



Archived at the Flinders Academic Commons:

<http://dspace.flinders.edu.au/dspace/>

'This is the peer reviewed version of the following article:

Pigou, P., Dennison, G. H., Johnston, M., & Kobus, H. (2017). An investigation into artefacts formed during gas chromatography/mass spectrometry analysis of firearms propellant that contains diphenylamine as the stabiliser. *Forensic Science International*, 279, 140–147. <https://doi.org/10.1016/j.forsciint.2017.08.013>

which has been published in final form at

<http://dx.doi.org/10.1016/j.forsciint.2017.08.013>

© 2017 Elsevier. This manuscript version is made available under the CC-BY-NC-ND 4.0 license <http://creativecommons.org/licenses/by-nc-nd/4.0/>

Accepted Manuscript

Title: An Investigation into Artefacts Formed During Gas Chromatography/Mass Spectrometry Analysis of Firearms Propellant that contains Diphenylamine as Stabiliser

Authors: Paul Pigou, Genevieve Dennison, Martin Johnston, Hilton Kobus



PII: S0379-0738(17)30312-2
DOI: <http://dx.doi.org/10.1016/j.forsciint.2017.08.013>
Reference: FSI 8956

To appear in: *FSI*

Received date: 16-11-2016
Revised date: 14-8-2017
Accepted date: 14-8-2017

Please cite this article as: Paul Pigou, Genevieve Dennison, Martin Johnston, Hilton Kobus, An Investigation into Artefacts Formed During Gas Chromatography/Mass Spectrometry Analysis of Firearms Propellant that contains Diphenylamine as Stabiliser, Forensic Science International <http://dx.doi.org/10.1016/j.forsciint.2017.08.013>

This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

An Investigation into Artefacts Formed During Gas Chromatography/Mass Spectrometry Analysis of Firearms Propellant that contains Diphenylamine as Stabiliser

Paul Pigou^{1,2}, Genevieve Dennison¹, Martin Johnston¹ and Hilton Kobus¹

¹School of Chemical and Physical Sciences, Flinders University, PO Box 2100, Adelaide SA 5001, Australia

²Forensic Science South Australia, 21 Divett Place, Adelaide SA 5000, Australia

Corresponding Author

Hilton Kobus

Email: Hilton.Kobus@flinders.edu.au

Highlights

- Identification of artefacts formed during gas chromatography analysis of firearms propellant.
- Identification of a previously unreported diphenylamine derivative.
- Identity of artefact confirmed by synthesis.

ABSTRACT

In the course of providing assistance to legal counsel in a matter that involved the analysis of firearms propellant by gas chromatography/mass spectrometry it was noticed that phenoxazine was reported as a component of 0.22 calibre propellant that contained diphenylamine as the stabiliser. The research was conducted to find how phenoxazine was formed. The results showed that the compound was not phenoxazine but a diphenylamine derivative 4-(phenylimino) cyclohexa-2,5-dien-1-one that formed in the injection port of the gas chromatograph. In addition a second artefact was found to form in the ion source of the mass spectrometer. While the presence of the artefacts does not affect the ability to identify particles as propellant they may impact on comparison with source ammunition.

Keywords: Gunshot residue, Firearms propellant analysis, forensic, chemistry

INTRODUCTION

Firearm propellant particles vary widely in colour, morphology and composition between different calibres of ammunition and between manufacturers. Their shape can include balls, flattened balls, disks, tubes and rods and colours can include green, yellow, grey and white (1). Propellant particles are often coated with a layer of graphite as an external lubricant and therefore will appear black, with the underlying colour only revealed when the coating is removed. The bulk of the chemical composition of firearms propellants is provided by the propellant compounds nitrocellulose and nitroglycerine (NG). Additives that include stabilisers, plasticizers and burn modifiers are present and contribute to the compositional variation (2,3). If partially burnt particles are located following a shooting incident examination of their physical appearance and analysis to determine their chemical profile can provide definitive evidence of a firearms origin and potential indication of a source ammunition,(2,3,4). Gas chromatography/mass spectrometry (GCMS) is a widely used analytical technique to determine the chemical profile of propellants (4,5).

Ethyl centralite and diphenylamine (DPA) are common stabilisers added to propellants. Their function is to remove nitrogen oxides produced by the degradation of nitrocellulose and nitroglycerine and so prevent autocatalytic decomposition of the propellant. DPA, the stabiliser studied in this paper, produces a range of nitro and nitroso derivatives (6-8) from the reaction with the propellant decomposition products. The reaction pathways for this process are shown in Figure 1 with the primary reaction/degradation product being N-nitroso DPA (NnDPA). This compound undergoes further reactions to produce the nitrodiphenylamine (NDPA) products shown in Figure 1. The detection of NDPAs as components of propellant can be an indicator of the level of degradation and a possible measure of age. .

Figure 1.here

In the course of providing assistance to legal counsel in a matter where the analysis of firearms propellant by GCMS was involved it was noticed that phenoxazine (Figure 2) was reported as a component of 0.22 calibre propellant that contained diphenylamine as the stabiliser (9). No reference to phenoxazine being a component of propellant formulations could be found and it was difficult to explain as a possible by-product of the manufacturing process. The formation of this compound is also not indicated in the reaction scheme for the nitration of DPA in the cartridge case as shown in Figure 1. While it is recognised that the elevated temperatures within the injection port of the gas chromatograph would lead to formation of the products shown in figure 1 (10) the possibility of the formation of other DPA-related products beside those shown in figure 1 should be considered. It had been noted during the course of routine work within the laboratory that the gas chromatographic profile of propellants was influenced by the cleanliness of the inlet path and this could also contribute to artefact formation.

Figure 2 here

In this paper we report on investigations undertaken to determine whether phenoxazine or other DPA related products could be formed as artefacts in the injection port of the GC.

EXPERIMENTAL

Propellant Sample

Propellant was obtained by unpacking a 0.22 rimfire cartridge of PMC (Korea) manufacture known to consist of nitrocellulose, nitroglycerine, diphenylamine (stabiliser) and dibutyl phthalate (plasticizer). GCMS analysis confirmed this composition.

