Osimertinib as a third-generation EGFR TKI in non-small cell lung cancer: selective targeting of T790M mutation and clinical advantages

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Abstract:

Non-small cell lung cancer is the most common type of lung cancer, accounting for around 85% of all cases (Testa et al.). Osimertinib is a third-generation epidermal growth factor receptor (EGFR) tyrosine kinase inhibitor (TKI) used to treat non-small cell lung cancer (NSCLC) that harbors EGFR mutations, such as the T790M mutation. T790M mutation, which is the substitution of threonine with methionine at amino acid position 790 in the EGFR protein due to a mutation in exon 20, is the most common mechanism that resists first- or second-generation TKI drugs. Approximately 50%-60% of patients receiving early-generation TKIs develop resistance (Fu et al.). Preclinical studies have shown that Osimertinib may be a favorable treatment option for patients who have developed resistance to first- or second-generation TKI drugs. This paper reviews the action and clinical advantages of third-generation TKI Osimertinib.

Keywords: Epidermal Growth Factor Receptor (EGFR), Tyrosine Kinase Inhibitor (TKI), Non-Small Cell Lung Cancer (NSCLC), T790M mutation, Osimertinib

Introduction:

The epidermal growth factor receptor (EGFR) is a transmembrane protein that is found on the surface of many cells, including those in the lungs. It plays an important role in regulating cell growth and survival through signal transduction pathways when bound to its ligand. Sometimes, the EGFR is overexpressed and needs a signal to stop the signal transduction. This is where a tyrosine kinase inhibitor (TKI) is required. A tyrosine kinase inhibitor is a type of enzyme that blocks the EGFR's active site, effectively inhibiting the EGFR's function. There are several generations of TKIs, including 1st-, 2nd-, and 3rd-generation TKIs. The reason behind multiple generations of TKIs is that if previous generations are ineffective, a different TKI is required. For example, in NSCLC, Osimertinib is a third-generation TKI because the last two generations were not effective in shrinking tumors. In the case of NSCLC, Osimertinib is used because the previous two TKIs introduced another mutation, T790M, which altered the structure of the EGFR in the ATP binding pocket, allowing ATP to continue powering the signaling cascade. Therefore, Osimertinib was developed to combat this structural change. Third-generation TKI such as Osimertinib represent a major step towards overcoming resistance in EGFR-mutant NSCLC, and this review will evaluate its therapeutic advantages as well as limitations.

Many mutations have been identified in EGFR that promote cancerous growth as the common outcome. There were an estimated 235,760 cases of NSCLC (Non-Small Cell Lung Cancer) in 2021, leading to 131,880 deaths due to mutations in EGFR. One of the most commonly studied mutations is the T790M mutation, a point mutation in the EGFR gene in NSCLC, where methionine is replaced with threonine at amino acid position 790 (Castañeda-González et al.). This specific mutation, the T790M mutation, will be reviewed in detail in this article.

The Role of Tyrosine Kinase Inhibitors (TKIs) in Cancer Treatment:

As the signaling molecule binds the EGFR extracellularly, the signal transduction pathway is activated. The site of signal transduction is in the cell cytoplasm. The enzyme tyrosine kinase plays a crucial role in initiating signal transduction. Tyrosine kinases are enzymes that add phosphate groups to the cytoplasmic domain, a binding pocket, of the EGFR, thereby activating the signal transduction pathway and promoting cell division. If EGFR is mutated, the risk of uncontrolled cell division can be eliminated by inhibiting the action of Tyrosine kinases. Tyrosine kinase inhibitors (TKIs) are the drugs designed to be used commercially to inhibit the action of tyrosine kinase in the case of mutated EGFR. Tyrosine kinases usually promote cell signaling and growth. On the other hand, Tyrosine Kinase Inhibitors (TKIs) specifically attach to the binding pocket of the mutated EGFR, which couldn't bind to its original ligand because of the structural change in the EGFR. The binding of the TKI inhibits the cell from growing and dividing further. This mechanism is highly effective in preventing the progression of uncontrolled cell growth, which can lead to tumor formation.

