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Review article

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The development of potential targets in the treatment of non-small cell lung cancer

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Abstract: The oncogenic driver mutations have been found that not only have potential sensitivity to epidermal growth factor receptor but also can inhibit anaplastic lymphoma kinase tyrosine kinase; more and more interest has been evoked in discovering additional targets to non-small cell lung cancer (NSCLC). Recently, many novel underlying oncogenic gene alterations have been identified, such as HER2 insertions, BRAF mutations, PIK3 mutations, FGFR1 amplifications, DDR2 mutations, KRAS mutations, MET amplification, ROS1 rearrangements, ALK rearrangements, and RET rearrangements. In this review, we will discuss the discovery of these potential targets and the application of each in NSCLC and of small molecular inhibitors on these potential targets.

Keywords: NSCLC, Potential target, HER2, BRAF, KRAS

1 Introduction

Lung carcinoma is the main reason for tumor-associated mortality worldwide, and it leads to more deaths than a combination of those caused by colorectal cancer, breast cancer, and prostate cancer [1]. NSCLC is a main origin of tumor-associated death and 85% or more patients are diagnosed with NSCLC at an advanced stage [2]. Typical therapy agents have been approached through two primary methods block VEGF- VEGFR binding or low molecule weight tyrosine kinase inhibitors (TKIs) that inhibit downstream VEGFR mediated signaling [3]. Recently, important advances have been discovered, including finding oncogene driver mutations [4]; the EGFR–TKIs inhibitor in people who carry vigorous EGFR mutations [5]; EGFR tyrosine kinase inhibitor erlotinib and the anaplastic lymphoma kinase (ALK) TKI crizotinib [6]. However, survival rate and morbidity of patients who carry advanced NSCLC tumors are still to be improved and reduced. In this review, we will make an introduction to new and key advances of the application of drugs for treating NSCLC recently.

2 Human epidermal growth factor receptor 2

HER2 is a representative member of the EGFR family relating to tumor growth, development, and neoplasm recurrence [7]. Due to the absence of a ligand, so far there is no signaling-related drug to other HER receptors by producing heteromeric complexes with HER1, HER3, and HER4 to be discovered [8]. Among all the complexes which may include HER2, HER2/HER3 heterodimers exhibit the potential to contribute to tumor cell growth. The remarkable signaling potency of HER2/ HER3 heterodimers uncovers the fact that this dimer can promote development via the ras/raf/MAPK pathway and escape from apoptosis through the PI3K/Akt pathway [9].

Through sequencing the gene which encodes the transmembrane protein tyrosine kinase HER2, we have discovered 4% mutations in the kinase domain from 120 primary lung tumors [10]. For the adenocarcinoma type of tumor, there are 10% mutations. So far, HER2 inhibitors have been shown to be ineffective in those with no HER2 mutation. So it will be re-evaluated in a clinical setting in lung cancer patients with HER2 mutations.

There are 1.6% (11 of 671) HER2 mutations in patients with NSCLC and there are none in other tumors. In addition, it is discovered that NCI-H1781 has a mutation. There are more HER2 mutations in never smokers and adenocarcinoma histology compared with smokers and with people with no tumor [11], which is similar to

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EGFR TK domain mutations [12]. Importantly, we found that targeted expression of the HER2 kinase domain duplication/insertion YVMA mutant in murine pulmonary tissue is intensely oncogenic, promoting quick tumor growth mainly in proximal and distal airway epithelia of most mice [13].

To date, several inhibitors that target EGFR and HER2 mutations have been studied in phase III trials, including neratinib and dacomitinib. Takeshi has proved that neratinib can inhibit growth by arresting G1 and induce apoptotic cell death in the HER2-mutant cell [14]. Jeffrey confirmed that dacomitinib, an irreversible Pan-HER2 inhibitor, is effective against HER2 mutations which gefitinib had failed to treat [15]. Moreover, De Grève implies afatinib is a promising novel drug for patients who have nonsmoking history and metastatic lung tumors with HER2 mutations, especially when other EGFR and HER2 drugs are ineffective [16]. Interestingly, TKI-dacomitinib which is a second-generation quinazalone-based irreversible HER2 family inhibitor, is more effective in patients who express both EGFR and HER2 but are resistant to erlotinib. However, only 3 of 18 patients had a response to dacomitinib, which implies that the effect of inhibiting cancer gene remains to be deliberated [17].

