LIQUID CHROMATOGRAPHY-TANDEM–MASS SPECTROMETRY BIOANALYTICAL TECHNIQUE FOR ESTIMATING FAVIPIRAVIR IN PHARMACEUTICAL DOSE FORMS, BOTH IN BULK AND IN COMBINATION

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ABSTRACT

Objectives: The aim is to develop a simple, straightforward, quick, and accurate LCMS/MS bioanalytical technique for estimating favipiravir in pharmaceutical dose forms, both in bulk and in combination.

Methods: Phenyl column using an isocratic separation with a 40:60 buffer and acetonitrile combination as the mobile phase and a buffer containing 1 mL of formic acid in 1 L of water.

Results: An excellent linear range of 2 ng/mL to 40 ng/mL (r^2=0.9997) was analyzed in the 10-min analysis. Favipiravir shows an LOD of 1 ng/ml (s/n = 4) and an LOQ of 10 ng/ml (s/n = 24). Accuracy, linearity, specificity, precision, system suitability, stability, and recoveries study results are all within the limits.

Conclusion: In accordance with the U.S. Food and Drug Administration criteria, the proposed method was concluded to be suitable for regular evaluation and used for determining the amount of a specific drug component in tablet dosage form. The medicine remained stable during the stability experiments.

Keywords: Liquid chromatography-tandem mass spectrometry, Favipiravir, U.S. Food and Drug Administration (USFDA) guidelines, Mobile phase, Stability.

INTRODUCTION

An antiviral medication called favipiravir was developed to treat a number of viral diseases, including COVID-19 and influenza [1,2]. Its chemical name is 6-fluoro-3-hydroxypyrazine-2-carboxamide, and its molecular weight is 157.104 g/mol. Its molecular formula is $C_5H_4FN_3O_2$ [3]. It is a white powder with a pKa value of 5.1 that is soluble in organic solvents and only weakly soluble in water [4]. Favipiravir is an organic molecule that is a member of the pyrazine carboxamides class [5]. Favipiravir stops viral transcription and replication by blocking the RNA-dependent RNA polymerase enzyme [6]. Therefore, the aim of the present studyis liquid chromatography-tandem–mass spectrometry (LC-MS/MS) bioanalytical technique for estimating favipiravir in pharmaceutical dose forms, both in bulk and in combination in compliance employing analytical characteristics listed in the International Council for Harmonisation (ICH) guidelines [7,8], including matrix factor, stability, limit of detection (LOD), limit of quantitation (LOQ), recovery, specificity, system suitability, precision, accuracy, linearity, and stability. Compared to HPLC or UV procedures, LC–MS/MS is faster, more selective (avoiding interference with excipients or other pharmaceuticals), and more sensitive (detecting ng/mL levels), making it the preferred method for estimating favipiravir in bulk and combination dose forms. HPLC-UV performs well with single, pure samples. In contrast, LC-MS/MS provides precise, reliable outcomes [9].

The novelty of the LCMS method improves throughput for stability and quality control investigations by providing broader linearity, faster run times, and simpler sample preparation. This improvement over current methods guarantees a reliable, stability-indicating, and legally valid way to quantify favipiravir in bulk and combination goods.

METHODS

Chemicals and reagents

In Mumbai, Maharashtra, we bought LCMS grade acetonitrile (ACN) from Sisco Research Laboratories Pvt. Ltd. Merck (India) Ltd. provides methanol and formic acid. The supplier of favipiravir was Biophore India Pharmaceuticals Pvt Ltd., Andhra Pradesh, Vishakapatnam.

Equipment

We used Alliance-type HPLC equipment in combination with a SCIEX, Framingham, MA, USA, QTRAP 5500, triple quadrupole LCMS/MS. The Waters e 2695-Empower software 2.0 versions (Waters Corporation, Milford, MA, USA) were used to complete the process.

