Bringing Ayahuasca to the Clinical Research Laboratory†

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Abstract—Since the winter of 1999, the authors and their research team have been conducting clinical studies involving the administration of ayahuasca to healthy volunteers. The rationale for conducting this kind of research is twofold. First, the growing interest of many individuals for traditional indigenous practices involving the ingestion of natural psychotropic drugs such as ayahuasca demands the systematic study of their pharmacological profiles in the target species, i.e., human beings. The complex nature of ayahuasca brews combining a large number of pharmacologically active compounds requires that research be carried out to establish the safety and overall pharmacological profile of these products. Second, the authors believe that the study of psychedelics in general calls for renewed attention. Although the molecular and electrophysiological level effects of these drugs are relatively well characterized, current knowledge of the mechanisms by which these compounds modify the higher order cognitive processes in the way they do is still incomplete, to say the least. The present article describes the development of the research effort carried out at the Autonomous University of Barcelona, commenting on several methodological aspects and reviewing the basic clinical findings. It also describes the research currently underway in our laboratory, and briefly comments on two new studies we plan to undertake in order to further our knowledge of the pharmacology of ayahuasca.

Keywords—ayahuasca, clinical pharmacology, DMT, human, psychedelics, review

In 1994, the authors had the unusual opportunity of directly witnessing the arrival of a religious tradition to Spain which involved the use of what was at that time a relatively obscure psychotropic plant concoction called ayahuasca. This tradition was being imported by a very motivated small nucleus of people involved in self-knowledge and spiritual practices who were experimenting with a peculiar mind-modifying tea in the context of a Brazilian religion known as the Santo Daime. Sessions, or "works," as participants called them, were held twice a month and interested outsiders were able to attend. Most participants seriously considered their commitment to the group as a path to self-discovery and personal betterment and had no notion of engaging in "drug taking" or any kind of illicit practices. To the external observer, nothing remarkable occurred during these sessions. After ingesting a small volume of a brownish tea, participants would sit, separated by sex, in rows of chairs where they would remain for most of the session in a kind of slumber that would force them to close their eyes and yawn from time to time. This was only interrupted by periods of chanting and dancing. Sporadically, someone would become upset and start sobbing or

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leave the room in order to lie down, or they would be accompanied to the bathroom if they felt nauseated. These rather quiet introspective sessions were watched over by experienced participants. Several hours later, after two or three intakes of the tea, they would end over a snack with everyone vividly discussing what appeared to be remarkable psychotropic effects with powerful visions and overwhelming emotions.

The fact that these ayahuasca sessions did not apparently interfere with the participants' everyday lives and social integration made this pattern of psychotropic drug use a remarkable social phenomenon. Furthermore, the relatively short-lived and a priori controllable nature of the subjective effects suggested that the drug could be amenable to study in a clinical research setting. Virtually no data were available on the effects of ayahuasca in humans, but that same year Strassman's papers on DMT appeared (Strassman & Qualls 1994; Strassman et al. 1994), providing very valuable information on the pharmacology of what had been described as the main psychotropic ingredient in ayahuasca brews. We thus considered that scientific inquiry into the effects on humans would be a stimulating challenge that could provide useful data to both users and the research community.

AYAHUASCA, AN UNUSUAL CHALLENGE TO THE PHARMACOLOGIST

When reviewing the botanical and chemical bibliography on the "ayahuascas," one is immediately confronted with the degree of complexity that these brews can attain. While the *Banisteriopsis caapi* liana appears to be the ubiquitous ingredient, as already described in the nineteenth century by Spruce (1908), the number of additional plants that can be used as additives to the tea is very extensive. Ott (1993) has listed up to 90 different species belonging to 38 families. As is well known, *B. caapi* contributes three major alkaloids to the tea, namely harmine, tetrahydroharmine (THH) and harmaline and, as in the samples we have analyzed, also harmol and harmalol (see below). Among the admixtures identified by Ott (1993) are found powerful psychotropic plants such as the nicotine-containing *Nicotiana tabacum* and *Nicotiana rustica*, *Erythroxylum coca* containing ecgonine alkaloids such as cocaine, *Ilex guayusa* and *Paullinia yoco* (both rich in caffeine), the solanaceous *Brugmansia sauveolens* and *Brugmansia insignis* which contain tropane alkaloids like atropine and scopolamine, and many others. However, the most frequently employed additives throughout the Amazon Basin are the rubiaceous *Psychotria viridis* and the malpighiaceous *Diplopterys cabrerana*, which lend the tea the visionary compound *N,N*-dimethyltryptamine or DMT.

In view of the above, when planning to study ayahuasca, it was important to establish which ayahuasca was be evaluated. Happily, the Santo Daime in Brazil agreed to support this research project by generously donating the ayahuasca samples from which the lyophilizate that was later administered in the clinical trials was obtained. Thus it was possible to focus on brews that were of the same geographical origin, were all elaborated following the same methodology and most importantly, always contained the same plant ingredients, i.e., exclusively *B. caapi* and *P. viridis*.

So far, two batches of ayahuasca from the Santo Daime have been analyzed and freeze-dried in this study. One was "Daime" and the other was "*Daime doblado*" or "double Daime" (which differ in strength, the latter having higher alkaloid concentrations). The present report focuses on the composition of the latter. The "double Daime" batch was the source of the ayahuasca used throughout the pilot and final double-blind studies described below. It had a volume of 9.6 liters and yielded 611 grams of powder after the freeze-drying process. One gram of freeze-dried material contained 8.33 mg DMT, 14.13 mg harmine, 0.96 mg harmaline and 11.36 mg THH, which corresponded to the following alkaloid concentrations in the original tea: DMT 0.53 mg/ml, harmine 0.90 mg/ml, harmaline 0.06 mg/ml and THH 0.72 mg/ml. In posterior analyses it was found that the lyophilizate also contained small amounts of harmol (0.28 mg/g) and harmalol (0.08 mg/g), corresponding to concentrations in the tea of 0.02 mg/ml and 0.005 mg/ml, respectively.

Even the study of brews prepared exclusively from *B. caapi* and *P. viridis* would thus involve the interplay of DMT with at least three major and two minor b-carbolines for which MAO inhibition and serotonin reuptake inhibition had been described (Buckholtz & Boggan 1977a, b). As will be discussed below, in the studies undertaken to date the authors have addressed the pharmacology of complete brews in the form of lyophilizates. In future studies, when we have adequately characterized the pharmacological profile of the ayahuasca lyophilizates, these standard preparations could be used as ideal "positive controls" against which the effects of the individual alkaloids or combinations thereof could be compared.