3 v-raf murine sarcoma viral oncogene homolog B1, BRAF

BRAF produces the B-Raf protein, which takes part in sending signals inside cells, which are important for proliferation. BRAF is an important transduction factor of the RAS–RAF–MEK–ERK–MAP kinase pathway located in chromosome 7q34, about 190kb long. Davies et al. show that the frequency of BRAF somatic missense mutations is 66% for malignant melanomas [18]. However, they have lower probability in other human tumors. At nucleotide 1796, A replaces T in more than 90% in BRAF mutations. The change leads to replacement of valine by glutamic acid at position 599. The mutations at position 599 can drive endogenous ERK1/2 to phosphorylate more intensely than ^{G463V}BRAF or ^{L596V}BRAF. This mutation may produce a dummy phosphorylation in the activation segment when they insert a negatively charged residue near to the regulatory phosphorylation position S598. Activation of BRAF signal, partially, can promote proliferation by the MAPK signal pathway [18]. Because BRAF is usually caused by a somatic point mutation in human tumors and at the same time it is a serine/threonine kinase, it could offer a novel treatment strategy of NSCLC.

The interest in mutations in NSCLC was aroused by the great probability of mutation in malignant melanoma. Brose et al. found that the frequency of BRAF mutations in NSCLC was 3 of 195 and 2 of 242 for exons 11 and 15 respectively, and that NSCLC BRAF mutations are more likely to be non-V599. Additionally, the investigators discovered that 3% and 4.9% of 696 [19] and 739 [20] lung tumor patients respectively have BRAF mutations. As we expected, current and former smokers have higher risk of mutation. Importantly, the non-V600E mutation rate in lung cancer is much higher than that of melanoma [20] and smokers have much higher risk of non-V600E mutations [20,21]. Moreover, V600E mutations were obviously more prevalent in females than in males (8.6% vs. 0.9%) [21]. Based on the above-mentioned discovery, a phase II clinical trial is evaluating the BRAF kinase inhibitor dabrafenib for patients who have BRAF V600E mutation positive metastatic (Stage IV) NSCLC.

4 Phosphatidylinositol 3-kinase mutations

Phosphatidylinositol 3-kinases (PI3Ks) are lipid kinases and can modulate signaling pathways, which is key for the cell growth, adhesion, survival, and motility of neoplasia [22]. To identify if the PI3Ks genes of cancers are changed, Samuels et al. compared PI3K genes of human tumors with related normal tissue [22]. They found that only PIK3CA, which can encode the p110 catalytic subunit, has somatic mutations. The positions of the mutations within PIK3CA imply that they are related to increasing kinase activity.

In addition, Yamamoto et al. and Rekhtman et al. identified mutations in 4.7% [23] and 4% [24] of NSCLC cell lines, respectively. Amazingly, we have enough firm evidence to determine that EGFR/KRAS mutations are closely related to carcinomas which have glandular differentiation, while they were not correlated with simple squamous cell carcinoma (SQCC). However, there are PIK3CA mutations both in adenocarcinoma and in SQCC, and compared with adenocarcinoma, they appear at high frequency in SQCC [24], indicating that PIK3CA mutations are not the true driver mutations, but the second mutations based on the driver mutations. Furtherly, PIK3CA mutations can induce lung carcinoma in the transgenic mouse model.

Additionally, the tumor regressed after knocking out the PIK3CA [25]. PIK3 is more active in the PIK3CA mutation lung cancer cell lines, whereas RNA interference can downregulate PIK3 and inhibit the lung cancer cell. Jeffrey et al. imply inhibitors of the PIK3-mTOR pathway

are probably effective for PIK3CA mutated tumors and, when administered with MEK inhibitors together, may provide therapy for lung cancers with KRAS mutations [25]. Although we have proved that inhibiting the phosphorylation of PIK3 can induce tumor response, it remains to be controversial whether it is effective in breast cancer cells. In order to solve these problems, some clinical trials are being conducted involving single PIK3 inhibitor (NCT01501604), PIK3 inhibitor combination with chemotherapy or other targeted drugs (NCT00974584).

