# Physical And Chemical Characteristics Of Cannabis Found In Trinidad And Tobago

R Maharaj, G Singh, C Thomas, D John

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**Abstract**

The analysis in 2002 of representative samples taken from flowering and fruiting tops of cannabis material seized by law enforcement officials in Trinidad and Tobago showed that the mean content of 9 tetrahydrocannabinol (THC) was 5.26%, range 0.53 – 11.58% with a standard deviation of 2.98%. The mean THC content of the Cannabis samples seized in 2002 was significantly higher than the content found Cannabis representative samples seized and analyzed in Trinidad and Tobago in 1993 (mean content of 9 tetrahydrocannabinol (THC) was 0.079%, range 0.01 – 0.18% with a standard deviation of 0.04%) .The specialized glands present on the aerial structures of the plant responsible for the biosynthesis of the cannabinoids were observed in all the samples.

## INTRODUCTION

Cannabis may have been the first cultivated plant. Records indicate use of this crop for paper, textiles, food and medicine throughout human history (Abel, 1980). It is a dioecious annual with distinctive palmate leaves, usually composed of an odd number of leaflets. Mature height ranges from 1 to 5 meters, according to environmental and hereditary factors. As with other plant species, the male plant is somewhat taller and more obviously flowered. These flowers have five yellowish tepals, and five anthers that hang at maturity, dispersing their pollen to the wind. The female plant has shorter branches and dense growth of leaves and flower-associated bracts. A single achene is produced per flower and shed or dispersed as a result of bird predation. The life cycle of the male is completed soon after anthesis, but the female survives until full seed ripeness.

Much confusion exists over the various terms used to describe the Cannabis plant. Terms such as vulgaris, pedemontana, lupulus, mexicana and sinensis have been used in the last centuries. Cannabis is the botanical genus of all these plants. Hemp is used to describe Cannabis plants high in fibre content whereas marijuana is used to describe Cannabis plants high in psychoactive components (Shultes and Hofmann, 1992) and (Stafford, 1992). Botanists agree however that there are hundreds of unique variants of the various Cannabis species. Unfortunately extensive hybridization and cultivation has made them hard to identify.

Secondary metabolic compounds are produced by Cannabis. A variety of alkanes have been identified (Adams Jr., Jones, 1973), as well as nitrogenous compounds (El Sohly et al, 1982), flavonoids (Gellbert, 1974)) and other miscellaneous compounds (Hanus, 1976). Terpenes appear in abundance and contribute to the characteristic odor of the plant (Hendricks et al, 1975) and some of its crude preparations, such as hashish. The compounds which comprise the active drug ingredients are apparently unique to this genus and are termed cannabinoids. There are over 60 of these type compounds present in the plant (Turner et al, 1980).

Delta-9-tetrahydrocannabinol (THC) is the cannabinoid responsible for the main psychoactive effects of most Cannabis drug preparations (Mechoulam, 1970). THC is thought to be produced by the plant from cannabidiol (CBD) which, in turn, is derived from cannabigerol (CBG) generated from non-cannabinoid precursors (Hammond and Mahlberg, 1994). CBG is also the biogenetic precursor of cannabichromene (CBC). Other cannabinoids are probably degradation products of the enzymatically produced cannabinoids such as CBD, THC and CBC.

The major sites of cannabinoid production appear to be epidermal glands (Fairbain, 1972). These epidermal glands seem to fall into two broad categories: stalked and sessile. The glandular cells are covered with a “sheath” under which the resins are secreted via vesicles (Mahlberg and Kim, 1992). A spherical structure forms as the resins accumulate

until the sheath bulges away from the secretory cells. The resin is then released by rupture of the membrane or through pores in its surface (De Pasquale et al, 1974). Bracts subtending the female flowers contain a greater density of glands than the leaves. The bracteole enclosing the pistil has the highest cannabinoid content of any single plant part (Kimura and Okamoto, 1970).

The widespread use of the sinsemilla growing technique (Marnell, 1997)) has increased the average concentration of THC from 1 – 3% in the 1960s to 6-14% in the 90s (DEA, 1997). There are several reports in the literature on the THC content of cannabis and some of these are summarized in table I. In the present context the origin refers to the country in which the plant was ground and not the country of origin of the seeds. The quoted THC levels are expressed as a percentage of dry weight of cannabis.

**Figure 1**

Table 1: Summary of published data on THC contents of Cannabis

| | | THC CONTENT (%) | | |
|---|---|---|---|---|
| NUMBER OF SAMPLES | COUNTRY OF GROWTH | RANGE | MEAN | REFERENCE |
| 13 | Argentina | 0.1-8.3 | 2.2 | 17 |
| 36 | Jamaica | 0.04-7.9 | 2.8 | 22 |
| 13 | Mexico | 0.1-3.0 | 1.7 | 23 |
| 30 | United States | 0.1-0.5 | | 24 |

Most of the data in table are derived from cannabis legitimately grown and processed as part of scientific studies of cannabinoids in the 1970s.

The present work is concerned with the identification of the epidermal glands (both stalked and sessile) as well as non-glandular trichomes in plant material seized by law enforcement officials in Trinidad and Tobago in the years 1993 and 2002. The level of THC content in the representative samples will be quantitated and the mean concentration of each year statistically compared to detect any changes in THC concentration over the period.

