Prehistoric psychotropic consumption in Andean Chilean mummies

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Hallucinogenic plants are often regarded as the main source of psychoactive drugs in antiquity to reach deep altered states of consciousness^{1,2}. Many researchers believe this was particularly true during the Tiwanaku empire expansion, circa (500-1000 A.D.), along the Atacama Desert of Chile. Highly decorated snuffing tablets and tubes are often found as grave goods during this period^{3,4,5,6,7,8}. Until now the type of drugs consumed in this paraphernalia has been unclear. From the modern city of Arica, naturally mummified human bodies with abundant hair provided a unique opportunity to test for hallucinogenic plants consumed in Andean prehistory. Analysis by gas chromatography and mass spectrometry demonstrated the presence of harmine. The Banisteriopsis vine, commonly called Ayahuasca, was the probable source. This is the first confirmed evidence of psychoactive plant consumption in pre-Hispanic Andean populations along the Atacama coastal region. Of the 32 mummy hair samples analyzed 3 males tested positive for harmine. This alkaloid aids in the catalysis and synergic effects of powerful hallucinogenic drugs. The consumption of harmine was likely related to medicinal practices and not exclusively ingested by shamans. Another important

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aspect of this evidence is that *Banisteriopsis* is an Amazon plant. It does not grow in the Atacama coastal region. Thus, our findings reveal extensive plant trade networks in antiquity between the coast, desert, highlands, and Amazon basin. The excellent preservation of human organic specimens, the use of gas chromatography and mass spectrometry allowed us to map and demonstrate the consumption of psychoactive compound plants in Andean prehistory. In addition, our findings open the door for future studies to debate the consumption and social role of ancient psychoactive and hallucinogenic plants.

Psychoactive alkaloids accumulate in hair and other body tissues, therefore if ancient Andeans consumed hallucinogenic plants their hair should test positive. To test the hypothesis that ancient South American people consumed hallucinogenic plants we used gas chromatography and mass spectrometry to detect the presence of harmalinic and triptaminic alkaloid molecules in the hair of thirty two pre-Hispanic mummies from Arica, Chile. Radiocarbon dated mummies from the Tiwanaku Empire Middle Period (500–1000 A.D.) are housed at the Universidad de Tarapacá Archaeology Museum. Mummies with hair were selected and grouped according to the presence or absence of snuffing tablets and lavish grave goods indicating their social differentiation (Supplementary Figs. 1-3). Three modern local hair samples (child, 12 years old; young adult, 24 years of age, and adult, 45 years old) were used as controls.

The Tiwanaku state rose in the Lake Titicaca area, what is now Bolivia, and expanded its influence, into what is now Chile, by religious control or militaristic activities^{3,4}. The lavish and intricately decorated textiles, ceramic and stone iconography depicting trophy heads, plants and supernatural beings, indicates a complex interplay of state control and religion³. Highland rain has destroyed most organic materials in the heartland of Tiwanaku, however, mummies and other organic materials are ubiquitous in the Tiwanaku periphery, particularly in the Atacama Desert.

During the Tiwanaku period we see the highest evidence of highly decorated snuffing implements. In northern Chile they were commonly deposited as grave goods, particularly in San Pedro de Atacama^{5,7,8} and to a lesser extent in the Arica valley⁹. There is a considerable amount of visual and tangential evidence for the use of narcotic plants in the past^{8,9}. However, actual proof of ingestion of hallucinogenic plants in the area remained elusive. Studies from Chiu-Chiu mummies dated to 100 A.D. tested negative for hallucinogenic plant consumption¹⁰ (Figs. 1 and 2).

Figure 1| Harmine standard with matrix effect. The upper graph shows the mass spectrum analysis and the lower graph the gas chromatograph analysis.

Figure 2 | Graphic representation of the three mummies that tested positive for the consumption of harmine. (a) AZ-140 T75, (b) AZ141 T30, (c) AZ141 T33.

