

# Cystic Fibrosis: Treatment with CFTR Modulators

---

**Westphal, Anna Philine**

**Master's thesis / Diplomski rad**

**2023**

*Degree Grantor / Ustanova koja je dodijelila akademski / stručni stupanj:* **University of Rijeka, Faculty of Medicine / Sveučilište u Rijeci, Medicinski fakultet**

*Permanent link / Trajna poveznica:* <https://um.nsk.hr/um:nbn:hr:184:380526>

*Rights / Prava:* [In copyright](#)/[Zaštićeno autorskim pravom.](#)

*Download date / Datum preuzimanja:* **2025-02-06**



*Repository / Repozitorij:*

[Repository of the University of Rijeka, Faculty of Medicine - FMRI Repository](#)



**UNIVERSITY OF RIJEKA**  
**FACULTY OF MEDICINE**

**INTEGRATED UNDERGRADUATE AND GRADUATE UNIVERSITY STUDY OF**  
**MEDICINE IN ENGLISH**

**Anna Philine Westphal**

**CYSTIC FIBROSIS: TREATMENT WITH CFTR MODULATORS**  
**GRADUATION THESIS**

Rijeka, June 2023

**UNIVERSITY OF RIJEKA**  
**FACULTY OF MEDICINE**

**INTEGRATED UNDERGRADUATE AND GRADUATE UNIVERSITY STUDY OF**  
**MEDICINE IN ENGLISH**

**Anna Philine Westphal**

**CYSTIC FIBROSIS: TREATMENT WITH CFTR MODULATORS**  
**GRADUATION THESIS**

Rijeka, June 2023

Thesis Mentor: Prof. Dr. sc. Srdan Banac

The graduation thesis was graded on June 28<sup>th</sup> 2023 in Rijeka, before the Committee composed of the following members:

1. Prof. dr. sc. Goran Palčevski (Committee Head)
2. Izv. Prof. dr. sc. Ljiljana Bulat-Kardum
3. Prof. dr. sc. Dinko Vitezić

The graduation thesis contains 40 pages, 2 figures, 2 tables, 32 references

## Table of Content

<b>1. Introduction</b> .....	<b>6</b>
<b>2. CFTR and genetic mutations</b> .....	<b>8</b>
<b>3. CFTR Modulators</b> .....	<b>10</b>
<i>a. History and Development</i> .....	<i>11</i>
<i>b. Pharmacodynamics</i> .....	<i>13</i>
<b>4. Results and Achievements</b> .....	<b>15</b>
<i>a. Effects on Lung Function</i> .....	<i>15</i>
<i>b. Effects on Gut Function</i> .....	<i>18</i>
<i>c. Effects on Macrophage Function</i> .....	<i>19</i>
<i>d. Ivacaftor</i> .....	<i>21</i>
<i>e. Lumacaftor-Ivacaftor</i> .....	<i>23</i>
<i>f. Tezacaftor-Ivacaftor</i> .....	<i>25</i>
<i>g. Elexacaftor/Tezacaftor/Ivacaftor</i> .....	<i>26</i>
<i>h. General Approach</i> .....	<i>28</i>
<b>5. Discussion</b> .....	<b>30</b>
<b>6. Conclusion</b> .....	<b>33</b>
<b>7. Summary</b> .....	<b>34</b>
<b>8. Literature</b> .....	<b>35</b>
<b>9. Curriculum Vitae</b> .....	<b>39</b>

## **Abbreviations and Acronyms**

AE – Adverse Effects

ASL – Airways Surface Liquid

ATP – Adenosine Triphosphate

AUC – Area Under the Curve

*B. cenocepacia* – *Burkholderia cenocepacia*

BMI – Body Mass Index

cAMP – cyclic Adenosine Monophosphate

CF – Cystic Fibrosis

CF MDM – Cystic Fibrosis Monocyte-derived Macrophage

CFQ-R – Cystic Fibrosis Questionnaire-Revised (Application)

CFRD – CF-related diabetes mellitus

CFTR – CF transmembrane regulator

CSR – Cell Stress Response

CYP3A – Cytochrome P450 3A

*E. Coli* – *Escherichia Coli*

ELX – Elaxacaftor

ELX/TEZ/IVA – Elaxacaftor/Tezacaftor/Ivacaftor

ER – Endoplasmic Reticulum

ERAD – endoplasmic-reticulum-associated degradation

EMA – European Medicine Agency

FDA – Food and Drug Administration

$\Delta$ F508 – Deletion of a phenylalanine residue at position 508 in the CFTR protein

GSNO – S-Nitrosoglutathione

*H. influenzae* – *Haemophilus Influenzae*

HBAE cell – Human Bronchial Airway Epithelial Cell

HBEC – Human Bronchial Epithelial Cell

HSP – Heat Shock Protein

IV – intravenous

IVA – Ivacaftor

LUM – Lumacaftor

LUM/IVA – Lumacaftor/Ivacaftor

mRNA – messenger Ribonucleic Acid

MSD-1 – Membrane Spanning Domain

M2-PK – Pyruvate-Kinase M2

*P. aeruginosa* – *Pseudomonas Aeruginosa*

ppFEV<sub>1</sub> – Percent Predicted Forced Expiratory Volume in 1 Second

QoL – Quality of Life

*S. Aureus* – *Staphylococcus Aureus*

*S. pneumoniae* – *Streptococcus pneumoniae*

SNOs – S-Nitrosothiols

TEZ – Tezacaftor

TEZ/IVA – Tezacaftor/Ivacaftor

## 1. Introduction

Cystic Fibrosis (CF) is an inherited disease, which is most prevalent in Northern parts of Europe. In fact, it is the second most common inherited metabolic disease and the most common lethal genetic condition in the Caucasian population. (1) Currently, around 70.000 cases are known worldwide, whereas around 1.000 new cases are added yearly. (2)

The disease is caused by an autosomal recessive mutation in the CF transmembrane regulator (CFTR) protein, leading to an absent or dysfunctional protein, or a decrease in its quantity. The consequence is a disturbed regulation of chloride in different organ tissues of the human body, mainly within the respiratory epithelium and the gastrointestinal tract, but also other organs such as sweat glands can be affected.

Due to the inability of the ATP-guided chloride channels to transport chloride across the cell membrane and the consequent accumulation in sodium within the cells, CF is characterized by an increased water reabsorption and a subsequent thickened mucus. The accumulation of the malfunctioning mucus makes the patient vulnerable to bacterial infections presenting with pulmonary exacerbations, chronic obstructive pulmonary disease with bronchiectasis and chronic sinusitis secondary to a dysfunctional mucociliary transport, stasis of the mucus and the provision of an optimal environment for bacteria to produce inflammation. Chronic bacterial infection is no rare consequence, giving rise to end-stage lung disease, which is simultaneously the most common cause of morbidity and mortality in cystic fibrosis diseased patients. Generally, respiratory symptoms are more common to present in adulthood, compared to symptoms occurring in childhood. Further typical respiratory symptoms include chronic sinusitis, eventually associated with nasal polyps, recurrent or chronic productive cough, hemoptysis and the above mentioned recurring pulmonary infections. Common pathogens for pulmonary infections in cystic fibrosis patients are *Burkholderia cepacia* (*B. cepacia*), *Streptococcus pneumoniae* (*S. pneumoniae*) and *Haemophilus Influenzae* (*H. Influenzae*). Furthermore, CF patients are vulnerable to opportunistic pathogenic infections, which include infections with *Pseudomonas aeruginosa* (*P. aeruginosa*) and *Aspergillus*. (3)

The classic clinical triad of cystic fibrosis that might start presenting in infancy or early childhood consists of a progressive obstructive lung disease, often accompanied by pulmonary infections, an exocrine pancreatic insufficiency, and an elevated sweat chloride level.

In newborns, cystic fibrosis commonly presents with a meconium ileus. In fact, almost all cases of meconium ileus are related to cystic fibrosis, being the underlying disease.

In infants and older children the disease is characterized by a failure to thrive or as a clinical picture, which might be a consequence of an interaction of different pancreatic diseases. These



pancreatic diseases are mostly defined by an exocrine insufficiency, presenting with a foul-smelling steatorrhea, malabsorption, and a deficiency of fat-soluble vitamins. Also, an endocrine dysfunction may occur, which is represented by a CF-related diabetes mellitus (CFRD). Additional intestinal symptoms are intestinal obstruction, cholecystolithiasis or cholestasis, fatty metamorphosis of the liver and biliary cirrhosis.

Since the chloride channels in sweat glands are responsible for transporting chloride inside the cell, a dysfunction in these channels has the consequence of excessive salt loss and increased NaCl levels within sweat. Nowadays, screening in newborns is mandatory in most Western countries, leading to an early diagnosis of CF. A significant loss of salt via sweat can possibly lead to electrolyte wasting. (3)

Other symptoms to mention include the musculoskeletal and urogenital systems. Due to osteopenia, frequent fractures can occur, as well as kyphoscoliosis. The urinary tract is affected in the form of nephrolithiasis, nephrocalcinosis and frequent urinary tract infections, while the genital system might be affected in the form of infertility in men, and reduced fertility in women. An obstructive azoospermia in men can occur, even though the spermatogenesis may be intact. The vas deferens might be absent, and the testicles might be undescended. In women the thick cervical mucus can obstruct the fertilization. Amenorrhea can additionally occur. (3)

The approach to the management of Cystic Fibrosis changed during the last decade. Since the disease is based on genetic mutations, it was treated symptomatically for a long period of time. Preservation of lung function, optimization of nutrition and screening for complications are some of the goal-directed interventions used to prevent patients from severe complications. (4) CFTR modulators are a new group of drugs targeting specific defects in the CFTR protein to optimize their function. Depending on their mechanism of action, they can also be combined to synergistically improve the CFTR protein's function. Since the introduction of CFTR modulators, many studies show their effectiveness regarding an improve in pulmonary function and subsequent patient-reported respiratory symptoms, reduction in pulmonary exacerbation and the overall nutritional status. (5)

This thesis will focus on the presentation of the different CFTR modulators, their history of development and some inspects towards the future evolution of what is yet to be developed.

## 2. CFTR and genetic mutations

Cystic Fibrosis is a disease that can be caused by three different types of mutations, which are divided into six classes. Depending on the mutation type, the disease presents in different severities and clinical pictures.

The mutations can be either of a deletion, nonsense or gating type. Deletions are by far the most common mutations, making up to 88% of all mutations causing cystic fibrosis. (6)

### a. Classes of CFTR Mutations

*Table 1: CFTR Mutation Classification*

Type of Mutation	Type of CFTR Mutation	Percent of people with CF who have at least 1 mutation
<b>Normal</b>	CFTR protein is created and transported to the cell membrane, normal transfer of chloride and water	
<b>Class I</b>	No functional CFTR protein is produced	22%
<b>Class II</b>	Production of misfolded CFTR proteins, keeping it from moving to the cell surface (trafficking defect)	88%
<b>Class III</b>	CFTR protein is produced and transported to the cell surface, but the channel gate does not open (defective channel regulation)	6%
<b>Class IV</b>	CFTR protein is produced and transported to the cell surface, but the channel function is malfunctioning (decreased channel conductance)	6%
<b>Class V</b>	Normal CFTR protein is produced and transported correctly to the cell surface but in an inadequate amount (reduced synthesis of CFTR)	5%
<b>Class VI</b>	CFTR protein is produced, but it does not work properly at the cell membrane (decreased CFTR stability)	5%

Source: based on Anas Z, ElSaygh J, ElSORI D, et al. A review of Trikafta: Triple Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) Modulator Therapy. Juli 03, 2021 [Internet]. [accessed November 19, 2022]; Cureus 13(7): e16144. DOI 10.7759/cureus.16144 Available from [https://www.researchgate.net/publication/352952690\\_A\\_Review\\_of\\_Trikafta\\_Triple\\_Cystic\\_Fibrosis\\_Transmembrane\\_Conductance\\_Regulator\\_CFTR\\_Modulator\\_Therapy](https://www.researchgate.net/publication/352952690_A_Review_of_Trikafta_Triple_Cystic_Fibrosis_Transmembrane_Conductance_Regulator_CFTR_Modulator_Therapy)

The first class of mutations includes frameshift, splicing and non-sense mutations with the consequence of reduced messenger ribonucleic acid (mRNA) amount and dysfunctional CFTR protein syntheses. This class of mutations can be subclassified in class IA and class IB, while Class IA describes large deletion mutations with absent mRNA production and class IB describes non-sense mutations, which result in a premature termination codon and the synthesis

of a rather unstable mRNA. Usually, the abnormality of the mRNA sequences is detected rapidly and therefore subsequently degraded.

