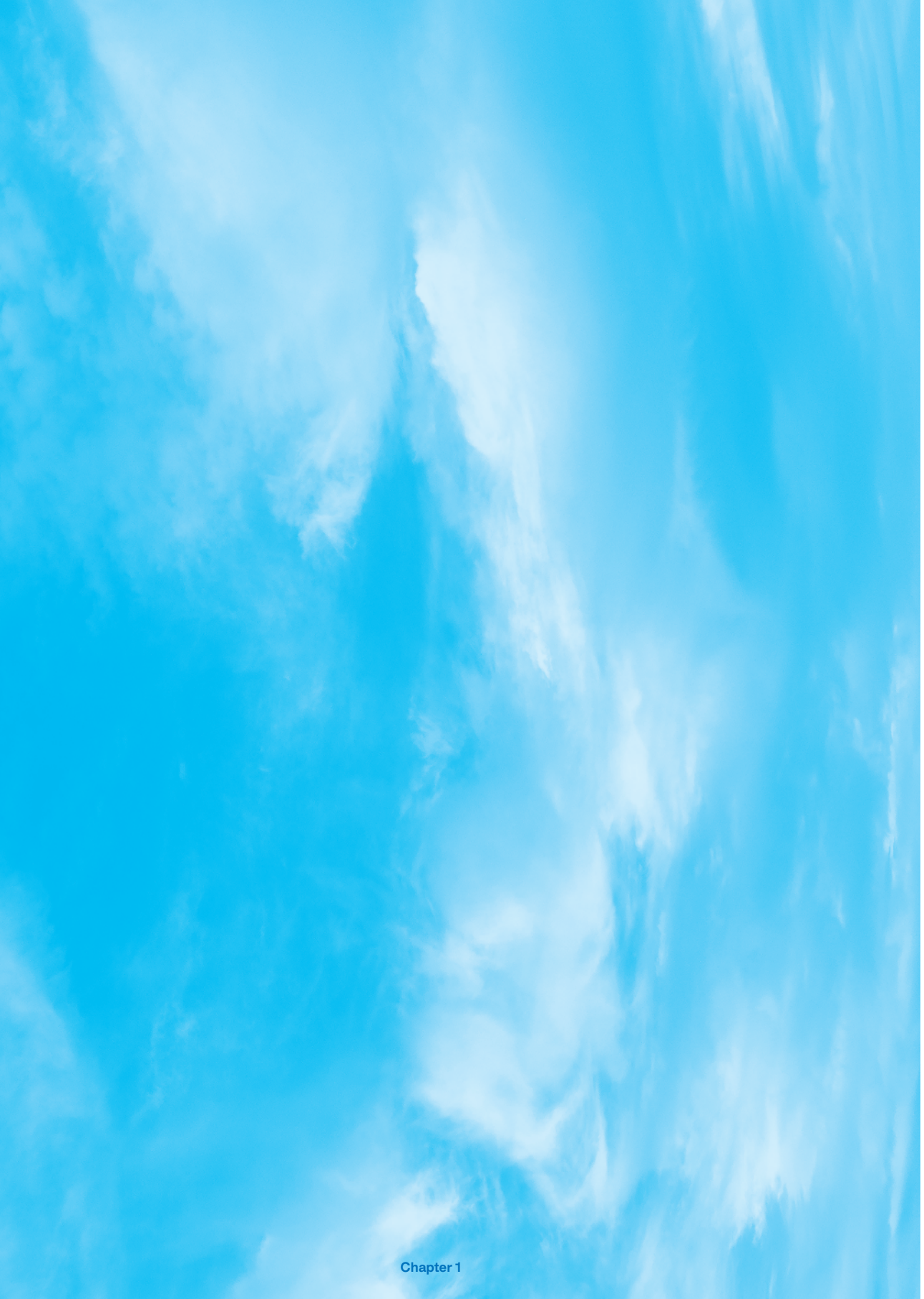
Primary and secondary resistance mechanisms in EGFR mutated non-small cell lung cancer

Veggel, B.A.M.H. van



Chapter 1

General introduction and outline of this thesis

General aspects of lung cancer

In 2020, lung cancer remained the leading cause of cancer death worldwide with an estimated 1.80 million deaths (1). Also in the Netherlands, lung cancer has a high mortality rate; although survival rates are still improving, only 21% of the patients are alive 5 years after diagnosis (based on the period 2011 – 2018) (2). In 2019, more than 14.000 people were diagnosed with lung cancer in the Netherlands. Approximately 50% of them has stage IV disease at diagnosis, this means the cancer has spread to distant organs and is in general incurable (2). Lung cancer is mostly a disease of the elderly, around 70% of patients with stage IV disease is 65 years or older (2).

