White Paper 10476

Determining Nitrosamines Using GC-MS/MS with Electron Ionization

David Steiniger and Alexander Semyonov
Thermo Fisher Scientific, Austin, TX

Key Words

AutoSRM, Electron Energy, GC-MS/MS, Nitrosamines, Triple Quadrupole

Introduction

Traditionally, nitrosamines have been analyzed by an ion trap in the MS/MS mode, using chemical ionization (CI) to provide a "soft" ionization of the precursor ion (e.g., U.S. Environmental Protection Agency (EPA) Method 521). Most of these methods require a large volume, 10–20 µL, injection. With the highly sensitive Thermo Scientific™ TSQ™ Duo triple quadrupole mass spectrometer this may no longer be necessary. Using automatic selected reaction monitoring (AutoSRM) software to fully optimize the choice of transitions and collision energies, and decreasing the electron energy to 40 eV to soften the ionization, equivalent detection limits can be achieved.

Using AutoSRM Software to Optimize Transitional Parameters

Compounds with labile chemical bonds need extra consideration when being analyzed by GC-MS/MS. Because of their nature, their standard 70 eV EI mass spectrum exhibits a lot of low abundance and low mass ion fragments with few good choices for precursor and product ions that would provide the sensitivity for low-level analysis.

AutoSRM for the TSQ Duo GC-MS/MS system provides two modes of collision energy (CE) optimization: full range and targeted. Full range CE optimization starts at 5 V and increases by 5-V steps during the experimental portion of the process. Targeted optimization uses smaller CE increments of 2 V, providing better optimization for those compounds that exhibit sharper dependencies on collision energy.

Seven nitrosamines targeted (by U.S. EPA Method 521) and two deuterated nitrosamines were run using AutoSRM both in full range and targeted optimization modes. The results are presented in Figures 1 and 2. Running these compounds in full range mode does not provide an apex, which would allow the user to know that the optimum collision energy has been reached. Targeted mode did produce an apex, assuring the best setting of the collision voltage for that transition. A total of seven different transitions were discovered to react in this manner.

Thermo SCIENTIFIC

Figure 1. Full range CE optimization mode.

