FORENSIC APPLICATIONS OF
DIFFERENTIAL MOBILITY SPECTROMETRY

A thesis presented

by

Samantha Mosley

to
The Department of Chemistry and Chemical Biology

In partial fulfillment of the requirements for the degree of
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ABSTRACT

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*Salvia divinorum* can be mostly found in southern Mexico where it is a native plant to the Oaxaca region. Presently, *Salvia divinorum* is the only known source of Salvinorin A, a hallucinogen which is quite potent. For people, an ingestion of a small dosage of 200-500µg can cause a person to go into a hallucinogenic state. Currently, forensic labs are using GC/MS in order to identify the presence of *Salvia divinorum*, which takes the instrument approximately 30 minutes to analyze.

Smokeless powder is one of the most common types of explosives today and can be used many ways, including ammunition. Since this is so common, the forensic field has become interested in being able to detect and identify the powder. Smokeless powder is a mixture of different components, including several different stabilizers, which make it sometimes difficult to identify what is used. Currently, the most common instrumentation used is GC/MS, which takes approximately 25 minutes.

Differential Mobility Spectrometry (DMS) is a growing technology for gas phase ion separation. When paired with mass spectrometry, DMS offers many advantages that could help with ion separation, including improvements in mass spectral signal to noise. This thesis presents a new method for ESI-DMS in order to provide a rapid ion separation. Instead of having a run time of over 25 minutes, DMS can help detect and identify the desired ion in one minute.
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LIST OF ABBREVIATIONS

DMS    differential mobility spectrometry
FAIMS  field asymmetric waveform ion mobility spectrometry
IMS    ion mobility spectrometry
MEMS   micro-electro-mechanical systems
NIST   National Institute of Standards and Technology
ESI    electrospray ionization
Vc     compensation voltage
kg     kilogram
g      gram
NMR    nuclear magnetic resonance
g/mol  grams per mole
µg     microgram
w/w    weight to weight
GC/MS  gas chromatography-mass spectroscopy
µL     microliter
mL     milliliter
µL/min microliter per minute
kV     kilovolt
nL/mL  nanoliter per milliliter
L/min  liter per minute
V          volt
ng/mL      nanogram per milliliter
mg         milligram
1. Introduction

1.1 Differential Mobility Spectrometry

Differential Mobility Spectrometry (DMS), also referred to as field asymmetric waveform ion mobility spectrometry (FAIMS), is a new technology that is very similar compared to Ion Mobility Spectrometry (IMS) [1-4]. DMS is a tool that is used for ion separation and identification of gas phase ions. Mostly, DMS is used with a combination of mass spectrometry [5]. The technique of DMS ion separation was first developed in Russia and was first reported in journal publications in the early 1990s [6, 7]. There are a few different instrumentation designs for the DMS. Two main types are planar and coaxial [45]. For this experiment, the instrumentation used was developed by Miller et. al. using microelectronics micro machined (MEMS) technology, which manufacture planar DMS sensors [8]. Sionex Corporation has commercially manufactured the MEMS electrode DMS sensors and is available as the microDMx™ [6, 44]. Figure 1 shows a microDMx™ sensor, which was produced by Sionex Corporation.

![Figure 1: Sionex Corporation manufactured microDMx™ sensor](image-url)
As mentioned before, DMS is similar to IMS in the way that the ions are separated by a drift gas at atmospheric pressure. However, DMS differs because there is an asymmetric electric field waveform that is applied to two parallel electrode plates [2, 3]. Between these plates, the ions pass through in a continuous manner at a non-pulsed rate. The electrical waveform consists of a high field strength of one polarity, which could go up as high as 30,000V/cm, then a low field duration of opposite polarity, which could go less than 1000V/cm. Figure 2 shows the asymmetric electrical waveform and demonstrates the duration of the high field and low field portions is applied so that the net voltage being applied to the electrode is zero. The high and low voltages of opposite polarity are identified as the Rf voltage [8, 10]. Since the ions are distinguished by the difference between mobilities at low and high electric fields, this shows that the ion mobility is dependent on the applied field strength [11].

