participates in epithelial-mesenchymal transition to drive the onset of brain metastasis in lung cancer [25]. Bcl-2 (B-cell CLL/lymphoma 2), an anti-apoptotic protein, was uncovered as a predictor for NSCLC under the condition of weak MALAT1 expression [26]. MALAT1 was also identified as a prognostic marker for patients with lung squa-

mous cell carcinoma [24] and early-stage NSCLC [27].

In cigarette smoke extract (CSE)-exposed human bronchial epithelial (HBE) cells, it was found that MALAT1 was upregulated with the decrease of miR-217 level. MiR-317 was predicted to target the 3' end region of MALAT1 and attenuated the CSE-induced enhancement of luciferase activity of this region, suggesting that the expression level of MALAT1 is negatively regulated by miR-217. When treated with si-MALAT1, malignant transformed HBE cells induced by CSE showed reduced expression of EZH2 (enhancer of zeste homolog 2), H3K27me3 and mesenchymal marker (N-cadherin, Vimentin) and simultaneously increased expression of epithelial marker (E-cadherin). Moreover, EZH2-enhanced H3K27me3 binds to the promoter of E-cadherin [28]. In a word, the peroration may be given that miR-217-silenced MALAT1 regulates the development of epithelial-mesenchymal transition (EMT) through the interaction between EZH2-mediated H3K27me3 and the markers of EMT. These data proposed lncRNA MALAT1 to be a promising therapeutic target.

*ANRIL: antisense non-coding RNA in INK4 locus*

ANRIL is 3.8 kb long and transcribed reversely from INK4A-ARF-INK4B gene cluster [Gibb, 2011 #5]. Collecting evidences suggest that ANRIL directly recruits polycomb repressive complex PRC1 and PRC2 to *in cis* or *trans* epigenetically silence tumor suppressor genes, such as p15, p16 and ARF, which are encoded from INK4A-ARF-INK4B gene locus. ANRIL is involved in a wide variety of human cancers, such as leukemia, melanoma, lung and bladder cancers [9, 29, 30]. ANRIL is also activated by transcription factor E2F1 during DNA damage [30]. Highly upregulated ANRIL expression was indicated in NSCLC tissues. The aberrant expression of ANRIL related to TNM stage, lymph node metastasis and tumor size. It could powerfully predict the prognosis of NSCLC patients. Si-ANRIL was found to suppress cell proliferation via cell cycle arrest and apoptosis advancement. The effect of ANRIL on oncogenesis was further confirmed in experiments *in vivo*. Moreover, the enforced expression level of ANRIL forced the ability of cell migration. The

researches about functional mechanisms of ANRIL showed that it inhibited KLF2 (Kruppel-like factor 2) and p21 transcription via EZH2 interaction. KLF2, a member of KLF family, serves as tumor suppressor to enhance cell apoptosis and inhibit cell proliferation and migration. These data demonstrate that KLF2 deletion maybe the downstream component of ANRIL and plays the oncogenic role for tumor development and progression [29, 31]. However, another research apparently had an inverse result that PLD (Phospholipase D) inhibition-induced upregulation of ANRIL stimulated lung cancer cell apoptosis and autophagy. In this respect, ANRIL functions as tumor suppressors to promote cell death [32]. Therefore, there is a intriguing decision that ANRIL has a dual role in tumorigenesis and development, which provides a base to develop promising therapeutic strategies to control human lung cancer for patients.

*HOTAIR: HOX transcript antisense intergenic RNA*

HOTAIR, a ~2.2 kb lncRNA localized to HOXC cluster on chromosome 12, possesses two important binding sites for PRC2 and LSD1 (histone demethylation lysine specific demethylase 1)-CoREST-REST complex at the 5'- and 3'-end, respectively. Binding to them, HOTAIR results in H3K27 methylation or H3K4me2 demethylation, so it epigenetically silences the expression of target genes, such as HOXD cluster, through reprogramming chromatin state [9, 10].

Several researches investigated the expression pattern of HOTAIR in primary NSCLC tissues and cell lines compared with paired non-cancerous and metastatic counterparts using qRT-PCR. It was revealed that HOTAIR expression was the highest in metastatic ones and the lowest in normal ones [33-35]. The differential expression was indicative that HOTAIR conferred aggressive characteristics to lung cancer cells. Moreover, a recent study showed higher expression level of HOTAIR in pure SCLC compared with combined tissues [36]. HOTAIR expression is regulated by several factors such as TFs. A recent study demonstrated that hypoxia-inducible factor-1 $\alpha$  (HIF-1 $\alpha$ ) induced lncRNA HOTAIR expression through binding to the hypoxia-responsive element (HRE) in the promoter region of HOTAIR under hypoxic condi-

tions, which was associated with the aggressive phenotype of hypoxic NSCLC cells [37]. In the NSCLC microenvironment, type I collagen (Col-1) also upregulated the expression of HOTAIR through activating the reporter gene regulated by HOTAIR promoter [38].

The upregulated level of HOTAIR consumedly correlates with tumor size, TNM stage and lymphatic and/or vascular metastasis [34, 35]. Patients with upregulated HOTAIR expression are disposed to be recurrent and high HOTAIR expression nearly means poor prognosis. In SBC-3 cells, knocking down HOTAIR led to reduced cellular proliferation and invasion by regulating cell adhesion-related genes [36]. However, other reports thought forced HOTAIR did not influence or even reduced the proliferation of NSCLC cells [34, 35]. *In vitro* migration and invasion assays and *in vivo* models proved that HOTAIR was responsible for the potential of motility and invasion essential for metastasis [33, 35, 36].

The knockdown of HOTAIR resulted in a decrease of matrix degradation *in situ*, but the different expression of the hallmarks (E-cadherin, N-cadherin and Vimentin) of EMT process between si-HOTAIR and si-control group was not observed. In addition, inhibition of HOTAIR upregulated the HOXA5 protein expression, which participated in the migration and invasion of NSCLC. These data are suggestive of the fact that HOTAIR promotes cell migration through regulating matrix metalloproteinases (MMPs) activity and HOXA5 expression but not EMT process [33, 35].

Taken together, HOTAIR promotes cell proliferation, tumor metastasis and replase, and induces drug resistance and hypoxia-activated malignant transformation in lung cancer.

### *Sox2ot: Sox2 overlapping transcript*

Sox2ot is located in an amplified region on chromosome 3q26.33 [39] and regulates pluripotency via interaction with pluripotency key factors Sox2 and OCT4 gene [40, 41]. Sox2ot is upregulated with its DNA copy number increase particularly in lung squamous cell carcinomas (SCC). In the investigation of the correlation between clinicopathologic factors and Sox2ot, it was revealed that only histological type was correlated to Sox2ot expression level when sta-

tistical *P* value <0.05. For NSCLC patients, the higher Sox2ot expression level is, the shorter overall survival (OS) is. Cox hazard regression analyses showed Sox2ot expression as a independent predictive factor for survival rate. Knocking down Sox2ot expression inhibited lung cancer cell proliferation and accumulated cells in G2/M phase due to lower expression of Cyclin B1 and Cdc2 protein. Additionally, treatments with si-Sox2ot could decrease the expression of EZH2 mRNA and protein. Interestingly, overexpressed EZH2 rescued Sox2ot silence-induced G2/M arrest by restoring cell cycle-regulating proteins. Together, the loss of Sox2ot mitigated cell proliferation by EZH2 regulating cell cycle [39]. Besides, si-Sox2ot caused a significant decline in cell mobility [42]. More investigations on Sox2ot function and the role of Sox2ot as a drug target indeed will promote to develop the therapy of NSCLC.

### *CCAT2: Colon cancer-associated transcript 2*

CCAT2 is transcribed from a highly conserved region on chromosome 8q24. It was previously described as an oncogene in colorectal and breast cancer. It was reported that CCAT2 was significantly overexpressed in lung cancer cell lines NCI-H1975 and had nothing to do with clinicopathological factors except for histological types. That is, the upregulation of CCAT2 was remarkably associated with lung adenocarcinoma. However, researchers observed that its expression level was not obviously in higher level in lymph node-negative NSCLC. The down-regulation of CCAT2 in NCI-H1975 cell lines transfected with CCAT2-targeting siRNA suppressed cell proliferation and invasion. Moreover, together with tumor biomarkers in serum such as CEA, CCAT2 functions efficiently as a predictor for lymph node metastasis in lung cancer (AUC=0.681) [7].