First, Second, and Third Generation TKIs

The effectiveness of this otherwise effective drug is questioned in advanced forms of cancer, such as NSCLC. First, second, and third-generation TKIs represent different stages of targeted cancer therapies. Second- and third-generation TKIs are developed if the first-generation TKI does not effectively halt the signaling cascade. Similarly, EGFRs in NSCLC sometimes develop resistance to these initial TKIs, the most common one being the introduction of a point mutation T790M. The T790M mutation forms as a response to first or second-generation TKIs such as gefitinib (first generation) or afatinib (second generation), even though the TKIs initially showed "substantial clinical benefit in advanced NSCLC patients with other mutations such as Ex19del and L858R"(Koulouris et al.).

Osimertinib–Third Generation TKI

To address the acquired mutation T790M, a third-line TKI has been developed to fix it. This specific TKI is called Osimertinib or sold under the brand name Tagrisso. Osimertinib is an oral, irreversible third-generation TKI that binds to the EGFR, blocking the signal transduction from occurring. It binds covalently to the cysteine-797 (C797) residue within the EGFR's ATP-binding site, thereby inhibiting the receptor's ability to signal and promote cell growth.

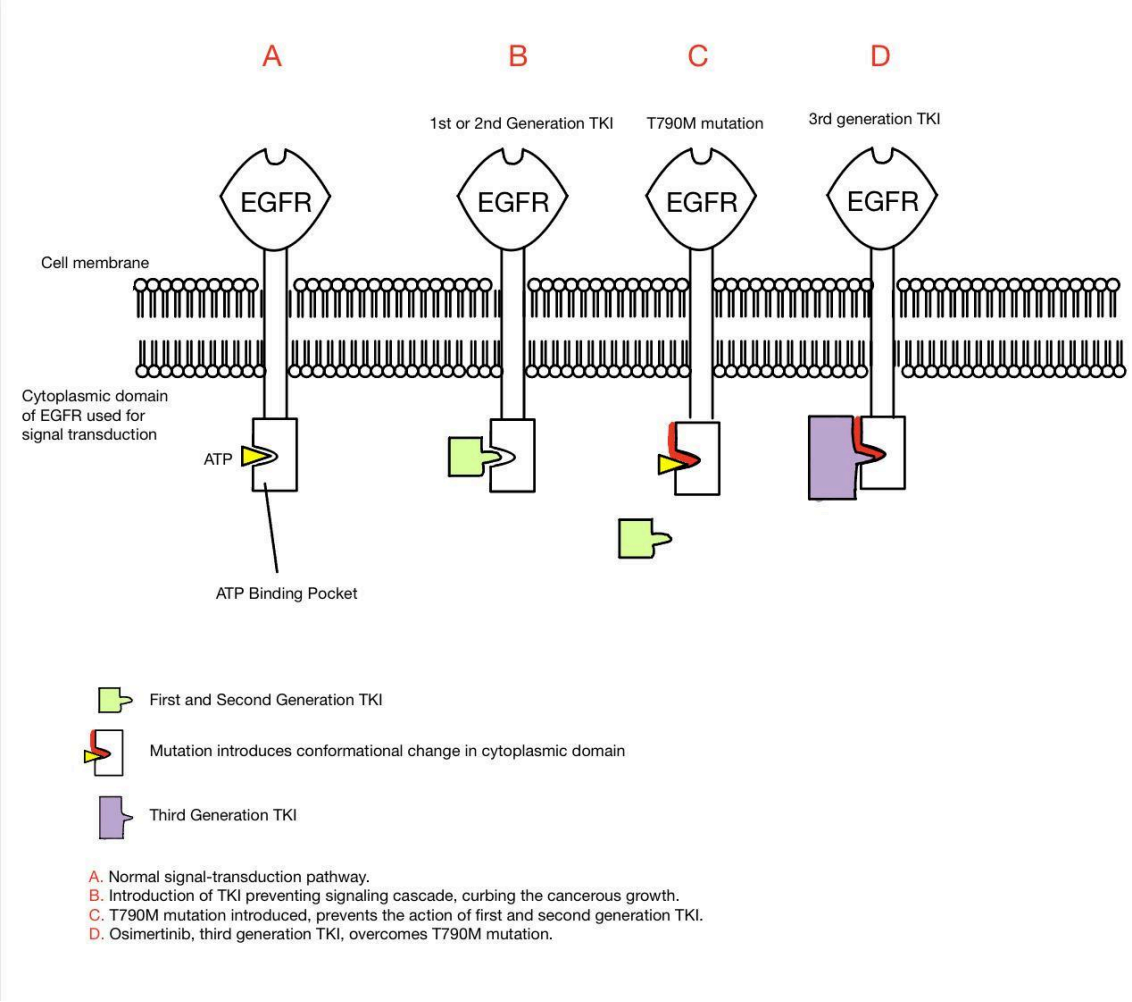

Figure 1: Action of first, second, and third-generation TKIs

Figure 1 represents how first, second, and third-generation TKIs work. Additionally, the T790M mutation typically occurs after the introduction of a first- or second-generation drug (Koulouris et al.).This mutation prevents the first- or second-generation TKIs from binding to the ATP-binding pocket, thereby allowing the signaling to continue in the cell. Third-generation drugs, such as Osimertinib, can overcome this problem as it can bind to the mutated EGFR ATP binding pocket, inhibiting phosphorylation, thus regulating cell growth. As indicated in Figure 1, A represents the function of a wild-type EGFR, where ATP binding to the pocket is unregulated. B represents the system when a first or second-generation TKI is introduced, effectively blocking the ATP from powering the system. Next, as a side effect of the first or second-generation TKI, the T790M mutation is