While traveling through the electrode plates, an ion may have an occurrence of a net mobility towards one of the plates. Because of this, a compensation voltage (Vc) is applied. After determining the desired Vc for a given sample, the Vc is applied as a constant DC voltage in order to maintain a safe path for the ion to pass through the plates without touching either one of them. The ions can pass through the plates in two different ways. The ions are either pulled through by a pump downstream from the electrodes or they are pushed using a pressurized gas flow upstream of the electrode plates [8, 10].
In DMS, the smaller the ion is, it will experience few collisions as the drift gas pulls the ions through. The drift gas has been discovered to be an influential role for the field dependence of the ion mobility [11, 12]. Miller et. al. has described the mobility to be related to the clustering reaction as the ion goes through the high and low electric fields as demonstrated in Figure 2. An ion will experience clustering in the drift gas during low field, which will then lead to an increase of the cross sectional area. For the high field, the clustering is dissociated which will then reduce the ion cross sectional area [10].

Using the microDMx™ sensor, it is possible to identify the ideal Rf and Vc voltage settings for the ion of interest. While scanning both the Rf and Vc voltages, the ion signal should be monitored which will then give off a plotted 3-dimensional plot called a dispersion plot. The plot demonstrates that as the Rf increases then there is typically an increase as well in ion separation as a function of differences in Vc. Typically, the higher the Rf voltage, a more definite peak should be able to be seen.
1.2 History and Botany of *Salvia divinorum*

*Salvia divinorum* is a native plant to the Oaxaca region, but over the years has started to spread north and is beginning to be cultivated in numerous parts of the U.S. [13]. *Salvia divinorum* is part of over 1000 species of *Salvia* in the world and comes from the Lamiacaea family. *Salvia divinorum* is considered a perennial herb in which it can reach up to 1.5 meters in height [14]. The plant consists of a stem, which is hollow and square, and sometimes flowers. On the stem, leaves grow as opposite pairs. Flowering is not really common, but when they are observed, they are white with a purple calyx [14].

In 1953, Mr. R. Gordon was the first to get the attention on *Salvia divinorum* while he was on ethnobotanical visits to the Sierra Mazateca region of Oaxaca, Mexico [15]. Gordon was actually supposed to be researching the practice of the hallucinogenic mushrooms which were used by the shamans, medicine men, of the Mazatec tribe. While doing research, he was informed of another plant that was used when the mushrooms were not available. However, even with this information, Gordon did not look at *Salvia divinorum* the 1960s. In years 1960-1961, the plant was collected then sent to Dr. Carlos Epling and Dr. R.E. Shultes in order to get identification [16]. Gordon’s attempt for identification was not the first to be reported. In 1938, J.B. Johnson had observed ritual rites that were using the plant being used, however the identification could not be provided [16]. Also, in 1952, R.G. Wietlaner observed ritual rites in which a plant was used, but that could not be identified either [17]. It was not until 1962 when more plant specimens were collected, did Dr. Epling finally identify that the plant was
botanically classified [16]. Dr. Epling sited *Salvia divinorum* into the Dusenostachys section of the Calosphace subgenus [18].

*Salvia divinorum* is considered to be a weaker hallucinogen compared to both the morning glory seeds and the different species of mushrooms used for divination, diagnosis and healing [27]. To the Mazatec Indians, *Salvia divinorum* is referred to as ska Maria, ska Pastora, hierba de Maria, hojas de la Pastora, which all refer to the belief that *Salvia divinorum* is the incarnation of the Virgin Mary [39]. Small doses of *Salvia divinorum* is used by the Mazatec healers for medicinal purposes because of its hallucinogenic effects. The Mazatec Indians believe that the some of the medicinal purposes include the improvement of headaches, anemia, and treatment of a swelling belly known as panzon de barrego, which is caused by a shaman’s curse [27]. When using *Salvia divinorum*, counting is always done by pairs. These medicinal purposes are prepared by rubbing fresh leaves in bare hands and placing them into water, which is not boiled [16]. Generally for medicinal use, the number of leaves seem to be about five pairs. For divination rituals, larger amounts of leaf pairs, which were prepared in the same manner, allow for the hallucinogenic properties of the plant to be experienced [27].

