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Analysis of adulterants in prepared bhang from different Authorized bhang shops

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Abstract:

Cannabis is a tall, erect, annual, dioecious herb, provided with an open sunny environment, light, well-drained composted soil and ample irrigation. Bhang is obtained from cut tops of uncultivated plants and is least potent. The potency depends upon the concentration of Tetrahydrocannabinol (THC). It is a narcotic drug and is analgesic in nature. Bhang is prepared by crushing the leaf of cannabis plant and its effects are described as sedative, stimulant and hallucinogenic. In present time, the adulterants like Dhatura, Tobacco or chemical substances such as sleeping pills are added to increase or decrease its quality or quantity for benefits. These adulterants are harmful for human body and sometimes it becomes toxic. Therefore, the present study was carried out with the objective of examining the adulterants present in the collected samples of Bhang through several chemical tests and chromatographic methods. The results were both positive and negative that indicated the presence and absence of the adulterants respectively. More than 50% samples were found to be adulterated with Dhatura and tobacco alkaloids.

Keywords- *Cannabis, Bhang, Nicotine, Sedative, Hallucinogenic, Stimulant,*

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Introduction

Cannabis is a tall upright annual herb. It is generally dioecious i.e. producing separate male and female plants but fiber hemp varieties have been specifically bred to be monoecious (hermaphrodite). The leaves are palmate, and in the iconic image of a cannabis leaf there are seven lobes, the lowest pair showing as backwards facing spurs. However, this number and shape is not fixed. On seedlings the first pair of leaves is typically monophylous (single lobed), the second pair having three lobes and the next pair five. In many plants, especially of central Asian origin, the number does not extend beyond five while in others the number can extend to around thirteen. The genera Cannabis and Humulus (hops) belong to the same family (*Cannabaceae*, sometimes known as *Cannabinaceae*). Generally, cannabis is considered to be monospecific (*Cannabis sativa L.*) which is divided into several subspecies (*C. sativa subsp. sativa, C. sativa subsp. indica, C. sativa subsp. ruderalis, C. sativa subsp. spontanea, C. sativa subsp. afiristanca*). However, the chemical and morphological distinctions by which cannabis has been split into these subspecies are often not readily discernible, appear to be environmentally modifiable, and vary in a continuous fashion. For most purposes, it will suffice to apply the name Cannabis sativa to all cannabis plants encountered. Cannabis is an annual, dioeciously, flowering herb. Staminate (male) plants are usually taller but less robust than pistillate (female) plants. Stems are erect and can vary from 0.2-2.0 m. However, most of the plants reach heights of 1-3 m. The extent of branching, like the plant height, depends on environmental and hereditary factors as well as the method of cultivation. Cannabis is a tall, erect, annual herb, provided with an open sunny environment, light well-drained composted soil, and ample irrigation. Cannabis will deteriorate in about two years if exposed to light, air or heat. It should always be stored in cool places. Cannabis is a wind pollinated species. The males, which are generally taller than the females commence flowering first. The plant was grown in still conditions and leaves appear yellow under the deep covering of pollen. When mature, the sepals on the male flowers open to expose the anthers, which hang freely on fine filaments. The female plants tend to be shorter and have more branches than the male. Female plants are leafy to the top with many leaves surrounding the flowers, while male plants have fewer leaves near the top with few if any leaves along the extended flowering limbs. The sticky resin produced by the flowers and top leaves contains a number of psychoactive substances, collectively known as cannabinoids, these collectively make up the drug called cannabis. The gland of the female flower secret drop of resin, which are produce under hot condition male plants die after the pollination in the absence of male plant, female ones produced more flower covered with THC producing glands. A function of resin gland is the protection of the plants from animals. The greenish or brownish fruit is physically an achene. It is ellipsoid, smooth slightly compressed about 2.5-5cm long and the diameter is 2-3.5cm chemical constituent cannabis (**Buchanan and O'Connell, 1998).**

The potency of the cannabis obtained from a plant is dependent on the content of delta-9tetrahydrocannabinol (THC), the most important of the cannabinoids. THC content is dependent on the part of the plant used, the method of cultivation, and the preparation of the extract:

a) Bhang obtained from cut tops of uncultivated plants with low resin content is the least poten.
b) Ganja or marijuana from flowering tops and leaves from specially cultivated plants has higher resin content and is more potent. Both of these herbal preparations (also known as 'grass' or 'weed') are usually smoked in hand-rolled cigarettes ('joints' or 'reefers'). Potency is variable, with a THC content of 1-10 per cent.
c) Cannabis resin (hashish) is the resin itself, in the form of a sticky brown cake, which can be smoked or eaten. Liquid cannabis or hashish oil is extracted from cannabis resin, and is more potent. Tobacco is dipped in this before smoking. It may contain up to 60 percent THC, and is a Class A drug. The Cannabis plant and its

products consist of an enormous variety of chemicals. Some of the 483 compounds identified are unique to Cannabis, for example, the more than 60 cannabinoids, whereas the terpenes, with about 140 members forming the most abundant class, are widespread in the plant kingdom. Cannabis contains over 300 compounds. At least 66 of these are cannabinoids, five important cannabinoids found in the cannabis plant are:

1. Tetrahydrocannabinol (THC)
2. Cannabidiol (CBD)
3. Cannabinol (CBN)
4. β-caryophyllene
5. Cannabigerol.

Bhang is prepared by crushing the leaf of cannabis plant. The effect of cannabis are confusing, it is described as sedative, stimulant and hallucinogen. In present time the adulterants like Dhatura, Tobacco or chemical substances such as sleeping pills are added to increase or decrease its quality or quantity for benefits. These adulterants are harmful for human body and some-times it becomes toxic.

