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ANALYTICAL METHOD DEVELOPMENT AND VALIDATION OF TENELIGLIPTINE IN PURE FORM BY USING UV SPECTROPHOTOMETRY

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Abstract

A simple, specific, accurate and precise UV spectrophotemetric method was developed and validated for the estimation of Teneligliptine in pure form. The stock solution was prepared by weighing 100mg of standard Teneligliptine in 100ml of Water in volumetric lask. The final stock solution was made to produce 1000µg/ml with water. Further dilution were prepared as per procedure and were scanned at 243nm. The linearity was found in the concentration range of 10-60µg/ml. the correlation coefficient was 0.999. The regression equation was found to be Y=0.017x. Recovery of Teneligliptine was found to be in the range of 98-102%. The method was validated for limit of detection limit of quantification for estimation of Teneligliptine was found to be 0.56µg/ml and 0.17µg/ml respectively. The Proposed method can be successfully applied for the quantitative determination of eneligliptine in pharmaceutical dosage form.

Keywords: Teneligliptine , UV spectrophotemetric method, Regression equation

Introduction

Analytical chemistry (1) is often described as the area of chemistry responsible for characterizing the composition of matter, both qualitatively (what is present) and quantitatively (how much is present). Analytical chemistry is not a separate branch of chemistry, but simply the application of chemical knowledge. Pharmaceutical Analysis (2) is the branch of chemistry involved in separating, identifying and determining the relative amounts of the components making up a sample of matter. It is mainly involved in the qualitative identification or detection of compounds and quantitative measurements of the substances present in bulk and pharmaceutical preparation. The technique (3) employed in quantitative analysis is based upon the quantitative performance of suitable chemical reactions and either measuring the amount of reagent needed to complete the reaction, or ascertaining the amount of reaction product obtained.

Quality (4) is important in every product or service but it is vital in medicine as it involves life. Unlike ordinary consumer goods there can be no “second quality” in drugs. Quality control is a concept, which strives to produce a perfect product by series of measures designed to prevent and eliminate errors at different stages of production.

Physico-chemical methods (5, 6) are used to study the physical phenomenon that occurs as a result of chemical reactions. Among the Physico-chemical methods, the most important are ptical (Refractometry, Polarimetry,

Emission, Fluorescence methods of analysis, Photometry including Photocolorimetry and Spectrophotometry covering UV-Visible and IR regions and Nephelometry or Turbidimetry) and chromatographic (Column, Paper, TLC, GLC, HPLC) methods. Methods such as Nuclear Magnetic Resonance and Para Magnetic Resonance are becoming more and more popular. The combination of Mass Spectroscopy with as Chromatography and Liquid Chromatography are the most powerful tools available. The chemical methods include the gravimetric and volumetric procedures which are based on complex formation; acid - base, precipitation and redox reactions. Titrations in non-aqueous media and complexometry have also been used in pharmaceutical analysis.

The number of new drugs is constantly growing. This requires new methods for controlling their quality. Modern pharmaceutical analysis must need the following requirements.

1. The analysis should take a minimal time.
2. The accuracy of the analysis should meet the demands of Pharmacopoeia.
3. The analysis should be economical.
4. The selected method should be precise and selective.

These requirements are met by the Physico-chemical methods of analysis, a merit of which is their universal nature that can be employed for analyzing organic compounds with a diverse structure. Of them, Visible Spectrophotometry is generally preferred especially by small scale industries as the cost of the equipment is less and the maintenance problems are minimal.Instrumental methods of Chemical analysis:

Instrumental method is an exciting and fascinating part of chemical analysis that interacts with all areas of chemistry and with many other areas of pure and applied sciences. Analytical instrumentation plays an important role in the production and evaluation of new products and in the protection of consumers and environment. This instrumentation provides lower detection limits required to assure safe foods, drugs, and water air. Instrumental methods are widely used by Analytical chemists to save time, to avoid chemical separation and to obtain increased accuracy.

Materials and Methods

Requirements:

The following materials used were either AR/LR grade or the Possible Pharma grade available as supplied by the manufacturer or supplier without further purification or investigation.