introduced, as seen in C. This mutation blocks the TKI, but allows the ATP to continue powering the signaling cascade system. Finally, Osimertinib, a third-generation drug, is introduced to bind to the mutated ATP-binding pocket, effectively blocking ATP from powering the signaling cascade; the signal for cell growth will halt at the EGFR, as seen in D.

Discussion:

Table 1: Advantages of Osimertinib

Advantages	Explanation	References
Selective for mutant EGFR	Inhibits EGFR-sensitizing mutation	(Wang et al.)
	T790M resistant mutations	(Fu et al.)
Target-specific	The wild-type EGFR function is not affected	(Fu et al.)
Penetration into the Central Nervous System	Can penetrate the blood-brain barrier	(Fu et al.)
Effective component of combination therapy	Osimertinib combined with MET inhibitor can overcome TKI resistance	(Cross et al.)
Dosage	Oral dosage of 80mg once a day	(Chmielecki et al.)

Epidermal Growth Factor Receptor (EGFR) is a protein responsible for cell growth signaling pathways. It is a transmembrane receptor on the cell waiting for the signal molecule to activate the downstream protein cascade. A mutation in the EGFR gene is a common cause of the development of many forms of cancer (Ou et al.).

One of the most common mutations in the EGFR protein is found in non-small-cell lung cancer (NSCLC), which is the most prevalent form of lung cancer. It consists of many subtypes such as adenocarcinoma, large cell carcinoma, and squamous cell carcinoma. One of the characteristics of NSCLC is that it grows and spreads slower than small-cell lung cancer. The majority of the cases are correlated to smoking, although it can also occur in non-smokers who happen to have a genetic mutation in the EGFR. Targeted therapies, such as EGFR tyrosine kinase inhibitors, have significantly helped patients who have specific mutations in the EGFR. A mutation, known as T790M, is a substitution in the EGFR gene where Threonine is replaced by Methionine. The

T790M mutation alters the structure of EGFR, especially in the ATP binding pocket, in a way that prevents Tyrosine Kinase Inhibitors (TKIs) from binding and inhibiting cell growth. One way to address this problem is by taking drugs that can either restore the effects of the mutation or make the mutation impossible to happen in the first place.