## MATERIALS AND METHOD

Fifty (50) and Sixteen (16) samples of plant material were collected in 1993 and 2002 respectively. These samples were seized from different parts of Trinidad and Tobago by Police Officers and submitted to the Trinidad and Tobago Forensic Science Centre for analysis.

Segments of leaves, stem, bracts and roots were immersed in 5% chloral hydrate solution and gently heated for 30 mins. The sections were examined with a Zeis Microscope (magnification 100). Samples of Cannabis were removed from the flowering and fruiting tops of the plant and were chopped and mixed to give a homogeneous mixture prior to analysis by gas chromatography.

The extraction process was carried out for each sample as follows:

Plant material (3g) was placed in a conical flask and approximately 50mls toluene added. The conical flask was stoppered and mounted on a Gallenkamp orbital shaker, at 140 revolutions per minute for two hours. The solution was filtered undergravity. The filtrate was transferred to a round bottom flask and evaporated to dryness, under vacuum, using a Buchi Rotary Evaporator at 70°C.

Each extract was made up to 50ml using an internal standard solution of 0.50mg/ml tetracosane in chloroform: methanol (1:1) and 2ml of this solution analysed by gas chromatography (Fairbairn and Liebmann, 1973). A Hewlett Packard model 6890 instrument was fitted with a HP-5 capillary column cross link 5% PHME siloxane (30m x 3.2mm x 0.25um film thick). The carrier gas flow rate was 2ml/min and the column temperature was 250ºC. Detection was by flame conisation and the detector was calibrated for THC response using a primary standard of 0.50 mg/ml. THC in an internal standard solution of 0.50mg/ml tetracosane in chloroform:methanol (1:1).

## RESULTS AND DISCUSSION

The microscopic analysis of the segments of plant material revealed the presence of glandular and non-glandular trichomes on most of the aerial parts of the plant. None of the characteristic features were observed on the root surfaces. The non-glandular trichomes observed included the cystolithic and non-cystolithic types. The stalked glands appeared as a single cell or a small group of cells on a single or multicellular pedestal whereas the sessile glands possess no stalk. Cystolithic and non-cystolithic Most of the structures were found at the apical tip and on the underside of the leaves. These observations are consistent with previous work done (De Pasquale et al, 1974).

The results generated from the gas chromatography analysis revealed that the mean THC (%) content for the sixteen (16) samples collected in 2002 was 5.26%, range 0.53 – 11.58% with a standard deviation of 2.98% and that the mean THC (%) content for the fifty (50) samples collected in 1993 was 0.079%, range 0.01-0.18% with a standard deviation of 0.04%

Statistical analysis of the data was done suing the Statistical Package for Social Scientist (SPSS) version 13.0. An independent two-sample t-test also referred to as the Aspin-Welch Unequal-Variance Test was conducted to evaluate the hypothesis that the purity of drugs produced in 2002 was higher to that produced in 1993. The test was significant at the conventional levels of significance of 0.0001, 0.01, 0.05 and 0.10. The value of the t-test statistic computed was 6.964 (degrees of freedom = 15) and the p-value obtained was 0.000002. As a result, one can strongly conclude that the concentration of the psychoactive drug THC in the samples seized in 2002 was significantly higher than those produced in 1993 (Mean = 0.0790, SD = 0.04032, n = 50).

The widespread use of the sensimella technique (Marnell, 1997) as well as other heredity factors (Small and Beckstead, 1973) can be attributed to the increase in THC content of Cannabis found in Trinidad and Tobago. The Sinsemilla technique (Marnell, 1997) refers the process of removing male plants from the grow environment before they have a chance to fertilize the females. The resultant unfertilized female cannabis plants contain more psychoactive THC. The amount of THC in sinsemilla is considerably higher to cannabis that has been grown in a pollinated environment, as if left unpollenated a female plant will divert all her energy to calyx production in an effort to catch pollen instead of the production of seeds. Due to the fact that the calyces contain a high density of drug producing trichomes, the greater the number of calyces the more drug is produced by the plant.

Ecological factors have long been thought to have an important influence in THC concentration by stressing the Cannabis plant and hence its THC content (Bouquet, 1950). The resultant increased biosynthesis of the cannabinoid and terpene containing resin, in most cases, seems likely of advantage to the organism in adapting it to a variety of survival-threatening situations. They act as barriers (dessication) to water loss in dry conditions, insect predation and microorganisms such as bacteria and fungi. Terpenes have been shown to suppress the growth of surrounding vegetation and hence reduce competition from other species. Environmental stresses such as Ultra Violet radiation, temperature and variation in soil nutrients all influence the determination of cannabinoid content in plants.

## CONCLUSION

The epidermal glands responsible for the biosynthesis were observed in the Cannabis samples examined in the study. The level of THC in the samples analyzed in 2002 was higher than that observed in 1993. Although the chemistry of Cannabis has come under extensive investigation, more work is needed to probe the relationship of its resin content to biotic and abiotic factors in the environment.

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## Author Information

**Rean Maharaj**
University of Trinidad and Tobago, (UTT) O'Meara

**Gizelle Singh**
Trinidad and Tobago Forensic Science Centre, Ministry of National Security

**Clive Thomas**
Trinidad and Tobago Forensic Science Centre, Ministry of National Security

**Deborah John**
Trinidad and Tobago Forensic Science Centre, Ministry of National Security