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TEMÓTEO LUIZ LIMA DA SILVA

HÁ UMA BASE BIOLÓGICA NA SELEÇÃO DE PLANTAS MEDICINAIS?

UMA AVALIAÇÃO A PARTIR DE SISTEMAS MÉDICOS LOCAIS.

Recife – PE

2020

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Tese apresentada ao Programa de Pós-graduação em Etnobiologia e Conservação da Natureza (UFRPE, UEPB, URCA e UFPE) como parte dos requisitos para obtenção do título de doutor.

Orientador:

Prof. Dr. Ulysses Paulino de Albuquerque

Universidade Federal de Pernambuco

Coorientador:

Prof. Dr. Washington Sorares Ferreira Junior

Universidade de Pernambuco

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Temóteo Luiz Lima da Silva

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Orientador

Dr. Ulysses Paulino Albuquerque
(Universidade Federal Rural de Pernambuco)

Examinadores

Dr. Thiago Antônio de Souza Araújo
(Uninassau)

Dra. Ivanilda Soares Feitosa
(Universidade Federal Rural de Pernambuco)

Dra. Flávia Rosa Santoro
(Laboratório de Ecologia e Evolução de Sistemas Socioecológicos)

Dr. Gilney Charll dos Santos
(Instituto Federal de Pernambuco)

Hoje rasgo os dicionários
Vou denegrir o mundo
Tarjas pretas não cobrirão vergonhas
Pois, não há vergonhas para cobrir
Será uma honra estar
Em cadernos e listas negras
Toda família torcerá
Para que a próxima criança a nascer
Seja uma ovelha negra
Não nos chamaram da cor do pecado
Pois, se há pecado e ele tem cor, é rubro
Rubro como o sangue derramado
Em todas as guerras ditas santas,
movidada por um Deus bélico
Rubro como o sangue de nossos irmãos
Derramados nas periferias, em nome do Estado
Não teremos mais *Mães de maio*
Pois, balas perdidas ou achadas,
só as das festas de Cosme de Damião
Voltando as ovelhas
não há
Repito:
Não há um Deus que divida o seu rebanho
Por cor ou identidade de gênero
No mundo denegrado Deus (es) é frutacor
Hoje rasgo os dicionários
Queimo os sentidos sem sentido
E faço de toda palavra
... Poesia.

Por Patrícia Ashanti

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RESUMO

A presente tese tem como objetivo entender a relação entre o gosto das plantas medicinais e sua seleção para compor farmacopeias locais. Para isso, utilizamos duas vias de investigação: 1) avaliar se a percepção de gosto influencia a seleção de plantas medicinais para tratar conjunto de doenças específicas e se a aceitabilidade de gostos varia de acordo com cada tipo de gosto presente nas plantas medicinais; 2) avaliar se a base biológica da percepção quimiossensorial de gosto amargo influencia características dos sistemas médicos, especificamente a atribuição do papel social de especialista local, o hábito de ser experimentador novas plantas medicinais e o conhecimento sobre plantas com gosto. Coletamos os dados com moradores maiores de 18 anos em cinco comunidades localizadas no Parque Nacional do Catimbau. Registramos o conhecimento sobre plantas medicinais utilizando listas-livre e os atributos de gosto, aceitabilidade de gosto e alvos terapêuticos por meio de entrevistas. Os especialistas locais foram identificados por meio da validação local e os experimentadores por duas perguntas objetivas. A base biológica foi mensurada por meio de um teste de limiar de identificação de gosto amargo, utilizando soluções com concentrações crescentes de feniltiocarbamida. Encontramos que plantas de gosto amargo e travoso são usadas para tratar um conjunto de doenças específicas, corroborando as evidências da literatura. No entanto, as associações foram distintas entre as cinco comunidades estudadas, indicando que provavelmente que a associação entre o gosto da planta e a finalidade terapêutica dependa mais de fatores culturais do que outros aspectos. Plantas com gosto doce possuem maior aceitabilidade enquanto plantas amargas possuem menor aceitabilidade de gosto. Provavelmente mecanismos psicofisiológicos e culturais influenciam a aceitabilidade de gostos amargos ou repugnantes, favorecendo a ingestão de plantas com esse tipo de gosto. Encontramos que a sensibilidade na percepção de gosto amargo não influencia a atribuição do papel social de especialistas locais nem o hábito de experimentar novas plantas medicinais. Essa sensibilidade também não está associada ao conhecimento sobre plantas medicinais com gosto. Provavelmente essa base biológica teve papel fundamental no início do uso de plantas medicinais pela espécie humana, mas não influencia a relação entre gosto e a seleção de plantas nas farmacopeias atuais. Futuros estudos poderiam buscar entender se a percepção quimiossensorial de gosto influencia a seleção de plantas por populações sedentárias e semisedentárias, uma vez que essas comunidades costumam lidar com ambientes que variam no tempo e espaço.

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ABSTRACT

This thesis aims to understand the relationship between the taste of medicinal plants and their selection to compose local pharmacopoeias. For this, we use two ways of investigation: 1) evaluate whether the perception of taste influences the selection of medicinal plants to treat a set of specific diseases and whether the acceptability of tastes varies according to each type of taste present in medicinal plants; 2) to assess whether the biological basis of chemosensory perception of bitter taste influences characteristics of medical systems, specifically the attribution of the social role of a local specialist, the habit of being an experimenter of new medicinal plants and knowledge about plants with taste. We collected data from residents over 18 years of age in five communities located in the Catimbau National Park. We recorded knowledge about medicinal plants using free lists and the attributes of taste, acceptability of taste and therapeutic targets through semi-structured interviews. Local experts were identified through local validation and experimenters were asked two objective questions. The biological basis was measured using a threshold test to identify bitter taste, using solutions with increasing concentrations of phenylthiocarbamide. We found that plants with a bitter and “travoso” taste are used to treat a set of specific diseases, corroborating the evidence in the literature. However, the associations were distinct between the five communities studied, indicating that the association between the taste of the plant and the therapeutic purpose is likely to depend more on cultural factors than other aspects. Plants with a sweet taste have greater acceptability while bitter plants have less acceptability of taste. Probably, psychophysiological and cultural mechanisms influence the acceptability of bitter or disgusting tastes, favoring the ingestion of plants with this type of taste. We found that sensitivity in the perception of bitter taste does not influence the attribution of the social role of local specialists or the habit of experimenting new medicinal plants. This sensitivity is also not associated with knowledge about medicinal plants with taste. This biological basis probably played a fundamental role in the beginning of the use of medicinal plants by the human species, but it does not influence the relationship between taste and plant selection in current pharmacopoeias. Future studies could seek to understand whether the chemosensory perception of taste influences the selection of plants by sedentary and semi-sedentary populations, since these communities usually deal with environments that vary in time and space.

INTRODUÇÃO GERAL

1.1 OBJETIVOS E QUESTIONAMENTOS

Evidências na literatura etnobotânica apontam que o gosto das plantas pode guiar sua seleção para tratar doenças específicas, como as plantas amargas que tratam doenças do trato digestório (AKLIN et al., 1999; BRETT e HEINRICH, 1998; MEDEIROS et al., 2015) além de doenças parasitárias (HUFFMAN, 1997). Esses estudos defendem que os atributos de gosto das plantas medicinais é uma característica utilizada pelas pessoas para identificar o potencial farmacológico de plantas (MEDEIROS et al., 2015; MORALES e LADIO, 2009). Para Gollin (2004), o gosto e outras propriedades da planta (como odor) são constructos sociais que geralmente possuem uma base biológica. Assim, a seleção de plantas por meio do gosto se daria por dois processos: o biológico, referente a percepção quimiossensorial de gosto (GLENDINNING, 1994), e o cultural, relacionado a interpretação, avaliação e validação local dos estímulos percebidos (BRETT e HEINRICH, 1998).

