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A Novel RP-HPLC Method for the Simultaneous Estimation of Ivacaftor and Lumacaftor in Bulk and Pharmaceutical Dosage Forms with Stability Studies

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ABSTRACT

A simple, accurate, precise method was developed for the simultaneous estimation of the Lumacaftor and Ivacaftor in bulk and Tablet dosage form. Chromatogram was run through Agilent C18 150 x 4.6 mm, 5µ. Mobile phase containing Buffer Na_2HPO_4: Acetonotrile taken in the ratio 65:45 was pumped through column at a flow rate of 1 ml/min. Buffer used in this method was Na_2HPO_4. Temperature was maintained at 30°C. Optimized wavelength selected was 245 nm. Retention time of Lumacaftor and Ivacaftor were found to be 2.116 min and 2.707. %RSD of the Lumacaftor and Ivacaftor were and found to be 0.8 and 0.8 respectively. %Recovery was obtained as 100.02% and 100.36% for Lumacaftor and Ivacaftor respectively. LOD, LOQ values obtained from regression equations of Lumacaftor and Ivacaftor were 1.00, 3.03 and 0.09, 0.26 respectively. Regression equation of Lumacaftor is y = 11463x + 11639, and y = 6775.7x + 7013.2of Ivacaftor. Retention times were decreased and that run time was decreased, so the method developed was simple and economical that can be adopted in regular Quality control test in Industries.

INTRODUCTION

DRUG PROFILE [1-4]

Ivacaftor

Description: Ivacaftor (also known as Kalydeco or VX-770) is a drug used for the management of Cystic Fibrosis (CF) in patients aged 2 years and older. Cystic Fibrosis is an autosomal recessive disorder caused by one of several different mutations in the gene for the Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) protein, an ion channel involved in the transport of chloride and sodium ions across cell membranes. CFTR is active in epithelial cells of organs such as the lungs, pancreas, liver, digestive system, and reproductive tract. Alterations in the CFTR gene result in altered production, misfolding, or function of the protein and consequently abnormal fluid and ion transport across cell membranes. As a result, CF patients produce a thick, sticky mucus that clogs the ducts of organs where it is produced making patients more susceptible to complications such as infections, lung damage, pancreatic insufficiency, and malnutrition.

STRUCTURE

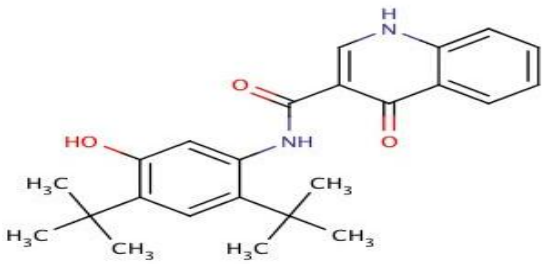

Figure No. 1: Ivacaftor structure

Application: A CFTR activator

CAS Number: 873054-44-5

Purity: ≥98%

Molecular Weight: 392.49

Molecular Formula: $C_{24}H_{28}N_2O_3$

IUPAC name: N-(2,4-di-tert-butyl-5-hydroxyphenyl)-4-oxo-1,4-dihydroquinoline-3-carboxamide

Appearance: Crystalline powder

Physical State: Solid

Solubility: Water Solubility (0.002 mg/mL) Soluble in DMSO (78 mg/ml at 25 °C), water (<1 mg/ml at 25 °C), and ethanol (<1 mg/ml at 25 °C).

Storage: Store at room temperature

Melting Point: 253.58° C (Predicted)

Boiling Point: ~550.5° C at 760 mmHg (Predicted)

Density: ~1.2 g/cm3 (Predicted)

pK Values: 6.57

Indication: When used as monotherapy as the product Kalydeco, ivacaftor is indicated for the management of CF in patients age 2 years and older who have a mutation in the CFTR gene that is responsive to ivacaftor potentiation. When used in combination with the drug Lumacaftor as the product Orkambi, ivacaftor is indicated for the management of CF patients age 6 years and older who are homozygous for the F508del mutation in the CFTR gene.