### *CARLo-5: cancer-associated region long non-coding RNA*

CARLo-5 was firstly identified in colon cancer. CARLo-5 is located on chromosome 8q24.21, a gene desert region containing an oncogenic MYC family, which is indicative of the fact that MYC regulates CARLo-5 transcriptional expression by binding to its promoter region. Through the analysis of expression profile of CARLo-5, largely upregulated level was demonstrated in NSCLC cell lines (SPC-A1, NCI-H1975) and tis-

sues, and the tight correlation between CARLo-5 and clinical characteristics (advanced stage and lymph node metastasis) suggested the role of CARLo-5 as a negative predictor for overall survival. Suppression of CARLo-5 mitigated NCI-H1975 cell proliferation by increasing p16, p21 and p27 proteins expression and subsequently blocking G0/G1 phase transition. Furthermore, the CARLo-5 replaces the epithelial behavior with the mesenchymal action and regulates transcriptional factors mediating EMT in NSCLC cells. Therefore, CARLo-5 as a promoter in EMT process leads to cell invasion phenotype and tumor metastasis [43].

In a word, the lncRNA CARLo-5 exercises the positive effect on tumorigenesis and tumor progression by inducing cell cycle arrest and EMT.

### *MVIH: microvascular invasion in HCC*

The lncRNA MVIH is known to be transcribed from the intron of the RPS24 gene encoding ribosomal proteins. Mounting MVIH expression was originally found in HCC and promoted microvascular invasiveness, tumor growth and metastasis [44]. As expected, MVIH was demonstrated to be upregulated in NSCLC tissues compared with matched noncancerous tissues. And the ectopic expression implied later stage, large tumor size and distant metastasis, which endowed MVIH with the role as a biomarker for predicting prognosis. The exploration results identified that reduced MVIH expression became an impediment to cell proliferation and invasion, while pcDNA-MVIH transfected cells exhibited the opposite phenotype. Further studies about underlying mechanism of metastasis manifested that MVIH regulated MMPs expression to alter extracellular matrix (ECM) degradation activity [45]. Therefore, MVIH may modulate multiple oncogenic properties and lung cancer progression.

### *ZXF1*

ZXF1, also known as uc001kfo, is located in chromosome 10 and proved by qRT-PCR to be highly expressed in lung adenocarcinoma samples, the level of which was distinctively associated with clinicopathological characteristics, such as lymph node metastasis and pathological stage. Thus ZXF1 can serve as a prognostic marker for patients with lung cancer. The down-

regulation of ZXF1 was described to impair cell migration and invasion but not affect cell vitality. Moreover, ACTA2 (Homo sapiens actin, alpha 2) is the antisense transcript of ZXF1 and encodes a-smooth muscle actin 2 ( $\alpha$ -SMA), which is monitored by transforming growth factor  $\beta$ 1 (TGF- $\beta$ 1). So lncRNA ZXF1 probably interacts with ACTA2,  $\alpha$ -SMA and TGF- $\beta$  signaling pathway to impact lung adenocarcinoma development [46].

### *HNF1A-AS1: HNF1A-antisense RNA 1*

HNF1A-AS1 (C12orf27) is 2455-bp long and located on chromosome 12, which is transcribed from HNF1A gene in the antisense direction. The higher expression level in NSCLC tissue and cell lines was detected and significantly correlated with poor survival. The knockdown of HNF1A-AS1 inhibited cell viability and clonogenic survival partially via promoting G0/G1 arrest and reducing Cyclin D1 in A549 cells but not in SPC-A1 cells. However, the apoptosis of cells with HNF1A-AS1 knockdown had no significant difference compared with that of control group. After injection of sh-HNF1A-AS1 into male nude mice, tumor size and weight was lower than that of control mice. Additionally, the downregulation of HNF1A-AS1 depressed lung adenocarcinoma cell motility *in vitro* and tumor metastasis *in vivo* through regulating EMT-associated markers' expression. The direct interaction between HNF1A-AS1 and DNA methyltransferase 1 (DNMT1) was confirmed by RNA immunoprecipitation (RIP) assays, while chromatin Immunoprecipitation (ChIP) assays identified the binding of DNMT1 to E-cadherin, suggesting the mechanism that HNF1A-AS1 can recruit DNMT1 to E-cadherin and mediate EMT by reducing E-cadherin expression [47].

### *BCYRN1: brain cytoplasmic RNA 1*

BCYRN1 (BC200) is a 200 nt transcript and possibly plays a pivotal role in repressing translation of metastasis-associated suppressors. BCYRN1 expression is significantly upregulated in NSCLC compared with that of normal control group, accompanied by high c-MYC expression level. Recent data show that c-MYC binds to the promoter of BCYRN1 and activates BCYRN1 transcription, which means that BCYRN1 is positively correlated to c-MYC, an oncoprotein. When the combination between c-MYC and

BCYRN1 is upregulated by transfection with pWPXL-c-MYC in A549 cells, the ability of cell migration and invasion is enhanced, suggesting BCYRN1 regulates cell motility through c-MYC-induced activation. Further detailed experiment demonstrated that upregulation of BCYRN1 level regulated cancer cell metastasis via the promotion of the expression of MMP9 and MMP13, ECM-degrading enzymes digesting the basement membrane and collagens [48].

### H19

It has been investigated that lncRNA H19 gene, belonging to imprinted cluster 11p15.5 [49] and encoding a 2.3 kb transcript [9, 49], is expressed on the maternal allele and in cis maintains epigenetic silencing of neighboring IGF2 (insulin-like growth factor) gene [10]. The expression of lncRNA H19 is relatively high throughout embryo and fetus development, but is reduced in most tissues shortly after birth. Many studies reported that loss of imprinting (LOI), at least in part due to hypomethylation of the promoter region, underlay strong H19 expression [9, 49, 50] [Matouk, 2015 #44; Gibb, 2011 #5; M, 1995 #50]. The anomalous expression of H19 was found in a variety of human cancers, including lung cancer. In hypoxic microenvironment of lung cancer with wild type p53, p53 expression is suppressed and HIF1- $\alpha$  is overexpressed, which simultaneously upregulates H19 lncRNA [49]. Furthermore, c-Myc induces lncRNA H19 expression via promotion of histone acetylation and transcriptional initiation in the H19 promoter [9, 10, 49, 51, 52]. Mdig (mineral dust-induced gene) reduces the level of the trimethylation of H3K9 in the promoter and elevates the expression of H19 [49]. The elevated expression of lncRNA H19 was also observed in the airway epithelium of smokers, which explained the inevitable correlation of lung cancer development with lncRNA H19 [53].

lncRNA H19 was confirmed to have abnormally higher expression in NSCLC tissues than corresponding non-tumorous tissues. The significant linkage between H19 level and tumor size and TNM stage was previously investigated. Besides, patients with high H19 level present shorter overall survival and poorer prognosis than those with the low expression level [51].

Previous studies have found that H19 silencing depresses cell proliferation and stalls cell cycle progression at the G0/G1 phase, which may result from H19-mediated dysregulated expression of miR-107 via their common base pairing [52]. Tumor formation assay *in vivo* showed tumor growth was slower and tumor weight was lighter in shRNA-H19 group compared to that in the control group [51]. The knockdown of H19 significantly inhibited anchorage-independent growth and attenuated the tumorigenic and scattering effect of HGF/SF. More importantly, lncRNA H19 contributes to the EMT process via Slug upregulation and E-cadherin downregulation, which thus increases the ability of lung cancer cells to metastasize. In addition, lncRNA H19 also regulates target gene expression by functioning as precursor for miR-675 [49]. These accumulating evidences suggest that lncRNA H19 plays an oncogenic role in lung cancer. Recently the high level and LOI of H19 expression is considered to be an early marker for the malignant transformation of airway epithelium and represent poor survival [49, 53].

### LUADT1: LUAD transcript 1

The LUADT1 gene, located on chromosome 6q24.3, processes 453 nt long non-coding RNA, whose expression level was higher in lung adenocarcinoma than lung small cell cancer and adjacent normal tissues as well as associated positively with T stage. It was observed that si-LUADT1 treatment dramatically suppressed lung adenocarcinoma cell proliferation and colony formation. Moreover, cell cycle was arrested by LUADT1 knockdown through decreasing cyclin D1, cyclin-dependent kinase 4 (CDK4) and cyclin-dependent kinase 6 (CDK6) expression, leading to increased percentage of cells in G0/G1 phase and thus opposite status in S phase. Further experiments *in vivo* validated these results.

The molecular mechanisms by which LUADT1 plays the potential role in lung adenocarcinoma were preliminarily discussed. Like HOTAIR, LUADT1 recruited PRC2, particularly SUZ12 (suppressor of zeste 12 protein homolog), to p27 promoter region and induced H3K27 trimethylation, followed by inhibited p27 expression. Namely, the level of LUADT1 was correlated negatively with that of p27. These data demonstrated LUADT1 epigenetically sup-

pressed p27 expression and subsequently foster lung adenocarcinoma proliferation ability [54].

