A guide to the acid-base extraction of Peruvian incense (by Litmus)

Version 1.0: This easy-to-follow guide will take you from your raw starting material (dry chips of a blue-green cactus species from Matucana, Peru) all the way up to glistening white needles of olfactory essence. Credits go out to people like foaf, me!, ion, fmoc and others for their pioneering work. The main improvements in this technique are the accurate titration and the low-loss purification steps. Following this method, it is possible to obtain 4,4g of essence-sulphate from 340g of dry cactus-flour. Probably, yields could even get a little better by implementing some possible improvements I thought of while performing this procedure for the first time. They are put in italics. If they prove to be beneficial, they will be integrated into the following protocol. I advise you to read through this manual and ‘rehearse’ the extraction, so you can acquire or improvise all the needed materials and arrange the process to fit your specific situation. Good luck!

1) Obtain 1kg of dried cactus-chips and reduce them to dust in a coffee-blender. Mix the resulting powder by shaking it inside a Ziploc bag. Now you will have a homogenous ‘cactus-flour’ that holds the same amount of cactus-essence in every gram. Next, you weigh 1/3 of this material and go to the next step. The reason for making 3 times the needed amount of cactus-flour is that after finishing the total extraction of the first batch, you will have accurate estimates of the amounts of reagents to be used for the following two batches. This is optional however, since this technique proves to be already accurate enough from the first batch.

2) In a 3l recipient, dissolve 300g of sodium hydroxide in 1900ml of demineralised water (warning: very caustic! From now on, always use safety-goggles and gloves during the manipulation of the reagents). The solution will tend to generate heat, so add the NaOH a little at a time. Normally, to convert all the essence to its free-base and then bring the pH of this solution to 13, it would only require 5-15g of NaOH. However, one uses the capacity of a very strong NaOH to break down the organic material and free all the essence. What’s more, these high NaOH-concentrations do not seem to desintegrate the essence which has got a rather high pKa.

3) Integrate the cactus-flour into this solution. The mixture will turn very slimy and it becomes very difficult to get it homogenous. Eventually, I put a mixer/blender into the mixture (careful for splattering!!). Let this mixture stand for at least 1 hour, this will prevent the xylene to wet any dry material and thus make it unextractable. Possible improvement: first put the flour into a small Ziploc bag and add a small quantity of demineralised water. Knead the water into the flour and then add it to the NaOH solution.

4) Now pour 500ml of xylene on top of this suspension and close the recipient. Make sure you have got a tight grip on it, and shake the mixture violently until you no longer see any separate layers or drops. I use a 3l canning jar for this. However, the rubber band cannot
withstand the xylene, so I lock a special solvent-glove underneath the lid when closing. Be sure to test the resistance of the glove beforehand because some ‘special’ gloves will nonetheless deform and get weak. Let the emulsion stand for 24 hours.

5) If the layers separated well (they never did with me), proceed to the next step. If not, put the jar into a large pot and fill with water. Put on a stove on moderate heat. Now you will start to see air bubbles and xylene-streams rising at the sides of the jar. Heat the jar for about an hour (but do not bring to a boil!) and let the jar cool down in the pot over the next 12 hours. This will bring about a complete separation.

6) With the help of a pipette, move 275ml of clear xylene from the top layer into a tall 75cl recipient. Next, in another smaller recipient, put another 75-125ml of xylene from the extraction jar (take care not to introduce organic material from the bottom layer). You must know how to work with a pipette. Considering the chemicals we are working with, it is imperative to use a rubber ‘pear’ with arrows for filling and emptying the pipette. As you will see, the manipulation of the pipette is essential for working accurately.

7) Prepare a plate on which you place little pieces of blue litmus-paper (Ah, this is new!). You will need no more than 10. You will put a drop of liquid on them consecutively, so make sure there is enough space between them. Also, prepare a solution of 2ml of sulphuric acid (96%) in 38 ml of demineralised water. You will only need 10 ml now, so store the rest of the solution in a small bottle for the next days. This way, you will only need to manipulate the concentrated acid once. Thirdly, add 75 ml of distilled water to the 200ml of brown xylene in the 75cl recipient.