Chromatographic conditions

Chromatographic separation was performed on an X-Bridge Phenyl column (150 × 4.6 mm, 3.5 μm; Waters Corporation, Milford, MA, USA) using an Agilent 1260 Infinity HPLC system equipped with a UV detector (Agilent Technologies, Santa Clara, CA, USA). The separation was carried out in isocratic mode at 25 °C. The phenyl stationary phase was selected over a conventional C18 column owing to its stronger π–π interactions with the aromatic ring of favipiravir, thereby improving selectivity, peak shape, and resolution, particularly in combination formulations. The mobile phase consisted of 0.1% formic acid and acetonitrile (40:60, v/v), delivered at a flow rate of 1.0 mL/min. An injection volume of 10 μL was used, and favipiravir was eluted at a retention time of 2.8 min, as detected by the UV detector. [10].

Preparation of favipiravir stock solution

The precisely weighted standard component was dissolved in methanol to produce a stock solution of the antiviral drug favipiravir that was

utilized throughout the LCMS method development stage. The standard solution of favipiravir has a concentration of 1 mg/mL. The working solutions used for the development of methods, calibration curves, and QC samples were prepared by appropriately diluting the stock solution with the mobile phase. In polypropylene vials, working standard solutions were preserved in a freezer (Thermo Scientific, USA) set at -20 °C [11, 12].

Preparation of internal standard (IS) for favipiravir

Zanamivir is used as an internal standard because its structural and physicochemical similarity to favipiravir ensures similar extraction, ionization, and recovery, giving accurate and reliable quantification. The IS is one of the most crucial components of analysis in the bioanalytical approach. Analyte similarity was assessed using the IS for the favipiravir LCMS technique [13].

m/z determination of favipiravir

ACN was used to dissolve favipiravir to create a stock solution with 1 mg/mL of the drug. Methanol was used to further dilute (10 µg/mL) this stock solution. This solution was taken in vials, and aliquots were examined for m/z in a mass detector (SCIEX 5500 QTRAP, USA) [14].

Extraction of favipiravir from a plasma sample

200 µL of blank plasma is treated with 300 µL of acetonitrile for protein precipitation (acetonitrile gives faster and high, consistent recovery of favipiravir with acceptable matrix effects), followed by the addition of Favipiravir (20 ng/mL), 500 µL Internal standard (I.S.) (30 ng/mL) and 500 µL of diluent then the mixture is vortexed for 5 minutes resulting mixture was centrifuged for 20 minutes at 5000 rpm, (Thermo Scientific Sorvall ST 16R / ST 40R ,USA) at controlled temperature 6oC then 2.5 mL Supernatant was gathered and put into the HPLC for analysis [15].

Bio-analytical method validation

To make sure the device was operating correctly, the system's suitability was investigated as well. By evaluating QC samples at various concentrations (low quality control [LQC], medium QC [MQC], and high QC [HQC]) on several days, accuracy and precision were ascertained. Favipiravir in human plasma, the new bio-analytical method was assessed for precision, selectivity, linearity, sensitivity, accuracy, recovery, and matrix effect using LCMS (SCIEX, Framingham, MA, USA) coupled with an HPLC (Waters Corporation, Milford, MA, USA). A range of circumstances, namely freeze-thaw, bench-top, wet extract, and auto-sampler stability, were used to assess the drug's stability in plasma. The method is verified for pharmacokinetic investigations of drugs in spiked human plasma because it was validated in compliance with regulatory standards [16,17].

System suitability

System suitability testing ensures the HPLC system is performing properly by checking precision and consistency using samples with 20 ng/mL Favipiravir and 30 ng/mL IS. Every batch is finished in the beginning, middle, and end, and the relative standard deviation (%RSD) for peak area and retention time must be below 2% and 5%, respectively, over six injections [18].

Stability of stock solution

80 ng/mL favipiravir and 120 ng/mL Zanamivir were combined to produce an aqueous stock solution in diluent. Three containers of the solution were created; the first was kept at the ambient temperature, the second was kept in a deep freezer (discuss the temperature), and the third was kept at −20°C (said to be stable as a freshly made solution). At specified intervals (0, 12, 24 h), the drug and IS solutions in each storage condition were removed and injected into the HPLC. Each sample's area of peaks from the chromatogram was contrasted with samples that had just been made [19].