Review of Literature

Mobarak *et al.*, (1974) suggested chromatographic methods for detection and identification of hashish (*Cannabis sativa L.*) constituents by two dimensional chromatography, withremarkable resolution Gas chromatography separation of some of constituents of hashish.

Joseph *et al.*, (1979) studied young plants of five Nicotiana tabacum L. genotypes were examined for activity of nicotine biosynthetic enzymes. Genotypes near isogenic except at two loci each with two alleles controlling nicotine level were used in a comparison of the four homozygous allelic combinations producing high, high intermediate, low intermediate, and low nicotine levels in a 'Burley 21" background.

Tiwari and Sharma (1982) has described TLC technique for the separation and identification of the extract of the plant flowering tops and the resin of cannabis as well as from the residue of smoking-pipes. Alumina plates developed by benzene-chloroform (1:1 v/v) gave good separation of the components of hashish. Ultraviolet light (254nm) and Fast-blue-B chromogenic reagent were used for the location of the spots. The R_f value the colour of the spots of the separated components are recorded.

McPartland and Ethan (2001) stated the use of botanical remedies is that herbs contain many active ingredients. Primary active ingredients may be enhanced by secondary compounds, which act in beneficial synergy. Other herbal constituents may mitigate the side effects of dominant active ingredients. We reviewed the literature concerning medical cannabis and its primary active ingredient, □^{9}-tetrahydrocannabinol (THC). Good evidence shows that secondary compounds in cannabis may enhance the beneficial effects of THC. Other cannabinoid and non-cannabinoid compounds in herbal cannabis or its extracts may reduce THC-induced anxiety, cholinergic deficits, and immunosuppression. Cannabis terpenoids and flavonoids may also increase cerebral blood flow, enhance cortical activity, kill respiratory pathogens, and provide anti-inflammatory activity.

Fraser and Worth, (2002) suggested that Cannabinoids can be detected by numerous and various analytical methods, including immunoassays (EMIT, ELISA, radioimmunoassay).

Teska *et al.* (2002) stated that planer chromatography technique: the classical thin layer chromatography (TLC), the optimum performance laminar chromatography (OPLC) and the automated multiple development (AMD).

Grinspoon and Twert (2003) reported that cannabis is most frequently prepared for human consumption from the dried flowers of the plant commonly referred to in the United States as marijuana. It is usually ingested by smoking but is also consumed

orally. As cannabinoid are not water soluble, cannabis preparations are not suitable for injection.

Hall and Pacula (2003) describe Cannabis as one of the oldest psychotropic drugs known to humanity with cultivation and use noted in archeological discoveries from more than 6000 years ago. One cannabinoid, delta 9-tetrahydrocannabinol, was the first cannabinoid to be isolated and is the primary psychoactive ingredient in cannabis. It is commonly referred to as THC. Unlike THC. The majority of known cannabinoid has mild to no psychoactive properties and do not lead to intoxication.

McPartland (2008) stated that shifting demographic of people admix cannabis with cholinergic agents, intent upon enhancing cannabimimetic effects or reducing adverse effects. Augmentation of cannabimimetic effects with tobacco (or nicotine) has been corroborated by in vitro mechanistic studies, animal behavior studies, anecdotes from patients, and one clinical trial.

Mabroukah *et al.*, (2015) the present research is focused on determination of nicotine content in international tobacco brands and growth tobacco in Libya locally sold in Libyan markets. Extraction method was used to extract the nicotine from the thirteen tobacco brands, three local produced tobacco and ten international brands of locally sold tobacco.

Methodology

Collection of sample:-

Standard sample- Fresh green leaves of bhang were collected from Gangotri Nagar, Allahabad and grind it for use as standard sample.

Suspected sample- The twenty samples of bhang (cannabis) was collected from different government authorized bhang shops of Allahabad. 8g of sample were collected from twenty different shops of Allahabad in wet and crushed form.

Physical Examination:

The Standard and suspected samples were physically visualized for the colour and leaves of the plant and the result was recorded.

Microscopic Examination:

Standard and Suspected sample of cannabis (bhang) were examined under the Microscope for the characteristic features of the cannabis.

Extraction of cannabis (bhang) Sample:

The samples were extracted by acid-ether extraction method.

About 2g of sample is transferred to separating funnel and acidified with 2N HCL by shaking it for 1 min.

After acidifying 10 volumes of Ether is added into the separating funnel and shaken for 1 min.