4.1 Fibroblast growth factor receptor 1

Fibroblast growth factor receptor 1 is a membrane-bound receptor tyrosine kinase which modulates the growth of cells via the MAPK and PI3K pathways, similar to EGFR. There are higher rates of FGFR1 amplification in SCLC than that of lung cancer. Additionally, we found that lung tumor can survive relying on the activity of the FGFR1 kinase [26]. Investigating low molecular weight FGFR1 suppressors has achieved some progress. The FGFR inhibitor PD173074 was found to suppress the proliferation of lung tumors with FGFR1 amplification, but not of those without FGFR1 amplification [26]. In the clinical setting, the most investigated FGFR inhibitor was brivanib and it can not only inhibit FGFR but also inhibit the VEGFR pathway. However, there is the similar secondary action which is the same as other VEGF inhibitors [27]. The investigators have conducted a randomized discontinuation study of 396 patients who have advanced solid tumors; there are 42 patients not responsive to brivanib who have made a partial recovery from NSCLC [28]. The new aim is to research novel FGFR inhibitors with less toxicity [29].

4.2 DDR2 mutations

DDR2 is a membrane-bound receptor tyrosine kinase binding to collagen and can modulate the growth and metastasis of tumors. Recently, via gene screening, DDR2 gene mutations were discovered in 2.2% of squamous cell lung cancers [30]. To determine the oncogenic function of DDR2, the researchers discovered that the proliferation of cells with DDR2 mutation was damaged after DDR2 expression was knocked down, suggesting that DDR2 is involved in the proliferation.

While DDR2 has not been regarded as a main target of drug research, a recent report suggests that ABL kinase suppressors suppress DDR2, including imatinib, nilotinib, and dasatinib [31]. In addition, dasatinib can inhibit proliferation of lung tumor cells with DDR2 mutations. Unluckily, there is no special effect of dasatinib on NSCLC, with only 34 patients partially responsive to it, of which only six were SCLC patients, in the phase II study [32]. Interestingly, a patient subject to SCLC responded to dasatinib and erlotinib in the single phase II study, and the tumor tissue had a point DDR2 mutation as discovered through gene sequencing [30].

4.3 KRAS mutations

KRAS is a guanine nucleotide (GDP/GTP)-binding protein which is a self-inactivating signal transducer. Cell surface receptors can drive the switch of bound GDP for GTP, with the result that it can activate the wildtype KRAS instantly. KRAS oncogenes contain single point mutations leading to amino acid replacement and produce proteins with intensely reduced GTPase activity. Thus, KRAS-mutated proteins are fixed in a composite activating GTP-bound state. [33].

There are 25% KRAS mutations in lung tumors of patients in western countries; the percentage in Asian countries is much lower. The rate of KRAS mutation is not related to age, gender, or smoking history. There are higher risks of transition mutation (G to A) in non-smokers compared with that of smokers. This discovery subverts the previous view that these mutations are smoking-related [34]. The investigators have found that k-RAS oncogene somatic mutations caused thorough resistance to EGFR tyrosine-kinase inhibition in NSCLC patients. Although it was first identified that KRAS can be regarded as an oncogene in tumors, there is no special treatment for NSCLC with KRAS mutation [35]. Selumetinib combined with docetaxel provides hope for treatment, although it can have more side effects compared with docetaxel alone, in the study of therapy for advanced NSCLC with KRAS mutation. These discoveries require clinical experiments to confirm the effectiveness of selumetinib in combination with docetaxel in KRAS-mutant non-small cell lung cancer [36].

4.4 MET amplification

MET is the receptor for hepatocyte growth factor and it is usually highly produced in non-small cell lung cancer (HGF) which can promote cell proliferation, invasion motility, and angiogenesis in in vitro studies [37]. There was 22% MET amplification in lung cancer specimens resistant to gefitinib and erlotinib and 1.4% to 20% in NSCLC. The

resistance is driven by activation of PI3K by its specific receptors ERBB3. Thus, we suggest MET amplification is likely to stimulate other ERBB-driven tumors' resistance to the drug; in addition, EGFR-TKI can generate resistance by MET gene amplification [38]. Furthermore, there was higher risk of death in MET-positive patients compared with MET-negative patients in many models. Gefitinib and erlotinib are ineffective in 21% of patients and there were only 3% untreated patients due to MET amplification; these findings suggest that MET may be a novel promising breakthrough for patients who are resistant to EGFR-TKIs [37].

There are long progression-free survival (PFS) and overall survival (OS) of combining tivantinib with erlotinib compared with placebo combination with erlotinib for patients with non-squamous history and MET overexpression [39]. The study of Lim et al. shows that MET mutations were significantly correlated with decreased survival [40]. In addition, the inhibition of tumor proliferation would be significantly improved through targeting to MET/RON kinases especially in various in vitro and in vivo models of NSCLC. These findings provide enough firm evidence that LY2801653 is a potential low molecular weight drug for MET/RON targeting therapy for NSCLC which can target MET/RON kinases especially [41].