FINDING AN OPTIMAL PHARMACEUTICAL FORM

Before we could start the clinical phase of the project, we spent two years in the planning stage, writing the study protocol, requesting the necessary permits and contacting potential volunteers. A considerable part of this period was also devoted to deciding whether to use "natural" ayahuasca or rather combinations of purified active compounds. Once the former option had been chosen, we had to obtain a pharmaceutical form of ayahuasca that could be administered in accordance with certain methodological requirements. The decision to study natural ayahuasca was based on the fact that this is the product to which users

were normally exposed. However, working with a natural extract like ayahuasca poses a series of problems such as progressive alkaloid degradation with time or the difficulty of establishing accurate dosages. Ideally, variations in alkaloid contents in the final pharmaceutical form should be kept to a minimum and dosing units should be easily stored and administered, and always contain the same amount of product. We were also confronted with the masking problem. Clinical research methodology requires that expectancy on the part of the study participant and on the part of the researcher be the same across experimental sessions. This is related to the fact that expectancy (i.e., knowledge about what one is going to receive or to administer) is believed to influence the effects reported by a volunteer and the experimenter's attention or motivation during the experimental session, respectively. Thus, when we finally decided to go for the completely natural product it became obvious that in its usual liquid form it would easily degrade and with its characteristic taste and smell masking would be virtually impossible. So it was decided that ayahuasca would be administered as a standardized encapsulated lyophilizate. Thus, although we plan to study combinations of pure DMT plus pure β-carbolines in the future in order to assess specific drug-drug interactions, in the research conducted to date we have evaluated the human pharmacology of ayahuasca as a whole. While some findings might not have changed much from one approach to the other, we believe that other aspects of the clinical picture certainly would have. A natural extract such as ayahuasca contains a myriad of substances. Some are pharmacologically active, but inert plant material is likely to influence at least the bioavailability and gastrointestinal tolerability of the tea. The extensive literature on personal experiences with the tea describes vomiting as a very common event. In some rituals and also in certain nonreligious contexts, this behavior is encouraged as it is considered a form of purification or an acceptable form of somatization, an essential part of the ayahuasca session. In contrast, self-reports on the effects of DMT plus harmine combinations describe nausea or vomiting as rare (Ott 1999). Thus, although not ideal for the precise study of drug-drug interactions, the ingestion of the natural tea may at least lead to a different experience in terms of somatic effects, nausea and vomiting, compared with the simple combination of pure compounds in a capsule. The use of the lyophilizate, where only the original water is lost, seemed to be a reasonable compromise allowing for a classical clinical trial design and at the same time being closer to the original tea than pure alkaloid combinations.

Lyophilization, or freeze-drying, is a method of removing water from a substance or material at low temperatures. In this case, ayahuasca is first frozen in an air-tight chamber, after which the frozen material is kept under high vacuum for around 24 hours. Under these conditions, the water in the samples changes directly from ice to vapor, leaving behind a porous solid. The fact that the whole process takes place at a low temperature prevents active compounds from being destroyed by heat, and the remaining material can be analyzed to accurately establish the alkaloid concentrations and encapsulated. Once made, the capsules can be tested for uniformity of weight and content. The use of capsules also allows perfect masking and ayahuasca effects can be compared with those of placebos or positive controls (i.e., capsules containing lactose, for example, or an active compound, respectively).

Despite these advantages, working with the ayahuasca lyophilizate poses a difficulty: as the product is extremely dry, it acts as a sponge to humidity. It attracts any moisture in the atmosphere and becomes a sticky paste which can not be redissolved or freeze-dried over again. Indeed this occurred in front of our very eyes on removing the first samples of ayahuasca from the freeze-drying chamber. The lyophilizate must consequently be stored under vacuum or, alternatively, in a dry atmosphere. Consequently, in subsequent freeze-drying rounds the bulk freeze-dried ayahuasca was first stored under vacuum and the encapsulation process was later conducted under dry nitrogen.

ESTABLISHING THE OBJECTIVES OF THE CLINICAL RESEARCH PROJECT

As mentioned above, this project aimed to obtain basic information on the human pharmacology of a rather standard form of ayahuasca. Specifically, it would study teas obtained exclusively from *Banisteriopsis caapi* and *Psychotria viridis*. We decided to focus on acute effects following a single dose administration and that the experimental part should follow the methodology of a typical controlled clinical trial, i.e., incorporate a placebo and the use of single-blind or double-blind randomized designs. We also decided at this stage not to address any possible adverse psychopathological effects derived from repeated consumption, or study any hypothetical therapeutic potential of ayahuasca.

The design of the clinical trial was underway by late 1996, at a time when virtually all data available on ayahuasca dealt only with its botany and chemistry. The only information on its effects in humans were self-administration reports and no data were available regarding its acute administration to healthy humans in controlled studies. Although as yet unpublished at that time, James Callaway, a researcher at the University of Kuopio, generously shared with us data he and his colleagues had obtained in the Hoasca Project, and this, together with our own field observations, helped to give us an approximate idea of the time course of effects following the administration of a single dose. Furthermore, blood drawing points were based in part on the pharmacokinetic parameters that Callaway and colleagues had calculated and later published (Callaway et al. 1999).

We drew up the following specific objectives:

1. Assess the general tolerability of ayahuasca, i.e., the volunteers' vital signs after the drug administration, and any event, either physical or psychological, regarded as unpleasant by the participant. Evaluate the impact of drug administration on standard hematological and biochemical analytical parameters.
3. Describe the pharmacokinetics of DMT and the β-carbolines present in ayahuasca after the oral administration of various doses of the preparation.
4. Measure *in vivo* the inhibition of MAO provoked by acute ayahuasca administration.
5. Measure the nature and time course of subjective effects elicited by acute ayahuasca administration.
6. Assess the effects of ayahuasca on the central nervous system (CNS) by means of quantitative pharmaco-electroencephalography (q-EEG).
7. Conduct an exploratory analysis of the brain cortical regions responsible for the observed q-EEG effects by means of low resolution electromagnetic tomography (LORETA).
8. Measure the effects of ayahuasca on sensory and sensorimotor gating (theoretical constructs dealing with the capacity of the brain to filter out irrelevant sensory information).

To accomplish these objectives it was necessary to examine the goodness of the main tool used to assess subjective effects, and to validate the analytical technique used to determine the plasma concentrations of the ayahuasca alkaloids. Thus, we also addressed the following:

1. The translation into Spanish of the Hallucinogen Rating Scale (HRS; Strassman et al. 1994), a then recently developed self-report instrument designed to evaluate the subjective effects elicited by psychedelics. The sensitivity of the instrument to psychedelics other than intravenous DMT had still to be tested and certain psychometric properties (i.e., its reliability and construct validity) had not been assessed either for the original American version or any of its translations.
2. The development and validation of an analytical methodology to adequately quantify ayahuasca alkaloids in plasma in order to characterize their pharmacokinetics in humans following oral administration of the tea. As the method developed by Callaway and colleagues (1996) was published, this was used as a starting point for our own methodology.