### *AFAP-AS1: actin filament associated protein 1 antisense RNA1*

Recently some studies found a significantly upregulated novel long non-coding RNA in lung cancer, AFAP-AS1, interfering with AFAP1 protein level [55]. It was verified that the expression of AFAP-AS1 was influenced by some clinical parameters, including clinical stage, smoking history and TNM stage [56], showing that AFAP-AS1 may serve as a prognostic factor for overall survival and recurrence-free survival of patients [55, 56]. Functionally, AFAP-AS1 enhances the migratory and invasive potential by regulating actin filament integrity and Rho/Rac GTPase family members-associated molecules, but has no effect on cell viability, cycle or apoptosis [55].

These data shows AFAP-AS1 is actually an oncogene for development of lung cancer and thereby can become a therapeutic target for lung cancer intervention.

### *PVT1*

LncRNA PVT1 was found to be located at 8q24.21 and upregulated in numerous human cancers, including NSCLC. The expression of PVT1 is positively related to MYC protein expression. When exploring the correlation between LncRNA PVT1 level and clinicopathological features, the researchers observed that higher PVT1 expression resulted in large tumor size, advanced histological grade, lymph node metastasis and poor overall survival. Loss-of-function assays utilizing PVT1-specific siRNA investigated the biological role for LncRNA PVT1 in NSCLC and ascertained that si-PVT1 abrogated cell proliferation, migration and invasion and induced apoptosis and cell cycle arrest [57, 58], which determined PVT1 to support the progression of NSCLC.

### *RGMB-AS1: repulsive guidance molecule b-antisense 1*

LncRNA RGMB-AS1 is located in chromosome 5 and has an inverse expression level with RGMB, the knockdown of which promotes the development and metastasis of several cancers such as prostate and breast cancer partly

through controlling BMP signaling pathway. The expression level of LncRNA RGMB-AS1 was demonstrated to be higher in NSCLC than normal tissues and correlated to differentiation status, lymph node metastasis and TNM stage [59]. Silencing RGMB-AS1 leads to the inhibition of cell proliferation via cell cycle arrest at G0/G1 phase in A549 and SPC-A1 cells. The reduction of RGMB-AS1 also weakens migratory and invasive rate. RGMB-AS1 expression is negatively related to RGMB level through regulating exon 2 sequence of RGMB [60]. These results suggest that RGMB-AS1 and RGMB affect the development of lung cancer through some signaling pathways, which needs further exploration to provide a new insight into lung cancer therapy.

### *GHSROS: growth hormone secretagogue receptor opposite strand*

LncRNA GHSROS, 1.1 kb in the full length, is an opposite strand containing a single exon of the intron of GHSR gene. LncRNA GHSROS doesn't possess the capability of translating any proteins despite of its encoding some very small peptides. Examination of GHSROS expression using qRT-PCR presented a higher GHSROS level in lung cancer. Overexpressing GHSROS could significantly cause the promotion of the migration ability of lung cancer cell but not alter cell proliferation, while migration was inhibited in the Beas-2B cells with forced expression of GHSROS [61]. These data showed that GHSROS could function as an oncogenic lncRNA in lung cancer.

### *Lnc\_bc060912*

A nuclear-retained long non-coding RNA as shown by isolation of the nuclear and cytosolic fractions displays increased expression in lung cancer. Si-Lnc\_bc060912 led to a marked reduction in A549 cell viability. Moreover, the knockdown of Lnc\_bc060912 had a promoting effect on cell apoptosis and increased caspase 3 cleavage while overexpressing Lnc\_bc060912 exerted an opposite effect, suggesting that Lnc\_bc060912 might expedite lung cancer cell proliferation via the inhibition of caspase 3-dependent apoptosis. It was confirmed that doxorubicin (dox)-induced p53 decreased the RNA level of Lnc\_bc060912 in A549 cell, and there was similar tendency in H1299 cell with homozygously deleted p53 when p53 was

enhanced. In brief, the tumor suppressor p53 reduces the level of Lnc\_bc060912 expression. Furthermore, CHIRP and RIP assays showed that Lnc\_bc060912 had a physical interaction with proteins PARP1 (poly (ADP-ribose) polymerase 1) and NPM1 (nucleophosmin), which synergistically prevent cell apoptosis in consistent with the role of Lnc-bc060912. Therefore, Lnc\_bc060912 impairs cell apoptosis in lung cancer by interacting with PARP1 and NPM1 to contribute to tumorigenesis and cancer progression [62].

### *NEAT1: nuclear enriched abundant transcript 1*

LncRNA NEAT1, as a component of nuclear paraspeckle, was identified to be implicated in mRNA editing and thus regulate gene expression. Pan *et al.* found its elevated expression in NSCLC tissues when compared to that of corresponding normal lung tissues. It has been investigated that the expression level of NEAT1 depends on patient age, lymphatic metastasis status, vascular invasion degree and TNM stage, whereas the association of NEAT1 expression level and survival time has little significance. Therefore, NEAT1 may play an oncogenic role and has a potential value for diagnosis in lung cancer in spite of no detectable effect on patient outcome [63].

Finally, NEAT1 had a negative interaction with the expression level of miR-449a. The knockdown of miR-449a led to the upregulation of NEAT1 while the lower expression level of NEAT1 was displayed in miR-449a-overexpressed lung cancer cell lines, demonstrating miR-449a negatively regulates NEAT1 in lung cancer [64].

### *DLX6-AS1: distal-less homeobox 6-antisense 1*

It is of note that by using microarray assay and qRT-PCR analysis, expression pattern of lncRNAs identified the upregulation of DLX6-AS1 in lung adenocarcinoma. The knockdown of DLX6-AS1 with siRNA has been confirmed to abolish the expression level of DLX6 mRNA and protein, indicating that DLX6-AS1 is positively associated with DLX6. Clinical data analysis has corroborated the explicit relationship of lncRNA DLX6-AS1 and DLX6 mRNA with both histological differentiation and TNM stage. Moreover, since the detail molecular mechanism underlying the involvement of DLX6-AS1

in the process of cell biology has been waiting for clear and definite explanation, KEGG pathway analysis has pointed that lncRNA DLX6-AS1 may regulate the proliferation ability of lung cancer cells [65]. To acquire better understanding of lncRNA DLX6-AS1, further studies are needed to uncover its function in lung cancer tumorigenesis.

### *LINC01133*

Recent identification of differentially expressed lncRNAs between lung adenocarcinoma (LAD) and lung squamous carcinoma (LSCC) demonstrated that a novel lncRNA LINC01133 was substantially upregulated in LSCC. Moreover, qRT-PCR assay was performed to validate the overexpressed expression of LINC01133 in LSCC, without significantly different expression level between LAD tissues and corresponding normal tissues. It has been observed that the aberrant expression of LINC01133 is correlated with poor survival time of patients with LSCC. The loss-of-function assay of LINC01133 by siRNAs in LSCC cell line, H1703, confirmed that its silence inhibited cell invasion [66].

However, another study identified the characterizations and functions of LINC01133 in both NSCLC type. Enhanced expression level of LINC01133 is linked to tumor size, advanced pathological stage and lymph node metastasis. The inhibition of proliferation in si-LINC01133-transfected NSCLC cells was induced by cell cycle arrest and high apoptotic rate. The knockdown of LINC01133 impedes the capability of NSCLC cell migration and invasion. LINC01133 binds to EZH2 and LSD1. EZH2 mediates H3K27me3 in KLF2, p21 and E-cadherin promoters, and LSD1 induces H3K4me2 demethylation in KLF2 and p21 promoters, which cause their epigenetic silencing. Moreover, silence of KLF2 was demonstrated to be take part in the promoting function of LINC01133 in cell growth. Hence, LINC01133 may regulate cell proliferation and apoptosis partly through restraining KLF2 and p21, while promote cell migration and invasion via reducing E-cadherin expression in NSCLC cells [67].

Accumulated data indicate that LINC01133 is regarded as an oncogene involving in the occurrence and metastasis of NSCLC and serve as a potential biomarker for the diagnosis, therapy and prognosis of NSCLC.

### *IRAIN*

Recent research on lncRNA in lung cancer, particular in NSCLC, identified a novel and functional transcript IRAIN, which is transcribed in antisense orientation from an intron within the IGF-1R locus and has been defined as a tumor suppressor in AML. IRAIN was found to be highly upregulated in NSCLC tissues, and the higher expression level of IRAIN in NSCLC patients exhibited a dramatic association with tumor size and smoking history. The downregulation of IRAIN via si-RNA showed the inhibitory effect on A549 cells through blocking cells in G1 phase. IRAIN modulates an important regulatory pathway influencing cell growth, IGF-1R signaling, to increase the incidence of NSCLC [68]. These data establish that IRAIN exerts the oncogenic action and further sets the stage for the efficient therapy for NSCLC.

