# Visible Spectrophotometric Methods for the Determination of Nateglinide in Tablets through Schiff's Base Formation

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**Two simple spectrophotometric methods for the analysis of nateglinide in pure form or pharmaceutical formulations have been developed based on the reaction of the drug with aromatic aldehydes, vanillin and paradimethyl amino cinnamaldehyde in acidic medium producing coloured Schiff's bases having λmax at 580 nm and 420 nm, respectively. Good agreement with Beer's law was found in the range of 25-150 μg/ml for method A and 20-140 μg/ml for method B. The methods are simple, precise and accurate with excellent recovery of 98-102% and also do not require any separation of soluble excipients in pharmaceutical preparations. The results obtained are reproducible with coefficient of variation of less than 1.0%.**

Nateglinide (NTG) is chemically N-(trans-4-isopropylcyclohexanecarbonyl)-D-phenylalanine. It is a new drug and is not official in any pharmacopoeia. It is a non–sulfonylurea insulin secretagogue[1]. Literature survey reveals the presence of HPLC[2,3] methods for its estimation. As there is no report of nateglinide analysis by visible spectrophotometry, the need for a fast, low cost and selective method are obvious especially for routine quality control analysis of pharmaceutical products containing NTG. In this communication, the authors report two simple, sensitive and reproducible spectrophotometric methods for the determination of NTG in pure form as well as in tablets containing nateglinide. The amino group present in the drug reacts with vanillin and para dimethylaminocinnamaldhyde (PDAC) in acidic conditions to give a Schiff's base by condensation which has absorption maximum at 580 nm (method A) and 420 nm (method B).

An Elico SL 171 spectrophotometer with 1 cm matched quartz cells was used in the present study. All reagents used were of analytical grade. A 0.4% w/v PDAC (BDH, Mumbai) solution was prepared by dissolving 400 mg of the reagent in 100 ml of chloroform. A 0.3% w/v vanillin (Qualigens, Mumbai) was prepared by dissolving 300 mg of the reagent in 100 ml of chloroform. Concentrated sulfuric acid and methanol were obtained from Qualigens, Mumbai and were used as such. All reagents were freshly prepared and used. NTG tablets (glinate and natelide) were obtained from local market.

A standard solution containing 1 mg/ml of NTG was prepared in methanol by dissolving 100 mg of pure NTG in 100 ml of methanol. From this solution, working standard solutions were prepared by dilution with methanol, 500 μg/ml (method A) and 400 μg/ml (method B). Two brands of commercial tablets of NTG were analyzed by the proposed methods. In each method, 10 tablets were accurately weighed and powdered. In each case, tablet powder equivalent to 100 mg of NTG was treated with sufficient quantity of methanol and diluted to 100 ml with the same solvent and filtered. The filtrate was suitably diluted and analyzed as given under the assay procedure for bulk samples.

Method A comprised of transferring into a series of 10 ml volumetric flasks, aliquots of NTG (0.5–3.0 ml, 500 μg/ml). This was followed by addition of 2 ml of vanillin and 3 ml concentrated sulfuric acid and then making up the total

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volume in each volumetric flask to 7 ml with methanol. The solutions were mixed and placed in a hot water bath for 15 min. The flasks were then cooled and the volume made up to 10 ml with methanol. After 10 min, absorbance was measured at 580 nm against a reagent blank. The concentration of NTG present was deduced from the calibration graph.

In method B, aliquots of standard drug solution (0.5–3.5 ml, 400 μg/ml) were transferred into a series of 10 ml volumetric flasks. Then 3 ml of PDAC and 3 ml concentrated sulfuric acid were added and the total volume in each flask was brought to 7 ml with methanol and the flasks were placed in a hot water bath for 10 min. The flasks were cooled to room temperature and volume in each flask was made up to 10 ml with methanol. After 15 min, absorbance was measured at 420 nm against a reagent blank. The concentration of NTG present was computed from calibration curve.

Optimum operating conditions used in the procedures were established adopting variation of one variable at a time method. The optical characteristics of the methods are presented in Table 1. The precision and accuracy of the methods was tested by measuring six replicate samples of the drug in Beer's law limits. Tablets containing NTG were successfully analyzed by the proposed methods. The results are presented in Table 2. None of the excipients usually employed in the formulation of tablets interfered in the analysis of NTG by the proposed methods. As an additional check of accuracy, recovery experiments were performed by standard addition method. When tablets containing NTG were analyzed, the results obtained by the proposed methods were in good agreement with the labeled amounts. The recovery with the methods was found to be 99–101%.