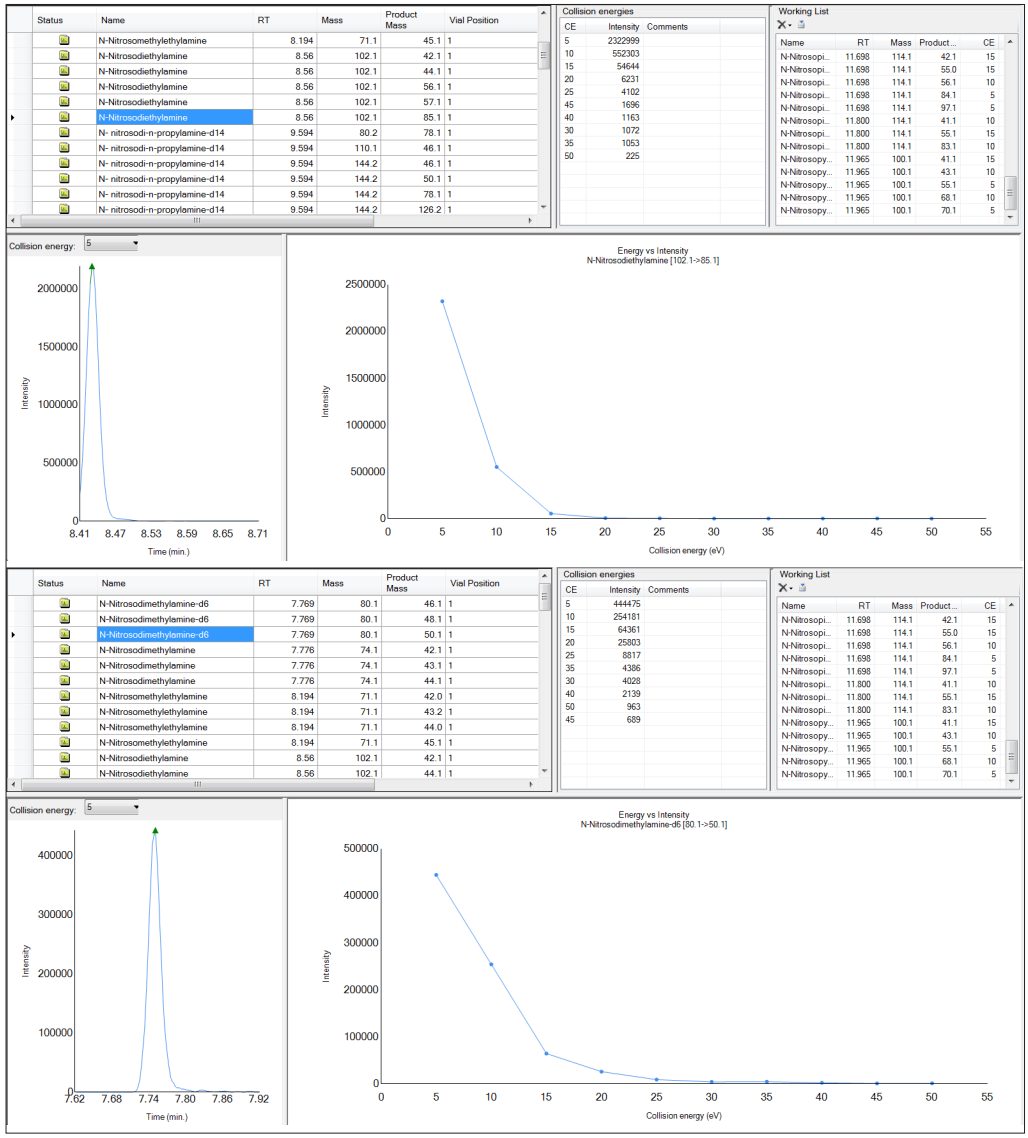
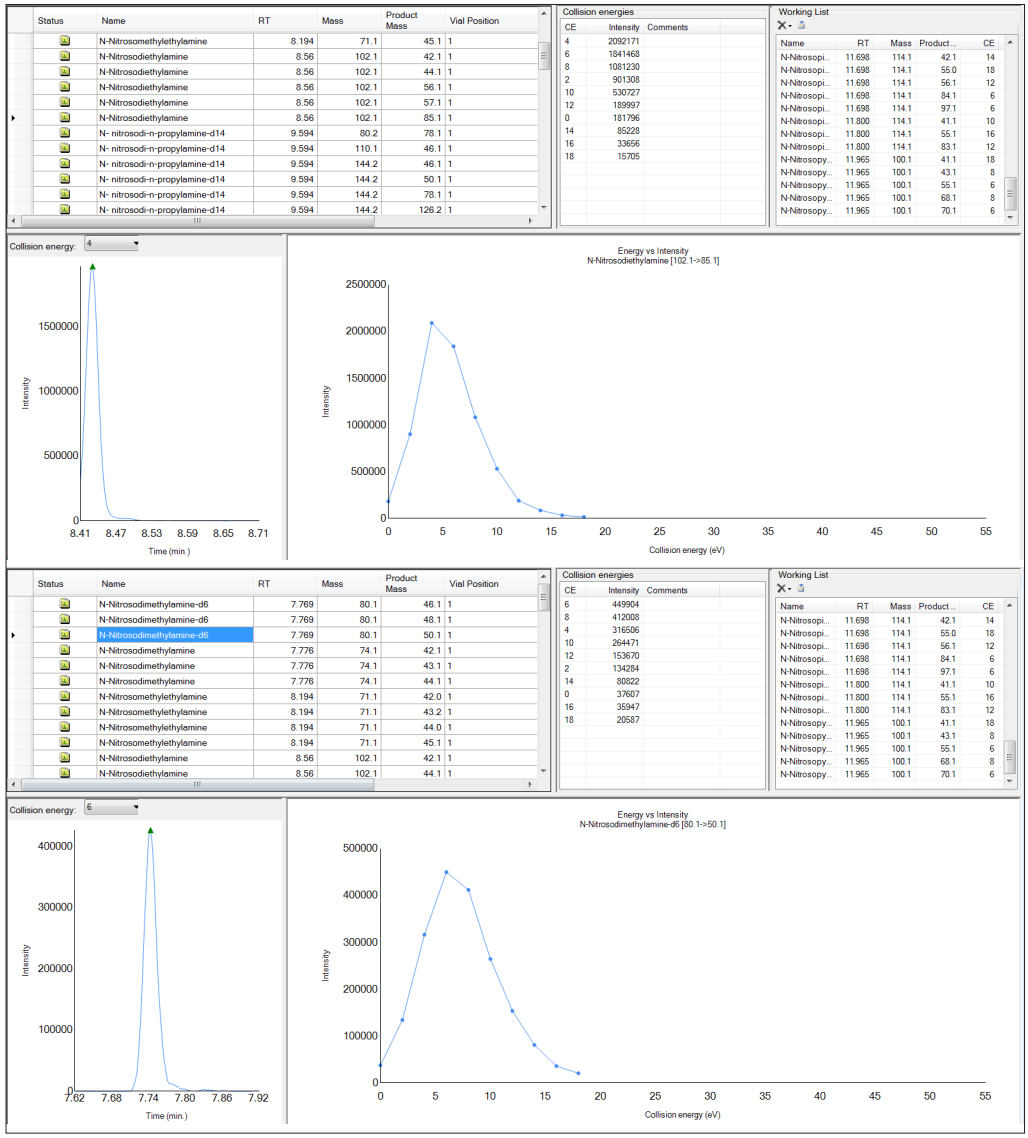


Figure 2. Targeted CE optimization mode.