Lung cancer can be divided into two subgroups, non-small cell lung cancer (NSCLC) and small cell lung cancer. Eighty five % of lung cancer patients are classified as NSCLC, a collective name for different subtypes: including adenocarcinoma, squamous cell carcinoma and other less common subtypes. Most lung carcinomas are adenocarcinomas.

Tobacco smoking remains the leading cause of lung cancer (3). The number of lung cancer patients who have never smoked varies widely, from more than 50% in women in Southeast Asia, to approximately 2-6% in men in Western countries (4). Lung cancers that occur in never smokers, defined as less than 100 cigarettes over lifetime, are considered a distinct entity (4, 5). Causative factors in this population are still poorly understood. The demographics of non-smokers with lung cancer are different compared with smokers, with more women and individuals of younger age. The vast majority are adenocarcinomas. In addition, never-smoker lung cancers show a high prevalence of targetable oncogenic driver alterations in nearly 80% of patients of whom approximately 50% harbors an epidermal growth factor receptor (*EGFR*) mutation (6, 7).

The majority of NSCLC patients are diagnosed with advanced disease on first presentation and are unsuitable for curative treatment. Until 2004, the standard treatment for advanced stage NSCLC was platinum based chemotherapy, with modest effects on life expectancy (8).

Over the past two decades, the therapeutic landscape of lung cancer has changed tremendously. The discovery of small-molecule receptor tyrosine kinase inhibitors (TKIs) was regarded as a landmark finding and has brought impressive clinical activity in specific subgroups with oncogenic drivers activated by mutation, translocation or fusion. Molecular analyses to detect oncogenic drivers have become standard in patients with advanced stage disease. The Dutch guideline on NSCLC recommends to test every patient with stage IV adenocarcinoma or never-smokers, regardless their histology, for the following driver alterations: *KRAS*, *EGFR, BRAF, HER2, ALK, MET, ROS1, RET, NTRK1-3* and *NRG1* (9). In the Netherlands, 50% of patients harbors a molecular alteration, mostly KRAS (37.9%), followed by *EGFR* (10.5%) and *BRAF* (4.1%) (10). Unfortunately, only a minority of lung cancer patients harbor a targetable genetic alteration; therefore, most NSCLC patients are not eligible for targeted treatment. This thesis focusses on patients with metastatic NSCLC harboring an *EGFR* mutation.

EGFR mutations

The EGF receptor is a member of the ErbB family, which consists of four related receptor tyrosine kinases: EGFR or HER1 (ErbB-1), HER2neu (ErbB-2), HER3 (ERbB-3) and HER4 (ErbB-4). It is a transmembrane protein receptor activated by binding its specific ligands, including EGF and transforming growth factor-α (11). Binding of EGFR to its ligands results in receptor dimerization followed by autophosphorylation, with leads to downstream activation of the Ras-Raf mitogen-activated protein kinase and the phosphatidyl inositol 3' kinase and Akt pathways. These pathways regulate cell proliferation, angiogenesis and inhibition of apoptosis (12). The physiological activity of the EGF receptor is tightly regulated, otherwise it can lead to uncontrolled cell growth and cancer formation (13).

The epidermal growth factor receptor is frequently overexpressed in common solid tumors and in 50% of NSCLCs (13). This observation made EGFR an interesting target for the development of targeted agents. In May 2003, the EGFR inhibitor gefitinib was approved as third-line therapy for non-small cell lung cancer (14, 15). In general, response rates were low in approximately 10% of patients. However, a small subset of patients showed dramatic, long-lasting responses, independent of EGFR overexpression levels (16, 17). Thereafter, Lynch et al. sequenced the entire coding region of EGFR in tumour samples from patients with a response and found different somatic gain-of-function mutations within the tyrosine kinase domain of EGFR (18). This was the starting point of the discovery of targeted therapy in lung cancer.

