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Dermal Reduction of Urushiols Using an Activated Charcoal Formulated Dermal Care Patch

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Honors Thesis

Dermal Reduction of Urushiols Using An Activated Charcoal Formulated Dermal Care Patch

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Abstract

For centuries, activated Charcoal (AC) has been used externally in a poultice form to adsorb "poisons" trapped in the outer layers of skin and internally to relieve intestinal discomfort and to remove toxic materials. The largest use for activated charcoal, in our society, is as a filter bed in air and water remediation cartridges. Recently, scientists have formulated AC into a nonstick dermal bandage called the Charcoal Patch (CP), but the adsorption properties are not well understood for this new formulation. Experiments have been conducted to see if these dermal bandages can be used to adsorb oils such as poison ivy and other toxic oils. Since poison ivy is an extreme dermal irritant, experiments were performed with a surrogate compound, 3- Pentadecylphenol (3-PDP), which has a similar chemical structure to urushiol, the active irritant in poison ivy.

The main goal of this research is to reduce the amount of urushiol that is absorbed by the skin by using an activated charcoal dermal patch. This particular project's objective is to determine the characteristics and quantity of materials that can be adsorbed and contained in a dermal patch. Experiments were conducted by cutting out square pieces of the charcoal patch and measuring the mass gained by various substances over varying amounts of time using a precision analytical balance, capable of recording mass changes down to 100 µg. The amount of substance being adsorbed in grams per gram of CP is calculated and compared among different analytes. This is the first stage in determining the effective adsorbing ability for nonpolar analytes using charcoal that is integrated into a non-messy, non-stick dermal bandage.

Introduction

Activated Charcoal (AC) has been used for centuries in a poultice form to adsorb "poisons" trapped in the outer layers of skin. Activated charcoal is the highly porous form of carbon, which imparts a surface area within these particles to nearly 500 $m²$ per gram of charcoal¹. Its hydrophobicity, high surface area, and low cost have made it an effective choice absorbing hydrophobic materials from water and air. It can also be taken internally to adsorb toxins that may have been ingested. Activated Charcoal is rated "Safe and Effective" (Category I) by the FDA for acute toxic poisoning². Activated Charcoal has also been used to aid in drawing out toxins in overdose patients and is recommended by the Poison Control Center². It is listed in the U.S. Pharmacopoeia and is recognized as the universal antidote². Activated Charcoal does adsorb most organic and inorganic chemicals, and does not adsorb beneficial nutrients.

Past experiments conducted have shown activated charcoal to be harmless on the skin and to cause no ill- side effects². Some experiments that were performed involved mixing activated charcoal with Cobra venom and injecting it into to a laboratory animal². The animal was not harmed showing the charcoal did adsorb the toxin and essentially inactivated the toxin. Other experiments involved mixing the activated charcoal with arsenic and strychnine to be ingested by humans under laboratory conditions². Subjects had survived despite the dosage being 5 to 10 times lethal. There have also been experiments that tested the adsorptive properties of activated charcoal showing that it can adsorb phenolic compounds under different isothermic condtitions³.

AC has also been shown to adsorb highly toxic oxyanions, but through modification with a cationic surfactant⁴. It is an adsorbent capable of binding other substances in a relatively large amount onto their surface. This property is often used in pharmacy as well as in the studies of the structure, biological activity relationships, where active charcoal serves as a model substance for the study of hydrophobic interactions. The adsorptive properties of AC have been investigated involving basic esters of phenylcarbamic acid with a local anesthetic effect⁵.

Activated charcoal is made from burning carbon rich sources such as woods, shells, and husks to name a few natural sources. There have been studies done in which activated charcoal was prepared from Aleppo Pistacia Vera shells using different percentages of zinc chloride at temperatures of 873 Kelvin in the absence of $air⁶$. The different percentages of zinc chloride were found to affect the pore size of the activated charcoal created. Using gases such as steam or air at high temperatures, to oxidize insoluble carbonized wood, can also create activated charcoal⁶. Dried coconut shells without meat, can also be used to create activated charcoal. These shells would need to burn at temperatures of about 600-900 degrees Fahrenheit, and then combined with 25 % concentrated solution of calcium chloride or zinc chloride for 24 hours⁶.