### *UCA1: urothelial carcinoma-associated 1*

The lncRNA UCA1, which was first known in human bladder carcinoma, has oncogenic property in non-small cell lung cancer. It was elevated in NSCLC tissues and cell lines (A549, H1299, H446, H460, and NCI-H1650). The association between the upregulated expression of UCA1 and tumor size as well as TNM stage was so significant that the expression level of UCA1 could represent severity of lung cancer. It was probed that UCA1 was an independent prognostic indicator for overall survival in NSCLC [69].

UCA1 induces cell proliferation when overexpressed in NSCLC cell lines. However, overexpressed miR-193a-3p reversed the promoting effect of UCA1, which brings a speculation that UCA1 may function as miR-193a-3p sponge to promote cell growth. Further mechanistic investigation showed that UCA1 increased the protein level of ERBB4, which is the molecular target of endogenous miR-193a-3p [69]. All the results indicate that UCA1 is involved in enhancing NSCLC progression by establishing and regulating UCA1-miR-193a-3p-ERBB4 signaling pathway.

### *AK001796*

AK001796 has been found to be the most distinctly downregulated lncRNA in A549 cells treated with resveratrol, a herbal drug from various plants. This lncRNA was defined as an

oncogene because of its upregulation and malignant ability in lung cancer tissues and cells. Its overexpression could contribute to lung cancer development. Inhibition of AK001796 could reduce lung cancer cell (A549, H446, 16HBE-T) proliferation *in vitro* and tumor growth *in vivo* and inhibit cell cycle progression, but have no significant effect on cell apoptosis. As the normal expression level of each cell cycle-associated gene ensures cell cycle to progress without a hitch, AK001796 was demonstrated to be involved in the dysregulation in a broad range of cell cycle-related genes and thus lead to cell cycle arrest. AK001796 was downregulated by the treatment of resveratrol in AK001796-shRNA-NC-transfected cells but not in AK001796-shRNA-transfected cells. Moreover, the inhibitory rate of cell proliferation by resveratrol in AK001796 knockdown group was lower than that in control group, suggesting that AK001796 serves as a mediator of the role for resveratrol in blocking lung cancer cell growth [70]. This report reveals that the anomalous expression of lncRNAs may affect the efficacy of chemoprevention and chemotherapy of lung cancer.

### *LINC01207*

LINC01207, a recently observed histological type-specific lncRNA located in an intergenic region on chromatin 4, is specifically upregulated in lung adenocarcinoma, and the aberrant expression of LINC01207 is correlated with tumor stage and poor survival of LAD. Si-LINC01207 not only inhibited the proliferation and colony formation but also increased the apoptotic percentage and activated caspase 3 in LAD cell (A549). The knockdown of LINC01207 also inhibited tumor growth *in vivo*. Furthermore, LINC01207 could guide EZH2 to bind to Bad, a pro-apoptotic gene in Bcl2 family, and induce the H3K27me3 at the promoter. Therefore, LINC01207 may hinder cell apoptosis partly through eliciting EZH2-mediated silencing of Bad [71]. These findings indicated that LINC01207 may play an oncogenic role in the modulation of LAD progression.

### *PCAT-1: prostate cancer-associated transcript 1*

PCAT-1 is an lncRNA that was originally identified at the chromosome 8q24 gene desert in prostate cancer [72]. A recent study suggested

that PCAT-1 was upregulated in NSCLC tissues and cell lines as compared to adjacent normal tissues and 16HBE, respectively. The growth of cell was significantly decreased in sh-PCAT-1 transfected A549 cells, while proliferation of A549 cells was enhanced in the pcDNA3.1-PCAT-1 group compared with respective controls. In addition, forced expression of PCAT-1 increased NSCLC cell migration and invasion [73]. Thus, the oncogenic function exerted by lncRNA PCAT-1 shows the evidence that it promotes NSCLC tumorigenesis and progression.

### *TATDN1: homo sapiens TatD DNase domain containing 1*

LncRNA TATDN1 was detected to be highly expressed in 95D cells compared to 95C cells. The inhibition of TATDN1 hindered the capability of proliferation in 95D cells by decreasing oncogene human epidermal growth factor receptor 2 (HER2) expression. The upregulated metastatic suppressor Nm23-H1 and downregulated  $\beta$ -catenin and Ezrin proteins were demonstrated to be involved in impediment of migratory and invasive phenotype of si-TATDN1-transfected 95D cells. *In vivo* tumor xenograft assay, the growth and metastasis of sh-TATDN1-derived tumors were inhibited, which were consistent with these data *in vitro* [74]. Undoubtedly, the merit of lncRNA TATDN1 should not be overlooked in promoting tumor metastasis of NSCLC.

### *Linc00673*

Linc00673 was commonly upregulated in NSCLC tissues and cell lines except H1703 and H226 with lower linc00673 expression. The correlation of linc00673 with patients' tumor size, lymph node metastasis and TNM stage was evaluated to be significant. Linc00673 was demonstrated to be potential diagnostic biomarker with AUC of 0.683. Loss and gain-of-function assays in NSCLC cell lines displayed that linc00673 might serve as an oncogene to have a positive role in accelerating cell proliferation. SiRNA-mediated depletion of linc00673 led to G0/G1 arrest by reducing CDK6 expression, which appears to be responsible for the alteration of cell viability. Xenograft mouse model confirmed that linc00673 could govern tumor growth. Gene expression profile analysis showed the expression of NCALC was reversely related to linc00673. Further researches about mechanisms determined that linc006-

73 recruited LSD1 to mediated H3K4me2 demethylation in neurocalcin delta (NCALD) promoter region, which inhibited the transcription of NCALD. Additionally, overexpressing NCALD could present the inhibitory effect on cell proliferation. The expression of NCALD was decreased in NSCLC tissues and predicted NSCLC patients overall survival [75]. These data elucidate that LSD1-induced repression of NCALD is involved in the upregulation of linc00673-mediated tumor progression in NSCLC.

### *LINC00473*

LKB1, a tumor suppressor gene, usually sustains mutational inactivation in lung cancer. Compared to LKB1-expressing NSCLC cells, the expression of LINC00473 was detected to be elevated in LKB1-null cell lines, which indicated that LINC00473 was a LKB1-related lncRNA and could predict LKB1 functional status in NSCLC. LINC00473 was demonstrated to elevate cell proliferation and tumor growth by knockdown in LKB1-null cells and overexpression in LKB1-WT cells. Moreover, LINC00473 may be a predictor for the survival time of NSCLC patients. LKB1 loss could promote dephosphorylated transcription coactivator (CRTC) to enter nucleus and ensuing enrichment of cyclic AMP-responsive element-binding protein (CREB) in the CRE sites of LINC00473 promoter region, which induced the transcriptional enhancement of LINC00473. Thus, CRTC/CREB activation mediated by LKB1 loss could induce the high expression of LINC00473 [76]. NONO was previously identified as a component of cAMP-signaling pathway [77]. Through RNA pull-down and RIP assays, NONO was confirmed to be LINC00473-related protein, which binds to CRTC via LINC00473 recruitment to stimulate CREB-mediated downstream gene transcription start [76]. LINC00473 is not only regulated by LKB1/CRTC/CREB signaling but also influences the expression of their target genes by introducing NONO protein. These data show that LINC00473 functions as potential marker for predicting tumor LKB1 inactivation and poor prognosis and as therapeutic target for depressing tumor growth in NSCLC.

### **Tumor suppressive lncRNAs**

#### *HMLincRNA717*

LncRNA HMLincRNA717, located at 18p11.228 with 818 nt in the length was identified as an

downregulated non-coding RNA in NSCLC tissues and cell lines by qRT-PCR assays, which indicated the promotion of the decreased HMIincRNA717 expression for lung cancer cell growth and the development of NSCLC. The correlation analysis between HMIincRNA717 and clinical factors provided accurate evidence that the low expression level of HMIincRNA717 was significantly related to advanced histological grade and TNM stage, which implied that HMIincRNA717 might impede the metastasis and progression of lung cancer. Kaplan-Meier analysis assessed the prognostic potential of HMIincRNA717 with the results that low expression of HMIincRNA717 was in accordance with short survival life for NSCLC patients. Further Cox hazard regression analysis determined HMIincRNA717 as an independent factor for NSCLC patients. All the results revealed that HMIincRNA717 impaired the development and progression of lung cancer through functioning as an tumor suppressor [78].

