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ORIGINAL ARTICLE **OPEN ACCESS**

Method Development and Validation of Tofacitinib Bulk Drug By Using UV Visible Spectrophotometer

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ABSTRACT
Tofacitinib is a drug used for the treatment of rheumatoid arthritis. Tofacitinib is a janus kinase inhibitor (JAK). A sensitive, simple, rapid, accurate and economical UV spectrophotometric method has been developed for the method development and validation of tofacitinib. Tofacitinib was estimated using uv visible double beam spectrophotometer at the wavelength maxima are 286nm in water. The developed method obeyed beers law in a concentration range of 2-10 µg/ml with correlation coefficient (R^2) of 0.9978. The limit of detection was found to be 10.45µg/ml and The limit of quantification was found to be 31.67µg/ml.
Key words: Tofacitinib, analytical method validation, ICH guideline, Specrtophotometric

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INTRODUCTION

The name of active pharmaceutical ingredient is tofacitinib. The category of tofacitinib is janus kinase inhibitor, antineoplastic and immunomodulating agent and it is used in treatment of rheumatoid arthritis. Tofacitinib are soluble in water, DMSO[3,4,7].

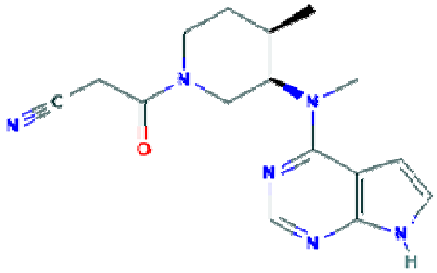

Fig. 1 Structure of Tofacitinib (Source: Google Images)

Tofacitinib is chemically 3-[(3R,4R)-4-methyl-3-[methyl (7H-pyrrolo[2,3-d]pyrimidin-4-yl)amino] piperidin-1-yl]-3-oxopropanenitrile. The molecular weight of tofacitinib is 312.37 g/mol and the molecular formula is $C_{16}H_{20}N_6O$.the half life is approximately 3 hrs.
Tofacitinib is a medication used to treat rheumatoid arthritis, ulcerative colitis, psoriatis arthritis, psoriasis. Tofacitinib is an inhibitor of the enzyme janus kinase 1 and janus kinase 3.Adverse effects include diarrhea, headache and high blood pressure.
Estimation of tofacitinib citrate by reported, however there was no method reported for tofacitinib by UV spectrophotometer for analytical method development and validation. It is robust, simple and accurate method and also economical analytical method.

MATERIAL AND METHODS

Experimental

Instrument and Material

Instruments- A double beam UV-1800 (Shimadzu) UV visible spectrophotometer. It consists of two similar, matched sample holder and it is made up of glass, quartz and silica. It is widely used for analytical

purpose.[1,2,5,8,9]. Following components are used for the measuring absorbance of an analytical solution:

1. Source
2. Detector
3. Monochromator
4. Sample holder or cuvette

Solution Preparation:

Preparation of standard stock solution -

10 mg of tofacitinib were weighed and transferred in to a 10 ml of volumetric flask, add small amount of water then, shake the flask. Drug is dissolved in water and make up the volume up to the mark with same solvent (water).these solution is make 1000µg/ml.

Preparation of sample solutions-

From standard stock solution 1 ml was taken and transferred to a 10ml volumteric flask and make up the volume up to the mark with water. These solution is make 100µg/ml.

From 100µg/ml withdraw 0.5ml, 1ml, 1.5ml, 2ml and 2.5ml solution with the help of pipette and dilute with water to 10ml and then to achieve 2µg/ml, 4µg/ml, 6µg/ml, 8µg/ml and 10µg/ml dilutions.

Method:

UV spectrophotometric method -

As per ICH guideline validation of an analytical procedure is to demonstrate that

The prepared 100µg/ml solution was taken and withdraws 1ml and dilute with water upto 10 ml The absorbance spectra of standard and test solution of tofacitinib was recorded in the range of 400-200 nm. The scanning was carried out in spectra mode of UV visible spectrophotometer. And it show lambda max 286 nm. (figure 2).

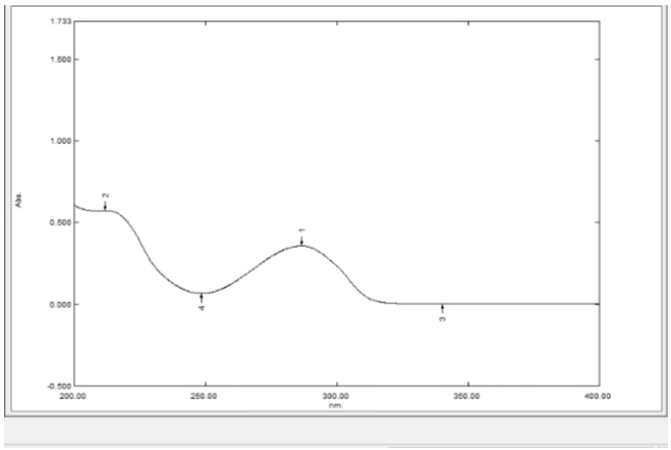

Fig.2UV visible Spectrum of tofacitinib in water.