Of the 32 mummy hair samples analyzed none tested positive for triptaminic alkaloids and 3 males tested positive for harmine (AZ-140 T75, 35-40 years old; AZ141 T30, 1-2 years old; and AZ141 T33, 35-40 years old). The three positive individuals belonged to the group with prestigious grave goods. This is the first proof of actual consumption of psychoactive substances in pre-Hispanic Andean populations along the Atacama coastal region. The *Banisteriopsis* vine species, the source of the harmine, contains three main active alkaloids: harmine, harmaline and tetrahydroharmine. *Banisteriopsis* can be used for medicinal or psychotropic purposes depending on dose and concentration ingested². Also, the plant is often mixed with *Psychotria viridis*, a plant containing the hallucinogenic compound DMT. When DMT is taken orally it is inactivated by the stomach enzyme aminomonooxidase (MAO). However, harmine inhibits the action of this MAO enzyme promoting the absorption of the hallucinogenic mixture².

What other drugs were consumed? Previous mummy hair analysis showed the presence of benzoylecgonine (BZE) metabolites providing evidence for consumption of coca leaves, or *Erythroxylum coca*, a non hallucinogenic plant, in northern Chile beginning 350 – 250 B.C.¹¹ Analysis of the snuffing tablets' powder from San Pedro de Atacama shows the presence of *Anadenanthera colubrina* var. cebil⁶, a plant rich in triptaminic alkaloid such as dimethyltriptamine or DMT¹². Finally, as far as we know, we show the first evidence of *Banisteriopsis* consumption^{10.}

The three plants, *Erythroxylum coca, Anadenanthera* and *Banisteriopsis* are recurrent in the ethnographic milieu and religious activities. All are nonexistent in the Atacama Desert; they are predominantly of selvatic origin. *Erythroxylum coca* grows at 1,000 to 2,000 meters above sea level on the western slopes of the Andes. *Anadenanthera* grows in tropical savannah like environments, and the *Banisteriopsis* vine is widely prepared and consumed in Amazon areas today.

Our data reveals extensive trade networks in antiquity; plant import networks reaching from Arica, to as far away as the Amazon region. This is particularly true for *Banisteriopsis* empirical ingestions. Contrary to the San Pedro de Atacama analyses of snuffing tablet powder⁶, we did not find presence of *Anadenanthera* in the Arica mummies.

Finally, our data validates archaeological propositions of drug consumption in antiquity. We proved, for the first time, that Ancient Andeans consumed psychoactive compounds related to the neurochemistry of hallucinogenic processes. Interestingly enough, the three positive individuals belonged to the elite group. However, the positive harmine result on the infant was a surprising result. This opens the question whether the plants were used as medicine to alleviate suffering as well as for their hallucinogenic properties. Future studies need to decipher if the child was given the hallucinogen as medicine or more likely acquired the hallucinogenic drug through his mother's milk.

Methods

We followed a similar procedure as reported previously^{10,13}, with minor variations. A lock of hair was cut directly from 32 mummies. Two hundred milligrams of hair were cleaned with a soft brush to remove debris, weighted and treated with 2 ml of 0.1M hydrochloric acid solution for 24 hours at 37°C. The solution was centrifuged at 4,000 rpm for 10 minutes. The aqueous acid solution was neutralized with 0.1N NaOH and adjusted with a phosphate buffer, pH 7. The aqueous analites were extracted and preconcentrated using a 3ml Bond Elut C18 (SPE) cartridge, then evaporated to dryness in a nitrogen environment. The residue was dissolved in 40 micro liters of acetonitrile. Two micro liters of analite were injected into a Variant 3800 CG gas chromatograph with a SE-30 30m ID 0.53 mm 1.2 μ film column, and the oven temperature ramped from 80°C for 2 min to 280 °C. The alkaloids were confirmed and run in tandem on a Variant Saturn 2000 GC/MS mass spectrometer. The lower detection value of 10 mg/l was employed to differentiate between positive and negative results.

Standards used: **Triptaminic**: t_R 9.717, peaks 130 m/z,160 m/z, 103 m/z, and 77 m/z. **5-metoxi-N,N-dimetiltriptamine:** t_R 13.968, peaks 58 m/z, 219 m/z, 160 m/z, 130 m/z, 103 m/z. **Harmina**: t_R 16.767, peaks 212 m/z, 197 m/z; 169 m/z.

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