RESEARCH ARTICLE

Formulation of Generic Atorvastatin Calcium Tablet by Reverse Engineering Technique

Furquan N. Khan*[1], Mirza S. Baig[1], Girish Nihalani[2], Mohammed Imran[1], Nikita Deshingkar[1]

[1]*YB Chavan College of Pharmacy, Aurangabad, Maharashtra, India*
[2]*Teva Pharmaceuticals, Florida, USA.*

Received: 8th April, 2021; Revised: 14th April, 2021; Accepted: 15th May, 2021; Available Online: 25th June, 2021

ABSTRACT

The present study involves preparinga generic dosage form of Atorvastatin calcium and establishing the similarity profile with the reference product. It involves a complete study of innovators' products from literature. Excipients from reference products were separated using differential solubility techniques as different excipients are soluble in different solvents. The study on reverse engineering of Atorvastatin calcium tablets was done using the marketed dosage form of 40 mg strength from the local market. The solvents used for separation were methanol, ethanol, dichloromethane, benzene, water, etc. The quantities of the drug and excipients present in the marketed dosage form after performing reverse engineering are lactose 22.33, MCC 11, croscarmellose sodium 0.55 mg, magnesium stearate 0.2 mg, calcium carbonate 4.2 mg, and drug 0.39 mg. DSC analyzes the separated API, and the results indicated a glass transition temperature of 140°C, and degradation was seen as a decrease in baseline at 190°C. The powder X-ray diffractogram indicated that the drug is X-ray amorphous. The separated excipients and drug were characterized by interpretation of Fourier-transform infrared spectroscopy (FTIR) spectra and melting point determination. Finally, the generic form was prepared, and it is characterized by performingdifferent physical and chemical tests; finally, F2 value in dissolution was determined and was found to be 57.14.

Keywords: Atorvastatin, Dissolution, Generic dosage form, Reverse engineering

International Journal of Pharmaceutical Quality Assurance (2021); DOI: 10.25258/ijpqa.12.2.03

How to cite this article: Khan FN, Baig MS, Nihalani G, Anees MI, Deshingkar NV. Formulation of Generic Atorvastatin Calcium Tablet by Reverse Engineering Technique. International Journal of Pharmaceutical Quality Assurance. 2021;12(2):14-20.

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

Atorvastatin calcium (ATC) belongs to the Statin classof cholesterol-lowering drugs, and it is the drug of choice in the management of elevated blood cholesterol level. Chemically ATC is [R-(R*,R*)]-2-(4-Fluorophenyl)-b,d-dihydroxy-5-(1-methylethyl)-3-pheny1-4[(phenylamino)carbonyl]-1H-pyrrole-1-heptanoic acid, calcium salt (2:1) trihydrate. The chemical structure of ATC is shown in Figure 1.[1]

ATC is ablockbuster lipid loweringsynthetic drugbelongs to BCS class II and acting as HMG-CoA (3 hydroxy-3-methylglutaryl coenzyme A) reductase inhibitor. The HMG-CoA enzyme is implicated in cholesterol biosynthesis as a catalyzing agent and takes part in the change reaction of HMG-CoA to mevalonate. The function of reduction in the numberof cholesterol results in removing the LDP (low-density lipoprotein) cholesterol in the blood by a surge in LDL receptors. The atorvastatin used as a calcium salt is utilizedto managethe initial phases ofhypercholesterolemia and dyslipidemia.[2] There is substantial scope for developing the generic dosage form of such drug concerningmore economic value and bioequivalence to that of innovators products. The keyadvantages of developing the generic brand are attributed to its low cost alternative as it bypasses the cost[3,4] of a clinical