RESULT AND DISCUSSION

General analytical procedure

Linearity -

Linearity of the analytical method is the ability, within a given range to obtain test result which is directly proportional to the concentration of analyte present in the sample within the given range.In this parameter linear correlation are obtained between concentration and absorbance in the selected wavelength of 2 -10 µg/ml. The linearity of developed method were determined at five different Concentration levels. Ranging from 2 – 10 µg/ml.

Table No.1: Linearity data of Tofacitinib

Sr. No.	Concentration (µg/ml)	Absorbance
1.	Blank	0.000
2.	2 PPM	0.073
3.	4 PPM	0.152
4.	6 PPM	0.226
5.	8 PPM	0.301
6.	10 PPM	0.361

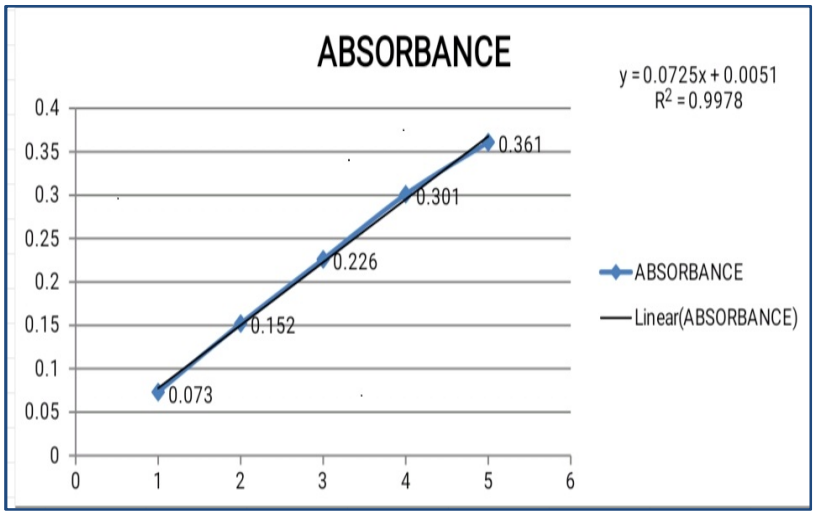

Table No. 2: Linearity of Tofacitinib

Sr. No.	Concentration (µg/ml)	Absorbance
1.	2	0.073
2.	4	0.152
3.	6	0.226
4.	8	0.301
5.	10	0.361
Regression data	M	0.0725
	C	0.0051
	R^2	0.997

Limit of detection:

LOD is an individual analytical process is the lowest amount of analyte in a sample which can be detected but not quantitated as an exact value.

It is the analytical parameter which is determine fromlinearity data. LOD was determine using the formula LOD = 3.3× SD/Slope. The LOD by using UV visible spectroscopy was found to be 10.42µg/ml.

LOD = 3.3× SD/slope

Table No.3: LOD of Tofacitinib

Sr. No.	Concentration (µg/ml)	Absorbance
1.	2	0.073
2.	4	0.152
3.	6	0.226
4.	8	0.301
5.	10	0.361
SD		0.11454365
Slope		0.03625
LOD		10.42

Limit of Quantification:

LOQ is the analytical parameter which is determine from linearity data. LOQ was determine using the formula LOQ = 10× SD/Slope. The LOQ by using UV visible spectroscopy was found to be 31.59µg/ml.

LOQ = 10×SD/Slope

Table No.4: LOQ of tofacitinib

Sr. No.	Concentration (µg/ml)	Absorbance
1.	2	0.073
2.	4	0.152
3.	6	0.226
4.	8	0.301
5.	10	0.361
SD		0.11454365
Slope		0.03625
LOD		31.59

Precision

As per ICH guideline, precision of an analytical process is usually expressed the variance, standard deviation of variation of a series of measurement.

The interday and intraday precision was performed by analyzing the 3 times responses on same day and on 3 different days for 3 different days for 3 different concentration of standard solution of Tofacitinib. The result obtained was reported in term of the relative standard deviation. The result obtained for interday and intraday variation by using UV – spectroscopy.