8) Now add 1ml of diluted sulphuric acid. Place your xylene-resistant glove over the mouth of the bottle and shake violently for 10 seconds. Let stand. The layers will nicely separate and the bottom layer will stay colourless. Put your pipette in the jar, pear inflated, and
when you descend towards the bottom layer, push the ‘filling’-arrow while compressing the pear. This will blow air out of the pipette and prevent it from filling due to the hydrostatic pressure. When you reach the water-layer, stop bubbling and release the ‘filling’-arrow. Now compress the pear some further and fill the pipette with a few ml’s of solution. Make sure you fill enough, because when taking out the pipette, you must once again open the filling-arrow and slowly press some solution out of the pipette until you reach the surface. Phew! Now take the pipette out of the recipient and put a drop of acid on one of the litmus-papers. It does not matter if some xylene runs from the side of the pipette; this will not affect the outcome.

In the above picture you can see the big extraction-jar, the gloves, a glass with diluted sulphuric acid solution and a glass syringe, the titration-bottle with two layers, the 25ml pipette and a plate with pieces of litmus on it (notice the red one!).

9) If the litmus-paper stays blue, repeat step 8. You will notice that once you add the required amount of acid, the bottom layer will take on a slightly yellow colour. If you test now, it will surely be acidic. Possible improvement: this discoloration may be a sign of unwanted extraction; perhaps it would be better to stop the titration just before the watery layer tests acidic. This is definitely possible by doing the necessary calculations and pipetting a smaller third ‘backup’-amount of xylene from the jar.

10) When the litmus-paper tests acidic (most likely after 4-6 mls of acid solution, depending upon the strength of your cactus), you have neutralised all the free bases present in the xylene and there is an excess of sulphuric acid in the watery solution. To minimise this amount, add the ‘backup’-amount of xylene (the 100ml) to the titration recipient. If you used steps of 1ml diluted sulphuric acid, this amount will be enough to regain the excess of free-base in the xylene. Now do some calculations: Suppose you added 4ml of acid solution to the 275ml xylene and it still tested neutral. After adding the 5th ml, the solution turns acidic. Suppose you were able to get another 125ml of xylene from the jar. This would bring the total xylene volume to 400ml. You know you needed between 4 & 5ml to neutralise 275ml, so you will need between (4x400/275=5,8)ml and (5x400/275=7,3)ml to neutralise the full amount of xylene. You already added 5ml, so you can safely add 0,8ml and test the solution. If it tests neutral, you can add acid in steps of 0,2ml until the litmus-paper turns pink. This will bring the excess acid to a maximum of (0,2ml x dilution of 1/20 x 96%)=0,01ml sulphuric acid =1/5 of a drop. Of course, you can dilute your acid solution even further for the second titration, so your error will even get smaller. I hope this explanation is clear. Also, as mentioned before, it might also be possible to stop a little before this expected neutralization equilibrium. This keeps the watery layer clean and might reduce the influx of unwanted products. You could also keep a small amount (20ml) of backup-xylene at hand to add after the second neutralization. This will be investigated during the next extraction procedure.
11) So now you have a quite precisely titrated solution of all kinds of essence-sulphates and impurities in the bottom watery layer. The point is now to get this layer into a vaporising dish without getting any xylene there too. This is accomplished by first washing your pipette with hot water and detergent to get any remaining xylene out. Then you apply the bubbling technique to reach the watery layer and fill your pipette. Now you use the above-mentioned pressing-technique to get the pipette through the upper layer without any xylene coming in. You will unavoidably have some xylene running down the pipette when you pulled it out of the solution, so make sure you have some tissue-paper ready to clean the pipette before it can suck some xylene in from the bottom. Now empty the solution into a flat vaporising-dish. You will notice that even though you gave the layers some time to separate, there are still some drops of watery solution clinging to the sides of the glass. There can also remain a small emulsion layer. To get all the essence out, add another 75ml of demineralised water to the xylene, shake, let separate and get this layer into the vaporising dish as well. Make sure you put the dish in a dust-free environment while the beautiful and large essence-sulphate crystals form. However, some impurities will cling to them and to the vaporising dish as well.