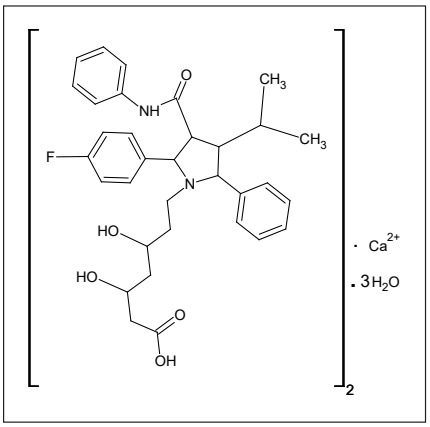

Figure 1: Chemical structure of Atorvastatin calcium

**Author for Correspondence: furkhankhan11@gmail.com*

trial, and manufacturers earntheir profit even at a much low cost. A clinical trial is one of the costlyresearch segments,and the clinical trial data is required to be submitted to Food Drug Association. As soon as the patent protection ends, the identical dosage form can be manufactured by demonstrating bioequivalency. Therefore, we can develop the generic dosage form that finally helps the community, particularly in developing countries.[5,6]

MATERIAL AND METHODS

Reagents and chemicals: ATC sample was procured as a gift from Lupin research park, Aurangabad, Maharashtra, India. ATC Tablets 40 mg strength was procured from the local market. The rest of theingredients and excipients used are laboratoryreagents.

METHODS

Characterization of ATC

Fourier Transform Infrared Analysis

The Fourier Transform Infrared Analysis (FTIR) of ATC was done on Jasco FTIR Spectrophotometer. The drug sample was triturated with fully dried Potassium bromide (KBr) in mortar and pestle in a moisture-free environment and then filled in the sample holder. Then the infrared spectrum was taken between in the range of 4000 cm^{-1} to 400 cm^{-1} wavenumbers.

Melting point characterization

The melting point of a compound gives an idea about the purity of the compound. The open capillary technique characterized the melting point of ATC.[7]

UV Visible Spectroscopy

Determination of Absorption Maxima

ATC equivalent to 50 mg is measured and solubilized in 50 mL of methanol to make a 1mg/mL solution. A quantity of 10 mL of the above solution is withdrawn in a 100 mL volumetric flask,and volume is made up to 100mL with methanol. Suitable dilutions are preparedusing methanol to obtain 10 µg/mL concentration and analyzed in UV range from 200–400 nm, the result and the spectrum obtained will be used for further analysis.[8]

Preparation of Standard Stock Solution: ATC in pure form (50 mg) was precisely weighed and taken in a 50 mL volumetric flask. Methanol is added to dissolve the drug, and the rest of the volume was filled up to 50 mL. The drug concentration obtained was 1 mg/mL. 2.5 mL solution was pipet ted in a 25 mL volumetric flask, and the remaining volume with methanol up to the mark. Finally, a stock solution of 100 µg / mL strength was prepared.

Preparation of Calibration Curve of Pure Drug: From the above-prepared stock solution,various dilutions of 0.5 mL, 1 mL, 1.5 mL, 2 mL, 2.5 mL dilutions were prepared in a 10 mL volumetric flask, and volume was filled up to the mark using methanol and solutions in the range of 5–25 µg/mL concentrations were obtained. The spectrum was taken, absorbance were obtained at 246 nm, and a calibration curve was prepared, graphically.[9]

Characterization of Marketed Dosage Form

The marketed dosage form of ATC 40 mg strength was purchased from the local market as reference dosage form and characterized for various physicochemical tests.[10,11]

Weight variation test

It is done to determine the deviation of each tablet's weight with an average weight in percent. Weigh 20 tablets and determined average weight. Repeat the procedure with other 19 tablets. Determine the average weight of the tablets. The individual weights should not deviate from not more than two as compared to the average by more than the percentage deviation, and none deviates by more than twice that percentage; for example,if the average weight of the tablet is 80 mg or less, 80 mg to 250 mg and 250 mg or more the percent deviation allowed is 10.0%, 7.5%, and 5% respectively.

Friability Test

Randomly selected 10 tablets were added in the friability drum of a tablet friability test apparatus, Roche friabilator. The drum was programmed at a rotation of 100 times in 4 minutes. After the test, the tablets are taken out of the drum, de dusted, and checked for weight loss. The weight loss is calculated in percentage, and if the loss percentage is greater than 1, the test has to be repeated twice, and the mean of the 3 tests results is determined. An upper limit of loss of 0.5% is considered to be acceptable for most products.

Hardness Test

Tablet hardness was found out by Monsanto hardness tester. Place the tablet between the jaws for every determination; similarly keep it in the direction of applying the force. Perform the test on 3 tablets, and care should be taken that all fragments of the tablets must be cleaned before each test.

