Application Note

Materials

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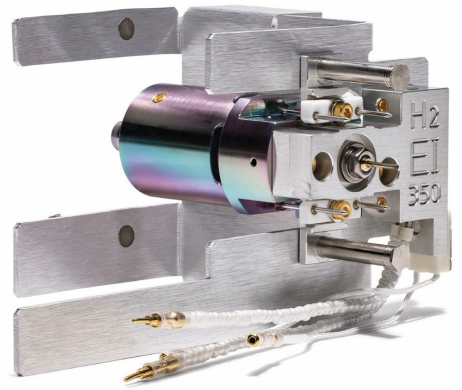
Analysis of Aromatic Amines in Textile Samples

Using hydrogen carrier gas and an Agilent 8890 gas chromatography system coupled with an Agilent 5977C GC/MSD with an Agilent HydroInert source



Authors

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Abstract

This application note demonstrates the use of an Agilent 8890 gas chromatography system coupled with an Agilent 5977C GC/MSD with the Agilent HydroInert source to detect and quantify residues of aromatic amines in textile samples using hydrogen as the carrier gas. The sample was extracted using organic solvents through ultrasonication, followed by a GC/MS analysis. A limit of quantification (LOQ) of 1 µg/g was achieved in textile samples for 24 amine compounds using internal standard (IS) calibration. Aqueous calibration range was established from 0.25 to 20 µg/mL.

Introduction

Azo dyes, characterized by the azo group (–N=N–), represent the largest class of synthetic organic colorants, accounting for approximately 70% of the global dye market with over 3,000 variants in use.[1] Their widespread application in textiles, leather, plastics, cosmetics, food, and pharmaceuticals is attributed to their low cost, colorfastness, and structural versatility. Global textile dye production exceeded 1 million tons in 2020.[1] Despite their industrial significance, azo dyes pose serious environmental and health concerns.[1,2] Up to 15% of these dyes are discharged into wastewater or solid waste streams, and as much as 40% may be lost during textile processing. Resistant to degradation, they persist in ecosystems and can release harmful aromatic amines under certain conditions.[1] Numerous GC/MS-based analytical methods are available for the estimation of amine compounds. These methods use helium as the carrier gas. For a sustainable analytical solution, there is a need for an alternative carrier gas to overcome the challenges occurring due to helium scarcity.

The method adopted for this application note demonstrates the use of hydrogen as a carrier gas in the Agilent 8890 gas chromatography system coupled with the Agilent 5977C GC/MSD using a HydroInert source for the quantification of aromatic amines residues in textile samples.

Experimental

Standard preparation

A 40 µg/mL reference mix solution (Mix 1) of 24 amines procured from Dr. Ehrenstorfer was prepared by mixing and diluting the stock solution of individual amine compound in acetonitrile. Mix 1 solution was diluted with acetonitrile to achieve 4.0 µg/mL concentration (Mix 2). A 10 µg/mL solution of anthracene-d10 (IS) was prepared by diluting the stock solution in acetonitrile.

Seven calibration levels (0.25, 0.5, 1.0, 2.5, 5.0, 10.0, and 20.0 µg/mL) were prepared as per the details mentioned in Table 3.

Instrument parameters

Table 1. GC/MS parameters.

Parameters	Value
Agilent 8890 Gas Chromatography System	
Inlet Temperature	290 °C
Analytical Column	Agilent J&W DB-35ms column (20 m × 0.18 mm, 0.18 µm) (p/n 121-3822)
Injection Volume	2 µL
Column Flow	Hydrogen; 1.0 mL/min, constant flow
Injection Mode	Split (10:1)
Oven Program	Start at 100 °C (hold for 2.0 min), ramp at 15 °C/min to 135 °C (hold for 1 min), then at 45 °C/min to 240 °C (hold for 1 min), and then at 15 °C/min to 290 °C (hold for 3 min)
MS Transferline Temperature	300 °C
Agilent 5977C GC/MSD	
Ion Source	Agilent HydroInert source (G7078-67930)
Ion Source Temperature	250 °C
Quadrupole Temperature (Q1 and Q2)	150 °C

Sample preparation

Approximately, 0.5 ±0.01 g of sample was taken in stoppered glass tubes. A citrate buffer of 30 mL was added to the sample, which was then placed in a water bath at 70 °C. Next, 15 mL of sodium dithionite was added, and the sample was again placed in the water bath for 30 minutes. Then, extraction was performed using methyl tert-butyl ether (MTBE), followed by concentrating the extract to 2 mL under a gentle stream of nitrogen. This extract was then injected into the 5977C GC/MSD.

