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Phytochemical Analyses of *Banisteriopsis Caapi* and *Psychotria Viridis*†

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Abstract—A total of 32 *Banisteriopsis caapi* samples and 36 samples of *Psychotria viridis* were carefully collected from different plants on the same day from 22 sites throughout Brazil for phytochemical analyses. A broad range in alkaloid distribution was observed in both sample sets. All *B. caapi* samples had detectable amounts of harmine, harmaline and tetrahydroharmine (THH), while some samples of *P. viridis* had little or no detectable levels of *N,N*-dimethyltryptamine (DMT). Leaves of *P. viridis* were also collected from one plant and analyzed for DMT throughout a 24-hour cycle.

Keywords—*ayahuasca*, circadian, *hoasca*, *yajé*, *mariri*, *caupuri*, *tucunacá*

Decoctions of a traditional South American sacrament are made from the pounded woody portions of the liana *Banisteriopsis caapi*, and typically brewed with the carefully washed leaves of the shrub *Psychotria viridis*. Use of these plants and the resulting decoctions form the core spiritual doctrines of many religious groups in this region of the world. Althoughit has been known by many other names, and many variations of this "tea" have been described (Ott 1994), the most common identifying feature is the presence of harmala alkaloids from *B. caapi* (Schultes 1982). Although these harmala alkaloids are not particularly psychoactive on their own, they can facilitate the activity of *N,N*-dimethyltryptamine (DMT) from *P. viridis* by the inhibition of the enzyme monoamine oxidase (MAO) in the liver and central nervous system (Ott 1994; McKenna, Towers & Abbot 1984; Holmstedt & Lindgren 1967).

The purpose of this article is to provide a phytochemical overview of the alkaloid content in *B. caapi* and *P. viridis*, in order to have a better understanding of these useful plants. There are much deeper issues concerning the relationship between these plants, their chemical composition and subsequent religious revelations that are well beyond the scope of this article.

MATERIALS AND METHODS

Plant Material

Samples of *B. caapi* (known locally as *mariri*) and *P. viridis* (known as *chacrona*) were collected from 22 sites throughout Brazil between 6:00 and 9:00 AM on October 7, 1995 by experienced members of the União do Vegetal (UDV), a religious group that uses these plants. The collection area was within 23- 8° S and 67-38° W. All plant samples were carefully dried at ambient temperatures (25 to 35°C) over subsequent days before storage in paper envelopes. All samples were carefully stored in a dark, dry place at room temperature until analysis. In addition, leaf samples of *P. viridis* were collected from opposing stems on the same bush over a 24-hour period, in order to examine possible circadian variations in DMT content. Most plant samples were obtained from garden specimens that have been maintained near UDV temples for up to 20 years. A conscientious attempt was made to select samples from a wide variety of specimens, to insure a representative sample of "typical" ingredients for the production of ***hoasca***.

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TABLE 1
Overall Alkaloid Composition for *B. Caapi* (i.e., Harmine, Harmaline and THH) and *P. Viridis* (DMT) Samples, Presented as Milligram of Alkaloid per Gram of Dried Plant Material

	Harmine	Harmaline	THH	DMT
Min.	0.31 mg/g	0.03 mg/g	0.05 mg/g	0.00 mg/g
Max.	8.43	0.83	2.94	17.75
Mean	4.83	0.46	1.00	7.50
±S.D.	2.06	0.19	0.79	5.01

Analytical Methodology

High pressure liquid chromatography (HPLC) with fluorescence detection was used to analyze all samples in a method previously described (Callaway et al. 1996). Briefly, the chromtographic column was packed with C-8 material and the mobile phase was 20% methanol, 20% acetonitrile and 60% 0.1 M ammonium acetate buffer, adjusted to pH 6.9 with acetic acid. Individual signals in the chromatogram wère verified according to retention time and molecular weight by liquid chromatographic mass spectrometry (LC-MS), using a VG thermospray-plasma probe coupled to a VG Trio-2 quadropole mass spectrometer.

