Electrospinning of Formic Acid/Acetic Acid and Nylon-6 Solutions for Wearable Hydration Sensors

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Electrospinning of Formic Acid/Acetic Acid and Nylon-6 Solutions for Wearable Hydration Sensors

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Executive Summary

Dehydration is not an uncommon experience, but it is one that is difficult to measure and one that can have unfortunate consequences. It is possible for hydration to be quantified by measuring the amount of salt and other ions present in a person’s sweat. RooSense is a company centered in Akron, Ohio with the goal of creating a wearable sensor that will be able to inform the wearer of their hydration level, helping to prevent cases of dehydration. The current iteration of the sensor is created through electrospinning a solution of nylon-6 dissolved in formic and acetic acid. Forty-eight solutions were created and electrospun into mats, with variations being made to the solution in the weight percent of nylon, the ratio of formic to acetic acid, the temperature of the solution while dissolving, and the time after dissolution in which the solution is spun. The intent of these variations was to determine the effects of varying these parameters on the final sensor response, coat weight, and thickness, and to be able to account for these changes with calibration curves.

The results for solutions showed that solution viscosity increases linearly with respect to nylon content with an R² of 0.96 for 25C, 0.94 for 35C, and 0.78 for 45C. Solution conductivity increases with respect to formic acid content in a linear fashion with R² values of 0.94 for 25C, 0.99 for 35C, and 0.98 for 45C. Tensile testing showed that there was a positive linear correlation between coat weight and the maximum force the sample could undergo. This correlation is less clear as temperatures increase, with R² values of 0.93 at 25C, 0.58 at 35C, and 0.48 at 45C. It does not, however, appear as though coat weight, the weight of polymer on the sensor per unit area, is affected by any one factor. ANOVA analysis shows that significant statistical difference between coat weight and thickness when formic acid content if varied, but it is recommended that more tests be done if greater predictability is desired. Sensor current
response data was generally good, with an average $R^2$ value of 0.80 and a standard deviation of 0.15. Finally, it does not appear that solution dissolution time had any significant impact on these quantities. When comparing data from the instant solutions and 24-hour solutions, trends were almost identical. For example, solution viscosity shows a linear correlation to Nylon-6 with a slope of 13979 and $R^2$ of 0.96 for instant solutions, and a slope of 17803 and $R^2$ of 0.87 for 24-hour solutions. ANOVA analysis shows a P-value of 0.896 for these correlations, meaning there is almost no statistical significance.

This work involved learning new laboratory procedures, experimental methods, and data analysis methods. During the process of learning these, greater technical aptitude has been achieved, along with the necessary technical knowledge to apply these methods to the correct situations. Additionally, this project has led to the development of greater soft skills as constant and effective communication between the authors of this paper, lab partners, and the leaders of RooSense was required. Additionally, this work will likely have great impacts in the field of wearable sensors and athletic technology. Being able to detect the salt content of an athlete’s sweat will allow for a greater amount of dehydration prevention and thus safety in training.

It is recommended is that further testing be done, with special care taken to ensure that coat weight is kept consistent, as later tests were more consistent in coat weight than earlier tests. If it is desired to identify trends in coat weight aside from the trends identified with the ANOVA tests, further experiments should be conducted. There are some trends identifiable, but it is likely that more data would clarify these trends into a more legible dataset. Additionally, for those sensors that have a low $R^2$ value for their sensor response testing, it is recommended that they be re-tested with the Potentiostat. If low $R^2$ values persist, it is likely that respinning these mats would be required to generate a usable calibration curve.
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Introduction