The steps taken to fulfill the above objectives were as follows. First, the Spanish version of the HRS questionnaire was administered both to ayahuasca users and to users of psychedelics in order to explore the psychometric properties of this instrument. The next stage was the initiation of the clinical studies with a pilot study conducted to explore the general tolerability (i.e., safety) of the lyophilizate, and its capacity to elicit psychotropic effects. The information obtained was used to establish the doses to be administered in the larger final study. This involved a greater number of volunteers and study variables, and the implementing of an optimal design. The analytical method was developed once the clinical phase of the project was completed. Further details can be found in a report by Yritia and colleagues (2002).

Questionnaire Assessment Studies

This involved administering the Spanish version of the HRS to a large sample of volunteers in two different studies, without manipulating their pattern of drug use. In Study 1, 75 users of ayahuasca participated. They were asked to answer the HRS approximately four hours after intake, in what is known as an immediate retrospective assessment of effects. The reliability of the different HRS scales was assessed. In Study 2, 56 volunteers with experience in the use of different kinds of psychotropic drugs were asked to answer the HRS and an already-validated questionnaire, the Addiction Research Center Inventory (ARCI; Martin et al. 1971), recalling the effects of their last previous psychedelic drug intake. This approach is known as a delayed retrospective assessment of effects. Again, reliability was assessed and correlations between HRS and ARCI scales were also calculated.

The result was that four of the six HRS scales were found to have an acceptable level of internal consistency. Two HRS scales also showed significant, though limited, correlations with the ARCI LSD scale, indicating the construct validity of the questionnaire. Finally, the HRS appeared to be sensitive to psychedelic drug effects other than those elicited by intravenous DMT, for which it was originally designed. Perhaps the most important limitation of the HRS is the high degree of intercorrelations found between scales, indicating that what they intend to measure is not totally independent but shows a marked overlap (for more information on the HRS assessment study, see Riba et al. 2001a). However, this problem is also present for other questionnaires measuring psychedelic effects, such as the APZ (see Dittrich 1998).

THE CLINICAL STUDIES, METHODOLOGY AND RESULTS

All participants enrolled in the pilot and final double-blind studies were healthy young volunteers with prior experience in the use of psychedelics. Volunteers were recruited by word of mouth. The extent and nature of their past psychedelic drug use was assessed in an initial interview in which the purposes of the study and the routine of the experimental sessions were explained to them. Insufficient or prior traumatic experience with psychedelics precluded participation. Eligible candidates subsequently underwent an interview with a psychiatrist to

rule out present or past psychiatric disorders, and alcohol and drug dependencies. A psychiatric disorder in first degree relatives was also considered a cause for exclusion. After the psychiatric assessment, volunteers underwent a general medical examination involving anamnesis, physical exploration, electrocardiogram and standard laboratory tests. This selection phase was also useful for the volunteer to become familiar with the research team and the environment where the sessions would be conducted. As an additional precaution and to reduce the anxiety associated with the interventions, it was decided that ayahuasca would not be administered on the first day of participation of any volunteer. In the course of the pilot single-blind study this was accomplished by administering the placebo on the first day of participation, and in the final double-blind study by administering an additional nonrandomized placebo on the first day of participation in a single-blind manner (see below).

All sessions were conducted in a quiet, dimly-lit room. The experimenter stayed with the volunteer, or remained in an adjacent room when no measurements were being conducted if the volunteer preferred. Room temperature and light were also adjusted to the participant's comfort. The volunteers were informed that nausea might occur and that they should try to avoid vomiting as this would modify the experiment results, i.e., alkaloid plasma level measurements. Nevertheless, preparations were made in case this was unavoidable.

Every effort was made to make the experimental sessions as comfortable and pleasant as possible for the volunteers within the inevitable limitations of a clinical research laboratory. An aspect that was important in achieving this goal was that the volunteers share the researchers' view on the value of the information to be gained through this kind of studies. A high degree of trust and empathy has to develop between the participant and the person conducting the study so that the volunteer feels safe. During the initial familiarization phase, potential volunteers who wished to enroll in the clinical trials despite overt distrust towards science in general or towards the clinical research approach to psychedelics were recommended not to participate. All volunteers received detailed information on the nature of ayahuasca and the general effects of psychedelics, including potential adverse effects. A signed informed consent form was required from all volunteers, participation was totally free and anyone could withdraw from the study at any time if they so wished. The clincial trials were approved by the hospital's ethics committee and the Spanish Ministry of Health.

The Pilot Clinical Trial

The first participants enrolled in the clinical phase of this project were six male volunteers, all of them with experience in psychedelic drug use and more specifically with ayahuasca. The aims of this pilot study were to assess the general tolerability, i.e., the safety of ayahuasca administration and to obtain initial information on the time course of effects and intensity thereof, in order to decide the final doses to be used in a larger subsequent study. For safety reasons, the doses were administered in increasing order, i.e., first the placebo, followed by the low dose, the medium dose and the high dose. On this occasion, the more demanding double-blind design was not used, but rather a single-blind approach, in which the experimenter but not the volunteer knew which dose was being administered on each day. The doses used were 0.5, 0.75 and 1.0 mg DMT/kg body weight. To measure subjective effects, the HRS and ARCI questionnaires were used together with visual analogue scales (VAS). Systolic (SBP) and diastolic (DBP) blood pressure and heart rate (HR) were recorded as physiological measures. Furthermore, blood samples were drawn after each experimental session to conduct complete hematological and biochemical determinations.

From this study it became evident that ayahuasca followed a dose response pattern with subjective effect intensity and alkaloid plasma levels increasing with dose. Cardiovascular effects appeared moderate in intensity and did not reach statistical significance for any variable. The laboratory analyses conducted after each session did not find any alterations in hematological indices or in biochemical indicators of liver function or other standard analytical parameters. Given the specific prior experience of this group of volunteers with ayahuasca, the doses to be used in the final study were decided based on their reports. Participants indicated that the 1.0 mg DMT/kg dose was exceedingly high. It was thus determined that the two doses to be used in the final larger study would lie between 0.5 and 0.75 mg/kg (low dose) and between 0.75 and 1.0 mg/kg (high dose). The detailed results of the pilot study have been published by Riba and colleagues (2001b).

THE FINAL DOUBLE-BLIND CLINICAL TRIAL

In this larger study, 18 volunteers (three female) were enrolled. They all had prior experience with psychedelic drug use, although not necessarily with ayahuasca. This time the study followed a double-blind design and doses of 0.6 and 0.85 mg DMT/kg body weight were administered following a randomized balanced order. As study variables, subjective and cardiovascular effects and pharmacokinetics were again measured, together with electroencephalography, evoked potential recordings, and levels of excreted neurotransmitter metabolites. The latter were quantified to evaluate the MAO inhibition pattern of ayahuasca. Hematological and biochemical parameters were determined this time at the beginning and at the end of the study and not after every single session. The main findings of this second larger study are described below.