Mechanism of action: Ivacaftor exerts its effect by acting as a potentiator of the CFTR protein, an ion channel involved in the transport of chloride and sodium ions across cell membranes of the lungs, pancreas, and other organs. Alterations in the CFTR gene result in altered production, misfolding, or function of the protein and consequently abnormal fluid and ion transport across cell membranes. Ivacaftor improves CF symptoms and underlying disease pathology by potentiating the channel open probability (or gating) of CFTR protein in patients with impaired CFTR gating mechanisms. The overall level of ivacaftor-mediated CFTR chloride transport is dependent on the amount of CFTR protein at the cell surface and how responsive a particular mutant CFTR protein is to ivacaftor potentiation.

Absorption: Following administration of ivacaftor with fat containing foods, peak plasma concentrations were reached at 4 hours (Tmax) with a maximum concentration (Cmax) of

768 ng/mL and AUC of 10600 ng * hr/mL. It's recommended that ivacaftor should be taken with fat-containing foods as they increase absorption by approximately 2.5- to 4-fold.

Volume of distribution: After oral administration of 150 mg every 12 hours for 7 days to healthy volunteers in a fed state, the mean (±SD) for apparent volume of distribution was 353 (122).

Protein binding: Ivacaftor is approximately 99% bound to plasma proteins, primarily to alpha 1-acid glycoprotein and albumin. Ivacaftor does not bind to human red blood cells.

Metabolism: Ivacaftor is extensively metabolized in humans. *In vitro* and clinical studies indicate that ivacaftor is primarily metabolized by CYP3A. M1 and M6 are the two major metabolites of ivacaftor in humans. M1 has approximately one-sixth the potency of ivacaftor and is considered pharmacologically active. M6 has less than one-fiftieth the potency of ivacaftor and is not considered pharmacologically active.

Route of elimination: Following oral administration, the majority of ivacaftor (87.8%) is eliminated in the feces after metabolic conversion. The major metabolites M1 and M6 accounted for approximately 65% of the total dose eliminated with 22% as M1 and 43% as M6. There was negligible urinary excretion of ivacaftor as unchanged parent.

Half-life: The apparent terminal half-life was approximately 12 hours following a single dose.

Uses: This medication is used to treat cystic fibrosis in certain people (those with an abnormal "CFTR" gene). It may help to improve breathing, reduce the risk of lung infections, and improve weight gain.

Side Effects: Dizziness, headache, and nausea may occur. If any of these effects persist or worsen, tell your doctor or pharmacist promptly.

Name: LUMACAFTOR

Description: Lumacaftor is a drug used in combination with Ivacaftor as the fixed dose combination product Orkambi for the management of Cystic Fibrosis (CF) in patients aged 6 years and older. Cystic Fibrosis is an autosomal recessive disorder caused by one of several different mutations in the gene for the Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) protein, a transmembrane ion channel involved in the transport of chloride

and sodium ions across cell membranes of the lungs, pancreas, and other organs. Mutations in the CFTR gene result in altered production, misfolding, or function of the CFTR protein and consequently abnormal fluid and ion transport across cell membranes. As a result, CF patients produce thick, sticky mucus that clogs the ducts of organs where it is produced making patients more susceptible to infections, lung damage, pancreatic insufficiency, and malnutrition. Lumacaftor improves CF symptoms and underlying disease pathology by aiding the conformational stability of F508del-mutated CFTR proteins, preventing misfolding and resulting in increased processing and trafficking of mature protein to the cell surface.