After adding of Ether the mixed sample was left for 2 min to settle down.

After that the ether was decanted off and the fraction was used for analysis after evaporating the solvent.

Colour test:

The suspected samples were subjected to colour tests, the results were noted down, and the following tests were performed for detection of cannabinoids and other suspected adulterants in the samples.

Test for Cannabis (Bhang)-

Fast blue B salt Test:

A small amount of extracted sample residue in taken in a test tube and add very small amount of solid reagent and 1 ml of chloroform. Shaken and add 1 ml of solution-II, shaken and allow the test tube to stand for two min, the resulting colour was noted down.

Nigam's Test:

Few drops of 8% solution of alcoholic vanillin solution and 2 drops of 2% aqueous acetaldehyde solution was added to the small amount of sample residue, then 2 ml of conc. HCL was added and the resulting colour was noted down.

Test for Dhatura Alkaloids-

Vitalis Test:

In a porcelain dish small amount of extracted sample residue was taken and 2-3 drops of acetic acid solution were taken into it and evaporated. To the dry residue one drop of conc. HNO_3 was added and again evaporated to dryness on a hot water bath. After cooling, a drop of alcoholic caustic potash was added to it and the resulting colour was noted down.

Test for Tobacco Alkaloids-

Schindelmeister Test:

A small amount of ethereal extract was taken in a test tube and evaporated to dryness.

One drop of formaldehyde solution and a drop of concentrated H_2SO_4 acid are added and the resulting colour was noted down.

Result

On the basis of the results of different chemical tests shows that bhang (cannabis) commonly used for drugging was adulterated with different adulterants like tobacco and other drugging and toxic components.

In this work, twenty samples of prepared bhang were analyzed for the adulterate in which eleven samples found to be adulterated and the adulterant which were found in the sample is tobacco. More than fifty percent sample were found to be adulterated. The retailors of authorized bhang shop add intentionally these type of adulterants to increase their quantity of product for the purpose of more profit.

Schindelmeister' test results shows that the suspected sample collected from different shops was adulterated with tobacco. It was observed that, In sample S1, S5, S6, S8, S9, S11, S13, S14, S15, S16 & S17 nicotine was present which shows that the samples were of adulterated with tobacco, the similar result were found in the research of **McPartland and Ethan (2001) McPartland (2008).**

Table 4.4: Schindelmeister Test for Tobacco Alkaloids (nicotine)

Sr. No.	Sample No.	Observation	Result
1.	Standard sample(S)	Fade yellow	Nicotine was absent
2.	Sample 1 (S1)	Reddish brown	Nicotine was present
3.	Sample 2 (S2)	Mild yellow	Nicotine was absent
4.	Sample 3 (S3)	Mild yellow	Nicotine was absent
5.	Sample 4 (S4)	Mild yellow	Nicotine was absent
6.	Sample 5 (S5)	Reddish brown	Nicotine was present
7.	Sample 6 (S6)	Reddish brown	Nicotine was present

8.	Sample 7 (S7)	Mild yellow	Nicotine was absent
9.	Sample 8 (S8)	Reddish brown	Nicotine was present
10.	Sample 9 (S9)	Reddish brown	Nicotine was present
11.	Sample 10 (S10)	Mild yellow	Nicotine was absent
12.	Sample 11 (S11)	Reddish brown	Nicotine was present
13.	Sample 12 (S12)	Mild yellow	Nicotine was absent
14.	Sample 13 (S13)	Reddish brown	Nicotine was present
15.	Sample 14 (S14)	Reddish brown	Nicotine was present
16.	Sample 15 (S15)	Reddish brown	Nicotine was present
17.	Sample 16 (S16)	Reddish brown	Nicotine was present
18.	Sample 17 (S17)	Reddish brown	Nicotine was present
19.	Sample 18 (S18)	Mild yellow	Nicotine was absent
20.	Sample 19 (S19)	Fade yellow	Nicotine was absent
21.	Sample 20 (S20)	Pale yellow	Nicotine was absent

Conclusion

As we know that some drugs are more toxic when it combined with another drug, it effects the functioning of the brain as a result mass disaster may happens. When bhang and tobacco combines, the effect of the drug increases resulting disorder in brain.

Combined use of bhang and tobacco is highly prevalent in today's population. Individual use of either substance is linked to structural brain changes and altered cognitive function, especially with consistent reports of hippocampal volume deficits and poorer memory performance. Due to effect of these drugs person can face mental disorders. Excess does of these drugs can leads to person's death.

In this study, twenty samples of bhang were analyzed for its genuinity and adulterants, In which all 20 samples found as genuine bhang eleven of them found to be adulterated and the adulterant which was found in the samples is tobacco. More than fifty percent sample were found to be adulterated. The retailors of authorized bhang shop add intentionally

these type of adulterants to increase their quantity of product for the purpose of more profit.

From the observation it was concluded that the presumptive test i.e., **Schindelmeister test** is enough for identification of adulterants in bhang (Cannabis) although this test is specific for the tobacco.

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