Tolerability

Cardiovascular effects. Throughout the experimental sessions, cardiovascular measures were obtained with the volunteers seated. In both the pilot and the final study, increases in SBP, DBP and HR mean values were observed at specific time points following ayahuasca administration as compared with mean placebo values. However, these increases did not appear to be a robust effect of ayahuasca as they were not large in magnitude or systematic. Statistically significant results were only obtained for DBP in the final double-blind study, where the sample was larger. This variable showed a moderate 9 mm Hg increase at 75 minutes after the 0.85 mg DMT/kg body weight dose. For SBP the largest, though not significant, differences with placebo were observed at 75 minutes and corresponded to 6 mm Hg increases after the high dose. Heart rate was even less affected. The largest differences with placebo were seen at 60 minutes and corresponded to four beats/minute increases after the high dose. In the pilot study, maximum differences with placebo for cardiovascular measures were observed after the high 1 mg DMT/kg dose and corresponded to 14 mm Hg for SBP, 10 mm Hg for DBP and 9 bpm for HR.

In the pilot and final study combined, two volunteers showed SBP values above 140 mm Hg at some time point and four showed DBP values above 90 mm Hg, the diagnostic criteria of hypertension. One volunteer showed HR values above 100 bpm, the diagnostic criterion of tachycardia. The maximum values recorded at any time point were 146 mm Hg for SBP, 96 mm Hg for DBP and 101 bpm for HR.

In view of these moderate cardiovascular effects, ayahuasca seems relatively safe from a cardiovascular point of view, but it should also be kept in mind that the results obtained refer only to single dose administrations in young healthy volunteers and recorded in the absence of any physical exercise. The cardiovascular picture could differ following repeated dose administration, a common practice in ayahuasca rituals, or were ayahuasca ingested by older individuals or those with cardiovascular conditions, or while performing physical exercise such as dancing.

Other aspects of the physical tolerability of ayahuasca. In the course of the experimental sessions, volunteers commonly reported a series of somatic-dysphoric effects associated with drug intake. The most common were altered bodily sensations, nausea, sensations of heat or cold, pins and needles, and gastrointestinal disturbances. Despite the nausea experienced by many volunteers, actual vomiting was only observed in four of 53 occasions in which ayahuasca was administered (pilot and final studies combined). Regarding laboratory determinations, laboratory blood analyses were conducted after each experimental session in the course of the pilot study without any alteration being found in standard hematological and biochemical parameters.

Unpleasant psychological effects. Most volunteers regarded the ayahuasca experience as pleasant, although they felt some degree of anxiety at some moment or another during the session, most often as effects increased towards a maximum. In no case did subjective effects last longer than the usual four to six hours, nor did any participant become concerned that they might not wear off. The most distressing event was experienced by one volunteer during the pilot study after having received the medium 0.75 mg DMT/dose. The volunteer experienced an intensely dysphoric reaction with transient disorientation and anxiety. Verbal support was sufficient to get him through this state, which lasted around 20 minutes. His displeasure with the experience led him to voluntarily withdraw from the study.

Alkaloid Plasma Levels Following Ayahuasca Administration

As part of the secondary objectives of the research project, analytical methods were validated to determine the plasma concentration of the different alkaloids (for the technical details see Yritia et al. 2002). Following this methodology, three major ayahuasca alkaloids (DMT, harmaline and THH) were measured in plasma following ayahuasca administration. Unexpectedly, however, levels of harmine were negligible and only observed in a small subset of volunteers. Levels of harmol and harmalol, the *O*-demethylated analogues of harmine and harmaline, were detected in all volunteers. For all measured compounds, the maximum plasma concentration (C_{max}) and area under the curve (AUC) values for the alkaloids increased with dose. C_{max} values for DMT were 12.14 ng/ml and 17.44 ng/ml after the low and high ayahuasca doses, respectively. The C_{max} was reached 1.5 hours after both doses and coincided with the peak of subjective effects. Indeed, the time course of DMT plasma concentrations closely paralleled that of subjective effects. The AUC normalized by dose index increased with dose for DMT. Conversely, the apparent volumes of distribution (V_z/F) and clearance (Cl/F) values calculated for DMT decreased with dose. These decreases were statistically significant for V_z/F and showed a tendency for Cl/F (Riba et al. 2003). Such results are suggestive of a nonlinear increment of DMT levels following the administration of larger doses of ayahuasca and could be due to the action of the higher amounts of β-carbolines administered. The authors plan to address the role of the β-carbolines in the possible nonlinearity of DMT levels between ayahuasca doses in a repeated dose administration study.

Based on the intravenous (i.v.) DMT plasma level data generously provided by Rick J. Strassman, we estimated the percentage of the total DMT amount in ayahuasca that finally reached systemic circulation (bioavailability). This calculation yielded a tentative value of around 10% to 15%

(Riba 2003). The approach used to calculate bioavailability was not ideal since i.v and oral data were obtained in different groups of volunteers and calculations had to be performed with grand-mean values rather than on an individual basis. However, if the real value is close to the value we obtained, it would be surprisingly low and would pose questions regarding the metabolism of DMT in vivo. It would mean that a very small fraction of the DMT in ayahuasca actually reaches systemic circulation. MAO activity has either only partially been inhibited by the β-carbolines, or DMT is redirected to other metabolic routes. Indeed, MAO-independent metabolites have been detected in in vitro experiments (Fish et al. 1955). However, the metabolic fate of DMT when administered alone to humans has not been investigated systematically. Szára (1956) found the MAO-dependent 3-indoleacetic acid in the urine of his volunteers after receiving DMT, but this amounted to a very low percentage of the administered dose. An interesting experiment would be to first identify the metabolic products of oral DMT when administered alone and then assess how the metabolic profile is modified by the concomitant administration of increasing doses of the different β-carbolines.

Another intriguing aspect of ayahuasca pharmacology is the absence of measurable levels of harmine in plasma in the present sample. This would suggest the alkaloid has an efficacious metabolism. A rapid turnover of harmine to harmol has been observed in vitro (Yu et al. 2003). However, that this is the cause of the lack of harmine levels cannot be unequivocally established, as low, but nevertheless measurable, levels of harmol and harmalol were found in the ayahuasca sample used in this study (see above). Interestingly, Callaway and colleagues (1999) found high harmine plasma levels in their sample. Ethnical differences in the metabolism of harmine could perhaps be an explanation. Phenotyping or genotyping volunteers for the CYP2D6 and CYP1A1 involved in harmine *O*-demethylation could shed some light on the differences observed between the present sample and that of Callaway and colleagues (1999).