4.5 ROS1 rearrangements

ROS1 is a receptor tyrosine kinase and down-regulates the MAPK signaling cascade via phosphorylation of RAS. The ROS1 gene can produce ROS protein, which is found within the membrane of human cells and plays a key role in proliferation of cancer and cell specialization. Mutation of ROS1 gene can result in NSCLC. There are 1% to 2% of individuals with NSCLC with ROS1 gene mutation which is fused to parts of another gene**.** Through a phosphoproteomic screen, the investigators discovered that the ROS1 gene is an oncogene within NSCLC [42]. ROS1 rearrangements were discovered previously in glioblastoma [43] and in NSCLC it was also reported.

Recently, ROS1 rearrangement was researched in 1,073 NSCLC patients by the break-apart FISH assay [44]. Subsequently, it was identified in 1,116 patients who were subject to lung cancer via the same method. There were 13 patients who carry rearrangements, among them 11 patients verified through RT-PCR [45]. ROS1 was famous as an oncogene of glioblastoma for many years [43]; however, the development of targeting ROS1 inhibitors did not make progress in clinical trials. While the investigators have suggested ALK inhibitors are effective in cells with ROS1 rearrangements, such as TAE684, a selective ALK inhibitor 602 cell line has sensitivity. Amazingly, we found that it can kill more than 50% of cells in 10 cell lines after the cells were exposed to the ALK inhibitors for 72h [46]. Even though the great majority of cell lines are sensitive to ALK alterations, only HCC-78 was discovered to cover ROS1 translocation so far [42]. After it was confirmed that HCC-78 harbors a ROS1 translocation, we found that the HCC78 ROS1-rearranged NSCLC cell line showed sensitivity to crizotinib [44]. Now more and more ongoing phase I trials have shown that 64% of patients with ROS1-rearranged lung tumors responded to it strongly [47].

4.6 ALK rearrangements

The ALK gene can direct anaplastic lymphoma kinase production. ALK was first identified as a fusion partner of nucleophosmin (NPM) in anaplastic large-cell lymphoma. Combinating EML4 with ALK can produces the aberrant EML4-ALK fusion gene, which are diversse chimeric variants, which can produce the similar fraction ALK, however it includes various volume of EML4. 6.7% of NSCLC patients carry the EML4–ALK fusion transcript; these patients were different from those with mutations in EGFR [48]. EML4-ALK place-changing can induce mutation because it is in charge of switching on and retaining lung tumors. The frequency of EML4-ALK-positive patients is 5.7% in western countries and 2.9%~6.7% in Asian countries [45]. ALK rearrangements were related to various clinical pathologic features, such as age, smoking history, and cancer history. EGFR and KRAS mutations generally did not coexist with EML4–ALK fusion gene; if they did, it implied resistance to EGFR and TKI. However it did after taking crizotinib. Portions of NSCLC patients are likely expressing the transforming fusion kinase which is not only a novel potential therapeutic target but also a diagnostic molecular marker of NSCLC [49]. Crizotinib has recently become a standard therapy in several countries for patients who are subject to advanced and ALK-positive NSCLC [50]. Ceritinib can be administered orally and is a potent second-generation ALK inhibitor. Preclinical experiment suggested impressive inhibition of cancer activity in crizotinib-resistant clones, and based on available data, ceritinib could represent a suitable option in crizotinib-resistant NSCLC [51].

4.7 RET

RET is the third receptor tyrosine kinase established and RET rearrangements were first identified in thyroid cancers

[52]. RET means that it will "rearrange during transfection"; the DNA of RET was first discovered rearranged in a 3T3 fibroblast cell line and transfection took place after it. Recently, RET rearrangements were discovered in NSCLC by using different screening strategies from cancer and normal tissue which originated from a lung cancer patient, in whom there were no driver mutation or fusion, who had no smoking history, and whose family has no tumor history [53]. KIF5B is a new chimeric fusion transcript, which was originally discovered as a new driver mutation in NSCLC by four independent research groups simultaneously. Soon afterwards, the investigators discovered in-frame fusion transcripts of KIF5B and RET oncogene. It was present in 1–2% of lung cancers from Japan and USA as detected by whole-transcriptome sequencing [54]. Furthermore, by screening 561 lung cancers, we confirmed another 11 cancer patients who carry KIF5B-RET gene mixture from one of 24 NSCLC specimens studied. Importantly, we discovered that multi-kinase inhibitors not only can inhibit RET but also are effective against cells that can express oncogenic KIF5B-RET [55]. As well as ALK rearrangements and ROS1 rearrangements, there is a high rate of RET rearrangements in young, non-smoking patients with adenocarcinomas [56].