A percepção quimiossensorial de gosto é a capacidade de detectar e identificar estímulos químicos de gosto na cavidade oral. Essa habilidade possui bases genéticas e provavelmente pode ter favorecido a sobrevivência da espécie humana pois permitiu evitar a ingestão de compostos amargos na natureza, que geralmente são tóxicos (GLENDINNING, 1994). A percepção quimiossensorial de gosto amargo também pode ter sido importante no início do uso de plantas medicinais, haja vista as evidências na literatura etnobotânica de que o gosto pode ser uma das características que indica se uma planta tem potencial medicinal. Baseado nas ideias de Jonhs (1990) as bases da medicina humana se deram em nosso passado evolutivo, no uso de plantas para alimentação. De acordo com o autor, enquanto ingeriam as plantas na alimentação, os humanos identificaram alguns recursos que tratavam sintomas de doenças. assim, o estudo da percepção de gosto amargo em humanos pode nos fazer avançar no entendimento de como os humanos descobriram o uso de plantas medicinais (FERREIRA-JUNIOR et al., 2015).

A diversidade alélica do gene *TAS2R* (BEHRENS e MEYERHOF, 2006; MENNELLA et al., 2005) afeta as variações na percepção do gosto amargo, e seus fenótipos podem ser mensurados por meio do limiar de gosto amargo, que equivale a menor concentração de um estímulo químico amargo que um indivíduo consegue detectar numa solução. Assim, podem existir indivíduos com baixo limiar de gosto amargo (ou seja, com alta sensibilidade na detecção) e indivíduos com alto limiar. De acordo com Ferreira-Junior et al. (2015), indivíduos mais sensíveis na detecção e identificação do gosto amargo possivelmente eram os xamãs ou pessoas detentoras de vasto conhecimento sobre plantas medicinais e teriam participado

ativamente na construção das tradições médicas nas populações humanas. É possível, então, que essa variação ainda esteja relacionada com a seleção de recursos medicinais, principalmente atuando na atribuição de papéis sociais de xamãs e especialistas locais nos sistemas médicos tradicionais atuais.

Na espécie humana existem cerca de 25 receptores distintos da família *TAS2R*, que são ativos por diferentes ligantes químicos amargos. Essa alta diversidade de ligantes pode estar associada a diversidade de substâncias encontradas na natureza que determinam o gosto amargo das plantas, como polifenóis, alcaloides, terpenóides (DRAGOS e GILCA, 2018), metilxantinas e sulfonamidas (DREWNOWSKI, 2009). Desta forma, é possível que indivíduos mais sensíveis na percepção do amargo sejam favorecidos na identificação de plantas para tratar doenças e isso seja refletido em diferentes características das farmacopeias tradicionais.

Baseado nisso, com essa tese buscamos entender a importância do gosto das plantas no seu uso medicinal. Nos debruçamos sobre dois eixos principais: 1. a relação entre a percepção sobre as características de gosto das plantas por população locais e seu uso para tratar doenças. Aqui, buscamos entender se a percepção sobre os gostos das plantas está relacionada ao tratamento de doenças específicas e se a aceitabilidade de gostos varia de acordo com cada tipo de gosto presente nas plantas medicinais. E 2. o papel da percepção quimiossensorial de gosto amargo na seleção de plantas medicinais. Aqui, buscamos entender se essa característica biológica influencia alguns aspectos da seleção de plantas nos sistemas médicos locais, como a atribuição do papel social de especialista local, o hábito de descobrir novas plantas medicinais por meio da experimentação e a atribuição de indicações terapêuticas às plantas. No melhor de meu conhecimento, é o primeiro estudo empírico que busca entender se essa característica genética influencia diferentes aspectos da seleção e uso de plantas medicinais. Buscamos então contribuir com evidências que possam melhorar o entendimento de como ocorre a seleção de plantas medicinais.

1.2 ESTRATÉGIAS DE PESQUISA

No nosso estudo, empregamos duas principais ferramentas para medir as variáveis utilizadas para alcançar os objetivos da tese. A primeira delas foi a técnica *lista-livre* (ver ALBUQUERQUE et al., 2014), usada para registrar o conhecimento individual sobre as plantas medicinais. Optamos por essa técnica por ser uma das principais ferramentas utilizada nos estudos etnobotânicos, além de ser uma ferramenta simples, de fácil aplicação e replicação e que consegue gerar dados objetivos. Os indivíduos são estimulados, por meio de uma pergunta norteadora, a citar itens de determinado domínio cultural, que no nosso caso foi o conhecimento sobre plantas medicinais.

A segunda ferramenta foi o teste de limiar de identificação de gosto amargo, usada para identificar a sensibilidade na percepção de gostos dos participantes da pesquisa. A literatura já possui aproximações teóricas buscando explicar a relação entre a sensibilidade na percepção de gosto e o uso de plantas medicinais (FERREIRA-JUNIOR et al., 2015; JONHS, 1996), no entanto estudos empíricos ainda não foram realizados. Aqui, trazemos o primeiro estudo que mensurou a base biológica da quimiopercepção de gosto amargo *in situ* para buscar entender aspectos do uso de plantas medicinais, assim, consideramos ser a primeira aproximação empírica entre os campos de estudo da quimiopercepção de gosto amargo e da seleção de plantas medicinais.

Optamos por protocolos que já são validados na literatura de quimiopercepção. Assim, preparamos as soluções utilizadas nessa etapa seguindo o método proposto por Harris e Kalmus (1949) com a substância feniltiocarbamida (PTC), que é amplamente utilizada nesse tipo de estudo e mais facilmente comercializada no Brasil (ao invés do Propiltiouracil - PROP, substância que também é comumente utilizada para mensurar limiar de gosto amargo). E seguimos o protocolo de Mennella et al. (2005) para conduzir o procedimento para mensurar o limiar de gosto amargo.

1.3 ESTRUTURA DA TESE

A tese será organizada em quatro capítulos. No capítulo um, apresentamos a fundamentação teórica que está por trás de nossos questionamentos e objetivos. É aqui que explicamos com detalhes como ocorre a percepção quimiossensorial do gosto amargo e suas implicações para a espécie humana. E como o gosto das plantas está relacionado a sua seleção para tratar doenças.

No capítulo dois, apresentamos um estudo empírico sobre o papel da percepção de gosto das plantas no seu uso medicinal em cinco comunidades rurais do Nordeste do Brasil. Nesse estudo, não investigamos a percepção quimiossensorial do gosto, mas sim a forma como as pessoas percebem e qualificam os atributos de gosto das plantas. Os atributos de gosto seriam as definições individuais que as pessoas dão para os estímulos químicos de gosto que elas identificam nas plantas, baseadas em seu contexto ecológico e cultural. Com esse capítulo, buscamos contribuir com evidências que mostram que existe associação entre o gosto das plantas e seu uso para tratar doenças específicas, além de tentar compreender a aceitabilidade das pessoas aos diferentes perfis de gosto de plantas medicinais. Esse capítulo é necessário na tese porque nos ajuda a compreender com mais detalhes os aspectos locais da percepção de gosto e conhecimento sobre plantas medicinais nas comunidades estudadas.

No capítulo três, realizamos um estudo sobre a relação entre a percepção quimiossensorial de gosto amargo e características da seleção e uso de plantas em duas comunidades rurais do Nordeste do Brasil. Nesse estudo, mensuramos *in situ* o limiar de identificação de gosto amargo dos participantes para entender sua relação com o hábito ser experimentador de plantas medicinais, com o papel social de especialista local bem como na identificação do gosto das plantas medicinais.