Table 2. Selected ion monitoring (SIM) parameters.

Compound	Retention Time (min)	Quantifier Ion	Qualifier Ion 1	Qualifier Ion 2
Aniline	1.734	93	66	65
o-Toluidine	2.649	106	107	77
2,4-Xylidine	3.603	121	120	106
2,6-Xylidine	3.652	121	120	106
2-Methoxyaniline	3.906	108	123	80
p-Chloroaniline	4.242	127	129	65
p-Cresidine	4.922	122	137	94
2,4,5-Trimethylaniline	4.965	135	120	134
1,4-Phenylenediamine	5.409	108	80	107
4-Chloro-o-toluidine	5.423	141	106	143
2,4'-Toluenediamine	6.469	121	122	94
2,4'-Diaminoanisole	6.962	123	138	95
2-Naphthylamine	7.242	143	115	116
p-Nitro-o-toluidine	7.471	152	122	77
4-Aminobiphenyl	7.908	169	168	170
Anthracene-d10 (IS)	7.987	188	160	80
p-Aminoazobenzene	9.467	92	197	120
4,4'-Oxydianiline	9.817	200	108	171
4,4'-Diaminodiphenylmethane	9.879	198	197	106
Benzidine	9.925	184	183	185
o-Aminoazotoluene	10.33	106	225	134
3,3'-Dimethyl-4,4'-diaminodiphenylmethane	10.742	226	225	211
3,3'-Dimethylbenzidine	10.893	212	106	213
4,4'-Thiodianiline	11.475	216	184	215
4,4'-Methylene-bis(2-chloroaniline)	11.691	231	266	140

Table 3. Preparation of calibration solutions.

Calibration Level	Amines Mix Solution Volume (µL)	Anthracene-d10 (IS) Volume (µL)	Acetonitrile Volume (µL)	Final Concentration (µg/mL)
L1	62.5 (from Mix 2)	100	837.5	0.25
L2	125 (from Mix 2)	100	775	0.5
L3	250 (from Mix 2)	100	650	1.0
L4	62.5 (from Mix 1)	100	837.5	2.5
L5	125 (from Mix 1)	100	775	5.0
L6	250 (from Mix 1)	100	650	10.0
L7	500 (from Mix 1)	100	400	20.0

Results and discussion

Figure 1 shows the total ion chromatogram (TIC) of the amines standard mix at 5 µg/mL concentration. Figure 2 shows quantifier and qualifier peaks, along with the calibration curves of all amines ranging from 0.25 to 20 µg/mL concentration. Table 4 shows the %RSD for the concentration of six replicates at 1 µg/mL and Table 5 shows the compounds found in a few textile samples.