### *SPRY4-IT1: SPRY4 intronic transcript 1*

SPRY4-IT1 is transcribed from an intronic region within SPRY4. It was previously found and highly-expressed in melanoma cells, increasing cell growth and invasion and decreasing apoptotic rate. However, another study reported significantly downregulated SPRY4-IT1 in NSCLC tissues, which was relevant to some clinicopathological factors, such as tumor size, advanced stage and lymphatic metastasis, and thus played a predictive role for poor survival rate of NSCLC patients. The investigation corroborated that overexpressed SPRY4-IT1 inhibited the abilities of NSCLC cell (SPC-A1, A549) proliferation and colony forming, meanwhile promoted cell apoptosis, which was determined by mouse models *in vivo* indicating smaller and lighter tumors in pCDNA-SPRY4-IT1-transfected groups. Wound-healing and transwell assay verified the suppressive effect of high SPRY4-IT1 level on NSCLC cell migration and invasion, which corresponded to metastatic assay *in vivo*. Immunostaining analysis showed that the protein level and activity of Ki67, a nuclear antigen associated with mitosis, was lower in tumors developed from pCDNA-SPRY4-IT1-transfected SPC-A1 cells. In contrast, the depletion of SPRY4-IT1 by targeted siRNA led to results as apposed to the above except for not influencing cell proliferation. Moreover, as repre-

sentative markers of EMT, the low E-cadherin expression and the high Vimentin expression in NSCLC may provide one optional pathway, by regulating which SPRY4-IT1 contributes to metastasis of cancer cells to distant position. Importantly, the transcriptional regulation of SPRY4-IT1 is mediated by EZH2, which binds to the promoter region of SPRY4-IT1 and trimethylates lysine 27 on histone 3, resulting in the epigenetic silencing of SPRY4-IT1. Hence, the expression and function of EZH2 and SPRY4-IT1 are exactly opposite in lung cancer development [79]. Briefly, SPRY4-IT1 is epigenetically silenced by EZH2 binding to the promoter region, and influences cell proliferation and metastasis partly via EMT.

### *GAS5: growth arrest-specific transcript 5*

GAS5, a lncRNA initially extracted from subtraction cDNA library and then considered to be an upregulated tumor suppressors during growth arrest [80-82], is derived from exon 12 sequence within gene *gas5* at 1q25 and is around 650 nt long. GAS5 expression is epigenetically downregulated through the CpG islands methylation in the promoter region, while the effect of histone methylation on GAS5 expression has no statistic significance. Besides, GAS5 has inhibitory interaction with miR-21. Using qRT-PCR, GAS5 expression was determined and showed largely lower level in NSCLC tissues and cell lines (A549, H1650, H1299, H1975 and SK-MES) compared with adjacent noncancerous samples and HBE respectively, but a little higher in SPC-A1 cells. The downregulated GAS5 expression is related to advanced stage, large tumor and poor tumor differentiation, which identifies the diagnostic and predictive role for GAS5 in early-stage NSCLC.

A recent study indicated that GAS5 overexpression impaired cell viability and colony formation in pCDNA-GAS5 transfected H1650 and A549 cells, together with decreased cells in S phase and increased cells in G1 phase. On the contrary, the capacity of cell proliferation and forming colony was slightly hindered in GAS5-deficient SPC-A1 cells by using siRNA. In addition, upregulated GAS5 level expedited cell apoptosis but didn't affected migration and invasion at all. Mouse model was conducted through subcutaneous injection with pCDNA-GAS5 or empty vector transfected A549 cells.

The tumors formed from pcDNA-GAS5 group showed smaller size and lower weight than those in control group. Moreover, GAS5 overexpression by qRT-PCR analysis and poor Ki67 positivity by immunohistochemical staining result were demonstrated in GAS5-transfected tumor tissues.

A p53-dependent or p53-independent pathway is involved in the functional regulation of GAS5 in lung cancer. Firstly, in GAS5-transfected A549 cells, the upregulated protein but not mRNA level of p53 and its target gene p21 are confirmed, which suggests that GAS5 post-transcriptionally modulates p53 and p21 expression. Secondly, in GAS5-transfected H1650 cells lacking wild-type p53, GAS5 reduces transcript factor E2F1 expression via the Pc2 polycomb protein and decreases cyclin D1 protein expression, which gives rise to the above mentioned cell cycle arrest [82].

Taken together, these data suggest that upregulated GAS5, as a tumor suppressor, inhibits lung cancer cell proliferation and tumor growth and mediates cell apoptosis.

### *GAS6-AS1: growth arrest-specific gene 6 antisense RNA 1*

GAS6-AS1 is located at chromosome 13q34 and is transcribed in the antisense direction from GAS6. GAS6-AS1 expression was detected to be commonly lower in NSCLC tissues. In addition to histological grade and classification, GAS6-AS1 is inversely correlated to lymph node metastasis and TNM stage. Like the majority of lncRNAs, GAS6-AS1 is also an independent predictor for patients' survival. Little functional role of GAS6-AS1 has been shown, but it is speculated that GAS6-AS1 maybe regulates GAS6 gene to exert its effects on NSCLC progression [83].

### *BANCR: BRAF activated non-coding RNA*

BANCR transcript is 693 bp at the length and localized at chromosome 9, which was previously found to facilitate cell motility in melanoma. The qRT-PCR analysis was conducted to detect the relatively low BANCR expression level in NSCLC tissues and cell lines, which was negatively correlated to tumor size, lymph node metastasis and TNM stage. Univariate and multivariate analysis suggested that BANCR was

an independent predictive factor with statistic significance for overall survival in NSCLC. Researches was performed and found that histone deacetylation 3 (HDAC3)-induced silencing could be the major cause of the downregulation of BANCR. The proliferation of pcDNA-BANCR transfected SPC-A1 and A549 cells was inhibited, which was ascertained by colony forming assays. In addition, the overexpressing BANCR suppressed cell growth by increasing the apoptosis of pcDNA-BANCR transfected cells. On the other hand, the forced expression of BANCR hindered cell migration and invasion by affecting EMT, such as increasing E-cadherin expression and decreasing N-cadherin, Vimentin and MMP2 expression, and blocked tumor metastasis *in vivo* [84].

### *PANDAR: promoter of CDKN1A antisense DNA damage-activated RNA*

PANDAR shows p53-dependent induction from the CDKN1A promoter when DNA damage occurs. The existence of wild type p53 response element in the PANDAR promoter region indicates that the transcriptional regulation and functional role of PANDAR are mediated by WT p53. However, p53 is usually absent or mutated in a variety of cancers. Recent studies reported that PANDAR was downregulated in NSCLC, and the dysregulated expression of PANDAR was linked to TNM stage, tumor size and poor prognosis of NSCLC. PANDAR overexpression inhibited cell proliferation and induced cell apoptosis, meanwhile its depletion could rescue p53-increased growth inhibition and apoptosis enhancement. PANDAR has an interaction with the transcription factor NF-YA (nuclear transcription factor Y, alpha), which binds to and regulates the expression of Bcl-2 to take part in tumorigenesis. In NSCLC, downregulation of PANDAR increases the binding of NF-YA to Bcl-2, which results in promotion of cell proliferation and inhibition of caspase-3 activation-mediated cell apoptosis. Therefore, PANDAR/NF-YA/Bcl-2 pathway plays an important role in regulating lung cancer cell growth [85].

### *TUG1: taurine-upregulated gene 1*

TUG, a 7.1 kb lncRNA, was firstly found to be upregulated by taurine in developing mouse retinal cells and was essential to retinal development. TUG1 expression was determined

using qRT-PCR to be downregulated in NSCLC in comparison with corresponding adjacent noncancerous tissues and normal 16HBE. The low expression level of TUG1 is remarkably associated with advanced clinical stage, larger tumor size and poor survival for NSCLC patients, indicating that the downregulated TUG1 contributes to NSCLC occurrence and development. Studies have shown that TUG1 presents WT p53 binding sites in the promoter region, which putatively suggests the transcriptional regulation of TUG1 expression is in p53-mediated manner. Further experiments that dox-induced p53 expression promotes the upregulation of TUG1 confirm the speculation. Si-TUG1 causes the knockdown of TUG1 and further promote cell cycle transition and subsequent cell growth. When shTUG1-transfected SPC-A1 cells are injected into nude mice, the developed tumors are larger in size and heavier in weight at the experiment end than those from control group. TUG1, enriched in the nucleus, directly binds to PRC2 and epigenetically regulates the expression of HOXB7 at the transcriptional level via H3K27 methylation at the promoter region. Si-HOXB7 transfected cells displays not only a reduction of growth and G0/G1 phase arrest but also apoptosis enhancement through affecting AKT and MAPK pathways with decreased p-ERK, p-AKT and p-GSK3 $\beta$ , which is in accordance with the effect of TUG1 on cell growth. These findings shows TUG1 can be involved in the modulation of HOXB7 by binding to PRC2 and then influence AKT and MAPK pathways, which are mechanisms of TUG1 controlling cell growth [86].

### *MEG3: maternally expressed gene 3*

MEG3 is a paternally imprinted gene located at chromosome 14q32.3 [87]. One of its transcriptional products is MEG3 transcript, a lncRNA broadly detectable in various normal human tissues but downregulated in a number of human cancers [9], including NSCLC [88].