Por fim, no capítulo quatro, apresentamos as considerações finais da tese que englobam as contribuições da tese para a ciência, as limitações do estudo e as propostas de investigações futuras.

CAPÍTULO I: FUNDAMENTAÇÃO TEÓRICA

1. PERCEPÇÃO DE GOSTO

O paladar é um dos sentidos pelo qual os seres humanos e outros animais percebem o seu ambiente (MENNELLA et al., 2013). Herbívoros comumente encontram substâncias e plantas tóxicas em sua alimentação e sua capacidade de identificar gosto pode ser uma estratégia para evitar o consumo desses recursos (GLENDINNING, 1994). Os seres humanos são capazes de identificar e distinguir cinco qualidades básicas de gosto: amargo, doce, salgado, azedo e umami. Essa capacidade pode ter desempenhado um papel importante na sobrevivência e bem-estar dos indivíduos pois fornecia informação útil para evitar a ingestão de compostos tóxicos e na busca por recursos energéticos. Assim, os genes que determinam variação na sensibilidade do gosto amargo provavelmente têm sido alvo de pressão seletiva desde o passado evolutivo dos seres humanos, selecionando indivíduos que conseguiam identificar substâncias potencialmente tóxicas por meio do paladar (BOYD, 1950; SORANZO et al., 2005).

As primeiras evidências de variação genética no paladar surgiram da descoberta da “cegueira do paladar” (*taste blindness*) (FOX, 1932). Em sua pesquisa, Fox (1932) evidenciou que alguns indivíduos percebiam soluções diluídas de feniltiocarbamida (PTC) com possuindo gosto amargo (*tasters*), enquanto outros não percebiam gosto algum (*non-tasters*). A partir dessa evidência, Harris e Kalmus (1949) propuseram um método para mensurar a capacidade de perceber gostos, baseado numa medida de limiar, utilizando também diluições de PTC. Esse método é utilizado ainda hoje em muitas pesquisas (FAREED et al., 2012; HONG et al., 2005; LEITE et al., 2018) e consiste em oferecer para os participantes da pesquisa degustarem soluções com concentrações crescentes de PTC e registrar as respostas. O limiar de gosto seria, assim, a menor concentração na qual um indivíduo perceberia um gosto distinto na solução de PTC.

Algumas bases genéticas da percepção do gosto já são conhecidas pela ciência. A diversidade alélica do gene *TAS2R* (BEHRENS e MEYERHOF, 2006; MENNELLA et al., 2005) afeta as variações na percepção do gosto amargo. Assim, existem indivíduos que naturalmente são muito sensíveis na percepção do gosto amargo, identificando o estímulo químico em uma solução mesmo que ele esteja em pequena concentração, e indivíduos pouco sensíveis, identificando o estímulo químico apenas em elevada concentração (MENNELLA et al., 2005). A diversidade alélica dos genes *TAS1R3* e *GNAT3* afeta a capacidade de perceber estímulos de gosto doce (JOSEPH et al., 2015). Essas bases biológicas estão envolvidas em diferentes comportamentos humanos como a rejeição do gosto amargo na população humana

(MENNELLA et al., 2005), a aversão de crianças por medicamentos amargos (MENNELLA et al., 2013), preferências alimentares (DUFFY et al., 2000; TEPPER, 1998) e baixa ingestão de vegetais (FORESTELL et al., 2017).

2. PERCEPÇÃO QUIMIOSSENSORIAL DE GOSTO AMARGO

O gosto amargo é muito estudado na literatura científica, principalmente devido ao comportamento inato de rejeição ao amargo, presente em algumas espécies animais (GLEDINNING, 1994; NISSIM et al., 2017), que pode estar relacionado ao fato de que na natureza o gosto amargo está relacionado a compostos tóxicos. Essa ideia é atualmente a mais aceita e muitos compostos tóxicos são conhecidos por possuírem gosto amargo (DREWNOWSKI, 2009). No entanto, nem todas substâncias tóxicas naturais são amargas (como o Tálho, ver PETER e VIRARAGHAVAN, 2005); e muitos compostos amargos podem trazer benefícios a saúde em concentrações nas quais são tipicamente consumidas, como a cafeína no café e os fenóis em vinhos e chás. Além disso, existem evidências de automedicação com plantas amargas entre primatas (HUFFMAN, 2003) e ruminantes (VILLALBA et al., 2014). É possível então que a capacidade de detectar o gosto amargo não seja apenas um mecanismo biológico para evitar a ingestão involuntária de substâncias amargas tóxicas (BEHRENS et al., 2018) mas possua também outras funções como a ingestão de itens com atividade biológica para tratar doenças em humanos e outros animais.

Os argumentos apresentados por Glendinning (1994) reforçam essa ideia. O autor examinou a relação entre o amargor e a toxicidade de plantas consumidas por mamíferos, avaliando os custos e benefícios ecológicos do comportamento animal de rejeição ao amargo usando estudos previamente publicados. Um conjunto de evidências mostra que existe consumo de plantas tóxicas entre os herbívoros e isso poderia revelar uma falha no mecanismo de detecção e rejeição de gosto amargo para evitar a ingestão desse tipo de alimento. Para Glendinning (1994) os herbívoros consomem esses itens quando: i) existe escassez de alimento, que levaria os animais a consumirem plantas não preferidas; ii) existe um delay na resposta a toxicidade, ou seja, os efeitos das toxinas são produzidos dias ou semanas após a ingestão e isso não resultaria na aversão a determinado item amargo e iii) as toxinas falham em provocar uma rejeição ao amargo em concentrações letais, então o comportamento não é expressado. Processos semelhantes podem ter ocorrido no passado evolutivo da espécie humana e, assim, permitindo a ingestão de compostos amargos que possuíam atividade biológica.

Muitas substâncias amargas encontradas em plantas possuem atividade biológica. O amargor das plantas pode ser determinado por uma grande diversidade de compostos, como os da classe dos terpenóides, flavonóides e taninos (HEINRICH et al., 1992), glicosídeos

cianogênicos, polifenóis, terpenóides e alcaloides (DRAGOS e GILCA, 2018), além de metilxantinas e sulfonamidas (DREWNOWSKI, 2009). No entanto, possuímos também uma grande diversidade de receptores da família TAS2R, localizados na cavidade oral, principalmente na língua e que podem ser ativados por diferentes estímulos químicos de gosto amargo (BEHRENS et al., 2009). Substâncias amargas encontradas em plantas, como os terpenóides andrografólido e amarogentina, ativam o receptor hTAS2R50 (BEHRENS et al., 2009). Anatomicamente, esses receptores estão organizados em estruturas multicelulares denominadas papilas gustativas (MENNELLA et al., 2013). Na língua, as papilas gustativas estão incorporadas em três tipos diferentes: as fungiformes, foliáceas e circunvaladas (MILLER, 1995). Cada célula receptora de gosto amargo expressa um subconjunto dos 25 genes humanos do receptor de gosto amargo (hTAS2Rs) e a maioria desses receptores possuem perfil de ligação com vários ligantes de gosto amargo (MEYERHOF et al., 2010).