Several newer treatments are being researched that combine targeted therapy with immunotherapy in an effort to enhance patient response to treatment and bypass resistance, including the T790M mutation. One of the ways to treat resistance from EGFR mutations, like T790M, is with immunotherapy or by using targeted drugs that either block the mutation's effects or try to stop the mutation from happening in the first place. The first-generation EGFR-TKIs, including gefitinib and erlotinib, work by reversibly binding to the EGFR receptor to block its signaling in cancer cells that contain frequent activating mutations. The second-generation TKIs, including afatinib and dacomitinib, bind irreversibly and are stronger but also affect normal EGFR in normal cells, so side effects are increased. Both generations are unfortunately not good at treating tumors that develop the T790M resistance mutation. That's where newer strategies, like combining EGFR-targeted therapy with immunotherapy, come in. These newer strategies are being studied to see if they can get around resistance, including T790M, and can improve long-term results for the patient. Osimertinib is an epidermal growth factor receptor (EGFR) tyrosine kinase inhibitor (TKI) of the third generation, which is designed to be highly selective for mutant EGFR. This is a reference to the fact that it works against cancer cells with EGFR mutations without affecting the normal (wild-type) EGFR, thus reducing the side effects.

It's highly effective at targeting common mutations like exon 19 deletions and L858R, which, similar to the T790M mutation, allow the signalling cascade to continue without any restraint. Osimertertinib is not just effective in T790M, but also in other common mutations, and is therefore a suitable drug for treating non-small cell lung cancer (NSCLC) with these mutations (Wang et al.). Perhaps the most significant benefit of osimertinib is that it works against the T790M resistance mutation. T790M is a mutation that occurs typically after patients have received first- or second-generation TKI treatment, making the previous drugs ineffective. Osimertinib overcomes this restriction by specifically targeting the T790M mutation, thus it's typically given after resistance to prior EGFR-TKIs is confirmed (Fu et al.). It creates a covalent bond with C797 residue within the ATP pocket of the EGFR, preventing the signal from being transferred (Cross et al.). Osimertinib has the important additional advantage of being capable of crossing into the central nervous system (CNS). This enables it to penetrate the blood-brain barrier and prevent or treat brain metastases, which in EGFR-mutant lung cancer are common (Fu et al.). Efflux transporters become responsible for the entry of certain drugs that are considered toxic chemicals. Efflux transporters move some drugs out of the brain, pumping out toxic or foreign chemicals in the brain. However, in-vivo and in-vitro tests of osimertinib were demonstrated to be a poor substrate and had the highest BBB penetrance; it was demonstrated to possess a rate of 0.21 Kpuu (Kpuu is the rate of drug distribution) compared to other TKIs, which were generally below 0.12 Kpuu, enabling the TKI to remain in the brain (Colclough et al.). Kpuu is the proportion, or ratio of drug concentration in tissue to drug concentration in blood

(Loryan et al.). Thus, osimertinib has been more effective in maintaining drug levels in target tissue.

The AURA2 trial demonstrated that 70% of patients who received a daily dose (80mg) of osimertinib had a partial or complete response, reducing or even eliminating the tumor. In EGFR-mutated non-small cell lung cancer (NSCLC) patients, Osimertinib is also being tested in combination therapies. For example, when resistance occurs through MET (Mesenchymal Epithelial Transition) amplification, osimertinib in combination with a MET inhibitor has been found to be promising. MET amplification is one of the frequent mechanisms of resistance to targeted drugs such as osimertinib. When there is amplification of MET, the cancer cells use an alternate signaling pathway that circumvents EGFR through which the tumor continues to grow even if EGFR is being inhibited well. The AURA3 trial showed that MET amplification was one of the most commonly acquired resistance mechanisms in patients who progressed on second-line osimertinib. About 19% of patients had MET amplification at resistance (Coleman et al.).