Sample Preparation

Banisteriopsis caapi. Approximately 30 g from each sample of *B. caapi* was milled and then dried in the dark at room temperature for about one week. For analysis, 100 mg of each milled sample was sonicated in 2 ml of methanol for 10 minutes at room temperature and then soaked for 24 hours in the same solution, in the dark at room temperature. The extract was centrifuged at 300 g for five minutes. An aliquot of the supernatant from each sample was diluted by 100 fold in the HPLC mobile phase and injected directly for the analysis of harmine, harmaline and tetrahydroharmine (THH).

Psychotria viridis. Whole leaves from each sample of *P. viridis* were weighted (0.60-2.60 g) and homogenized in 67% methanol, 11% acetonitrile and 22% 0.1M ammonium acetate at pH 8.0. The mixture was otherwise prepared and analyzed for *N,N*-dimethyltryptamine (DMT) by HPLC.

RESULTS AND DISCUSSION

Banisteriopsis Caapi

According to experienced individuals in the UDV, two different varieties of *B. caapi* are recognized; *mariri caupuri* and *mariri tucunacá*. The *caupuri* grows near the equator while the *tucunacá* thrives in the cooler climes of southern Brazil. Botanically speaking, they are considered to be the same species. These two varieties are morphologically distinct and impart different sensations to the body and mind from the resulting teas. From the analytical results, a slight trend was observed towards higher levels of all harmala alkaloids in the *caupuri* samples, but this trend was not statistically significant. The subjective difference in teas from these two varieties is still a mystery.

In the *B. caapi* sample set (N = 33), a broad range of alkaloid profiles and concentrations were observed for the three most prominent components: harmine, harmaline and THH (see Table 1 and Figures 1-3). A few minor signals were observed in most samples, but the major components and distributions were always harmine, which was greater than THH and, to a much lesser extent, harmaline. Proportional levels of harmine and harmaline displayed a surprisingly uniformity in all samples, as illustrated in Figures 1 and 2, where harmine was consistently present at a level of approximately one magnitude over harmaline (ca. 10:1). Levels of THH showed a more variable distribution in these samples and had no clear relationship with the other two harmala alkaloids (Figure 3). Two samples were consistently low in all harmala alkloids (Figures 1-3); these specimens were from older plants, which were known to be seven and nine years old.

Psychotria Viridis

In the *P. viridis* (known locally as *chacrona*) sample set (N = 37), a broad range of alkaloid concentrations was observed for DMT (Table 1 and Figure 4). It was surprising to see absolutely no DMT in one sample and less than 0.60 mg/g in eight samples. Although some species of *Psychotria* do not contain detectable levels of DMT (Verotta et al. 1999), it is quite certain that the species analyzed in the present study was, indeed, *P. viridis*.

P. viridis leaf samples were also collected from a single plant, at three hour intervals, for the better part of a 24-hour period (from midnight until 9 p.m. of the same day). These samples were carefully selected from the same nodal order from the same the plant, and considered to be identical for the purpose of phytochemical circadian analysis. In Figure 5 it can be seen that *P. viridis* DMT levels increased until about 6 a.m., with a decline in DMT from 6 to 9 a.m., then gradually increasing again from 9 a.m. to a zenith at 6 p.m., and sharply decline after 6 p.m. to basal levels again at 9 p.m. It is not known why this fluctuation occurs, although circadian variations have already been