The second class of mutations leads to misfolding of CFTR proteins. This inhibits a normal trafficking towards the apical cell surface and therefore reduces the amount of functional CFTR proteins on the cell membrane. The defect is detected early, resulting in proteasomal degradation, which is called endoplasmic-reticulum-associated degradation (ERAD). This class of mutations makes up the most common type of cystic fibrosis causing genetic defects, being present in about 88% of cases. The most common mutation is the F508del mutation.

In class III of genetic mutations, the CFTR proteins are trafficked to and integrated correctly into the apical cell membrane, but a defect in CFTR gating channel is limiting the normal CFTR channel opening. An atypical connection of ATP to the nucleotide binding domains (NBD1 and NBD2), additionally to a reduction in ATP hydrolysis is causing the dysfunction in gating. In most cases, the mutation is an exchange of glycine to aspartate at the gene position 551 in NBD1 of the CFTR gene. (7) This leads to a hundred times lower open probability compared to a healthy individual. Therefore, this gene mutation is associated with a very severe phenotype, causing classical manifestations of CF. Common mutations in this class are G551D or S549N. (8)

In class IV of gene mutations, a reduction in chloride and bicarbonate conduction is caused by a mutation causing a dysfunctional anion selectivity of the CFTR protein. Usually, the phenotype is not as severe as in class III mutations because some conductance is preserved. R117H mutation is the most common in this class, being a missense mutation.

In class V of mutations, promotor mutations, as well as alternate or noncanonical splicing defects or missense mutations can lead to abnormal mRNA transcripts with the consequence of greatly diminished amounts of functional CFTR proteins at the cell membrane. (7) This can be the consequence of either an increased deactivation of functional proteins or an insufficient protein production, both leading to a paltry number of proteins on the apical cell membrane. Usually, the clinical manifestations are mild. (8)

Class VI includes mutations that produce a normally working CFTR protein migrating regularly into the cell membrane, but with an abnormal stability. The endocytosis or turnover is accelerated, diminishing the density and function of the integrated CFTR protein. (9)

A second classification system classifies the mutations according to their disease severity. Residual function mutations still retain some CFTR function, therefore they are rather associated with a milder, non-severe type of disease presentation. These patients, compared to

other classes are more prone to be pancreatic sufficient and might present with a later onset of a clinical presentation. Commonly, patients with these types of mutations respond to CFTR potentiators, such as Ivacaftor (IVA). Studies suggest that residual function mutations are defined as patients having a sweat chloride level  $<86$  mEq/l and pancreatic insufficiency in less than 50% of cases. Additionally, it might be determined when an increase in chloride transport of  $>10\%$  appears after introduction of IVA.

All mutations with trivial function, and which do not respond to CFTR modulators can be classified in the class of minimal function mutations. For instance, in studies it was shown that this class includes mutations leading to no full-length proteins or a baseline chloride transport with less than 10% of normal CFTR and a less than 10% increase after incubation with Tezacaftor (TEZ), IVA or the combination of both. (8)

### **3. CFTR Modulators**

CFTR modulators are a certain group of drugs that direct at molecular defects in the CFTR protein to intensify the CFTR activity. Three classes of CFTR modulators are yet known and in use.

CFTR Potentiators increase the open probability and the time period that the channel spends in the open position of the mutated CFTR channels that have either gating or conductance mutations and are therefore indicated in class III or IV mutations. Additionally, potentiators magnify the flux of chloride and bicarbonate ions along the cell surface. (8) Studies suggest that potentiators can also be used in the F508del mutation and can therefore be indicated in class II mutations. For this to be efficient, it is required that the dysfunctional CFTR channel is already present on the cell membrane. Therefore, the potentiator alone cannot be used to treat class I or II mutations but is used in combination with correctors. The most common potentiator used is called Ivacaftor. It mainly targets class III mutations, especially G551D.

CFTR Correctors improve the folding, the trafficking of CFTR or the transport towards the cell surface by stabilizing the three-dimensional conformation of the protein, even though it might be misfolded. This type of modulator is especially recommended in class II mutations, for example in the F508del mutation. Approved Correctors are Tezacaftor (TEZ), Lumacaftor (LUM) and Elexacaftor (ELX), while ELX has the ability to work as a corrector, as well as a potentiator. Hereby, it works synergistically with TEZ as a corrector, and synergistically with IVA as a potentiator. (8)

CFTR Read-Through-Agents suppress premature stop codons, leading to an inhibition of premature stop during translation and therefore allowing a stable reading flow and the creation of a full-length CFTR protein. Consequently, they are indicated in class I mutations. The most common drug used from this group is Ataluren. (6)

Additionally, there are also studies about amplifiers and stabilizers, but yet none of these is approved. Amplifiers should improve the presentation of the dysfunctional CFTR mRNA and the subsequent establishment of the final CFTR protein.

Stabilizers should decrease the elimination and deactivation of CFTR protein from the apical cell membrane. Both classes should work in the mutation groups II to V, acting on processing, gating, conduction or insufficient protein mutations. (8)

#### a. History and Development

In 1989, the CFTR gene was cloned for the first time. For many years, scientists have been searching for a solution to correct the dysfunctional gene. Up to then, gene therapy to replace the abnormal gene within the pulmonary epithelium with a wild-typed DNA seemed to be the best option. Generally, this is tolerated by patients, but unfortunately it does not achieve an adequate clinical outcome. (10)

Within the last 10 years, the introduction of CFTR modulators has meaningfully ameliorated the course of cystic fibrosis and yielded remarkable improvement in the clinical outcome of patients in all age groups.

In 2012, the first CFTR modulator Ivacaftor was approved on the market (Kalydeco®). It had a significant impact on cystic fibrosis patients while decreasing the sweat chloride concentration under the cystic fibrosis diagnostic range and increasing the general lung function of about 10%. (11) IVA is a drug that mainly works on gating channel mutations, covering class III of mutations. Accordingly, it is applicable in only 10% of the cystic fibrosis population and consequently cannot be utilized by the majority of CF patients.

Three years later, a new drug was approved by the Food and Drug Administration (FDA), named IVA/LUM (Orkambi®), consisting of a combination of the drugs Ivacaftor and Lumacaftor. LUM is a CFTR corrector and therefore it has a significant effect on F508del mutations. By stabilizing the three-dimensional conformation of the CFTR protein and by supporting the trafficking of the protein towards the cell membrane, it leads to a modest clinical improvement for patients with a F508del mutation of the CFTR gene. (6,11)

Since LUM/IVA is effective on homozygous F508del mutations, it can be utilized by around 40% to 50% of the cystic fibrosis population. Generally spoken, it showed positive, but less

impressive results regarding the clinical outcome when compared to the first success of IVA. (10)

2018, a new drug combination was approved in the US with IVA and TEZ as active substances. (12) This combination is called Symdeko® and seems to achieve a better tolerance and a better drug-drug interaction profile compared to LUM/IVA, while presenting the same efficiency. TEZ/IVA was approved for several residual-function CFTR mutations in addition to F508del. It helped CF patients by decreasing the number of pulmonary exacerbations and by increasing FEV<sub>1</sub> up to 3-4%. It did not result in significant Body Mass Index (BMI) changes. (11)

New dimensions for the treatment of cystic fibrosis showed up when the triple combination of CFTR modulators in heterozygous CF patients with an F508del mutation and minimal function mutation was approved, consisting of Elexacaftor, Tezacaftor and Ivacaftor. ELX acts synergistically to IVA as a CFTR potentiator. (13) It was demonstrated that with this combination of drugs the results for homozygous and minimal function F508del mutation patients were similar, with an improvement in FEV<sub>1</sub> of 10,4% to 13,8%, decreased sweat chloride concentrations and increased quality of life (QoL). (11)

Important to mention is that the BMI and the number of exacerbations significantly improved in patients with minimal function mutations, when compared to the homozygous genotype. Generally, the triple CFTR modulations presented with an acceptable tolerability.

Subsequently, one can conclude that the combination therapy of LUM/IVA or TEZ/IVA is efficient for patients with a homozygous genotype of F508del mutation, but rather inefficient in heterozygous patients. For these group of patients, the triple therapy of Elexacaftor/Tezacaftor/Ivacaftor (ELX/TEZ/IVA) showed pronounced clinical improvement, establishing a life-changing treatment option for patients with the minimal function genotype of CF. Additionally, ELX/TEZ/IVA can be applied to many CF patients due to its ability to target all mutation groups with at least one copy of F508del mutation, which represent more than 90% of the cystic fibrosis population.

The future of the treatment of CF is not yet to be determined. Hopefully, a next generation of CFTR modulators will be also efficient for patients with rarer mutations which might show a resistance towards LUM/IVA. Yet, 10% of all Cystic Fibrosis patients have no opportunity to be treated with CFTR modulators. Additionally, the treatment of infections in the CF is still a challenge. (11)

b. Pharmacodynamics

Genetic Mutations in Cystic Fibrosis depend on the class they belong to. As mentioned above, there are different types of mutations: deletions, nonsense and gating mutations. Consequently, the CFTR modulators targeting different types of mutations also need to meet different mechanisms of actions to fulfill their function.

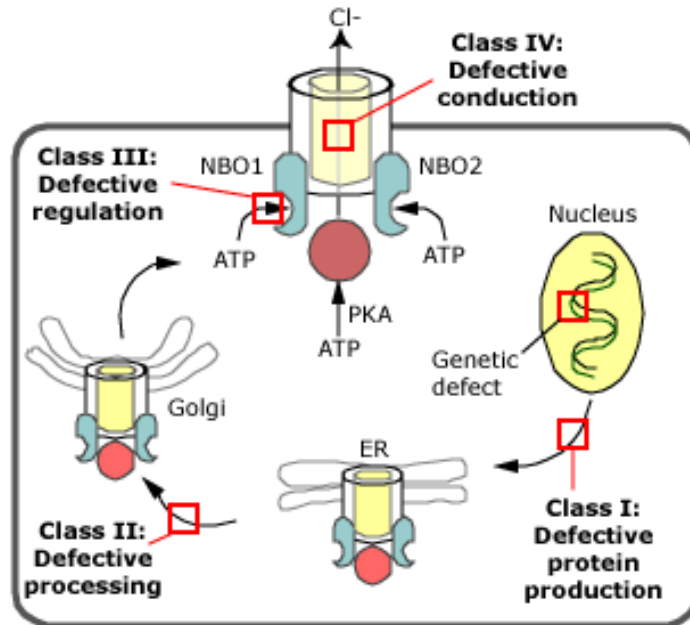


Figure 1: Defects of different CFTR mutations

Source: “Cystic Fibrosis” (1994) in *Metabolic and molecular basis of inherited disease*. McGraw-Hill, p. 3801.