Disintegration Test

Place 1 tablet into every tube of the basket and add a disc into every tube. Hang the basket assembly in the beaker comprising water and run the apparatus as per the specified time. After the run is over the basket assembly is taken out of the liquid, and observed for disintegration. The complete disintegration of 6 tablets indicates passing of the test.

In-vitro Dissolution Test

Dissolution test was performed as per the procedure given in the USP type II apparatus. 900 mL of dissolution media comprising of 3% SLS, 0.5 % NaCl, pH made up to 6 by using phosphoric acid, the prepared media should be free from dissolved air, and this media is transferred into the dissolution bowl of the apparatus and heated to get the temperature of the media to 37.5°C. The dissolution test was performed on 3 marketed formulations for about 90 minutes, and drug dissolved in percent Vs time graph was constructed. Every tablet was tested for the active amount dissolved in solution as a percentage of the specification in which more than 2 tablets are placed together. For each test , the quantity of active

ingredients in solution for each tablet was determined and amount in percent of the specified quantity was determined. The results should conform to the requirement at stage S1 given in the accompanying acceptance table if not, then continue testing with additional tablets through II and III stages, and the results conform at II stage.

Separation of API and Excipients

All excipient are separated as per the differential solubility technique; solubility of each component and amount was analyzed by solvent evaporation method of soluble component and then confirmed by various physical and chemical tests of each separated fraction. Ten tablets were taken and triturate. The exact amount of 10 tablet powder was found to be 400 mg. This content was then extracted in different solvents for the separation of excipients.[12,13]

Separation of ATC: The tablet powder equivalent to 10 tablets was dissolved in 30 mLAR grade Methanol. Drug powder and methanol dispersion were sonicated for about 15 minutes and then filtered through whatmann filter paper. Both filter paper deposit and filtrate are subjected to air dry. Methanol gets evaporated within 30 minutes and drug fraction gets separated. The quantity of separated drug fractions was measured.

Separation of Magnesium Stearate: The filter paper deposit obtained after filtration is allowed to react with warm ethanol. Magnesium stearate is freely soluble in warm ethanol. About 15 mL of warm ethanol was taken in which the residue obtained after separation with methanol was dissolved. This solution is then passed through whatmann filter paper. As Magnesium stearate is soluble in warm benzene passed through filter paper and residue contains residual excipient. The filtrate is air-dried for 30 minutes, and subsequently, the separated quantity of magnesium stearate was estimated.

Separation of Polysorbate 80: As polysorbate 80 is freely soluble in ethyl acetate, the remaining filter paper deposit was treated with ethyl acetate to separate polysorbate 80. About 15 mL of ethyl acetate was taken, in which residue obtained was dissolved. This solution is then passed through whatmann filter paper. The soluble portion of polysorbate 80 was passed through the filter paper, and the residue contains residual excipients. The filtrate is air-dried for 30 minutes, and then the quantity of separated polysorbate 80 was estimated.

Separation of Croscarmellose Sodium: Croscarmellose sodium can be separated with DMF help; hence, the remaining fraction of residue was treated with DMF. The filtrate obtained was air-dried for 30 minutes, and then the quantity of separated croscarmellose sodium was determined.

Separation of Hydroxypropyl Cellulose: The residue after treating with DMF is dissolved in propylene glycol to separate hydroxyl propyl cellulose. This solution was passed through whatmann filter paper. As HPC is freely soluble in propylene, glycol was passed through filter paper, and residue contains remaining excipients. Both filtrate and residue are subjected to air dry for 10–15 minutes, and then the amount of separated fraction of HPC was determined.

Separation of Microcrystalline Cellulose: As micro-crystalline cellulose is easily solubilized in Methylene chloride, the remaining fraction of residue was treated with Methylene chloride for Separation of MCC. About 15 mL of Methylene chloride was taken, and the remaining residue was dissolved in it. This solution was allowed to pass through whatmann filter paper, as MCC is freely soluble passed through filter paper. Both residue and filtrate were subjected to get air dry for 30 minutes, and then the quantity of separated fraction of HPC was calculated.