Structure:

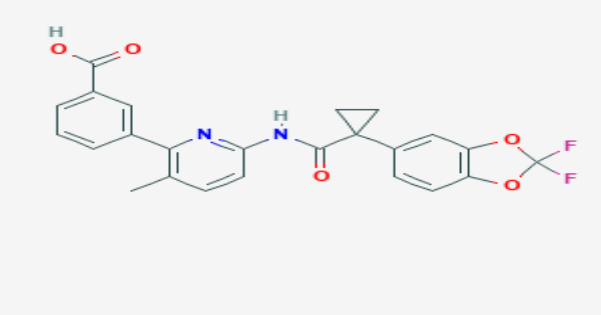

Figure No. 2: Lumacaftor Structure

CAS number: 936727-05-8

Molecular Weight: 452.414

Chemical Formula: $C_{24}H_{18}F_2N_2O_5$

IUPAC Name: 3-[6-[[1-(2,2-difluoro-1,3-benzodioxol-5-yl)cyclopropanecarbonyl]amino]-3-methylpyridin-2-yl]benzoic acid

Indication: When given in combination with Ivacaftor as the fixed dose combination product Orkambi, lumacaftor is indicated for the treatment of cystic fibrosis (CF) in patients age 6 years and older who are homozygous for the F508del mutation in the CFTR gene.

Pharmacodynamics: Results from clinical trials indicated that treatment with Orkambi (lumacaftor/ Ivacaftor) results in improved lung function, reduced chance of experiencing a pulmonary exacerbation, reduced sweat chloride, increased weight gain, and improvements in CF symptoms and quality of life. Label Orkambi was not found to increase the QTc interval to any clinically relevant extent.

Mechanism of action: Lumacaftor improves CF symptoms and underlying disease pathology by aiding the conformational stability of F508del-mutated CFTR, resulting in increased processing and trafficking of mature protein to the cell surface. Label more specifically, lumacaftor acts as a protein-folding chaperone, preventing misfolding of CFTR ion channels and consequent destruction during processing in the endoplasmic reticulum.

Absorption: Following administration of Orkambi (lumacaftor/Ivacaftor) with fat containing foods, peak plasma concentrations were reached at 4 hours (Tmax). It's recommended that Orkambi should be taken with fat-containing foods as they increase absorption of lumacaftor by approximately 2-fold, and [DB08820 by 3-fold.

Volume of distribution: Following oral administration of 200 mg of lumacaftor every 24 hours to cystic fibrosis patients in a fed state for 28 days, the mean (+/-SD) for apparent volumes of distribution was 86.0 (69.8) L.

Protein binding: Lumacaftor is extensively protein bound in the plasma (99%), and binds primarily to albumin.

Metabolism: Lumacaftor is mostly excreted unchanged in the feces and is not extensively metabolized. When metabolism does occur, oxidation and glucuronidation are the main processes involved.

Route of elimination: Lumacaftor is primarily excreted unchanged in the feces (51%). A minimal amount of the parent compound and its metabolites are excreted in the urine.

Half life: The half-life of lumacaftor is approximately 26 hours.

Brand name: ORKAMBI

MATERIALS AND METHODS [5-8]

Materials:

- Lumacaftor and Ivacaftor pure drugs (API), Combination Lumacaftor and Ivacaftor tablets, Distilled water, Acetonitrile, Phosphate buffer, Methanol, Potassium dihydrogen orthophosphate buffer, Ortho-phosphoric acid. All the above chemicals and solvents are from Rankem.

Instruments:

- Electronics Balance-Denver
- p^H meter -BVK enterprises, India
- Ultrasonicator-BVK enterprises
- WATERS HPLC 2695 SYSTEM equipped with quaternary pumps, Photo Diode Array detector and Auto sampler integrated with Empower 2 Software.
- UV-VIS spectrophotometer PG Instruments T60 with special bandwidth of 2 mm and 10mm and matched quartz cells integrated with UV win 6 Software was used for measuring absorbances of Lumacaftor and Ivacaftor solutions.

Methods:

Diluent: Based upon the solubility of the drugs, diluent was selected, Acetonitrile and Water taken in the ratio of 50:50.