GCMS Analysis

Instrumentation and Conditions

Analysis was performed using an Agilent Technologies 7890A gas chromatograph with a 7693 autosampler and a 5975C mass selective detector (MSD) operated from 40 to 500 amu in electron impact (EI) mode with an ionization energy of 70eV and source temperature of 230°C (unless otherwise stated). Helium was used as the carrier gas at constant flow of 68 cm/s with an inlet split ratio of 20:1; the column was a 30 m x 0.25mm x 0.25µm HP-5ms fused-silica capillary and the injection port was held at 200°C (unless otherwise stated). GC conditions were (i) for the large number of sequential samples the initial column temperature was 60°C for 2 minutes and then ramped at 40°C/min to 250°C (ii) later work investigating possible co-elution used an initial column temperature of 140°C for 1 minute and then ramped at 10°C/min to 250°C.

Production of Artefacts

An attempt was made to induce the appearance of artefacts in the analysis profile due to inlet contamination through 200 injections of propellant solutions without changing the injection port liner. To promote inlet port soiling, a concentrated solution of propellant particles dissolved in acetonitrile (20mg/mL) was prepared for repetitive analysis through an SGE split/splitless liner with silanised glass wool. It is recognised that such a large number of repetitive injections and using a concentrated propellant solution does not reflect routine analytical practice but this was done to ensure the formation of any artefacts.

Similar studies were made by co-injecting nitroglycerine with diphenylamine, nitroglycerine with 2-NDPA and nitroglycerine with N-nitrosodiphenylamine.

Chemicals

Standards of phenoxazine (TCI Co. Ltd.), diphenylamine (AnalaR, BDH Chemicals Ltd.) and N-nitroso diphenylamine (Sigma-Aldrich) were used.

Synthesis of 4-(Phenylimino)cyclohexa-2,5-dien-1-one

This compound (Figure 3) was identified as a possible GC artefact in the experiments and it was synthesised as follows to provide a standard for comparison with the suspected artefact.

4-Hydroxydiphenylamine (500mg, 2.7 mmol) was dissolved in dichloromethane (30 mL) and Dess-Martin periodinane (1.15 g, 2.7 mmol) was added with stirring at room temperature. An immediate colour change to deep red was noted and the mixture left to stir for a further 15 minutes. The reaction mixture was diluted with dichloromethane (70 mL) and quenched with sodium thiosulphate (2.0 g) in saturated sodium bicarbonate solution (aq, 100 mL) and the mixture stirred vigorously for

15 minutes. The organic phase was isolated, washed with water (1x25 mL), dried (Na_2SO_4) and the solvent removed in vacuo. The dark residue was purified by column chromatography (neutral alumina, Brockman II) eluting with chloroform to yield the product as a deep orange to red solid (0.46 g, 93% yield).

The identity of the desired compound was confirmed by GCMS analysis (see Figure 4) and NMR spectroscopy.

Figure 3 here

RESULTS AND DISCUSSION

The Effect of a Soiled Liner on the Chromatographic Profile

The amount of DPA detected decreased significantly with the number of injections and as expected this was accompanied by an increase in the amount of 2N and 4N DPA. This was assumed to be an indication of soiling of the inlet liner and is shown in Figure 4. Notably, phenoxazine (retention time of 6.77 min and base peak m/z 183 Da) was not detected in these chromatograms but another compound with a prominent m/z 183 Da appeared at a retention time of approximately 6.2 minutes after about 60 injections. The retention times of the propellant components, artefacts and phenoxazine are shown in table 1.

Figure 4 here

Identification of the artefact at R_t 6.2 min and m/z 183 Da

Phenoxazine shows the same base peak at 183 Da but the remainder of the mass spectrum shows clear differences, for example phenoxazine has a prominent ion at m/z 127 while the spectrum of the compound at R_t 6.2' shows an ion at m/z 129. Other differences can be seen as shown in Figure 5. A mass spectral library search (NIST11) gave the compound 4-(phenylimino)cyclohexa-2,5-dien-1-one (or N-phenyl-1,4-benzoquinone) as the highest ranked candidate; with phenoxazine ranked second. Similarity between the two spectra suggests that a library search could place phenoxazine as the first choice in the absence of other possibilities. Therefore if a phenoxazine standard was not run it is possible that it was misidentified from a library search.

Figure 5 here

GCMS analysis of the synthesised 4-(phenylimino)cyclohexa-2,5-dien-1-one gave both retention time and mass spectrum corresponding to that of the propellant artefact peak. This clearly excludes phenoxazine and strongly supports the identity of 4-(phenylimino)cyclohexa-2,5-dien-1-one as the artefact. However, this did not explain the presence of an ion of 185 Da.

Identification of the source of the ion at m/z 185 Da

Subtraction of the contribution from 4-(phenylimino)cyclohexa-2,5-dien-1-one from the mass spectrum of the component at R_t 6.2 min left a mass spectrum resembling 4-(phenylamino)phenol (Figure 6). However, this compound had a retention time of 6.9 minutes and so could not be co-eluting with the artefact at 6.2 min. Investigations to determine whether there was co-elution with

other components revealed that it was actually a further artefact induced by the high temperature (230°C) of the ion source. With an ion source temperature of 130°C, the 185 Da contribution was minimized as illustrated in Figure 7. Subtraction of this spectrum from the mass spectrum derived with an ion source temperature of 230°C provided a spectrum once again similar to 4-(phenylamino)phenol.

Figures 6 and 7 here

Possible Formation of the 4-(phenylamino)cyclohexa-2,5-dien-1-one Artefact.

The GCMS analysis of a mixture of DPA and NG using the soiled liner is shown in Figure 8. The artefact peak at 6.2 minutes is clearly visible indicating a DPA reaction with NG is a contributing source. The expected production of 2 and 4NDPA is also evident.