The *EGFR* gene is located on chromosome 7 short arm q22 and consists of four domains: an extracellular domain (EGF binding), transmembrane domain and two intracellular domains, the tyrosine kinase and autophosphorylation regions (19). The tyrosine kinase domain can be divided into an N-lobe and a C-lobe, with an ATP-binding site located between the two lobes (19). *EGFR* mutations in NSCLC are almost exclusively clustered within the tyrosine kinase domain of EGFR, and more specifically between exon 18 and 21, around the ATP-binding pocket.

EGFR in-frame deletions in exon 19 are the most prevalent *EGFR* mutations, accounting for 45% of all *EGFR* mutations, followed by *EGFR* L858R missense mutation in exon 21, which is found in 40% of *EGFR* kinase domain mutations. Exon 20 insertions (ex20ins) comprise 4-12% and are the third most common category of *EGFR* mutations found in NSCLC. The remaining *EGFR* mutations consist of a small group of nucleotide substitutions in exon 18 and 20 (13, 20, 21).

Classic *EGFR* mutations

Approximately 85% of *EGFR* mutations comprise the two classic activating *EGFR* mutations: in-frame deletions in exon 19 and L858R substitution mutations in exon 21. Activation of wild-type EGFR is primarily driven by ligand-binding-induced dimerization. Classic *EGFR* mutations hyperactivate the kinase activity of EGFR by stabilizing ligand-independent dimerization with ERBB family receptors (19). Of all *EGFR* mutations, *EGFR* exon 19 deletions exhibit the highest catalytic activity, followed by L858R mutations (22-24). These mutations increase the relative affinity for EGFR TKIs over ATP compared with wild-type EGFR.

Treatment

Two classes of EGFR directed therapies have been developed since 1984. First anti-EGFR monoclonal antibodies (mAb), and in 2003 early generation reversible EGFR TKIs were introduced (25, 26).

The role of anti-EGFR mAb's in the treatment of *EGFR* -mutated NSCLC is limited as they are ineffective as a single agent (27). Anti-EGFR mAb's bind to the extracellular domain of EGFR and block ligand-induced EGFR tyrosine kinase activation by occluding the ligand-binding region (25, 26). Cetuximab was the first anti-EGFR human-to-murine chimeric mAb tested in patients with EGFR overexpression determined by immunohistochemical analysis. Treatment with cetuximab alone was well tolerated. The most frequent adverse events were fever and chills, asthenia, transaminase elevation, nausea, skin toxicities (flushing, dermatitis and acneiform rashes) and allergic reactions (28).

Small-molecule EGFR TKIs like gefitinib and erlotinib, compete reversibly with ATP to bind to the receptor's kinase pocket, which inhibit EGFR autophosphorylation and prevents downstream signaling (26, 29). Gefitinib and erlotinib, two first-generation EGFR TKIs, have shown an improvement of objective response rate (ORR) and progression free survival (PFS) compared to platinum-based chemotherapy and became the standard of care as first-line treatment for advanced *EGFR* mutated NSCLC (30-32). Dose-dependent and reversible diarrhea and acneiform rashes are the most frequent side effects. Thereafter, irreversible second-generation EGFR TKIs (afatinib and dacomitinib) did show a slight clinical improvement compared to first-generation TKIs, however they were associated with higher toxicity rates (33-35). Until the discovery of third generation EGFR TKIs, first- or second generation EGFR TKIs were both used without a clear preference.

Despite significant clinical and durable benefit after treatment with early-generation EGFR TKI's, patients inevitably develop disease progression, mostly driven by acquisition of an exon 20 T790M resistance mutation. This mutation, also referred to as 'gatekeeper' mutation, sterically hinders the binding of early-generation EGFR TKIs to the ATP-binding site of EFGR. Osimertinib, a third generation EGFR TKI, was developed to selectively inhibit both EGFR-TKI-sensitizing and *EGFR* exon 20 T790M resistance mutations while sparing wild-type EGFR (36). As first-line treatment, it also demonstrated a significantly PFS and overall survival (OS) benefit relative to first-generation EGFR TKIs. In addition, osimertinib showed superior activity against brain metastases and has a favorable toxicity profile (37-40). With a median overall survival of more than three years, first-line osimertinib is the preferred option for *EGFR*-positive NSCLC patients at this time (41).

