



**INTERNATIONAL JOURNAL OF
PHARMACEUTICAL SCIENCES**
[ISSN: 0975-4725; CODEN(USA): IJPS00]
Journal Homepage: <https://www.ijpsjournal.com>



Research Article

Spectral Insights: Bathochromic Shifts in Visible Spectra of Nicotinamide with Ninhydrin

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ARTICLE INFO

Published: 10 Aug 2025

Keywords:

Nicotinamide, Ninhydrin,
Ammonium molybdate, and
Bathochromic shift

DOI:

10.5281/zenodo.16789120

ABSTRACT

Most of the pharmaceuticals are analyzed using UV-Visible methods due to their simplicity, better understanding for students, and economy. Ammonia and primary amino groups, whenever heated with ninhydrin in the presence of ammonium molybdate, form a rhenium purple color. The simple, precise, and cost-effective UV-Visible method has been developed and validated for the determination of Nicotinamide in bulk drugs. The method is based on the reaction of ninhydrin with amide present in Nicotinamide in the presence of ammonium molybdate to form a blue chromogen, which is determined spectrophotometrically at 761nm. The effects of variables such as temperature, heating time, concentration of colour-producing reagent, and stability of colour were investigated to optimize the procedure. The method is validated for linearity, accuracy, and precision parameters. The rhenium purple color of chromogen is shifted towards longer wavelengths (from 570 nm to 761 nm). Thus, Nicotinamide shows a Bathochromic shift due to the presence of the amide group. This reaction is quantitatively proceeding at $970 \pm 1^\circ\text{C}$ in 5 minutes. Beer Lambert's law is obeyed in the concentration range of 0-125 $\mu\text{g/ml}$ and is described by the regression equation $y = 0.0047x$ with a regression coefficient (r^2) of 0.9997. For Nicotinamide, the value of molar absorptivity and Sandell's sensitivity are $2.3487 \times 10^2 \text{ L/mol/cm}$ and $0.2166 \mu\text{g/cm}^2$, and LOD and LOQ are found to be 0.0238 and $0.0722 \mu\text{g/ml}$, respectively. The statistically validated results indicate that the proposed method is simple, precise, green, and cost-effective.

INTRODUCTION

Nicotinamide (NCT), also called niacinamide, is 3-pyridine carboxamide (Figure. 1-A). NCT is an amide form of nicotinic acid or niacin (Figure 1-

B). NCT is a white crystalline powder or colorless crystals; it has a molecular weight of 122.12 g/mol. It is also one of the hydrophilic B vitamins, called vitamin B₃. Many multivitamins and supplementary pharmaceutical preparations

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Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.



contain NCT¹. The main sources of niacin include meat, liver, green leafy vegetables, wheat, oats, palm kernel oil, legumes, yeast, mushrooms, nuts, milk, fish, tea, and coffee². It is also found in many dermal preparations, such as solutions, creams, and gels, whether as a single active ingredient or with other active ingredients. NCT is used for many skin conditions, including acne with variable concentration (3/4/5 % w/w).

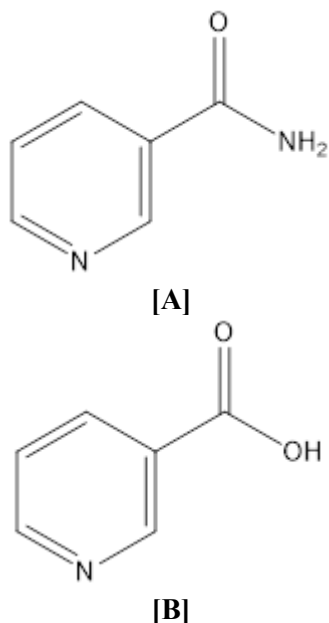


Figure 1: Chemical Structures of Nicotinamide [A] and Nicotinic acid [B]

Several classical and advanced analytical methods are available for Nicotinamide estimation in various samples, like Titrimetry³, Colorimetry^{4,5}, UV Spectroscopy⁶⁻¹⁰, HPLC¹¹⁻¹⁶, HPTLC¹⁷⁻¹⁹ Capillary Electrophoretic Chromatography^{20,21}. These methods, individually or in combination, are crucial for ensuring the quality, efficacy, and safety of Nicotinamide-containing formulations. Each method has its unique advantages depending upon the sample type, required sensitivity, and specific application.