Figure 8 here

The primary nitration product of DPA is NnDPA (4) and co-injection of a solution of NnDPA and NG produced a chromatographic profile essentially identical to that provided in Figure 8; showing DPA not NnDPA due to the thermal instability of the latter. So, while a reaction between NG and NnDPA must be considered as a possible source of the artefact, since there is reversion to DPA, the primary reaction may be between DPA and NG. Conversely, co-injection of a mixture of NG and 2-NDPA or 4-NDPA did not produce the artefact component at 6.2 minutes.

We have not been able to explain how the artefact is formed and are unable to propose a reaction mechanism at this time. Further research will investigate this issue.

The effect of a soiled liner can also be induced by using elevated injection port temperatures. Figure 9 shows the change in analysis profile as thermal decomposition is induced in the injection port. The production of nitroDPA compounds with the associated loss of NG and DPA is the most prominent feature with rising temperature but the formation of 4-(phenylimino)cyclohexa-2,5-dien-1-one is also indicated.

Figure 9 here

CONCLUSION

Two diphenylamine analogues have been shown to be produced as artefacts during GCMS analysis of firearms propellant that contains diphenylamine as the stabiliser.

(i) 4-(phenylimino)cyclohexa-2,5-dien-1-one is formed as an artefact in the injection port by reaction of DPA with the nitro-esters or their decomposition products. The injection port liner conditions and temperature are important contributory factors to the formation of this artefact

(ii) 4-(phenylamino)phenol is formed as an artefact in the ion source of the mass spectrometer. Higher temperatures in the ion source (230C) favour the formation of this artefact.

It should also be remembered that the amount of 2N and 4N DPA can be enhanced over that present in the propellant sample due to reactions in the injection port.

While the occurrence of artefacts does not affect the ability to identify a particle as a propellant from its chemical profile, caution must be exercised if any quantitative or semi-quantitative comparisons with a source propellant are to be made. GCMS is a valuable technique for propellant analysis due to its sensitivity and resolution as long as an awareness of the potential for artefact formation is maintained. Contamination of the inlet liner and any artefact formation can be easily monitored by routine quality management procedures where blanks and standards are interspersed between samples.

References

- (1) **online Smokeless Powders Database [internet]. National Center for Forensic Science, University of Central Florida. Available from: <http://www.ilrc.ucf.edu/powders/>**
- (2) Wallace JS (2008), *Chemical Analysis of Firearms, Ammunition and Gunshot residues*, CRC Press.
- (3) DMK Dennis, MR Williams and ME Sigman (2016), Assessing the evidentiary value of smokeless powder comparisons, *Forensic Science International* 259: 179-187
- (4) RM Heramb and B McCord (2002), The manufacture of smokeless powders and their forensic analysis: A brief review, *Forensic Science Communications*, 4(2), 1-4
- (3) Akhavan J (2006), *The Chemistry of Explosives*, Royal Society of Chemistry,
- (4) Goudsmits, E, Sharples G and Birkett J (2015), Recent trends in organic gunshot residue analysis. *Trends in Analytical Chemistry* 74: 46-57.
- (5) Dalby O, Butler D, Birkett J (2010), Analysis of Gunshot Residue and Associated Materials - A Review, *Journal of Forensic Sciences*, 55, 924-943
- (6) Lindblom, T. (2002). "Reactions in Stabilizer and Between Stabilizer and Nitrocellulose in Propellants." *Propellants Explosives Pyrotechnics* 27(4): 197-208.
- (7) Lussier LS and Gagnon H (2000), On the chemical reactions of diphenylamine and its derivatives with nitrogen dioxide at normal temperature conditions, *Propellant Explosives Pyrotechnics* 25, 117-125.
- (8) Curtis NJ and Rogasch PE (1987), Determination of derivatives of diphenylamine in Australian gun propellants by high performance liquid chromatography, *Propellants Explosives Pyrotechnics*, 12, 158-163
- (9) Strobel N (1993), *The characterisation of 0.22 calibre ammunition propellant by GCMSD*, MSc thesis, RMIT University, Melbourne, Australia
- (10) Gassener and C Weyerman (2016), LCMS method development and comparison of sampling materials for the analysis of organic gunshot residues, *Forensic Science International*, 264, 47-55

LIST OF FIGURES

Figure 1: Reaction scheme for the nitration of diphenylamine. DPA – diphenylamine, NnDP – N-nitrosodiphenylamine, nDPA – nitrodiphenylamine, DNDPA - dinitrodiphenylamine

Figure 2: Structure of phenoxazine and diphenylamine

Figure 3: 4-(Phenylimino)cyclohexa-2,5-dien-1-one

Figure 4: Top: Comparison of chromatograms of same PMC propellant solution using new and dirty (200 injections) inlet liners. The component marked 'DBP' is dibutylphthalate. Lower: Reconstructed ion chromatograms for m/z 183. Traces show the appearance of a component at 6.2 minutes using GC conditions (ii) as described in the experimental section.

Figure 5: Mass spectra of phenoxazine (top), 6.2 minute peak (ion source 230°C) and the search result (middle).

Figure 6: Structure and mass spectrum of 4-(phenylamino)phenol.

Figure 7: 4-(Phenylimino)cyclohexa-2,5-dien-1-one reference material analyzed with the ion source at 230°C (top spectrum) and 130°C (middle spectrum), with the difference spectrum (bottom).

Figure 8: Chromatogram of a mixture of nitroglycerin and diphenylamine which was introduced through a dirty inlet liner (upper) with the mass spectrum of the 6.2' artefact (lower).

Figure 9: A mixture of nitroglycerin and diphenylamine analysed by GCMS at low and high inlet temperatures (retention times differ due to changed conditions).