The image reminds about the certain classes of mutations that can be targeted by CFTR modulators. Class I shows to have the dysfunction in protein production, while class II mutations are affected by a dysfunctional protein folding and trafficking towards the cell surface. Class III mutations have a defective regulation of channel opening, whereas class IV mutations suffer from a defective conduction within the channel opening. (14) All these different circumstances of pathophysiology have to be targeted by different pharmacodynamics of each drug. Yet, little is known about the exact mechanisms of actions of this group of drugs. Generally spoken, IVA as a CFTR potentiator enhances the channel-opening probability of the target protein at the apical cell membrane surface. It is indicated in patients presenting with at least one genetic variant affecting the gating capacity, classified in class III of genetic mutations. These mutations have the consequence of a normal transport of the CFTR proteins towards the cell surface, but the following cyclic Adenosine Monophosphate (cAMP) - mediated activation of the protein is dysfunctional. The exact mechanism of how IVA enables a prolonged opened status of the protein is unknown, but it is speculated that a certain

decoupling process of the gating cycle and ATP hydrolysis cycle may enable the chloride transport. (15)

LUM, as a CFTR corrector targets directly on the F508del CFTR mutation with the result of an increase in the cellular processing and trafficking. With that, the number of functional CFTR proteins at the cell surface rises. Other CFTR correctors, such as TEZ bind to the first Membrane Spanning Domain (MSD-1) of CFTR and then have the same mechanism of action as LUM. ELX as the third CFTR corrector to mention binds to different sites on the CFTR protein, resulting in an additive effect and finally giving rise to an increase of CFTR proteins on the cell surface. But also, here the exact mechanism is yet unknown. (16)

Generally spoken, for the F508del mutation there are different discussions about a cell rescue mechanism for cell stress responses (CSR). In particular, heat shock proteins (HSPs), osmolytes and low temperature and its synergy are focused on in certain studies. All of these play an important role within CSR. Focusing on modulation of one of these entities inducing, or being involved in the induction of CSR, that might be a possible mechanism of correcting trafficking defects of CFTR-mutated proteins. (17)

Additionally, S-Nitrosothiols (SNOs) are discussed of being involved in the cells rescue of trafficking CFTR proteins towards the cell surface, since they increase the expression of F508del CFTR proteins on the cell surface of human bronchial epithelial airway (HBAE) cells in a dose-dependent manner. It is shown that the presence of S-nitrosoglutathione (GSNO) is associated with an increase in the expression, function and maturation of F508del CFTR proteins on the HBAE. As mentioned above, low temperature plays an important role in research about the molecular pathophysiology of CF. Studies proofed that low temperature is efficient for rescuing the trafficking of not properly folded F508del CFTR proteins by increasing their stability and slowing down the process of degradation by proteasomes. (18)

All this pathophysiological knowledge of cystic fibrosis has led and might lead pharmacists to the development of new correctors for the treatment of class II diseased CF patients.



## 4. Results and Achievements

### a. Effects on Lung Function

The most common symptoms of patients suffering from CF are related to lung function. This is due to a large quantity of CFTR proteins in the human bronchial epithelial cells (HBEC), which are mutated and dysfunctional in CF patients. Dysfunctional CFTR proteins are not able to secrete chloride effectively, therefore also sodium and water is secreted less. Consequently, dehydration occurs, and the mucus is thickened and prone to stasis. This creates the perfect surrounding for bacteria such as *P. aeruginosa* to colonize and to produce an inflammation. Finally, a structural damage to the bronchial epithelium and general airway system is the aftermath, commonly associated with bronchiectasis and respiratory failure.

CFTR modulators were ideated to offer a causal solution for patients with CF, instead of only treating symptoms as it was done before the approval of Ivacaftor 2012. In clinical trials it is common to name endpoints to which the trial is primarily or secondarily focused on. Lung function measured in FEV<sub>1</sub> is a frequent primary endpoint, while the quantity of pulmonary exacerbations is a common secondary endpoint in most trials. (19)

Gramegna et al. compared a total of 23 papers in regard to a systematic review of the effect of certain CFTR modulators. In total, 4219 patients were part of these studies, performed in a time period of 15 years, between 2005 and 2020.

Here, IVA, given in a dose of 150mg twice daily showed an enhanced pulmonary function in different trial groups. In a group of patients with a G551D gating mutation, children aged 6-11 years showed an improvement of the percent predicted forced expiratory volume in 1 second (ppFEV<sub>1</sub>) of +12,5 percentage points, while patients aged more than 12 years old presented improvements of +10,6 percentage points. Patients with a non-G551D mutation, older than 6 years achieved an overall improvement of FEV<sub>1</sub> with +8,3 percentage point, and patients having a R117H mutation, older than 6 years +2,1 percentage points.

LUM/IVA, administered to patients homozygous for F508del in a phase II and phase III clinical trial, showed an improve in FEV<sub>1</sub> of +6,1 and +4,8 percentage points, respectively. TEZ/IVA, given to patients homozygous for F508del and older than 12 years showed an improve of ppFEV<sub>1</sub> of +4,0 percentage points, and +6,8 percentage points in F508del residual function mutations. ELX/TEZ/IVA was tested in patients beyond 12 years. One group was homozygous for F508del mutation and showed an improvement of +9,7 percentage points, while the other group had a residual function F508del mutation and improved about +13,3 percentage points.

Pulmonary Exacerbations showed to be the most reduced when treated with ELX/TEZ/IVA. Here, the pulmonary exacerbations in patients homozygous for F508del mutation reduced about 63%. In patients with G551D mutations receiving IVA, the pulmonary exacerbation rate

reduced about 55%, while in patients homozygous for F508del treated with LUM/IVA the rate reduced only about 39% and in patients treated with TEZ/IVA only about 35%.

To conclude, according to Gramegna, patients with one gating mutation receiving IVA and patients with F508del mutation (homo- and heterozygous) receiving ELX/TEZ/IVA showed the most beneficial effect in regard to overall lung function, pulmonary exacerbations and symptom improvement. (20)

Most clinical trials included patients who have a lung function with a value of  $FEV_1 >40\%$  and  $<90\%$ . Since a majority of existing patients today are no children, the severity of lung disease increases, and consequences of the significant lung damage evolve more and more. This includes structural changes in lung parenchyma, chronic pulmonary infection, malnutrition and frequent hospitalizations with intravenous (iv) antibiotic treatment.

Shteinberg et al. summarized the effects of CFTR modulator therapy in a subgroup of patients suffering from a severe and advanced lung disease, with a lung function value of  $<40\%$  (See table below). A study of Barry et al. focused with under on an extended access program (n=21) including people with severe CF pulmonary disease ( $FEV_1$  % pred  $<40\%$ ) with a G551D mutation. Here, the treatment with IVA led to an improvement of  $FEV_1$  of 4,2%, compared to a group with a similar pulmonary insufficiency, not G551D positive and not treated with IVA. Additionally, a significant increase in lung function and a noteworthy reduction of needed days with iv antimicrobial treatment was mentioned.

Wrainwright et al. did researches on patients homozygous for F508del mutation. LUM/IVA was administrated in these patients, showing a lesser increase in lung function compared to IVA for G551D mutation, but a powerful result for the reduction in hospitalizations (30-61%) and for the reduction in iv antibiotic use (45-56%). A subgroup of patients had a pulmonary function value  $<40\%$  and showed similar results as the patients with better pulmonary function.

The TRAFFIC and TRANSPORT studies from Elborn et al. inform about a subgroup of patients with severely reduced lung function. Here, an increase of +3,7 and +3,3 percentage points of lung function improvement compared to a placebo group was shown.

The EVOLVE Study compared the administration of TEZ/IVA to a placebo group, showing an overall improvement in lung function of +4%, and a decrease in pulmonary exacerbation of -35% in patients with a baseline lung function of  $<40\%$ .

Last but not least, the triple combination of ELX/TEZ/IVA was tested in patients with severely reduced lung function, which showed a markedly improved lung function after administration, with an increase of 10-14%, compared to placebo. (21)

Table 2: Comparison of CFTR modulators in patients with severe lung disease from published trials

First author [ref.]	Population; study design: RCT/open	Overall effect: lung function	Overall effect: exacerbation reduction	Patients with severe disease (FEV <sub>1</sub> <40% pred)	Lung function increase [% points] in severe disease	Exacerbation reduction in severe disease	Effect on weight or BMI in severe disease	Effect on quality of life (mean CFQ-R) in severe disease
<b>IVA</b>								
RAMSEY [6]	G551D/any; RCT	10.6% <sup>#</sup>	55% <sup>#</sup>	NA	NA	NA	NA	NA
DE BOECK [13]	Non-G551D gating/any; RCT	10.7% <sup>#</sup>	NA	0	NA	NA	NA	NA
HEBESTREIT [10]	G551D/any; open, EAP	NA	NA	14 (100%)	5.2±5.6 <sup>§</sup> ; median 3.9 [-4.1-16.8] <sup>*</sup>	NA	2.1±2.4 kg <sup>§</sup> ; median 1.1 [-0.3-6.3] kg <sup>*</sup>	NA
BARRY [9]	G551D/any; open, EAP	NA	NA	21 (100%)	4.2 <sup>§</sup>	49% <sup>§,†</sup>	1.8 kg <sup>§</sup> ; median 2.3 [-0.4-4.2] kg <sup>*</sup>	NA
TAYLOR-COUSAR [11]	G551D/any; open, EAP	NA	NA	44 (100%)	4.8±0.5 <sup>§</sup> ; [-13.1-22.7] <sup>*</sup>	NA	3.3±4.0 kg <sup>§</sup> ; [-2.3-14.4] kg <sup>*</sup>	NA
SALVATORE [12]	Non-G551D gating/any; open, EAP	NA	NA	13 (100%)	11.5 <sup>§</sup>	51% <sup>§,¶¶</sup>	3.0 kg <sup>§</sup>	NA
<b>LUM/IVA</b>								
WAINRIGHT [14] and ELBORN [15]	F508del/F508del; RCT, subgroup analysis	2.6-4.0% <sup>#</sup>	39-61% <sup>#</sup>	81 (7.3%)	3.3 [0.2-6.4]-3.7 [0.5-6.9] <sup>§,††</sup>	53-41% <sup>§,§</sup>	BMI 0.3 [-0.2-0.8] and 0.6 [0.1-1.2] kg·m <sup>-2§,††</sup>	3.3 [5.2-11.7] and -4.2 [-12-3.7] <sup>§,†††</sup>
HUBERT [16]	F508del/F508del; open	NA	NA	53 (37 completed 3 months)	3.19 <sup>§</sup>	NA	BMI 0.03 kg·m <sup>-2§</sup>	NA
TAYLOR-COUSAR [17]	F508del/F508del; open	NA	NA	46 (35 completed 24 weeks)	-0.4 [-1.9-1.1] <sup>§,††</sup>	64% <sup>§,¶¶</sup>	BMI 0.29 [0.17] kg·m <sup>-2§,††</sup>	2.5 [-1.0-5.9] <sup>††</sup>
MURER [18]	F508del/F508del; open	NA	NA	20 (10 completed)	+2.5 <sup>§#</sup>	60% <sup>§,¶¶</sup>	BMI 0.9 kg·m <sup>-2##</sup>	NA
DIAB-CACERES [19]	F508del/F508del; open	NA	NA	20		61% <sup>§,¶¶</sup>		
<b>TEZ/IVA</b>								
TAYLOR-COUSAR [21]	F508del/F508del; RCT	4.0 [3.1-4.8] <sup>§,††</sup>	36% <sup>§,†††</sup>	27 (9.4%)	3.5 [1.0-6.1] <sup>§,††</sup>	NA	NA	NA
ROWE [22]	F508del/RF; RCT (crossover)	6.8 [5.7-7.8] <sup>§,††</sup>	46%	22 (9%)	4.4 [1.1-7.8] <sup>§,††</sup>	NA	NA	NA
<b>ELX/TEZ/IVA</b>								
MIDDLETON [23]	F508del/MF; RCT	13.8 [12.1-15.4] <sup>§,††</sup>	63%	18 (9%)	15.2 [7.3-23.1] <sup>§,††</sup>	NA	NA	NA