The quantification of Nicotinamide has been a fascinating area of research, and visible spectrophotometry indeed stands out as a method of choice. Its ability to strike a balance between

accessibility and analytical effectiveness makes it highly favoured. Researchers have leveraged it for its simplicity in operation and excellent selectivity for distinguishing compounds. Additionally, its cost-effectiveness makes it a practical option for various industrial and academic laboratory settings, including those with budget constraints. Its accuracy and precision ensure reliable results, which are crucial in pharmaceutical analysis.

As per the author's knowledge, to date, there is no documented method showing bathochromic effect on Nicotinamide when it is reacted with ninhydrin in the presence of ammonium molybdate. The objective of this research work is to determine how the molecular environment created by ninhydrin and ammonium molybdate affects Nicotinamide's electronic transitions, leading to the bathochromic shift

The reaction involving ninhydrin and the amide group in Nicotinamide, catalysed by ammonium molybdate to produce a blue chromogen, is a brilliant choice for detection, as shown in Scheme 1. Measuring the absorbance spectrophotometrically at 761 nm ensures precise quantification due to the distinct peak of the chromogen at that wavelength. This approach combines chemistry and analytical techniques to achieve sensitivity and selectivity in the analysis.

The bathochromic shift from 570 nm to 761 nm, as observed in the rhenium purple color of the chromogen, reflects the interplay between molecular structure and light absorption. This specific shift due to the amide group in Nicotinamide underscores how structural nuances can influence the chromophore's properties. By leveraging this, the spectrophotometric method at 761 nm ensures both precision and specificity in detection, blending the art and science of analytical chemistry.

2. MATERIALS AND METHODS

2.1 Instrumentation

A SHIMADZU 1800 UV-Visible spectrophotometer with 10²mm matching quartz cells was used for absorbance measurements. The UV spectra were recorded over the wavelength range 400-800 nm.

2.2 Chemicals and Reagents

Reagents and all chemicals employed were of Analytical Grade. Nicotinamide was obtained as a gift sample from Wockhardt Pharmaceuticals Pvt. Ltd. (Aurangabad, India). All other reagents and chemicals used were of Analytical Grade.

2.3 Stock Solution of Nicotinamide

Weigh accurately 100 mg of Nicotinamide and dissolve in 100 mL of 0.1 M HCl (1000 ppm). Stock Solution [A], further dilute 10 mL of Stock Solution [A] with 0.1 M HCl up to 100 mL (100 ppm) and refer to as Stock Solution [B].

2.4 Preparation of 0.1 M Hydrochloric Acid (HCl) Solution

Dissolve 21.25 mL of Concentrated Hydrochloric Acid in 100 mL of Distilled Water.

2.5 Preparation of 2M Glacial Acetic Acid

Weigh accurately 11.5 mL of Glacial Acetic Acid and dissolve in 100 mL of Distilled water.

2.6 Preparation of 0.2 M Ninhydrin Solution

Take 50 g of Ninhydrin and dissolve in 23.75 g of Ethanol and stir until it gets completely dissolved. To this solution, add 5 mL of 2M glacial acetic acid.

2.7 Preparation of 0.2M Ammonium Molybdate Solution

Accurately weigh 12.35 g of Ammonium Molybdate and dissolve in 50 mL of deionized water or distilled water.