Preparation of Standard stock solutions: Accurately weighed 50 mg of Lumacaftor, 31.25 mg of Ivacaftor and transferred to 50 ml volumetric flask and 3/4 th of diluents was added to these flask and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (1000 µg/ml of Lumacaftor and 625 µg/ml of Ivacaftor)

Preparation of Standard working solutions (100% solution): 1ml from each stock solution was pipetted out and taken into a 10ml volumetric flask and made up with diluent. (100 µg/ml Lumacaftor of and 62.5 µg/ml of Ivacaftor)

Preparation of Sample stock solutions: 5 tablets were weighed and the average weight of each tablet was calculated, then the weight equivalent to one tablet was transferred into a 100 ml volumetric flask, 5ml of diluents was added and sonicated for 25 min, further the volume was made up with diluent and filtered by HPLC filters. (100 µg/ml of Lumacaftor and 62.5 µg/ml of Ivacaftor)

Preparation of Sample working solutions (100% solution): 1 ml of filtered sample stock solution was transferred to 10 ml volumetric flask and made up with diluent. (100 µg/ml of Lumacaftor and 62.5µg/ml of Ivacaftor)

Preparation of buffer:

0.1%OPA Buffer: 1 ml of orthophosphoric acid was diluted to 1000 ml with HPLC grade water.

Validation:

System suitability parameters: The system suitability parameters were determined by preparing standard solutions of Lumacaftor (100 ppm) and Ivacaftor (62.5 ppm) and the solutions were injected six times and the parameters like peak tailing, resolution and USP plate count were determined.

The % RSD for the area of six standard injections results should not be more than 2%.

Specificity: Checking of the interference in the optimized method. We did not find interfering peaks in blank and placebo at retention times of these drugs in this method. So this method was said to be specific.

Precision:

Preparation of Standard stock solutions: Accurately weighed 50 mg of Lumacaftor, 31.5mg of Ivacaftor and transferred to 50 ml volumetric flask and 3/4 th of diluents was added to these flask and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (1000 µg/ml of Lumacaftor and 625 µg/ml of Ivacaftor)

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Preparation of Sample working solutions (100% solution): 1 ml of filtered sample stock solution was transferred to 10 ml volumetric flask and made up with diluent. (100 µg/ml of Lumacaftor and 62.5 µg/ml of Ivacaftor)

Linearity:

Preparation of Standard stock solutions: Accurately weighed 50 mg of Lumacaftor, 31.25mg of Ivacaftor and transferred to 50 ml volumetric flask and 3/4 th of diluents was added to these flask and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (100µg/ml of Lumacaftor and 62.5µg/ml of Ivacaftor)

25% Standard solution: 0.25ml each from two standard stock solutions was pipetted out and made up to 10ml. (25µg/ml of Lumacaftor and 15.625µg/ml of Ivacaftor)

50% Standard solution: 0.5ml each from two standard stock solutions was pipetted out and made up to 10ml. (50µg/ml of Lumacaftor and 31.25µg/ml of Ivacaftor)

75% Standard solution: 0.75ml each from two standard stock solutions was pipetted out and made up to 10ml. (75µg/ml of Lumacaftor and 46.875µg/ml of Ivacaftor)

100% Standard solution: 1.0ml each from two standard stock solutions was pipetted out and made up to 10ml. (100µg/ml of Lumacaftor and 62.5µg/ml of Ivacaftor)

125% Standard solution: 1.25ml each from two standard stock solutions was pipetted out and made up to 10ml. (100µg/ml of Lumacaftor and 78.125µg/ml of Ivacaftor)

150% Standard solution: 1.5ml each from two standard stock solutions was pipettede out and made up to 10ml. (100µg/ml of Lumacaftor and 93.75µg/ml of Ivacaftor)

Accuracy:

Preparation of Standard stock solutions: Accurately weighed 50 mg of Lumacaftor, 31.25mg of Ivacaftor and transferred to 50ml volumetric flask and 3/4 th of diluents was added to these flask and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. (100µg/ml of Lumacaftor and 62.5µg/ml of Ivacaftor)

Preparation of 50% Spiked Solution: 0.5ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.0ml from each standard stock solution was pipetted out and made up to the mark with diluent.

Preparation of 100% Spiked Solution: 1.0ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.0ml from each standard stock solution was pipetted out and made up to the mark with diluent.

Preparation of 150% Spiked Solution: 1.5ml of sample stock solution was taken into a 10ml volumetric flask, to that 1.0ml from each standard stock solution was pipetted out and made up to the mark with diluent.