FIGURE 1
Variations in *Banisteriopsis Caapi* Harmine Levels in mg/g

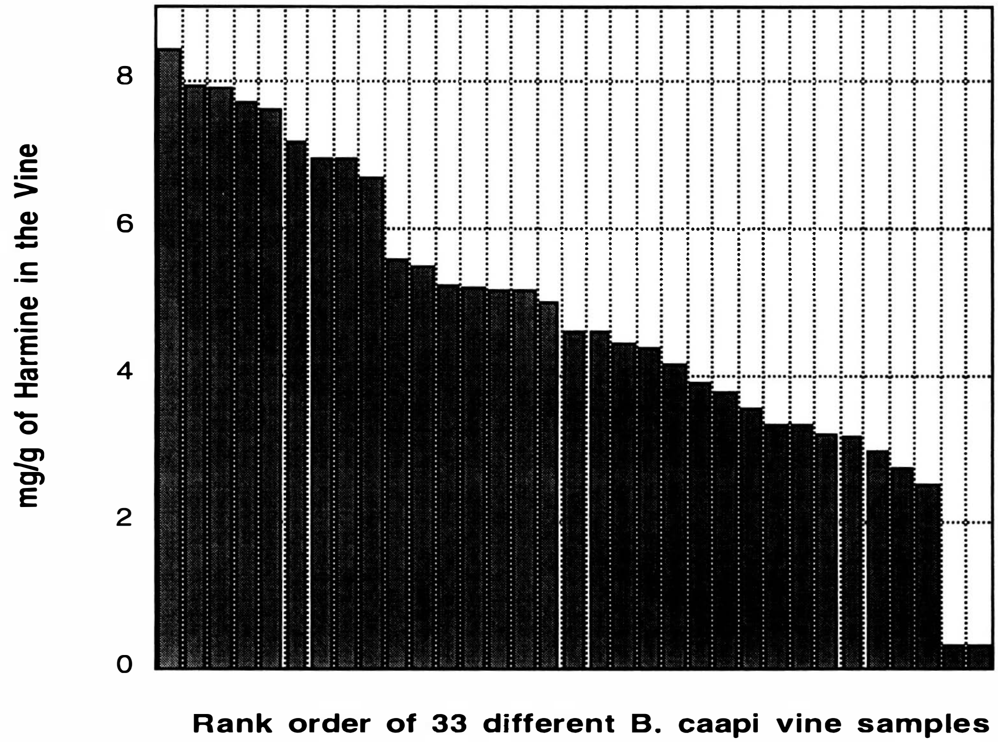


FIGURE 2
Variations in *Banisteriopsis Caapi* Harmaline Levels in mg/g

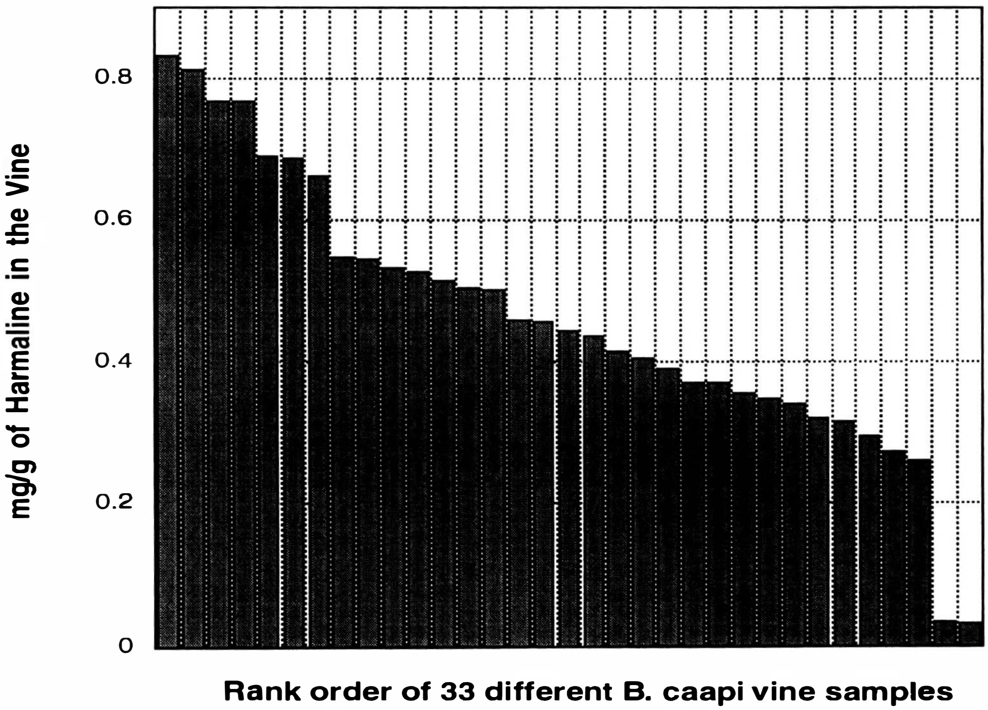


FIGURE 3