A few of the available TKIs on the market are effective against RET kinase. For example, in addition to inhibiting RET phosphorylation, vandetanib was confirmed to inhibit the proliferation of various cell lines transformed by KIF5B-RET [54]. Two VEGFR inhibitors, sunitinib and sorafenib not only are effective against RET kinase, but also can inhibit the proliferation of KIF5-BRET–transformed cell lines [55]. Cabozantinib, which is capable to inhibiting medullary thyroid cancer, can also suppress KIF5B-RET–transformed cell lines [57]. However, there are not enough data to establish the application of RET inhibitors clinically.

5 Conclusion

In a word, targeting oncogenic driver mutations of tumors has a great potential to make some progress against lung cancer and squamous cell tumors in the last decades. Many of these oncogenic alterations were discovered in other tumors, which can suggest to identify them in NSCLC as well. At present, the greatest challenge is expanding more effective experiments to confirm the level of gene alterations. In addition, how to select patients for clinical trials and testing is the key to take advantage of underlying drug sensitivity in new oncogenes that have been identified. Because screening for mutations of each oncogene will uncover personal differences, we are likely to eventually develop multigene tests and personalized tumor therapies. So in the coming years, we will make the greatest progress through conducting trials of promising targeted therapies.

Conflict of interest: Authors declare nothing to disclose.

References

[1] Khan I., Morris S., Hackshaw A., Lee S.M., Cost-effectiveness of first-line erlotinib in patients with advanced non-small cell lung cancer unsuitable for chemotherapy, BMJ Open, 2015, 5, e006733.
[2] Alberg A.J., Samet J.M., Epidemiology of Lung Cancer, CHEST, 2003, 123, 21S–49S
[3] Lee M., Ellis Daniel J., Hicklin, Pathways Mediating Resistance to Vascular Endothelial–Targeted Therapy, Clin. Cancer Res., 2008, 14 (20), 6371-6375.
[4] Kris M.G., Johnson B.E., Berry L.D., Kwiatkowski D.J., Iafrate A.J., Wistuba I.I., et al, Using Multiplexed Assays of Oncogenic Drivers in Lung Cancers to Select Targeted Drugs, JAMA, 2014, 311 (19), 1998-2006.
[5] Rafael R., Enric C., Radj G., Vergnenegre A., Massuti B., Felip E., et al, Erlotinib versus standard chemotherapy as first-line treatment for European patients with advanced EGFR mutation-positive non-small-cell lung cancer (EURTAC): a multicenter, open-label, randomized phase 3 trial, Lancet Oncol., 2012, 13, 239–46.
[6] Shaw A.T., Kim D.W., Mehra R., Daniel S.W., Tan, M.B., Enriqueta B.S., et al, Ceritinib in ALK-Rearranged Non–Small-Cell Lung Cancer, N. Engl. J. Med., 2014, 370, 1189-97.
[7] Normanno N., De Luca A., Bianco C., Strizzi L., Mancino M., Maiello M.R., et al, Epidermal growth factor receptor (EGFR) signaling in cancer, Gene, 2006, 366, 2–16.
[8] Thomas P.J., Garrett, Neil M., McKern, Meizhen Lou, Thomas C. Elleman, et al, The Crystal Structure of a Truncated ErbB2 Ectodomain Reveals an Active Conformation, Poised to Interact with Other ErbB Receptors, Mol. Cell, 2003, 11, 495–505.
[9] Ami C., Kochupurakkal B.S., Yosef Y., The deaf and the dumb: the biology of ErbB-2 and ErbB-3, Exp. Cell Res., 2003, 284, 54–65.
[10] Tatem A. J., Carlos A., Guerra, Atkinson P.M., Hay S. I., Momentous sprint at the 2156 Olympics, Nature, 2004, 431, 525-526.
[11] Shigematsu H., Takahashi T., Nomura M., MajmudarK., Suzuki M., Lee H., et al, Somatic Mutations of the HER2 Kinase Domain in Lung Adenocarcinomas, Cancer Res., 2005, 65 (5), 1642-1646.
[12] Shigematsu H., Lin L., Takahashi T., Suzuki M., Wistuba I.I., Fong K.M., et al, Clinical and Biological Features Associated With Epidermal Growth Factor Receptor Gene Mutations in Lung Cancers, J. Natl. Cancer Inst., 2005, 97 (5), 339-346.
[13] Pereraa S.A., Li D., Shimamura T., Raso M.G., Ji H.B., Chen L., et al, HER2YVMA drives rapid development of adenosquamous lung tumors in mice that are sensitive to BIBW2992 and