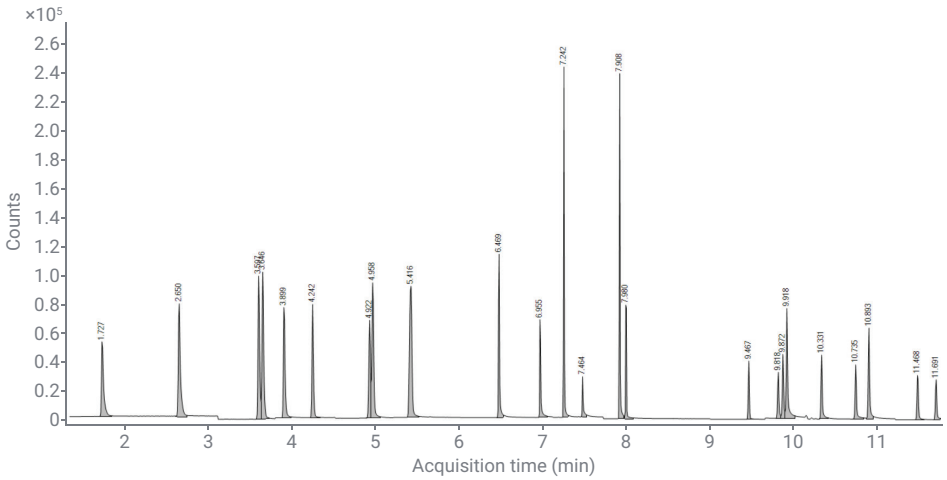
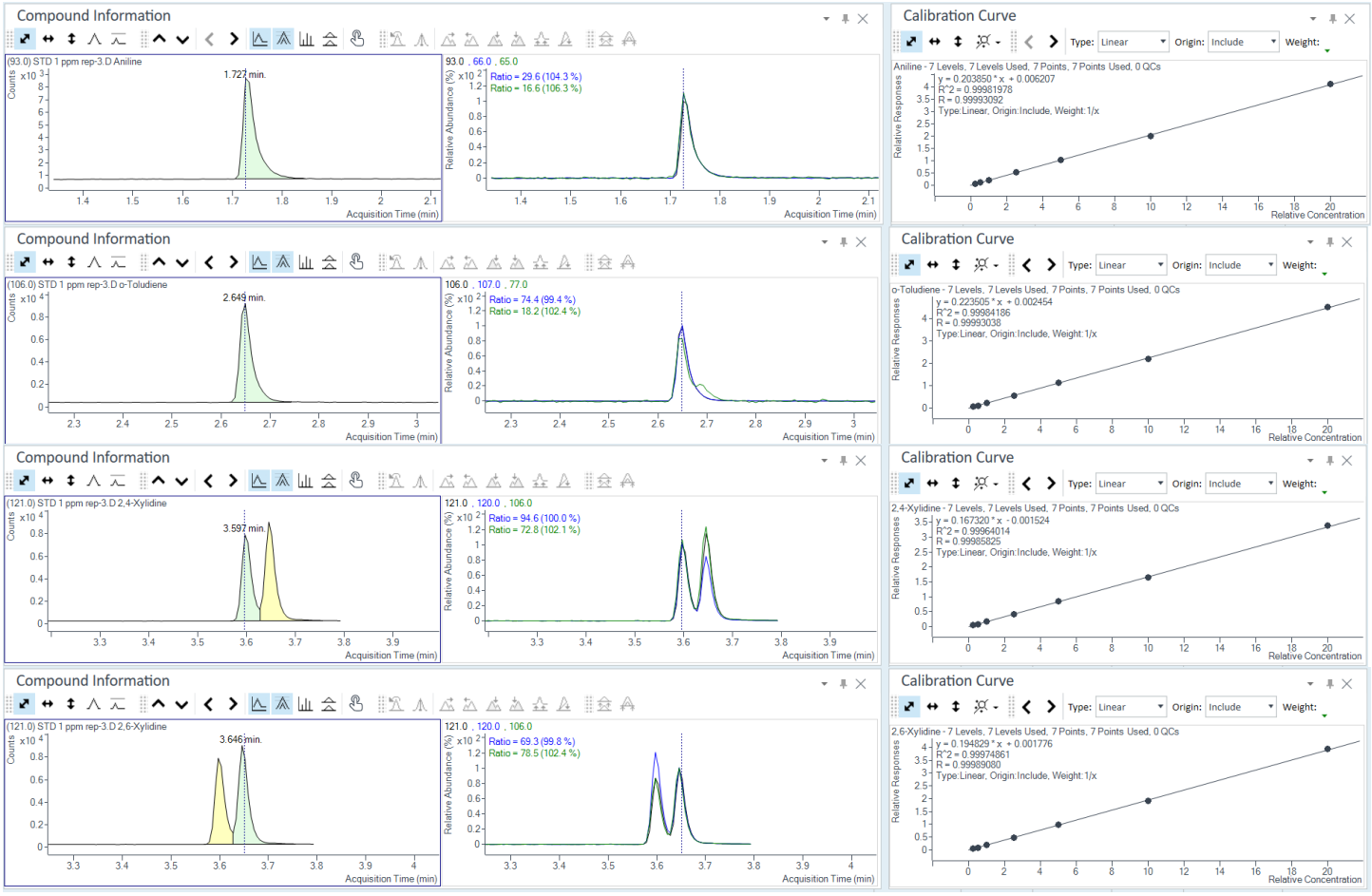
