

Eugenol Extraction from Cloves. You will be isolating the natural product, eugenol, from cloves. It is possible that there are some esters of eugenol present, such as acetyl eugenol, along with eugenol itself.

Steam Distillation of Essential Oils Weigh out 15 grams of cloves and place them in a 250 mL round bottom flask. Add about 125 mL of water. Set up a simple distillation apparatus. Heat the contents using a low flame (acceptable here because you are essentially distilling water) until the water starts to boil. Occasionally this mixture will foam, so boil at a rate that keeps the foam from bumping over. You can transfer out your distillate periodically to a graduated cylinder to keep it separate in case your distillation does bump over (and so you can monitor the amount of liquid that has distilled out). Collect about 80-100 mL total, making sure to periodically add water to the round bottom flask to replace the water distilled (simply remove the flame, lift the thermometer adapter, and pour water through the distillation head).

Dissolve Oils into an Organic Solvent (Extraction from Aqueous Layer) Place the distillate in a separatory funnel and extract with three portions (~15 mL each) of dichloromethane (methylene chloride, CH_2Cl_2). Remember, the bottom layer is the more dense liquid – the organic layer in this case! Shake gently at first since dichloromethane can form emulsions with organic solutes and water. If you get an emulsion, you can draw off the clear part of the dichloromethane (bottom layer) up to the emulsion layer on your first two extractions. On your third extraction, shake the mixture more gently and allow more time for separation (you can gently use a glass stir rod to break up the bubbles in the emulsion). If there are lots of bubbles at the interface, leave that part behind to avoid drawing off any water. Combine all of your dichloromethane extracts and add enough calcium chloride pellets so that it does not clump. Swirl the flask well to allow the drying agent to adsorb the water from the wet dichloromethane. If the calcium chloride is not powdered, you should be able to decant the solution from the drying agent. If you see fine powder, then do a gravity filtration with filter paper.

Analyze “Crude” Organic Layer This dried solution contains ALL of the volatile oil components steam distilled from the cloves (eugenol + eugenol esters + other organic molecules), and we will analyze a portion of this collectively as a class. Measure your volume of the dried dichloromethane solution in a graduated cylinder. Place approximately one-fifth into the tared round bottom flask that has been provided for the class. The instructor will rotovap this fraction (**crude oils #1**) and take an IR spectrum, and this solution can be used for TLC analysis.

Separate Eugenol from Other Essential Oils by Extraction with Aqueous Base Take the remaining 4/5 of your dichloromethane solution and add it to your 125 mL separatory funnel. EXTRACT it with three portions (~10 mL each) of 5% aqueous NaOH solution (NOTE: this causes the deprotonated eugenol to move to the aqueous layer!). Set aside the organic layer but do not discard anything until the entire procedure is completed. This fraction has any neutral organic material not extracted into the aqueous base solution.

Isolate Eugenol by Neutralizing and Extracting Back into Organic Acidify the aqueous NaOH extracts to a pH \approx 1 with concentrated HCl (use litmus paper to check the pH by touching the paper with the tip of a pipette or glass rod). EXTRACT the acidified aqueous layer with three portions (~8 mL each) of dichloromethane, being careful again with possible emulsions being formed (NOTE: this will move the neutral eugenol back into the organic layer!). Dry the combined organic layers (**eugenol #2**) with calcium chloride, as before, and gravity filter into a tared round-bottom flask. Use a rotovap to remove the dichloromethane. Analyze residual oil by IR and calculate a mass percent recovery of eugenol from the cloves.

Analyze your different fractions (#1, #2) using TLC (Develop with 1:1 dichloromethane/hexane or 90:10 hexane/ethyl acetate eluent).