Operating the TSQ Duo System at 40 eV to Soften Ionization

Analyzing nitrosamines on the TSQ Duo system at an electron energy of 40 eV has two advantages over the standard value of 70 eV. First, a higher abundance of higher-mass ions enables the selection of higher intensity precursor ions. Second, fragmentation of higher-mass ions usually provides higher-mass product ions, leading to improved selectivity and detectability. The use of softer ionization also increases selectivity through higher-mass precursor selection, as has been shown in analysis of dioxins.[1]

A comparative study was performed in AutoSRM software at the standard 70 eV and again at 40 eV electron energies. The results obtained using 70 eV and 40 eV are compared in Figure 3. In some cases different transitions and collision energies were used in the method developed at 40 eV.

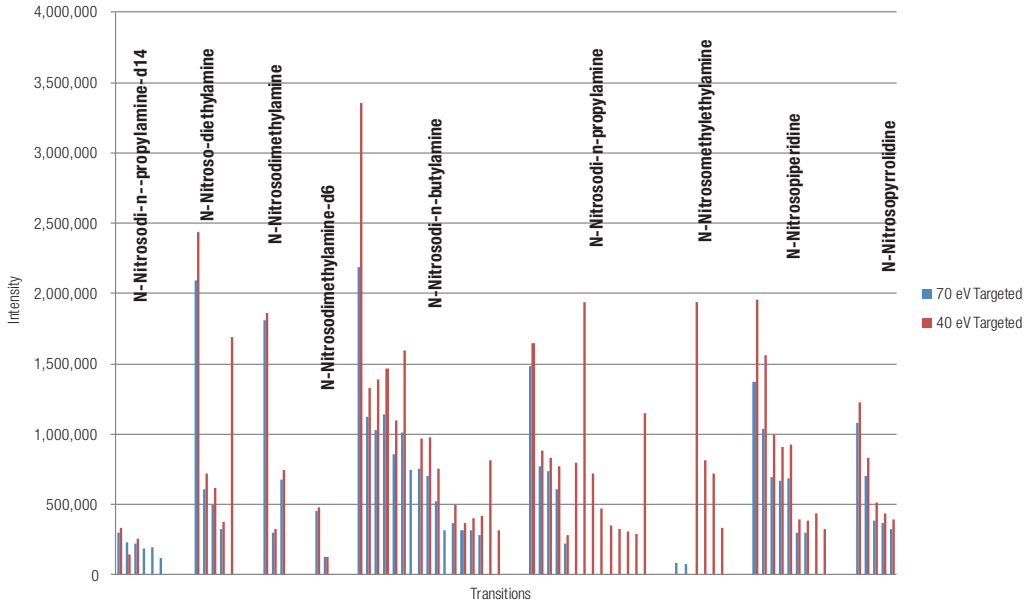


Figure 3. Comparison of AutoSRM results at 70 eV and 40 eV electron energy.

Results

The best results were achieved using targeted CE optimization and a soft electron energy of 40 eV. Calibration linear curves were generated for the concentration range from 1 to 50 pg injected on-column, with six calibration levels in methylene chloride. Of these standards, 1 µL of the the 1 ppb level was then injected in 10 replicates to calculate the IDL. The r^2 values for the seven analytes were greater than or equal to 0.999 (Table 1). The replicate injection results were used to calculate instrument detection limits (IDLs) for the seven targeted nitrosamines (Table 1). Table 2 provides the SRM transitions and the corresponding optimized collision energies used in this evaluation.

Table 1. Linearity and IDLs for the targeted nitrosamine compounds.

Compound	r^2	TSQ Duo System IDL	EPA Method 521
N-Nitrosodimethylamine	0.9997	0.33	0.28
N-Nitrosodimethylethylamine	0.9997	0.19	0.28
N-Nitrosodiethylamine	0.9999	0.13	0.26
N-Nitrosodi-n-propylamine	0.9996	0.26	0.32
N-Nitrosodi-n-butylamine	0.9998	0.32	0.36
N-Nitrosopiperidine	0.9998	0.28	0.66
N-Nitrosopyrrolidine	0.9990	0.44	0.35

Table 2. Transitions and collision energies for targeted and deuterated nitrosamines. Blue rows with bold indicate quantitation ions.