Valdés et al. declare that shamans use approximately 20 to 60 leaf pairs for divination rites [18]. The belief is that during the divination, the shaman is able to go to heaven, see the future and see where the illness originated as well as finding the cure [17].
1.3 Chemistry and First Extraction Techniques of *Salvia divinorum*

In 1980, Dr. Albert Hoffman was one of the first to attempt to identify the hallucinogenic compounds that are present in *Salvia divinorum* [18]. Hoffman was unsuccessful in the identity and it was not until 1982 when Ortega and Valdes were able to identify the compound by isolating Salvinorin A from dried samples of *Salvia divinorum* [19, 20]. Valdes did an extraction of 5.36kg of *Salvia divinorum* with ether. This was then followed with liquid-liquid separation between hexanes and 90% methanol. The liquid-liquid separation was done in order to remove any non-polar components from the crude extract. Extraction was then purified by flash column chromatography over silica gel. Recrystallization from ethanol gave Valdes 1.2g of Salvinorin A [20]. The structure was determined using $^1$H and $^{13}$C nuclear magnetic resonance (NMR) and X-ray crystallography. Ortega et. al. also performed an extraction, but not as complicated as Valdes. Ortega et. al. did an extraction of 200g of dried *Salvia divinorum* leaves with chloroform and then did a column chromatography by purification of activated clay. After doing a recrystallization from methanol, an unspecified amount of Salvinorin A was seen. The structure was determined by infrared analysis, X-ray crystallography, NMR [19]. Each group gave the compound different names where Valdes referred to it as ‘divinorum A’ and Ortega referred to it as “Salvinorin A”. Since Ortega had the earlier publication, it has been determined that the proper name for the compound is in fact Salvinorin A [20]. Figure 3 shows the structure of Salvinorin A.
Salvinorin A is a neoclerodan diterpene which is also known as *Salvia divinorum*, a psychoactive terpenoid, and is a unique component to the salvia plant [21]. Salvinorin A has been named and classified by the Chemical Abstract Service (CAS) as 2H-Naphtho[2, 1-c]pyran-7-carboxylic acid, 9-(acetyloxy)-2-(3-furanyl)dodecahydro-6a, 10b-dimethyl-4, 10-dioxo-, methyl ester, (2S, 4aR, 6aR, 7R, 9S, 10aS, 10bR)-(9CI). The CAS # is 83729-01-5 and the molecular formula is $C_{23}H_{28}O_8$ with a molecular weight of 432.47 g/mol.

Scheerer et. al. organically synthesized Salvinorin A [22] and many research groups have explored multiple analogues [23-26]. Salvinorin A is considered to be one of the most potent naturally occurring hallucinogens with an active dose as low as 200-500 µg [27, 28]. Siebert performed experiments in which he located Salvinorin A in the leaf's subcuticular space of the
glandular trichomes. The amount of Salvinorin A in a dry leaf ranges from 0.089 to 0.37\% (w/w) [29].

1.4 Smokeless Powder

Smokeless powder is a common kind of explosive which is frequently used in ammunition. Because it is such a common explosive, the detection of its identification has become a great interest in the forensic field [30, 31]. Smokeless powder has three special levels in which different chemicals are present. Single-base powders are propellants based solely on nitrocellulose. Double-base powders consist of a mixture of nitroglycerin which helps dissolve the nitrocellulose. Lastly, triple-base powders include nitroguanidine, which helps to have a flashless effect and is commonly used in military use [30, 32]. Along with the main component, there are also stabilizers such as diphenylamine, ethyl centralite, methyl centralite, and hydroquine [33-36]. The focus of this experiment will concentrate detecting two of the stabilizers present in smokeless powder. Figure 4 shows the structure of diphenylamine and structure 5 shows the structure of ethyl centralite.
Currently, GC/MS and nESI–MS are commonly used in order to identify and detect the stabilizers in smokeless powder. In 2007, Muller et. al. used GC/MS in order to detect and identify several different stabilizers. 10µm of smokeless powder was transferred into a 250µL
conical glass vial along with approximately 10µL of acetone. Vials were vortexed for an
unspecified amount of time. A Varian Star 3400CX gas chromatograph along with a Varian
Saturn 2000 Ion Trap was used to help identify the stabilizers. The GC conditions started at
100°C and increased to 250°C over a 30 minutes time period. The total ion chromatogram shows
diphenylamine eluting at approximately 13 minutes and ethyl centralite eluting at approximately
16 minutes [33].