Algumas variações na sensibilidade ao gosto amargo, além das genéticas, foram descobertas nos últimos anos. Existe variação ontogenética na sensibilidade de percepção de gosto amargo, com crianças sendo mais sensíveis que adultos (MENNELLA et al., 2005; MENNELLA et al., 2010; VENNERØD et al., 2018), com adolescentes no nível intermediário (MENNELLA et al., 2005), ou seja, a percepção de gosto é mais aguçada na infância e tende a atenuar-se ao longo do desenvolvimento ontogenético. Como a frequência alélica não muda da infância para a fase adulta, outros fatores, que não os genéticos, podem estar influenciando essa variação. Possivelmente, essa alta sensibilidade na infância está relacionada a identificação e aprendizagem precoce dos estímulos químicos de gosto presente no contexto em que a criança vive. A sensibilidade na percepção de gosto diminui marcadamente com o envelhecimento, e não apenas para o amargo, mas para os cinco gostos básicos (METHVEN et al., 2012). Evidências na literatura sugerem que, entre adultos, mulheres podem ser mais sensíveis do que os homens (BARTOSHUK et al., 1994). Já entre crianças, o sexo não parece promover diferenças na sensibilidade de percepção do amargo (MENNELLA et al., 2010). Também existem diferenças étnicas, com evidências de que hispânicos e afro-americanos são mais sensíveis que brancos não hispânicos, especialmente entre homens (Williams et al., 2016).

3. SELEÇÃO DE PLANTAS MEDICINAIS POR MEIO DO GOSTO

Um conjunto de evidências tem mostrado que o gosto das plantas é um forte atributo para identificar o seu potencial medicinal (ANKLI et al., 1999; BRETT e HEINRICH, 1998; CASAGRANDE, 2002; DRAGOS e GILCA, 2018; GECK et al., 2017; GILCA e BARBULESCU, 2015; MEDEIROS et al., 2015; MOLARES e LADIO, 2009; PIERONI e

TORRY, 2007). A seleção de plantas para tratar doenças pode ser guiada pelo gosto, haja vista que muitas substâncias que determinam gosto de plantas também possui atividade biológica (HEINRICH et al., 1992). Algumas evidências têm mostrado que o gosto é um importante característica utilizadas por populações humanas para distinguir plantas medicinais de plantas não medicinais (ANKLI et al., 1999; LEONTI et al., 2002). No entanto a casualidade não é muito clara. Ankli et al. (1999) não verificou diferenças na porcentagem de planta medicinais e não medicinais consideradas amargas; enquanto Casagrande (2000) encontrou uma evidência semelhante, indicando que mesmo que plantas com gosto amargo sejam amplamente utilizadas para tratar doenças gastrointestinais, o gosto amargo por si só não é suficiente para predizer o uso medicinal de uma planta. Isso vai de encontro a alguns achados da literatura que mostram que gosto amargo é uma característica particular de muitas plantas medicinais (Heinrich et al 1992) e que o gosto pode influenciar a seleção de plantas para tratar doenças específicas (MEDEIROS et al., 2015).

Algumas evidências mostram a importância do gosto na seleção de plantas para tratar doenças específicas. Morales e Ladio (2009) reportaram que problemas estomacais são tratados com plantas de cheiro forte e gosto amargo na região árida da Patagônia Argentina. Plantas com gosto amargo também são indicadas no tratamento de inflamações (Medeiros et al, 2015) e distúrbios gastrointestinais, como diarreia e disenteria (ANKLI et al., 1999; HEINRICH et al., 1992; MEDEIROS et al., 2015). As plantas doces são preferidas para o tratamento de doenças respiratórias (ANKLI et al., 1999). Mesmo em automedicação de primatas é possível verificar comportamento de automedicação em primatas para um conjunto de sintomas específicos (HUFFMAN, 2003).

Para tentar explicar o papel do gosto (e do odor) na seleção de plantas medicinais, Medeiros et al. (2015) propuseram três diferentes hipóteses. 1) o gosto das plantas medicinais são pistas de eficiência química da planta. O que determinaria o uso da planta seria a eficiência química e o gosto seria uma pista para que as pessoas identificassem essas plantas. 2) o gosto é determinante da seleção das plantas medicinais. Nesse caso, os seres humanos possuiriam uma capacidade inata para usar plantas com determinados marcadores de gosto, que seriam determinantes na sua seleção para tratar doenças. 3) gosto é um recurso mnemônico. Aqui, o gosto pode ser entendido como artefato de recordação e associação, e a associação gosto-indicação terapêutica serviria para que as pessoas recordem a indicação terapêutica da planta.

4. ACEITABILIDADE DE GOSTO DE PLANTAS MEDICINAIS

O gosto das plantas medicinais influenciar na sua aceitabilidade por populações humanas (CANIVENC-LAVIER et al., 2019; LESSCHAEVE e NOBLE, 2005; SIRÓ et al., 2008) e, assim, na sua ingestão (DINEHART et al., 2006). A presença de compostos amargos e adstringentes pode diminuir a aceitabilidade de plantas. Kaminski et al., (2000) evidenciou que quanto maior o amargor percebido em diferentes tipos de alimentos, menores eram os escores de aceitabilidade entre alunos da Universidade de Michigan. O gosto doce, por sua vez, está relacionado a maior aceitabilidade e ingestão de produtos vegetais. No entanto, comumente a interação com substâncias químicas que possuem outros gostos (geralmente amargo e adstringente), diminuem a doçura percebida, como diferentes terpenóides e alcaloides encontrados em plantas da medicina chinesa que inibem a percepção da doçura das mesmas (SUTTISRI et al., 1995).

A sensibilidade na percepção de gosto amargo pode influenciar a aceitabilidade de diferentes compostos químicos e gostos na população humana (DREWNOWSKI e ROCK, 1995). Fischer et al. (1961) encontraram evidências de que a sensibilidade no gosto amargo está ligada a aversão e baixa aceitabilidade de café e pães amargos. Kaminski et al. (2000) encontraram evidências de que indivíduos mais sensíveis na percepção de gosto amargo identificavam maior amargor em plantas, e isso resultava numa menor aceitabilidade desses recursos. De fato, bases genéticas podem influenciar a aceitabilidade e ingestão de plantas medicinais, no entanto, existem diferentes mecanismos que podem facilitar a ingestão de recursos que possuem pouca aceitabilidade. Alguns compostos fenólicos, por exemplo, interagem com proteínas salivares que ajudam a modular a percepção do gosto amargo (GLENDINNING, 2007; SOARES et al., 2018).

Outro mecanismo importante é a supressão de misturas, que é um fenômeno observado quando ocorre a mudança no gosto de substâncias quando elas são misturadas, mesmo que não haja reações químicas (BARTOSHUK, 1975). Quando ocorre a mistura de dois estímulos de gosto diferentes, suas intensidades são suprimidas, fazendo com que a intensidade percebida de cada estímulo na mistura seja menor que a percebida quando as substâncias são provadas fora da mistura (KEAST e BRESLIN, 2003). A maior parte das evidências na literatura sobre supressão de misturas é entre substâncias doces e amargas (GREEN et al., 2010; KAMEN et al., 1961; LAWLESS, 1977; PELLETIER et al., 2004). Substâncias com gosto doce, como a sacarose (GREEN et al., 2010) e a rebaudiosídeo (GAUDETTE et al., 2016), podem suprimir o amargor de soluções. De acordo com Gaudette et al. (2016) a supressão de misturas é um mecanismo importante para promover o aumento da aceitabilidade de compostos promotores

de saúde, levando a população humana a consumi-los mesmo se possuírem gostos com baixa aceitabilidade.

CAPÍTULO 2

THE ROLE OF TASTE PERCEPTION IN THE SELECTION OF MEDICINAL PLANTS IN RURAL COMMUNITIES IN NORTHEAST BRAZIL

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The role of taste perception in the selection of medicinal plants in rural communities in northeast Brazil

Temóteo Luiz Lima da Silva¹, Washington Soares Ferreira Junior², Ulysses Paulino Albuquerque^{1*}.