2.8 Working Principle of Chromogen Formation

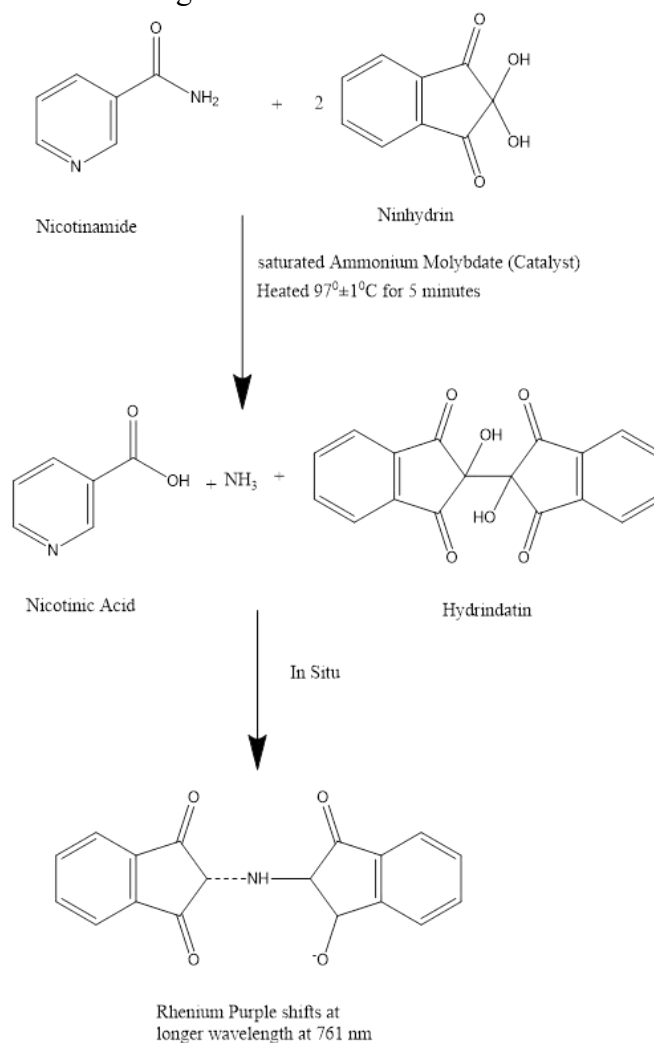
Recently added newly developed analytical methods with good properties, such as selectivity and sensitivity, were not sufficient, so modern analytical methods also need to be green. Hence, the scope of this present study is to develop a method concerning two different objectives: i] lowering the heating time, ii] and increasing the quantitative and qualitative analysis. Both factors are of great importance in reducing time and increasing the accuracy rate. In this present study, Nicotinamide developed a dark blue colored product with ninhydrin in the presence of ammonium molybdate, and further spectrophotometric analysis was carried out using 0.1M HCl as a Blank solution.

The reaction of an amide group with ninhydrin is less common compared to its reaction with free amino groups. However, under specific conditions, ninhydrin can interact with amides, especially when catalysed by agents like ammonium molybdate. Here's a general outline:

1. **Hydrolysis of Amide:** The amide group may undergo hydrolysis to release ammonia or an amine, which can then react with ninhydrin.
2. **Formation of Chromogen:** The released ammonia or amine reacts with ninhydrin to form a colored complex, such as Ruhemann's Purple or other chromogens, depending on the reaction conditions.



3. **Catalysis:** Catalysts like ammonium molybdate can enhance the reaction, leading to the formation of a distinct chromogen with a bathochromic shift, as observed with Nicotinamide.



Scheme: Reaction between Nicotinamide with Ninhydrin catalyzed by ammonium molybdate.

2.9 Procedure

In a 25 ml beaker, add 10 ml standard stock solution B, warm it in a boiling water bath at $97 \pm 1^{\circ}\text{C}$ for 5 minutes, to this solution add 5 ml of 0.2M Ammonium Molybdate and 5 ml of 0.2M Ninhydrin, and cool at room temperature. Use this solution for further dilutions as Stock solution C, which contains $1000 \mu\text{g/ml}$ Nicotinamide. Scan the spectrum, and absorbance was measured at λ_{max} at 761 nm versus the reagent blank (Figure 2).