rapamycin combination therapy, Proc. Natl. Acad. Sci. USA, 2009, 106, 474–479.
[14] Shimamura T., Ji H., Minami Y., Thomas R.K., Lowell A.M.,Shah K.,et al, Non-small cell lung cancer and Ba/F3 transformed cells harboring the ERBB2 G776insV_G/C mutation are sensitive to the dual-specific epidermal growth factor receptor and ERBB2 inhibitor HKI-272, Cancer Res., 2006, 66, 6487-6491.
[15] Engelman J.A., Zejnullahu K., Gale C.M., Lifshits E., Gonzales A.J., Shimamura T., et al, PF00299804, an irreversible pan-ERBB inhibitor, is effective in lung cancer models with EGFR and ERBB2 mutations that are resistant to gefitinib, Cancer Res., 2007, 67, 11924-11932.
[16] De Greve J., Teugels E., Geers C., Decoster L., Galdermans G., De Mey J., et al, Clinical activity of afatinib (BIBW 2992) in patients with lung adenocarcinoma with mutations in the kinase domain of HER2/neu, Lung Cancer ,2012, 76, 123-127.
[17] Kelly R.J., Carter C., Giaccone G., Personalizing therapy in an epidermal growth factor receptor– tyrosine kinase inhibitor–resistant non–small-cell lung cancer using PF-00299804 and trastuzumab, J. Clin. Oncol., 2010, 28, e507-e510.
[18] Davies H., Bignell G.R., Cox C., Stephens P., Edkins S., Clegg S., et al, Mutations of the BRAF gene in human cancer, Nature, 2002, 417, 949- 954.
[19] Brose M.S., Volpe P., Feldman M., Kumar M., Rishi I., Gerrero R., et al, BRAF and RAS mutations in human lung cancer and melanoma, Cancer Res., 2002, 62, 6997-7000.
[20] Paik P.K., Arcila M.E., Fara M., Sima C.S., Miller V.A., Kris M.G., et al, Clinical characteristics of patients with lung adenocarcinomas harboring BRAF mutations, J. Clin. Oncol. 2011, 29, 2046-2051.
[21] Marchetti A., Felicioni L., Malatesta S., Grazia S.M., Guetti L., Chella A., et al, Clinical features and outcome of patients with non–small-cell lung cancer harboring BRAF mutations, J. Clin. Oncol., 2011, 29, 3574-3579.
[22] Samuels Y. Waldman T., Oncogenic mutations of PIK3CA in human cancers, Curr. Topic. Microbiol. Immunol., 2010, 347, 21-41.
[23] Yamamoto H., Shigematsu H., Nomura M., Lockwood W.W., Sato M., Okumura N., et al, PIK3CA mutations and copy number gains in human lung cancers, Cancer Res., 2008, 68, 6913-6921.
[24] Rekhtman N., Paik P.K., Arcila M.E., Tafe L.J., Oxnard G.R., Moreira A.L., et al, Clarifying the spectrum of driver oncogene mutations in biomarker-verified squamous carcinoma of lung: Lack of EGFR/KRAS and presence of PIK3CA/AKT1 mutations, Clin. Cancer Res., 2012, 18, 1167-1176.
[25] Engelman J.A., Chen L., Tan X., Crosby K., Guimaraes A.R., Upadhyay R., et al, Effective use of PI3K and MEK inhibitors to treat mutant Kras G12D and PIK3CA H1047R murine lung cancers, Nat. Med., 2008, 14,1351-1356.
[26] Dutt A., Ramos A.H., Hammerman P.S., Mermel C., Cho J., Sharifnia T., et al, Inhibitor-sensitive FGFR1 amplification in human non-small cell lung cancer, PLoS ONE, 2011, 6, e20351.
[27] Jonker D.J., Rosen L.S., Sawyer M.B., de Braud F., Wilding G., Sweeney C.J., et al, A phase I study to determine the safety, pharmacokinetics and pharmacodynamics of a dual VEGFR and FGFR inhibitor, brivanib, in patients with advanced or metastatic solid tumors, Ann. Oncol., 2011, 22, 1413-1419.
[28] Ratain M.J., Schwartz G.K., Oza A.M., Rudin C.M., Kaye S.B., De Jonge M.J., et al, Brivanib (BMS-582664) in advanced solid tumors (AST): Results of a phase II randomized discontinuation trial (RDT), J. Clin. Oncol., 2011, 29, 213S.