¹ Laboratory of Ecology and Evolution of Social-ecological Systems, Center of Biosciences, Department of Botany, Federal University of Pernambuco, Cidade Universitária, Recife, Brazil, Zip code 50.670-901.

² University of Pernambuco, BR 203, Km 2, Cidade Universitária, Petrolina, Pernambuco, Brazil, Zip code 56.328-903

*Correspondence should be addressed to upa677@hotmail.com

Abstract

In this study, we investigated whether people's perception of plant taste influences their choice of plants for the treatment of specific diseases, their consensus on the taste of plants, and their acceptance of medicinal plants with different taste profiles. We used the free listing technique to record local knowledge about medicinal plants in five communities in the Catimbau National Park, Pernambuco, Brazil. Individuals over 18 years of age, who agreed to participate in the research, shared their knowledge about the therapeutic properties and taste attributes of medicinal plants and their assessment of registered tastes. We examined the association between the taste of plants and the body systems for which they are indicated using the chi-square test. We examined the correlation between the plant use consensus and the taste attribute consensus using Spearman's correlation. We analyzed the acceptability of the plants' taste attributes using the chi-square test. We detected an association between bitter tasting plants and the treatment of digestive tract and respiratory system diseases and between bad tasting plants and the treatment of infectious diseases; however, these associations varied among the five communities, suggesting that they may be mainly influenced by cultural aspects. We also discovered that there was greater consensus on the taste attributes of more important plants. Sweet tasting plants were more acceptable, while bitter tasting plants were less acceptable. Mechanisms that minimize the aversion to bitter and disgusting tasting plants are likely to favor their intake and acceptability.

Introduction

Ethnobotanical research has shown that the taste of plants can influence their selection for the treatment of diseases [1, 2, 3, 4]. This organoleptic property is often determined by bioactive compounds that have pharmacological activity. Thus, taste could be an important indicator of the presence of certain bioactive compounds in the plant. In some societies, taste is important in distinguishing between medicinal and non-medicinal plants [5]. In addition, there is evidence that taste is used as a criterion to select plants to treat specific diseases. Plants perceived as bitter are generally used to treat digestive system problems [3, 6], while stomach problems can be treated with plants the taste and smell of which are perceived as strong by people [4]. The taste of plants can also be important in providing clues as to the best way to prepare and administer them [7].

The perception of plant taste by humans occurs in two simultaneous processes: the biological and the cultural. The first process refers to the chemosensory perception of taste. Humans are able to detect chemical taste stimuli [8, 9]. This is an important biological function, as it has made it possible to avoid the ingestion of potentially toxic substances, which generally taste bitter, in addition to favoring the discovery of beneficial chemical compounds [10]. The cultural aspect is associated with the interpretation, evaluation, and local validation of the perceived stimuli [1]. Together, these processes can guide human populations in experimenting with and selecting different plants for the treatment of different diseases.

In addition to being important in the selection of medicinal plants, taste can also influence affective responses to and the intake of medicinal plants [11, 12, 13]. For example, foods that contain polyphenols, which have bitter and astringent qualities, are not readily accepted or ingested by children [14]. On the other hand, sweet substances, such as sucrose, activate positive affective responses in humans [15]. These responses may have evolved in the human species as a mechanism for dealing with nature-derived chemicals; in nature, the bitter and sour tastes are associated with toxic compounds [8], while the sweet and salty tastes are associated with energy resources [15]. When it comes to medicinal plants, it is possible that the acceptability of a particular taste may influence how local populations choose plants for therapeutic purposes.

In this context, our objective was to investigate the importance of plant taste in their medicinal use in five rural communities in northeastern Brazil. Specifically, we aimed to: (1)

understand whether certain taste attributes of plants are associated with the treatment of specific diseases. Based on evidence from the literature, we believe that, proportionally, some taste attributes will be associated with a set of specific diseases, revealing the importance of this organoleptic property in local therapeutic choices; (2) check if there is a consensus on the perception of the taste of the species used. We believe that if taste plays an important role in the medicinal use of a plant, there will be a consensus among the five local communities on the taste of the most important plants; (3) assess the variation in the acceptability of different tastes (bitter, sweet, salty, and sour). Based on the assumption that in nature aversion to bitter and sour tastes is indicative of avoidance of toxic substances, we hypothesized that bitter, astringent and sour tasting plants would be evaluated as having bad taste attributes, while sweet and salty tasting species would be more likely to be evaluated as having a pleasant taste.

Materials and Methods

Study Area

The study was carried out in five rural communities located within the Catimbau National Park (PARNA Catimbau): Sítio Igrejinha, Sítio Muquém, Sítio Breu, Sítio Dor de Dente, and Sítio Açude Velho. PARNA Catimbau is a protected conservation unit (UC), instituted by decree of December 13, 2002. Even though this is a UC area that does not allow for the presence of human populations, many communities reside in this territory. These communities depend on native resources for their survival, such as firewood and other wood resources, game, edible plants, pasture, and medicinal plants. Currently, approximately 56, 20, 20, 7, and 10 families live in Sítio Igrejinha, Sítio Muquém, Sítio Breu, Sítio Dor de Dente, and Sítio Açude Velho, respectively. All communities depend on land use to develop their economic activities, which consist mainly of subsistence farming and goat farming.

PARNA Catimbau covers about 62,000 ha and is located in the municipalities of Ibirimirim, Tupanatinga, and Buíque, in the state of Pernambuco, 295 km from Recife, the state capital. Currently, the Brazilian Institute for the Environment and Renewable Natural Resources (IBAMA) is responsible for managing the UC, without however having formulated a management plan for it yet. PARNA Catimbau is located in the transition zone between the wild land and the hinterland, with a BSh-type climate that transitions to a tropical rainy climate, according to the Köppen classification [16], with rainfall ranging from 650 to 1100 mm.

The communities were chosen due to their dependence on the use of native resources to treat diseases, given that not all families had access to the public health system. Health care services were available in a small village outside the PARNA Catimbau (Vila do Catimbau),

which is approximately 15 km from the communities, with access by unpaved roads, as well as in the city center of Buíque, which is approximately 27 km from the communities. In this context, families with few financial resources were unable to travel frequently to receive health care services, thus medicinal plants represented important resources for the treatment of the diseases of these populations.

Legal aspects

The study was assessed by the Research Ethics Committee (CEP) of the University of Pernambuco, which granted a favorable opinion for the execution of the research (CAAE: 89890917.1.0000.5207), indicating that we followed the guidelines and recommendations of the national health council (resolution 466/12). Our study was also appreciated by the Biodiversity Authorization and Information System (SISBio), which granted authorization for the development of research at PARNA Catimbau (N° 55107), as it is a UC.

Participant selection and data collection

We visited all the houses of the two communities in search of residents over 18 years old in order to invite them to participate in the research. The 140 individuals who agreed to participate (Igrejinha = 92, Muquém = 23, Açude Velho = 10, Dor de Dente = 7, Breu = 8) signed the Free and Informed Consent Form, after understanding the objectives of the study. Then, each participant was invited to share information on their knowledge about plants used to treat diseases. Data collection was carried out between January and December 2017 by researchers from the National Institute of Science and Technology - Ethnobiology, Bioprospecting, and Nature Conservation (INCT), with which this study is associated. INCT researchers conducted a survey to assess the knowledge about and use of natural resources by all of the PARNA Catimbau local populations. Data were collected on knowledge about timber, food, medicinal, and synergistic resources, as well as on knowledge about risk perception and individual well-being. For our analysis, we only used the data collected about the medicinal plants used by the population.