AC can be made from macadamia nut shells and coconut shells, by being heated at high temperatures to be carbonized then using oxygen in air oxidizes it⁷. The removal of carbon mass by the development of pores is responsible for the increases in surface area of the activated charcoal. The mass of carbon removal is related to the pore size. Due to these properties of AC, we want to study if this recently made charcoal patch would exhibit the same characteristics of

AC and if it could be used in the treatment of poison ivy exposure. We want to see if this charcoal patch would be effective in the dermal reduction of urushiols from skin.

Experiments were conducted to determine if these dermal bandages could adsorb oils such as poison ivy and other toxic oils. Since poison ivy is an extreme dermal irritant, experiments were performed with a surrogate compound, 3-Pentadecylphenol (3-PDP), which is similar in structure and hydrophobicity to urushiol, the active irritant in poison ivy. Similar urushiols can also be found in poison oak and sumac.

Experimental Section

Charcoal patches (CP) and charcoal patch formulation (CP-formulation) were obtained from Neobiotech (Berrien Springs, MI). The charcoal patches were manufactured using a heated transfer of the CP-formulation onto a cotton fabric backing then covered by wax paper. The charcoal patch formulation was processed using activated charcoal and a proprietary set of additives and then blended into a homogenous mixture. This formulation can be placed on top of skin but not leave any residue when removed. The charcoal patch (CP) is the completed product comprised of CP-formulation and woven backing made of cotton.

Materials needed for experiments performed include CP squares, tape, various substances and oils (listed below), glass plates, rulers, test tubes, Bunsen burner, Pasteur pipets, Erlenmeyer flasks (25 mL) and hot plates. Mass measurements were made on a Mettler Toledo (model AF-166) analytical balance that measures to 0.1 mg. Mineral, vegetable, walnut, peanut, and fish oil were used as surrogate oils for poison ivy. These were purchased from a local food market and used without further modification. ACS reagent grade acetone, ethanol, 2-propanol, and heptanes were obtained from Sigma Aldrich and used as is. Activated charcoal powder (AC) and 3-Pentadecylphenol (3-PDP) were also obtained from Sigma Aldrich and used without modification.

Methods

Procedure 1:

The first experiment was performed to determine the properties and characteristics of the AC relative to CP-formulation. This experiment was then repeated using activated charcoal powder, so that the properties of the AC could be compared with that of the CP formulation.

- 1. Obtain four test tubes, in two put a few milligrams of AC and in another two put a few milligrams of CP-formulation
- 2. In a each test tube containing AC and CP formulation add water.
- 3. In the other test tubes containing AC and CP-formulation add water + mineral oil.
- 4. Cover the test tubes with parafilm and leave for 24 hrs.

Procedure 2:

The next experiment involved using substances such as distilled water, heptane, mineral oil, acetone, 3-PDP, ethanol, and 2-propanol. This experiment was done in order to determine the adsorption properties these various substances in a short time frame to quickly screen materials. The outlined procedure is listed below. The charcoal patch was taped down to the plate glass to prevent substances from being absorbed by the cotton backing as well as to give an exposed surface area in which adsorption Is taking place. This ensures that the actual adsorption of the charcoal patch formulation is being observed. This procedure is similar to those of procedure 3, 5, and 6. It differs in that it is a more generalized procedure and the amount of substance that was added to be adsorbed was not measured; only the results were recorded. Procedure 3 is more specific and the amount of substance added has been recorded, the time intervals are longer, and the substances used have been narrowed down to specific oils. In procedure 5, a step is added in which the charcoal patch is pre-treated with water and soap to see if it will aid in the adsorption of oils. In procedure 6, a slygard, silicone gel is used instead of the tape. This was to test if previous results were accurate, and that charcoal patch formulation fibers were not being taken up with the tape.

- 1. Cut out about eight squares of charcoal patch squares about 3 cm by 3 cm.
- 2. Weigh and record the initial weight of the square.
- 3. Tapes the square to a plate glass with the charcoal portion faced upward, see Figure 2, and record the weight of the combination.
- 4. Place a drop of distilled water on another glass plate, then place the charcoal patch that is attached to the plate glass on top of the drop. Leave in contact for about 30 s.
- 5. Pick up the plate and dab the charcoal patch dry (If there is any excess liquid, make sure not to press too hard).
- 6. Weigh and record results.
- 7. Repeat steps 4-6 increasing the time intervals by 4 min.
- 8. Repeat steps 2-7 using the other substances listed above.