[29] Brooks A.N., Kilgour E., Smith P.D., Fibroblast growth factor signaling: A new therapeutic opportunity in cancer, Clin. Cancer Res., 2012, 18, 1855-1862.
[30] Hammerman P.S., Sos M. L., Ramos A.H., Xu C., Duct A., Zhou W.J., et al, Mutations in theDDR2 Kinase Gene identify a Novel therapeutic target in squamous cell lung cancer, Cancer Discov., 2011, 1 (1),78-79.
[31] Day E., Waters B., Spiegel K., Alnadaf A., Manley P.W., Buchdunger E., et al, Inhibition of collagen-induced discoidin domain receptor 1 and 2 activation by imatinib, nilotinib and dasatinib, Eur. J. Pharmacol., 2008, 599, 44-53.
[32] Johnson F.M., Bekele B.N., Feng L., Wistuba I., Tang XM., Tran H.T., et al, Phase II study of dasatinib in patients with advanced non– small-cell lung cancer, J. Clin. Oncol., 2010, 28, 4609-4615.
[33] Onno Kranenburg, The KRAS oncogene: Past, present, and future. Biochimica. Biophysica. Acta., 2005, 1756, 81–82.
[34] Riely G.L., Kris M.G., Rosenbaum D., Marks J.,Li A., Chitale D.A., et al, Frequency and Distinctive Spectrum of KRAS Mutations in Never Smokers with Lung Adenocarcinoma, Clin. Cancer Res., 2008, 14 (18), 5731-5734.
[35] Linardou H., Dahabreh I.J., Kanaloupiti D., Siannis F., Bafalouks D., Kosmidis P., et al, Assessment of somatic k-RAS mutations as a mechanism associated with resistance to EGFR-targeted agents: a systematic review and meta-analysis of studies in advanced non-small-cell lung cancer and metastatic colorectal cancer, Lancet Oncol., 2008, 9, 962–72.
[36] Jänne P.A., Shaw A.T., Pereira J.R., Jeannin G., Vansteenkiste J., Barrios C., et al, Selumetinib plus docetaxel for KRAS-mutant advanced non-small-cell lung cancer: a randomised, multicentre, placebo-controlled, phase 2 study, Lancet Oncol., 2013, 14, 38–47.
[37] Cappuzzo F., Marchetti A., Skokan M., RossiE., Gajapathy S., Felicioni L., et al, Increased MET Gene Copy Number Negatively Affects Survival of Surgically Resected Non–Small-Cell Lung Cancer Patients, J. Clin .Oncol., 2009, 27, 1667-1674.
[38] Engelman J.A., Zejnullahu K., Mitsudomi T., SongY., HylandC., Park JA., et al, MET Amp Resistance in Lung Cancer by Activating ERBB3 Signaling, Science, 2007, 316, 1039-1043.
[39] Scagliotti G.V., Novello S., Schiller J.H., Hirsh V., Sequist L.V., Soria J.C., et al, Rationale and Design of MARQUEE: A Phase III Randomized, Double-Blind Study of Tivantinib Plus Erlotinib Versus Placebo Plus Erlotinib in Previously Treated Patients with Locally Advanced or Metastatic, Non-squamous, Non–Small-Cell Lung Cancer, Clin. Lung Cancer, 2012, 13 (5), 391-395.
[40] Lim E.H., Zhang S.L.,Li J.L.,Yap W.S.,Howe T.C.,Tan B.P., et al, Using Whole Genome Amplification (WGA) of Low-Volume Biopsies to Assess the Prognostic Role of EGFR, KRAS, p53, and CMET Mutations in Advanced-Stage Non-small Cell Lung Cancer (NSCLC), J. Thorac. Oncol., 2009, 4,12–21.
[41] Kawada I., Hasina R., Arif Q., Mueller J., Smithberger E., Aliya N. Husain., et al, Dramatic Antitumor Effects of the Dual MET/RON Small Molecule Inhibitor LY2801653 in Non– Small Cell Lung Cancer, Cancer Res., 2014, 74 (3), 884-895.
[42] Rikova K., Guo A., Zeng Q., Possemato A., Yu J., Haack H., et al, Global survey of phosphotyrosine signaling identifies oncogenic kinases in lung cancer, Cell, 2007, 131, 1190-1203.