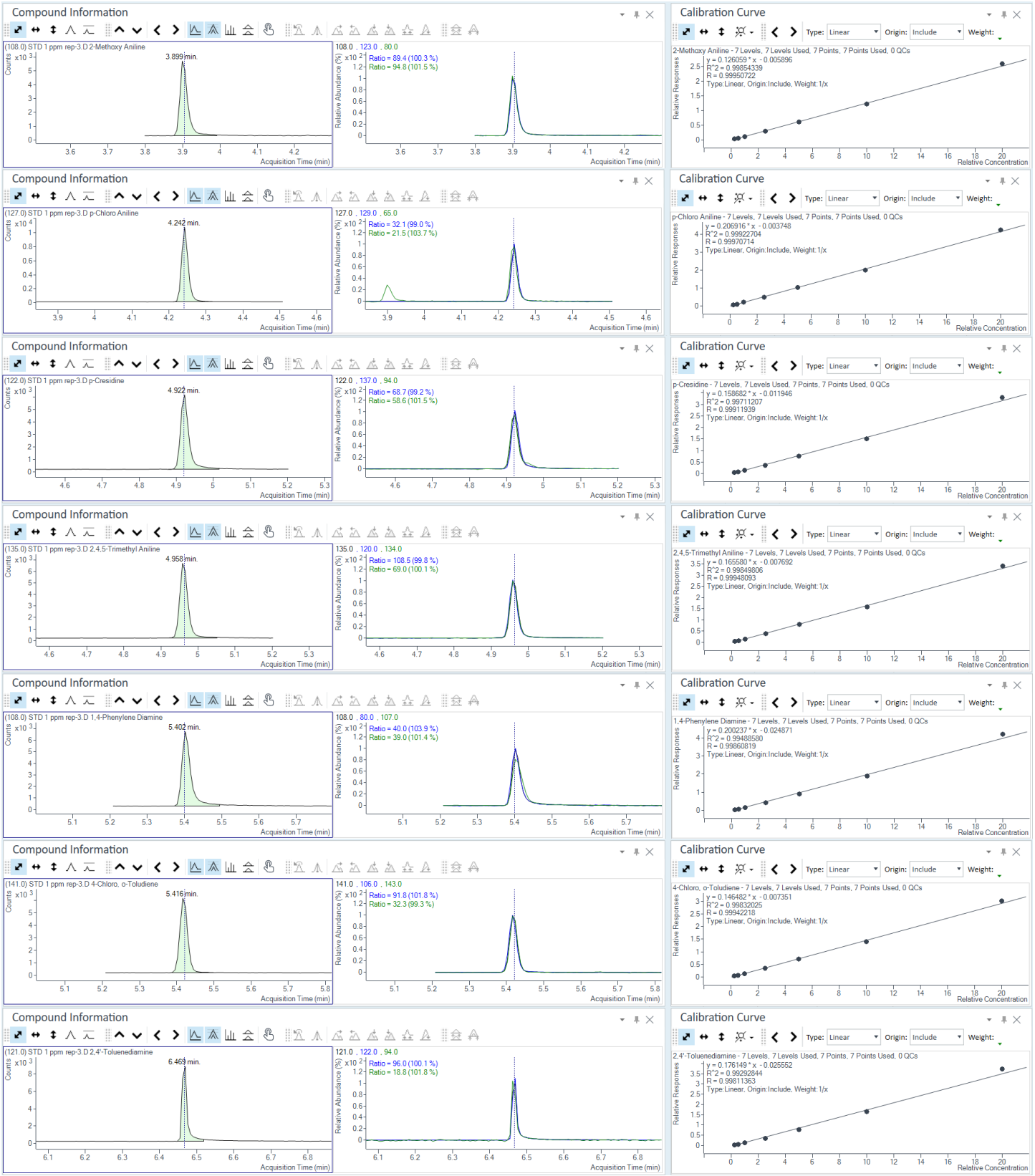