Brackets indicate 95% confidence intervals, unless otherwise stated. RCT: randomised controlled trial; FEV<sub>1</sub>: forced expiratory volume in 1 s; BMI: body mass index; CFQ-R: Cystic Fibrosis Questionnaire-Revised; NA: not assessed; EAP: expanded access programme; RF: residual function CFTR mutation; MF: minimal function CFTR mutation. <sup>#</sup>: increase over placebo; <sup>§</sup>: mean±SD, change from baseline; <sup>\*</sup>: range, change from baseline; <sup>§</sup>: mean change from baseline; <sup>†</sup>: days with antibiotics for pulmonary exacerbation, annualised; <sup>##</sup>: median, change from baseline; <sup>¶¶</sup>: exacerbations per year per patient; <sup>††</sup>: least mean square.

Source based on: Shteinberg, M. and Taylor-Cousar, J.L. (2020) "Impact of CFTR modulator use on outcomes in people with severe cystic fibrosis lung disease," *European Respiratory Review*, 29(155), p. 190112. Available at: <https://doi.org/10.1183/16000617.0112-2019>. (Accessed: January 29, 2023)

The table, offered by Shteinberg et al., shows an overview of the comparison of the different above-mentioned studies all focusing on clinical trials including patients with a severely reduced pulmonary function.

Airway infections are a well-known and common complication in patients with CF. It is a result of abnormal ion transport, which leads to a higher viscosity and acidity in the airways surface liquid (ASL), which is included in the innate immune system and which therefore acts as a first line of defense within the respiratory system. The consequence is a thickened mucus, which accumulates on the epithelium and therefore impairs mucociliary clearance. For bacteria, this condition is the perfect environment, leading to severe airway infection with a subsequent hyperinflammatory state of the airway system. Following this, the lung function declines constantly, eventually leading to respiratory insufficiency preceding premature death.

Common bacteria colonizing the lungs of CF patients are rather opportunistic microbes such as *P. aeruginosa*, *S. aureus*, or *B. epacian*. In some data is shown that CFTR modulators might have a direct effect against bacteria, in particular against *P. aeruginosa* and *S. pneumonia*.

TEZ/IVA treatment in children proved to recover the bacterial composition to similar levels as healthy individuals. Additionally, a study called GOAL validated the hypothesis that IVA aids in *P. aeruginosa* clearance in patients who were positive for this bacterium, as they proved to be negative after IVA administration. It underlines the assumption that CFTR modulators can aid in reducing the rate of acute airway infections.

Since IVA contains a quinolone ring as a central structural component, it might have similar properties as quinolone antibiotics. To accentuate this, it has been shown that IVA has some bactericidal activity towards *Streptococci* and bacteriostatic activity towards *S. aureus*.

Furthermore, it was reported that the combination of CFTR modulators and antibiotics have synergistic effects in killing *P. aeruginosa* colonization, leading to a 100-fold decrease in bacterial counts. Also, Lumacaftor showed to have the ability in producing reactive oxygen species, aiding secondarily in the killing of bacteria. (22)

As a limitation of these hypotheses, it was reported that bacteria such as *P. aeruginosa* reduce the effectiveness of CFTR modulators by the production of certain molecules decreasing the activity of certain correctors. (21)

Overall, CFTR modulators presented to be successful in improving the general lung function of patients with CF in all age groups, by significantly increasing FEV<sub>1</sub> values and reducing pulmonary exacerbations, also associated with hospital admission and bacterial infections.

#### b. Effects on Gut Function

Since CFTR channels are abundantly present in the intestine, the gut is significantly affected in patients with Cystic Fibrosis. Especially children are affected by pancreatic dysfunction, causing inflammation, exocrine dysfunction, CFRD, as well as meconium ileus, intestinal obstruction, and liver and bile abnormalities. (3)

Patients may also suffer in a way that the microbiota is changed, leading to complications that are predominantly ruled by a dysfunction of the host immunity and metabolic capacity. Intestinal Inflammation, malignancy, irreversible fibrotic liver changes and airway colonization are a few to mention.

Biomarkers such as stool calprotectin and Pyruvate-Kinase M2 (M2-PK) are important factors to follow up when researching the effect of CFTR modulators regarding the intestinal function. In Cystic Fibrosis patients stool calprotectin is observed to be elevated as in other inflammatory bowel diseases or malignancies, as well as M2-PK.

According to Ooi, studies proved the significant reduction of calprotectin levels, after the introduction of IVA. This might be suggestive, that the intestinal inflammation in CF patients may overall improve or be reversible.

Furthermore, bacterial colonization within the intestine is important to observe when researching the impact of IVA on the diseased gut. CF patients are known to present with a higher colonization of the *Enterobacteriaceae* family, compared to healthy individuals. Particularly *E. Coli* bacteria dominate the colonization of the gut, having a significant impact on the alteration of the microbiome. Unfortunately, IVA presented to have only a weak influence on the reduction of the bacterial abundance.

The general changes in the gut microbiome due to modulation of intestinal CFTR function demonstrate the close connection of bacterial dysbiosis and intestinal inflammation.

Another microbe to focus on is *Akkermansia*. This is a Gram-negative bacterium living in the human intestinal mucous layer with the ability to degrade mucin. Its abundance is significantly correlated with a healthy gut mucosa. Consequently, its abundance is inversely related to intestinal inflammation. *Akkermansia* shows to have a positive influence in host mucosal anti-inflammatory pathways. IVA shows to increase the abundance of *Akkermansia* bacteria, among other things by increasing bicarbonate secretion by CFTR and therefore creating a supporting environment for the colonization of *Akkermansia*. (23)

Yet, no information about the effect of other CFTR modulators on the CF-diseased gut is known.

### c. Effects on Macrophage Function

Most clinical trials and reviews of CFTR modulators focus on the outcome and improvement of respiratory function, gut function or sweat chloride levels. However, the host immune system is affected by the therapy with CFTR modulators in a CF diseased patient as well. Especially macrophages show an important role in fighting chronic bacterial infections in Cystic Fibrosis. Macrophages are affected in a way that they produce an excessive amount of pro-inflammatory cytokines and therefore stimulate inflammatory processes. Additionally, they express a reduced number of CFTR on the cell surface, are more prone to early apoptosis and have a decreased function of phagocytosis.

A research of Zhang et. al shows the effects of CFTR modulators on CF monocyte-derived macrophages (CF MDM) and the related apoptosis rate, CFTR expression, polarization, and cytokine production. It claims that after treatment with IVA or LUM/IVA the absolute number of CFTR protein expression increases in CF MDM, but does not reach the levels of non-CF MDM. Furthermore, the study showed that the higher rate of apoptosis in CF MDM can be improved by administration of IVA or LUM/IVA, while the effect was significantly more impressive with the CF MDMs receiving IVA, compared to the ones receiving LUM/IVA.

Polarization of macrophages defines the ability to switch from one functional state to another. M1 is a phase where macrophages are in a pro-inflammatory state, whereas M2 macrophages are in a rather anti-inflammatory or pro-fibrotic state. This ability depends on certain stimuli. In CF macrophages, these phases seem to be different than in non-CF MDMs. The M2 phenotype is higher, and the M1 phenotype lower compared to non-CF MDMs, according to Zhang et al.

Patients treated with IVA express a polarization that is similar to MDMs of healthy individuals, whereas patients treated with LUM/IVA therapy only present different levels of M2 phenotype. Zhang et al. focused additionally on the phagocytosis ability of CF macrophages. This was shown to be reduced in CF MDMs, together with a decreased killing of *C. cenocepacia* and *P. aeruginosa* bacteria. After treatment with IVA monotherapy, the phagocytosis levels of CF MDMs increased, as well as the phagocytosis of *C. cenocepacia* bacteria. The overall load of *P. aeruginosa* seems to get reduced for about 89% after IVA monotherapy. LUM/IVA therapy does not lead to an increase of *C. cenocepacia* phagocytosis and only to a non-significant reduction of *P. aeruginosa* phagocytosis, according to the beforementioned study.

Furthermore, the cytokine production in CF MDMs differs from non-CF MDMs. Measured in the supernatant, the baseline levels of CF- and non-CF MDMs showed to be similar, but after an infection with *C. cenocepacia*, the cytokine levels in CF patients differ. IL-6, TNF- $\alpha$ , IL-10, IL-12, and IL- $\beta$  levels increased significantly in CF-MDMs. After concomitant treatment with IVA monotherapy, IL-6, TNF- $\alpha$  and IL-12 cytokine production was decreased, while LUM/IVA therapy only changed the production of IL-6 cytokine. Systemically measured, less significant measures were achieved. Except an increased concentration of IL-8 at baseline and after CFTR modulator therapy and an increased IL-12 production after IVA therapy was no difference mentioned.

To conclude, the study of Zhang et al. proves that CFTR levels are reduced in CF MDM, which increase after CFTR modulator administration. An increase in CFTR expression is associated with a decrease in apoptosis of the mentioned cells, which suggests the claim that CFTR expression is correlated with cell stability.

Response of M2 activation is not increased by CFTR modulators, which might be caused by a loss of plasticity or a persistent M2 state of certain CF MDMs. MDMs with reduced M1 phenotypes at baseline show to be responsive to IVA administration. The phagocytosis ability of CF MDM is increased after IVA therapy, but not with LUM/IVA treatment. In fact, according to Zhang et al. it gets even more reduced in LUM/IVA treatment. (24)

#### d. Ivacaftor

Up to now, Ivacaftor is the only FDA approved CFTR potentiator. It was the first ever approved drug, which treated the basic defect of CF, declared in 2012. (7)

IVA is approved for the treatment of class III mutations, in which primarily the channel gating is dysfunctional, leading to an up to 100 times lower open probability. Unfortunately, this class of defects is only present in around 7% of the CF population. (25) The most common mutation in this class is the G551D mutation, where the amino acid glycine is substituted by aspartate at position 551 in the NBD1 of the CFTR gene. G551D is the 3<sup>rd</sup> most common CF-causing mutation, making up to 4% of all patients suffering from CF. (19) Unfortunately, this mutation is commonly associated with a severe phenotype, presenting with severe pulmonary dysfunction and pancreatic insufficiency. Especially in northern and central European countries this mutation occurs the most. (5)

IVA is indicated as monotherapy for patients older than 6 years and weighing more than 25kg presenting with G551D, R117H, G1244E or other CFTR gating channel mutations.

Additionally, IVA is also given in combination with other CFTR modulators, such as TEZ, LUM or ELX, as written below.