Figure 2: Glass slide with CP taped down

Procedure 3:

The experimental procedure more accurately determines the amount of substance in grams was being adsorbed per grams of the charcoal patch over a given time period. Based on the results from the previous experiment, specific substances can be used to study the adsorption of the patch. The main substances used included mineral oil and distilled water. Most of the experiments conducted used this basic procedure with few changes. Some changes involved

running experiments for 1 hr, 4 hrs, & 24 hrs. Other changes involved using different oils such as peanut oil, walnut oil, and fish oil. Experiments were also done with dish soap.

- 1. Cut out nine pieces of charcoal patch squares.
- 2. Weigh three charcoal patch squares and record.
- 3. Tape three individual squares to three individual glass plates.
- 4. Measure and record the exposed area of the charcoal patch squares.
- 5. Place two drops, approximately 1.0 g of distilled water onto each exposed area of the charcoal patch and place a glass plate over it.
- 6. After an hour, remove the glass plate (make sure to wipe off any excess water), and remove the tape from the patch.
- 7. Weigh and record the resulting charcoal patch.
- 8. Repeat steps 2-7 using mineral oil and vegetable oil.

Procedure 4:

This experiment was performed to determine how much oil could be adsorbed by activated charcoal powder. This experiment was also repeated with water to determine if water could be adsorbed by activated charcoal powder.

- 1. Put one piece of tape adhesive side up and tape down to a glass plate using two other pieces of tape adhesive side down. Figure 3.
- 2. Put two other pieces perpendicular to the other strips of tape to make an adhesive square in the middle.
- 3. Weigh and record the weight of the plate glass and tape.
- 4. Add charcoal powder and smooth down with lintless tissue wipe until there is a thin film of charcoal. Note: Try to make sure there isn't much charcoal residue coming off the tape.
- 5. Weigh and Record the plate glass, tape, and charcoal.
- 6. Put two drops/approximately 1.0 g of mineral oil and wait for 2 min.
- 7. Wipe off excess oil, trying not to take up any charcoal powder.
- 8. Weight and record the results.
- 9. Repeat procedures 1-8, three times.

Procedure 5:

This experiment was performed to determine if adding water to the charcoal patch square will aid in its adsorption to oils. When results showed that water did not aid in oil adsorption,

Figure 3: To the left, plate glass with tape, adhesive side up. To the right, plate glass with tape and charcoal, on adhesive side up.

this experiment was conducted again using dish soap instead of water. Results showed that dish soap did not aid in the charcoal patch adsorption of oils as well.

- 1. Cut out four pieces of charcoal patch squares.
- 2. Weigh and record each square.
- 3. Tape down each square lightly and to individual glass plates measure and record the exposed area.
- 4. Add two drops/approximately 1.0 g of distilled water.
- 5. Remove the tape from the charcoal patch square, then weigh and record the patch plus distilled water.
- 6. Following the indention of the tape lines, tape back the charcoal patch square to the glass plate.
- 7. Add about two drops/ approximately 1.0 g of mineral oil and place a glass plate over it.
- 8. After an hour, weigh and record the resulting charcoal patch. Note: Make sure procedure is done with each charcoal patch square individually.

Procedure 6:

This experiment was performed to see if previous experiments were accurate, when determining the adsorption of the patch. Experiments were performed by taping down the sides of the charcoal patch squares and it was unknown if fibers of the patch were being caught on the tape. It was decided to perform procedure 2, using a slygard 184, silicone elastomer, which is a gel used instead of tape to make a well to prevent the oil from getting into the backing of the CP.

- 1. Cut out four charcoal patch squares, weigh and record.
- 2. Cut out four sylgard gel squares, and cut out a second square in the middle and measure and record the area. Figure 4.
- 3. Place one of the squares on a charcoal patch and add two drops/ approximately 1.0 g of Peanut Oil.
- 4. Place the smaller square on top of the oil and place a glass plate on top of it.
- 5. Leave for an hour.
- 6. Peel back the gel and record the resulting patch.
- 7. Repeat steps 3-6, with three other patches.