Assessing the MAO-Inhibiting Effects of Ayahuasca in Vivo

Although MAO inhibition is generally acknowledged as the mechanism facilitating the absorption of DMT in the gastrointestinal tract, there is no direct proof that this enzymatic inhibition actually takes place in humans after the ingestion of ayahuasca. In order to measure MAO inhibition in our volunteers in vivo, we quantified the levels of excreted urine monoamine metabolites. This approach is based on the fact that the neurotransmitters noradrenaline, epinephrine and dopamine are themselves physiologically degraded by MAO and another enzyme, cathecol-*O*-methyltransferase (COMT), to produce deaminated and methylated metabolites, respectively. Serotonin, on the other hand, is exclusively metabolized by MAO to produce a deaminated compound. In vivo and in vitro studies have shown that when MAO is pharmacologically inhibited, the levels of MAO-dependent deaminated metabolites decrease while those of COMT-dependent methylated compounds increase. In humans, MAO inhibitors decrease, after acute administration, the urinary excretion of vanilmandelic acid (VMA), homovanillic acid (HVA) and 5-hydroxyindoleacetic acid (5-HIAA), the deaminated metabolites of noradrenaline/epinephrine, dopamine and serotonin, respectively; while increasing that of metanephrine and normetanephrine, the methylated metabolites of epinephrine and noradrenaline, respectively (Koulu et al. 1989; Pletscher 1966). Monoamine metabolites have both a CNS and a non-CNS origin and their assessment in urine does not give information regarding the organ where MAO was inhibited. Nevertheless, this approach can identify dose-response relationships after drug administration and allows for the study of the time course of MAO-inhibition.

However, the results we obtained after acute administration of ayahuasca could not establish an unequivocal pattern of variation in these levels based on these measures. Ayahuasca increased the excretion of all metabolites measured in 24 h urine. Nevertheless, the only statistically significant increase observed was that of normetanephrine, the noradrenaline COMT-dependent metabolite. Although the increase in normetanephrine is in line with an inhibition of MAO, the increases of deaminated compounds (VMA, HVA, 5-HIAA) are not. With the increase in normetanephrine as the only significant change, it is impossible to conclude there was a systemic inhibition of MAO. The increases in normetanephrine could, for instance, also be due to increased noradrenaline release while its degradation rate remained constant.

The lack of a clear MAO inhibitory profile may have been related to the amounts of β-carboline administered with the ayahuasca doses. These were established in terms of DMT administered per kg body weight. At the 0.85 mg DMT/kg dose which was used in the final study as the high dose, volunteers received 1.45 mg/kg harmine. This value is slightly below the 1.5 mg/kg found by Ott (1999) to be the threshold dose necessary to render DMT orally active in self-experiments. Although 1.45 mg/kg may have been sufficient to allow enough DMT to reach the systemic circulation unmetabolized and to elicit psychotropic effects, it may not have been sufficient to modify the profile of endogenous monoamine metabolites in urine. Thus, the harmine-DMT interaction may have taken place predominantly in the gastrointestinal tract. At least in our experiment, harmine cannot possibly have exerted any effects on the CNS, as it did not even reach the systemic circulation. The in vivo MAO inhibition effect could be reassessed with this methodology in future studies, using

ayahuasca batches with higher amounts of β-carboline. A repeated dose study would perhaps yield a greater degree of MAO inhibition that could be reflected as the expected variations in neurotransmitter metabolite excretion.

Subjective Effects

The approach used in this study to what the participants experienced subjectively in the course of the experimental sessions was several-fold. Interviews were conducted and recorded after any session when volunteers felt they would like to report on something they had experienced. These interviews were basically nonstructured and the interviewer tried to intervene only minimally, mainly to ask the volunteers if they would like to enlarge on a topic that had been raised. Although this approach should a priori provide a richer account of the experience, it relies heavily on the volunteer's ability to verbalize the nuances of the experience. Therefore, while in some cases rich descriptive reports were obtained, in others volunteers only commented on their having been more or less "high." To date, none of this material has been analyzed.

A more structured approach was inevitably needed to record subjective effects. While obviously reductionistic, this approach provides a quantitative measure of various aspects of the experience that may help in the search for dose-response relations. Thus, questionnaires like the ARCI and the HRS were administered. With the ARCI, the experimental drug can be classified as pertaining to a given class based on the subjective effects it elicits. Essentially, it provides information on whether a drug is more sedative-like or stimulant-like or whether it elicits dysphoric effects or euphoriant effects. On the other hand, the HRS was specifically developed to measure different facets of the psychedelic experience and was drafted from reports of subjects taking parenteral DMT, both self-administered and in a clinical research setting. In the present studies, these two questionnaires were always administered after at least four hours had elapsed following the administration of ayahuasca. To measure the time course of effects, i.e. the intensity of a certain effect at a given time point, visual analogue scales were administered.

The overall picture obtained from the structured approach showed that effects exhibited a similar time course between individuals. On average, psychological effects were first noted after 30 to 60 minutes, they peaked between 60 to 120 minutes and were resolved in four to six hours. These effects included feelings of increased activation (ARCI-A, VAS-stimulated), euphoria and well-being (ARCI-MBG, VAS-high, VAS-liking, VAS-good effects) and somatic modifications (ARCI-LSD), in addition to perceptual modifications (HRS-Perception, VAS-visions), changes in thought content (HRS-Cognition) and increased emotional lability (HRS-Affect).

The constellation of subjective effects elicited by ayahuasca could thus be effectively quantified by means of the battery of self-administered instruments used for this purpose. The HRS was sensitive to ayahuasca effects, as it has previously been shown to be sensitive to other psychedelics such as i.v. DMT and oral psilocybin (Gouzoulis-Mayfrank et al. 1999; Strassman et al. 1994). VAS data indicated that ayahuasca shows a distinct duration of effects in comparison with other prototypical psychedelic drugs (i.e., longer than those of i.v. DMT) but shorter than those of oral mescaline or LSD. Finally, ARCI results revealed that ayahuasca effects are not merely perceptual but share common features with psychostimulants and elicit marked somatic-dysphoric symptoms. The coexistence of stimulation with modifications in the sensorium clearly portrayed ayahuasca as a psychedelic, with its subjective effects following a classical dose-dependent pattern. Once this information has been established for ayahuasca, blockade studies with selective antagonists can be proposed to better characterize the neurochemical mechanisms involved in its effects. Additionally, it may now be possible to address the long neglected study of individual β-carbolines in humans and examine which of these and at what doses they can elicit subjective effects analogous to those of ayahuasca, if at all. Besides, the use of quantitative measures of subjective effects in studies involving the administration of pure β-carbolines with pure DMT may help tease apart the contribution of each of the β-carbolines in the facilitation of DMT absorption per os.