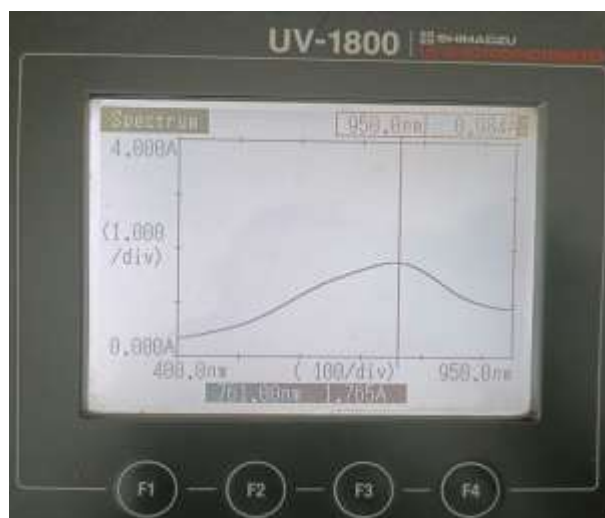


Figure 2: UV-Visible Spectrum of Nicotinamide.

3. METHOD VALIDATION

3.1 Effect of temperature on absorbance

The effect of temperature on the complexation reaction at 80° , 90° $97 \pm 1^{\circ}\text{C}$ was examined (Table 2). It was observed that the ninhydrin–NIC complex in saturated ammonium Molybdate required $97 \pm 1^{\circ}\text{C}$ for obtaining maximum and stable absorbance and remained constant for about 8 further hours.

Table 2: Effect of temperature on absorbance

Temperature ($^{\circ}\text{C}$)	Absorbance
80 ± 1	0.049
90 ± 1	0.804
97 ± 1	1.765

3.2. Linearity

The method's linearity was examined²² From the Standard Stock solution B ($100\mu\text{g/ml}$), different aliquots (2.5, 5.0, 7.5, 10.0 and 2.5) were taken in a series of 10 ml volumetric flasks and 1.0 ml ninhydrin was added to it followed by 1.0 ml saturated ammonium molybdate solution and volume made up with 0.1M HCl to get

concentration $25\text{--}125\mu\text{g/ml}$. All flasks were heated for 5 min in a boiling water bath and cooled to room temperature, and the absorbance was measured at 761 nm. Five replicates of analytes were measured, and the absorbance *versus* concentration is shown in Table 2. Plot a graph of concentration *versus* absorbance, a linear correlation was found, which obeys Beer-Lambert's Law in the concentration range of $25\text{--}125\mu\text{g/ml}$ (Figure 3). Regression analysis of Beer's law data using the method of least squares was made to evaluate the slope (b), intercept (a), and the correlation coefficient (r^2). So far, from the below graph, the correlation coefficient, v-intercept, and slope of the regression line are given in Table 3²³.

Table 2: Observation data for the calibration Curve of Nicotinamide

Concentration ($\mu\text{g/ml}$)	Absorbance
25	0.115
50	0.236
75	0.355
100	0.468
125	0.577

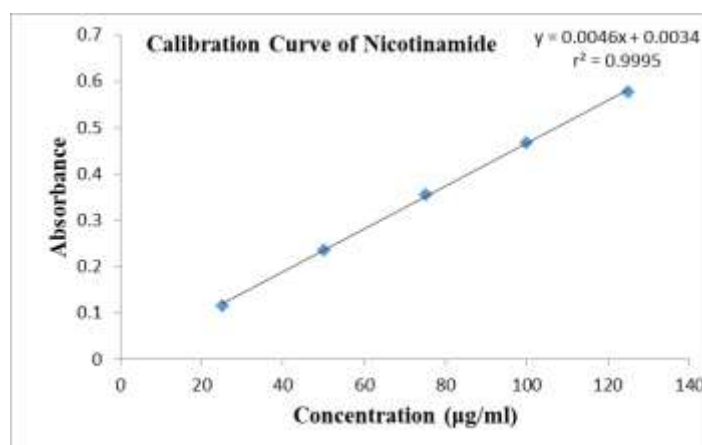


Figure 3: Calibration curve of Nicotinamide.