We used the free listing technique [17] to register the medicinal plants known to the participants. The stimulus given was the following guiding question: which medicinal plants do you know? Then, for each plant mentioned, we recorded which conditions they were used to treat, in which forms they were used, which parts were utilized, and which were their taste attributes. Regarding the taste attributes, we recorded information according to the characteristics expressed by the participants, using words from the emic perspective [18]. In

order to assess the acceptability of the plants' taste attributes, we asked the participants to evaluate each taste mentioned by them as good, bad, or neutral.

We carried out guided tours [19] in places where people collect medicinal plants, to collect plants for taxonomic identification. The species were identified by a specialist from the Agronomic Institute of Pernambuco and the specimens were incorporated into the collection of the Herbarium Dárdaro de Andrade Lima (IPA), in the same institution.

Data analysis

We used the chi-square test to check whether different sets of diseases were treated with medicinal plants with certain taste attributes. To this end, we categorized diseases according to the following World Health Organization classifications [20]: endocrine diseases, infectious and parasitic diseases, gastrointestinal diseases, injuries, general symptoms and signs, circulatory system diseases, respiratory system diseases, urogenital system diseases, and neoplasia. In addition to these categories, we included three other categories to cover the diversity of diseases mentioned: veterinary diseases, cultural syndromes, and others. We considered cultural or spiritual illnesses, such as open chest, cleanliness of the body, evil eye, etc., as cultural syndromes. The others category encompassed diseases that were not specific or did not fit into the other categories (*i.e.*, weakness in the body, warmth in the body, and aesthetic diseases). Similar taste attributes were grouped into broader taste categories, in order to not underestimate taste perception categories. For example, sweet and sweetie were included in the sweet category, bitter fine, mild bitter, and bitter were included in the bitter category, and astringent, lock, and mild astringent were included in the astringent category. Finally, the number of taste citations was recorded for each body system and then compared using the chi-square test.

We used the same body system classification to calculate the relative importance (IR) of the species with the method of [21] using the following formula: $IR = (NSCE/NSCEV) + (NPE/NPEV)$, where NSCE is the number of body systems treated by species x, NSCEV is the total number of body systems treated by the most versatile species, NPE is the number of diseases treated by species x, and NPEV is the total number of diseases treated by the most versatile species. The IR of the species calculation provides information on which plants were most versatile in the treatment of diseases by taking into account both the number of diseases as well as the number of different body systems that the plants treated.

We used Spearman's correlation to verify whether the most widely used species by the participants were those that had the greatest agreement in terms of taste perception. The

consensus on plant use (CU) among the participants was calculated by means of the plants' relative frequency (number of participants that cited the species $i \times 100/\text{total number of participants}$) [22]. Similarly, we calculated the participants' consensus on taste attributes (CG) (total number of citations of the taste attribute for species $i \times 100/\text{total number of participants who cited species } i$) [23]. For the correlation test, we used only the plants that were reported by at least five participants, given that plants that were mentioned by a few individuals could have higher CU and CG values.

To assess whether the acceptability of taste varies according to the type of taste, we calculated the sum of the qualities of taste (good or bad) attributed to bitter, astringent, sweet, salty, and sour tastes. We used the chi-square test to check if there were differences in the proportion of the qualities attributed to each type of taste. We considered the tastes that received a statistically higher attribution of the good quality as being more acceptable, while the tastes to which the bad quality was attributed were viewed as less acceptable.

Results and Discussion

Medicinal species and their taste attributes

We registered 114 medicinal plants that were associated with 45 different taste attributes and were recommended for the treatment of 211 different diseases. The most important plants in terms of IR were *Ximenia americana* L. (IR = 2), *Aloe vera* L. (IR = 1.85), *Hymenaea courbaril* L. (IR = 1.71), *Myracrodruon urundeuva* Allemão (IR = 1.60), *Amburanacearensis* A.C. Smith (IR = 1.57), *Croton heliotropiifolius* Kunth (IR = 1.50), and *Dysphania ambrosioides* (L.) (IR = 1.49) (Table 1).

Among the attributes of reported tastes, bitter had the highest number of citations (42.34%), followed by the astringent (26.78%), sweet (8.04%), and sour (1.84%) tastes. As we took into account the emic classification of the participants, many of the taste attributes mentioned only made sense locally and it was not possible to categorize them into the five scientifically established basic tastes (bitter, sweet, sour, salty, and umami). A set of attributes cited showed that people perceived that species have taste, but are unable to discriminate it (*i.e.*, yes/have taste, taste of itself, a different taste etc.). Other attributes were placed in the comparative category, in which the participants made a direct association with known tastes (*i.e.*, acerola, ginger, wet clay, sugar cane, mint). Even when we tried to delve deeper and understand better the association between plant tastes and known tastes, the known taste description was the only possible and sufficient for the participant. Another grouping of taste attribute was evaluations of taste rather than taste itself (*i.e.*, tasty, neutral, normal, light,

smooth, strong, etc.). Many participants (15.03%) also mentioned not knowing whether certain plants had a taste and about 11.74% of participants mentioned that certain plants had no taste at all.

Plant taste and use of medicinal plants

Our results showed an association between plant taste and some body systems in the five communities studied. At Sítio Igrejinha, gastrointestinal tract diseases were treated with bitter tasting plants (chi-square = 12.23; $p = 0.002$). Respiratory system diseases, as well as their symptoms and general signs (*i.e.*, cough, pain, etc.), were treated by sweet tasting plants (chi-square = 7.34; $p = 0.02$). At Sítio Muquém, genitourinary system diseases were treated with bitter tasting plants (chi-square = 0.125; $p = 0.0104$). At Sítio Açude Velho, infectious and parasitic diseases were treated exclusively by bad tasting plants; the same was true for Sítio Dor de Dente (chi-square = 30.25; $p = 0.0001$). At Sítio Breu, general symptoms and signs were treated with bitter plants (chi-square = 13; $p = 0.0046$). We did not detect an association between the taste attributes and the other bodily systems in any of the five communities.

The literature suggests that bitter tasting plants are used to treat diseases in all body systems. In the case of gastrointestinal disease treatment, the literature has already indicated an association with bitter tasting plants [2, 3, 5, 6]. One of the main effects of bitter plant compounds on the digestive system is the stimulation of the digestive tract's motility [24], in addition to their antibacterial, antifungal, antioxidant, and hematoprotective properties [25]. There is also evidence that astringent plants are used to treat various infections [24, 26], corroborating our results.

Although we detected an association between certain tastes and disease groups, our results do not allow us to make inferences about chance in the selection of plants, especially as we found different associations between taste and diseases in the five communities studied. Preferences, interpretations, and taste assessments are mainly determined by culture and can be learned through cultural familiarity with the plants used, as argued by [24]. In a study carried out in two groups with marked cultural differences in the Amazon, [27] observed that there were similarities between the identification and classification of tastes as well as between the ecological and taxonomic characteristics of the pharmacopoeias of the two groups; however, there were differences in the taste assessments of the two pharmacopoeias, as well as in the plants used, their form of administration, and their therapeutic purposes. Thus, even though the psychophysiological perception of chemical compounds is similar, different human groups can interpret the taste of medicinal plants differently, thus influencing their use in medical systems.

Based on this evidence and our findings, it is possible that the associations between the taste and the therapeutic purpose of medicinal plants depend more on cultural and individual aspects than on other mechanisms (*i.e.*, the biological perception of taste).