Separation of lactose: Lactose is soluble in ethanol; hence, the remaining residue was treated with ethanol, resulting in the separation of lactose.

Separation of Calcium Carbonate: For separation of calcium carbonate. The addition of ammonium salts increased the solubility of calcium carbonate in water. The residue was treated with hot water for separation of calcium carbonate, stirred well and this solution allowed to boil till all water content gets evaporated. The remaining fraction was consists of calcium carbonate. The quantity was measured.

Characterization of Separated Excipient

Characterization of Melting Point: The melting point of each excipient was characterized in the melting point apparatus. The separated excipient are said to be pure if they melt at the reported temperature.

Characterization of Procured and Separated API

API Analysis by Differential Scanning Calorimetry: Differential scanning calorimetry (DSC) was used to determine ATC's polymorphic form. The DSC was performed on shimadzu TA 60 WS (Model). Each sample was analyzed in an aluminum crimped and hermetically sealed pan consisting of precisely weighed (1–3 mg) compound, keeping an empty pan as reference. Calibration of the system was done by utilizing Indium (highly pure). The analysis of the samples was done at a higher heating rate of 2°C per minute between the temperature range of 80–280°C under N atmosphere, sharp alteration in peak is owing to endothermic change in substance. DSC also helps in determining the compatibility between API and excipients.

X-ray diffraction (Powder) Analysis: X-ray diffraction Powder (PXRD) patterns are determined to evaluate the crystalline/amorphous character of the untreated drug. Measurements were performed using XGW/30 kV/15 mA consisting of copper anode coupled to a computer interfaced diffractometer control unit. The vertical goniometer (PW3050/60) is used to determine scattered radiations.[14,15]

Preparation of Generic Form

The quantity of API and excipients is taken in the formulation of tablet based on the obtained quantities after the separation step as given in Table 1. Entire Ingredients, including API are precisely weighed and allowed to pass through a 30 number sieve to remove lumps, if any, and then uniformly mixed in a poly bag except magnesium stearate for about 30 minutes. The mixed powder was then collected in a tray,

and weight was determined on digital balance and recorded. The prepared blend was compressed into a tablet using 12 mm round shape punches and dies on 8 stations single rotatory tablet compression machine.[16-18]

Prepared Generic Form Characterization

The characterization of the prepared generic form was performed as per the tests mentioned under the characterization of a marketed dosage form such as Weight variation, friability, hardness, DT, and Disso test.[19,20]

Determination of F2 Value

It is the logarithmic transformation of
The sum - squared error of changes among the test T_t and reference products R_t at each time point. It signifies the nearness of two comparative dosage forms.[21]

Estimation of similarity factor is done by the following equation:

$$F2= 50+\log([1+(1/n)\sum_{t=1}*n(R_t-T_t)^2]^{-0.5}*100)$$

R_t and T_t are the cumulative % dissolved at every selected n time points of reference and test dosage form, respectively.

RESULTS AND DISCUSSION

Characterization of Procured At or vast at in Calcium

Fourier Transform Infrared Analysis

The FTIR spectra consisting of transmission (%) vs wave number of ATC is exhibited in Figure 1 with typical peaks of N-H (aromatic) stretching at 3363 and 3234 cm^{-1}, C=O stretching at 1646 cm^{-1}. Amidic groups at 1646 and 1427 cm^{-1}, OH stretching at 3671 cm^{-1}, Pyrrol ring stretching at 1157 and 1311 cm^{-1}. The IR spectrum of the sample complies with the standard. The FTIR spectra are exhibited in Figure 2.

Characterization of the Melting Point: The melting point of ATC,when found out with the help of a thistle tube, was found to be 153°C which corroborates with the scientific records.

Determination of Absorption Maxima and Calibration Curve: The absorption maxima of ATC were observed at 246 nm. The calibration curve shows that ATC obeys linearity between a range of 5–25 µg/mL concentration with $R^2 = 0.9978$ as shown in Figure 3.

Characterization of Marketed Dosage Form: The marketed dosage form of ATC 40 mg was characterized with various physicochemical tests,for example,weight variation, disso, and friability test,etc.