Effects on the EEG Power Spectrum

Electroencephalography consists of recording spontaneous brain electrical activity by means of electrodes placed on the scalp surface. Once the recordings are obtained and after adequate preprocessing of the signals, the energy or power present in a series of frequency bands can be quantified. This approach to the study of drug effects on the CNS, termed quantitative pharmacoelectroencephalography, is modest in its objectives despite its long tradition. The power spectrum variables derived from the spontaneous EEG are not directly associated with specific cognitive processes and thus the information gained by these means is somewhat unspecific. This technique essentially detects whether a drug has an effect on the CNS, assesses the time course of this effect, and determines whether there is a dose-response profile when doses are escalated. In some cases it may provide a specific neurophysiologic correlate of the drug's central actions, identifying from among the multiple variables that can be computed those that are most selectively and reliably modified by the drug in question. Furthermore, by comparing these changes with the pattern induced by other better known compounds, inferences can be made regarding the neurochemical mechanism of action of the drug under investigation This technique not only has the advantage of providing a quantitative output, but is objective, relatively inexpensive and it allows for an almost unlimited number

of measurements within a session. However, in order to obtain meaningful results it requires very strictly standardized procedures given the large interindividual variability in EEG measures. Recording conditions and the degree of vigilance during the recording session should be, among other aspects, carefully monitored. Furthermore, placebos should be implemented to control for circadian rhythms, measurements should be repeated along time to determine whether there is a time course of EEG effects compatible with the pharmacokinetics of the drug and finally, various doses of the same drug should be administered to make sure the observed effect varies with dose. Field conditions are not therefore the ideal setting for conducting this kind of research. Additionally, adequate preprocessing of the recordings in order to eliminate artifacts is a critical step that cannot be resolved by the mere visual inspection of the traces, and submitting the data to statistical analysis is mandatory (see Anderer et al. 1992, 1987; Ferber et al. 1999). In recent years, several studies attempting to evaluate the effects of ayahuasca on the CNS under field conditions have overlooked the above requirements. The reproducibility of results is therefore dubious and even more so considering that the only results found in some of these studies were in the frequencies above 30 Hz, where EEG energy is minimal and artifact susceptibility highest, thus imposing even more demanding recording conditions and preprocessing techniques.

Concerning our own research, we are still far from having established a specific neurophysiologic marker of ayahuasca effects on the EEG and we consider our results preliminary. Nevertheless, we have effectively been able to demonstrate that ayahuasca does indeed induce statistically significant changes on the EEG and have measured the time course of these effects. Non-parametric analysis combining all variables and leads together showed that the first changes relative to placebo appeared between 15 and 30 minutes. These are followed by a steep rise at 45 minutes, reaching the maximum effects between 45 and 90 minutes. EEG measures gradually decline thereafter to reach baseline values in around four to six hours. These ayahuasca-induced EEG modifications largely coincide with the time course of subjective effects and DMT plasma levels.

With regard to the individual EEG variables targeted, a general decrease was observed in absolute power in the entire 0.3-30 Hz frequency range studied. This decrease was more pronounced in the low frequency range, leading to a displacement of the center-of-gravity towards the higher frequencies. Absolute power decreases were most prominent in theta, delta and beta-1 bands at 90 to 120 minutes, while the alpha-2 band showed a highly significant decrease in all leads at 60 minutes. Relative power was most prominently increased in the beta-3 and beta-4 bands. Additionally, the alpha/delta-theta ratio, an index of activation, was found to be increased after ayahuasca. All these effects were more intense at the higher dose than at lower ayahuasca dose (Riba et al. 2002a).

The modification pattern of the individual variables is far from simple and further EEG studies will be needed to establish which variable better reflects the effects of ayahuasca on the CNS. However, some comparisons can be established with other psychoactive drugs. Early pharmaco-EEG research also found decreases in delta-theta and alpha activity after acute LSD administration (Itil & Fink 1966). Ayahuasca's effects would be similar to those described for LSD. The fact that ayahuasca decreases delta and theta power, which have traditionally been thought to reflect inhibitory activity and relaxed states, respectively, suggests an excitatory or arousing effect for the brew. This assumption is further supported by the fact that major tranquilizers with D_2 or mixed D_2/5-HT_2 antagonist activity, such as chlorpromazine and risperidone, are characterized by their delta and theta-promoting activity (Lee et al. 1999; Saletu et al. 1993).

Intracerebral Location of the Observed Scalp EEG Changes

Based on the scalp electrical potential distribution (topography) obtained by means of classical EEG measures it is possible to tentatively identify the cortical brain areas responsible for the observed changes. One technique that allows for the calculation of this "inverse solution" is called low resolution electromagnetic tomography or LORETA. This algorithm computes a unique three-dimensional intracerebral power density distribution for the different EEG frequency bands, making no a-priori assumptions about the number of sources involved, in contrast to dipole modeling techniques. Based on the assumption that neighboring neuronal sources are likely to be similarly active, the only constraints imposed by this technique is that of maximal smoothness of the solution and its anatomical restriction to cortical gray matter and hippocampus. Thus, the algorithm provides by definition a sparse map of activated (or deactivated) areas. Hence the "low resolution" (Pascual-Marqui, Michel & Lehmann 1994).

In the present study, intracerebral power density showed the maximum differences with placebo at 60 and 90 minutes. Ayahuasca decreased power density in the alpha-2, delta, theta and beta-1 bands. At 60 minutes, a widespread power reduction in the alpha-2 band was observed in extensive areas around the temporo-parieto-occipital junction in both hemispheres. At 90 minutes, the slow delta rhythm was decreased also in posterior brain regions around the temporo-parieto-occipital junction. Theta was found to be decreased in the medial frontal and medial temporal cortices. Finally, beta-1 decreases were found mainly in the parietal lobe (Riba et al. 2004).

Based on these results, it would seem that bioelectrical changes are found on cortical association areas in temporo-parietal and frontomedial areas, together with

paralimbic areas such as the cingulate and the temporomedial cortices, which play relevant roles in the neurobiology of attention, emotion, and memory. It is worth mentioning that in contrast with the widespread power density decreases observed here for ayahuasca in posterior regions with LORETA, PET and SPECT studies of psychedelic drug administration have shown blood flow and metabolic increases in the frontal cortex (see for instance Vollenweider et al. 1997). However, a recent study has shown that a direct anatomical relation between drug-induced bioelectric and blood perfusion/metabolic changes in the brain may not exist (Gamma et al. 2004; Nuñez & Silberstein 2000).