S. No	Materials	Source
1	Teneligliptine	Aurobindo pharma Pvt.Ltd, Jadacherla.
2	Methanol	Merck Mumbai Ltd, Mumbai.
3	Ethanol	Merck Mumbai Ltd, Mumbai.
4	Sulphuric acid	Merck Specialities Pvt Ltd, Mumbai.
5	Chloroform	Merck Specialities Pvt Ltd, Mumbai.
5	Hydrochloric acid	Merck Specialities Pvt Ltd, Mumbai.
6	Acetonitrile	Merck Mumbai Ltd, Mumbai.
7	Potassium dihydrogen orthophosphate	Qualigens,Mumbai.
8	Ortho phosphoric acid	Qualigens,Mumbai.

S. No	Equipments	Source
1	UV Spectrophotometer	Elico SL 210 UV/VIS Spectrophotometer and Systronics 117 UV-VIS Spectrophotometer.
2	Sonicator	Wensar

UV SPECTROSCOPY:

Systronics 117 UV-VIS spectrophotometer was used with 1 cm matched quartz cells. The data processing was performed using UV –probe software.

UV method development:

The parameters for the development were as follows:

1) Selection of solvent

2) Selection of wavelength

Validation of proposed method:

The parameters for the validation were as follows:

1) Linearity

2) Accuracy

3) Precision

4) Robustness

5) Ruggedness

6) Limit of Detection

7) Limit of Quantification

Selection of solvent:

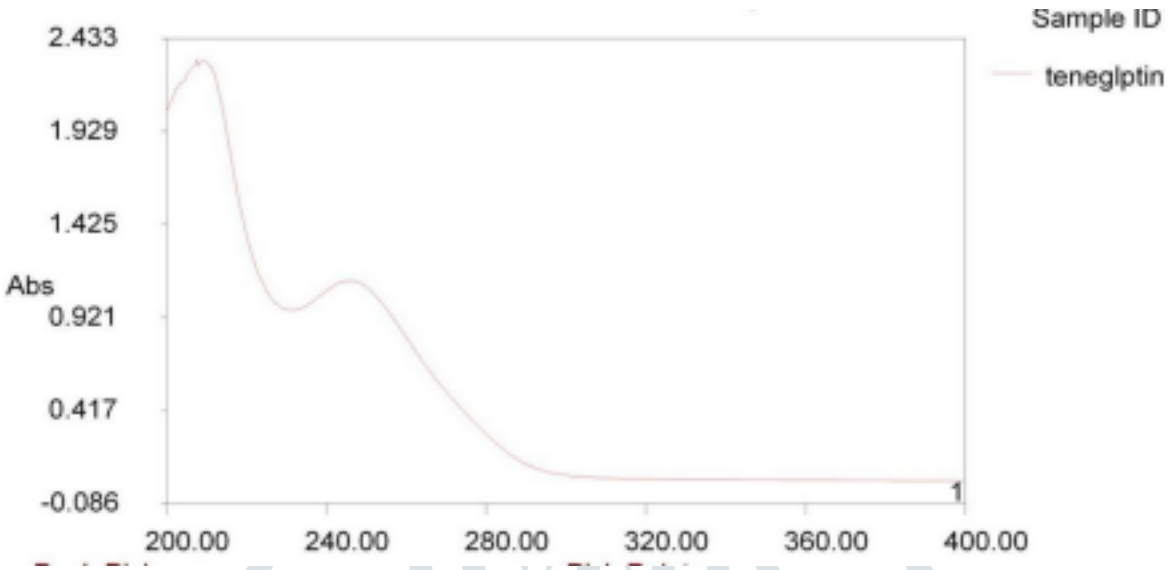
In order to select suitable solvent for determination of teneligliptine various solvent like methanol, acetonitrile, ethanol, sulphuric acid, hydrochloric acid Chloroform Buffer & Water tried for the solubility studies and it was found that teneligliptine was freely soluble in water. In the present investigation distilled water was selected as a solvent.

METHODOLOGY

Selection of wavelength:

10 µg/ml of teneligliptine was scanned in the range of 200-400 nm.

UV Spectrum for Teneligliptine



Wavelength(nm)

VALIDATION OF THE METHOD:

The method was validated in terms of parameters like Linearity, accuracy, precision, ruggedness, and robustness.

Preparation of Stock Solution:

100 mg of teneligliptine was dissolved in water in a 100 ml of volumetric flask and the solution was made up to volume with Distilled water.