The findings showed that while osimertinib is very potent at targeting EGFR mutations, a proportion of tumors become resistant to it by activating the MET pathway, allowing cancer growth to continue independently of EGFR signaling. As a result, the study indicated the promising osimertinib and MET inhibitors combination to overcome this type of resistance. (Cross et al.). This combination will be capable of overcoming resistance and keeping the treatment effective. Finally, dosing of Osimertinib is straightforward with an oral dosage of 80 mg once daily. This ease of administration, along with its therapeutic benefit and lower toxicity, has made osimertinib a highly popular targeted agent for EGFR-mutant NSCLC (Chmielecki et al.).

Table 2: Limitations of Osimertinib

Disadvantages	**Explanation**	**Reference**
Selective for T790M mutation	In some cases, osimertinib doesn't work because the patient develops another mutation that is not T790M	(Peng et al.)
Side effects	Side effects include rash, diarrhea, fatigue, and mouth sores	(Odogwu et al.)
Additional drug resistance through mutations	Osimertinib still may not ameliorate emerging drug-resistant mutations	(Zhang et al.)

Since Osimertinib is a second or third-line therapy that can only be used after initial treatment, by then, mutations such as T970 can form in the EGFR, even in wild-type EGFRs that haven't had that specific mutation to begin with. Then, it makes sense to use Osimertinib. However, it is not the case in every patient. Some patients do not have the T970 or do not develop that mutation after the first or second line of treatment. According to a previous study, Osimertinib treatment after initial treatment led to the T970 mutation only accounted for 60% of the surveyed patients. This excluded the remaining 40% of the patients, showing how still, Osimertinib doesn't work for many people (Peng et al.). Moreover, although Osimertinib has fewer side effects compared to first and second line treatments, it still has a significant amount of side effects. In a study conducted by the FDA (>20%) side effects in patients treated with osimertinib were diarrhea, rash, dry skin, nail toxicity, and fatigue (Odogwu et al.). Although Osimertinib has shown significant efficacy, additional mutations such as C797S mutations emerge in the EGFR, hindering its long-term effectiveness. In this specific example, serine SER797 replaces cysteine CYS797, which disrupts the covalent bonds formed by osimertinib to the ATP binding pocket (Zhang et al.). As a result of the new mutation, another TKI needs to be created to resolve this issue.

Conclusion:

The development of Osimertinib, a third-generation TKI, was groundbreaking; Osimertinib has changed the way EGFR-mutated NSCLC is treated, especially in patients with the T790M mutation in the ATP binding pocket. It is more targeted, can effectively cross the blood-brain barrier, and has fewer side effects relative to the previous generation TKIs. However, like most cancer treatments, resistance eventually develops. To overcome this resistance, current research is focusing on 4th-generation TKIs to target the C797S mutation in the EGFR specifically (Zhang et al.).

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References

Castañeda-González, Juan Pablo, et al. "Multiple Mutations in the EGFR Gene in Lung Cancer: A Systematic Review." *Translational Lung Cancer Research*, vol. 0, no. 0, Jan. 2021, https://doi.org/10.21037/tlcr-22-235. Accessed 25 Sept. 2022.

Chmielecki, Juliann, et al. "Analysis of Acquired Resistance Mechanisms to Osimertinib in Patients with EGFR-Mutated Advanced Non-Small Cell Lung Cancer from the AURA3 Trial." *Nature Communications*, vol. 14, no. 1, 27 Feb. 2023, https://doi.org/10.1038/s41467-023-35962-x.

Colclough, Nicola, et al. *Preclinical Comparison of the Blood–Brain Barrier Permeability of Osimertinib with Other EGFR TKIs*. Vol. 27, no. 1, 1 Jan. 2021, pp. 189–201, https://doi.org/10.1158/1078-0432.ccr-19-1871.

"Beyond Epidermal Growth Factor Receptor: MET Amplification as a General Resistance Driver to Targeted Therapy in Oncogene-Driven Non-Small-Cell Lung Cancer." ESMO Open, vol. 6, no. 6, Dec. 2021, p. 100319, https://doi.org/10.1016/j.esmoop.2021.100319.