Since IVA is a CFTR potentiator, it increases gating of the protein located on the apical cell membrane. Hereby it enhances the chloride transport in proteins having reduced channel-opening probability, such as proteins with a G551D mutation. Also, in R117H mutations IVA is efficient, where not only the gating of the protein is dysfunctional, but also the conductance is reduced. (26) For the high efficacy of the drug, it is speculated that the mechanism of action is the targeting of the PKA-phosphorylated CFTR. Binding seems to induce an ATP-independent channel opening associated with hydrolysis, leading to an increased chloride secretion within the respiratory epithelium. Additionally, the drug decreases the amiloride-sensitive current and the absorption of fluid within the diseased epithelium. Subsequently, the cilia show an increase in rhythm frequency. (19)

Ivacaftor (Kalydeco ®) is available as 75mg or 150mg film-coated tablets. For the IVA monotherapy, it is recommended to take one 150mg tablet twice daily, with 12h apart from each other together with fat-containing food. This is the indication for all patients older than 6 years and weighing more than 25kg. (26) IVA is also approved for patients older than 2 years, while children between 2 and 6 years are recommended to take one tablet of 50mg, or 75mg when weighing more than 14kg every 12 hours also together with fat-containing food.

The exposure of IVA increases every 12 hours from 25mg to 450mg every 12 hours. The exposure additionally increases, when administering IVA together with fat-containing food.

When distributed, the drug is primarily bound to plasma proteins, particularly alpha 1-acid glycoprotein and albumin to mention. The metabolization occurs to the majority by CYP3A enzyme, where M1-IVA and M6-IVA are produced as major metabolites. Here, M1 is the pharmacologically active product. 87% of IVA is excreted via feces, where 65% of it consist of metabolites.

Since IVA is a substrate of CYP3A, coadministration of inducers or inhibitors of this enzyme is not recommended. It could lead to the loss of IVA efficacy.

Very common AE are upper respiratory tract infections and nasopharyngitis, as well as headache, dizziness, oropharyngeal pain, abdominal pain, diarrhea, and transaminase elevations, as well as rash. As common AE are classified symptoms of influenza, rhinitis, hypoglycemia, any type of ear symptoms, abnormal breathing, flatulence and nausea, Aminotransferase elevations and pruritus. Uncommon AE are ear congestion, wheezing, breast symptoms and increased blood pressure. (26)

The efficacy of IVA was also tested in patients homozygous for a class II mutation. Due to the fact, that the F508del mutation does not only lead to a dysfunction in trafficking, but also in the overall function of the protein and the degradation time of the protein, this mutation was thought of to be also accessible by a CFTR potentiator. *In vitro* studies showed some activity of the drug in those mutations. However, no significant improvement of FEV<sub>1</sub> or reduction of pulmonary exacerbations could be associated with a regular administration of IVA monotherapy. (5)

In patients positive for at least one allele of a G551D mutation presented with an improved lung function, reduced levels of sweat chloride and a significant increase in the CFQ-R scores, which represent the improvement of health-related quality of life.

The lung function is the primary focus in most studies, measured in FEV<sub>1</sub>, showing a statistically significant treatment effect within a noteworthy period of time. The so-called STRIVE study, mentioned by Sermet-Gaudelus claimed the efficacy of IVA within only 15 days of administration, while showing maintenance afterwards. The channel function shows overall improvement, measured within the nasal and sweat gland epithelium. (5)

Additionally, it was proven that patients with poor pulmonary function showed similar improvements as patients with only mild pulmonary dysfunction.

Pulmonary exacerbations are significantly reduced by the administration of IVA, which in addition to the before mentioned effects of the drug support the hypothesis that an increase in CFTR function helps in intensification of airway clearance.



Patients in clinical trials of IVA administration also mentioned an improvement of the overall nutritional status, measured as BMI. (5)

According to Sermet-Gaudelus, clinical trials (n= 171) with the given dose of 150mg IVA for 48 weeks showed that the overall pulmonary function, measured in FEV<sub>1</sub> increased from baseline for +10,5 percentage points, compared to the placebo group. Additionally, it is mentioned that the drug-induced improvement was already showing after 15 days of administration.

Sermet-Gaudelus also reviewed about a clinical trial done in a pediatric population (n= 52), where a change of 10% in FEV<sub>1</sub> was achieved after 48 weeks of drug administration.

Important to mention is, that in this study the improvements in patients with rather poor pulmonary function were comparable to that of patients with rather mild functional impairment.

Furthermore, the rate of pulmonary exacerbations is described to be reduced by 55% in the adult patient group, as well as an increase in weight gain was noted in both studies.

An additional secondary endpoint in the clinical trials of IVA was the sweat chloride concentration. This was mentioned to be reduced by 48,1mmol/L<sup>-1</sup> when compared to placebo in the adult study. In the pediatric trial group, sweat chloride levels were reduced with a treatment difference of -53mmol/L<sup>-1</sup>.

IVA is described as to be overall safe, while the interruption rate of the clinical trials due to severe AE was low (13% in the adult group vs 6% in the children group). The reason for interruptions was elevated hepatic enzyme levels. (5)

#### e. Lumacaftor-Ivacaftor

LUM/IVA is a drug combination consisting of Lumacaftor and Ivacaftor. Lumacaftor is a CFTR channel corrector, and Ivacaftor a potentiator. The combination of these two drugs was approved in 2015, 3 years after the approval of Ivacaftor monotherapy. Since Lumacaftor monotherapy showed no significant improvements in pulmonary function or sweat chloride reduction, clinical trials focused on the combination of LUM and IVA, especially in patients with two positive alleles for the F508del mutation. (25)

LUM/IVA is indicated in patients aged more than 6 years and homozygous for the F508del mutation. It is not efficient in F508del minimal function mutations or heterozygosity.

As IVA improves the channel opening probability, LUM as a CFTR corrector is a synergistic partner. It aids in processing and trafficking of CFTR and therefore increases the number of CFTR proteins on the apical cell membrane. The exact mechanism of how these drugs work is

yet unknown and to be determined, but together they increase the quantity and quality of CFTR channels and therefore improve the chloride transport.

LUM/IVA (Orkambi®) is given in form of 2 tablets every 12 hours. In patients aged 6-11 years, 2 tablets of Lumacaftor 100mg and Ivacaftor 125mg should be taken each morning and each evening. This makes up a total daily dose of 400mg Lumacaftor and 500mg Ivacaftor. Patients aged 12 years and older are recommended to take 2 tablets of Lumacaftor 200mg and Ivacaftor 125mg every 12 hours, which makes up a total dose of 800mg Lumacaftor and 500mg Ivacaftor. Furthermore, this drug combination should be taken together with a fat-containing meal, to achieve the best possible absorption rate. (27)

Regarding the pharmacokinetics of LUM/IVA, important to reiterate is that Lumacaftor is a strong inducer of the Cytochrome P450 3A (CYP3A), while Ivacaftor is metabolized by this enzyme. This leads to a decreased exposure of Ivacaftor in a dose-dependent manner. Furthermore, Lumacaftor induces other enzymes such as CYP2B6, CYP2C8 or CYP2C9, which proves to increase the risk for drug interactions immensely. Consequently, dose adjustments of other drugs that get metabolized by these enzymes need to be considered when co-administering. (25)

Very common AE are nasopharyngitis, headache and dizziness, nasal congestion, dyspnea, cough and other respiratory system symptoms, as well as abdominal pain, diarrhea and nausea. Common AE include upper respiratory tract infections, ear symptoms, pharyngeal erythema, abnormal respiration, flatulence and vomiting, transaminase elevations and abnormal menstruation cycles with dysmenorrhea. Uncommon AE are hypertension, hepatic encephalopathy and cholestatic hepatitis. (27)

Lumacaftor in monotherapy did not achieve satisfying results in clinical trials. It showed a change in ppFEV<sub>1</sub> of only +2,5 – 4,1%.

Overall, clinical studies according to Kuk and Taylor-Cousar showed significant changes in sweat chloride secretion, but only modest improvement of lung function. Phase III studies of the TRANSPORT and TRAFFIC studies tested the combination of LUM/IVA in 1122 randomized patients and compared the drug to placebo. The observed mean absolute change in FEV<sub>1</sub> % showed an improvement of +2,6-4%. This improvement occurred already after 15 days and maintained until the end of the study (24 weeks). The pulmonary exacerbation rate reduced to about 30% with a daily dose of 600mg Lumacaftor and 39% with 400mg Lumacaftor dose. Additionally, significant improvements were seen according to BMI. (25)

LUM/IVA was the first drug that was approved for the majority of CF patients, by addressing the F508del mutation. Therefore, this was an important step in the development of further

CFTR modulator research. However, the outcomes it achieved were only small and are nowadays obsolete by other combination therapies, such as ELX/TEZ/IVA.

f. Tezacaftor-Ivacaftor

TEZ/IVA is a drug combination consisting of a CFTR corrector and a CFTR potentiator. This drug is indicated in patients aged 6 years or older, having a homozygous F508del mutation or being heterozygous with one F508del mutation and one other mutation, for example P67L or R117C.

Since TEZ is a CFTR corrector, it aids in processing and trafficking of the CFTR protein within the cell. For that, it binds selectively to the first Membrane Spanning Domain (MSD-1) of the CFTR gene. Hereby, it increases the quantity of proteins sent to the apical cell surface.

IVA is a CFTR potentiator, functioning on the CFTR proteins, which were transported towards the cell surface with the aid of TEZ and improves their open probability.

TEZ/IVA is administered orally in form of tablets. For children aged 6-12 years weighing less than 30 kg, it is recommended to take one tablet of TEZ/IVA in the morning with 50mg TEZ and 75mg IVA dosage. For children aged older than 6 and weighing more than 30kg and for patients older than 12 it is suggested to take one pill containing 100mg TEZ and 150mg IVA in the morning. Additionally, all patients should take one pill of IVA in the evening, with the same dosage as their morning combination pill is containing. Since the Area Under the Curve (AUC) of IVA increases significantly when administering it together with fat-containing food, the combination drug in the morning, as well as IVA alone in the evening should be given together with fatty food.

Within 4 hours, TEZ gets absorbed to its maximum concentration ( $t_{max}$ ). IVA gets absorbed in a median time of about 6 hours ( $t_{max}$ ). Both constituents of the drug are 99% bound to plasma proteins, TEZ primarily to albumin and IVA to alpha 1-acid glycoprotein and albumin. The metabolization of TEZ occurs primarily by CYP3A4 and CYP3A5 enzymes.

Three metabolites are produced, while one is completely inactive, one minimally active, and one is considered pharmacologically active. This pharmacologically active metabolite is called M3-TEZ.

IVA is also metabolized by CYP3A4 and CYP3A5, while M1-IVA and M6-IVA are the important metabolites to mention, M1-IVA being pharmacologically active. The excretion of TEZ/IVA occurs primarily after 167 hours for TEZ and after 9,3 hours for IVA. Both excretions occur primarily via feces.

It is advised to adjust the dose of TEZ/IVA when co-administering it with other CYP3A inhibitors, for example Fluconazole, Erythromycin or Verapamil, which are moderate CYP3A inhibitors. Strong CYP3A inhibitors are for example Ketoconazole and Clarithromycin.