Specificity and selectivity

For this test, blank plasma specimens from six separate sources were analyzed to look for signs of chromatographic interference at the IS and favipiravir retention periods [20].

Calibration curve

Appropriate volumes of working solution were spiked into the blank plasma to produce an 8-point calibration curve, yielding the ultimate concentrations of 2, 5, 10, 15, 20, 25, 30, and 40 ng/ml for the favipiravir. The plotting of the peak area ratio of the favipiravir to the IS transition pair versus the nominal level of calibration standards allowed for the creation of the calibration curve. A linear regression analysis was performed on the results. Except for lower limit of quantification (LLOQ), which was set at ±20%, each one of the back-calculated standard values had an acceptable criterion of ±15% standard deviation (SD) of the nominal value (Food and Drug Administration of the United States, 2001) [21].

Precision and accuracy

Six duplicates were examined on the same day as well as three different days, respectively, at six QC standards (LQC, MQC, HQC, and LLOQ). The data had to be accurate within ±15% of the nominal values and precise within ±15% of the RSD, except for LLOQ, which should not be more than ±20% of the SD [22].

Estimating LOD and LOQ (Signal-to-noise method)

The analyte concentration required to generate a signal with a certain noise-to-signal ratio value was obtained by measuring the signal-to-noise ratio within the analyte retention time using the signal-to-noise method. The highest peak of the blank plasma close to the favipiravir retention time was used to compute the noise value. The chromatogram printout was used to manually measure the noise magnitude. In general, the quantity of analyte for which the ratio of signal to noise was equal to or >3 times has been designated as LOD, and the amount of analyte at which the signal-to-noise ratio was equal to or >5 times was recognized as LOQ.

Matrix effect

Obtaining the matrix effect from six different drug-free plasma samples of favipiravir was achieved by comparing their height-area ratios. Six separate plasma lots were used in triplicate experiments conducted at MQC levels with an acceptable accuracy of <15% [23].

Recovery

By assessing the analytical outcomes for extracted samples at three distinct concentrations (LQC, MQC, and HQC) in six replicates with extracted standards (unprocessed), which indicate 100% recovery, the process for the extraction recovery from the analytes was identified. Whereas a recovery experiment was carried out for IS at a single concentration of 0.1 ng/mL. The recovery study results were calculated as the average, SD, and %RSD [24].

Stability studies

To evaluate the stability of favipiravir in human plasma under several circumstances, stability experiments were carried out. Stability experiments were performed by storing spiked plasma samples at −20 °C in a freezer (Thermo Fisher Scientific, USA). Autosampler stability was evaluated using an Agilent 1260 Infinity HPLC autosampler (Agilent Technologies, USA). Plasma samples were centrifuged at 4,000 rpm for 10 min at 4 °C using Eppendorf 5810 R refrigerated centrifuge (Eppendorf, Hamburg, Germany) prior to storage. The stability of the analyte was assessed at -20°C under benchtop, auto-sampler, short-, long-, and freeze-thaw cycles. Spiked quality control samples were stored at -20 °C for 24 hours to evaluate the stability of the wet extract and dry extract [25, 26].

RESULTS AND DISCUSSION

m/z determination

Fig. 1 shows a mass spectrum of favipiravir ion peaks at different m/z values, with the highest peak at m/z 158.63, indicating the most abundant fragment. Other peaks represent different ionized fragments of the compound. Fig. 2 shows the mass spectrum for Zanamivir, base peak at m/z 332.65, indicating the most abundant ion. Other significant fragment ions are observed at m/z 193.48, 272.61, 428.27, 505.34,

and 607.33. Favipiravir shows a lighter spectrum, consistent with its smaller molecular structure. Zanamivir, being larger and more complex, exhibits a higher base peak and heavier fragment ions, confirming its more elaborate structure.