3.3. Limit of Detection (LOD) and Limit of Quantification (LOQ)

Two important parameters, that is, Limit of Detection (LOD) and Limit of Quantification

(LOQ), were determined to test the performance limits²⁴. The detection limit of a singular analytical method is defined as the minimum concentration of an analyte present in a sample that can be recognized, albeit not necessarily quantified as an

exact measurement. The quantitation limit of a singular analytical method refers to the minimum concentration of an analyte in a sample that can be determined quantitatively with appropriate precision and accuracy. The quantitation limit serves as a critical parameter in quantitative assays aimed at identifying trace levels of compounds within sample matrices and is particularly employed in the assessment of impurities and/or degradation products. The values of the limit of detection (LOD) and limit of quantitation (LOQ) are established utilizing the standard deviation of the response in conjunction with the slope method, as delineated in the guidelines provided by the International Conference on Harmonization (ICH) and presented in Table 3.

Table 3: Optical Characteristics data and validation parameters of Nicotinamide

Parameter	Analytical data
Range ($\mu\text{g/ml}$)	25-125
λ max (Linearity nm)	761
Molar extinction coefficient, L/mole cm^{-1}	2.3657×10^2
Sandell's sensitivity, $\mu\text{g/cm}^2$	0.5199
Slope	0.0046
Standard deviation about regression	± 0.0082
Standard deviation of Slope (Sb)	± 0.0063
Standard deviation of intercept (Sa)	± 0.0051
Correlation of coefficient (r^2)	0.9996
Limit of detection (LOD, $\mu\text{g/ml}$)	3.3444
Limit of quantification (LOQ, $\mu\text{g/ml}$)	10.1346

3.4. Precision

3.4.1. Intraday Precision

Precision quantifies the veracity of a model's positive predictions. The ratio of correct guesses to the total number of correct and incorrect predictions is used to determine it. Particularly

useful in scenarios where minimising false positives is critical, precision emphasises the accuracy of positive predictions for every occurrence predicted as positive by the model²⁵. A common approach to demonstrate intraday precision, where measurements are taken from the same batch at consistent intervals-initially after 24 hours and after 48 hours (Table 4). This method ensures that variability within the same day and across the day is carefully observed.

3.4.2. Interday Precision

Interday precision of the test method is demonstrated by three samples of the same batch (same concentration) on three successive days (Table 4).

Once the data is collected, the precision can be assessed by calculating the **Relative Standard Deviation (RSD%)** for the replicate measurements over the three days. A low RSD% value would confirm that the method is reliable and reproducible over multiple days.

A low RSD% value in UV spectroscopy is indicative of high precision and reliability in the method. While the acceptable threshold can vary depending on regulatory guidelines and the specific application, in most cases:

- **RSD% \leq 2%** is generally considered excellent precision for UV spectroscopy.
- **RSD% between 2% and 5%** may still be acceptable, depending on the context and requirements of the analysis.

Table 4: Evaluation of intra-day and inter-day accuracy and precision

NIC taken ($\mu\text{g/ml}$)	Intraday Accuracy and Precision			Interday Accuracy and precision		
	NIC found	RE %	RSD %	NIC found	RE %	RSD %
	($\mu\text{g/ml}$)			($\mu\text{g/ml}$)		
100	91.29	89.19	1.59	86.74	83.74	1.45
150	141.82	94.83	0.97	138.62	92.38	1.02



200	192.34	96.16	0.76	187.54	94.57	0.69
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3.5. Accuracy

The recovery study method represents²⁶. By adding different percentages (80%, 100%, and 120%) of Nicotinamide bulk sample within the linearity range, we ensure a robust evaluation of how well the method measures the actual amount of analyte present. The results compiled in Table 5 likely show recovery percentages close to 100%, indicating the accuracy of the method.

Recovery within 98-102%: This range is generally considered excellent accuracy in analytical chemistry.

Recovery within 95-105%: May still be acceptable depending on regulatory guidelines and the specific application.