[43] Birchmeier C., Sharma S., Wigler M., Expression and rearrangement of the ROS1 gene in human glioblastoma cells, Proc. Natl. Acad. Sci. USA, 1987, 84, 9270-9274.
[44] Bergethon K., Shaw A.T., Ou S.H., Katayama R., Lovly C.M., McDonald N.T., et al, ROS1 rearrangements define a unique molecular class of lung cancers, J. Clin. Oncol., 2012, 30, 863-870.
[45] Takeuchi K., Soda M., Togashi Y., Suzuki R., Sakata S., Hatano S., et al, RET, ROS1 and ALK fusions in lung cancer, Nat. Med., 2012, 18, 378-381.
[46] McDermott U., Iafrate A.J., Gray N.S., ShiodaT., ClassonM., Maheswaran S., et al, Genomic alterations of anaplastic lymphoma kinase may sensitize tumors to anaplastic lymphoma kinase inhibitors, Cancer Res., 2008, 68, 3389-3395.
[47] Shaw A.T., Camidge D.R., Engelman J.A., Tsai C.M., Mok T.S., Schuler M., et al, Clinical activity of crizotinib in advanced non-small cell lung cancer (NSCLC) harboring ROS1 gene rearrangement, J. Clin. Oncol., 2012, 30, 482S.
[48] Soda M., Choi Y.L., Enomoto M., Takada S., Yamashita Y., Ishikawa S., et al, Identification of the transforming EML4–ALK fusion gene in non-small-cell lung, 2007, 448, 561-566.
[49] Robert C.D., Pilling A.B., Aisner D.L., KutateladzeT.G., Le A.T., Weickhardt A.J., et al, Mechanisms of Resistance to Crizotinib in Patients with ALK Gene Rearranged Non-Small Cell Lung Cancer, Clin. Cancer Res., 2012, 18 (5), 1472–1482.
[50] Shaw A.T., Varghese A.M., Solomon B.J., Costa D.B., Novello S., Mino-Kenudson M., et al, Pemetrexed-based chemotherapy in patients with advanced, ALK-positive non-small cell lung cancer, Ann. Oncol., 2013, 24, 59–66.
[51] Landi L., Cappuzzo F., Ceritinib for the treatment of non-small lung cancer, Drugs Today (Barc), 2014, 50, 465-473.
[52] Samuel A., Wells J.R., Santoro M., Targeting the RET Pathway in Thyroid Cancer, Clin. Cancer Res., 2009, 15 (23), 7119–7123.
[53] Ju Y.S., Lee W.C., Shin J.Y., Lee S., Bleazard T., Won JK., et al, Fusion of KIF5B and RET transforming gene in lung adenocarcinoma revealed from whole-genome and transcriptome sequencing, Genome Res., 2012, 22, 436-445.
[54] Kohno T., Ichikawa H., Totoki Y., Yasuda K., Hiramoto M., Nammo K., et al, KIF5BRET fusions in lung adenocarcinoma, Nat. Med., 2012, 18, 375-377.
[55] Lipson D., Capelletti M., Yelensky R., Otto G., Parker K., Jarosz M., et al, Identification of new ALK and RET gene fusions from colorectal and lung cancer biopsies, Nat. Med., 2012, 18, 382-384.
[56] Suehara Y., Arcila M., Wang L., Hasanovic A., Ang D., Ito T., et al, Identification of KIF5B-RET and GOPC-ROS1 Fusions in Lung Adenocarcinomas through a Comprehensive mRNA-Based Screen for Tyrosine Kinase Fusions, Clin. Cancer Res., 2012, 18 (24), 6599–6608.
[57] Drilon A., Wang L., Hasanovic A., Suehara Y., Lipson D., Stephens P., et al, Response to Cabozantinib in Patients with RET Fusion-Positive Lung Adenocarcinomas, Cancer Discov., 2013, 3(6), 630–635.