Analytical method development of favipiravir by LCMS method
To develop a new LC procedure with mass detection, a mobile phase based on a 0.1% formic acid buffer was initially studied. Following several trials, the best buffer for the mobile phase of the suggested LCMS method was determined. The optimized mobile phase was identified to be 0.1% formic acid and ACN in the ratio of 40:60 v/v, and it shows a broad peak. The shorter retention time (2.808 min) of favipiravir suggests it is more hydrophilic or less retained by the column's stationary phase than

Fig. 1: Mass spectrometry spectra of favipiravir

Fig. 2: Mass spectrometry spectra of zanamivir

zanamivir (3.998 min) shown in Fig. 3. Symmetrical peaks suggest good column performance. No tailing or fronting, indicating the analytes are not interacting adversely with the mobile phase. So, this separation is efficient, with good resolution between the two analytes.

System suitability
Favipiravir's area ratio and percentage RSD were determined to be 0.88% and 0.172, respectively. As a result, the system suitability was passed. Table 1 summarizes the findings. The %RSD for the RT must be <2.00% to meet the acceptance criteria. The area ratios' percentage RSD ought to be <5.00.

Specificity and selectivity
The Food and Drug Administration's criteria were followed when conducting both selectivity and specificity tests. The remaining blank serum sample was examined for the selectivity research, and the chromatogram of the blank showed no interfering peaks in the retention periods related to favipiravir and the IS (Zanamivir). Excellent specificity was demonstrated by the chromatograms derived from the analysis, which revealed no interference peaks at the drug and IS retention durations (Fig. 4). According to this observation, the overall run time was 5 min, and the drug and IS retention times were approximately 2.865 and 3.976 min, respectively. This demonstrates that the suggested procedure is capable of detecting and quantifying favipiravir within human plasma and other dosage forms.

Sensitivity and LLOQ
The sensitivity of the bioanalytical method was determined by assessing the limit of quantification. It was discovered that the LLOQ is the lowest amount of favipiravir that can be measured with a reasonable level of accuracy and precision, which was 1 ng/mL. This illustrates how the technique can identify and measure favipiravir at low levels in human plasma, which qualifies it for pharmacokinetic research and efficacy studies. The %RSD for favipiravir and its area ratio was found to be 1.012% and 115%. Hence, it passed the sensitivity. Table 2 shows the sensitivity results of favipiravir. This indicates that LLOQ meets precision and accuracy criteria, assuming that the method was validated. The % Mean Accuracy of 115% at LLOQ exceeds the ±20% limit, likely due to matrix effects, low-concentration handling errors, or signal enhancement in the LC–MS/MS analysis.

Precision and accuracy
The findings suggested that the %RSD of intra-day and inter-day precision proved within reasonable bounds, and Table 3 shows that the procedure was repeatable and reproducible. Furthermore, the method's precision further supports its dependability for measuring favipiravir in human plasma.

Matrix effect
When the analyte was ionized, a matrix of ionized constituents was
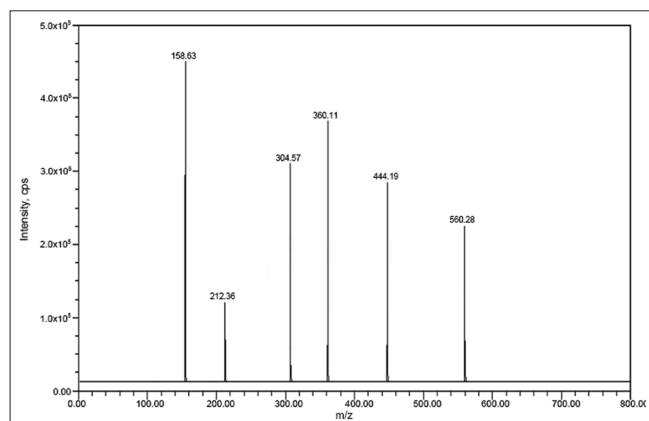