Cross, Darren A E, et al. "AZD9291, an Irreversible EGFR TKI, Overcomes T790M-Mediated Resistance to EGFR Inhibitors in Lung Cancer." Cancer Discovery, vol. 4, no. 9, 2014, pp. 1046–61, www.ncbi.nlm.nih.gov/pubmed/24893891/, https://doi.org/10.1158/2159-8290.CD-14-0337.

Fu, Kai, et al. "Therapeutic Strategies for EGFR-Mutated Non-Small Cell Lung Cancer Patients with Osimertinib Resistance." *Journal of Hematology & Oncology*, vol. 15, no. 1, 8 Dec. 2022, jhoonline.biomedcentral.com/articles/10.1186/s13045-022-01391-4, https://doi.org/10.1186/s13045-022-01391-4.

Koulouris, Andreas, et al. "Resistance to TKIs in EGFR-Mutated Non-Small Cell Lung Cancer: From Mechanisms to New Therapeutic Strategies." *Cancers*, vol. 14, no. 14, 8 July 2022, p. 3337, pubmed.ncbi.nlm.nih.gov/35884398/, https://doi.org/10.3390/cancers14143337. Accessed 24 Oct. 2022.

Loryan, Irena, et al. "Unbound Brain-To-Plasma Partition Coefficient, Kp,Uu,Brain—a Game Changing Parameter for CNS Drug Discovery and Development." *Pharmaceutical Research*, vol. 39, no. 7, 11 Apr. 2022, pp. 1321–1341, https://doi.org/10.1007/s11095-022-03246-6. Accessed 6 Sept. 2023.

Odogwu, Lauretta, et al. "FDA Benefit-Risk Assessment of Osimertinib for the Treatment of Metastatic Non-Small Cell Lung Cancer Harboring Epidermal Growth Factor Receptor T790M Mutation." *The Oncologist*, vol. 23, no. 3, 1 Mar. 2018, pp. 353–359, pubmed.ncbi.nlm.nih.gov/29242281/, https://doi.org/10.1634/theoncologist.2017-0425. Accessed 3 Aug. 2021.

Ou, Xuejin, et al. "Mechanisms of Resistance to Tyrosine Kinase Inhibitor-Targeted Therapy and Overcoming Strategies." *MedComm*, vol. 5, no. 9, Winter 2024, p. e694, pubmed.ncbi.nlm.nih.gov/39184861/, https://doi.org/10.1002/mco2.694.

Peng, Mu-Han, et al. "Osimertinib as Second- and ≥Third-Line Treatment in Advanced and Recurrence EGFR-Mutant NSCLC Patients Harboring Acquired T790M Mutation." *Cancers*, vol. 16, no. 24, 14 Dec. 2024, pp. 4174–4174, www.mdpi.com/2072-6694/16/24/4174, https://doi.org/10.3390/cancers16244174. Accessed 17 Dec. 2024.

Testa, Ugo, et al. "Lung Cancers: Molecular Characterization, Clonal Heterogeneity and Evolution, and Cancer Stem Cells." *Cancers*, vol. 10, no. 8, 27 July 2018, p. 248, https://doi.org/10.3390/cancers10080248.

"The Cancer Therapy Arms Race | Bio-Rad." *Bio-Rad*, 2019, www.bio-rad-antibodies.com/blog/cancer-therapy-arms-race.html. Accessed 5 July 2025.

Wang, Chunsheng, et al. "Patterns and Treatment Strategies of Osimertinib Resistance in T790M-Positive Non-Small Cell Lung Cancer: A Pooled Analysis." *Frontiers in Oncology*, vol. 11, 2 Mar. 2021, https://doi.org/10.3389/fonc.2021.600844. Accessed 16 Apr. 2022.

Zhang, Die, et al. "Fourth-Generation EGFR-TKI to Overcome C797S Mutation: Past, Present, and Future." *Journal of Enzyme Inhibition and Medicinal Chemistry*, vol. 40, no. 1, 2 Apr. 2025, https://doi.org/10.1080/14756366.2025.2481392. Accessed 6 Apr. 2025.