Table 5: Recovery Data

Level	Amount of NIC added (µg)	Amount of NIC found (µg)	% Recovery	% RSD
80%	80	823.1	103.87	0.71
100%	100	102.75	102.75	0.83
120%	120	115.56	96.3	0.92

*An average value \pm relative standard deviation of 5 observations

4. RESULT AND DISCUSSION

Amides are more stable and do not spontaneously release ammonia. Under catalyzed conditions, the amide group might hydrolyze, releasing ammonia or an amine, which then reacts with ninhydrin to form a different chromogen, such as the blue product observed for Nicotinamide.

The chromogen produced can exhibit a bathochromic shift (e.g., to 761 nm for Nicotinamide), resulting in a color shift toward

longer wavelengths, influenced by the amide group's electronic properties. The reaction with amides is more selective, as it requires specific conditions and catalysts, making it less common but useful in targeted analytical applications.

The research method is validated, viz. stability, linearity, LOD, LOQ, Precision, and Recovery data for Nicotinamide in bulk drugs, and found within acceptable limits as per ICH Guidelines²¹

CONCLUSION

As a versatile analytical tool, based on the results and discussion, it can be concluded that the reaction between ninhydrin and the amide group in Nicotinamide is indeed a striking example of chemical ingenuity. The catalytic role of ammonium molybdate highlights how carefully selected reagents can enhance sensitivity and reactivity. The newly developed spectrophotometric method for determining Nicotinamide is precise, economical, and environmentally friendly. By highlighting its eco-friendliness, cost effectiveness, and the avoidance of organic solvents, we have shown how this method aligns with sustainable and economical laboratory practices. Overall, this innovative method suggests a practical solution for quality control of Nicotinamide in bulk quantities for research and academic laboratory settings with budget constraints. Its accuracy and precision ensure reliable results, which are crucial in pharmaceutical analysis.

ACKNOWLEDGEMENT

The authors gratefully acknowledge the receipt of pure Nicotinamide as a Gift sample from Wockhardt Pharmaceuticals Ltd.(Chhatrapati Sambhajanagar). We extend our heartfelt gratitude



to Dr. S. S. Angadi, the esteemed principal of Yash Institute of Pharmacy, Chhatrapati Sambhajnagar, for his unwavering support and encouragement throughout this project.