Figure 1. TIC (SIM) of amine standard mix at 5 µg/mL concentration.




Compound Information
(108.0) STD 1 ppm rep-3.D 2-Methoxy Aniline
3.899 min.
108.0 , 123.0 , 80.0
Ratio = 89.4 (100.3 %)
Ratio = 94.8 (101.5 %)
Calibration Curve
Type: Linear
Origin: Include
Weight:
2-Methoxy Aniline - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.126059 * x - 0.005896
R^2 = 0.99854339
R = 0.99950722
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(127.0) STD 1 ppm rep-3.D p-Chloro Aniline
4.242 min.
127.0 , 129.0 , 65.0
Ratio = 32.1 (99.0 %)
Ratio = 21.5 (103.7 %)
Calibration Curve
p-Chloro Aniline - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.206916 * x - 0.003748
R^2 = 0.99922704
R = 0.99970714
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(122.0) STD 1 ppm rep-3.D p-Cresidine
4.922 min.
122.0 , 137.0 , 94.0
Ratio = 68.7 (99.2 %)
Ratio = 58.6 (101.5 %)
Calibration Curve
p-Cresidine - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.158682 * x - 0.011946
R^2 = 0.99711207
R = 0.99911939
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(135.0) STD 1 ppm rep-3.D 2,4,5-Trimethyl Aniline
4.958 min.
135.0 , 120.0 , 134.0
Ratio = 108.5 (99.8 %)
Ratio = 69.0 (100.1 %)
Calibration Curve
2,4,5-Trimethyl Aniline - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.165580 * x - 0.007692
R^2 = 0.99849806
R = 0.99948093
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(108.0) STD 1 ppm rep-3.D 1,4-Phenylene Diamine
5.402 min.
108.0 , 80.0 , 107.0
Ratio = 40.0 (103.9 %)
Ratio = 39.0 (101.4 %)
Calibration Curve
1,4-Phenylene Diamine - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.200237 * x - 0.024871
R^2 = 0.99488580
R = 0.99860819
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(141.0) STD 1 ppm rep-3.D 4-Chloro, o-Toludiene
5.416 min.
141.0 , 106.0 , 143.0
Ratio = 91.8 (101.8 %)
Ratio = 32.3 (99.3 %)
Calibration Curve
4-Chloro, o-Toludiene - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.146482 * x - 0.007351
R^2 = 0.99832025
R = 0.99942218
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(121.0) STD 1 ppm rep-3.D 2,4'-Toluenediamine
6.469 min.
121.0 , 122.0 , 94.0
Ratio = 96.0 (100.1 %)
Ratio = 18.8 (101.8 %)
Calibration Curve
2,4'-Toluenediamine - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.176149 * x - 0.025552
R^2 = 0.99292844
R = 0.99811363
Type:Linear, Origin:Include, Weight:1/x
Counts
Relative Abundance (%)
Acquisition Time (min)
Relative Responses
Relative Concentration