Over the past decade, ESI has started to become an important technique for forensic
applications [46]. In 2009, Scherperel et. al. used nESI – MS to differentiate the various
components. Five milligrams of each powder was added to 1mL of methanol and vortexed for
10 seconds. Sample was centrifuged and then the powder was dried under vacuum. A Thermo
Scientific model LCQ quadrupole ion trap mass spectrometer (San Jose, CA, USA) was used to
help with detection. The flow rate was set to 1µL/min with the spray voltage set to 2.5 kV and
the heated capillary temperature at 125 °C. Scherperel et. al. found that the capillary voltage and
heated capillary temperature played a significant role on the abundance of the protonated
precursor ions and also on the in-source fragmentation [30].

These methods show that it is in fact possible to detect and identify both diphenylamine
and ethyl centralite using different techniques. This research investigates the usage of DMS in
hopes to create a more rapid way to detect and identify some components of smokeless powder.
2. Experimental

2.1 Instrumentation

The ESI-DMS-MS system consists of a modified Sionex Corporation microDMx™ differential mobility sensor and a Micromass ZQ detector. The ESI source includes a Valco microconnector union attached to a Newport micro movement plate. The ESI voltage is applied to the union where a liquid-liquid junction is created between peak tubing and a Proxeon steel emitter. The DMS is simply placed onto the mass spectrometer inlet and is stationed using a ring stand. The DMS is approximately 3” in length, 1” in height and ¼” in width. Samples were directly infused using a Hamilton 50µL syringe on a Harvard Apparatus 22 syringe pump at a flow of 300nL/min. All spectra were recorded in negative-ion mode at a capillary temperature of 125.0°C and a spray voltage of 2.0 kV. A drift gas line that came from the dopent had a constant flow of ultra high purity nitrogen at approximately 145 L/min. The flow was monitored using a helium Aalborg mass flow meter. In MS/MS measurements, the collision voltage varied between 30 and 60%. Data was accessed through the Xcalibar software.

DMS was used by the means of several different components. An Agilent E3612A DC power supply was set to 50V. An hp 6205B Dual DC Power Supply that varied in increments of 20V from 0-40V.
2.2 Extraction Technique of Salvinorin A

Purified Salvinorin A was obtained from Adam Hall. This standard was prepared at 10ng/mL and used in order to determine the experimental parameters to use for the ESI/DMS.

One ounce of *Salvia divinorum* was obtained from Adam Hall and was used for of the experiments. Samples of whole leaves were photographed and are shown in Figure 6.

![Salvia divinorum leaves in comparison to a quarter](image)

Figure 6: *Salvia divinorum* leaves in comparison to a quarter

Extractions of whole leaves were done in methanol, acetone, methylene chloride and ethanol. These four solvents were chosen because of previous experiments that were done for *Salvia divinorum* extractions [21,36-38]. All chemicals were obtained from Fisher Scientific (Pittsburgh, PA).
Siebert had determined that the most concentrated form of Salvinorin A was found in the glandular trichomes of the *Salvia divinorum* plant [29]. Therefore, all extractions were done with just whole leaves and it was not necessary to use crushed or powdered leaves. 100mg samples of whole leaves were placed into three 250 Erlenmeyer flasks. 5.0mL of acetone was added to each flask. Each flask was swirled once per minute for 30 minutes. After the last swirl, 1mL was transferred to a 2mL Fisherbrand microcentrifuge tube and placed into a Savant Speed Vac for 30 minutes. After 30 minutes, tubes were placed into a freezer until further use. This procedure was repeated with methanol, ethanol and methylene chloride. Each solvent gave off different shade of green due to chlorophyll. Methanol solutions were the darkest green and acetone solutions were the lightest. All chemicals obtained from Fisher Scientific (Pittsburgh, PA) and were used without any further purification.