Clinical trials (n(adults) = 1042, n(children) = 130) showed that the drug TEZ/IVA is considered as safe. Very common side effects included headache and nasopharyngitis, upper respiratory tract infections, dizziness, rash, abdominal pain and diarrhea, as well as transaminase elevations. Common and rather uncommon AE were rhinitis, ear pain and discomfort, nausea and breast mass. (28)

g. Elexacaftor/Tezacaftor/Ivacaftor

ELX/TEZ/IVA (Trikafta®, Vertex Pharmacology) is a combination of two correctors and one potentiator. The correctors are TEZ and ELX, which act directly on the F508del-CFTR polypeptide. The combined CFTR potentiator is IVA. (7)

The drug is approved in patients with one copy of F508del mutations and is indicated in patients older than 6 years.

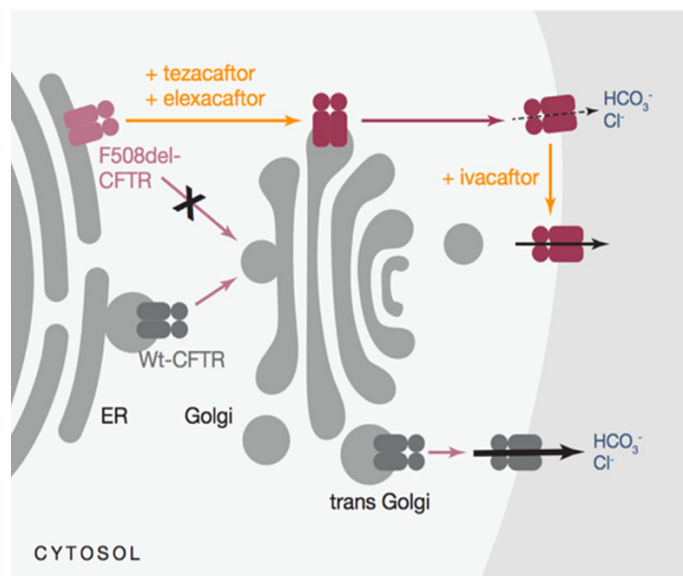


Figure 2: Mechanism of Action of ELX/TEZ/IVA on a cellular level

Source: Anas Z, ElSaygh J, ElSORI D, et al. A review of Trikafta: Triple Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) Modulator Therapy. Juli 03, 2021 [Internet]. [accessed November 19, 2022]; Cureus 13(7): e16144. DOI 10.7759/cureus.16144 Available from [https://www.researchgate.net/publication/352952690\\_A\\_Review\\_of\\_Trikafta\\_Triple\\_Cystic\\_Fibrosis\\_Transmembrane\\_Conductance\\_Regulator\\_CFTR\\_Modulator\\_Therapy](https://www.researchgate.net/publication/352952690_A_Review_of_Trikafta_Triple_Cystic_Fibrosis_Transmembrane_Conductance_Regulator_CFTR_Modulator_Therapy)

The figure demonstrates the mechanism of action of the combination drug ELX/TEZ/IVA. TEZ and ELX, as CFTR correctors potentially support the CFTR protein in trafficking and transport towards the apical cell membrane, while IVA is essential in establishing a functional channeling of the protein, so that bicarbonate and chloride ions can be excreted in a sufficient manner.

Together they increase the amount and function of CFTR channels on the apical cell surface of epithelial cells mainly within the lung, the pancreas, and the intestine.

ELX/TEZ/IVA (Kaftrio®) / Trikafta®) is given in a fixed manner of dosage. In children 6 to 12 years old, weighing less than 30kg, two tablets are given in the morning, containing 37,5mg IVA, 25mg TEZ and 50mg ELX. In the evening, one tablet of 75mg IVA is administered.

In children 6-12 years old, weighing more than 30kg, two tablets are given in the morning, containing 75mg IVA, 50mg TEZ and 100mg ELX. Additionally, one tablet of IVA 150mg is administered in the evening. The same is implied in patients older than 12 years. The drug administration should be 12 hours apart from each other, while it is suggested to take the medication together with fatty meals. (29)

Pancreatic-insufficient patients are advised to additionally take pancreatic enzymes for an optimal efficacy. (7)

After the first administration of ELX/TEZ/IVA, the plasma concentration is reached in 7, 8 and 3-5 days, respectively. The bioavailability of ELX in oral administration is 80%. Its maximal concentration ( $t_{max}$ ) is reached within 6 hours, while  $t_{max}$  of IVA is reached within 3 hours. The bioavailability of ELX increased 1.9 – 2,5-fold when administered together with fatty food, compared to a sober circumstance. The bioavailability of IVA increases to the 2,5 – to 4-fold, when administered together with fatty food. The bioavailability of TEZ cannot be influenced by food.

99% of ELX and TEZ is bound to plasma proteins during distribution, primarily to albumin. Also the majority of IVA is bound to plasma proteins, but in comparable amounts to albumin and alpha-1-acid glycoprotein and human gamma-globulin. (30)

AE of ELX/TEZ/IVA are divided into 3 classes. Very common AE are infections of the upper respiratory tract, headache and dizziness, oropharyngeal pain and nasal congestion, diarrhea, abdominal pain, transaminase elevation, rash and sputum positive for bacteria. Common AE are rhinitis and influenza infection, ear pain and discomfort, rhinorrhea and abnormal breathing, nausea, abdominal pain, flatulence, acne and pruritus, breast mass and an increase in blood creatine phosphokinase. Uncommon AE would be ear congestion, wheezing, breast inflammation, gynecomastia and arterial hypertension. (29)

According to Zaher et. al, it was shown by clinical trials that ELX/TEZ/IVA enables an improvement of overall lung-function, as well as in the quality of life of diseased patients.

Additionally, also the pulmonary exacerbation rate decreased, despite of a decrease in sweat chloride.

The European Medicine Agency (EMA) published a study, n(=403), which was randomized, double-blind and placebo-controlled. It focused on patients with an F508del mutation on one allele and a minimal function mutation on the remaining allele. All patients were 12 years and older and had an average baseline ppFEV<sub>1</sub> of 61,4%. The result was very positive. The ppFEV<sub>1</sub> increased up to +14,3 percentage points, the sweat chloride levels reduced by -41,8 percentage points and the BMI changed with 1,04 kg/m<sup>2</sup>.

Another study published by EMA n(=258) was focusing on patients older than 12 years, heterozygous for the F508del mutation and a mutation on the second allele with a gating defect or residual CFTR activity. The study was double-blind, randomized, and active-controlled. The patients received either IVA or TEZ/IVA in combination with IVA, while being in a 4-week open label run-in period. Afterwards they got randomized to receive either ELX/TEZ/IVA in combination with IVA or remained on the first applied drug. Compared to the control group, the ELX/TEZ/IVA group n (=132) improved their ppFEV<sub>1</sub> by +3,7 percentage points, while the sweat chloride value reduced by -59,5 percentage points. (29)

To summarize, the approval of ELX/TEZ/IVA in 2019 did an immense change within the therapy options for patients with class II mutations of cystic fibrosis. It significantly improves the pulmonary function tests of affected patients, consequently increasing their quality of life and diminishing the pulmonary exacerbations with subsequent hospital admissions. (7)

Up to now, the safety and efficacy for children younger than 6 years is not known.

#### h. General Approach

The first decision if a patient is approved for the treatment with CFTR modulators depends on the patient's age and genotype. For patients presenting with genotypes that are qualified for more than one certain therapy, it is suggested on the regimen with the greatest number of modulators. For instance, a triple combination therapy with ELX/TEZ/IVA. If a patient is not qualified for a certain therapy due to a young age, it is suggested to wait for that therapy until meeting the required age.

Patients presenting with F508del mutations need to be classified in being genotypically homo- or heterozygous.

In patients with a F508del homozygous mutation being older than 6 years, ELX/TEZ/IVA is preferred over a dual therapy. Both treatment regimens are efficient, but in certain studies, ELX/TEZ/IVA showed more impressive results regarding an improvement of FEV<sub>1</sub> and the symptom related QOL in a time period of 4 weeks. In patients younger than 6 years with the

same genotype, the dual therapy of lumacaftor-ivacaftor shows to be the most efficient. Yet, ELX/TEZ/IVA is not approved for children under six years, while LUM/IVA is approved from the age of 2 years. When reaching the age of 6 years, the regimen can be switched to ELX/TEZ/IVA.

In patients with a F508del heterozygous mutation being older than 6 years ELX/TEZ/IVA is recommended. In this case, the second mutation does not constitute to the treatment decision, as long as one mutation being F508del. In patients younger than 6 years and older than 4 months, IVA Monotherapy is the suggested treatment regime, with the requirement of the responsiveness to IVA of the second mutation.

Patients presenting with one of the other 180 approved mutations for CFTR modulator therapy are recommended to start with the maximal therapy that is approved and available for their age group. Usually, this is to start with ELX/TEZ/IVA.

Patients can also present with non-approved mutations. Here, the use of CFTR modulators is recommended to be reduced to the minimum, which is the participation in clinical trials. (8)

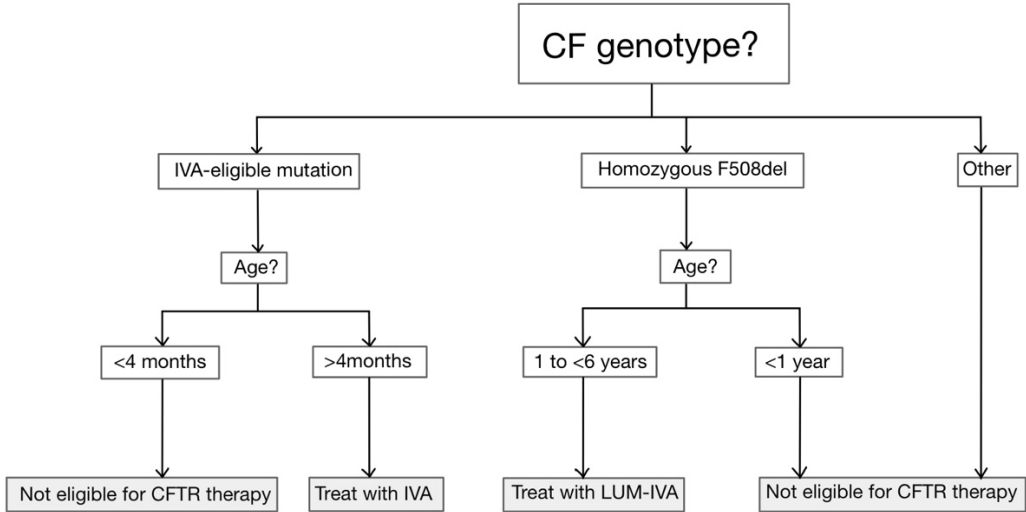


Figure 3: Selection of CFTR modulator Therapy

Source based on: Simon, R.H. (no date) *Cystic Fibrosis: Treatment with CFTR modulators*. UpToDate. Available at: <https://www.uptodate.com/contents/cystic-fibrosis-treatment-with-cftr-modulators#H2400702249> (Accessed: November 23, 2022).

## 5. Discussion

This review paper reports the development and effectiveness of CFTR modulators as the first causal therapy of Cystic Fibrosis. Within 10 years, 4 different medicamentous therapy options were approved by FDA, so that 90% of CF patients can benefit of a gene modulator therapy with significant benefits.

Ivacaftor, being the first CFTR modulator to be developed in 2012 is addressing the mutations affecting the channeling of CFTR proteins. The most common mutation in this class III is G551D, affecting around 4% of CF patients. (19) The development of IVA was paving the way to further medications being developed, also addressing patients with different mutation classes so that the majority of CF patients could benefit from the newly developed drug invention.