FIGURE 1

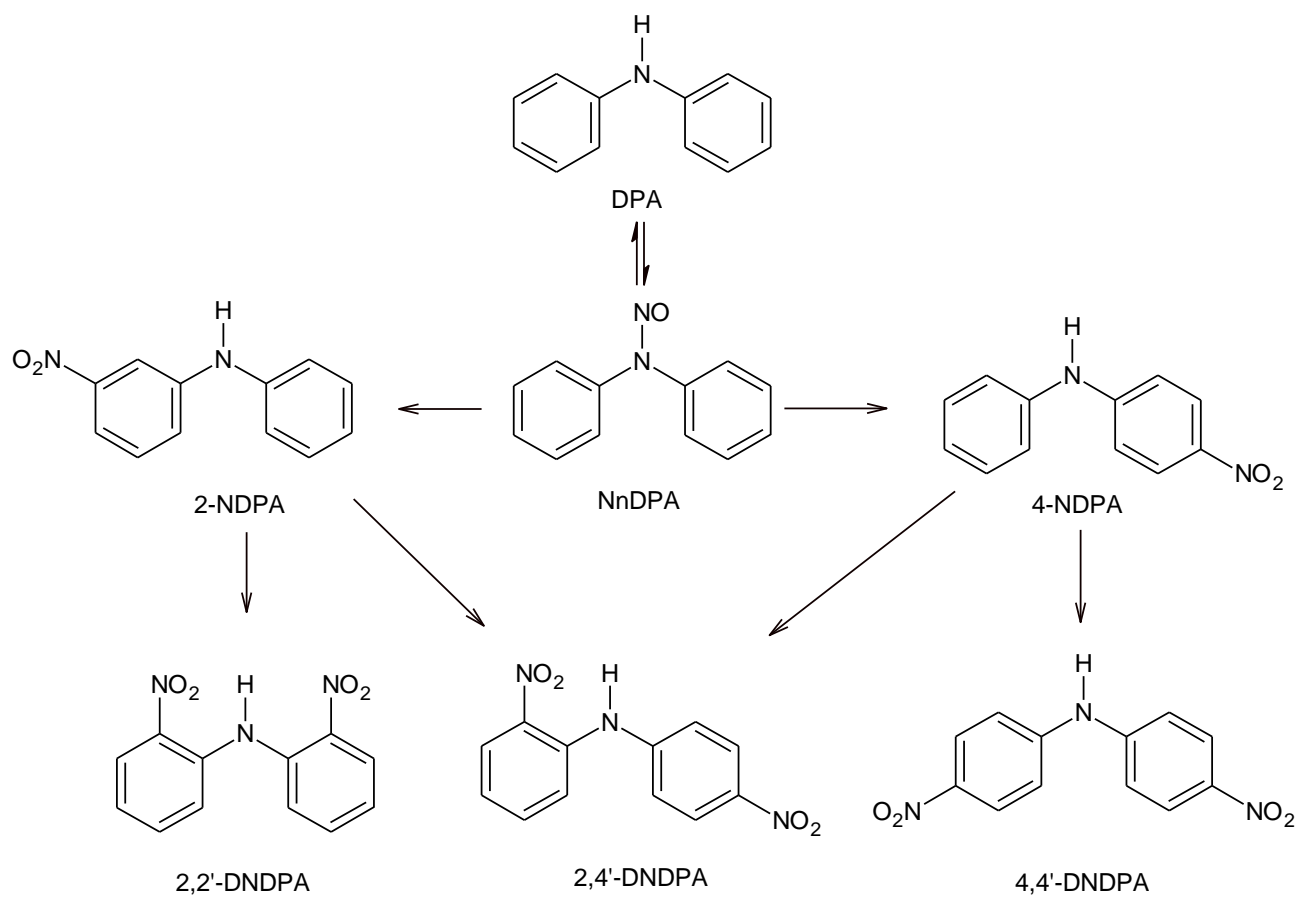
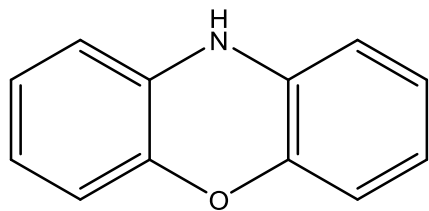