2.3 Sample Preparation for Smokeless Powders

Diphenylamine and ethyl centralite were obtained Cerrilliant (Round Rock, TX). Diphenylamine came as a 5000µg/mL sample in 1.2mL MeOH and ethyl centralite came as 500µg/mL sample in 1.2mL MeOH. Standards were prepared at 10ng/mL in 70:30:0.1 MeOH:H₂O:FA and used in order to determine parameters for smokeless powder samples.

Three different samples of smokeless powders were obtained from Adam Hall; Alliant blue dot smokeless, powder from a bullet cartridge and powder from the National Institute of Standards and Technology (NIST). Each powder was prepared in the same manner. Four flakes
of the smokeless powder were transferred into a 2mL Fisherbrand screw thread vial. 500µL of acetone was added to vial and mixed using a Baxter Vortex Mixer for approximately 2 minutes until all flakes were dissolved. 200µL was transferred into a separate 2mL screw thread vial and the rest was placed into a freezer for later use. In the separate vial, 800µL of a 70:30:0.1 methanol:water:formic acid (MeOH:H₂O:FA) mixture was added. Vial was vortexed for 30 seconds. Because of the polymers of the nitrocellulose that is found in the smokeless powders, the vials were placed in a Galaxy Mini 6000rpm centrifuge and were centrifuged for an hour to allow the separation of the polymer.

2.4 Procedure

2.4.1 Salvinorin A

Vials from freezer were reconstituted with either 1mL 70:30:0.1 MeOH:H₂O:FA or 1mL 50:50:0.1 ACN:MeOH:FA. The Rf voltage was set to either 1000V or 1500V and the compensation voltage was scanned from -43 to +15V in 0.19V increments over 10 minutes. The MS signal was collected for the m/z range of 400-500 looking for the 433 ion during the Vc scan. Once scan was complete, a set Vc was able to be calculated by taking the highest peak of the spectrum, multiplying it by 60 (change from minutes to seconds), multiplying by 0.096 (the increments of the voltage at each second) and subtracting from -43V. After calculating the desired Vc, the compensation voltage was set to selected values and then ran for one minute. Mass was also collected for each sample condition with the DMS off and also in-line for
comparisons. Blanks of the 70:30:0.1 MeOH:H₂O:FA 50:50:0.1 ACN:MeOH:FA were also run to determine that there was no carry over and any of the desired analytes in the blank.

2.4.2 Smokeless Powder

Due to little sample preparation, the smokeless powder samples were able to be made on the same day in which they were going to be run through the DMS. The Rf voltage was set to 1000V and the compensation voltage was scanned from 43 to +15V in 0.19V increments over ten minutes. The MS signal was collected for the m/z range of 50-300 looking for the 170 ion for diphenylamine and also for the 269 ion for the ethyl centralite during the Vc scan. Once both samples were separated, just like previously explained, the calculation was done in order to determine each ion’s specific Vc. Once again, blanks of the 70:30:0.1 MeOH:H₂O:FA were run along with the mass for each sample condition with the DMS off and also in-line for comparisons.