The length of the above crystals goes up to 10cm!
12) Return the xylene to the big jar, shake until the emulsion is homogenous, let stand 24 hours and repeat steps 5-11 two more times. To get rid of the water in time, I put the dish in a pre-cleaned HEPA-box with a fan blowing over the surface of the water, a dessicator for large rooms and a halogen-light for warming the box. This setup evaporates 150ml of water in 10 hours or less. Here are some numbers from my first extraction: starting material: 340g dried cactus (outer skin only). Quantity of acid needed for first neutralization: probably 6,8ml diluted sulphuric acid (I overshot badly the first time, I began to doubt my calculations and went from 6ml straight to 10ml!). The amount of extract you get throughout the 3 neutralizations is pretty linear though, that’s why I can guess it’s probably 6,8ml. After scraping the crystals from the dish: 3,7g. Second neutralization: exactly 2,6ml diluted acid, amount of crystals: 1,4g. Third neutralization: I diluted the solution even further in order to work more precisely. 1 part of acid for 2 parts distilled water. From this solution I used exactly 3,0ml. Weight of the crystals after scraping: 0,5g.

13) Now it’s time to do the first purification step. Put all the crystals from the 3 neutralizations into a glass beaker and add 30-50ml of anhydrous acetone. You can make the acetone anhydrous by baking some magnesium sulphate (Epsom salt) in the oven until it releases no more water and then add this to 1l of acetone and shake the bottle. I used 50g of anhydrous salt, this is probably a large excess. Let it settle to the bottom for a few hours, and then you can take some anhydrous acetone out of the recipient with your pipette. I used 30ml, but I washed the first extract separately from the following two. It is good to crush the crystals some further in the acetone so that all the excess of acid and impurities are liberated from the crystals. Now put this beaker into a hot water bath and bring the acetone to a boil. Let it boil for a few seconds and then put the beaker in an angle of 45° on top of a cup or something, so that it stays in this tilted position. Put this setup into the freezer for 20 minutes. When you take it out, take care not to move the beaker too much. This will prevent the fine crystal-dust from suspending itself once again. Now put a large diameter needle onto a glass syringe (or use your pipette) to carefully suck up the acetone without disturbing the crystal layer. Unavoidably, you will get some clouds of dust in your solution as well, but these quantities are negligible. When you have taken most of the solution out, pour in another 30-50ml of acetone and repeat the process. You will notice that each acetone-washing becomes clearer and clearer. I washed the crystals 3 times, but four times could be better. Don’t worry about your essence-sulphate, it will not dissolve at all (it doesn’t dissolve in anhydrous acetone, and even if some water was present, the freezing step will recrystallize it out of solution). After the last washing, leave the beaker to dry. This brought my original 5,6g of crystals down to 5,1g, visually more white.