Compound	RT	Precursor Mass	Product Mass	Collision Energy
N-Nitrosodimethylamine	**7.766**	**74.1**	**44.1**	**6**
N-Nitrosodimethylamine	7.766	74.1	42.1	14
N-Nitrosodimethylamine	7.766	74.1	43.1	12
N-Nitrosodimethylamine-d6	**7.779**	**80.1**	**50.1**	**6**
N-Nitrosodimethylamine-d6	7.779	80.1	46.1	14
N-Nitrosomethylethylamine	**8.194**	**88.1**	**71.1**	**4**
N-Nitrosomethylethylamine	8.194	88.1	42.1	16
N-Nitrosomethylethylamine	8.194	88.1	43	8
N-Nitrosodiethylamine	**8.46**	**102.1**	**85.1**	**4**
N-Nitrosodiethylamine	8.46	102.1	44.1	12
N-Nitrosodiethylamine	8.46	57.1	42.1	6
N-Nitrosodi-n-propylamine-d14	**9.594**	**78.1**	**50.1**	**6**
N-Nitrosodi-n-propylamine-d14	9.594	78.1	46	10
N-Nitrosodi-n-propylamine-d14	9.594	144.2	50	10
N-Nitrosodi-n-propylamine	**9.665**	**130.1**	**113.1**	**4**
N-Nitrosodi-n-propylamine	9.665	101.1	70.1	6
N-Nitrosodi-n-propylamine	9.665	70.1	43.1	6
N-Nitrosodi-n-buytlamine	**11.27**	**116.1**	**99.1**	**6**
N-Nitrosodi-n-buytlamine	11.27	115.1	84.1	6
N-Nitrosodi-n-buytlamine	11.27	158.2	99.1	8
N-Nitrosopiperidine	**11.7**	**114.1**	**84**	**8**
N-Nitrosopiperidine	11.7	114.1	41.1	12
N-Nitrosopiperidine	11.7	114.1	97.1	6
Nitrosopyrrolidine	**11.995**	**100.1**	**55.1**	**6**
Nitrosopyrrolidine	11.995	100.1	43.1	10
Nitrosopyrrolidine	11.995	100.1	70.1	6

Conclusions

The sensitivity of the TSQ Duo GC-MS/MS system makes nitrosamines analysis easier due to:

- No requirement for large-volume injections
- The complexity of CI is unnecessary
- No need for methane or ammonia gas
- Simple, straightforward EI analysis
- AutoSRM software for optimization of compound transitions and collision energies

The TSQ Duo triple quadrupole GC-MS/MS system can analyze nitrosamines in EI mode like most other GC-amenable compounds as an alternative to using more complex, difficult methods.

References

1. Thermo Scientific Application Note 10406: Validation of GC-MS/MS for Detection and Confirmation of Low-Level Dioxins. Runcorn, UK. http://www.thermoscientific.com/content/dam/tfs/ATG/CMD/cmd-documents/sci-res/app/ms/GC-MS/AN-10406-GC-MS-Low-Level-Dioxins-AN10406-EN.pdf (accessed August 26, 2015).

www.thermoscientific.com/TSQDuo

Thermo Fisher Scientific, Austin, TX USA is ISO 9001:2008 Certified.

Africa +43 1 333 50 34 0
Australia +61 3 9757 4300
Austria +43 810 282 206
Belgium +32 53 73 42 41
Canada +1 800 530 8447
China 800 810 5118 (free call domestic) 400 650 5118
Denmark +45 70 23 62 60
Europe-Other +43 1 333 50 34 0
Finland +358 10 3292 200
France +33 1 60 92 48 00
Germany +49 6103 408 1014
India +91 22 6742 9494
Italy +39 02 950 591
Japan +81 45 453 9100
Korea +82 2 3420 8600
Latin America +1 561 688 8700
Middle East +43 1 333 50 34 0
Netherlands +31 76 579 55 55
New Zealand +64 9 980 6700
Norway +46 8 556 468 00
Russia/CIS +43 1 333 50 34 0
Singapore +65 6289 1190
Spain +34 914 845 965
Sweden +46 8 556 468 00
Switzerland +41 61 716 77 00
UK +44 1442 233555
USA +1 800 532 4752

Thermo SCIENTIFIC
A Thermo Fisher Scientific Brand

WP10476-EN 0915M