Aromatic aldehydes have lead to numerous applications as analytical reagents. Aldehydes have been applied to the colorimetric determination of primary alkylamines[4] and primary aromatic amines in acidic medium. More specifically, PDAC has been used for the determination of trace amounts of hydrazine. Further, the condensation of indole derivatives in acidic medium gives coloured products[6] that can also be estimated spectrophotometrically. The proposed methods are simple, convenient, accurate, sensitive and reproducible. These methods can be employed for routine analysis of NTG either in its pure form or from tablets.

TABLE 1: OPTICAL CHARACTERISTICS AND PRECISION OF THE PROPOSED METHODS

| Parameter | Method A | Method B |
|---|---|---|
| λmax (nm) | 580 | 420 |
| Beer's law Limit (μg/ ml) | 25 – 150 | 20 – 140 |
| Molar absorptivity ($1mol^{-1}$ $cm^{-1}$) | $3.92 \times 10^4$ | $8.14 \times 10^3$ |
| Sandell's sensitivity | | |
| (mg $cm^{-2}$ per 0.001 absorbance unit) | 0.010 | 0.0480 |
| Regression equation (y = a + bC)* | | |
| Slope (b) | $1 \times 10^{-2}$ | $1.8 \times 10^{-3}$ |
| Intercept (a) | $4 \times 10^{-3}$ | $3.1 \times 10^{-2}$ |
| Correlation coefficient (r) | 0.9987 | 0.9991 |
| Relative standard deviation (%)** | 0.32 | 0.781 |
| % Range of error (confidence limits)** | | |
| 0.05 level | 0.339 | 0.318 |
| 0.01 level | 0.532 | 0.512 |
| % Error in bulk samples*** | 0.85 | 0.59 |

*Y=a+bC, where C is concentration of analyte and Y is absorbance unit, **average of six determinations, ***average of three determinations.

TABLE 2: ASSAY OF NTG IN TABLETS

| Formulation | Label claim mg/tablet | Amount found by proposed methods | | % Recovery by proposed Method* |
|---|---|---|---|---|
| | | Method A | Method B | |
| Tablet 1 | 60 | 59.7±0.05 | 59.5±0.06 | 99.9±0.08 |
| Tablet 2 | 120 | 119.9±0.08 | 119.8±0.05 | 99.9±0.92 |

Tablet 1 is Glinate, 60 mg manufactured by Glenmark Pharma, Mumbai and tablet 2 is Natelide, 120 mg, Alembic, Vadodara. *Recovery of 10 mg added to the pre-analyzed pharmaceutical dosage forms (average of 3 determinations).


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## REFERENCES


1. Meenakshi, A. and Seth, R.K., Eds; In; Current Index of Medical Specialities, Mumbai, 2002, 4, 21
2. Weaver, M.L. and Orwing, B.A., **Drug Metab. Dispos.,** 2001, 29, 415.
3. Horton, E.S. and Clinkingbeard, C., **Diabetes Care,** 2000, 23, 1660.
4. Pesez, M. and Petit, A., **Bull. Soc. Chem. Fr.,** 1947, 122, 58.
5. Sawicki, E. and Johnson, H., **Chemist–Analyst,** 1966, 101, 165.
6. Byrum, P. and Turnbull, H., **Talanta,** 1963, 10, 1217.


# FTIR-Spectrum of Galactomannan Extracted from *Trigonella foenum - graceum*


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**Guar is used in millions of tones annually by various industries and is employed medicinally as well. The galactomannan portion of the guar seed is found to be active and responsible for its numerous industrial and medicinal applications. Guar galactomannan has a galactose-mannose ratio (G:M) of 1:2 and possesses a very high viscosity. The objective of this study was to explore some other source of galactomannan having different G:M ratio and to study its effect on medicinal applications. Fenugreek is one such promising source that offers a galactomannan having a G:M ratio of 1:1. The extracted galactomannan from fenugreek was compared with guar galactomannan on the basis of FTIR-spectra. The peaks and their accompanying shoulders, in the FTIR-spectra of the two compounds, were found to overlap closely.**


The literature survey on *Trigonella foenum-gracium* or fenugreek, reports its numerous (about 30) medicinal applications[1,2]. The mechanism and the specific constituent responsible for most of its activities are not known or simply the whole seed powder is reported to have a particular activity in question in most of the cases[3-7]. Hence, there is a need to carry out research work to correlate the active constituents of this little seed with its numerous therapeutic actions.


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