Determining Weight Variation: It is determined on 20 tablets. The average weight of tablets was obtained to be 410.7 mg, and the standard deviation was found to be 1.494. It was found that the result shows that all parameters were within specification. All the tablets possess uniform weight with minor SD values.

Hardness Test (Tablet): The tablet hardness was observed at 3.4Kg/cm^2. This guarantees the good behavior of tablets.

Friability Test: The percent friability was found to be 0.17%. As this value was less than 1% it confirms the mechanical stability of the tablets.

Disintegration Test: Disintegration time (DT) of ATC tablet was performed in water as a disintegrant media set at 37°C. DT of ATC was observed at 4 minutes and 20 seconds.

Table 1: Separation of excipients using differential solubility technique

Sr. No.	*Excipient separated*	*Solvents used*	*Quantity (g)*
1.	ATC	Methanol	0.39
2.	Magnesium stearate	Warm ethanol	0.2
3.	Polysorbate 80	Ethyl acetate	0.53
4.	Crosscarmellose sodium	Dimethyl Formamide	0.55
5.	HPC	Propylene glycol	0.75
6.	Lactose	Ethanol	22.33
7.	Calcium carbonate	Hot water	4.2
8.	MCC	Methylene chloride	11
9.	Total	-	39.950

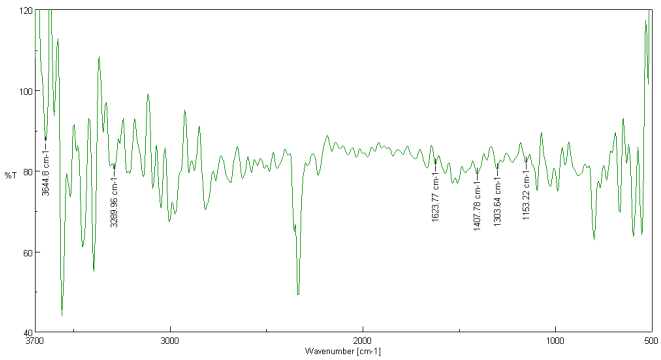

Figure 2. FT Infrared spectrum of ATC

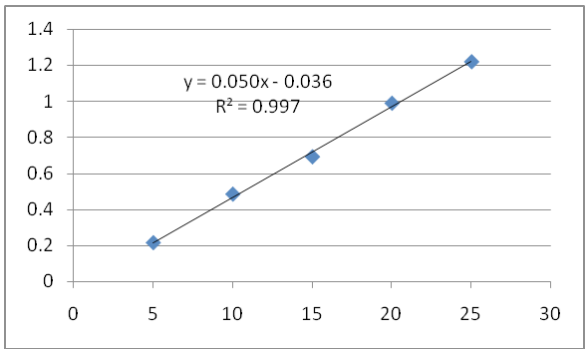

Figure 3: Calibration curve of ATC pure drug sample

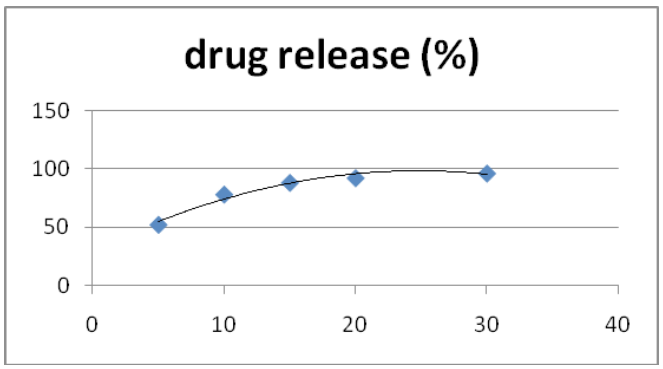

Figure 4: Percent drug dissolved of marketed dosage form

Dissolution Test: Dissolution test was done in buffer (phosphate) 6.8 pH media (Potassium dihydrogen phosphate 11.45 gm and disodium hydrogen phosphate 28.85 gm for 1000 mL), dissolution study was carried out on marketed dosage form within time range 5–30 minutes and percent drug release graph was plotted as shown in Figure 4.