Acceptance Criteria:

The % Recovery for each level should be between 98.0 to 102.

Robustness: Small deliberate changes in method like Flow rate, mobile phase ratio, and temperature are made but there were no recognized change in the result and are within range as per ICH Guidelines.

Robustness conditions like Flow minus (0.9ml/min), Flow plus (1.1ml/min), mobile phase minus, mobile phase plus, temperature minus (25°C) and temperature plus (35°C) was maintained and samples were injected in duplicate manner. System suitability parameters were not much effected and all the parameters were passed. %RSD was within the limit.

LOD sample Preparation: 0.25 ml each from two standard stock solutions was pipetted out and transferred to two separate 10 ml volumetric flasks and made up with diluents. From the above solutions 0.1ml each of Lumacaftor, Ivacaftor, solutions respectively were transferred to 10ml volumetric flasks and made up with the same diluents.

LOQ sample Preparation: 0.25 ml each from two standard stock solutions was pipetted out and transferred to two separate 10 ml volumetric flask and made up with diluent. From the above solutions 0.3ml each of Lumacaftor, Ivacaftor, solutions respectively were transferred to 10ml volumetric flasks and made up with the same diluent.

Degradation studies:

Oxidation:

To 1 ml of stock solution of Lumacaftor and Ivacaftor, 1 ml of 20% hydrogen peroxide (H2O2) was added separately. The solutions were kept for 30 min at 60^0C. For HPLC study, the resultant solution was diluted to obtain 100µg/ml & 62.5µg/ml solution and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of sample.

Acid Degradation Studies:

To 1 ml of stock s solution Lumacaftor and Ivacaftor, 1 ml of 2N Hydrochloric acid was added and refluxed for 30 mins at 60ºC. The resultant solution was diluted to obtain 100 µg/ml & 62.5 µg/ml solution and 10 µl solutions were injected into the system and the chromatograms were recorded to assess the stability of sample.

Alkali Degradation Studies:

To 1 ml of stock solution Lumacaftor and Ivacaftor, 1 ml of 2N sodium hydroxide was added and refluxed for 30 mins at 60^0C. The resultant solution was diluted to obtain 100 µg/ml & 62.5µg/ml solution and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of sample.

Dry Heat Degradation Studies:

The standard drug solution was placed in oven at 105°C for 1 h to study dry heat degradation. For HPLC study, the resultant solution was diluted to 100 µg/ml & 62.5 µg/ml solution and10 µl were injected into the system and the chromatograms were recorded to assess the stability of the sample.

Photo Stability studies:

The photochemical stability of the drug was also studied by exposing the 1000 µg/ml & 625 µg/ml solution to UV Light by keeping the beaker in UV Chamber for 1days or 200 Watt hours/m^2 in photo stability chamber. For HPLC study, the resultant solution was diluted to obtain 100 µg/ml & 62.5 µg/ml solutions and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of sample.

Neutral Degradation Studies:

Stress testing under neutral conditions was studied by refluxing the drug in water for 1hrs at a temperature of 60º. For HPLC study, the resultant solution was diluted to 100 µg/ml & 62.5 µg/ml solution and 10 µl were injected into the system and the chromatograms were recorded to assess the stability of the sample.

RESULTS AND DISCUSSION

Method Validation: The validation of the Process carried out was validated as per ICH guidelines and the following parameters were reported as follows:

System suitability: All the system suitability parameters were within the range and satisfactory as per ICH guidelines.

According to ICH guidelines, plate count should be more than 2000, tailing factor should be less than 2 and resolution must be more than 2. All the system suitable parameters were passed and were within the limits.