FIGURE 2



Phenoxazine

FIGURE 3

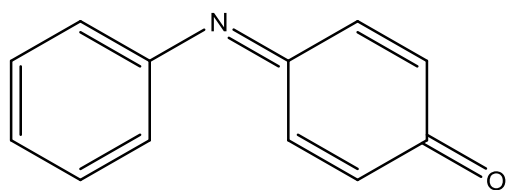


FIGURE 4

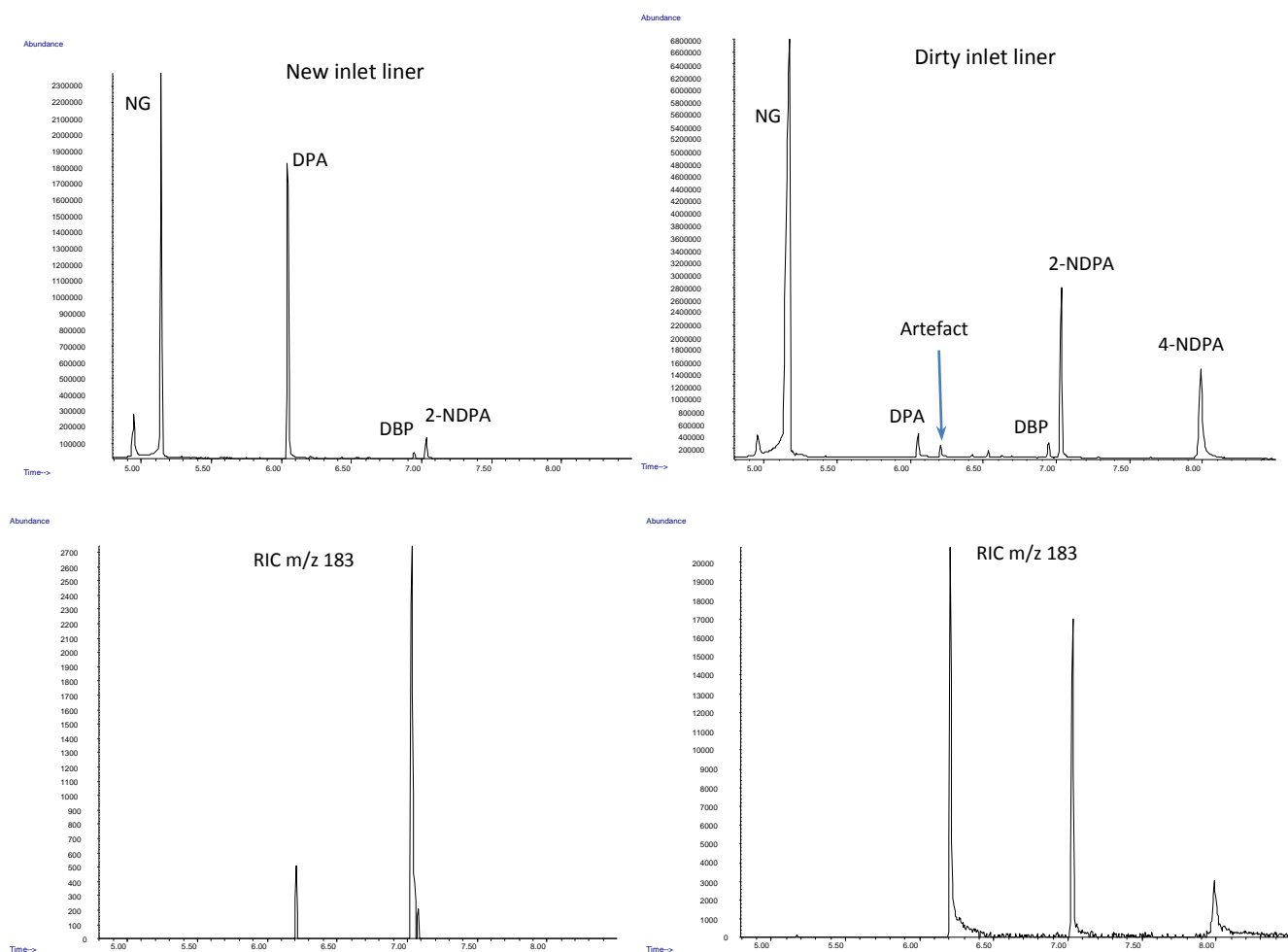


FIGURE 5

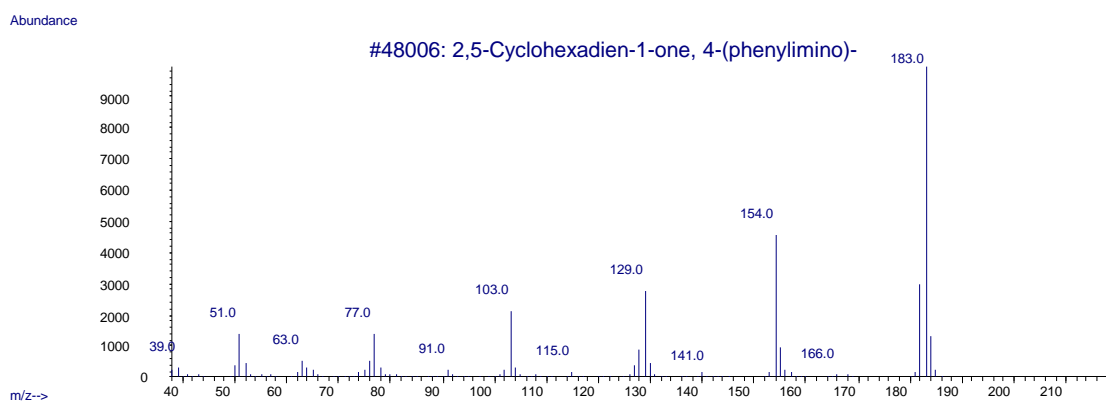
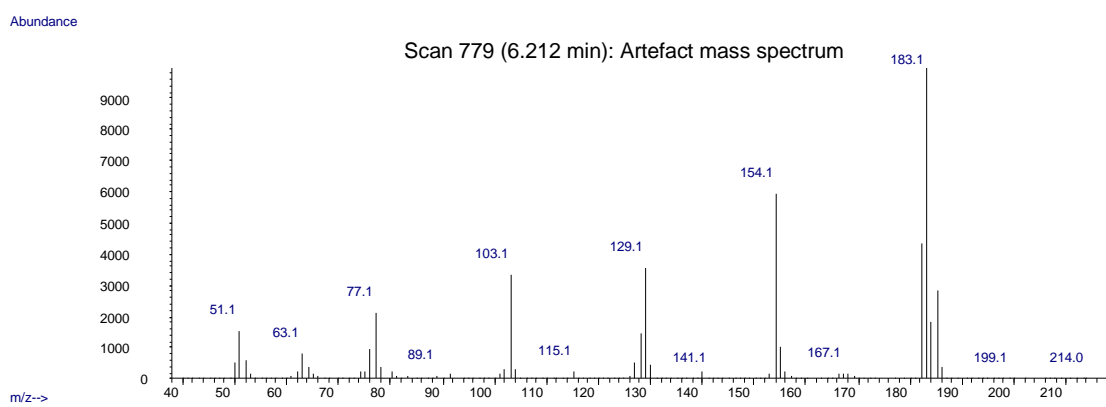
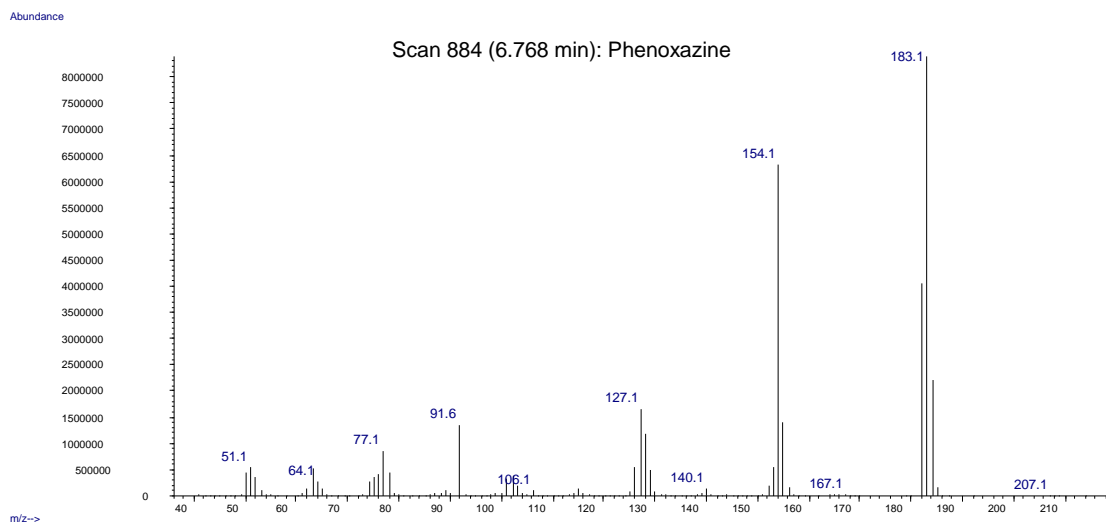