Compound Information
(123.0) STD 1 ppm rep-3.D 2,4'-Diamino Anisole
6.955 min.
123.0 , 138.0 , 95.0
Ratio = 53.6 (96.5 %)
Ratio = 31.0 (91.9 %)
Calibration Curve
Type: Quadratic
Origin: Include
Weight:
2,4'-Diamino Anisole - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.002665 * x ^ 2 + 0.094366 * x - 0.002208
R^2 = 0.99971550
R = 0.99481186
Type:Quadratic, Origin:Include, Weight:1/x
Compound Information
(143.0) STD 1 ppm rep-3.D 2-Naphthylamine
7.242 min.
143.0 , 115.0 , 116.0
Ratio = 41.1 (102.5 %)
Ratio = 13.3 (92.0 %)
Calibration Curve
Type: Linear
Origin: Include
Weight:
2-Naphthylamine - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.325483 * x - 0.030887
R^2 = 0.99693277
R = 0.99906173
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(152.0) STD 1 ppm rep-3.D p-Nitro, o-Toludine
7.464 min.
152.0 , 122.0 , 77.0
Ratio = 156.1 (98.2 %)
Ratio = 71.5 (83.8 %)
Calibration Curve
Type: Quadratic
Origin: Include
Weight:
p-Nitro, o-Toludine - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 4.884671E-004 * x ^ 2 + 0.021669 * x + 9.826969E-004
R^2 = 0.99931613
R = 0.99541538
Type:Quadratic, Origin:Include, Weight:1/x
Compound Information
(169.0) STD 1 ppm rep-3.D 4-Aminobiphenyl
7.908 min.
169.0 , 168.0 , 170.0
Ratio = 21.5 (99.8 %)
Ratio = 13.3 (100.4 %)
Calibration Curve
Type: Linear
Origin: Include
Weight:
4-Aminobiphenyl - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.422395 * x - 0.054757
R^2 = 0.99369730
R = 0.99821838
Type:Linear, Origin:Include, Weight:1/x
Compound Information
(92.0) STD 1 ppm rep-3.D p-Aminoazobenzene
9.467 min.
92.0 , 197.0 , 120.0
Ratio = 57.7 (96.8 %)
Ratio = 26.5 (99.8 %)
Calibration Curve
Type: Quadratic
Origin: Include
Weight:
p-Aminoazobenzene - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.001702 * x ^ 2 + 0.054759 * x - 8.830296E-004
R^2 = 0.99990589
R = 0.99459041
Type:Quadratic, Origin:Include, Weight:1/x
Compound Information
(200.0) STD 1 ppm rep-3.D 4,4'-Oxydianiline
9.818 min.
200.0 , 108.0 , 171.0
Ratio = 33.7 (90.6 %)
Ratio = 19.8 (92.0 %)
Calibration Curve
Type: Quadratic
Origin: Include
Weight:
4,4'-Oxydianiline - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.002752 * x ^ 2 + 0.059999 * x - 0.001068
R^2 = 0.99994571
R = 0.99168713
Type:Quadratic, Origin:Include, Weight:1/x
Compound Information
(198.0) STD 1 ppm rep-3.D 4,4'-Diaminodiphenylmethane
9.872 min.
198.0 , 197.0 , 106.0
Ratio = 74.8 (106.2 %)
Ratio = 43.2 (97.5 %)
Calibration Curve
Type: Quadratic
Origin: Include
Weight:
4,4'-Diaminodiphenylmethane - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs
y = 0.002141 * x ^ 2 + 0.063240 * x - 0.005609
R^2 = 0.99926303
R = 0.99500553
Type:Quadratic, Origin:Include, Weight:1/x
Counts
Relative Abundance (%)
Acquisition Time (min)
Relative Responses
Relative Concentration