Lu *et al.* have already confirmed that the transcriptional inactivation of lncRNA MEG3 may be attributed to hypermethylation of CpG island in MEG3-DMR loci. Cell proliferation and colony formation was significantly impaired in pCDNA-MEG3 transfected SPC-A1 cells or A549 cell, while proliferation of H1299 cell was increased in the si-MEG3 group compared with respective controls. In addition, forced expression of

MEG3 enhanced cell cycle arrest at the G1/G0 phase and decreased the percentage of cells in G2/S phase. Moreover, MEG3 overexpression resulted in an increase in the apoptotic rate of SPC-A1 and A549 cells, but there was no alteration in the ability of cell invasion. *In vivo* tumor formation assay in a nude mouse model showed pcDNA-MEG3 stably-transfected tumors were substantially smaller and lower compared to empty vector group [88]. All data indicated that MEG3 could reduce tumor growth.

The results of further exploration of the underlying mechanisms identified the upregulated expression of p53 and the inhibition of MDM2 (mouse double minute 2 homolog) expression in pCDNA-MEG3 transfected SPC-A1 cells [88]. These results exhibit that MEG3 mediates p53 activation and thus suppresses the development of NSCLC. Another report demonstrated that lncRNA MEG3 inhibited cell growth by suppressing the expression of Skp2 via cooperation with miR-3163. Skp2 is a component of the E3 ubiquitin ligase that contributing to ubiquitination-related degradation of p27 [87].

### *CASC2: cancer susceptibility candidate 2*

CASC2 was firstly screened as a tumor suppressor gene located at chromosome 10q26 in endometrial cancer [89] and suppressed malignant phenotype in human gliomas by miR-21 [90]. It was recently indicated that CASC2 was significantly downregulated in NSCLC tissues and cell lines. The reduced expression of CASC2 remarkably declared the advanced clinical stage and greater tumor size. Overexpressing CASC2 could preclude lung cancer cell proliferation by stalling cell cycle at the G0/G1 phase *in vitro* and decrease tumor growth and the proliferation index Ki-67 *in vivo*. CASC2 was capable of predicting prognosis and overall survival and regarded as a target for treating lung cancer [91].

### *ENST00000434223*

ENST00000434223 was peculiarly downregulated in NSCLC tissues and cell lines. Low expression level of ENST00000434223 in NSCLC patients was strikingly associated with tumor size, lymph node metastasis and advanced TNM stage. ENST00000434223 overexpression could reduce NSCLC cell proliferation, and pcDNA3.1-ENST00000434223 may cause

obvious G0/G1 phase arrest and induce apoptosis by flow cytometric analysis. Tumor formation assay *in vivo* verified that upregulated ENST00000434223 accelerated tumorigenesis. Furthermore, enhanced expression of ENST00000434223 could decelerate NSCLC cell invasive nature and reverse EMT [92]. Cumulatively, lncRNA ENST0000043223 is a crucial tumor suppressor in NSCLC.

### Malignant transformation-associated lncRNAs

#### AF118081

AF118081 was found to have an oncogenic effect on lung cancer carcinogenesis induced by exogenous carcinogen, such as tobacco smoke containing a large number of chemicals, which may be reported for the first time. Although AF118081 was identified as significantly overexpressed lncRNA by qRT-PCR in malignantly anti-benzo [a] pyrene-trans-7,8-dihydrodiol-9,10-epoxide (anti-BPDE)-transformed 16HBE-T cells, lung cancer cell lines and patient samples, the expression level of AF118081 had no correlation with clinicopathological factors. Downregulation of AF118081 expression by siRNA could suppress cell proliferation and colony formation but accelerate cell apoptosis. Si-AS118081 also inhibited the ability of malignant cells to migrate and invade. Meanwhile, nude mouse xenograft models verified that AF118081 promoted tumor growth, corresponding to assays *in vitro*.

In summary, AF118081 contributes to malignant transformation and tumorigenesis. It can help to assess the prognosis of lung cancer patients, also be the valuable biomarker for lung cancer diagnosis and ideal target for specific therapy [93].

#### SCAL1: smoke and cancer-associated lncRNA-1

SCAL1 is upregulated in cells (CL-5) predisposed to metastasis and bronchial epithelial cells from smokers, suggesting that this lncRNA is associated with smoke exposure and aggressive phenotype in lung cancer. Consistent with the opinion, HBE1 and NHBE cells present a high expression of SCAL1 after being exposed to CSE. NRF2 (nuclear factor erythroid 2-related factor), a transcription factor binding to NF-E2 (nuclear factor erythroid-derived 2) motif in the promoter of SCAL1, is increased in many

lung cancer cell lines in parallel with SCAL1. After oxidative stress-induced activation, upregulated NRF2 modulates the expression of oxidation suppressors and frees cells from the toxic effect induced by external stressor. Collectively, SCAL1 may be a vital downstream regulator of NRF2 in defending the cytotoxicity generated by cigarette smoke and weaken malignant behaviors in lung cancer cells [94].

#### Lnc-DQ786227

It has found that the expression of lnc-DQ786227, similar to AF118081, was higher in lung cancer cells and human bronchial epithelial cells malignantly induced by anti-BDPE than normal cells. Knocking-down lnc-DQ786227 by specific siRNA decreased BEAS-2B-T cells viability and colony formation rate, which was clarified to be dependent on increased apoptosis rate as cell cycle was not affected. Moreover, *in vivo*, the tumors developed from shRNA transfected cells were smaller and lighter compared with control group. These data showed that lnc-DQ786227 might be associated with malignant transformation and identified as an oncogene [95].

### Drugs resistance-associated lncRNAs

#### AK126698

There was differential expression level of many lncRNAs by comparing A549 with cisplatin-resistant A549 cell lines. Of these, the lncRNA AK126698 was investigated and found its significantly low level using qRT-PCR. Pathway analysis and co-expression network highlighted Wnt signaling pathway that could be involved in cisplatin resistance. Further studies indicated that the inhibition of AK126698 by specific siRNA reduced NKD2 (naked cuticle) expression level, which resulted in high level of  $\beta$ -catenin, transcript factor in Wnt signaling pathway. Intriguingly, it was found that si-AK126698 regulated Wnt signaling pathway by blocking  $\beta$ -catenin degradation. Moreover, it was evident that AK126698 knockdown could suppress cell apoptosis after cisplatin treatment. Taken together, cisplatin resistance was induced by low lncRNA AK126698 expression regulating Wnt signaling pathway [96].

#### HOTAIR

Some studies showed that the lncRNA HOTAIR manipulated the resistance to chemotherapy in

lung cancer. The expression level of HOTAIR is higher in anticancer drugs-resistant lung cancer cells than that in parental cells [6, 97]. And the upregulation of HOTAIR is in a cisplatin dose-dependent manner [6]. Transfection with specific si-HOTAIR into resistant cells led to reduction of IC50 values of chemotherapeutic drugs including CDDP, ADM and VP-16 through enhancing drugs-induced apoptosis and G0/G1 arrest. Conversely, overexpressing HOTAIR generated the resistance of parental A549 cells to cisplatin and inhibited cell apoptosis and cell cycle arrest. The above mentioned results were verified by assays *in vivo* [6, 97].

In NSCLC, HOTAIR mediates the recruitment of PRC2 to tumor suppressor p21, which contributes to the silence of p21. That is, enhanced expression of HOTAIR is correlated to the downregulation of p21. Based on this, transfection with pcDNA-p21 or si-p21 into A549/DDP or A549 cells was performed and showed that the effects of upregulation or downregulation of p21 on the sensitivity of A549/DDP or A549 cells to cisplatin was respectively similar to si-HOTAIR or pcDNA-HOTAIR [6]. These studies point to the truth that p21 could be a crucial downstream regulator involved in HOTAIR-mediated cisplatin resistance of lung adenocarcinoma cells. In SCLC, HOXA1, one of the HOX family members, was identified to participate in chemoresistance. HOTAIR is associated with HOXA1 methylation and epigenetically causes its silence through altering the levels of DNA methyltransferase DNMT1 and DNMT3b [97]. So HOTAIR regulates the methylation of HOXA1 to affect multidrug resistance in SCLC. These statements in part accounts for acquired chemotherapy resistance for clinical lung cancer patients.

### GAS5

In addition to enhance tumorigenesis and progression, GAS5 downregulation was confirmed to be in connection with primary epidermal growth factor receptor-tyrosine kinase inhibitors (EGFR-TKIs) resistance in lung adenocarcinoma with wild-type EGFR expressing. Since the expression level of GAS5 in A549 cell line with gefitinib-resistant property is significantly lower than in sensitive cell line, resistant A549 cells were transfected with pcDNA-GAS5, which led to decrease in cell viability and increase in

cell apoptosis, as well as cell cycle arrest after gefitinib treatment. Furthermore, A549 treated with GAS5 plus gefitinib displayed reduction of EGFR/PI3K/Akt pathway proteins and IGF-1R, suggesting IGF-1R could be the downstream factor of GAS5 to abrogate EGFR-TKIs efficacy. All the studies indicated that GAS5 overexpression sensitized lung adenocarcinoma cells to EGFR-TKIs through inhibiting IGF-1R level, which provides a new therapeutic approach for EGFR-TKIs-resistant patients with lung adenocarcinoma [4].