Immune checkpoint inhibitors has revolutionized the treatment of NSCLC, however the efficacy of immunotherapy in *EGFR* -mutated NSCLC appears to be quite limited (42). In addition, the use of immune checkpoint inhibitors prior to or concurrent with osimertinib increases the risk of pneumonitis (43). This underlines the importance of molecular testing at diagnosis to enable personalized treatment.

EGFR exon 20 insertion mutations

EGFR ex20ins mutations comprise approximately 1 to 2% of all NSCLC cases and 4 – 12% of all *EGFR* mutated lung cancers, representing the third most common type of *EGFR* mutations (20, 21, 44, 45).

Similar to the classic *EGFR* mutations, *EGFR* ex20ins are more common among never-smokers and females. The increased incidence among Asian patients is less pronounced. In addition, the ex20ins mutation was more common in older patients compared to *EGFR* exon 19 deletions and L858R mutations (20, 46). The prognosis of patients harboring an *EGFR* ex20ins mutation is worse, compared to patients with common or other uncommon *EGFR* mutations. The median OS ranged from 4.8 – 20 months, similar or slightly better than the *EGFR* wild-type population (46-48).

EGFR ex20ins mutations are a heterogeneous group of activating mutations within the tyrosine kinase domain of EGFR. They represent a combination of in-frame insertions and/or duplications, ranging from three to 21 base pairs, clustered between amino acid positions 762 and 774 of exon 20 (20). This region of exon 20 contains two important regions: the regulatory C-helix (762 – 766) and the adjacent loop that follows it (767-774) (20, 21, 45, 49). To date, more than 60 unique variants are identified, mostly located in the loop following the C-helix (45).

PD-L1 status is negative in approximately 75% of patients with an *EGFR* ex20ins mutation and median tumor mutational burden is low (48, 50). Although little data is available, they seem to derive less benefit from immunotherapy (50).

Treatment

EGFR ex20ins are generally associated with de-novo resistance to early generation EGFR TKIs, except for variant A763_Y764insFQEA (20, 46, 51, 52). Classic *EGFR* mutations directly affect the structure of the ATP-binding pocket and increase the relative affinity for EGFR TKIs over ATP compared with wild-type EGFR. This leads to a large therapeutic window for EGFR TKIs. Targeting *EGFR* ex20ins is challenging because they activate EGFR without diminishing ATP affinity, which makes it difficult to selectively target the ex20ins mutant over wild-type EGFR. In addition, *EGFR* ex20ins mutations induce steric hindrance of the drug-binding pocket by pushing the C-helix into its inward (active state) position, which prevents binding of EGFR TKIs (53). Insertions before residue 764, like the A763_Y764insFQEA variant, do not reveal this stabilized active formation and do not induce drug resistance (21). The role of osimertinib had not yet been established. 3D modeling of the crystal structures of *EGFR* ex20ins showed changes within the drug-binding pocket, similar to T790M (53). Given these similarities, osimertinib could be a treatment option. However, in contrast with T790M mutation, *EGFR* ex20ins mutations are characterized by changes of the a-C helix in an inward, activated position. In addition, 3D modeling showed a shift of the P-loop into the drug-binding pocket, resulting in steric hindrance of the drug-binding pocket from different directions (53). Osimertinib has a large inflexible terminal 1-methylindole group that reduces the binding ability to *EGFR* ex20ins. So based on preclinical data, the role of osimertinib for this patient category was unclear. Overall, platinum-based chemotherapy is to date the most efficacious first-line treatment for patients with an *EGFR* ex20ins mutation (48, 54).

Outline of this thesis

The primary goals of this thesis were to gain insights in primary and secondary resistance mechanisms in patients with *EGFR* mutated NSCLC and to develop new treatment strategies.

The first part of this thesis focuses on a specific subgroup of *EGFR* mutated lung cancer patients, namely patients harboring an *EGFR* ex20ins mutation. We retrospectively and prospectively analyzed the activity of two different treatment regimens for this specific mutation subtype.

The second part focusses on genomic profiling of resistance mechanisms after treatment with a third generation EGFR TKI. In addition, we investigated the activity of combination treatment after cMET driven resistance.