3. Results and Discussion

3.1 Salvinorin A

Since DMS is dependant on the dopent that is used, finding the right dopent took some time. ESI-MS data obtained from a 10ug/mL Salvinorin A standard in 70:30:0.1 MeOH:H₂O:FA with isopropanol alcohol (IPA) as the dopent is shown in Figure 7.
Figure 7: Mass spectrum of Salvinorin A standard in 70:30:0.1 MeOH:H₂O:FA with DMS on, 20V, Rf=1000, Full Scan, and IPA 145

ESI-MS data obtained from a 10ug/mL Salvinorin A standard in 70:30:0.1 MeOH:H₂O:FA with ethyl acetate (EtOAc) as the dopent is shown in Figure 8.
Figure 8: Mass spectrum of Salvinorin A standard in 70:30:0.1 MeOH:H₂O:FA with DMS on, 20V, Rf=1000, Full Scan, and EtOAc 145

ESI-MS data obtained from a 10μg/mL Salvinorin A standard in 70:30:0.1 MeOH:H₂O:FA with methylene chloride (CH₂Cl₂) as the dopent is shown in Figure 9.
In the IPA and EtOAc dopents, the desired ion of 432.8 +/- 0.5 m/z is present but not at a great abundance, so it was determined that both were not goods dopent for the separation of Salvinorin A. Looking at Figure 9, it is seen that that the desired ion is seen along with the presence of a solvent peak. Because of this, it was determined that CH$_2$Cl$_2$ would be the dopent in hopes for good separation for Salvinorin A.

Figure 10 shows mass spectrum of a MeOH extraction sample, where a pair of leaves was soaked in MeOH for 30 minutes. The compensation voltage is set to the calculated voltage is
which in theory should only show the 432.8 m/z. The ion is present, but the voltage has also picked up a lot of other ions. This shows that MeOH may not be the best solvent to use in order to extract Salvinorin A from *Salvia divinorum*.

![Mass spectrum of MeOH extract with DMS on, 20V, Rf=1000, Vc=-0.78, and CH₂Cl₂ 145](image)

Since MeOH did not seem like a great solvent, others were used to attempt to extract Salvinorin A. Figure 11 shows an ethanol extraction, Figure 12 shows a CH₂Cl₂ extraction and Figure 13 shows an acetone extraction.
Figure 11: Mass spectrum of ethanol extract with DMS on, 20V, Rf=1000,
Vc=-1.41, and CH2Cl2 145

Figure 12: Mass spectrum of CH2Cl2 extract with DMS on, 20V, Rf=1000,
Vc=-0.84, and CH2Cl2 145
Each of the last three figures show that the extraction solvent has separated Salvinorin A successfully from *Salvia divinorum*. The 415 m/z is a solvent peak and seems to appear in all the samples, but when ran through MS/MS, it disappears. Either extraction solvent could be used and be able to obtain good data. In this case, the best solvent is CH$_2$Cl$_2$ giving an abundance of 5.28E4 and ethanol being the worst of the three giving an abundance of 1.34E4. However, all three were better than MeOH, which gave an abundance of 3.73E3. Figure 14, Figure 15 and Figure 16 show the mass spectra for the CH$_2$Cl$_2$ extract, acetone extract and ethanol extract,
respectively, after being filtered once in order to see if it is possible to minimize the noise in the mass spectra.

Figure 14: Mass spectrum of CH$_2$Cl$_2$ filtered extract with DMS on, 20V, Rf=1000, Vc=-0.84, and CH$_2$Cl$_2$ 145
Figure 15: Mass spectrum of acetone filtered extract with DMS on, 20V, Rf=1000, Vc=-1.07, and CH₂Cl₂ 145

Figure 16: Mass spectrum of ethanol filtered extract with DMS on, 20V, Rf=1000, Vc=-1.41, and CH₂Cl₂ 145
Figures 14-16 show that filtering the extract actually increases the abundance, in some cases quite a lot, and also minimizes the noise a little bit. This could be that the filtering is removing some of the other components in the plant, which could include chlorophyll.

### 3.2 Smokeless Powder

For this part of the experiment, it was determined to start with CH$_2$Cl$_2$, since it worked so well for Salvinorin A. After the first couple of runs, CH$_2$Cl$_2$ turned out to work quite well and was used as the dopent for smokeless powder as well. ESI-DMS data obtained from a 10µg/mL ethyl centralite standard in 70:30:0.1 MeOH:H$_2$O:FA is shown in Figure 17.
Figure 17: 10μg/mL ethyl centralite standard in 70:30:0.1 MeOH:H₂O:FA with DMS on, 20V, Rf=1000, Full Scan, CH₂Cl₂.