AUTHORSHIP CONTRIBUTION STATEMENT

Vandana Patil: Designed the overall study concept and methodology

Krishna Chopde: Conducted experimental trials and quality control assessments

Amruta Jadhav: Contributed to the literature review and Data Synchronization

Suwarna Kale: Assisted in manuscript revision and provided the critical ideology

Sachidanand Angadi: Funding and Data Curation

REFERENCES

1. Sarkis N, Sawan A. Development and validation of derivative UV spectroscopic method for simultaneous estimation of nicotinamide and tretinoin in their binary mixtures and pharmaceutical preparations. *BMC Chemistry* [Internet]. 2022 Dec [cited 2025 May 11];16(1):15. Available from: <https://bmcchem.biomedcentral.com/articles/10.1186/s13065-022-00809-x>
2. Wohlrab J, Kreft D. Niacinamide - Mechanisms of Action and Its Topical Use in Dermatology. *Skin Pharmacol Physiol*. 2014;27(6):311–5.
3. Mohamed F, Sidhom M, Hussein S. TITRIMETRIC AND SPECTROPHOTOMETRIC DETERMINATION OF MEBENDAZOLE AND FLUBENDAZOLE USING N-BROMOSUCCINIMIDE. *Bulletin of Pharmaceutical Sciences Assiut* [Internet]. 1986 Dec 31 [cited 2025 May 11];9(2):92–104. Available from: http://bpsa.journals.ekb.eg/article_74580.htm
4. Nwanisobi G, Egbuna SO. Colorimetric determination of nicotinamide with dichlorodicyano benzoquinone. *International Journal of ChemTech Research*. 2015 Jan;8(3):1139–41.
5. Gloria N, Nwaiwu J. A COST EFFECTIVE AND SIMPLE METHOD FOR THE DETERMINATION OF NICOTINAMIDE USING PICRIC ACID.
6. Kumar V SV, J K, Ks L. UV Spectrophotometric Quantification of Niacinamide in Pharmaceutical Dosage Form by Multivariate Calibration Technique. *RJPT* [Internet]. 2021 Apr 29 [cited 2025 May 11];2013–20. Available from: <https://rjptonline.org/AbstractView.aspx?PID=2021-14-4-33>
7. Chanda I, Bordoloi R, Chakraborty DD, Chakraborty P, Chanda Das SR. Development and Validation of UV-Spectroscopic Method for Estimation of Niacin in Bulk and Pharmaceutical Dosage Form. *J App Pharm Sci* [Internet]. 2017 [cited 2025 May 11];7(09):081–4. Available from: http://www.japsonline.com/abstract.php?article_id=2409
8. Dr. K. B, Anila B, Saileela S, MogiliSumakanth Dr. METHOD DEVELOPMENT OF NIACIN IN BULK AND DOSAGE FORM BY UVVISIBLE SPECTROPHOTOMETER. *IJMPR*. 2022;6(9):41–3.
9. Trivedi MK, Branton A, Trivedi D, Nayak G, Bairwa K, Snehasis J. Spectroscopic Characterization of Disulfiram and Nicotinic Acid after Biofield Treatment. *J Anal Bioanal Tech* [Internet]. 2015 [cited 2025 May 11];6(5):6:5. Available from:



- https://www.omicsonline.org/open-access/spectroscopic-characterization-of-disulfiram-and-nicotinic-acid-after-biofieldtreatment-2155-9872-1000265.php?aid=60834
10. Alahmad S, Elfatratry HM, Mabrouk MM, Hammad SF, Mansour FR. Development and Validation of Chemometric Spectrophotometric Methods for Simultaneous Determination of Simvastatin and Nicotinic Acid in Binary Combinations. CDDT [Internet]. 2018 Apr 26 [cited 2025 May 11];15(2):149–55. Available from: <http://www.eurekaselect.com/154829/article>
11. Tokunaga H, Okada S, Kimura T. [Determination of nicotinic acid in injections by high-performance liquid chromatography]. Eisei Shikenjo Hokoku. 1989;(107):108–12.
12. Sangshetti JN, Aqeel M, Zaheer Z, Ahmed RZ, Dehghan MHG, Gonjari I. Development and validation of RP-HPLC method for determination of Atorvastatin calcium and Nicotinic acid in combined tablet dosage form. Journal of Saudi Chemical Society [Internet]. 2016 Sep [cited 2025 May 11];20:S328–33. Available from: <https://linkinghub.elsevier.com/retrieve/pii/S1319610312001949>
13. Tsang PK, Zeng L. Quantitative analysis of nicotinic acid, nicotinamide and 3-cyanopyridine in industrial effluent by high performance liquid chromatography. Front Chem China [Internet]. 2007 Jul [cited 2025 May 11];2(3):270–3. Available from: <http://link.springer.com/10.1007/s11458-007-0048-y>
14. Pescagliani M, Micheli V, Simmonds HA, Rocchigiani M, Pompucci G. Nicotinic acid phosphoribosyltransferase activity in human erythrocytes: Studies using a new HPLC method. Clinica Chimica Acta [Internet]. 1994 Sep [cited 2025 May 11];229(1–2):15–25. Available from: <https://linkinghub.elsevier.com/retrieve/pii/0009898194902259>
15. Iammarino M. Validation of an Analytical Method for the Determination of Ascorbic Acid and Nicotinic Acid in Fresh Meat Preparations by HPLC-UV-DAD. JFNS [Internet]. 2015 [cited 2025 May 11];3(1):7. Available from: <http://www.sciencepublishinggroup.com/journal/paperinfo?journalid=154&doi=10.11648/j.jfns.s.2015030101.12>
16. Dhadve AZ, Dashetwar AR, Kardile DP, Shete RV. Stability Indicating RP-HPLC Method for Simultaneous Determination of Atorvastatin and Nicotinic acid from Tablet Dosage Form. Rese Jour of Pharm and Technol [Internet]. 2020 [cited 2025 May 11];13(2):604. Available from: <http://www.indianjournals.com/ijor.aspx?target=ijor:rjpt&volume=13&issue=2&article=018>
17. Tiwari PK, Sathe P. Development and validation of HPTLC method for niacin and simvastatin in binary combination. ABB [Internet]. 2010 [cited 2025 May 11];01(02):131–5. Available from: <http://www.scirp.org/journal/doi.aspx?DOI=10.4236/abb.2010.12018>
18. Dołowy M, Pyka A. Validation of an RPHPTLC-Densitometric Method Using Silica Gel 60 RP18WF254 for Simultaneous Determination of Nicotinamide in Selected Pharmaceutical Formulations. Journal of Analytical Methods in Chemistry [Internet]. 2015 [cited 2025 May 11];2015:1–9. Available from: <http://www.hindawi.com/journals/jamc/2015/631025/>
19. Banik K, Rath S, Gupta BK. Chromatographic Estimation of Niacinamide in Anti-aging Dermatological product. Asian Jour Pharmac