### UCA1

The development of acquired resistance to EGFR-TKIs may occurs about 12 months after the treatments for most patients with EGFR-mutated NSCLC. Although the related mechanisms still remain to be illuminated, it was found that lncRNA UCA1 was upregulated in acquired EGFR-TKIs resistant NSCLC patients and cells with or without T790M mutation, whereas its downregulation was observed in patients with primary resistance. As higher UCA1 expression caused a lower objective response rate and poorer survival than low expression before treatment of EGFR-TKIs, indicating lncRNA UCA1 could be referred to as a prognostic indicator. However, after acquired resistance arose, lncRNA UCA1 was associated with prognosis only when NSCLC patients had no T790M mutation [98]. These data declared that lncRNA UCA1 may be involved in acquired resistance to EGFR-TKIs. Co-treatment with si-UCA1 and gefitinib could lead to a reduction in the proliferation of EGFR-TKIs resistant cells *in vitro* and tumor growth *in vivo*, as well as an increase in gefitinib-induced cell apoptosis [98], which indicated that UCA1 had a pivotal effect on viability of EGFR-TKI-resistant cells.

Further research put forward the molecule mechanisms by which UCA1 influences the efficiency of EGFR-TKIs. Firstly, si-UCA1 suppresses the expression of p-AKT, p-mTOR and p-ERK, the downstream mediators of EGFR, to regulate AKT/mTOR and ERK signaling pathways. Secondly, EMT process is associated with EGFR-TKIs resistance and gets an obvious inhibition when resistant NSCLC cells are treated with si-UCA1 [98]. Taken together, in EGFR-mutant NSCLC, UCA1 may contribute to non-T790M acquired resistance to EGFR-TKIs at least partly by stim-

ulating AKT/mTOR signaling, ERK pathways and EMT.

### MEG3

MEG3 has already been verified to be involved in the resistance of LAD cells to cisplatin. In detail, firstly, there was a lower expression level of MEG3 in A549/DDP cells compared to parental A549 cells. When transfected with pcDNA-MEG3, A549/DDP showed a significantly decreased IC50 to cisplatin compared to control group. However, MEG3 knockdown led in an increased IC50 in A549 cells. Secondly, in pcDNA-MEG3-transfected A549/DDP cells combined with cisplatin treatment, the apoptotic rate showed a remarkable cisplatin dose-dependent increase, while cell proliferation was impaired and cell cycle was arrested at G1/G0 phase. The knockdown of MEG3 expression level using si-MEG3 resulted in the promotion of cell proliferation and the impairment in cisplatin-induced apoptosis of A549 cells. Cell cycle got a progression with decreased G1/G0 phase and increased S phase in the same context [99]. All the above results remind us that MEG3 is a contributing factor in the development of cisplatin resistance of A549/DDP.

Furthermore, the underlying mechanisms stressed that MEG3 had an implication in mitochondrial apoptosis pathway and induced cisplatin-resistant LAD cells apoptosis by regulating p53 and pro-survival protein Bcl-xl expression [99]. The role of MEG3 overexpression in augmenting cisplatin chemosensitivity of LAD cells was also elucidated in nude mice models. The analysis of 41 LAD patients revealed the result that MEG3 expression had a crucial effect on the response of patients to cisplatin-based therapy [99]. Taken together all the evidence, the long non-coding RNA MEG3 is conspicuously correlated with the resistance of human lung adenocarcinoma to cisplatin.

### LINC00635-001

Linc00635-001 with 2119 nt length is located in chromosome 3. In a study on EGFR-TKIs-resistant lung cancer, it has been proposed that linc00635-001 is overexpressed in gefitinib-resistant HCC827-8-1 cells. Si-linc00635-001 inhibited resistant cell proliferation and colony formation and increased the apoptosis rate and cleaved caspase 3, which revealed

that silenced linc00635-001 could re-sensitize HCC827-8-1 cells to gefitinib treatment by promoting drug-induced apoptosis. Further investigation about mechanism, by which the apoptosis of HCC827-8-1 cells is induced via si-linc00635-001 plus gefitinib, reported that p-Akt expression reduced [100]. The expression of linc00635-001 is related to pre-B-cell leukemia homeobox (PBX)-3, one transcriptional factor involved in EGFR/Akt signaling pathway and tumor growth [101]. These data collectively imply that linc00635-001 is implicated in the resistance of lung cancer to EGFR-TKI through PBX regulating p-Akt.

### Blood-based/circulating lncRNAs

#### UCA1

As mentioned above, lncRNA UCA1 is associated with NSCLC progression and acquired EGFR-TKIs resistant NSCLC. Also, Wang et al. found that UCA1 was overexpressed in tissues and plasma from patients with non-resistant NSCLC. Advanced histological grade and distant lymph node metastasis have a tendency to develop in patients with a higher expression level of UCA1, which suggests UCA1 may facilitate the progression of lung cancer functioning as an oncogene and predict overall survival for patients. Moreover, the expression level of UCA1 between tumor tissues and plasma had a highly positive correlation. As a result, lung cancer could be conveniently and easily diagnosed by detecting the level of UCA1 in plasma from patients. UCA1 might be developed a promising diagnostic biomarker and therapeutic target in the future clinical application for NSCLC patients [102].

#### MALAT1

MALAT1 in the whole blood, including cell fraction and plasma, could serve as a potential biomarker with an AUC more than 0.70 for discrimination between NSCLC patients and healthy people. MALAT1 theoretically might be derived from the cells indirectly affected by tumor in the immune system such as leucocytes. The expression level of MALAT1 in the blood cells was downregulated when compared to that of controls, which may due to a weak response of immune system caused by tumor. MALAT1 also might originate from the release of vesicles including lncRNA from *in situ* tumor and exist in

the plasma of patients. Recent observations demonstrated that the expression of MALAT1 was significantly associated with metastasis. A stronger expression of MALAT1 was confirmed in the whole blood with bone or brain metastasis than that with lymph node metastasis, because the circulating tumor cells (CTCs) with abundant MALAT1 transcripts didn't colonize in the lymph node. Moreover, it is sure that the combinational application of MALAT1 and other biomarkers is more sensitive for the diagnosis of lung cancer [103, 104].

### *XIST & HIF1A-AS1*

By microarray assay, XIST and HIF1A-AS1 are verified to be overexpressed in NSCLC tissues. Also, the two lncRNAs could reach the circulation, as examined by qRT-PCR, and their expression level in serum is positively correlated with that in tumor tissues. Resecting tumor could result in the decrease of circulating XIST and HIF1A-AS1, which implies that blood-based lncRNAs may be from released tumor cells. These data introduce XIST with an AUC of 0.834 and HIF1A-AS1 with an AUC of 0.876 to be potentially predictive biomarkers for NSCLC. There is an intriguing discovery that the combination of XIST and HIF1A-AS1 in serum could provide an more accurate diagnose for NSCLC, with an improved AUC of 0.931 [105].

### *Prospective application of lncRNAs in lung cancer*

As lung cancer has become one of malignant diseases damaging human physiological and mental health, it is urgent to find non-invasive cancer biomarkers with high sensitivity and specificity for the earliest diagnosis. lncRNAs were defined as distinct transcripts partly due to their cancer or tissue-specific expression [10], building up a good basis for cancer prediction. Circulating lncRNAs may be secreted from tumor cells, these blood-based lncRNAs could be utilized as fingerprinting molecules for lung cancer diagnosis [106]. It turns out that the combination of lncRNAs with each other indicates a higher value [107, 108]. Additionally, dysregulated lncRNAs could be predictors for the prognosis of lung cancer patients.

The finding of misregulated lncRNAs in lung cancer is conducive to endowing cancer patients with the individual and normative treat-

ment. Multiple assays suggest that silenced oncogenic lncRNAs or overexpressed tumor suppressive lncRNAs may inhibit lung cancer cell growth. In this regard, RNAi-based strategies will open a new avenue for lung cancer therapy [109, 110]. It is well established that lncRNAs have abilities to recruit chromatin or histone modification complexes to epigenetically silence particular genes, which gives us a hint that we may develop targeted drugs acting as obstacles to epigenetic modification-mediated signaling pathways. Another approach could be translated in clinical therapy that targeting aberrant RNA secondary structure motifs via modularly assembled small molecules to intervene in their protein-binding or other potential [111].

Given that there have not yet been comprehensive knowledge about the functional mechanisms of lncRNAs, we need further exploratory to identify and validate their biologic robustness in diagnostic, prognostic and therapeutic aspects.