Dehydration is something that likely everyone has dealt with at one point or another. Dehydration often occurs when a person is exercising and is not intaking enough water to keep up with the amount of fluid that is being lost and thus the body does not have enough fluids to carry out its normal functions. As the person is losing fluids, they are also losing electrolytes such as sodium chloride. The number of electrolytes lost vary large amounts from person to person and depend on the type and intensity of the exercise (Baker, 2017). Some symptoms of dehydration include extreme thirst, less frequent urination, fatigue, dizziness, and confusion (Cheuvront 2014). When considering exercise, excessive sweating is the cause of dehydration as water is being lost in the sweat and the fluids are not being replaced. When exercising in hot, humid weather the amount fluid lost is increased. While simple dehydration can be easily fixed by intaking fluids, if not taken care of, dehydration can lead to heat injury such as cramps, exhaustion, heatstroke; urinary and kidney problems like urinary tract infections, kidney stones, and kidney failure; seizures due to the unbalanced electrolytes that occur in dehydration; and low blood volume shock, which is when low blood volume causes a drop in blood pressure and a drop in the amount of oxygen in the body which can be life threatening (Mayo Clinic Staff, 2019). Dehydration is something that should be avoided if possible as the complications can be drastic. Preventing dehydration appears easy enough, however a hot day or a long bout of exercise can make it difficult. Depending on the intensity of exercise, type, and climate the amount of sweat lost will vary and the amount of sodium in sweat can vary up to 100 times from person to person (Blow 2020, Bates 2008). Dehydration is often found once it has already occurred but if there was a way to track the amount salt leaving the body to correlate to the amount of fluid, dehydration would be more easily prevented. By monitoring the level of sweat leaving someone’s body in their sweat will allow them to know how much water to intake to offset the loss.

Background

Over the past several months, students have been working on the development of wearable hydration sensors at the startup company RooSense in downtown Akron, Ohio. RooSense is a company founded based on research from the Department of Chemical and Biomolecular Engineering at the University of Akron. The goal of the company is to develop a lightweight
wearable sensor that enables the user to know their hydration level, thus elevating their performance goals. To measure the hydration of the wearer, the sodium and other salt ions are collected from the user’s sweat as they exercise. This sweat is analyzed as it is collected to give the user an exact level of hydration, so they know how much fluids they need. The production of this ion sensor requires numerous steps and iteration upon the analysis of results. The process of the makeup of every sensor starts with electrospinning a carbon fiber mat in an enclosed chamber in a lab. Once the mats are electrospun, quality control tests are run including tensile testing to show the durability of the sensors and sensor response testing is completed to show how the sensor responds to exposure to sodium ions so that a calibration curve can be created for use in the final sensors. When an athlete wears the sensor the sodium ions in the sweat are measured which helps the athlete better understand their hydration level to improve their athletic performance and stay healthy. This project built on previous work done by students in the RooSense laboratory and quality control of the produced mats for the sensor response was the main goal.

**Methods**

To begin the electrospinning, the solution needs to be prepared which consists of formic acid, acetic acid, and nylon 6. As determined by the written procedure the specified amount of each of the three components are weighed out and the weights are recorded for consistency. A stir bar is added to the vial and the vial is placed in a hot water bath on a heated plate where the solution is stirred and heated to dissolve. In this process, several variations of solutions are made. The percent of nylon in the solution is changed, the ratio of formic acid to acetic acid is changed, the temperature of the hot water bath the vial is placed in, and whether the solution is stirred and heated just until dissolved or for 24 hours is changed. When the solution is stirred only until dissolved, the total dissolved solids, the conductivity, and the viscosity is measured four times throughout the dissolving, whereas those are only measured once at the end of the 24 hours of heating. Each of these alterations were analyzed to see what difference they have in the end product. Before electrospinning, the collection surface needs to be made by using aluminum foil in the middle and coated paper on either side connected with electrical tape. This surface is wrapped tightly around the drum and taped in place to collect the fibers produced during electrospinning. The picture below depicts the electrospinning set up.
Figure 1. The electrospinning set up used to make the nylon mats is shown above. On the left in the syringe pump and on the right is the needle and drum where the mat is spun. The right showcases the foil that the nylon solution is collected on and the drums that spins to evenly collect the material.

Once preparations are complete, plastic tubing with a needle on the outlet is connected to the machine, pointed toward the collection surface. A recorded amount in volume and weight of the solution prepared earlier is drawn in a syringe and connected to the other side of the tubing. The syringe is placed in the clamp of the pump and the pump is set to 5 mL/hr flow rate so that the pump will slowly and continuously feed solution through the needle to the drum. The needle has a voltage applied to it, which affects the size, shape, and speed of the jet of polymer, affecting how it disperses onto the drum. The desired coat weight, or polymer in weight per unit area, often measured in g/m², is predetermined, the density of the solution is known, thus these together are used to find how long the machine needs to be run. Every half an hour the humidity and temperature are recorded to account for any abnormalities in the data.