Side by side comparison of AC and CP-formulation ability to absorb water and mineral oil.

Utilizing Procedure 1, the CP-formulation and AC were evaluated in a side-by-side comparison. It is generally accepted that activated charcoal powder tends to adsorb oils, i.e.

Figure 4: Plate glass with a charcoal patch with a slygard, silicone gel, on top.

hydrophobic compounds, and does not adsorb water or hydrophilic compounds, ions, or metals. This first procedure was implemented to verify this behavior and to establish a correlation between AC and CP-formulation through a simple, side-by-side test tube comparison. This experiment compared the characteristics of AC to that of the CP formulation.

Each material was placed in deionized water or a deionized water-mineral oil mixture. The charcoal based substances were added and some manual mixing (swirling or spatula) was

utilized to mix the materials with both the water and oil layers, if present.

AC was first evaluated to visualize the standard behavior of this material in water. AC in water, see Figure 5-Part A, was suspended throughout the water with manual mixing. After a few minutes, a portion of the AC settled to the bottom of the tube, while other portions rose to the top and rest on top of the water. Some material clung to the sides of the tubes. AC in the water and mineral oil mixture was thoroughly mixed and allowed to settle over minutes and hours to observe its behavior in this mixed solvent system. About half of the AC was suspended or dissolved in the mineral oil layer on top, see Figure 5-Part B, while the rest of the AC fell to the bottom of the tube as it did with water alone.

Figure 5: AC and CP-Formulation compared in test tubes containing water $(A \& D)$ or in a water-mineral oil mixture (B&C). Test tubes A and B contain AC while test tubes C & D contain CP-formulation.

CP-formulation was evaluated in test tubes in a very similar manner utilizing approximately the same about of material. Figure 5-Part C shows the results after the CP-formulation has been in contact with water for many hours. It appears to have absorbed all the water. It has expanded to the size of the volume of water initially in the test tube. There was about 10 times the mass of water relative to the mass of the CP-formulation. This material appears to absorb water very well. In the water and mineral oil mixture, Figure 5-Part D, the oil formed a layer on top of the water layer. Upon addition of the CP-formulation, it settled to the bottom of the test tube and over a period of hours. During this time, the CP-formulation absorbed all the water with the same characteristics listed above. The mineral oil layer was resting on top, undisturbed.

These results utilizes procedure 4, in which an experiment was conducted with AC to compare its characteristics to that of the CP. (Refer to Figure 3) Results showed that activated charcoal does not adsorb water and do adsorb oils. Four trials were run and it can be seen that the AC powder can adsorb about 5 g/g of oil.

Quantitative Evaluation of CP Ability to Absorb Various Materials.

In the next set of experiments, the goal was to evaluate the various type of substances that could be adsorbed by the CP as well as determine how much grams of substance per grams of CP-formulation could be adsorbed. In Figure 6, one can see the amount of adsorption in grams of substance per grams of CP plotted against time. It can be seen that there was a direct relationship between the adsorbance in g/g as time progressed.

adsorption in grams of substance per grams of CP plotted against time in seconds. The time was taken in intervals of 5 min from 30 s to 840 s.

Results showed that water adsorbed very well, while other substances adsorbed poorly. The acetone, heptane, 2-propanol, vegetable oil and ethanol were barely adsorbed, while the mineral oil did adsorb to some degree. Once the acetone, heptane, 2-propanol, and ethanol were placed on the CP, the solution would spread out and start evaporating right away. With the CP background also changing, losing weight, it was difficult for an accurate reading to take place. When the oils were added they would rest on top of the patch and there was no change in appearance seen. When the water was added it would remain as a droplet on the CP and it can be seen that it was adsorbing right away. The volatile substances would evaporate too fast for an adsorption reading to take place, so the focus was more on nonpolar oils that do not evaporate at room temperature. There was a weight gain with the 3-PDP and although it was not a lot the patch did adsorb enough of the substance.

Evaluating the Adsorption Capacity of the CP Using Various Substances.