When dealing with bitter taste, our findings showed great expressiveness in all analyzed body systems, although it was not proportionally the most important taste in all systems. A great diversity of bioactive compounds determines the bitter taste in plants [28], so it is possible that bitter plants are selected through experimentation to treat specific diseases and are subsequently tested for other therapeutic indications. Therefore, the mnemonic association between the bitter taste and the plant is not lost.

CU and CG

We found a strong positive correlation between the CU and the CG of plants ($r_s = 0.62$; $p = 0.003$). This means that the species with the greatest CU according to the participants were those with the highest agreement in terms of their CG. The species that showed the greatest CU were *X. americana* L. (CU = 63.63), *H. courbaril* L. (CU = 60.60), *M. urundeuva* Allemão (CU = 57.57), and *Sideroxylon obtusifolium* (Roem. & Schult.) T.D. Penn. (CU = 57.57). The species with the greatest CG were *Ziziphus joazeiro* Mart. (GC = 75.75), *Ximenia americana* L. (GC = 60.60), *A. vera* L. (CG = 54.54), *M. urundeuva* Allemão (CG = 48.48), *Prosopis juliflora* (Sw.) DC. (CG = 42.42), and *Croton conduplicatus* Kunth (CG = 42.42). All the plants with higher CG values were predominantly bitter; the characteristic of bitter taste is probably striking and easily spread. The relevance of these species in terms of CU and CG may be associated with the great diversity of chemical compounds that have a bitter taste, given that different classes of compounds that have pharmacological activity, such as terpenoids, polyphenols, alkaloids [29], methylxanthines, and sulfonamides [28], can determine the bitter taste in plants.

Acceptability of the taste of medicinal plants

Our results indicated a greater acceptability of the sweet taste (chi-square = 74 $p = 0.0001$), while bitter tasting plants were less acceptable (chi-square = 15.06 $p = 0.001$), that is, the participants proportionately evaluated the sweet taste as being good and the bitter taste as being bad. We found no differences between the acceptability of sour, astringent and, salty tasting plants. The results on the acceptability of sweet and bitter tastes corroborate evidence from the literature [14, 30] and indicate that the intake of bitter tasting medicinal plants or even foods, which however are beneficial to health, may be discouraged [14]. However, the literature suggests that people can respond positively to bitter taste if there is health related information, that is, they can suppress the taste preference for health [30, 31].

There are processes that can promote a greater acceptability of bitter or disgusting tastes, making them more acceptable and palatable. One of these processes is taste habituation, which refers to an increase in the acceptability of a taste after repeated exposure to it [8, 13]. In a study by [32], children aged 4 and 5 years old were exposed to a new food (tofu) twice a week, for 9 weeks, in 3 different contexts: one group tried simple tofu, another group tried sweetened tofu, and the third tried salted tofu. Rather than preferring the sweet version, as the sweet taste is biologically more pleasant and preferred [15, 33, 34], the children learned to prefer the version they were exposed to compared with the other versions. Possibly, the children learned to interpret the family stimulus as normal after repeated exposures. It is possible, therefore, that bitter tasting plants may be widely accepted and used by local populations.

Another process that can influence the acceptability of the taste of plants is mixture suppression. This process occurs when two different taste stimuli are mixed and the perceived intensity of each stimulus in the mixture is less than the perceived intensity if the same stimuli were presented individually [35]. Some studies provide evidence of the occurrence of mixture suppression in relation to different taste attributes [36, 37, 38, 39]. A study by [37] on mixture suppression showed that the sweet taste of sucrose can reduce the bitterness of caffeine when mixed in a solution. Other sweet substances, such as rebaudioside, can also decrease the bitterness of solutions [12]. In the communities studied, adding sugar and/or honey to infusions, as well as using various plants in medicinal preparations (liquors and bottles), were common practices. Adding sugar to liquors and mixing different plant species in the preparation of herbal medicine could be a strategy employed by locals to deal with the taste of medicinal species, in an effort to make them more palatable and acceptable.

Conclusions

Even though the different communities examined existed in a similar environmental context, their associations of plant taste and specific diseases varied; this suggests that these associations may be mainly influenced by cultural aspects. Although bitter tasting plants were less acceptable in the studied communities, psychological and cultural mechanisms can minimize the aversion to bitter and disgusting tastes, suggesting that bitter taste is not an impediment to the intake of medicinal plants.

Data Availability

The datasets used and analyzed during the current study are available by sending email to the corresponding author.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Authors' Contributions

TS, WSFJ and UPA contributed equally to this study.

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Table 1. List of medicinal plant species of greater relative importance from the five communities studied in PARNA Catimbau (Northeast Brazil) and their respective common names, therapeutic indications and taste attributes.

Scientific name	Relative Importance	Common name	Therapeutic indication	Taste attributes
<i>Aloe vera</i> L.	1.85	Babosa	headache, body ache, fever, flu, hair, inflammation, worm, hemorrhoid, phlegm, injury, cough, cancer onset	bitter
<i>Amburana cearensis</i> A.C. Smith.	1.57	imburana de cheiro	inflammation, flu, cough, fever, uterus inflammation, prostate, tiredness, cold, phlegm, bruise	bitter, tasty, astringent, sweet
<i>Anacardium occidentale</i> L.	1.21	cajueiro roxo	whole body inflammation, vaginal infection, injury, inflammation, woman inflammation, heart	astringent, bitter, sour, like acerola
<i>Commiphora leptophloeos</i> (Mart.) J.B. Gillett	1.47	imburana de cambão	stomach ache, belly ache, flu, hemostatic, heart, injury	sour, astringent, bitter, sweet

<i>Croton conduplicatus</i> Kunth	1.39	quebra-faca do sertão	itching, bellyache, toothache, stomach, constipation, inflammation, digestion problems, liver, spine, flu	bitter
<i>Dysphania ambrosioides</i> (L.) Mosyakin & Clemants	1.49	mastruz	blow, inflammation, spine, twist, injured, side pain, flu, fracture, cough, worm, healing, gastritis, prostration, phlegm	Green leaf, bitter, distinctive, burning
<i>Hymenaea courbaril</i> L.	1.71	jatobá	cough, bronchitis, fertility problems (man and woman), stomach, inflammation, anemia, flu, infection	bitter, tasty, astringent, distinctive, tasty, sweet, like honey
<i>Maytenus opaca</i> Reissek	1.22	bom nome	kidneys, Spine, Stroke, Cancer	Bitter, astringent, sour
<i>Myracrodruon urundeuva</i> Allemão	1.60	aroeira	inflammation, belly ache, spine, swollen belly, flu, injury	astringent, bitter, kind of soft
<i>Myroxylon peruiferum</i> L.f.	1.10	bálsamo	astringent, bitter, kind of soft	rancid, bitter, sweet, astringent, weird, dry.

<i>Ocotea odorifera</i> (Vell.) Rohwer	1.29	sassafrás	fever, belly pain, gastritis, inflammation, kidneys, infection, general pain, rheumatism, ulcer	different, bitter, taste of ground, astringent, strong, nauseous, bad, sweet.
<i>Sideroxylon obtusifolium</i> (Roem. & Schult.) T.D. Penn.	1.21	quixabeira	bang, inflammation injury, backache, healing, cut, woman inflammation, fall	like milk, bitter, astringent, sour
<i>Ximenia americana</i> L.	2	ameixa	wound, inflammation, infection, bath, itching, cut, sore throat, skin inflammation, blood thinner, phlegm, flu, cough	bitter, astringent, distinctive, rancid
<i>Ziziphus cotinifolia</i> Reissek	1.1	juazeiro	dandruff, flu, cough, phlegm, brushing teeth, inflammation, fever, cold, headache, skin	bitter, good, taste of ground, astringent

CAPÍTULO 3

IS THERE A BIOLOGICAL BASIS IN THE SELECTION OF MEDICINAL PLANTS IN THE HUMAN SPECIES? AN INITIAL APPROACH BASED ON CHEMOSENSORY PERCEPTION OF TASTE

Artigo publicado no periódico Ethnobiology and Conservation

IS THERE A BIOLOGICAL BASIS IN THE SELECTION OF MEDICINAL PLANTS IN THE HUMAN SPECIES? AN INITIAL APPROACH BASED ON CHEMOSENSORY PERCEPTION OF TASTE

Temóteo Luiz Lima da Silva^{1*}, Washington Soares Ferreira Junior², Ulysses Paulino Albuquerque^{1*}

¹ Laboratório de Ecologia e Evolução de Sistemas Socioecológicos, Centro de Biociências, Departamento de Botânica, Universidade Federal de Pernambuco, Cidade Universitária, Recife, Brazil, 50.670-901.