For tensile testing, sensors of approximate size 1.2 by 7 cm were created for the top, middle, and bottom of each mat. The dimensions, thickness and coat weight were recorded for each and approximately 1 cm was marked on either end of the sensor to show where the clamps will go. On the machine, the grip clamps are separated at a distance of approximately 2.3 inches, using
the control in the software to change the distance. The sample is clamped in the grips, so the sample is loose but taut. In the software, the test type is set to “Tensile Stress at Break and Elongation Rate” and the test name, specimen length, specimen width, and specimen thickness are each inputted into the parameters. The sample is ready for testing, the start button is pressed, the upper clamp raises, stretching the sample until the sensor breaks. This data is saved, and the process is repeated for all samples. For each sample, the maximum force value in newtons, the displacement in millimeters (mm), the strain in percent, and the elastic modulus in kilogram force per square centimeter (kgf/cm²) are recorded.

In general, most solution preparation and all of the tensile testing was conducted by Nichole Cooper. Sensor response testing was conducted by Jonathan Doak. Electrospinning was a time-consuming but not attention-requiring procedure, and as such was conducted fairly evenly by whoever was in the lab at the moment.

**Results and Discussion**

Various comparisons were made to find correlations and relationships components of the mats. Throughout the data analysis, evidence showed that the coat weight is likely affected by more than one variable. Each of these variables were independently graphed and graphed with multiple variables to showcase the effect on the coat weight of the mat. The results of the analysis and the variables compared are examined below. At each temperature, the coat weight versus the weight percentage of nylon in the sample was plotted and can each be seen below in Figures 2-4.
**Figure 2.** The above graph shows coat weight of the sensor versus the percentage of nylon in the sensor at 25°C for varying percentages of formic acid. There is a slight positive trend with the nylon and coat weight at 50% and 70% formic acid, a slight negative trend for 60% formic acid.

![Graph 35°C](image)

**Figure 3.** The above graph shows coat weight of the sensor versus the percentage of nylon in the sensor at 35°C for varying percentages of formic acid. At 35°C, for 50% and 60% formic acid there is a slight positive trend and a negative trend for 70%.

![Graph 45°C](image)

**Figure 4.** The above graph shows coat weight of the sensor versus the percentage of nylon in the sensor at 45°C for varying percentages of formic acid. A slight negative trend is seen for
samples with 50% formic acid, and a more prevalent negative trend is found for 60% and 70% formic acid.

To further examine the potential correlation of nylon weight percent and coat weight, a regression analysis was completed for these variables at the three temperatures. The following three figures, Figures 5-7 show the results of the regression analyses.

**Figure 5.** The regression analysis for the correlation of nylon percent and the coat weight at 25°C. The R square is 0.0178 which corresponds to virtually no correlation.
Figure 6. The regression analysis for the correlation of nylon percent and the coat weight at 35°C. The R square is 0.0071 which corresponds to virtually no correlation, even less than at 25°C.

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Figure 7. The regression analysis for the correlation of nylon percent and the coat weight at 45°C. The R square is 0.1055 which corresponds to virtually no correlation, slightly more than 25°C and 35°C.

Figure 8. These tables show the ANOVA analysis for thickness and coat weight of the mats with respect to formic acid content: notably, the 60/40 FA/AA and 70/30 solutions. Very small P-values in the ‘columns’ section of each analysis means the null hypothesis can be rejected. From this the conclusion can be drawn that formic acid content can have a significant impact on the coat weight of the sample.

After analyzing data and looking at the trends in the previous three graphs, the conclusion that there is no direct correlation between the percentage of nylon in the solution and the coat weight of the produced mat was drawn. In the regression analysis, the R square value was
looked at to see the amount of correlation in the two variables, with closer to 1 being good correlation and closer to zero is very little correlation. At each of the temperatures, the nylon weight percent and coat weight have a R square of 0.1 or less, thus there is little to no correlation in the variables. In Figure 3, there are three outliers from the same mat of 17% nylon and 50% formic acid, thus this mat should be recreated and tested to see where the data actually lies. This information, combined with the ANOVA tests and trendlines suggesting formic acid percent contributes to coat weight, has led us to the conclusion that more tests should be done to predict coat weight with greater accuracy.

**Figure 9.** The graph above shows the correlation between the percentage of nylon in the solution and the viscosity of the solution. As the amount of nylon increases, the viscosity of the solution increases, as expected due to nylon’s high viscosity. This trend holds true for all temperatures, although at 45°C there are significantly higher deviations.
**Figure 10.** Regression analysis of the correlation between nylon percent and viscosity of the electrospinning solution for solutions made at 25°C. The R square value is 0.955 which shows that there is almost perfect correlation between the nylon percent and solution viscosity.