Effects on Sensory and Sensorimotor Gating

Current human research with psychedelics and entactogens has explored the possibility that drugs displaying agonist activity at the 5-$HT_{2A/2C}$ sites temporally disrupt inhibitory neural mechanisms thought to intervene in the normal filtering of information. Suppression of the P50 auditory evoked potential and prepulse inhibition of startle (PPI) are considered operational measures of sensory and sensorimotor gating, respectively. According to this model, serotonergic psychedelics would interact with brain structures involved in the gating mechanisms, temporarily decreasing their functionality and giving rise to the characteristic perceptual and cognitive effects elicited by these agents. In the final double-blind study we evaluated the effects of ayahuasca on these measures.

At the doses administered, however, ayahuasca induced a different pattern of effects on PPI and P50. Ayahuasca produced dose-dependent reductions of P50 suppression measured both as amplitude difference between the response to the conditioning and testing stimuli and as percent inhibition of the testing response. On the contrary, no statistically significant effects were found on the magnitude of the startle response, its habituation rate or on the percentage prepulse inhibition at any of the prepulse-to-pulse intervals studied (Riba et al. 2002b).

STUDIES CURRENTLY UNDERWAY

Since the research project described above was completed, we have undertaken new experiments to study issues such as changes in regional cerebral blood flow during the peak effects of ayahuasca, the modulation of the neuroendocrine and immune system parameters after acute ayahuasca administration, and the effects of daytime ayahuasca use on sleep architecture. The clinical part of these experiments is now complete and we are currently analyzing the data generated.

We studied regional cerebral blood flow by means of single photon computerized tomography or SPECT. A radiotracer was administered at the peak of subjective effects following the administration of a 1 mg DMT/kg body weight ayahuasca dose. The uneven uptake of the radiotracer in the different brain regions is dependent on blood flow, which is in turn believed to reflect neural activity, with increased uptake reflecting increased activation in a given brain region. In fact, this technique, together with positron emission tomography (PET) and functional magnetic resonance imaging (fMRI) are considered the "gold standards" in neuroimaging. By this means we hope to expand our knowledge of which brain areas are involved in the genesis of the effects of ayahuasca.

Another interesting aspect of ayahuasca phamacology we are currently addressing is the effects of the tea on the neuroendocrine and immune systems. We are currently analyzing the variations in cortisol, growth hormone and prolactin release, and lymphocyte subpopulations in blood samples taken after the acute administration of ayahuasca. Although these analyses on the acute effects of the tea on neuroendocrine variables and the immune system will not provide information regarding the changes experienced by users after years of ayahuasca ingestion, important initial information will be gained on the capabilities of ayahuasca to modulate these systems.

Finally, we are also exploring the effects of daytime consumption of ayahuasca on the architecture of sleep on the following night. It is well known that the serotonergic system, which is targeted by ayahuasca alkaloids, plays a relevant role in the regulation of sleep. Our approach to assessing the impact of ayahuasca on sleep regulation is based both on the analysis of variables derived from the classical sleep stage classification (macroscopic changes in a 30 s scale) and on the study of the variations of energy content in the different frequency bands throughout the night (microscopic changes in a continuous scale).

FUTURE STUDIES

Two studies are planned for the near future. We are currently about to begin a repeated dose study in which two identical doses of ayahuasca will be administered following a certain time interval, a pattern of use which is very common in ayahuasca sessions. A primary focus of this study will be on cardiovascular measures and on alkaloid pharmacokinetics in order to determine a possible nonlinearity in the DMT plasma levels associated with the administration of the second dose. After controlling for this possibility, we will explore whether subjective and EEG effects after the second dose are disproportionally increased (indicating sensitization), disproportionally decreased (indicating tolerance), or simply linearly superimposed on the residual effects of the first dose.

In 2006, we plan to conduct a study involving the administration of ayahuasca after pretreatment with a serotonin-2 receptor antagonist and evaluate how this

modifies subjective effects and EEG measures. This study should help clarify whether mechanisms other than the activation of this receptor are responsible for the entire or at least part of the pharmacological picture of ayahuasca.

Finally, in the more distant future, we plan to undertake the study of the individual ayahuasca alkaloids and their interactions. Here, as already mentioned, the ayahuasca lyophilizate could be used as a potential positive control. This approach could help tease apart the respective contributions of the different alkaloids to the final psychotropic effects of this fascinating Amazonian preparation.

REFERENCES

Anderer, P.; Semlitsch, H.V.; Saletu, B. & Barbanoj, M.J. 1992. Artifact processing in topographic mapping of electroencephalografic activity in neuropsychopharmacology. *Psychiatry Research Neuroimaging* 45:79-93.

Anderer, P.; Saletu, B.; Kinsperger, K. & Semlitsch, H. 1987. Topographic brain mapping of EEG in neuropsychopharmacology - Part I. Methodological aspects. *Methods and Findings in Experimental and Clinical Pharmacology* 9:371-84.

Buckholtz, N.S. & Boggan, W.O. 1977a. Monoamine oxidase inhibition in brain and liver produced by β-carbolines: Structure-activity relationships and substrate specificity. *Biochemical Pharmacology* 26: 1991-96.

Buckholtz, N.S. & Boggan, W.O. 1977b. Inhibition by β-carbolines of monoamine uptake into a synaptosomal preparation: structure-activity relationships. *Life Science* 20: 2093-99.

Callaway, J.C.; McKenna, D.J.; Grob, C.S.; Brito, G.S.; Raymon, L.P.; Poland, R.E.; Andrade, E.N.; Andrade, E.O. & Mash, D.C. 1999. Pharmacokinetics of *hoasca* alkaloids in healthy humans. *Journal of Ethnopharmacology* 65: 243-56.

Callaway, J.C.; Raymon, L.P.; Hearn, W.L.; McKenna, D.J.; Grob, C.S. & Brito, G.S. 1996. Quantitation of N,N-dimethyltryptamine and harmala alkaloids in human plasma after oral dosing with ayahuasca. *Journal of Analytical Toxicology* 20: 492-97.

Dittrich, A. 1998. The standardized psychometric assessment of altered states of consciousness (ASCs) in humans. *Pharmacopsychiatry* 31 (Suppl 2): 80-4.

Ferber, G.; Abt, K.; Fichte, K. & Luthringer, R. 1999. IPEG guideline on statistical design and analysis for pharmacodynamic trials. International Pharmaco-EEG group. *Neuropsychobiology* 39: 92-100.

Fish, M.S.; Johnson, N.M.; Lawrence, E.P. & Horning, E.C. 1955. Oxidative N-dealkylation. *Biochemical and Biophysical Acta* 18: 564-65.

Gamma, A.; Lehmann, D.; Frei, E.; Iwata, K.; Pascual-Marqui, R.D. & Vollenweider, F.X. 2004. Comparison of simultaneously recorded [$H_2{}^{15}O$]-PET and LORETA during cognitive and pharmacological activation. *Human Brain Mapping* 22: 83-96.