Variations in *Banisteriopsis Caapi* THH Levels in mg/g

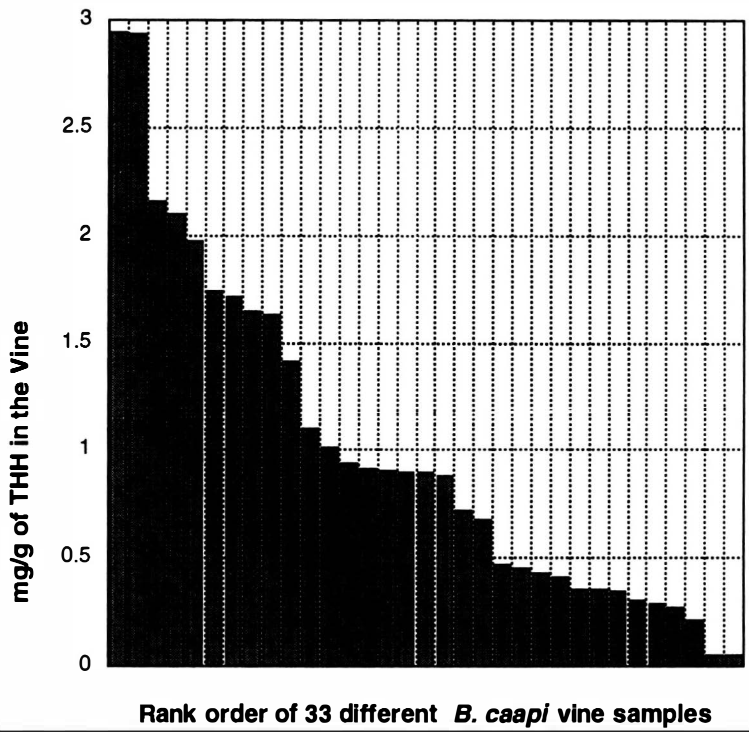


FIGURE 4
Variations of DMT Levels in Leaves of *Psychotria Viridis* in mg/g

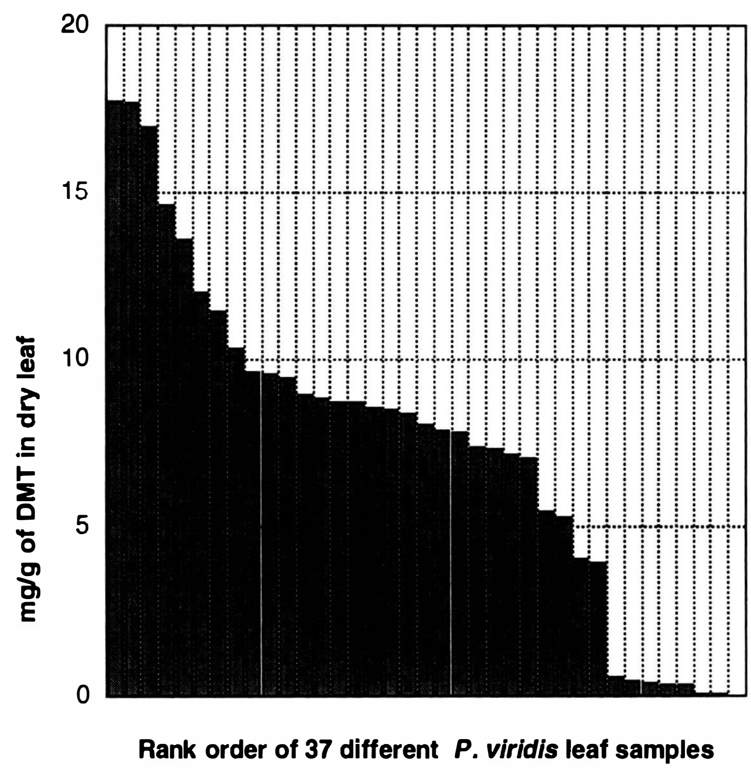
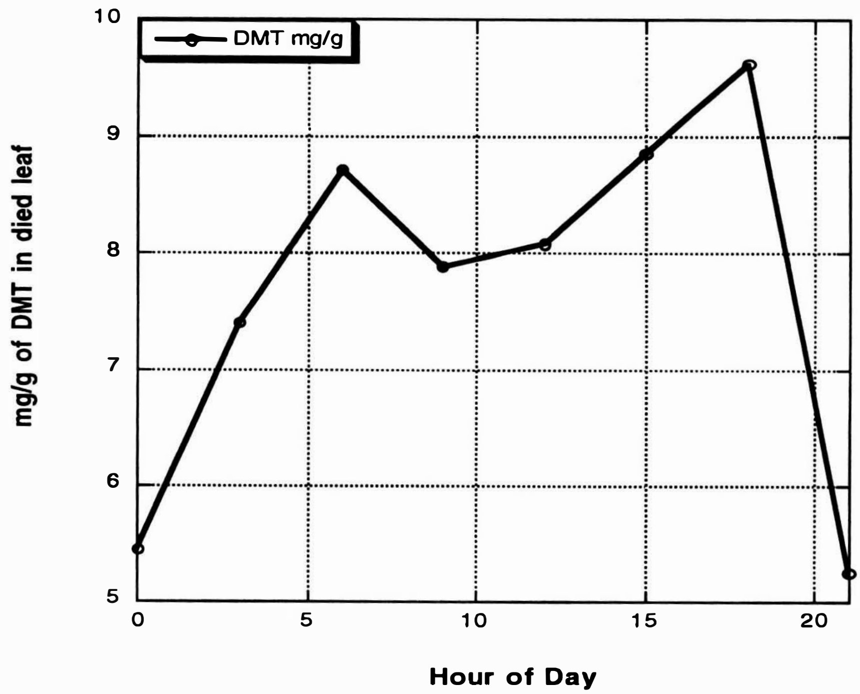