Separation of API and Excipients: The exact amount of different ingredients separated based upon the differential solubility technique is given in Table 1. The weight of the powder of 10 tablets of ATC before separation is 39.998 g. This content was then extracted in different solvents for the separation of ATC and excipients. The weight of the total ingredients separated is 39.950 g. The loss of very minute quantities of the ingredients may be attributed to handling during experimentation.

Characterization of Separated Excipients: The separated excipients purity was ascertained by melting behavior and refractive index test.

Melting Point Determination: The melting behavior of separated excipients is analyzed to confirm the purity. The results of the melting point test indicate that the excipients are separated successfully; slight variation in melting point from the reported value may be due to the presence of minor quantities of the impurities. The results are given in Table 2.

Characterization of Separated ATC

Fourier Transform Infrared Analysis

It shows peaks of aromatic N-H stretching at 3363, 3234 cm^{-1} and C=O stretching at 1646 cm^{-1}. Amidic groups at 1646 and 1427, OH stretching at 3671, Pyrrol ring stretching at 1157 and 1311. The IR spectrum of the sample complies with the standard as depicted in Figure 5.

Determination of Melting Point: The Melting point of separated ATC was observed at 155 °C and complies with the reference.

Differential Scanning Calorimetry: DSC results exhibited glass transition temperature, which starts at 140°C, and the degradation is observed as a decrease in baseline at 190° C. No re crystallization was seen in the heating duration. The DSC Thermogram is shown in Figure 6.

X-ray diffraction (Powder) Analysis: X-ray diffraction(Powder) pattern of separated ATC was confirmed to be X-ray amorphous in comparison to X-ray diffraction spectrums of the reported amorphous ATC form. This confirms that the separated ATC is an amorphous solid (Figure 7).

Characterization of Prepared Generic Dosage Form

Weight variation: The test was conducted on 20 tablets. The average weight of tablets was 411.2 mg, and the standard deviation was 5.76. The results show that all parameters were within specification. All the tablets possess uniform weight with minor SD values.

Hardness: The hardness of tablet was observed at 3.2 Kg/cm^2. This guarantees decent behavior characteristic of tablets.

Friability: The percent friability was 0.17. As this value was less than 1% it guarantees mechanical tablet stability.

Disintegration: Disintegration time of ATC was found out in water as a disintegrant media at 37°C. Disintegration time of ATC was found to be 5 minutes and 10 seconds.

Dissolution: The dissolution study of the reference product is performed and samples were withdrawn at an interval of 5,10, 15, 20, and 30 minutes. The release profile is depicted in figure 8.

Determination of F2 Value: The resemblance may be related by model-independent or model-dependent methods. For example, the statistical method of multiple variation comparative parameters (weibull function) or the drug dissolved percent at various time intervals or by estimating resemblance factor i.e. F 2 value. The dissolution data will help to find out the F2 value for marketed and prepared tablets. The dissolution test was performed,and the percent drug dissolved was calculated.

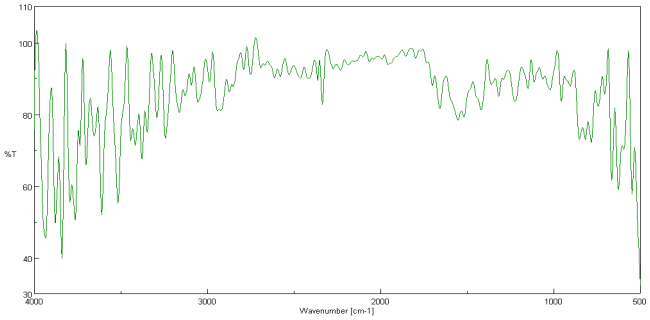

Figure 5: FTIR analysis of separated API

Table2: Melting point of various separated excipients

Sr. No.	*Excipients*	*Melting point Reported °C*	*Melting point Obtained °C*
1	Lactose	232	230
2	Calcium carbonate	Decomposes	N.D
3	Magnesium stearate	117-150	138
4	Croscarmellose sodium	Decomposes	N.D
5	MCC	260-270	258
6	HPC	260	262
7	Polysorbate 80	>100	129

(* ND- not determined due to high melting point)
D- not determined due to high melting point)