During these set of experiments an adsorption screening was set up To determine what will be the optimum time to record adsorption, researchers set up a screening technique, in which the adsorption of vegetable oil as well as distilled water were monitored for 1 hr, 4 hrs, and 24 hrs. (Refer to Figure 7) After an hr, the patch begins to lose weight and some evaporation occurs (in the case of water). At an hour there was a minimum change in the CP weight and the maximum adsorption took place that could be recorded.

Our goal was to determine the optimal time to record mass changes, thus time versus adsorption changes were studied. At an hour there was a minimum change in the background CP mass and the maximum adsorption of analyte. We noted that CP patches changed mass in time periods greater than one hour depending on the relative humidity of the testing environment. This graph shows our justification for selecting a 60-minute adsorption time period.

The Fatty Acid Composition of Oils.⁸

These results utilize procedure 3, 4, and 6. This next set of results refers mainly to Figure 8, which shows the CP adsorption capacity using various substances as well as the urushiol surrogate, 3-PDP. The CP had the greatest capacity to absorb water. Distilled water is the best substance to be adsorbed by the CP due to its hydrophilic tendencies. Results from experiments showed that the CP adsorbed more hydrophilic molecules as opposed to hydrophobic molecules. Dish soap was the next best substance to be adsorbed. This was due to its hydrophobic and hydrophilic properties. Peanut oil was the best oil to be adsorbed, although the amount was few. The fatty acid percent composition in the oils played a role in its adsorption capacity by the CP (Refer to Table 1 Above). It was found that oils containing a higher composition of a single fatty acid were more likely to be adsorbed than the oils with a wider variety of fatty acids. The poison ivy surrogate 3-PDP did adsorb although it was in limited amounts. Enough substance was adsorbed for our project to be considered successful. It adsorbed around 500 µg of substance and it takes about 50 µg of urushiol to cause an irritation.

To determine techniques that would aid in CP adsorption the CP was pre-treated with distilled water and dish soap. Results showed that water and soap did not aid in adsorption, instead no adsorption took place when oil was added. (Refer to Figure 8) When the oil was added it looked as though the droplet just rested on top of the water and the dish soap.

Since experiments were done with taping down the charcoal patch, researches wanted to see if fibers were being taken up on the tape. This would make sure that the results from previous experiments were accurate. The experimental procedure was performed using sylgard 184, silicone elastomer gel instead of tape. (Refer to Figure 4) There was no significant difference. (Refer to Figure 8)

Evaluating the Characteristics of CP

After viewing the results of some experiments performed it was noticed that there seemed to be a decrease in the weight of the patch as time progressed. This led to a procedure in which the weight of 1.0 g of mineral oil and a 0.2771 g piece of a charcoal patch square was monitored over 2 days by periods of time. Results showed that the mineral oil drop remained constant and lost 0.0003 g within the second day. The charcoal patch was seen to lose 0.0019 g of weight only 30 min later. This led to the conclusion that the charcoal patch was losing weight as time progressed.

Conclusion

The experiments conducted in this study allow us to confirm that AC powder does adsorb oils well, but the CP formulation preferentially adsorbs hydrophilic materials with some limited hydrophobic molecule adsorption. Untreated AC does not adsorb water and has an effective capacity of 5 g/g of oil/AC. This is the benchmark oil adsorption for AC. AC in the CP formulation has a much lower affinity for oils. We measured oil adsorption capacities around 0.01 g/g for a variety of oils. Oils containing a higher composition of a single fatty acid were more likely to be adsorbed than the oils with a wider variety of fatty acids. Peanut oil is the best oil to be adsorbed, although the amount is low. Our target molecule, 3-PDP did appear to have a low capacity to adsorb into the CP, around 0.05 g/g. However, this may be enough adsorption to remove urushiols from skin. Since the CP formulation exhibits the opposite adsorption characteristics relative to AC, the other formulation ingredients within the CP are reducing the oil adsorbing capacity of the AC within the patch. The limited 3-PDP adsorption was evidence that enough oil-adsorbing properties remain and this could be sufficient to reduce dermal concentration of urushiols on skin. These dermal patches greatly reduce the mess and hassle of AC poultices, but further modification of the formulation is suggested to increase the oil adsorbing capacity of these CP.

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