PART I. Targeted treatment of *EGFR* exon 20 insertion mutations

This part of the thesis focuses on finding new targeted treatment options for patients with advanced stage NSCLC harboring an *EGFR* ex20ins mutation. Although the clinical activity of osimertinib for classical *EGFR* mutations is impressive, data about effectiveness of third generation EGFR TKIs for patients harboring an *EGFR* ex20ins mutation were conflicting in pre-clinical studies. Limited clinical data were available. Firstly, we retrospectively collected clinical and molecular data of 21 patients harboring an *EGFR* ex20ins mutation who were treated with osimertinib to gain insight into effectiveness. In **chapter 2**, we described the results of this case series. Almost all patients were treated with osimertinib 80 mg once daily with limited efficacy. Preclinical data showed that the IC50 values of osimertinib for *EGFR* ex20ins mutations were 10 to 100 fold higher compared to classical *EGFR* mutations. We hypothesized that a higher dose of osimertinib might be necessary to effectively treat those patients. In **chapter 3**, we present the results of the POSITION-20 trial, a prospective trial in which 25 patients harboring an *EGFR* exon 20 mutation were treated with high-dose osimertinib in first or second line.

A different strategy to target *EGFR* ex20ins positive NSCLC could be to intensify EGFR blockade by concurrent TKI and EGFR monoclonal antibody treatment. Afatinib is an interesting drug to combine with other EGFR directed therapies like anti-EGFR mAbs, given its broad and irreversible inhibitory profile and low potential for drug interactions (55). First-line afatinib plus cetuximab already showed efficacy in heavily pretreated *EGFR* -mutant NSCLC patients with acquired resistance to first generation EGFR TKIs (56). Dual EGFR blockade with afatinib and cetuximab may also induce tumor responses in *EGFR* ex20ins mutations, based on the treatment outcomes of four *EGFR* exon 20 insertion positive NSCLC patients treated with this combination, presented in **chapter 4**. We tried to validate these results in a dedicated clinical trial. In **chapter 5**, we present the results of a single-arm, phase II trial, where 37 advanced *EGFR* ex20ins positive treatment-naïve or pretreated NSCLC patients were treated with afatinib in combination with cetuximab.

In recent years, several TKIs directed against *EGFR* ex20ins mutations have been developed. However, *EGFR* ex20ins mutated NSCLC cannot be seen as a single disease entity, due to marked heterogeneity in this subgroup. There is a need to expand the knowledge on the structure-function relationship of different *EGFR* ex20ins mutants on drug sensitivity. **Chapter 6** provides a comprehensive assessment of the available literature of pre-clinical and clinical drug sensitivity to currently available TKIs across different *EGFR* ex20ins mutations.

PART II. Analysis an atment of resistance mechanisms after third generation EGFR TKI treatment.

Despite robust activity of osimertinib in first- and second-line settings, eventually acquired resistance occurs. In **chapter 7**, we retrospectively analyzed pre- and post-treatment biopsies in patients who received a third-generation EGFR TKI in second line to gain more insights in innate and acquired resistance mechanisms. Bypass-track mechanisms after third-generation EGFR TKI treatment are most commonly MET amplifications. Little was known about the efficacy of cMET inhibitors in the acquired resistance setting. Finally, in **chapter 8** we retrospectively analyzed the treatment results of patients with acquired cMET amplification after EGFR-directed therapy, who are treated with crizotinib.