14) The second purification step: recrystallization. Start by getting most of the crystals out of the beaker and divide them into two test tubes of 20-25ml. You could put them into a single test-tube of larger volume, but I didn’t have one. The point is the recipient needs to be very narrow and tall for the decantation step. Rinse the beaker with 10ml of warm demineralised water and pour into one of the test-tubes. Then rinse the beaker a second time with 10 ml of warm demineralised water and pour into the second test tube. Heat these tubes in a hot-water bath (60-70°C) under regular shaking until the crystals are dissolved. Now add 2 ml of acetone to each test-tube. You will see a cloudy layer of crystals forming on top of the water. Shake. The crystals will dissolve. Add 2 more mls of acetone and shake again. If the crystals do not redissolve, heat the tubes a little more. Now add acetone ml by ml until you cannot heat the tubes any longer because of the acetone boiling out of solution (around 55-60°C). I added between 7-10mls of acetone to each tube. If you still observe undissolved crystals, keep on boiling acetone out of the solution until they are gone. Now let the test-tubes cool inside the hot-water bath and you will see
many needle-like crystals forming throughout the solution. Let cool to room temperature and then put in the refrigerator, still keeping the tubes inside the water-bath to act as a temperature buffer to slow down the cooling. Let stand for a few hours. Now with a clean metal wire, loosen the crystals from the sides of the tubes and break up the beautiful architecture of the needles. Not too much, you just want them to settle into a dense layer. Put the tubes in the freezer, together with 2 more tubes filled each with 10 ml of cold demineralised water. Now check the temperature of the water every 20 minutes until it approaches 0°C. Do not freeze the solution. Take the tubes out of the water bath and simply pour off the supernatant solution. You will see that the crystals remain packed in the bottom of the tubes. If you really want, you can collect the liquid and the subsequent rinsings for further recrystallization, but returns are minimal. Now pour the water from the tubes you also put in the freezer over the crystals, 10 ml into each tube and shake vigorously. Let the crystals settle for a while in the refrigerator, put in the freezer and decant the water-layer when reaching near-zero temperatures. Repeat this rinsing process two more times with anhydrous acetone. After the last rinsing, pour some acetone into the test-tubes to mobilize the crystals and pour the slurry into a Petri-dish for evaporation. Repeat the rinsing of the tubes until no more crystals remain. After evaporation, you will observe a glistening fluffy white tapestry of pure-white needles, softly interwoven with one another. Weight: 4.4g. Congratulations: you can stop here and use these small needles for dosing, or repeat the recrystallization-procedure to further purify the crystals.
The above pictures show the final steps of the procedure. After this, I wanted to obtain large crystals like I saw in the first evaporation-dish. So I dissolved the crystals in an excess (80ml) of demineralised water and poured this solution into a Petri-plate for slow evaporation. However, upon concentrating, the solution crystallised out in a fine layer on top of the liquid, thus preventing further evaporation. The result was a mess. It might very well be possible to grow large crystals out of less saturated solutions, but I decided not to try this. Instead, I redissolved the crystal mess in 40 (?)ml of water and in a hot-water bath added acetone (40ml) until clouding was evident. Then I let this solution cool slowly. It took much longer for the crystals to form out of this less-saturated recrystallization liquid. However, they were considerably larger! I need to get the recrystallization procedure right next time, for I suffered quite some losses. I only got 2.2g of ultra-pure crystals back from the liquid. I saved the liquids though, and after evaporation, some nice crystals were observed in the Petri-dishes (I will save them and add them to a second extraction-solution). For the moment, I enjoy having essence-sulfate crystals of this extreme purity. The grid underneath the crystals is made up of 1x1cm squares.
15) Dosing: There is an excellent little article on the internet on how to make a microgram (!)-
scale out of a galvanometer, a stabilised power-supply and a high-precision variable
resistor. I simplified this down to a comparator between the calculated weight of a
measured length of copper-wire with known mass to length-ratio and a mass of crystals. It uses a 12V DC-supply, a cheap variable resistor of 2KOhms and a precision variable resistor of 50KOhm in parallel. The extra 1KOhm resistors are there to allow for scaling. It is precise to 1 milligram.

16) Storage: In a glass canning jar of 0.5-1 litre with rubber ring and glass lid, put some desiccant on the bottom (you can use calcium chloride, or magnesium sulphate to which a small amount of anhydrous blue cobalt chloride is added as indicator). Then place a mini-rack or net or spacing of some kind on the desiccant and put the Petri-plate on top. Now take this setup and put it inside a big translucent plastic bag. In this bag, you also put a ceramic/metal setup to burn a piece of clean charcoal (pyrolised willow, or water-pipe tablets). Now put the lid and clips inside the bag, light the charcoal and close the bag. After 30 min or when the charcoal dies of suffocation, without opening the bag, place the lid with rubber ring on the glass jar and put the clips on it. Now you will have a more or less inert atmosphere of nitrogen, carbon dioxide and presumably a little carbon monoxide in the jar, that is kept anhydrous by the desiccant. Take the jar out of the bag and put in the freezer until use. After each olfactory exploration, use the same procedure to store the essence.

Please respect the transforming nature of this essence and put it to constructive use. May you fare well and enjoy illuminating experiences,

Litmus.