The TIC may not be the best looking, however it does show that fact that the extraction was done in one minute and gives a great mass spectrum of ethyl centralite for the preferred parameters. Peak A is the desired ion for ethyl centralite, whereas peaks B and C are from the solvent of 70:30:0.1 MeOH:H₂O:FA. MS/MS was done on all three peaks and it was determined that peaks B and C were nothing as they fell apart. Peak A however was determined to be positive for ethyl centralite. MS/MS spectrum of ethyl centralite standard can be seen in Figure 18.
At a collision energy of 30%, the ion breaks down and helps determine that the ion is in fact ethyl centralite.

ESI-DMS data obtained from a 10µg/mL diphenylamine standard in 70:30:0.1 MeOH:H₂O:FA is shown in Figure 19. The desired peak is D, and the other peaks are from the solvent of 70:30:0.1 MeOH:H₂O:FA. MS/MS was done on both peaks D and E and it was determined that the peak E was nothing as it fell apart during the process. Peak D however was
determined to be positive for diphenylamine, which was expected since it was the standard.

MS/MS spectrum of diphenylamine standard can be seen in Figure 20.

Figure 19: 10μg/mL diphenylamine standard in 70:30:0.1 MeOH:H₂O:FA with DMS on, 20V, Rf=1000 and Full Scan
After determining the parameters for each of the desired components from the smokeless powder, it was possible to test three different sources. Figure 21 shows a background mass spectrum of the 70:30:0.1 MeOH:H₂O:FA to show that neither diphenylamine nor ethyl centralite is present.
The first smokeless powder that was tested came from NIST. By extracting the 170 and 269 m/z ions from the TIC, Figure 22 shows a distinction between the two ions. At first glance, it looks as if only the 269 ion is present in the NIST smokeless powder. However, after doing the appropriate calculations to determine the correct volts, it was determined that diphenylamine came off at a voltage of -12.6V and ethyl centralite came off at a voltage of -9.5V. Figure 23 and Figure 24 show the mass spectra of diphenylamine and ethyl centralite, respectively.
22: TIC and MS of smokeless powder from NIST with DMS on, 20V, Rf=1500V, and Full Scan
Figure 23: Mass spectrum of diphenylamine in NIST smokeless powder

Figure 24: Mass Spectrum of ethyl centralite in NIST smokeless powder
Figure 25 shows the data that was collected using the same parameters as when the DMS was on. In this figure it can be seen that it looks as if only the 269 m/z ion has been registered. If zoomed in, it is possible to see the 170 m/z, however, the abundance is small. With the use of the DMS, the abundance of each ion is able to be optimized and also be separated from other ions and solvent peaks.
Figure 26: TIC and MS of smokeless powder from bullet cartridge with DMS on, 20V, Rf=1000V, and Full Scan
The second smokeless powder that was tested came from a bullet cartridge provided by Adam Hall. By extracting the 170 and 269 m/z ions from the TIC, Figure 26 shows a small distinction between the two ions. Even though Figure 26 shows a smaller distinction than the smokeless powder from NIST, there is still a voltage difference between the two ions. Unlike the first tested smokeless powder, both 170 and 269 m/z ions are seen. However, after doing the appropriate calculations to find the correct volts, it was determined that diphenylamine came off at a voltage of -2.9V and ethyl centralite came off at a voltage of -2.3V. Figure 27 and Figure 28 show the mass spectra of ethyl centralite and diphenylamine, respectively, from the bullet cartridge.
Figure 27: MS of smokeless powder from bullet cartridge
with DMS on, 20V, Rf=1000V, Vc= -2.3V