Preparation of Standard working solution:

5ml of standard solution was dissolved in 50 ml of volumetric flask and the solution was made up to volume with Distilled water.

1. Linearity:

To evaluate the linearity, serial dilution of analyte were prepared from the standard working solution was diluted with solvent to get a series of concentration ranging from 10, 20, 30, 40, 50 and 60 µg/ml. The prepared solutions were filtered through whatmann filter paper (No.41). Calibration curve was constructed by plotting the absorbance on Y- axis against the concentration on X- axis

2. Precision:

The precision of the analysed method was studied by analysis of multiple sampling of homogeneous ample. The precision is expressed as standard deviation (or) relative standard deviation. The precision of the method was demonstrated by intra-day and inter –day variation studies.

2.1 Intraday precision:

In the intraday studies, the standard solutions (10, 20, 30, 40, 50 and 60 µg/ml) was analysed for six times in different time interval within day. %RSD was calculated

2.2 Inter day precision:

In the inter-day variation studies, the standard solution (10, 20,30,40,50 and 60µg/ml) was analysed for six times in different days. % RSD was calculated presented

Accuracy:

Recovery studies by the standard addition method performed with a view to justify the accuracy of the proposed method. Previously analysed samples of teneligliptine (45,55,and 65µg/ml) were spiked with 80,100,120% extra teneligliptine standard and the mixture wereanalysed by the proposed method. The experiment was performed in triplicate and recovery of the pure drug. %RSD was calculated and reported

4. Sensitivity:

The sensitivity of measuring of teneligliptine by use of the proposed method was estimated in terms of the limit of detection (LOD) and the limit of quantitation (LOQ).The LOD and LOQ were calculated by the use of the equation LOD=3.3 × σ/s and LOQ=10 × σ/s where σ is the standard deviation of response and S is the slope of the calibration curve LOD & LOQ Values are reported in Table 7.10 & Table 7.11.

5. Ruggedness:

Ruggedness is a measure of the reproducibility of a test result under normal, expected operating condition from instrument to instrument and analyst to analyst. The ruggedness of the method was determined by carrying out the experiment by different operators. The result of ruggedness testing is reported

6. Robustness:

Robustness is a measure of capacity of a method to remain unaffected by small, but deliberate variation in the method condition, and is indication of the reliability of the method. A method is robustness, if it is unaffected by small changes in operating condition .To determine the robustness of this method, the experimental condition were deliberately altered at three different levels and responses were evaluated. Variation of wavelength (241nm and 245nm) had no significant effect on the absorbance of 50µg/ml solution, indicating that the method was robustness.

RESULTS & DISCUSSION

UV SPECTROSCOPY METHOD

Characteristic parameters of Teneligliptine for the proposed UV spectroscopy method:

Parameters	UV
Calibration range (µg/ml)	10-60(µg/ml)
Wavelength	243nm
Regression equation (y*)	0.017x
Slope	0.017

Correlation Coefficient((r2)	0.999
LOD (µg/ml)	0.56
LOQ (µg/ml)	1.7

Y*= bx+a where x is the concentration of Teneligliptine in µg/ml and Y is the absorbance at the respective ʎ max.

VALIDATION OF ANALYTICAL METHOD:

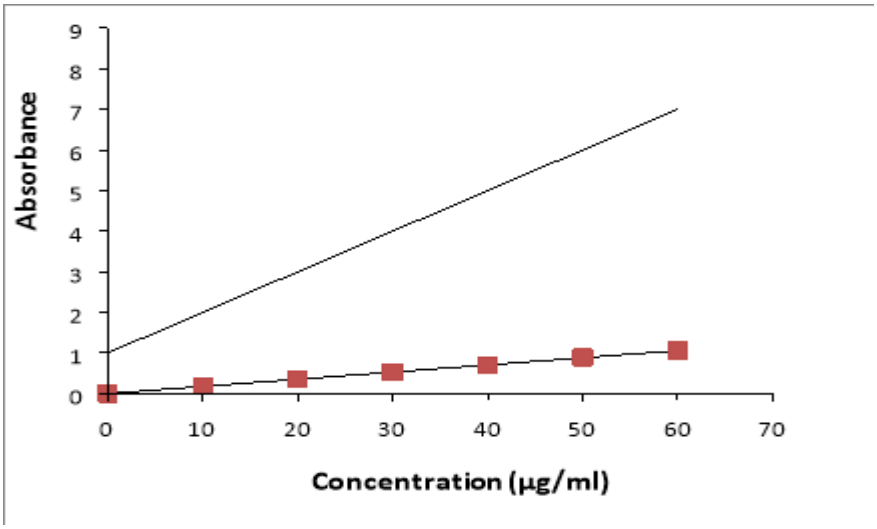
Validation of an analytical method is the process to establish by laboratory studies that the performance characteristic of the method meets the requirement for the intended analytical application .Performance characteristic were expressed in terms of analytical parameters.