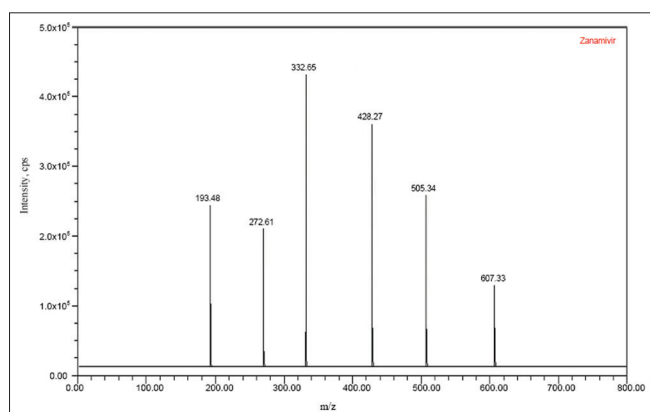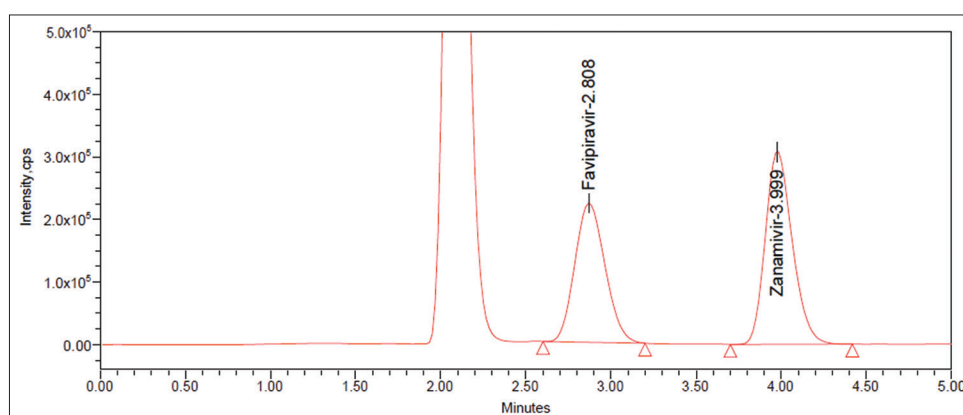**Fig. 3: LC–MS/MS chromatogram of Favipiravir (RT 2.808 min) and Zanamivir IS (RT 3.999 min) showing good peak shape with tailing factors of 1.12 (Favipiravir) and 1.08 (Zanamivir), indicating acceptable peak symmetry**
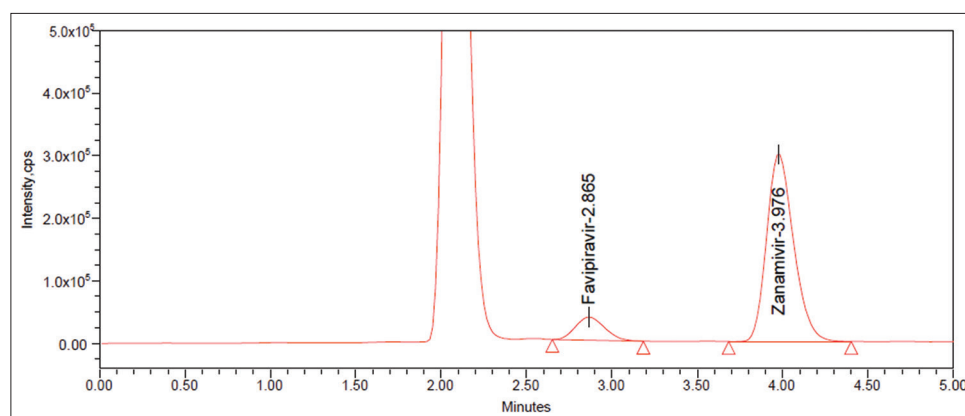
Fig. 4: Favipiravir-spiked blank rat plasma at lower limit of quantification and internal standard