References

1. https://www.who.int/news-room/fact-sheets/detail/cancer
2. https://iknl.nl/kankersoorten/longkanker/registratie/overleving
3. Alberg AJ, Brock MV, Ford JG, Samet JM, Spivack SD. Epidemiology of lung cancer: Diagnosis and management of lung cancer, 3rd ed: American College of Chest Physicians evidence-based clinical practice guidelines. Chest. 2013;143(5 Suppl):e1S-e29S.
4. Couraud S, Zalcman G, Milleron B, Morin F, Souquet PJ. Lung cancer in never smokers--a review. Eur J Cancer. 2012;48(9):1299-311.
5. Subramanian J, Govindan R. Lung cancer in never smokers: a review. J Clin Oncol. 2007;25(5):561-70.
6. Devarakonda S, Li Y, Martins Rodrigues F, Sankararaman S, Kadara H, Goparaju C, et al. Genomic Profiling of Lung Adenocarcinoma in Never-Smokers. J Clin Oncol. 2021;39(33):3747-58.
7. Couraud S, Souquet PJ, Paris C, Dô P, Doubre H, Pichon E, et al. BioCAST/IFCT-1002: epidemiological and molecular features of lung cancer in never-smokers. Eur Respir J. 2015;45(5):1403-14.
8. Chemotherapy in addition to supportive care improves survival in advanced non-small-cell lung cancer: a systematic review and meta-analysis of individual patient data from 16 randomized controlled trials. J Clin Oncol. 2008;26(28):4617-25.
9. Specialists DFoM. Non-Small Cell Lung Cancer - General. https://richtlijnendatabase.nl/richtlijn/niet_kleincellig_longcarcinoom/algemeen.html
10. https://iknl.nl/kankersoorten/longkanker/registratie/incidentie
11. Yarden Y, Sliwkowski MX. Untangling the ErbB signalling network. Nat Rev Mol Cell Biol. 2001;2(2):127-37.
12. Herbst RS. Review of epidermal growth factor receptor biology. Int J Radiat Oncol Biol Phys. 2004;59(2 Suppl):21-6.
13. Sharma SV, Bell DW, Settleman J, Haber DA. Epidermal growth factor receptor mutations in lung cancer. Nat Rev Cancer. 2007;7(3):169-81.
14. Fukuoka M, Yano S, Giaccone G, Tamura T, Nakagawa K, Douillard JY, et al. Multi-institutional randomized phase II trial of gefitinib for previously treated patients with advanced non-small-cell lung cancer (The IDEAL 1 Trial) [corrected]. J Clin Oncol. 2003;21(12):2237-46.
15. Kris MG, Natale RB, Herbst RS, Lynch TJ, Jr., Prager D, Belani CP, et al. Efficacy of gefitinib, an inhibitor of the epidermal growth factor receptor tyrosine kinase, in symptomatic patients with non-small cell lung cancer: a randomized trial. Jama. 2003;290(16):2149-58.
16. Giaccone G, Herbst RS, Manegold C, Scagliotti G, Rosell R, Miller V, et al. Gefitinib in combination with gemcitabine and cisplatin in advanced non-small-cell lung cancer: a phase III trial--INTACT 1. J Clin Oncol. 2004;22(5):777-84.
17. Herbst RS, Giaccone G, Schiller JH, Natale RB, Miller V, Manegold C, et al. Gefitinib in combination with paclitaxel and carboplatin in advanced non-small-cell lung cancer: a phase III trial--INTACT 2. J Clin Oncol. 2004;22(5):785-94.