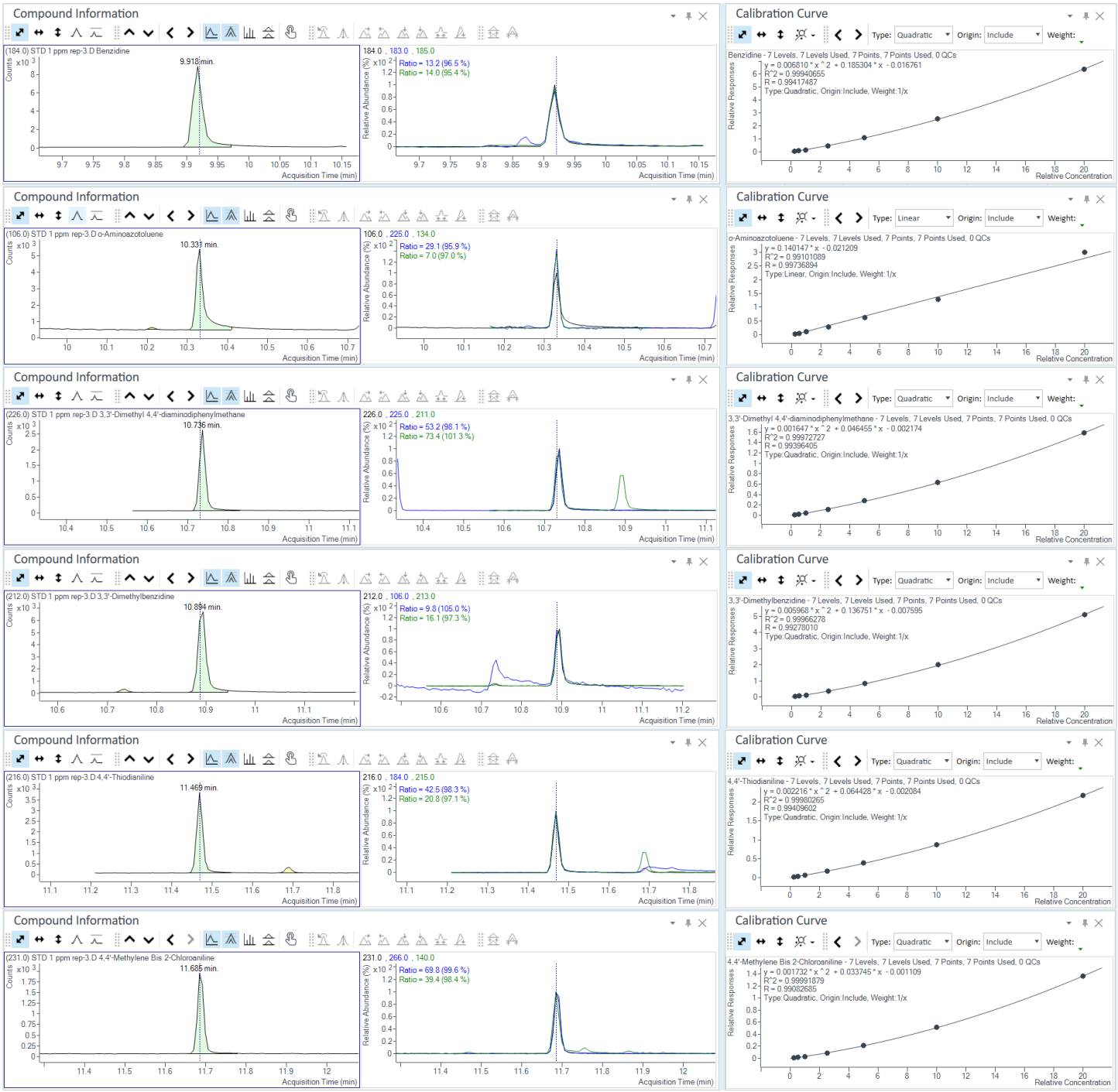


Figure 2. Peaks at 1.0 µg/mL concentration and calibration curves ranging from 0.25 to 20 µg/mL concentration for amine mix standard.

Table 4. %RSD for six replicates of 1.0 µg/mL amines mix standard.

Compound	Final Concentration (µg/mL)						%RSD
	Standard Replicate 1	Standard Replicate 2	Standard Replicate 3	Standard Replicate 4	Standard Replicate 5	Standard Replicate 6	
Aniline	0.955	0.943	1.065	0.848	0.893	0.833	9.219
o-Toluidine	0.963	0.965	1.026	0.911	0.920	0.868	5.796
2,4-Xylidine	0.951	0.959	0.990	0.923	0.923	0.902	3.347
2,6-Xylidine	0.965	0.970	1.000	0.915	0.931	0.895	4.109
2-Methoxyaniline	0.942	0.945	0.982	0.909	0.905	0.882	3.848
p-Chloroaniline	0.948	0.963	0.973	0.939	0.925	0.910	2.480
p-Cresidine	0.936	0.948	0.961	0.921	0.923	0.906	2.137
2,4,5-Trimethylaniline	0.940	0.954	0.961	0.932	0.915	0.914	2.064
1,4-Phenylenediamine	0.913	0.912	0.933	0.906	0.892	0.884	1.919
4-Chloro-o-toluidine	0.939	0.948	0.945	0.928	0.912	0.919	1.559
2,4'-Toluenediamine	0.921	0.889	0.887	0.888	0.886	0.875	1.733
2,4'-Diaminoanisole	0.968	0.979	0.989	0.979	0.956	0.942	1.804
2-Naphthylamine	0.936	0.910	0.926	0.887	0.916	0.931	1.947
p-Nitro-o-toluidine	1.002	1.009	1.023	1.002	1.049	1.019	1.761
4-Aminobiphenyl	0.894	0.899	0.876	0.901	0.879	0.895	1.205
p-Aminoazobenzene	0.966	0.976	0.964	0.974	0.951	0.966	0.912
4,4'-Oxydianiline	0.996	1.038	0.943	0.983	0.977	1.008	3.221
4,4'-Diaminodiphenylmethane	0.909	0.914	0.867	0.890	0.902	0.911	1.969
Benzidine	0.906	0.923	0.877	0.898	0.889	0.921	1.982
o-Aminoazotoluene	0.898	0.895	0.885	0.903	0.889	0.898	0.729
3,3'-Dimethyl-4,4'-diaminodiphenylmethane	0.937	0.970	0.920	0.938	0.943	0.984	2.512
3,3'-Dimethylbenzidine	0.930	0.938	0.907	0.939	0.932	0.957	1.719
4,4'-Thiodianiline	0.982	0.994	0.953	0.994	0.974	1.021	2.315
4,4'-Methylene-bis(2-chloroaniline)	0.993	0.987	0.963	0.987	0.970	1.011	1.731