1. Linearity:

Calibration graph were plotted using absorbance of standard drug versus concentration of standard drug solution. Linear regression data showed a good linear relationship over a concentration range 10-60µg/ml.

Calibration data of Teneligliptine

S.NO	Concentration(µg/ml)	Concentration
1	0	0
2	10	0.172
3	20	0.36
4	30	0.524
5	40	0.697
6	50	0.891
7	60	1.056



Calibration curve of Teneligliptine

Observation:

The correlation coefficient for Teneligliptine was found to be 0.999 respectively.

The linearity range for Teneligliptine was found to be 10-60µg/ml.

Precision:

The precision of the analytical method was studied by analysis of multiple sampling of homogenous sample. The precision results were expressed as standard deviation or relative standard deviation.

Intraday precision

Absorbance

S. No	Conc µg/ml	1	2	3	4	5	6	AVG	SD	%RSD
1	10	0.176	0.172	0.177	0.175	0.170	0.177	0.174	0.0029	1.67
2	20	0.365	0.368	0.367	0.364	0.363	0.364	0.365	0.0019	0.52
3	30	0.523	0.521	0.523	0.525	0.520	0.521	0.522	0.0018	0.34
4	40	0.692	0.693	0.699	0.693	0.692	0.697	0.694	0.003	0.43
5	50	0.891	0.884	0.919	0.915	0.881	0.89	0.896	0.0188	1.6
6.	60	1.056	1.049	1.053	1.051	1.056	1.052	1.053	0.0028	0.266

Acceptance criteria:

%RSD of the six replicate injections should not more than 2.0%

Inter day precision result for Teneligliptine

S. No	Conc µg/ml	Absorbance						AVG	SD	%RSD
		1	2	3	4	5	6			
1	10	0.178	0.172	0.173	0.174	0.179	0.174	0.176	0.0028	1.6
2	20	0.372	0.361	0.374	0.369	0.359	0.364	0.366	0.006	1.63
3	30	0.526	0.525	0.521	0.527	0.531	0.531	0.527	0.0038	0.72
4	40	0.691	0.697	0.689	0.685	0.693	0.695	0.691	0.0053	0.76
5	50	0.903	0.919	0.901	0.886	0.932	0.901	0.907	0.016	1.7
6	60	1.062	1.057	1.053	1.054	1.061	1.059	1.058	0.0052	0.491

Acceptance criteria:

%RSD of the six replicate injections should not more than 2.0%

Accuracy:

Observation for accuracy standard (50µg/ml)

S. No	Concentration (µg/ml)	Absorbance
1	Set-1	0.835
2	Set-2	0.848
3	Set-3	0.843
4	**AVG**	0.842
5	**SD**	0.0065
6	**%RSD**	0.77

Observation for accuracy for 80% (45µg/ml)

S. No	Concentration (µg/ml)	Absorbance
1	Set-1	0.776
2	Set-2	0.777
3	Set-3	0.770
4	**AVG**	0.77
5	**Result**	45.8
6	**%Rec**	101.8
7	**SD**	0.003
8	**%RSD**	0.38

Observation for accuracy standard 100% (55µg/ml)

S. No	Concentration (µg/ml)	Absorbance
1	Set-1	0.929
2	Set-2	0.927
3	Set-3	0.931
4	**AVG**	0.929
5	**Result**	55.1
6	**%Rec**	100.3
7	**SD**	0.002
8	**%RSD**	0.21

Observation for accuracy standard 120% (65µg/ml)

S .No	Concentration (µg/ml)	Absorbance
1	Set-1	1.123
2	Set-2	1.104
3	Set-3	1.102
4	**AVG**	1.109
5	**Result**	65.4
6	**%Rec**	101.3
7	**SD**	0.0115
8	**%RSD**	0.99

For accuracy summary

Sample (%)	Initial amount (µg/ml)	Amount added (µg/ml)	Amount recovered (µg/ml)	%Recovery ±SD·	%RSD
80	40	5	45.8	101.8±0.003	0.38
100	50	5	55.1	100.3±0.002	0.21
120	60	5	65.4	101.3±0.115	0.99

*Average of three determinations

Acceptance criteria:

%Recovery should be within the range of 99-102%

%RSD should be less than 2.