LUM/IVA was the first combination therapy, consisting of Ivacaftor and Lumacaftor and is indicated in patients homozygous for F508del mutations. Second being a CFTR corrector did not convince in monotherapy but showed significant effects in sweat chloride concentration reduction when combining it with Ivacaftor. Although, it was a great innovation and the right therapy for some patients, it did not achieve the revolutionary results that IVA achieved in patients with G551D mutations. Additionally, it is not effective in patients with only one F508del mutation and several drug-drug interactions make the drug to be used with caution. (27)

However, further research found the correctors Tezacaftor and Elexacaftor, which showed especially in a triple combination with Ivacaftor an even greater effect according to lung function, exacerbation reduction and sweat chloride change. Also, the BMI and quality of life changed in a significant positive way. (7)

Even though the invention of CFTR modulators was groundbreaking and a huge step for the therapy options of CF patients, there are still some limitations.

First, 10-15% of CF patients are still not able to be treated with CFTR modulators, due to the reason of having a mutation not accessible for the therapy with one of the approved modulators. These include mutations with a premature termination codon, frameshift and deletion mutations, canonical splice mutations and several missense mutations. (31)

Second, the treatment with CFTR modulators is very expensive. Yet not everyone in need of a therapy with CFTR modulators has the access and the availability to the proper treatment option. A therapy with CFTR modulators is extremely expensive. In 2018, the annual price for Kalydeco was 310000\$, for Orkambi 259000\$ and for Symdeko 292000\$. Until now, Vertex is the only pharmaceutical company producing approved CFTR modulators, which may partly



explain the high prices. A company, called Galapagos, a partner of AbbVie is currently in the process of developing new CFTR modulator products. (10)

For the 10-15% of patients with non-accessible CFTR mutations, there are several variant options for an alternative treatment in development, but presently CFTR modulator therapy is the only market-approved causal treatment for CF.

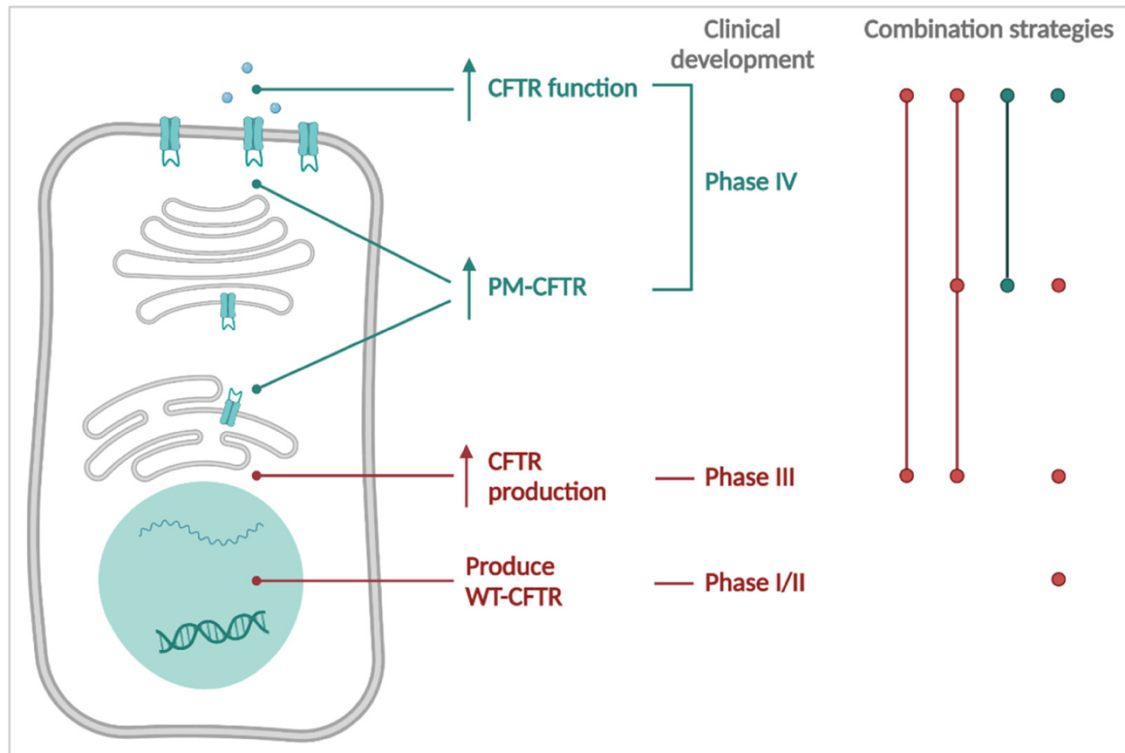


Figure 4: Summary of therapeutic strategies for causal CF therapy and their actual progress.

Source: Ensinck, M.M. and Carlon, M.S. (2022) "One size does not fit all: The past, present and future of Cystic Fibrosis Causal Therapies," *Cells*, 11(12), p. 1868. Available at: <https://doi.org/10.3390/cells11121868>. (Accessed: March 13, 2023).

The picture shows the strategy of several possibilities to increase the functional number of CFTR proteins on the cell surface, as well as its strategic combination options.

Lubiprostone is a drug that is initially indicated in patients with chronic constipations, but when combined with ELX/TEZ/IVA, it was reported to achieve enhanced F508del rescue in CF cells in the airway epithelium. The drug aids in stimulation of fluid secretion by the modulation of cAMP levels via prostaglandin receptor EP4 and consequent activation of CFTR channels. Other drugs that have the ability to enhance the flux of chloride within the CFTR channels by increasing cAMP are Forskolin and IBMX. These drugs belong to a class called CFTR activators.

Another class of drugs is called CFTR potentiators, which interact directly with CFTR. Deutivacaftor being one of those, is currently tested in clinical trials phase 3. (31)

Co-Potentiators include Genistein. It was shown that the combination of ivacaftor as a potentiator with genistein as a co-potentiator has a synergistic effect and improves the CFTR functional rescue in gating mutations. It might also be useful in mutations yet not accessible with current available modulators. Also, the relatively new modulator Elexacaftor shows to have potentiator- and corrector characteristics, which depends on the studied mutation. New studies and results are being expected here as well.

The company Galapagos is currently in the process of development of new CFTR modulators and other drugs. The approval of these drugs might help making CF medications available and accessible for all CF patients in every financial situation and with any CFTR mutation.

With under these modulators are two correctors. Type 1 correctors are directing the NBD1 membrane spanning domains interface, while type 2 correctors are choosing the NBD2. The type 2 corrector ABBV/GLPG2737 has the ability to save the V232D-CFTR gene, which describes a rare mutation causing misfolding of the protein and therefore leads to an dysfunctional channel maturation. Galicaftor (ABBV/GLPG2222) is the name of the type 1 corrector. This medication focuses on several rare CFTR mutants. Towards the mutation variants of E92K-, P67L- and V232D-CFTR, it acts similar as the Vertex compound Lumacaftor, notwithstanding with a lower potency. These drugs by Galapagos have an improved activity when combined, especially when applied in the F508del mutation.

Another CFTR modulator by Galapagos is GLPG3067, acting as a CFTR potentiator. The triple combination of GLPG2222, GLPG2737 and GLPG3067 considerably rises the chloride transport along the CFTR proteins compared to monotherapy.

GLPG2737 has already passed the safety assessment in healthy volunteers within clinical trials and was also already evaluated in F508del homozygous patients. Here it is presently tested as an add-on therapy for patients using LUM/IVA. (32)

Other compounds of AbbVie/Galapagos in recent studies on airway cells include drugs that target mutations that are rarer and focus on AC1 and type 2 correctors, called AC2-1 and AC-2. The I1234\_R1239del mutation is very rare, while pre-treatment with the just mentioned novel correctors in monotherapy ended in significant improvements in channel action. The greatest results were achieved in the combination therapy of AC1 and AC2-2.

Another company that might be a future concurrent for Vertex Pharmaceuticals is Proteostasis Therapeutics Inc. This company is developing a triple combination including a potentiator called Dirocaftor, a corrector Posenacaftor and Nesolicaftor. This combination is classified as a CFR amplifier and promotes CFTR protein synthesis in an agnostic way. The drug Nesolicaftor has already been tested in F508del CF patients in phase 1 and phase 2 clinical

trials. Here, according to Mergioti, an 8% ppFEV1 improvement, as well as a -29mmol/L decrease in sweat chloride in just 4 weeks of treatment compared to placebo was reported. (32) A lot did happen in the last 10 years when observing the development of the therapy in Cystic Fibrosis patients. A whole new therapy option has developed, and not only new medications are regularly approved by FDA, but also already approved medications are optimized constantly, to be accessible for the broadest CF population possible. As Lumacaftor was available only for patients older than 12 years when first approved, it is now also available for patients older than 6 years old.

Since more pharmaceutical companies are nowadays working on the development of new medications, the process will exponentially develop and change in the near future. There is hope for the accessibility and availability of appropriate CFTR therapy for the broad CF population, even for those with severe disease, financial instability and rare mutations.

## **6. Conclusion**

To conclude, CFTR modulators are a great possibility for CF patients to get appropriately treated. The best beneficial effect is achieved in patients having one gating mutation, treated with Ivacaftor and patients with F508del mutations being homo- or heterozygous and receiving the triple combination of Elexacaftor, Tezacaftor and Ivacaftor. Primary and secondary endpoints showed the best results in the above-mentioned patient groups, when comparing all clinical trials and studies. When focusing on the F508del homozygous CF patients, TEZ/IVA and ELX/TEZ/IVA showed better results when compared with LUM/IVA. Here important to mention is also the safety, since LUM/IVA has a large number of drug-drug-interactions and might to be taken with caution in patients with comorbidities and polypharmacy. (28)

The overall safety is good in all CFTR modulators, while adverse effects are similar in all 4 medications and the overall cancellation rate in the clinical trials was insignificant, with exception of LUM/IVA.

Yet, 10-15% of CF patients do not have access to CFTR modulators, which is why a lot of research and development for further inventions is ongoing. Furthermore, other companies than Vertex Pharmaceuticals are processing new treatment options for making CFTR treatment possibilities available and accessible for all CF patients.

## **7. Summary**

CFTR modulators have been a groundbreaking new invention starting with the approval of the CFTR corrector Ivacaftor in 2012. Nowadays, up to 90% of CF patients have the potential ability to get treated by an appropriate CFTR modulator.

Ivacaftor showed significant improvement in the treatment of CF patients with gating mutations, especially in G551D mutations. The combination of Elexacaftor, Tezacaftor and Ivacaftor showed tremendous benefits because it is appropriate for a broad spectrum of variant mutations, importantly to mention being effective in F508del mutations no matter if homozygous, heterozygous or with a minimal function mutation.

An early administration of CFTR modulators reduces the severity and extent of the clinical picture, which underlines the necessity and importance of the neonatal CF screening.

A lot has been achieved up to now, and a lot is yet to come. The range of accessible CFTR modulators will be increased in the near future, since new medications are in development and already in the last phases of clinical trials. The goal is to make therapy accessible and available for all cystic fibrosis patients to achieve equality and to reduce mortality and morbidity of this lethal genetic disease.