Table 1: Results of Favipiravir System suitability at MQC Level (Acceptance criteria: %RSD < 2% for retention time and peak area; %RSD < 5% for area ratio

Name of sample	Area of analyte	RT of the analyte (min)	IS area	IS RT (min)	Area ratio
MQC	2.505×10^5	2.808	3.071×10^5	3.999	0.8157
MQC	2.503×10^5	2.802	3.089×10^5	3.995	0.8103
MQC	2.510×10^5	2.800	3.050×10^5	3.992	0.8230
MQC	2.503×10^5	2.806	3.029×10^5	3.990	0.8263
MQC	2.507×10^5	2.804	3.031×10^5	3.998	0.8271
MQC	2.514×10^5	2.801	3.035×10^5	3.994	0.8283
Mean	2.507×10^5	2.803	3.050×10^5	3.994	0.8218
SD	0.0043	0.003	0.0244	0.0034	0.0072
%RSD	0.172	0.109	0.800	0.086	0.8819

MQC: Medium QC, RSD: Relative standard deviation, IS: Internal standard, SD: Standard deviation. *n=6

Table 2: Favipiravir sensitivity results

Replicate number	LLOQ
	Nominal concentration (ng/mL) 2.24
	Nominal concentration range (2.10–2.51)
	Area of analyte (ng/mL)
1	0.256×10^5
2	0.257×10^5
3	0.252×10^5
4	0.253×10^5
5	0.252×10^5
6	0.253×10^5
Mean	0.254×10^5
SD	0.00257
%RSD	1.012
% Mean accuracy	115%

RSD: Relative standard deviation, SD: Standard deviation, LLOQ: Lower limit of quantification. *n=6

Table 3: Precision and accuracy of the favipiravir data

QC sample	Area of analyte (ng/mL)	Mean (ng/mL)	SD	Accuracy (%)	RSD (%)
Intra-day					
LLOQ	0.251×10^5	0.255×10^5	0.017	96.6	6.92
LQC	1.255×10^5	1.248×10^5	0.014	101.75	1.14
MQC	2.529×10^5	2.534×10^5	0.021	100.12	0.83
HQC	3.742×10^5	3.749×10^5	0.024	99.48	0.66
LLOQ	0.251×10^5	0.255×10^5	0.017	96.6	6.92
Inter-day					
LLOQ	0.254×10^5	0.257×10^5	0.018	96.9	6.98
LQC	1.253×10^5	1.242×10^5	0.013	100.15	1.02
MQC	2.532×10^5	2.537×10^5	0.028	100.09	0.81
HQC	3.741×10^5	3.751×10^5	0.024	99.51	0.72

MQC: Medium QC, RSD: Relative standard deviation, SD: Standard deviation, LLOQ: Lower limit of quantification, HQC: High quality control, LQC: Low quality control, QC: Quality control. *n=6

Table 4: Favipiravir matrix effect

S. No.	Lot No.	HQC	LQC
		Nominal concentration (ng/mL) 30.42	10.43
		Nominal concentration range (ng/mL) (30.12–30.60)	(10.14–10.63)
		Area of analyte (ng/mL)	
1	Lot 1	3.741×10^5	1.279×10^5
		3.778×10^5	1.281×10^5
		3.774×10^5	1.264×10^5
2	Lot 2	3.747×10^5	1.282×10^5
		3.774×10^5	1.254×10^5
		3.734×10^5	1.234×10^5
3	Lot 3	3.774×10^5	1.256×10^5
		3.754×10^5	1.274×10^5
		3.717×10^5	1.264×10^5
4	Lot 4	3.747×10^5	1.261×10^5
		3.724×10^5	1.281×10^5
		3.747×10^5	1.287×10^5
5	Lot 5	3.771×10^5	1.291×10^5
		3.747×10^5	1.297×10^5
		3.798×10^5	1.242×10^5
6	Lot 6	3.787×10^5	1.297×10^5
		3.774×10^5	1.271×10^5
		3.742×10^5	1.269×10^5
n		18	18
Mean		3.757×10^5	1.271×10^5
SD		0.022	0.017
%CV		0.59	1.39
% Mean accuracy		99.29%	102.48%
No. of QC failed		0	0

