Original Article

Thin layer Chromatography of Ibogaine from plant extract of Tabernanthe iboga.

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Abstract
Ibogaine is a slightly psychoactive 5 to 6% of indole alkaloid derived from the Tabernanthe iboga plant native to Central Africa. The iboga plant contains the different chemical constituents such as the active alkaloids ibogaine, ibogamine, noribogaine, ibogaline, etc. Iboga & Ibogaine are used in various treatments of addictions, depression, drowsiness, fever, the flu, HIV-AIDS, high blood pressure, nerve disorder, fatigue, low sex drive. The Tabernanthe iboga root extract are necessary involve the separation of different chemicals using thin layer chromatography (TLC). TLC is useful separation, purification and identification of plant extract, by this means the ibogaine can be distinguished from the other iboga alkaloids and its relative concentration estimated. The various TLC methods were developed, in all methods uses different solvent system. Ibogaine is not itself addictive, and may thus be administered in monthly or similar doses over a period of time to help preserve a drug-free state.

Keywords: Ibogaine, Tabernanthe iboga, Indole alkaloid, TLC.

Introduction
The word chromatography means ‘to show in colour’in greek & was first introduced by Russian biotanist ‘michel tswett’ to describe that solvent according to adsorbance sequence are resolved into various colour zone. (Alliston, 1971) TLC is a chromatography technique used to separate mixtures (Vogel, 1989). It involves a stationary phase consisting of a thin layer of adsorbent material, usually silica gel, aluminum oxide, or cellulose immobilized onto a flat, inert carrier sheet. A liquid phase consisting of the solution is drawn up the plate via capillary action. The separation is based on the polarity of the components of the compound in question. (Dhahir, 1972, Kasture, 2008,) TLC is a simple, quick, and inexpensive procedure that gives the chemist a quick answer as to how many components are in a mixture. Ibogaine, classified under the ibogaterpenoids is a

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test and type of sample solvent (e.g. it contains high quantity of water) are also important disadvantages.

**Drug information:** (Bouso, 2012) (20, 21, 22, 23)

1) Structure:

![Ibogaine structure](image)

2) Solubility: Ibogaine is soluble in ethanol, ether, chloroform, acetone and benzene, but it is practically insoluble in water.

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4) Pka Value: 8.97

5) Melting Point: 1530C

6) Maximum absorbance wavelength: >290 nm.

7) Category: Hallucinogen and Stimulant.

**Materials and Methods**

**Collection and preparation of plant materials** (Jenks, 2002), (Dahir, 1972), (Alper, 2001), (Alliston, 1971)

The *Tabernanthe iboga* between root or root bark becomes available, consider that root contains about one-third bark, and only the bark contains the alkaloids. The root or root barks were washed thoroughly in running water to remove the soil and dust particles, finally washed with distilled water. The washed barks were dried and cut into very small pieces. These materials were stored in air tight polythene bags until use.


**i). Extraction:** 10gm of iboga root or root bark powder dissolve in 50ml of 0.5% of acetic acid solution. This is conveniently made by diluting 5ml of distilled vinegar containing 5% of acetic acid with 40ml of water. The extraction done at room temperature, since heating does not improve the ultimate yield. Powder & solvents are mixed every 15 min for 2h. The extract stays on overnight. The extract has been extracted nine times by which time there is approximately the original alkaloid- little enough to justify discarding the root. Extract should be made basic with ammonia solution, addition of 3.6ml of 5% ammonia with stirring the mixture. A gray or Brown milkiness should appear. When the amount of ammonia is adequate, the pH should be maintained at 9 & add ammonia solution to be filtered, mixture should no cause precipitation. This mixture filtered though Whatman filter paper, that material wash with water to rinse the alkaloids. The precipitate is carefully died at 500C on hot plate. The total alkaloid should be spread out with Spatula. The solid dies it becomes dark brown.

**ii). Separation of Ibogaine:** A portion (2gm) of dark brown powder in flask, add a potion of acetone in same flask, stirred the flask & attached to the distillation assembly. If the distillation is unavailable it is possible to evaporate all of the acetone. The dark syrup residue is dissolving in water & filtered through filter paper. Filtrate solution made basic with 12 ml of 5% of ammonia. Filter though filter paper yellow solid residue dries into light.

**iii). Purification:** Purification of ibogaine uses recrystallization of ibogaine residue from 95% ethyl alcohol.

C). **For TLC Method:** (Jenks, 2002), (Dahir, 1972), (Kasture, 2008), (Alliston, 1971)

To prepare a thin layer chromatogram, a pinch of root or bark powder is stirred with about one milliliter of a solvent like ethyl acetate, petroleum ether, chloroform, etc. A sample of the solution is applied using a thin (<1 mm) glass capillary to one end of a 2 cm by 8 cm piece of silica gel TLC sheet, and the sheet is placed in a sealed bottle with a little solvent, such various solvent system uses at the bottom, with the extract spot at the lower end. Once the solvents has climbed to the top of the sheet, the sheet is removed, dried, and the components on it made visible by using locating agent. By this means the ibogaine can be distinguished from the other iboga alkaloids and its relative concentration estimated. The various TLC methods were developed, in all methods uses different solvent system as following –
Table 1. Different solvent system and Locating Agent.

<table>
<thead>
<tr>
<th>No.</th>
<th>Solvent System</th>
<th>Locating Agent</th>
<th>Rf Value</th>
</tr>
</thead>
</table>
B). 5% w/v 4-dimethylaminobenzaldehyde in Methanol-HCl (1:1) | 0.61 ± 0.02  |
| 2.  | Cyclohexane : Ethyl acetate (1:1) | A). Dragendorff’s reagent  
B). DTNB –Ellman’s reagent | 0.49 ± 0.04  |
B). Pot. Permanganate reagent | 0.57 ± 0.09  |
B). Pot. Permanganate reagent | 0.45 ± 0.02  |
B). Uv-light | 0.16 ± 0.07  |

Fig. 1. Solvent 1:1 ethyl acetate & petroleum ether for total alkaloids (TA)  
(Spot A- Stating point, Spot B- Ending point of Ibogaine in plant extract).

Results and Discussion

In TLC has been included under both adsorption and partition chromatographic. TLC is a simple, quick, and inexpensive procedure that gives the chemist a quick answer as to how many components are in a plant extract. In various solvent system results gives Morpholine : Toluene (1:9), Cyclohexane : Ethyl acetate (1:1), Methanol : Benzene (20:80), Methanol : Chloroform (10:90), and Ethyl acetate : Petroleum Ether (1:1) Rf Value are 0.61 ± 0.02, 0.49 ± 0.04, 0.57 ± 0.09, 0.45 ± 0.02 and 0.16 ± 0.07 respectively. Separation may result due to adsorption or partition or by both phenomenon depending upon the nature of adsorbents used on plates and solvent systems for developments.

Conclusion

In case of Iboga root sample in chemical constituent are not very know even constituents know variability of naturally occurring samples may very considerable. Furthermore, the TLC is also suitable for rapid and simple authentication and comparison of the suitable difference among samples with identical plant resource but different geographic locations. Colour based test were standardized and screened for samples from plant extract.

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