**Keywords:** CFTR modulators, Cystic Fibrosis, Ivacaftor, G551D mutation, F508del mutation, CFTR correctors, CFTR potentiators

## 8. Literature

- (1) Sibley CD, Rabin H, Surette MG. Cystic fibrosis: A polymicrobial infectious disease. *Future Microbiology*. 2006 Jun 1; [Internet] 1 (1): 53–61. [accessed November 20, 2022] Available from: <https://pubmed.ncbi.nlm.nih.gov/17661685/>
- (2) Rafeeq MM, Murad HA. Cystic fibrosis: Current therapeutic targets and future approaches. *Journal of Translational Medicine*. [Internet] 2017 Apr 27; 15(1). [accessed November 20, 2022] Available from: <https://translational-medicine.biomedcentral.com/articles/10.1186/s12967-017-1193-9>
- (3) B., D. and E., M. (2013) “Cystic fibrosis: Does cftr malfunction alter PH regulation?,” *Genetic Disorders* [Preprint]. Available at: <https://doi.org/10.5772/52342>. [Accessed: March 27, 2023]
- (4) Yankaskas JR, Marshall BC, Sufian B, Simon RH, Rodman D. Cystic Fibrosis Adult Care Chest [Internet]. 2004; January 01, 2004 [accessed November 17, 2022]; 1S-39S Available from: [https://journal.chestnet.org/article/S0012-3692\(15\)32218-2/fulltext](https://journal.chestnet.org/article/S0012-3692(15)32218-2/fulltext)
- (5) Sermet-Gaudelus I. Ivacaftor treatment in patients with cystic fibrosis and the G551D-CFTR mutation. *European Respiratory Review* [Internet]. January 07, 2013 [accessed November 17, 2022]; 22: 66-71 Available from: <https://err.ersjournals.com/content/22/127/66>
- (6) Derichs N. Targeting a genetic defect: cystic fibrosis transmembrane conductance regulator modulators in cystic fibrosis, *European Respiratory Review* 22: 127, 58-65 [Internet] January 12, 2013 [accessed November 19, 2022]; Available from: <https://err.ersjournals.com/content/22/127/58>
- (7) Anas Z, ElSaygh J, ElSORI D, et al. A review of Trikafta: Triple Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) Modulator Therapy. Juli 03, 2021 [Internet]. [accessed November 19, 2022]; *Cureus* 13(7): e16144. DOI 10.7759/cureus.16144 Available from [https://www.researchgate.net/publication/352952690\\_A\\_Review\\_of\\_Trikafta\\_Triple\\_Cystic\\_Fibrosis\\_Transmembrane\\_Conductance\\_Regulator\\_CFTR\\_Modulator\\_Therapy](https://www.researchgate.net/publication/352952690_A_Review_of_Trikafta_Triple_Cystic_Fibrosis_Transmembrane_Conductance_Regulator_CFTR_Modulator_Therapy)

- (8) Simon, R.H. (no date) *Cystic Fibrosis: Treatment with CFTR modulators*, UpToDate. [Accessed: November 23, 2022]. Available at: <https://www.uptodate.com/contents/cystic-fibrosis-treatment-with-cftr-modulators#H2400702249>
- (9) Bergeron C, Cantin AM. New Therapies to Correct the Cystic Fibrosis Basic Defect. *Int J Mol Sci*. [Internet]. 2021; June 08, 2021 [accessed November 19, 2022]; PMID 34201249 Available from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8227161/>
- (10) Burgener EB, Moss RB. Cystic Fibrosis Transmembrane Conductance Regulator Modulators: Precision Medicine in Cystic Fibrosis. *Current Opinion in Pediatrics*. 2018; 30(3): 372-7. [Internet]. [accessed November 20, 2022] Available from: <https://pubmed.ncbi.nlm.nih.gov/29538046/>
- (11) Cuevas-Ocana S, Laselva O, Avolio J, et al. The era of CFTR modulators: improvements made and remaining challenges. *Breathe* 2020; 16: 200016 [Internet]. [accessed November 19, 2022] Available from: <https://breathe.ersjournals.com/content/16/2/200016>
- (12) Symdeko approved for CF patients with certain genetic mutations [Internet]. *Cystic Fibrosis News Today*. 2021 [accessed November 20, 2022]. Available from: <https://cysticfibrosisnewstoday.com/symdeko-tezacaftor-ivacaftor-combo-therapy/>
- (13) Shaughnessy CA, Zeitlin PL, Bratcher PE. Elexacaftor is a CFTR potentiator and acts synergistically with ivacaftor during acute and chronic treatment. *Scientific Reports*. [Internet]. 2021 Oct 6; 11 (1). [accessed November 20, 2022] Available from: <https://www.nature.com/articles/s41598-021-99184-1>
- (14) Welsh, MJ, Tsui, L-C, Boat, T, Beaudet, AL. Cystic fibrosis. In: *The Metabolic and Molecular Basis of Inherited Disease*, Scriver, CR, Beaudet, AL, Sly, WS, et al (Eds), McGraw-Hill, New York, 1995, p. 3801.
- (15) Fohner, A.E. *et al.* (2017) “Pharmgkb summary,” *Pharmacogenetics and Genomics*, 27(1), pp. 39–42. Available at: <https://doi.org/10.1097/fpc.0000000000000246>. [Accessed: January 22, 2023].
- (16) van der Meer, R., Wilms, E.B. and Heijerman, H.G.M. (2021) “CFTR modulators: Does One dose fit all?,” *Journal of Personalized Medicine*, 11(6), p. 458. [accessed April 23, 2023] Available at: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8224731/>

- (17) Nieddu, E. (2013) “Editorial (hot topic: cystic fibrosis: CFTR modulators and their mechanism of action),” *Current Pharmaceutical Design*, 19(19), pp. 3474–3475. [Accessed: May 06, 2023]. Available at: <https://www.eurekaselect.com/article/50849>
- (18) Zaman, K. *et al.* (2014) *S-nitrosothiols increases cystic fibrosis transmembrane regulator expression and maturation in the cell surface*, *Biochemical and biophysical research communications*. U.S. National Library of Medicine. [Accessed: January 22, 2023]. Available at: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3974270/?report=reader>
- (19) Kotha K, Clancy JP. Ivacaftor treatment of cystic fibrosis patients with the G551D mutation: A review of the evidence. *Therapeutic Advances in Respiratory Disease*. 2013;7(5):288–96. [accessed 2023 Jan21]. Available at: [https://journals.sagepub.com/doi/10.1177/1753465813502115?url\\_ver=Z39.88-2003&rfr\\_id=ori:rid:crossref.org&rfr\\_dat=cr\\_pub%20%20pubmed](https://journals.sagepub.com/doi/10.1177/1753465813502115?url_ver=Z39.88-2003&rfr_id=ori:rid:crossref.org&rfr_dat=cr_pub%20%20pubmed)
- (20) Gramegna, A. *et al.* (2020) “From ivacaftor to triple combination: A systematic review of efficacy and safety of CFTR modulators in people with cystic fibrosis,” *International Journal of Molecular Sciences*, 21(16), p. 5882. [accessed May 06, 2023] Available at: <https://doi.org/10.3390/ijms21165882>.
- (21) Shteinberg, M. and Taylor-Cousar, J.L. (2020) “Impact of CFTR modulator use on outcomes in people with severe cystic fibrosis lung disease,” *European Respiratory Review*, 29(155), p. 190112. [Accessed: January 29, 2023] Available at: <https://doi.org/10.1183/16000617.0112-2019>.
- (22) Harvey, C. *et al.* (2022) “The effect of CFTR modulators on airway infection in cystic fibrosis,” *International Journal of Molecular Sciences*, 23(7), p. 3513. [Accessed: March 31, 2023] Available at: <https://doi.org/10.3390/ijms23073513>.
- (23) Ooi C, Syed S, Rossi L, Garg M, Needham B, Avolio J, *et al.* Impact of CFTR modulation with ivacaftor on gut microbiota and intestinal inflammation. *Scientific Reports*. 2018Dec13;8(1) [Accessed March 31, 2023] Available from: [https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6292911/pdf/41598\\_2018\\_Article\\_36364.pdf](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6292911/pdf/41598_2018_Article_36364.pdf)
- (24) Zhang, S., Shrestha, C.L. and Kopp, B.T. (2018) “Cystic fibrosis transmembrane Conductance Regulator (CFTR) modulators have differential effects on cystic fibrosis

macrophage function,” *Scientific Reports*, 8(1). [Accessed: January 24, 2023]. Available at: <https://doi.org/10.1038/s41598-018-35151-7>.

(25) Kuk K, Taylor-Cousar JL. Lumacaftor and ivacaftor in the management of patients with cystic fibrosis: Current evidence and future prospects. *Therapeutic Advances in Respiratory Disease*. 2015;9(6):313–26. [accessed 2023 Jan21]. Available from: [https://journals.sagepub.com/doi/10.1177/1753465815601934?url\\_ver=Z39.88-2003&rfr\\_id=ori:rid:crossref.org&rfr\\_dat=cr\\_pub%20%20pubmed](https://journals.sagepub.com/doi/10.1177/1753465815601934?url_ver=Z39.88-2003&rfr_id=ori:rid:crossref.org&rfr_dat=cr_pub%20%20pubmed)

(26) *Annex I summary of Product Characteristics - European Medicines Agency* (no date) EMA Europa. [Accessed: January 27, 2023] Available at: [https://www.ema.europa.eu/en/documents/product-information/kalydeco-epar-product-information\\_en.pdf](https://www.ema.europa.eu/en/documents/product-information/kalydeco-epar-product-information_en.pdf)

(27) Ema (2023) *Orkambi, European Medicines Agency*. Available at: <https://www.ema.europa.eu/en/medicines/human/EPAR/orkambi> (Accessed: March 11, 2023).

(28) *Annex I summary of Product Characteristics - European Medicines Agency* (no date) EMA Europa. Available at: [https://www.ema.europa.eu/en/documents/product-information/symkevi-epar-product-information\\_en.pdf](https://www.ema.europa.eu/en/documents/product-information/symkevi-epar-product-information_en.pdf) (Accessed: January 27, 2023).

(29) Ema (2023) *Kaftrio, European Medicines Agency*. Available at: <https://www.ema.europa.eu/en/medicines/human/EPAR/kaftrio> (Accessed: March 9, 2023)

(30) Ema (2023) *Kaftrio, European Medicines Agency*. Available at: <https://www.ema.europa.eu/en/medicines/human/EPAR/kaftrio> (Accessed: March 31, 2023).

(31) Ensink, M.M. and Carlon, M.S. (2022) “One size does not fit all: The past, present and future of Cystic Fibrosis Causal Therapies,” *Cells*, 11(12), p. 1868. Available at: <https://doi.org/10.3390/cells11121868>. (Accessed: March 13, 2023).

(32) Mergioti, M. *et al.* (2022) “CFTR modulator therapy for rare CFTR mutants,” *Journal of Respiration*, 2(2), pp. 59–76. Available at: <https://doi.org/10.3390/jor2020005>. (Accessed March 14, 2023)



## **9. Curriculum Vitae**

Anna Philine Westphal was born on April 17<sup>th</sup> 1998, in Berlin, Germany. She finished her school career 2016 at “Kippenberg Gymnasium” in Bremen. 2017 she began to study medicine in the first program of medical studies in English, at the University of Rijeka, Croatia. Here, she received a scholarship for excellence in 2021, as one of the best students in her year.

In addition to her regular student obligations, she worked as a nursing assistant in a COVID ambulance in Bremen. Additionally, she participated as a student work job in the European COVID study “LEOSS”, via the “Klinikum Bremen Mitte”.

She absolved the practical parts of several subjects, such as Internal Medicine, Dermatology, Neurology, Surgery and Clinical Skills in German hospitals.

Furthermore, she accomplished several voluntary practicals in Germany, withunder at the Universitätsklinikum Eppendorf and Universitätsklinikum Göttingen.

Philine Westphal will receive her diploma in July 2023.