SD: Standard deviation. HQC: High quality control, LQC: Low quality control, QC: Quality control

determined by comparing the response of post-extracted plasma standard QC samples (n=6) with the response of the analyte from neat samples at equivalent concentrations. The rat plasma that has been chromatographically screened was used to evaluate the anticipated procedure. Precision of favipiravir (%CV) at HQC and LQC is 0.59% and 1.39%, respectively. The matrix effects of favipiravir quantitation are displayed in Table 4.

Linearity

Concentration had a direct correlation with the peak area ratio of the standards used for calibration. Over the 2.0–40.00 ng/mL of favipiravir concentration range, the standard graphs were linear. 0.999 was the

Table 5: Recovery of the analyte of favipiravir

Replicate number	HQC		MQC		LQC	
	Extracted response	Un extracted response	Extracted response	Un extracted response	Extracted response	Un extracted response
1	$3.725×10^5$	$3.849×10^5$	$2.525×10^5$	$2.678×10^5$	$1.206×10^5$	$1.325×10^5$
2	$3.765×10^5$	$3.865×10^5$	$2.558×10^5$	$2.675×10^5$	$1.219×10^5$	$1.317×10^5$
3	$3.725×10^5$	$3.835×10^5$	$2.569×10^5$	$2.697×10^5$	$1.243×10^5$	$1.323×10^5$
4	$3.735×10^5$	$3.875×10^5$	$2.587×10^5$	$2.699×10^5$	$1.273×10^5$	$1.341×10^5$
5	$3.762×10^5$	$3.869×10^5$	$2.581×10^5$	$2.723×10^5$	$1.239×10^5$	$1.321×10^5$
6	$3.797×10^5$	$3.881×10^5$	$2.562×10^5$	$2.733×10^5$	$1.224×10^5$	$1.343×10^5$
Mean	$3.752×10^5$	$3.862×10^5$	$2.564×10^5$	$2.701X10^5$	$1.234X10^5$	$1.328×10^5$
SD	0.028	0.017	0.021	0.023	0.023	0.010
%RSD	0.76	0.45	0.86	0.87	1.90	0.82
%Mean Recovery	99.19%	101.44%	100.09%	102.76%	100.21%	105.98%
Overall % Mean Recovery	101.61%					
Overall SD	0.020					
Overall %RSD	0.94					

SD: Standard deviation, HQC: High quality control, LQC: Low quality control, MQC: Medium quality control, RSD: Relative standard deviation, *n=6

Table 6: Stability studies of favipiravir

Stability conditions	Concentration level	Mean	SD	%CV	% Mean stability
Autosampler	HQC	$3.745×10^5$	0.030	0.48	99.58
	LQC	$1.245×10^5$	0.032	2.64	99.32
	MQC	$2.544×10^5$	0.028	1.11	101.47
Wet extract	HQC	$3.764×10^5$	0.034	0.91	100.09
	LQC	$1.239×10^5$	0.030	2.50	98.84
	MQC	$2.543×10^5$	0.033	1.31	101.44
Freeze-thaw	HQC	$3.737×10^5$	0.025	0.68	99.38
	LQC	$1.243×10^5$	0.020	1.63	99.16
	MQC	$2.52×10^5$	0.015	0.62	100.52
Bench Top	HQC	$3.763×10^5$	0.038	1.03	100.06
	LQC	$1.24×10^5$	0.046	3.75	100.95
	MQC	$2.531×10^5$	0.037	1.46	99.56
Dry Extract	HQC	$3.762×10^5$	0.032	0.87	100.04
	LQC	$1.241×10^5$	0.021	1.72	99.00
	MQC	$2.535×10^5$	0.034	1.36	101.12
Short term	HQC	$3.612×10^5$	0.007	0.20	96.05
	LQC	$1.144×10^5$	0.052	4.62	91.26
	MQC	$2.415×10^5$	0.007	0.31	96.33
Long term	HQC	$3.184×10^5$	0.003	0.12	84.67
	LQC	$1.050×10^5$	0.007	0.72	83.77
	MQC	$2.152×10^5$	0.006	0.32	85.84