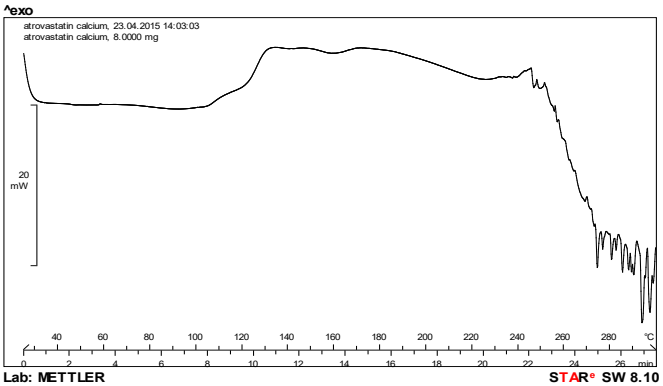

Figure 6: DSC thermogram of separated API

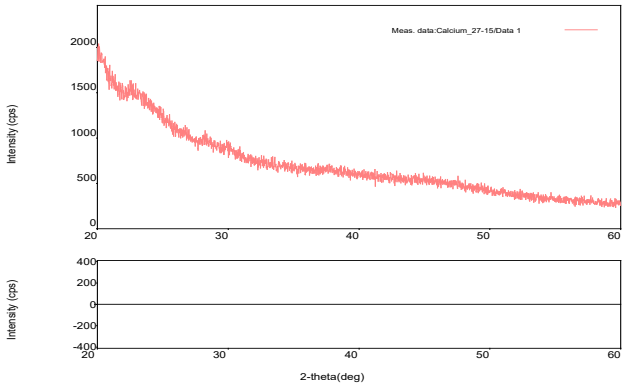

Figure 7: Powder X-ray diffraction of separated ATCr

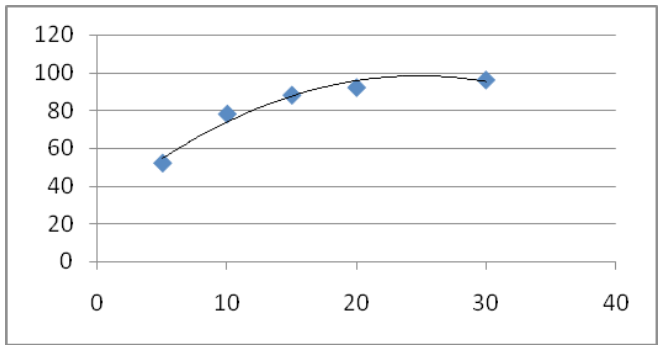

Figure 8: Percent drug release of prepared dosage form

The F2 value calculated was approximately 57.14. The prepared generic dosage form can be bioequivalant as the F2 obtained was over 50 and indicated the resemblance in dissolution profile with less than 15 % deviation.[24-28]

CONCLUSION

The Reverse Engineering of marketed formulation of ATC tablets is effectively carried out by using the differential solubility method. Different analytical methods confirmed the authenticity of the separated ATC and other ingredients. The quantities found after separating the various ingredients of the commercial dosage form of ATC were utilized to develop a generic ATC dosage form. The prepared generic dosage form is equal in strength purity and stability as compared to the commercial dosage form of ATC, which was ascertained with various characterization techniques. Hence, the reverse engineering process using differential solubility technique for preparation of ATC tablet formulation was successfully performed with the preparation of generic dosage form.

ACKNOWLEDGMENT

The authors are grateful to the late Chairman, Mrs. Fatma Rafiq Zakaria, Maulana Azad Educational Trust, Dr. Rafiq Zakaria Campus, to give the required infrastructure for the research work. We are also grateful to Lupin Pharmaceutical Ltd, Aurangabad, Maharashtra, India, for providing ATC sample.