Gouzoulis-Mayfrank, E.; Thelen, B.; Habermeyer, E.; Kunert, H.J.; Kovar, K.A.; Lindenblatt, H.; Hermle, L.; Spitzer, M. & Sass, H. 1999. Psychopathological, neuroendocrine and autonomic effects of 3,4-methylenedioxyethylamphetamine (MDE), psilocybin and *d*-methamphetamine in healthy volunteers. *Psychopharmacology* 142: 41-50.

Itil, T. & Fink, M. 1966. Klinische Untersuchungen und quantitative EEG-Daten bei experimentellen Psychosen. Arzneimittelforschung 16: 237-39.

Koulu, M.; Scheinin, M.; Kaarttinen, A.; Kallio, J.; Pyykkö, K.; Vuorinen, J. & Zimmer, R.H. 1989. Inhibition of monoamine oxidase by moclobemide: effects on monoamine metabolism and secretion of anterior pituitary hormones and cortisol in healthy volunteers. *British Journal of Clinical Pharmacology* 27 :243-55.

Lee, D.Y.; Lee, K.U.; Kwon, J.S.; Jang, I.J.; Cho, M.J.; Shin, S.G. & Woo, J.I. 1999. Pharmacokinetic-pharmacodynamic modeling of risperidone effects on electroencephalography in healthy volunteers. *Psychopharmacology* 144: 272-78.

Martin, W.R.; Sloan, J.W.; Sapira, J.D. & Jasinski, D.R. 1971. Physiologic, subjective, and behavioral effects of amphetamine, methamphetamine, ephedrine, phenmetrazine, and methylphenidate in man. *Clinical Pharmacology and Therapeutics* 12:245-258.

Nuñez, P.L. & Silberstein, R.B. 2000. On the relationship of synaptic activity to macroscopic measurements: does co-registration of EEG with fMRI make sense? *Brain Topography* 13: 79-96

Ott, J. 1999. Pharmahuasca: Human pharmacology of oral DMT plus harmine. *Journal of Psychoactive Drugs* 31: 171-77.

Ott, J. 1993. *Pharmacotheon: Entheogenic Drugs, Their Plant Sources and History*. Kennewick, Washington: Natural Products Co.

Pascual-Marqui, R.D.; Michel, C.M. & Lehmann, D. 1994. Low resolution electromagnetic tomography: A new method for localizing electrical activity in the brain. *International Journal of Psychophysiology* 18: 49-65.

Pletscher, A. 1966. Monoamine oxidase inhibitors. *Pharmacological Reviews* 18: 121-29.

Riba, J. 2003. Human pharmacology of ayahuasca. Doctoral dissertation, Barcelona, Universitat Autònoma de Barcelona.

Riba, J.; Anderer, P.; Jané, F.; Saletu, B. & Barbano, M.J. 2004. Effects of the South American psychoactive beverage *Ayahuasca* on regional brain electrical activity in humans: A functional neuroimaging study using low resolution electromagnetic tomography (LORETA) *Neuropsychobiology* 50: 89-101.

Riba, J.; Valle, M.; Urbano, G.; Yritia, M.; Morte, A. & Barbanoj, M.J. 2003. Human pharmacology of *Ayahuasca*: subjective and cardiovascular effects, monoamine metabolite excretion and pharmacokinetics. *Journal of Pharmacology and Experimental Therapeutics* 306: 73-83.

Riba, J.; Anderer, P.; Morte, A.; Urbano, G.; Jané, F.; Saletu, B. & Barbanoj, M.J. 2002a. Topographic pharmaco-EEG mapping of the effects of the South American psychoactive beverage *Ayahuasca* in healthy volunteers. *British Journal of Clinical Pharmacology* 53:613-628.

Riba, J.; Rodríguez-Fornells, A. & Barbanoj, M.J. 2002b. Effects of *Ayahuasca* on sensory and sensorimotor gating in humans, as measured by P50 suppression and prepulse inhibition of the startle reflex, respectively. *Psychopharmacology* 165: 18-28.

Riba, J.; Rodríguez-Fornells, A,; Strassman, R.J. & Barbanoj, M.J. 2001a. Psychometric assessment of the Hallucinogen Rating Scale. *Drug and Alcohol Dependence* 62: 215-23.

Riba, J.; Rodríguez-Fornells, A.; Urbano, G.; Morte, A.; Antonijoan, R.; Montero, M.; Callaway, J.C. & Barbanoj, M.J. 2001b. Subjective effects and tolerability of the South American psychoactive beverage *ayahuasca* in healthy volunteers. *Psychopharmacology* 154: 85-95.

Saletu, B.; Barbanoj, M.J.; Anderer, P.; Sieghart, W. & Grünberger, J. 1993. Clinical-pharmacological study with two isomers (*d*-, *l*-) of fenfluramine and its comparison with chlorpromazine and *d*-amphetamine: Blood levels, EEG mapping and safety evaluation. *Methods and Findings in Experimetal and Clinical Pharmacology* 15: 291-312.

Spruce, R. 1908. *Notes of a Botanist on the Amazon and Andes, Vol 2*. London: Macmillan.

Strassman, R.J. & Qualls, C.R. 1994. Dose-response study of *N,N*-dimethyltryptamine in humans, I. Neuroendocrine, autonomic and cardiovascular effects. *Archives of General Psychiatry* 51: 85-97.

Strassman, R.J.; Qualls, C.R.; Uhlenhuth, E.H. & Kellner, R. 1994. Dose-response study of *N,N*-dimethyltryptamine in humans, II. Subjective effects and preliminary results of a new rating scale. *Archives of General Psychiatry* 51: 98-108.

Szára, S. 1956. Dimethyltryptamine: Its metabolism in man; the relation of its psychotic effect to the serotonin metabolism. *Experientia* 12: 441-42.

Vollenweider, F.X.; Leenders, K.L.; Scharfetter, C.; Maguire, P.; Stadelmann, O. & Angst, J. 1997. Positron emission tomography and fluorodeoxyglucose studies of metabolic hyperfrontality and psychopathology in the psilocybin model of psychosis. *Neuropsychopharmacology* 16: 357-72.

Yritia, M.; Riba, J.; Ortuño, J.; Ramirez, A.; Castillo, A.; Alfaro, Y.; de la Torre, R. & Barbanoj, M.J. 2002. Determination of *N,N*-dimethyltryptamine and b-carboline alkaloids in human plasma following oral administration of *Ayahuasca*. *Journal of Chromatography: Biomedical Applications* 779:271-281.

Yu, A.; Idle, J.R.; Krausz, K.W.; Küpfer, A. & Gonzalez, F.J. 2003. Contribution of individual cytochrome P450 isozymes to the *O*-demethylation of the psychotropic β-carboline alkaloids harmaline and harmine. *Journal of Pharmacology and Experimental Therapeutic* 305: 315-22.