### **Conclusion**

The advent of high-throughput sequencing technologies makes lncRNA to be a noticeable superstar in RNA field. lncRNAs have emerging as influential regulators in the biological and pathological contexts. Hence, the regulation of tumor biology is not limited to protein-coding genes. The discoveries of lncRNA fuel the study in this field and pave the way for cancer therapies.

The lncRNA study has drawn increasing attention and gradually gained unprecedented prominence. Previous reports with high-throughput sequencing technologies have revealed the crucial function of lncRNA in lung cancer. In this review, we elucidated the dysregulation of lncRNA and its involvement in driving and progressing lung cancer (**Table 1**). The inaccurate guidance of lncRNA encourages us to understand the pathogenesis of lung cancer more definitely, supporting the inference that the application value of lncRNA is unexpectedly tremendous in the clinical. However, the clinical translation of lncRNAs as predictors, diagnostic biomarkers or anti-tumors drugs targets have not yet been put into practice. Frankly, Rome is not built in a day, so it is a long way for searching RNA world.

## LncRNAs in lung cancer

**Table 1.** Dysregulated lncRNAs and their function in lung cancer

LncRNA	Expression	Cell processes	Interaction	Clinical association	References
MALAT1 (NEAT2)	↑in NSCLC & CSE-exposed HBE cells	Migration, EMT	MIR-217, EZH2, CXCL5	Metastasis, biomarker, prognosis	[17, 22-25, 27, 28, 103, 104]
ANRIL	↑in NSCLC & PLD inhibition H460 cell	Proliferation, cycle, apoptosis, migration	EZH2, KLF2, p21, p15	TNM stage, lymph node metastasis, tumor size, prognosis	[29-32]
HOTAIR	↑in (metastatic/hypoxia) NSCLC & (pure)SCLC & DDP-resistant LAD	Proliferation, migration, invasion, EMT	HOXA5, MMP, Col-1, HIF-1 $\alpha$ , p21, HOXA1, cell-adhesion genes, DNMT1, DNMT3b	Metastasis, recurrence, tumor size, TNM stage and lymphatic, vascular metastasis, resistance to cisplatin, prognosis	[6, 33-36, 38, 97]
Sox2ot	↑in LSCC	Proliferation, cycle, migration	EZH2	Histological type, survival	[39, 42]
CCAT2	↑in NSCLC	Proliferation, invasion	-	Histological type, biomarker	[7]
CARLo-5	↑in NSCLC	Proliferation, cycle, EMT, invasion	P16, p21, p27	Advanced stage, lymph node metastasis, prognosis	[43]
MVIH	↑in NSCLC	Proliferation, invasion	MMP	Advanced stage, lymph node metastasis, tumor size, prognosis	[45]
ZXF1 (uc001kfo)	↑in LAD	Migration, invasion	$\alpha$ -SMA, ACTA2, TGF- $\beta$ 1	Lymph node metastasis, tumor pathological stage, prognosis	[46]
HNF1A-AS1 (C12orf27)	↑in LAD	Proliferation, cycle, migration, invasion, EMT	DNMT1	Tumor size, TNM stage, lymph node metastasis, prognosis	[47]
BCYRN1 (BC200)	↑in NSCLC	Migration, invasion	C-MYC, MMP9, MMP13	-	[48]
H19	↑in NSCLC	Proliferation, cycle, EMT	C-MYC, p53, HIF1- $\alpha$ , miR-107	Tumor size, TNM stage, prognosis	[49, 51, 52]
LUADT1	↑in LAD	Proliferation, cycle	SUZ12, p27	Tumor stage	[54]
AFAP-AS1	↑in NSCLC	Migration, invasion	Rho/Rac GTPase family members, actin cytoskeleton signaling pathway	Clinical stage, smoking history, infiltration degree, lymph node metastasis, distant metastasis, prognosis	[55, 56]
PVT1	↑in NSCLC	Proliferation, migration, invasion, apoptosis, cycle	MYC protein	Histological grade, lymph node metastasis, tumor size, prognosis	[57, 58]
RGMB-AS1	↑in NSCLC	Proliferation, cycle, migration, invasion	RGMB	Differentiation status, lymph node metastases, TNM stage	[59, 60]
GHSROS	↑in NSCLC	Migration	-	-	[61]
Lnc-bc060912	↑in NSCLC	Proliferation, apoptosis	P53, PARP1, NPM1	-	[62]
NEAT1	↑in NSCLC	-	MIR-449a	Patient age, lymphatic metastasis, vascular invasion, TNM stage	[63, 64]
DLX6-AS1	↑in LAD	-	DLX6	Histological differentiation, TNM stage	[65]
LINC01133	↑in NSCLC	Migration, invasion, proliferation, cycle, apoptosis, EMT	EZH2, LSD1, KLF2, p21	Prognosis, tumor size, advanced pathological stage, lymph node metastasis	[66, 67]
IRAIN	↑in NSCLC	Proliferation, cycle	IGF-1R signaling	Smoking status, tumor size	[68]
UCA1	↑in NSCLC & acquired EGFR-TKIs resistant NSCLC	Proliferation, apoptosis, EMT	MIR-193a-3p, ERBB4, Akt/mTOR & ERK signaling pathways	Tumor size, TNM stage, prognosis, resistance to EGFR-TKIs, biomarker	[69, 98, 102]
AK001796	↑in lung cancer & ↓in resveratrol-treated lung cancer cells	Proliferation, cycle	Cell cycle-related genes	-	[70]
LINC01207	↑in LAD	Proliferation, apoptosis	EZH2, Bad	TNM stage, prognosis	[71]

## LncRNAs in lung cancer

PCAT-1	↑in NSCLC	Proliferation, migration, invasion	-	-	[73]
TATDN1	↑in NSCLC	Proliferation, migration, invasion,	Nm23-H1, HER2, E-cadherin, β-catenin, Ezrin	-	[74]
linc00673	↑in NSCLC	Proliferation, cycle	LSD1, NCALD	Tumor size, lymph node metastasis, TNM stage	[75]
LINC00473	↑in NSCLC	Proliferation	LKB1/CRTC1/CREB signaling axis, NONO	Prognosis, biomarker	[76]
HMIlncRNA717	↓in NSCLC	-	-	Histological grade, lymph node metastasis, prognosis	[78]
SPRY4-IT1	↓in NSCLC	Proliferation, apoptosis, migration, invasion, EMT	EZH2	Tumor size, advanced stage, lymph node metastasis, prognosis	[79]
GAS5	↓in NSCLC	Proliferation, apoptosis	P53, E2F1, EGFR/PI3K/Akt pathway, IGF-1R	Tumor size, TNM stage, tumor differentiation, EGFR-TKIs resistance	[4, 82]
GAS6-AS1	↓in NSCLC	-	GAS6	Lymph node metastasis, advanced TNM stage, histological grade, histological classification, prognosis	[83]
BANCR	↓in NSCLC	Proliferation, apoptosis, migration, invasion, EMT	HDAC3	Tumor size, advanced stage, metastasis distance, prognosis	[84]
PANDAR	↓in NSCLC	Proliferation, apoptosis	P53, NF-YA, Bcl-2	Tumor size, advanced TNM stage, prognosis	[85]
TUG1	↓in NSCLC	Proliferation, cycle, apoptosis	P53, EZH2, HOXB7, Akt & MAPK pathways	TNM stage, tumor size, prognosis	[86]
MEG3	↓in NSCLC & cisplatin-resistant NSCLC	Proliferation, apoptosis, cycle	MDM2, p53, miR-3163, Skp2, Bcl-xl	Advanced stage, tumor size, prognosis	[87, 88, 99]
CASC2	↓in NSCLC	Proliferation, cycle	-	Tumor size, TNM stage, prognosis	[91]
ENST00000434223	↓in NSCLC	Proliferation, apoptosis, cycle, migration, invasion, EMT	-	Tumor size, lymph node metastasis, TNM stage	[92]
AF118081	↑in lung cancer & anti-BPDE transformed 16HBE cells	Proliferation, apoptosis, migration, invasion	-	Malignant transformation	[93]
SCAL1	↑in metastatic-prone lung cancer cells & CSE-exposed airway epithelial cells	Cell survival	NRF2	Weaken cigarette smoke toxicity	[94]
Lnc-DQ786227	↑in lung cancer cells & B [a] P-transformed BEAS-2B cells	Proliferation, apoptosis	-	Malignant transformation	[95]
AK126698	↓in cisplatin-resistant NSCLC	Cell survival	Cell cycle & MAPK & Wnt signaling pathway, NKD2	Cisplatin resistance	[96]
LINC00635-001	↑in EGFR-TKIs resistant LAD	Cell survival	Akt	EGFR-TKIs resistance	[100]
XIST & HIF1A-AS1	↑in NSCLC	-	-	Biomarker	[105]

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## Disclosure of conflict of interest

None.

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