Table 5. Results in textile samples.

Compound	Test Sample 1		Test Sample 2		Test Sample 3	
	RT (min)	Final Conc. (µg/g)	RT (min)	Final Conc. (µg/g)	RT (min)	Final Conc. (µg/g)
Aniline	ND	ND	1.734	91.8159	ND	ND
o-Toluidine	2.65	172.205	ND	ND	ND	ND
2,4-Xylidine	ND	ND	ND	ND	ND	ND
2,6-Xylidine	ND	ND	ND	ND	ND	ND
2-Methoxyaniline	ND	ND	ND	ND	ND	ND
p-Chloroaniline	ND	ND	ND	ND	ND	ND
p-Cresidine	ND	ND	ND	ND	ND	ND
2,4,5-Trimethylaniline	ND	ND	ND	ND	ND	ND
1,4-Phenylenediamine	ND	ND	ND	ND	ND	ND
4-Chloro-o-toluidine	ND	ND	ND	ND	ND	ND
2,4'-Toluenediamine	ND	ND	ND	ND	ND	ND
2,4'-Diaminoanisole	ND	ND	ND	ND	ND	ND
2-Naphthylamine	ND	ND	ND	ND	ND	ND
p-Nitro-o-toluidine	ND	ND	ND	ND	ND	ND
4-Aminobiphenyl	ND	ND	ND	ND	ND	ND
p-Aminoazobenzene	ND	ND	ND	ND	ND	ND
4,4'-Oxydianiline	ND	ND	ND	ND	ND	ND
4,4'-Diaminodiphenylmethane	ND	ND	ND	ND	ND	ND
Benzidine	ND	ND	ND	ND	9.918	46.4791
o-Aminoazotoluene	ND	ND	ND	ND	ND	ND
3,3'-Dimethyl-4,4'-diaminodiphenylmethane	ND	ND	ND	ND	ND	ND
3,3'-Dimethylbenzidine	ND	ND	ND	ND	ND	ND
4,4'-Thiodianiline	ND	ND	ND	ND	ND	ND
4,4'-Methylene-bis(2-chloroaniline)	ND	ND	ND	ND	ND	ND

Conclusion

This application note presents key strategies for the analysis of amines using GC/MS with hydrogen as the carrier gas, while maintaining sensitivity to meet the maximum residue limits (MRLs) as mentioned in part 1 of ISO:14362. The method includes an Agilent J&W DB-35ms column (20 m × 0.18 mm, 0.18 µm) and a hydrogen-compatible electron ionization source, the Agilent HydroInert source. The optimized setup with hydrogen showed good chromatographic resolution. The HydroInert source was shown to provide good sensitivity. The method presented here allowed for the quantitation of 24 target amines analytes at concentrations ranging from 0.25 to 20 µg/mL in an aqueous standard. Good repeatability was found with %RSD values below 5 for 22 of the 24 compounds. Real world textile samples were also analyzed using the same method and three target compounds—aniline, o-toluidine, and benzidine—were observed as present in those samples.

References

1. Nguyen, T. L.; Saleh, M. A. Thermal Degradation of Azobenzene Dyes. *Results Chem.* **2023**.
2. Nguyen, T.; Saleh, M. A. Detection of Azo Dyes and Aromatic Amines in Women Undergarment. *J. Environ. Sci. Health, Part A* **2016**, *51(9)*, 744–753. DOI: 10.1080/10934529.2016.1170446

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