**Figure 11.** Regression analysis of the correlation between nylon percent and viscosity of the electrospinning solution for solutions made at 35°C. The R square value is 0.818 which shows significant correlation between the nylon weight percent and solution viscosity.
Regression analysis of the correlation between nylon percent and viscosity of the electrospinning solution for solutions made at 45°C. The R square value is 0.503 which shows that there is some correlation between the two variables, however not drastically significant. From the visualizations, there is a direct response from the percentage of nylon to the viscosity of the solution. This is a strong trend that matches the expected response. Since nylon has a higher viscosity than the other two components, the overall viscosity of the solution will increase when more nylon is added (Berry 2006). The regression analysis for each temperature supports the trend from the graphs. The analysis also shows that as the temperature increases, the correlation between nylon percent and viscosity decreases. This could be due the decrease in viscosity of nylon with the increase in temperature. No correlation between the nylon percentage and the conductivity of the solution was found when graphed. The same data was plotted for the connection between the percent of formic acid and the viscosity and conductivity. There is no correlation between the percentage of formic acid and the viscosity, and this is due to the similar viscosities of formic acid and acetic acid. There was a slight trend with the formic acid and conductivity which can be seen in Figure 13 below.

**Figure 12.** Regression analysis of the correlation between nylon percent and viscosity of the electrospinning solution for solutions made at 45°C. The R square value is 0.503 which shows that there is some correlation between the two variables, however not drastically significant.

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Figure 13. The graph shows the conductivity of the solution versus the percentage of formic acid added. While standard deviations are large, there is a trend of increasing conductivity when the percentage of formic acid is increased.

Increasing the amount of formic acid and thus decreasing the amount of acetic acid increases the conductivity on the solution because formic acid is a stronger acid (Chemistry Community, 2011). The stronger the acid the more conductive the acid is (Stubbings). In acetic acid, the methyl group is electron donating towards the O-H bond which in turns makes the hydrogen harder to remove. Formic acid does not have an electron donating group, thus making formic acid the stronger acid and more conductive (Chemistry Community, 2011). There is a trend between the formic acid and conductivity, however with the significant number of outliers, more testing should be done to find better trends.

As previously mentioned, the solution was created in duplicate with one being spun on the heater just until dissolved and the other being spun for 24 hours. This was done to see what differences if any would be found due to temperature dependence. The resulting differences in the viscosity of the instant and 24-hour solutions are found in Figure 14 below.
Figure 14. The comparison of the solution viscosity and the nylon percentage for some instant and 24-hour solutions is shown. The trends for the two times are extremely similar and there is no significant difference.

The lack of difference above shows that the solution can be created and allowed to spin for longer than just the dissolving time. This allows solutions to be prepared ahead of time. More experiments should be conducted to see if the solution changes if allowed to sit before spinning, if spun less than 24 hours but more than dissolving time, and more than 24 hours.
Figures 15-16. These figures show the correlation between conductivity and viscosity plotted with respect to Nylon-6 percent and formic acid percent. Conductivity increases with formic acid percentage but does not seem to have an identifiable trend with respect to nylon composition. Conversely, viscosity increases as nylon-6 percentage increases, and does not appear to change with formic acid percent. As previously stated, this is the expected outcome due to nylon’s high viscosity. Note that these values are averages across temperatures and does not account for the variations that occur with temperatures noted in Figures 13-14.
Figure 17. This figure shows the average coat thickness of the mats that were electrospun. The mats have very large standard deviations when taking all of them into account, along with minimum and maximum values of 5.2 and 43.5 g/m², respectively. However, when looking into only the last third of mats that were produced, the standard deviations become much narrower, with minimum and maximums of 10.2 and 17.9 g/m². The narrowing in standard deviation is believed to be due to a combination of the methodology becoming more refined as tests were conducted, and the operators becoming more familiar with the equipment.

Enough sensors were made from each mat to do several testing experiments. Tensile testing was completed for the top, middle, and bottom of each mat and the max force and elastic modulus was found. Several variables were compared and little to no trend was found between max force and formic acid percentage, the elastic modulus and formic acid percentage, and the elastic modulus and coat weight. The big correlation that was found from tensile testing was the connection between coat weight and the max force. A graph displaying the results can be found below in Figure 18.
Figure 18. The correlation between the coat weight of the mat sample and the max force value in newtons is shown for every sample.