HQC: High quality control, LQC: Low quality control, MQC: Medium quality control. *n=6
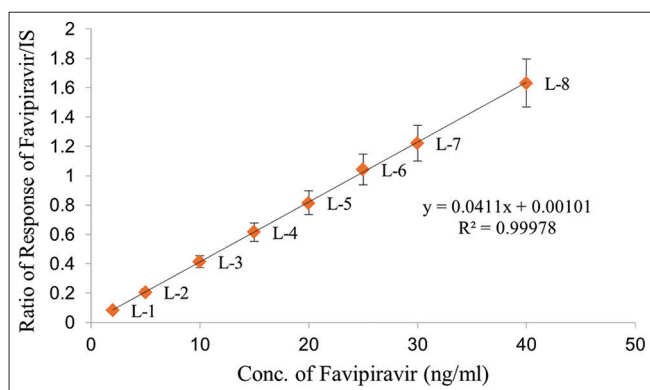

Fig. 5: Plot of calibration for favipiravir concentration versus area ratio

average correlation coefficient. The ratio of the analyte peak area to the IS peak area was used to quantify the samples. Plotting of peak area ratios against plasma concentrations was done. Fig. 5 shows its calibration plot, which seemed to be linear.

LOQ and LOD

The calibration curve approach was used to determine the LOD and LOQ independently. Using the developed LCMS technique, increasingly smaller quantities of standard solutions were injected to determine the compound's LOD and LOQ. Favipiravir has LOD concentrations of 1 ng/mL and s/n values of 4. Favipiravir has an LOQ concentration of 10 ng/mL and a s/n value of 24.

Recovery of analyte

Three concentration levels — low, medium, and high- QC — were used to assess the drug and IS recovery. By contrasting its reaction in samples that were repeated by standard solution responses, recovery was computed. The comparison of the analytical response from the quantity of analyte added to that ascertained from the sample matrix is known as the analyte recovered from a matrix of samples (extraction efficiency). ACN solvent was used for the extraction of favipiravir due to its fundamental characteristics. As shown in Table 5, the recovery parameter data were deemed adequate. The acceptable recovery range is between 98 and 102%. For each of the three concentrations, the results were well within the range. As a result, the recovery parameter showed that the suggested approach was verified.

Stability tests

To evaluate the stability of favipiravir in human plasma under several circumstances, stability tests were carried out. The stability of the analyte was assessed at −80°C under benchtop, auto-sampler, short-, long-, and freeze-thaw cycles. Spiked QC samples were left at room temperature for 18 h to evaluate the wet extract and dry stability. According to the findings, favipiravir did not significantly degrade in human plasma under those conditions, as shown in Table 6. This guarantees the method's dependability when handling and storing samples. Favipiravir (about 85% stability) cannot be maintained at -80 °C for an extended period of time; samples should be examined immediately or kept for a short amount of time under validated conditions.

CONCLUSION

A more sensitive LC-MS/MS technique has been developed to measure the favipiravir concentration. The approved technique was effectively used to measure drug levels in bulk and pharmaceutical dosage forms. This bioanalytical method is tough, quick, cost-effective, and provides quick quantification, good precision, accuracy, and sensitivity. The method can be used to check favipiravir therapeutically regularly. Above all, the parameters' outcomes followed the ICH guidelines. The analysis of favipiravir, both bulk

and formulations, was thus made possible by the suggested LC-MS/MS approach.

AUTHOR CONTRIBUTION

Kiran Kumar Ganta, the method, compilation of data and wrote the original draft. Raja Sundararajan, editing and Supervision.

CONFLICT OF INTEREST

No conflicts of interest among the authors.

AUTHOR FUNDING

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