FIGURE 6

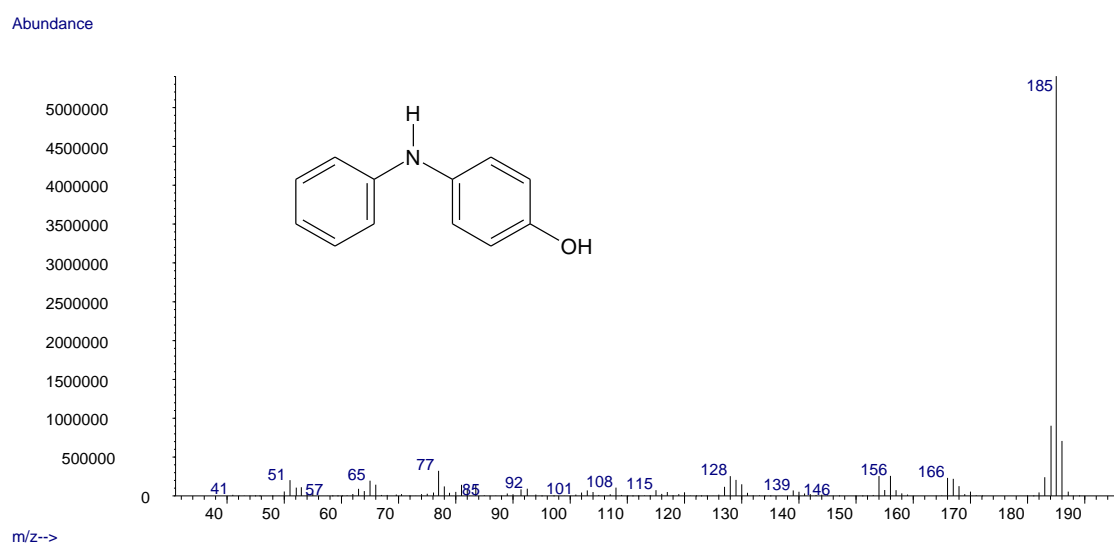


FIGURE 7

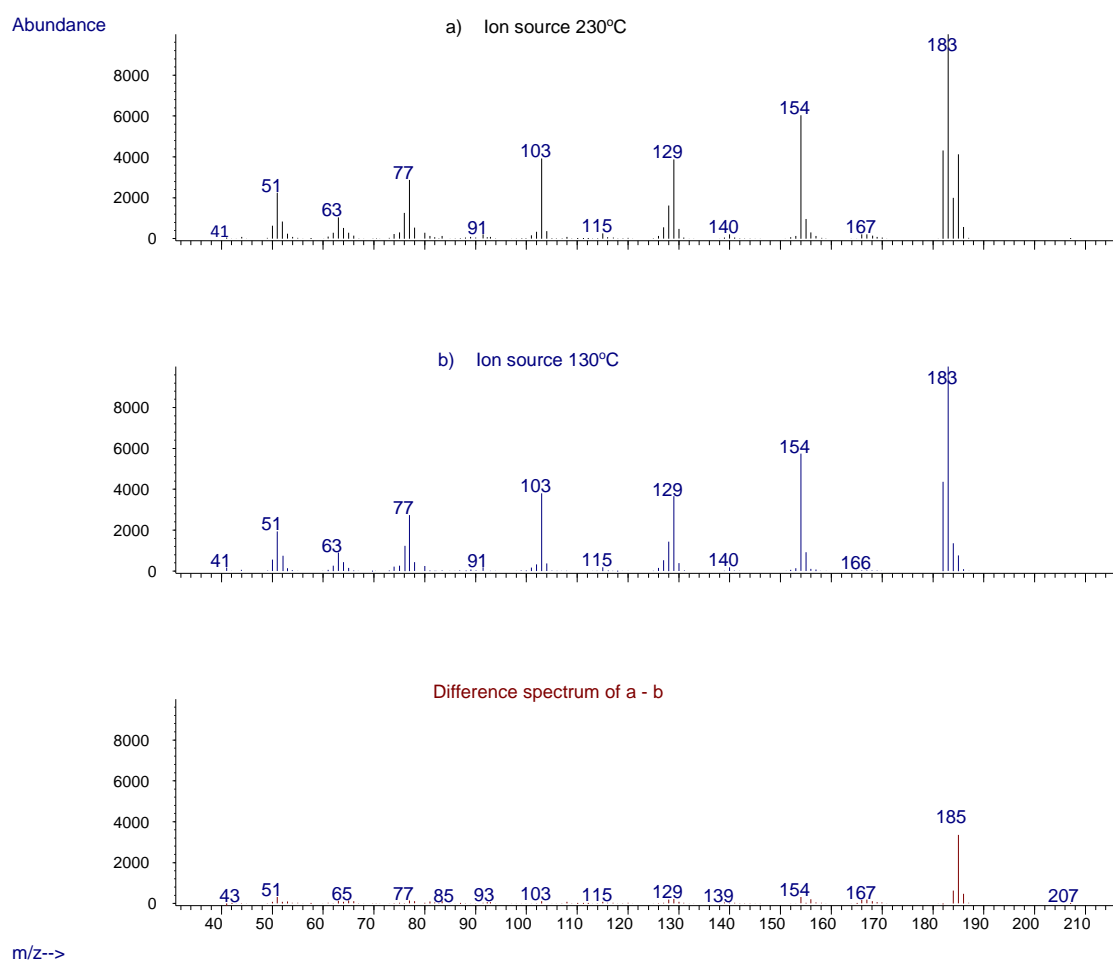