Table No.5: Intra-day precision

Sr. No.	Concentration (µg/ml)	Absorbance	Average	SD	RSD	%RSD
1	2	0.075	0.07433	0.00054	0.7264	72.64
2	2	0.074				
3	2	0.074				
4	4	0.162	0.3786	0.2652	70.036	7003.6
5	4	0.163				
6	4	0.161				
7	6	0.225	0.2243	0.0010	0.488	48.8
8	6	0.225				
9	6	0.223				

Table No.6: Inter-day precision

Sr.no	concentration	Absorbance	Average	SD	RSD	%RSD
1	2	0.076	0.1756	0.12320	70.15	7015
2	2	0.075				
3	2	0.074				
4	4	0.163	0.1626	0.0005830	0.3585	35.85
5	4	0.163				
6	4	0.162				
7	6	0.224	0.0.02045	0.25045	1224.4	122,440
8	6	0.226				
9	6	0.225				

Ruggedness

From stock solution, sample solution Tofacitinib were prepared and analyzed by two different analyst and by using the same operating condition and environmental condition.

Table No. 7: Ruggedness of Tofacitinib

Sr.no	Concentration(µg/ml)	Analysis 1	Analysis 2	Analysis 3
1	10	0.363	0.363	0.362
2	10	0.363	0.362	0.363
3	10	0.362	0.363	0.363
4	10	0.363	0.363	0.361
5	10	0.363	0.362	0.362
Average		0.3028	0.3626	0.3622
SD		0.00044	0.00173	0.00081
RSD		0.1212	0.4771	0.2236
%RSD		12.12	47.71	22.36

Robustness

As per ICH guideline, Robustness of an analytical procedure measure of its capacity to remain unaffected by small, variation in method parameter and provide an indication of reliability during normal usage.Robustness is an important parameter of analytical method as a small change in the method parameter like PH, solvent and composition etc.

Table No. 8: Robustness of Tofacitinib

Sr.no	Concentration(µg/ml)	286 nm	285 nm
1	8	0.303	0.300
2	8	0.301	0.301
3	8	0.301	0.299
4	8	0.303	0.300
5	8	0.303	0.300
Average		0.3022	0.3
SD		0.001095	0.000707
RSD		0.3623	0.23
%RSD		36.23	23

In the proposed method, Tofacitinib showed absorption maxima at 286 nm. The calibration curve was found to be linear in the concentration range of 2-10 µg/ml. The precision was calculated as intraday and interday variation for Tofacitinib. The proposed method was found to be accurate, simple, rapid, sensitive and economic for routine quality control analysis. The result of validation parameter was found to be satisfactory. Therefore this method can be applied successfully for the estimation of Tofacitinib in its pure form.

CONCLUSION

The Tofacitinib is the Janus Kinase Inhibitor and used to treat rheumatoid arthritis. The Tofacitinib bulk drug was studied by using UV visible spectroscopy. This technique is simple, robust, specific, accurate, precise and rapid for analysis. The developed spectrophotometric method was validated for Tofaciinib by using parameter like linearity, precision, limit of detection, limit of Quantification, Ruggedness, and Robustness as per the ICH guidelines. Hence, the proposed method was found to be more accurate, reproducible and sensitive.

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REFERENCES

1. Santosh V. Gandhi, BarkhaG.Kapoor, (2019). Development and validation of UV spectroscopic method for estimation of Baricitinib, journal of Drug Delivery and Therapeutics, 9:488-491.
2. N. V. Thakariya, S. B. Ezhava (2017). Development and Validation of UV spectrophotometric method for the estimation of Tofacitinib citrate, Niyanti et al/ pharma science monitor 8(2), 401-408.
3. http://pubchem.ncbi.nlm.nih.gov/compound/Tofacitinib
4. http://go.drugbank.com/drugs/DB08895
5. S. K. Sankar, P. Shanmugasundaram, B.Datchayani, N. Balakumaran, Mohammed Rilwan, R. Subaranjani, M. Sumithra, (2017). Stress Degradation studies and development of validated spectrometric – Assay method for Determination of Tofacitinib in pure and physical admixtures, Research J. pharm. and Tech.10(1);117:120.
6. K.Ortiz- Ibanez, M.M.Alsina, C. Munozsantos. (2013). Tofacitinib and other kinase inhibitor in the treatment of psoriasis. Actas Dermosifiliogr104(4):304-10.
7. Chemical Book, http://www.chemicalbook.com accessed on 20.3.2016.
8. International council on harmonization of technical requirement for the registration of pharmaceuticals for human use, validation of analytical procedures: text and methodology Q2(R1),2005.
9. ICH harmonized Tripartite Guideline, (1995). European medicines agency, validation of analytical procedures:definitions and methodology. CPMP/ICH/381/95.

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