From the graph a relatively strong positive correlation is found between coat weight and the maximum force value in newtons. This is logical as there is more material that would need to be broken, thus more force would be needed. The trend is not ideal, showing that more testing should be done to prove the connection. The R² value for the trendline decreases as the temperature increases, showing more variability at higher mix temperatures. For the most consistent max force, the solution should be mixed at 25°C, however more experiments should be done as well.

Sensor response testing was conducted on another sample of the top, middle, and bottom of each mat. These sensors were 1.2 by 1.2 cm in size. The sensors were prepared by placing them in a solution of carbon nanotubes and Triton X-100 and sonicated for 45 minutes. Once sonicated, the sensors were removed from solution and allowed to dry for at least a day in an aluminum pan covered with parafilm. Once the sensor was dry, the sensor response testing began. The sensor was washed with DI water and placed in a premeasured solution of salt water. These solutions had concentrations of 10, 20, 30, 40, 50, 80, and 100 mM. The sensor was allowed to soak for two minutes and then removed from the solution and laid on a glass slide. Two alligator clips were attached to the sensor to hold it onto the slide, taking special
care not to let the clips touch. Upon starting the test run, the Potentiostat runs a voltage through the clips and therefore the sensor, and the resistance for that sensor at the given concentration was found. The resistance was calculated by the constant voltage divided by the measured current for each sensor. Each mat’s top, middle, and bottom sensors were tested for each of the 7 concentrations. A calibration curve was generated from a generated coefficient and exponent based on the equation:

\[ \text{Response} = \text{Coefficient} \times \text{Concentration (mmol)}^{\text{Exponent}} \]

The R² value was found for each sensor response based on how well its experimental results match this theoretical response. In general, the fits were good, as the average R² value was 0.8, with a standard deviation of 0.15. There were, however, some exceptions.
Figures 19-21. These figures show the averaged responses of the top, middle, and bottom sensors of each mat. As expected, the resistance of the sensors shows exponential decay with respect to the sodium concentration. Notably, the samples prepared at 25°C show the most deviation from the theoretical results.
Figures 22-24. These figures show the sensor responses for a sensor with a good response, $R^2 = 0.996$, compared to a sensor with a poor response, $R^2 = 0.372$. 49 of 133 sensors that were measured had an $R^2$ value of greater than 0.90, and so would resemble sensor 33’s response. Only 15 sensors had an $R^2$ value of less than 0.50.
Figures 25-30. These figures show the same data as Figures 17-19 but separated to show the
solutions prepared instantly against the solutions that were allowed 24 hours before electrospinning.

**Figure 31.** This figure shows the ANOVA analysis conducted for the 60/40 FA/AA solutions against the 70/30 solutions with respect to the coefficients and exponents of their calibration curves. \( F \) being greater than \( F_{\text{crit}} \) for the 24-hour solutions but not for instant solutions suggests the preparation method does affect experimental results. As \( F \) is not greater than \( F_{\text{crit}} \) for any other cases, the null hypothesis cannot be rejected, and results are inconclusive.
**Conclusion**

In conclusion, mats were created through the electrospinning of solutions of nylon 6, dissolved in formic acid and acetic acid. The measurement of the variables of these solutions produced some expected results, such as the solution viscosity increasing as more nylon-6 is added. However, there were other trends in the data whose source cannot be identified. For instance, coat weight of the mat once spun has a large standard deviation of about a third of the average values, and extremely volatile maximums and minimums. It does however appear that the mats spun more recently are more consistent in their results. Taking only the final 16 of 48 mats into account, the standard deviations become about 12% of average values. It is also important to note that the coat weight corresponds directly to the maximum force and tensile strength of the resulting sample. For these reasons, it is recommended going forward that more tests be conducted, or possibly redoing some of the early tests, while paying special attention to keeping the electrospinning consistent. Sensor response was tested by measuring their resistance after being soaked in varying salt solutions to emulate human sweat. These results were fairly consistent, with the theoretical response having an $R^2$ value of 0.75 or greater for 77% of the samples taken. As with coat weight, there does not seem to be an identifiable trend in the sensors that have a poor response. Retesting of these sensors is therefore recommended, with the possibility of re-spinning those mats if the sensors still do not perform as expected. Going forward, the mats or samples that were identified as outliers should be recreated and tested again. Once the outliers have been retested and satisfactory results are obtained, methods for adapting the sensors into a wearable design should be researched and tested. Ideally this method should result in a sensor with a repeatable and consistent response to similar concentrations of sweat.
Works Cited