REFERENCES

1. Khan FN, Dehghan MH. Enhanced bio availability and dissolution of atorvastatin calcium from floating microcapsules using minimum additives. Scientia pharmaceutical. 2012;80:215-228.
2. Khan F.N., Dehghan M.H, Enhanced bio availability of atorvastatin calcium from stabilized gastric resident formulation. AAPS Pharmscitech. 2011;12(4):1077-1086.
3. Arvind KB, Vishal K. The role of reverse engineering in the development of generic formulations. Pharm Tech. 2005:55(2);708-713.
4. Peny JM. How Bright Is The Future For Generics? Aon Scrip Magazine. 2003;122: 144-117.
5. Kong Y and seldom J. Pseudo generic products and barriers to entry in pharmaceutical markets. Review of industrial organization. 2004;25:71-86.
6. Davit et al. Comparing generic and innovator drugs: a review of 12 years of bioequivalance data from the USFDA. Ann Pharmacotherapy. 2009;43(10):1583-1597.
7. Rowe RC, Sheskey PJ, Quinn ME. Hand book of pharmaceutical excipients. 6th ed. Pharmaceutical press publication. 2009; 56-57.
8. Rowe RC, Sheskey PJ, Quinn ME. Hand book of pharmaceutical excipients. 6th ed. Pharmaceutical press publication. 2009; 637-639.
9. York P. solid-state properties of powders in the formulation and processing of solid dosage forms. Int. J. Pharm. 1983;14(1): 1-28.
10. Koradia V, Chawla G, Bansal A.K. Comprehensive characterization of the innovator product: Targeting Bioequivalance Generics. J.Generic Med. 2005; 4(6):907-911.
11. Brian RH. The role of particle size in the development of generic dosage products, business briefing.Pharmagenerics. 2003; 56(21): 195-201.
12. Zahid Z et al. Reverse Engineering and Development of Generic Orlistat Formulation. Journal of Innovations in Pharmaceuticals and Biological Sciences. 2016:3(4); 17-25.
13. Ahmed Z, Khan FN & Shaikh, D.A. Reverse engineering and formulation by QBD of olopatadine hydrochloride ophthalmic solution. J. Pharm. Investig. 2018;48: 279-293.
14. Giron D. Thermal Analysis and Calorimetric Methods in Characterization of Polymorphs and Solvates Thermochim. Acta. 1995; 248 (2): 1-59.
15. Raw AS, Furness MS. Regulatory considerations of pharmaceutical solid polymorphism in abbreviated new drug application. Advance drug delivery reviews. 2006;56(6):397-414.
16. Hadkar UB. A handbook of physical pharmacy and physical pharmaceutics, 6th edn. Nirali Prakashan, Pune. 2010;35-36.
17. Rowe RC, Sheskey PJ, Quinn ME. Hand book of pharmaceutical excipients, 6th ed. pharmaceutical press publication. 2009; 656-657.
18. Raymond CR. Paul JS, Marian EQ;eds, handbook of pharmaceutical excipients, 6th edition, London: pharmaceutical press and American pharmacist Association, 2009: 244-246.
19. Shohin IE., Kulinich JI., Ramenskaya GV. dissolution technologies. 2011;18(1):26-29.
20. The Merck Index, 13 th edition, Merck and co. INC, Whitehouse station, NJ,1997, 868.
21. Qureshi F. F2 similarity factor (A deficiency), http//www. drug-dissolution-testing.com.html. [Cited on Saturday 25 April 2014]

22. Tiwary AK, Panpalia GM. Influence of crystal habit on trimethoprim suspension formulation. Pharmaceutical research. 1999 Feb;16(2):261-265.
23. Shohin IE., Kulinich JI., RamenskayaGV. dissolution technologies, 2011 issues, volume 18, issue I, February 2011.
24. Khan FN., Dehghan MH. Enhanced bioavailability and dissolution of atorvastatin calcium from floating microcapsules using minimum additives, Scientia pharmaceutica. 2012;80: 215-228.
25. Khan FN, Dehghan MH. Enhanced bioavailability of atorvastatin calcium from stabilized gastric resident formulation. Aaps Pharmscitech. 2011 Dec;12(4):1077-1086.
26. Raymond CR. Paul JS, Marian EQ;eds, handbook of pharmaceutical excipients,
27. 6th edition, London: pharmaceutical press and American pharmacist Association, 2009: 244-246.
28. The Merck Index, 13th edition, Merck and co. INC, Whitehouse station, NJ,1997, 868