18. Lynch TJ, Bell DW, Sordella R, Gurubhagavatula S, Okimoto RA, Brannigan BW, et al. Activating mutations in the epidermal growth factor receptor underlying responsiveness of non-small-cell lung cancer to gefitinib. N Engl J Med. 2004;350(21):2129-39.
19. Davies RL, Grosse VA, Kucherlapati R, Bothwell M. Genetic analysis of epidermal growth factor action: assignment of human epidermal growth factor receptor gene to chromosome 7. Proc Natl Acad Sci U S A. 1980;77(7):4188-92.
20. Arcila ME, Nafa K, Chaft JE, Rekhtman N, Lau C, Reva BA, et al. EGFR exon 20 insertion mutations in lung adenocarcinomas: prevalence, molecular heterogeneity, and clinicopathologic characteristics. Mol Cancer Ther. 2013;12(2):220-9.
21. Yasuda H, Park E, Yun CH, Sng NJ, Lucena-Araujo AR, Yeo WL, et al. Structural, biochemical, and clinical characterization of epidermal growth factor receptor (EGFR) exon 20 insertion mutations in lung cancer. Sci Transl Med. 2013;5(216):216ra177.
22. Galdadas I, Carlino L, Ward RA, Hughes SJ, Haider S, Gervasio FL. Structural basis of the effect of activating mutations on the EGF receptor. Elife. 2021;10.
23. Gilmer TM, Cable L, Alligood K, Rusnak D, Spehar G, Gallagher KT, et al. Impact of common epidermal growth factor receptor and HER2 variants on receptor activity and inhibition by lapatinib. Cancer Res. 2008;68(2):571-9.
24. Yun CH, Boggon TJ, Li Y, Woo MS, Greulich H, Meyerson M, et al. Structures of lung cancer-derived EGFR mutants and inhibitor complexes: mechanism of activation and insights into differential inhibitor sensitivity. Cancer Cell. 2007;11(3):217-27.
25. Masui H, Kawamoto T, Sato JD, Wolf B, Sato G, Mendelsohn J. Growth inhibition of human tumor cells in athymic mice by anti-epidermal growth factor receptor monoclonal antibodies. Cancer Res. 1984;44(3):1002-7.
26. Ciardiello F, Tortora G. EGFR antagonists in cancer treatment. N Engl J Med. 2008;358(11):1160-74.
27. Rossi A, Maione P, Gridelli C. Cetuximab in advanced non-small cell lung cancer. Crit Rev Oncol Hematol. 2006;59(2):139-49.
28. Baselga J, Pfister D, Cooper MR, Cohen R, Burtness B, Bos M, et al. Phase I studies of anti-epidermal growth factor receptor chimeric antibody C225 alone and in combination with cisplatin. J Clin Oncol. 2000;18(4):904-14.
29. Hynes NE, Lane HA. ERBB receptors and cancer: the complexity of targeted inhibitors. Nat Rev Cancer. 2005;5(5):341-54.
30. Maemondo M, Inoue A, Kobayashi K, Sugawara S, Oizumi S, Isobe H, et al. Gefitinib or chemotherapy for non-small-cell lung cancer with mutated EGFR. N Engl J Med. 2010;362(25):2380-8.
31. Mok TS, Wu YL, Thongprasert S, Yang CH, Chu DT, Saijo N, et al. Gefitinib or carboplatin-paclitaxel in pulmonary adenocarcinoma. N Engl J Med. 2009;361(10):947-57.
32. Rosell R, Carcereny E, Gervais R, 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 multicentre, open-label, randomised phase 3 trial. Lancet Oncol. 2012;13(3):239-46.
33. Park K, Tan EH, O'Byrne K, Zhang L, Boyer M, Mok T, et al. Afatinib versus gefitinib as first-line treatment of patients with EGFR mutation-positive non-small-cell lung