Sensitivity:

Limit of detection of (LOD) and limit of quantitation (LOQ) were determined from standard and slope method as per ICH guidelines, for Teneligliptine LOD was found to be 0.56 µg/ml and LOQ was found to be 1.7 µg/ml.

Observation of Limit of Detection:

S .No	Slope	SD of precision	LOD
1	0.017	0.0029	0.56

Observation of Limit of quantitation:

S. No	Slope	SD of precision	LOQ
1	0.017	0.0029	1.7

Ruggedness:

For Ruggedness (Analyst to Analyst)

S. No	Analyst-1		Analyst-2	
	Concentration (µg/ml)	Absorbance	Concentration (µg/ml)	Absorbance
1	50	0.899	50	0.911
2	50	0.876	50	0.931
3	50	0.899	50	0.922
	AVG	0.891	**AVG**	0.921
	SD	0.013	**SD**	0.0107
	%RSD	1.45	**%RSD**	1.08

Acceptance criteria:

%RSD of the six replicate injections should not more than 2.0%

For Ruggedness (Instrument to Instrument)

S. No	Systronics 117 UV-Spectrophotometry		ELICO 210 UV-Spectrophotometry	
	Concentration (µg/ml)	Absorbance	Concentration (µg/ml)	Absorbance
1	5	0.517	5	0.514
2	5	0.520	5	0.518

3	5	0.516	5	0.523
	AVG	0.51	**AVG**	0.518
	SD	0.002	**SD**	0.004
	%RSD	0.39	**%RSD**	0.77

Acceptance criteria:

%RSD of the six replicate injections should not more than 2.0%

For 241 and 245 wavelengths

S. No	Concentration	Absorbance (at 241 nm)	Absorbance (at 245 nm)
1	Set-1	0.882	0.892
2	Set-2	0.882	0.878
3	Set-3	0.894	0.882
	AVG	0.886	0.884
	SD	0.006	0.0072
	%RSD	0.7	0.8

Robustness Summary

S. No	Condition	Modification	Mean absorbance ± SD*	% RSD for absorbance
1	Wavelength(nm)	241	0.886±0.006	0.7
		245	0.884±0.007	0.8

*Average of the three determination.

Acceptance criteria:

%RSD should not be more than 2.

DISCUSSION

In the present work, an attempt was made to provide a newer, sensitive, simple, accurate and low cost UV-Visible Spectroscopic method. It is successfully applied for the determination of Teneligliptine in pharmaceutical preparations without the interferences of other constituent in the formulations.
The optimum wavelength for detection was 243 nm at which better detector response for the drug were obtained. The calibration was linear in concentration range of 10-60 µg/ml for Teneligliptine respectively**.** The sensitivity for the drug has been calculated and the LOD and LOQ of the Teneligliptine was found to be 0.56 µg/ml and 1.7 µg/ml .
The low values of % R.S.D. indicate the method is precise and accurate. The mean recoveries were found in the range of 99-102% in the Table 7.9 for Teneligliptine respectively.
Ruggedness of the proposed methods was determined by analysis of aliquots from homogeneous slot by different analysts, using similar operational and environmental conditions; the % R.S.D. reported was found to be less than 2 %
The proposed method was validated in accordance with ICH parameters and the results of all methods were very close to each other as well as to the label value of commercial pharmaceutical formulation. Therefore, there is no significant difference in the results achieved by the proposed method.
Hence it is suggested that the proposed is UV / VIS Spectrophotometric method can be effectively applied for the routine analysis of Teneligliptine in bulk and in tablet formulation.