FIGURE 5
Circadian Fluctuations of DMT Levels within the Leaves of a Single Living Specimen of *Psychotria Viridis* in mg/g over 21 Hours



noted for alkaloids in other plants (see Itenov, Mølgaard & Nyman 1999 and references therein). From Figure 5, it appears that a depression in DMT production begins and ends during the hottest times of day, and perhaps *P. viridis* produces DMT to protect itself from solar radiation. Another possibility is that *P. viridis* produces DMT to actually absorb solar radiation for purposes other than self-preservation. In either event, this is probably no accident, as the narrow range of UV-B radiation in the ultraviolet spectrum is between 315 and 280 nm, which also happens to be the range of greatest UV absorbance by DMT.

CONCLUSIONS

The English botanist Richard Spruce (1873) encountered the use of *B. caapi* decoctions while exploring tributaries of the Amazon River in 1851, which may or may not have contained the leaves of *P. viridis*. It is quite certain that he was aware of the most important component of the brew, which he correctly attributed to the vine, *B. caapi*. In 1853, Richard Spruce (1873: 186) wrote of hope for future investigations into the substance he referred to as *caapi* or *aya-huas-ca*, which must have been the vine B. *caapi* and the resulting brew, respectively: "Some traveller[s] who may follow my steps, with greater resources at his command, will, it is to be hoped, be able to bring away materials adequate for the complete analysis of this curious plant."

In fact, samples from Spruce's expedition were brought back and preserved at Kew Garderns in England, and subsequently analyzed by Bo Holmstedt and Jan-Erik Lindgren at the Karolinska Institute in Stockholm, Sweden with modern chromatographic methods in 1968 (Schultes et al. 1969). Soon afterwards, subsequent investigations focused on contemporary plant specimens and resulting decoctions (Riba, Saa & Caseido 1972; Rivier & Lindgren 1972; Rüf 1972; Ghosal, Mazumber & Bhattahcharaya 1971), further demonstrating the extant use of *B. caapi* and other plant additives in this native religious practice.

The present article presents the results of the largest phytochemical survey to date of these two plant species, *B. caapi* and *P. viridi*. The ability to coordinate and execute such a broad, regional collection on the same day stands as an example of dedication and group unity that is typical of UDV members. It is unlikely that these analyses are complete, or that everything can actually be known about these plants and their derivatives, but it is hoped that these results would have satisfied the academic curiosity of nineteenth century explorer Richard Spruce. These results may allow for additional insights into this ancient technology, and perhaps a better understanding of its use and application in modern cultures.

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