FIGURE 8

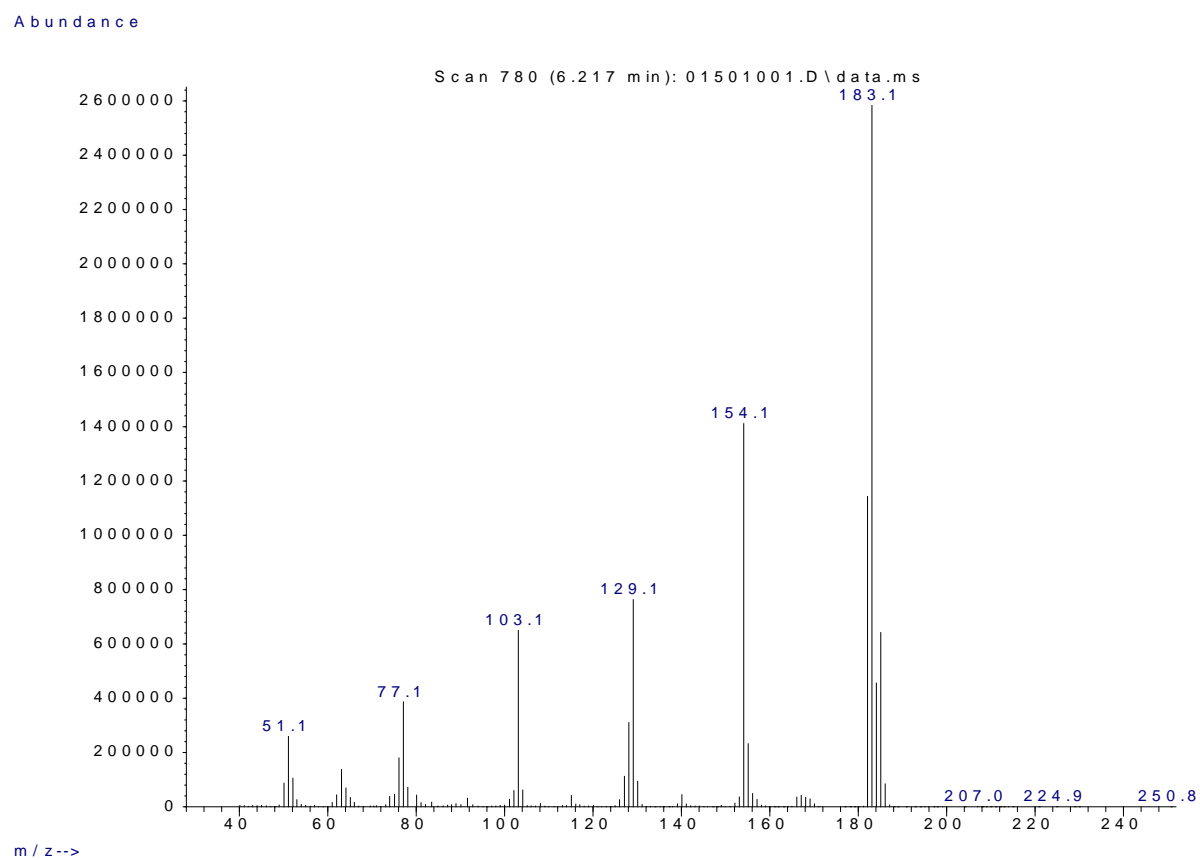
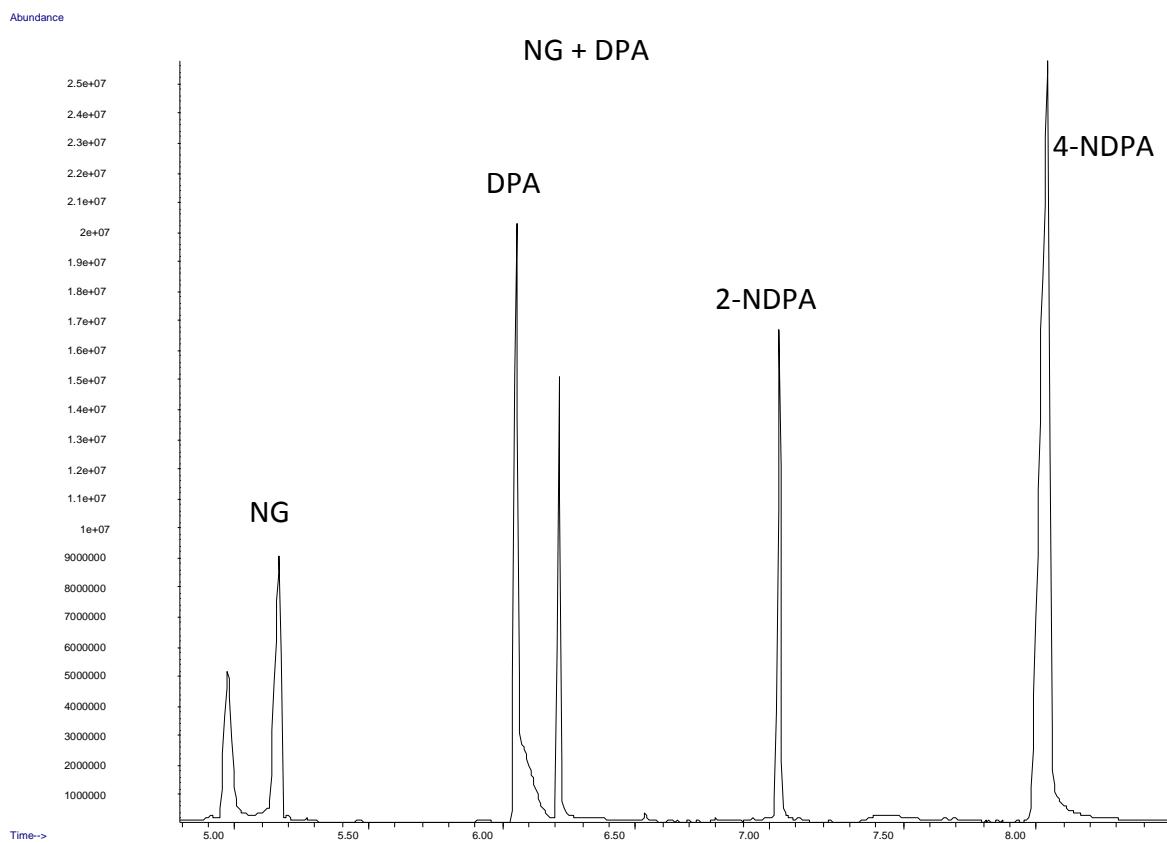