cancer (LUX-Lung 7): a phase 2B, open-label, randomised controlled trial. Lancet Oncol. 2016;17(5):577-89.
34. Sequist LV, Yang JC, Yamamoto N, O'Byrne K, Hirsh V, Mok T, et al. Phase III study of afatinib or cisplatin plus pemetrexed in patients with metastatic lung adenocarcinoma with EGFR mutations. J Clin Oncol. 2013;31(27):3327-34.
35. Wu YL, Zhou C, Hu CP, Feng J, Lu S, Huang Y, et al. Afatinib versus cisplatin plus gemcitabine for first-line treatment of Asian patients with advanced non-small-cell lung cancer harbouring EGFR mutations (LUX-Lung 6): an open-label, randomised phase 3 trial. Lancet Oncol. 2014;15(2):213-22.
36. Cross DA, Ashton SE, Ghiorghiu S, Eberlein C, Nebhan CA, Spitzler PJ, et al. AZD9291, an irreversible EGFR TKI, overcomes T790M-mediated resistance to EGFR inhibitors in lung cancer. Cancer Discov. 2014;4(9):1046-61.
37. Mok TS, Wu YL, Ahn MJ, Garassino MC, Kim HR, Ramalingam SS, et al. Osimertinib or Platinum-Pemetrexed in EGFR T790M-Positive Lung Cancer. N Engl J Med. 2017;376(7):629-40.
38. Ramalingam SS, Vansteenkiste J, Planchard D, Cho BC, Gray JE, Ohe Y, et al. Overall Survival with Osimertinib in Untreated, EGFR-Mutated Advanced NSCLC. N Engl J Med. 2020;382(1):41-50.
39. Reungwetwattana T, Nakagawa K, Cho BC, Cobo M, Cho EK, Bertolini A, et al. CNS Response to Osimertinib Versus Standard Epidermal Growth Factor Receptor Tyrosine Kinase Inhibitors in Patients With Untreated EGFR-Mutated Advanced Non-Small-Cell Lung Cancer. J Clin Oncol. 2018:Jco2018783118.
40. Soria JC, Ohe Y, Vansteenkiste J, Reungwetwattana T, Chewaskulyong B, Lee KH, et al. Osimertinib in Untreated EGFR-Mutated Advanced Non-Small-Cell Lung Cancer. N Engl J Med. 2018;378(2):113-25.
41. Planchard D, Popat S, Kerr K, Novello S, Smit EF, Faivre-Finn C, et al. Metastatic non-small cell lung cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol. 2018;29 Suppl 4:iv192-iv237.
42. Addeo A, Passaro A, Malapelle U, Banna GL, Subbiah V, Friedlaender A. Immunotherapy in non-small cell lung cancer harbouring driver mutations. Cancer Treatment Reviews. 2021;96:102179.
43. Schoenfeld AJ, Arbour KC, Rizvi H, Iqbal AN, Gadgeel SM, Girshman J, et al. Severe immune-related adverse events are common with sequential PD-(L)1 blockade and osimertinib. Ann Oncol. 2019;30(5):839-44.
44. Beau-Faller M, Prim N, Ruppert AM, Nanni-Metéllus I, Lacave R, Lacroix L, et al. Rare EGFR exon 18 and exon 20 mutations in non-small-cell lung cancer on 10 117 patients: a multicentre observational study by the French ERMETIC-IFCT network. Ann Oncol. 2014;25(1):126-31.
45. Riess JW, Gandara DR, Frampton GM, Madison R, Peled N, Bufill JA, et al. Diverse EGFR Exon 20 Insertions and Co-Occurring Molecular Alterations Identified by Comprehensive Genomic Profiling of NSCLC. J Thorac Oncol. 2018;13(10):1560-8.
46. Burnett H, Emich H, Carroll C, Stapleton N, Mahadevia P, Li T. Epidemiological and clinical burden of EGFR Exon 20 insertion in advanced non-small cell lung cancer: A systematic literature review. PLoS One. 2021;16(3):e0247620.
47. Lo P, Jackman D, Butaney M, Lindeman N, Johnson B, Jaenne P, et al. Clinical Behavior Of Lung Cancers Harboring EGFR Exon 20 Insertions. Journal of Thoracic Oncology. 2012;7:S206-S7.
48. Choudhury NJ, Schoenfeld AJ, Flynn J, Falcon CJ, Rizvi H, Rudin CM, et al. Response to Standard Therapies and Comprehensive Genomic Analysis for Patients with Lung Adenocarcinoma with EGFR Exon 20 Insertions. Clin Cancer Res. 2021;27(10):2920-7.
49. Meador CB, Sequist LV, Piotrowska Z. Targeting EGFR Exon 20 Insertions in Non-Small Cell Lung Cancer: Recent Advances and Clinical Updates. Cancer Discov. 2021;11(9):2145-57.
50. Geng D, Guo Q, Huang S, Zhang H, Guo S, Li X. Clinical and molecular characteristics of epidermal growth factor receptor exon 20 insertion mutations in non-small-cell lung cancer. Clin Transl Oncol. 2022;24(2):379-87.
51. Naidoo J, Sima CS, Rodriguez K, Busby N, Nafa K, Ladanyi M, et al. Epidermal growth factor receptor exon 20 insertions in advanced lung adenocarcinomas: Clinical outcomes and response to erlotinib. Cancer. 2015;121(18):3212-20.
52. Yang JC, Sequist LV, Geater SL, Tsai CM, Mok TS, Schuler M, et al. Clinical activity of afatinib in patients with advanced non-small-cell lung cancer harbouring uncommon EGFR mutations: a combined post-hoc analysis of LUX-Lung 2, LUX-Lung 3, and LUX-Lung 6. Lancet Oncol. 2015;16(7):830-8.
53. Robichaux JP, Elamin YY, Tan Z, Carter BW, Zhang S, Liu S, et al. Mechanisms and clinical activity of an EGFR and HER2 exon 20-selective kinase inhibitor in non-small cell lung cancer. Nat Med. 2018;24(5):638-46.
54. Yang G, Li J, Xu H, Yang Y, Yang L, Xu F, et al. EGFR exon 20 insertion mutations in Chinese advanced non-small cell lung cancer patients: Molecular heterogeneity and treatment outcome from nationwide real-world study. Lung Cancer. 2020;145:186-94.
55. Solca F, Dahl G, Zoephel A, Bader G, Sanderson M, Klein C, et al. Target binding properties and cellular activity of afatinib (BIBW 2992), an irreversible ErbB family blocker. J Pharmacol Exp Ther. 2012;343(2):342-50.
56. Janjigian YY, Smit EF, Groen HJ, Horn L, Gettinger S, Camidge DR, et al. Dual inhibition of EGFR with afatinib and cetuximab in kinase inhibitor-resistant EGFR-mutant lung cancer with and without T790M mutations. Cancer Discov. 2014;4(9):1036-45.