Table No. 1: System suitability parameters for Lumacaftor and Ivacaftor

S. No.	Lumacaftor			Ivacaftor			
Inj	RT(min)	USP Plate Count	Tailing	RT(min)	USP Plate Count	Tailing	Resolution
1	2.089	4473	1.36	2.678	5326	1.26	4.3
2	2.091	4527	1.39	2.681	5210	1.26	4.3
3	2.102	4310	1.37	2.695	5251	1.28	4.2
4	2.103	4294	1.37	2.695	5230	1.28	4.2
5	2.104	4415	1.40	2.697	5211	1.25	4.3
6	2.116	4332	1.39	2.707	5360	1.31	4.1

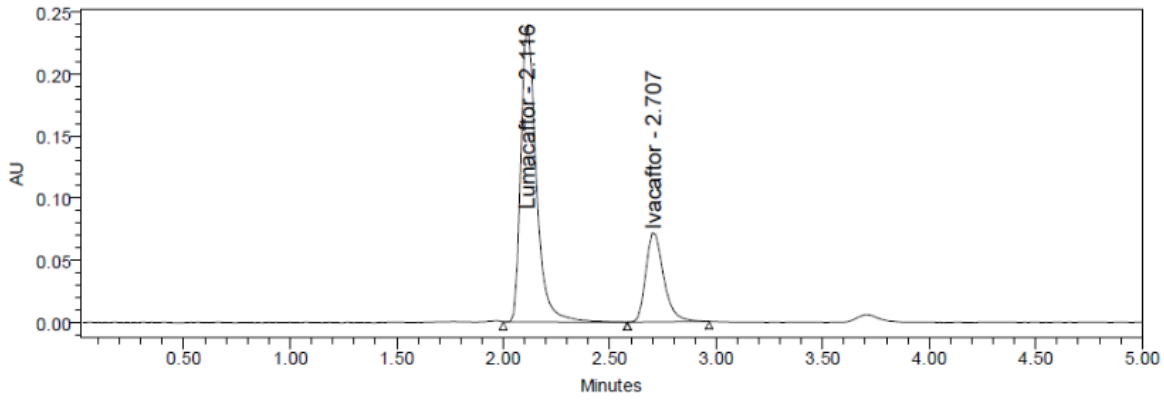

Figure No. 3: System suitability Chromatogram

Accuracy:

Three levels of Accuracy samples were prepared by standard addition method. Triplicate injections were given for each level of accuracy and mean % Recovery was obtained as 100.02% and 100.36% for Lumacaftor and Ivacaftor respectively.

Table No. 2: Accuracy table of Lumacaftor

% Level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery	Mean % Recovery
50%	50	49.72887	99.46	100.02%
	50	50.48705	100.97	
	50	49.86138	99.72	
100%	100	99.14542	99.15	
	100	100.3555	100.36	
	100	99.19419	99.19	
150%	150	152.6196	101.75	
	150	149.2695	99.51	
	150	150.1569	100.10	

Table No. 3: Accuracy table of Ivacaftor

% Level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery	Mean % Recovery
50%	20	31.22	99.89	100.36%
	20	31.78	101.70	
	20	30.82	98.62	
100%	40	62.786	100.46	
	40	62.351	99.76	
	40	63.079	100.93	
150%	60	94.123	100.398	
	60	94.789	101.108	
	60	94.114	100.388	

LOD and LOQ

The LOD and LOQ of the developed method were determined by injecting progressively low concentrations of the standard solutions using the developed RP-HPLC method. The LOD is the smallest concentration of the analyze that give the measurable response. The LOD for Lumacaftor and Ivacaftor was found to be and 1.00 and 3.03 respectively.

LOQ is the smallest concentration of the analyte, which gives response that can be accurately quantified signal to noise ratio of 10. The LOQ was 0.09 and 0.26 for Lumacaftor and Ivacaftor.

Linearity:

Six linear concentrations of Ivacaftor (15-90 µg/ml) and Lumacaftor (10-60 µg/ml) were injected in a duplicate manner. Average areas were mentioned above and linearity equations obtained for Lumacaftor was y = 11463x + 11639and of Ivacaftor was y = 6775.7x + 7013.2. Correlation coefficient obtained was 0.999 for the two drugs.