CONCLUSION

For routine analytical purpose it is always necessary to establish method capable of analysing huge number of samples in a short time period with due accuracy and precision.
Teneligliptine is not official in Pharmacopoeia. There is few analytical methods appeared in the literature for the determination of the Teneligliptine. In literature review we have method only for the estimation of the above drugs of concern in individually or in combination of others
In view of the above, a simple and specific analytical method was planned to develop with sensitivity, accuracy, precision and economical.
In the present investigation of UV spectrophotometric method for the quantitative estimation of Teneligliptine in pure drug and Pharmaceutical formulation has been developed and validated.
The proposed UV method is more sensitive, accurate and precise and is suggested for routine analysis.

References

- David Harvey. Modern Analytical Chemistry. 1st ed. United States of America: Harcourt Brace & Company; 1997. p. 2, 578-80
- Sharma BK. Instrumental Methods of Chemical Analysis. 13th ed. Meerut: Goel Publisher House; 2000. p.1, 7.
- Jeffery GH, Bassett J, Mendham J, Denney RC. Vogel's Text book of Quantitative Chemical Analysis. 5th ed. New York: John Wily & sons, Inc.; 1989. p.7, 668-9.
- Sethi PD. Quantitative Analysis of Drugs in Pharmaceutical Formulations. 3rd ed. New Delhi: CBS Publishers and Distributors; CBS Publishers and Distributors 1986.p.1, 20.Willard HH, Merrit LL, Dean JA, Settle FA. Jr. Instrumental Methods of Analysis. 6th ed.New Delhi: CBS Publishers and Distributors; 1989.
- Day RA, Underwood AL. Quantitative Analysis. 4th ed. New Delhi: Prentice Hall; 1986.
- Ravi Sankar S. Text book of pharmaceutical analysis. 3rd ed. Tirunelveli: Rx Publications; 2001. p. 1-1.
- Chatwal, Anand. Instrumental Methods of Chemicals Analysis. 1st ed. Mumbai: Himalaya Publishing House; 2000. p. 184. Elhance DN. Fundamentals of statistics, Kitab mahal; ed XLVII, 2003; 10.Shodhganga.inflibnet.ac.in > bitstream

- U Berry RI, Nash AR. Pharmaceutical Process Validation and Analytical method validation. 1st ed. New York: Marcel Dekker Inc; 1993. p. 411-2.
- Asian Guideline for Validation of Analytical Procedure Adopted from ICH Guideline, Q2A27 Oct. 1994 and ICH Q2B, 6th Nov. 1994.
- V.D.Barhate, spectrophotometric determination of an antidiabetic drug teneligliptine bulk and pharmaceutical formulations, World journal of pharmaceutical research 2016,vol-5,p- 1625 - 1632.
- Ashim kumar sen, Analytical method development and validation for simultaneous estimation of Teneligliptin hydrobromide hydrate and Metformin hydrochloridefrom it's pharmaceutical dosage form. Journal of Applied Pharmaceutical Science, 2016, Vol. 6 (09), p- 157-165.
- Vishnu C. Shinde, Development and validation of high performance thin layer chromatographic (HPTLC) method for estimation of teneligliptin hydrobromide in pharmaceutical preparation, scholars research library,Der pharmacia Letter, 2016, 8(8):291-301.
- Atul T. Hemke , Hplc estimation of teneligliptin from Tablet dosage form Asian journal of pharmaceutical Analysis and Medicinal chemistry. 2016, 4(3), 2016, 148-156.
- Sohan S.Chitlange, Estimation of anti-diabetic teneligliptin hydrobromide hydrate by rp-hplc and derivative spectroscopic method, Indo American journal of pharmaceutical research,2016, vol-6, p- 123-132.
- T.N.V. Ganesh kumar Method development, validation, andstability studies of teneligliptin by RP-HPLC and identification of degradation products by UPLC tandem mass spectroscopy, Journal of Analytical science and technology,2016, 6(5), 1-8.
- Shailesh V. Luhar Simultaneous Estimation of Teneligliptin Hydrobromide Hydrate and its Degradation Product by RP-HPLC Method, Journal of pharmaceutical science and Bioscientific research 2016,6(3), 254-261.
- Raja haranadha babu chunduri, Development and validation of Lc-ms/ms method for quantification of teneligliptin in human plasma and its application to apharmacokinetic study, World Journal of pharmacy and pharmaceutical sciences, 2016, 5(5), 838-850.
- https://en.wikipedia.org/wiki/Teneligliptine.