Introduction

Marijuana (Cannabis) is identified by macroscopic and microscopic tests based on its morphological characteristics and confirmed by the color reaction of the Rapid Modified Duquenois-Levine (D-L) test. The combination of the macroscopic, microscopic and D-L test makes the identification for marijuana highly specific. However, in the case of hashish, the loose cystolith hairs are of limited diagnostic value since they cannot be related to the plant structure from which they had originated. Therefore, the Chemistry Unit employs instrumental techniques such as Gas Chromatography/Mass Spectrometry (GC/MS) when hashish or hash oil is submitted.

Safety

- Personal protective equipment (PPE) including safety glasses/safety shield, gloves, and lab coat/scrubs must be worn when working with the potentially hazardous materials in this procedure. A fume hood must always be used when working with hazardous gases, solvents, dust, etc. Additional protective equipment may be used at the discretion of the analyst. Appropriate personal protective equipment is specified in the Safety Section of the Quality Assurance Manual (QAM).

- Disposal of all chemical and biohazardous waste must be performed in accordance with the procedures detailed in the Safety Section of the QAM.

Paraphernalia

- Paraphernalia such as pipes will be analyzed on a case by case basis with approval of the Chemistry Manager. Pipes will only be analyzed for marijuana if sufficient plant material is present to perform a macroscopic, microscopic, and color test for the presence of Cannabis.

Macroscopic Examination

- Gross morphological characteristics that may be observed include the palmate arrangement of the leaflets, the pinnate appearance of the leaflets, the serrated edges of the leaflet, the buds (with or without seeds) and if present, fluted stems and/or stalks.

- Positive macroscopic examination results are recorded in the analytical notes by using the abbreviation 'pos', '+', or 'Y'. Negative macroscopic examination results are recorded in the same location using 'neg', '-', or 'N'. The chemist will also document what macroscopic characteristics were observed leading to the positive result.
• In some cases, the condition of the plant material or quantity of plant material is insufficient to obtain macroscopic identification. Confirmation of plant material as Cannabis can be made based solely on the microscopic examination and a positive Rapid Modified Duquenois-Levine test.

Microscopic Examination (Plant Material):

• Place leafy material under the stereo zoom microscope, and observe the following botanical characteristics:
  ➢ Cystolith hairs (bear claw appearance), must be present
  ➢ Elongated hairs (underside of leaf), must be present
  ➢ Resin glands (glandular hair), may be present

• For the microscopic test to be considered positive, both cystolith and elongated hairs must be present. Positive identification of each type of hair is documented in the analytical notes as 'pos', '+', or 'Y' in the appropriate field. Negative identification of each type of hair is documented in the analytical notes as 'neg', '-', or 'N' in the appropriate field.

Microscopic Examination (Differentiation between Compressed Marijuana and Hashish)

• This examination only needs to be performed when the appearance of the item is similar to compressed marijuana. If item can be visually determined not to be compressed marijuana, this examination does not need to be performed.

• Place a portion of the suspected hashish on a microscopic slide.

• Apply chloroform dropwise and observe under a polarizing microscope.

• Look for an abundance of unattached cystolithic hairs or "bear claws".

• Using the crossed nichols position (polarizer and analyzer 90° to one another), observe the birefringence of the hairs or “bear claws”. The hairs should be bright white in appearance and be easily extinguishable from surrounding leaf material.

• The combination of the loose hairs and the birefringence is highly indicative of hashish and not compressed plant material.

Performing the Rapid Modified Duquenois-Levine Test
• Preparation of the D-L reagent (505 mL): Dissolve 5 mL of acetaldehyde and 10 grams of vanillin in 500 ml of 95% ethanol.

• Place a small portion of the material in a test tube.

• Add approximately 1 mL of the D-L reagent to the plant material. Optional: After waiting for a few minutes, decant solution off of leaf material into another test tube.

• Add at least equal portions of concentrated hydrochloric acid, as there is D-L reagent to the test tube.

• Observe the color development to purple within a few minutes. If the color does not develop, it is possible that the sample does not contain cannabinoids or the sample may be a seedling, old or moldy. Try extracting the sample with petroleum ether first, drying the ether, then performing the color test.

• Extract the purple color in the aqueous layer with chloroform (CHCl₃) and observe the purple color in the CHCl₃ layer of the test tube.

• The purple colors in both layers have to be observed for the test to be considered positive. Positive results are indicated in the analytical notes as either 'pos', '+', or 'Y'. Negative results are indicated in the analytical notes as either 'neg', '-', or 'N'.

• Results of the macroscopic, microscopic and D-L tests will be documented on the CH Marijuana Form.

Gas Chromatography/Mass Spectrometry (For Hashish or Hash Oil and Other Materials Potentially Containing Tetrahydrocannabinol. Optional Additional Test for Cannabis)

• Inject the extracted sample from which the IS has been added, into the GC/MS using the MSSCREN, QSCREN, or STEROID method along with the appropriate standards and blanks.

• Compare the fragmentation pattern of the sample to that of the cannabinoid standard. The base peak should be the same between the sample and standard spectra along with the general overall pattern between the two mass spectra. Tetrahydrocannabinol (THC) must be identified in order to report as hash.

Reporting Results

• Plant material: Report will state in similar form or other slight variation as “CANNABIS Schedule I Net Weight: # grams”.

• Hash and Hash Oil: Report will state in similar form or other slight variation as DELTA-9-ΤΕΤΡΑΗΔΡΟΚΑΝΝΑΒΙΝΟΛ Schedule I Net Weight: # grams.”
References


- *Analysis of Drugs Manual*; DEA, U.S. Department of Justice, Office of Forensic Sciences, 2nd ed.


- *Basic Training program for Forensic Drug Chemists*; DEA, U.S. Department of Justice, Office of Forensic Sciences, 3rd ed.