- Anal [Internet]. 2019 [cited 2025 May 11];9(2):62. Available from: <http://www.indianjournals.com/ijor.aspx?target=ijor:ajpa&volume=9&issue=2&article=004>
20. Iwaki M, Murakami E, Kakehi K. Chromatographic and capillary electrophoretic methods for the analysis of nicotinic acid and its metabolites. *Journal of Chromatography B: Biomedical Sciences and Applications* [Internet]. 2000 Sep [cited 2025 May 11];747(1–2):229–40. Available from: <https://linkinghub.elsevier.com/retrieve/pii/S0378434799004867>
 21. Abraham J. International Conference on Harmonisation of Technical Requirements For Registration Of Pharmaceuticals For Human Use. In: Tietje C, Brouder A, editors. *Handbook of Transnational Economic Governance Regimes* [Internet]. Brill | Nijhoff; 2010 [cited 2025 May 11]. p. 1041–53. Available from: https://brill.com/view/book/edcoll/9789004181564/Bej.9789004163300.i-1081_085.xml
 22. Sakhare RS, Marashivane RN, Wattamwar PB. 10.13040/IJPSR.0975-8232.16(2).457-65. *IJPSR*. 2025;16(2):457–65.
 23. Srikanth G, Phanindra VS, Sultana S, Vadayar KP, Akshitha GL, Kumar KN. METHOD DEVELOPMENT AND MODEST VALIDATION OF SULFADIAZINE BY REVERSE PHASE HPLC. *IJPSR*. 2025;16(2):475–80.
 24. Mastanamma SK, Chitturi S, Naseemunnisa MD, Anitha Kumari V. VALIDATED ANALYTICAL METHOD DEVELOPMENT AND CHARACTERIZATION OF FORCED DEGRADATIVE PRODUCTS OF MITAPIVAT AND THEIR TOXICITY PREDICTION BY IN-SILICO STUDIES. *IJPSR*. 2025;16(1):184–200.
 25. Khokrale S, Lulla J, Borse L. A Review Of The UV-Visible Spectroscopy's Method Development And Validation. *Int J of Pharm Sci*. 2024;2(6):527–38.
 26. Majumder S, Saha P. QUANTITATIVE ESTIMATION OF QUERCETIN FROM PHOENIX SYLVESTRIS USING A VALIDATED HPTLC METHOD. *IJPSR*. 16(1):162–6.

HOW TO CITE: Dr. Vandana Patil, Krishna Chopde, Amruta Jadhav, Nilay Shinde, Spectral Insights: Bathochromic Shifts in Visible Spectra of Nicotinamide with Ninhydrin, *Int. J. of Pharm. Sci.*, 2025, Vol 3, Issue 8, 883-892. <https://doi.org/10.5281/zenodo.16789120>