Table No. 4: Linearity table for Lumacaftor and Ivacaftor

Lumacaftor		**Ivacaftor**	
Conc. (µg/mL)	**Peak area**	**Conc. (µg/mL)**	**Peak area**
0	0	0	0
25	296407	15.625	117350
50	593976	31.25	223647
75	892547	46.875	321932
100	1146028	62.5	435959
125	1447345	78.125	533708
150	1723503	93.75	639785

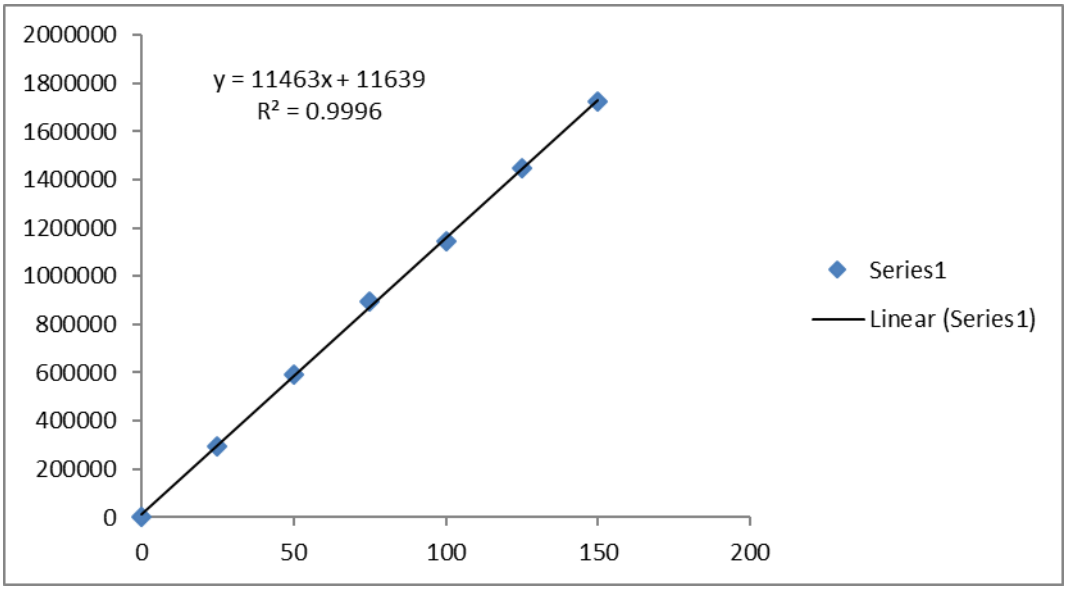

Figure No. 4: Calibration curve of Lumacaftor

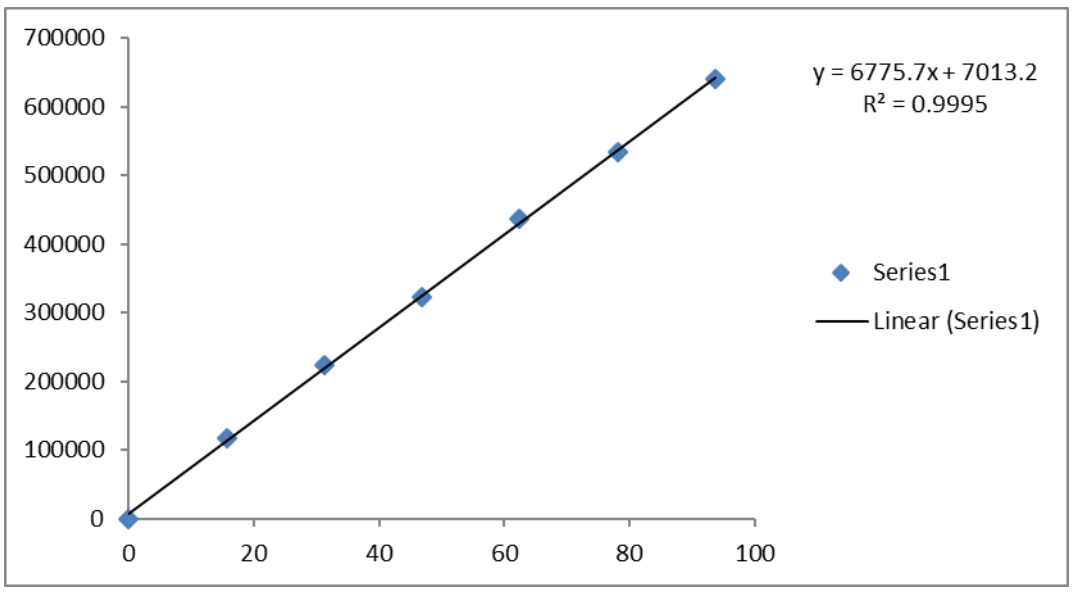

Figure No. 5 Calibration curve of Ivacaftor

System Precision:

From a single volumetric flask of working standard solution, six injections were given and the obtained areas were mentioned above. Average area, standard deviation and % RSD were calculated for two drugs. % RSD obtained as 0.8% and 0.8% respectively for Lumacaftor and Ivacaftor. As the limit of Precision was less than "2" the system precision was passed in this method.

Table No. 5: System precision table of Lumacaftor and Ivacaftor

S. No.	Area of Lumacaftor	Area of Ivacaftor
1.	1180469	412254
2.	1173827	410241
3.	1167054	415914
4.	1183940	418219
5.	1189202	413178
6.	1193266	417873
Mean	1181458	414613
S.D	9716.8	3226.8
%RSD	0.8	0.8

Robustness:

Robustness conditions like Flow minus (0.9ml/min), Flow plus (1.1ml/min), mobile phase minus (60B:40A), mobile phase plus (70B:30A), temperature minus (25°C) and temperature plus(35°C) was maintained and samples were injected in duplicate manner. System suitability parameters were not much affected and all the parameters were passed. %RSD was within the limit.

Table No. 6: Robustness data for Lumacaftor and Ivacaftor.

S. No.	Condition	%RSD of Lumacaftor	%RSD of Ivacaftor
1	Flow rate (-) 0.9ml/min	0.4	0.5
2	Flow rate (+) 1.1ml/min	1.3	0.6
3	Mobile phase (-) 60B:40A	0.6	0.5
4	Mobile phase (+) 70B:.35A	0.0	0.4
5	Temperature (-) 25°C	0.3	0.3
6	Temperature (+) 35°C	0.2	0.5

Assay: ORKAMBI, bearing the label claim Ivacaftor 125 mg, Lumacaftor 200 mg. Assay was performed with the above formulation. Average % Assay for Lumacaftor and Ivacaftor obtained was 100.11% and 100.05% respectively.

Table No. 7: Assay Data of Lumacaftor

S. No.	Standard Area	Sample area	% Assay
1	1180469	1178546	99.55
2	1173827	1181234	99.78
3	1167054	1198310	101.22
4	1183940	1197827	101.18
5	1189202	1179647	99.65
6	1193266	1175554	99.30
Avg	1181458	1185186	100.11
Stdev	9716.8	10150.8	0.857
% RSD	0.8	0.9	0.9

Table No. 8: Assay Data of Lumacaftor

S. No.	Standard Area	Sample area	% Assay
1	412254	411376	99.02
2	410241	414015	99.66
3	415914	422556	101.71
4	418219	417935	100.60
5	413178	417976	100.61
6	417873	410066	98.71
Avg	414613	415654	100.05
Stdev	3226.8	4699.9	1.13
% RSD	0.8	1.1	1.1

CONCLUSION

A simple, accurate, precise method was developed for the simultaneous estimation of the Lumacaftor and Ivacaftor in bulk and Tablet dosage form. Retention time of Lumacaftor and Ivacaftor were found to be 2.116 min and 2.707. % RSD of the Lumacaftor and Ivacaftor were and found to be 0.8 and 0.8 respectively. %Recovery was obtained as 100.02% and 100.36% for Lumacaftor and Ivacaftor respectively. LOD, LOQ values obtained from regression equations of Lumacaftor and Ivacaftor were 1.00, 3.03 and 0.09, 0.26 respectively. Regression equation of Lumacaftor is y = 11463x + 11639, and y = 6775.7x + 7013.2 of Ivacaftor. Retention times were decreased and that run time was decreased, so the method developed was simple and economical that can be adopted in regular Quality control test in Industries.

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