Figure 9

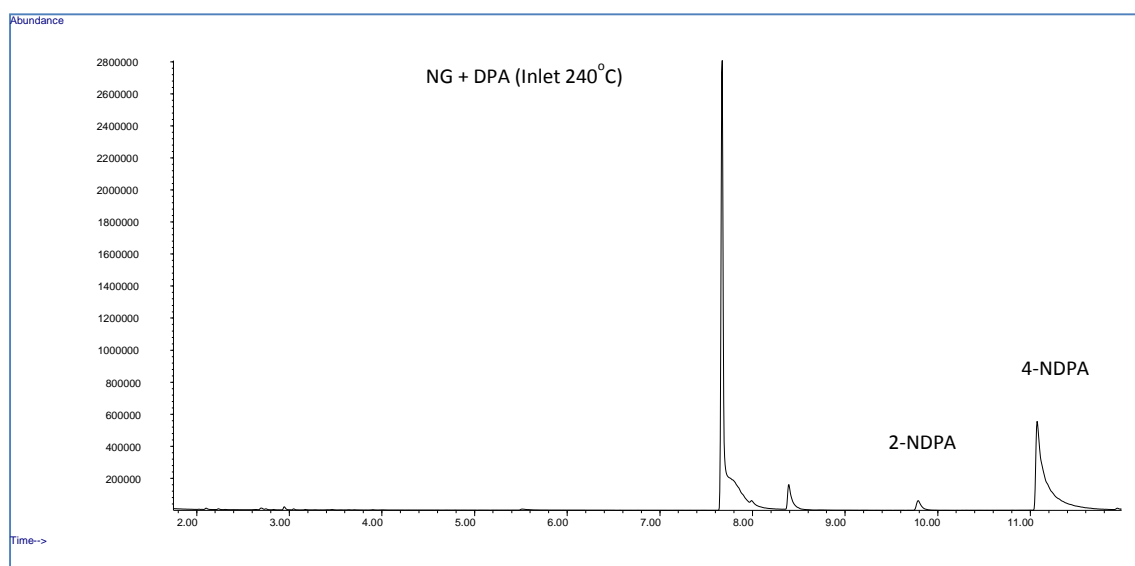
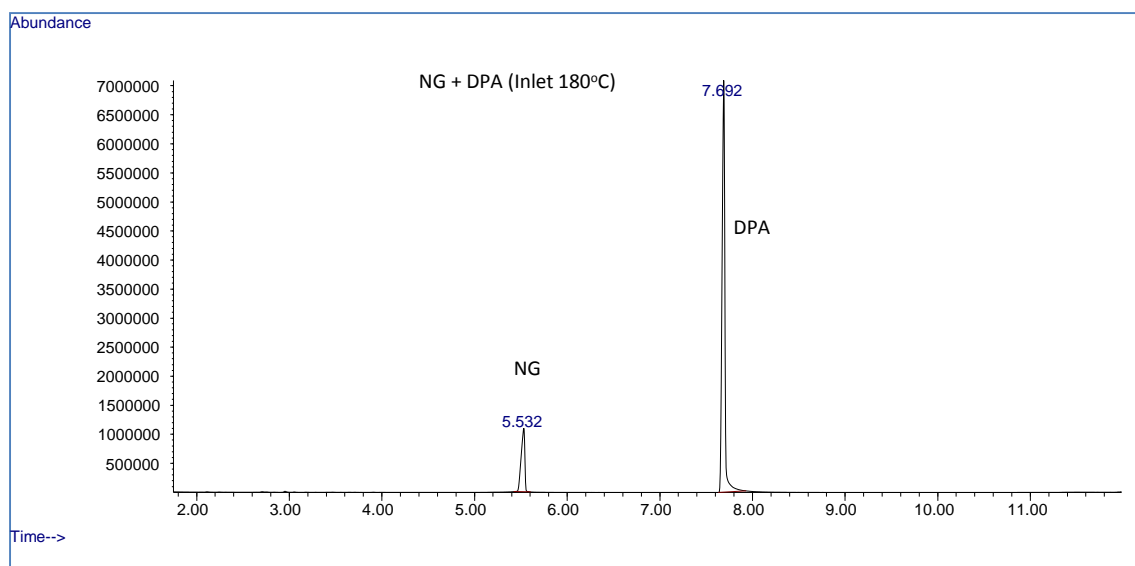


Table 1: Compounds and retention times for propellant chromatograms

Compound	Rt GC(i) (min)	Rt GC(ii) (min)
Nitroglycerine (NG)	5.15	5.53
Diphenylamine (DPA)	6.05	7.69
Artefact	6.22	8.00
Phenoxazine	6.77	9.12
4-(phenylamino)phenol	6.91	9.41
Dibutylphthalate (DBP)	6.95	9.64
2-nitroDPA (2NDPA)	7.04	9.74
4-nitroDPA (4NDPA)	7.99	11.1