Development of a Spectral Library for Nitrosamine ID and Quantification in Pharmaceuticals and Consumer Products

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Introduction

Nitrosamine impurities present ongoing analytical challenges in pharmaceutical development and quality control, particularly when confident structural discrimination of closely related species is required.

This work demonstrates that Molecular Rotational Resonance (MRR) spectroscopy enables structure-specific identification and targeted quantitation of nitrosamines within a simplified, orthogonal workflow.

Methods

ASA Workflow:

1. **Define candidate structures:** Theoretical rotational spectra were generated for target nitrosamines to create a predicted reference library.
2. **Acquire experimental spectra:** Samples were measured by automated MRR to obtain high-quality rotational spectra.
3. **Match theory to experiment:** Experimental spectra were compared with predicted spectra to confirm molecular identity.
4. **Build validated library:** Confirmed spectra were incorporated into an experimental nitrosamine reference library for future identification.

Quantitation Workflow:

Sample: Nitrosamine solutions prepared in deionized water.

Source Parameters: PTV-based sampling with a variable-temperature program to enable solvent purge, followed by analyte elution.

Acquisition Parameters: Targeted single-frequency MRR detection; average runtime ~19 minutes.

Results

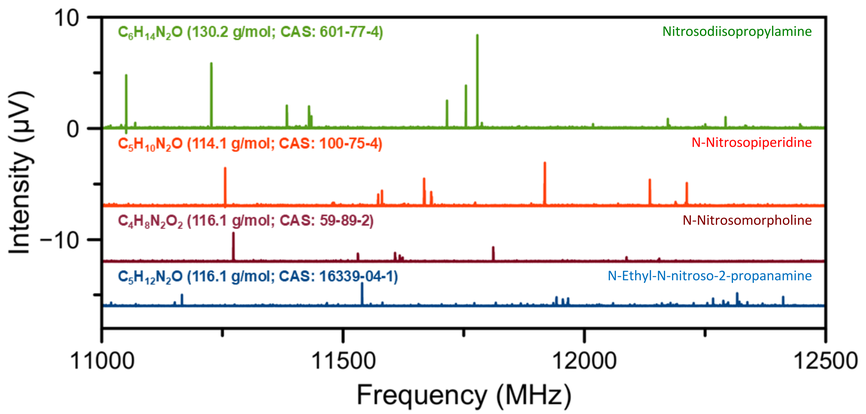
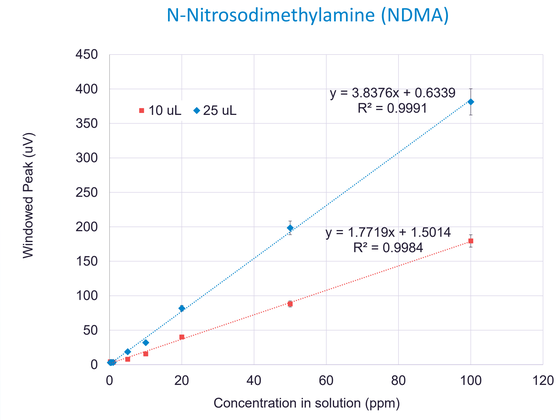


Figure 1. MRR spectra of Nitrosamines. Measured Molecular Rotational Resonance (MRR) spectra for representative nitrosamines generated using the spectraMRR: N-nitrosopiperidine, N-ethyl-N-nitroso-2-propanamine, N-nitrosomorpholine, and nitrosodiisopropylamine. The simulated rotational spectra illustrate the distinct spectral fingerprints arising from differences in molecular geometry and mass distribution, supporting structure-specific discrimination within closely related nitrosamine classes.



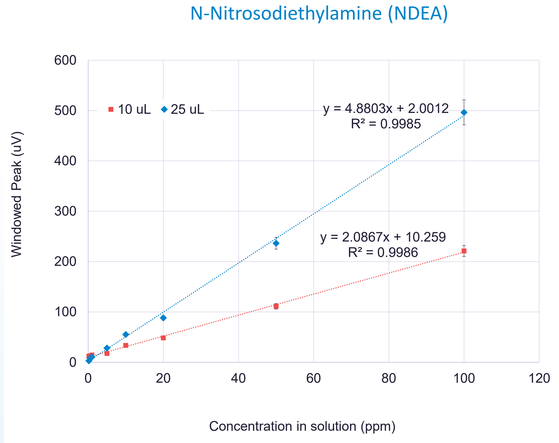


Figure 2. NDMA and NDEA Feasibility (1 – 100 ppm). Quantitative feasibility for representative nitrosamines using targeted MRR measurements. **(A)** Calibration curve for N-nitrosodimethylamine (NDMA). **(B)** Calibration curve for N-nitrosodiethylamine (NDEA). Single-frequency targeted acquisition demonstrates a linear response across the evaluated concentration range (1 - 100 ppm), supporting selective quantitation following structural confirmation. The response curves to two different injection volumes are given, demonstrating linearity of the response to the analyte mass.

Discussion

MRR enables structure-specific identification of nitrosamine impurities through direct measurement of gas-phase rotational transitions. Because rotational spectra are determined by molecular geometry and mass distribution, closely related nitrosamines can be differentiated without reliance on chromatographic retention behavior or fragmentation patterns.

Predicted spectra generated with BrightHub provide a framework for candidate screening and library development. Experimental measurements confirm structural identity through direct spectral matching. Following identification, targeted single-frequency measurements support selective and quantitative analysis across relevant concentration ranges.

These results demonstrate that MRR provides an orthogonal approach for nitrosamine analysis, combining structural confirmation with targeted quantitation in a simplified workflow.

Conclusion

- A validated MRR spectral library was developed for representative nitrosamines.
- Experimental spectra matched predicted rotational signatures, confirming structure-specific identification.
- Isomeric and structurally related nitrosamines were distinguishable without chromatographic separation.
- Targeted measurements demonstrated quantitative feasibility from 1 to 100 ppm.

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