Figure 28: MS of smokeless powder from bullet cartridge
with DMS on, 20V, Rf=1000V, Vc= -2.9V
The third sample that was run through the ESI-DMS was a blue alliant blue dot smokeless powder which was provided by Adam Hall. When extracting the 170 and 269 m/z ions from the TIC, Figure 29 shows that there is only a difference of one volt between the two
ions. Just like the smokeless powder from the bullet cartridge, there is still a voltage difference between the two ions which helps distinguish each individually. This particular smokeless powder sample behaves in a very similar manner in which both the 170 and 269 m/z ions can be seen right away, where the 170 m/z seems to be more dominate as well. Again after doing the calculations, it was determined that diphenylamine came off at a voltage of -2.9V and ethyl centralite came off at a voltage of -1.9V. Figure 30 and Figure 31 show the mass spectra of ethyl centralite and diphenylamine, respectively, from the blue dot alliant powder.

Figure 30: MS of smokeless powder from Blue Dot with DMS on, 20V, Rf=1000V, Vc= -2.9V
In order to be certain that the ions were in fact the desired ions of diphenylamine and ethyl centralite and not just solvent peaks, MS/MS was done using different collision energies to maximize the process. Figures 32 and 33 show the MS/MS spectra of the smokeless powder from the bullet cartridge for each of the ions. Figures 34 and 35 show the MS/MS spectra of the smokeless powder from the blue dot alliant. Tong et. al. had previously done
tandem MS on both diphenylamine and ethyl centralite and confirmed that the separation is consistent [35].

Figure 32: MS/MS of smokeless powder for diphenylamine from bullet cartridge with DMS on, 20V, Rf=1000V, Vc= -2.56V with 40% collision energy
Figure 33: MS/MS of smokeless powder for ethyl centralite from bullet cartridge with DMS on, 20V, Rf=1000V, Vc= -1.41V with 30% collision energy

Figure 34: MS/MS of smokeless powder for ethyl centralite from blue dot alliant with DMS on, 20V, Rf=1000V, Vc= -1.9V with 30% collision energy
Figure 35: MS/MS of smokeless powder for diphenylamine from bullet cartridge with DMS on, 20V, Rf=1000V, Vc= -2.9V with 40% collision energy

4. Conclusion

The extraction of Salvinorin A from *Salvia divinorum* can be done from whole leaf pairs using different extraction solvents, but methanol being the best. The extraction time that gives the optimal amount of Salvinorin A takes approximately 30 minutes, with minimal stirring. The presence of Salvinorin A has the best reading when reconstituted with a mixture of 70:30:0.1 MeOH:H₂O:FA. The extracted Salvinorin A is able to be seen just as well as the commercial standard when analyzed using ESI-DMS-MS.
This experiment has demonstrated the use of ESI-DMS-MS for the analysis of Salvinorin A and smokeless powder. DMS has proven that it is able to obtain successful gas phase ion separation at atmospheric pressure when choosing the correct drift gas. Analyte separation can be obtained when the drift gas is changed or altered depending on what is trying to be separated [7, 40-43] and in this experiment it was determined that methylene chloride was the best drift gas to use.

With the collaboration of the DMS with mass spectrometry, we have demonstrated a rapid-quantitative analysis with good accuracy. DMS is still in the early stages of being tested and hopefully in the future, more experiments will be conducted in order to better understand its role in separation science. With more knowledge of DMS, it can be applied to more applications in science.

5. Future Direction

Additional research will have to be done in the future on ESI analysis of both Salvinorin A and smokeless powder. Since there are so many different species of salvia, additional extraction will have to be performed and analyzed. For the smokeless powder, other experiments have to be done in order to identify nitroglycerin in double-base powders and nitroguanidine in triple-base powders.

Analysis should also be done on burnt samples of salvia divinorum in order to determine whether the identification of Salvinorin A is possible to detect in residue. Residue from explosives should also be analyzed to see if it possible to differentiate components used in
powders. Finally, a calibration curve should be created in order to determine the limit of detection.

Smokeless powder has three different based powders. In the future, additional research will have to be done in order to differentiate between the different bases. In order to complete this, the detection of nitroglycerin and nitroguanidine will have to be identified.
6. References