² Universidade de Pernambuco, BR 203, Km 2, S/N, Cidade Universitária, Petrolina, Pernambuco, Brazil, 56.328-903

Corresponding author:

E-mail address: TLLS (timoteobio@gmail.com), UPA (upa677@hotmail.com)

Abstract

The ability to identify tastes associated with plant chemicals may have favored humans in identifying plant chemists with pharmacological activity throughout human evolutionary history. The genetic basis of taste perception influences people's varying sensitivity to perceive chemical stimuli of taste. This biological basis can play an important role in plant selection to compose local medical systems, given the argument in the ethnobiological literature that plant taste can influence their selection as a medicinal resource. Thus, we aimed to understand whether this biological basis influences on the selection of medicinal plants. Our investigation was made through the survey of ethnobiological data on the knowledge of medicinal plants and sensitivity data on the perception of bitter taste in two local communities. We tested whether local experts and active experimenters of medicinal plants are more sensitive to the perception of bitter taste than the rest of the population. Additionally, we evaluated whether this biological basis influences on the number of citations of plants with taste and on the versatility attributed to medicinal plants. Our assumptions were not corroborated by our results. It is likely that the bitter taste threshold is not relevant for the selection of medicinal plants.

Evolutionary Ethnobiology - Ethnobotany - Taste threshold - Taste perception - Organoleptic property - Local medical systems

Introduction

The ability to detect tastes is a feature present in humans and other animals that enables them to perceive and deal with the surrounding chemical environment (Glendinning 1994; Mennella et al. 2013). Some behaviors, such as rejection of bitter taste and acceptability of sweet tastes, are innate and conserved in different animal species (Glendinning 1994; Hayes and Johnson 2017). When it comes to bitter taste rejection, this behavior represents an important biological function as it may allow humans to avoid involuntary ingestion of toxic compounds, as bitter taste is naturally associated with generally toxic secondary metabolites (Glendinning 1994).

The allelic diversity of the *TAS2R* gene (Behrens and Meyerhof 2006; Mennella et al. 2005) affects variations in the perception of bitter taste. Thus, there are individuals who are naturally very sensitive in the perception of bitter taste, identifying the chemical stimulus in a solution even if it is in small concentrations, whereas low sensitive individuals identify the chemical stimulus only in high concentration (Mennella et al. 2005). Bitter taste receptor cells are located in the oral cavity, mainly in the tongue, and have a set of *TAS2R* family receptors that can be activated by different chemical stimuli (Behrens et al. 2009). The *TAS2R* gene have probably been target of selective pressure since the evolutionary past of humans, favoring individuals who could identify toxic compounds and regulate their intake, avoiding poisoning (Richerson and Boyd 2005; Soranzo et al. 2005).

Through the taste threshold, it is possible to objectively measure the phenotypic expression of *TAS2R*. Two thresholds can be measured: taste detection threshold, which is the lowest concentration at which a substance can be distinguished from water (Harris and Kalmus 1949; Hong et al. 2005); and the taste identification threshold, which is the lowest concentration at which the taste quality of a substance can be identified (e.g. bitter, sour, etc.) (Hong et al. 2005). Thus, the taste threshold is a measure inversely proportional to the sensitivity in the perception of taste. The lower the threshold of an individual, the greater their sensitivity to perceive tastes. There is evidence in the literature that found a relationship between the bitter taste identification threshold and the other tastes threshold, especially sweet (Chang et al. 2006; Pasquet et al. 2002) and salty tastes (Pasquet et al. 2002).

The perception of bitter taste does not only appear to be a biological mechanism to prevent involuntary ingestion of toxic bitter substances (Behrens et al., 2018), but may play an important role in selecting plants for human populations and influencing the construction of local pharmacopoeias. Evidence has shown that plant taste is a strong attribute for identifying

their medicinal potential (Ankli et al. 1999; Brett and Heinrich 1998; Casagrande 2000; Dragos and Gilca 2018; Geck et al. 2017; Gilca and Barbulescu 2015; Medeiros et al. 2015; Molares and Ladio 2009). The reason for this is likely to be linked to the fact that compounds having biological activity (Behrens et al. 2009; Drewnowski and Gomez-Carneros 2000) determine the taste of plants. For bitter taste, for example, there is evidence of two terpenoids found in Chinese medicinal plants (andrografolide and amarogentin) that are known to have bitter taste by activating bitter taste receptor cells (Behrens et al. 2009). Besides bitter taste, among the ethnobotanical evidences, Ankli et al. (1999) and Leonti et al. (2002) reported the importance of plant taste attributes for the distinction between medicinal and non-medicinal plants among local populations. Morales and Ladio (2009), in turn, reported that stomach problems are treated with strong-smelling and intense taste, usually sweet plants, in the arid Patagonia Argentina. Bitter-tasting plants are also indicated for the treatment of inflammation (Medeiros et al, 2015) and gastrointestinal disorders such as diarrhea and dysentery (Ankli et al. 1999; Heinrich et al. 1992; Medeiros et al. 2015).

Plant taste is generally related to certain secondary compounds such as terpenoids, flavonoids, tannins and other chemical components (Heinrich et al. 1992) that have pharmacological activity (Brett and Heinrich 1998). However, the same taste attribute may be related to different chemical compounds. The bitter taste of plants, for example, can be determined by the presence of compounds of the following classes: cyanogenic glycosides, polyphenols, terpenoids and alkaloids (Dragos and Gilca 2018), as well as methylxanthines and sulfonamides (Drewnowski 2009). Individuals with lower thresholds of bitter taste who could distinguish different tastes in plants, and thus make more taste-therapeutic indication associations, could best distinguish the diversity of chemical compounds. Thus, we believe that individuals who have a higher sensitivity to the perception of bitter taste would assign more therapeutic functions to medicinal plants, increasing their versatility in medical systems.

The process of selecting plants to treat diseases can be interpreted as an adaptive strategy, considering that diseases can be considered selective pressures that have always been present in human history (Wiley and Allen 2009). Throughout evolution, the pursuit of plants for nutrition has led humans to develop strategies to deal with plant chemicals to maximize the nutritional benefit of low intake of toxic compounds (Jonhs 1990, 1996), directly influencing individual survival. In his model of human chemical ecology, the development and transmission of practices for dealing with toxic compounds, such as plant fermentation and domestication, played a key role in minimizing the effects of toxins while searching for food (Johns 1990; see also Ferreira Júnior and Albuquerque 2018). This may have facilitated the safety of tasting with

a larger set of new plants and favored the identification of resources that alleviated disease symptoms. Humans began to make an association between the taste of plants and their chemical substances, being able to identify possible toxins and plants with pharmacological properties (Ferreira Júnior and Albuquerque 2018).

It is possible that people with lower taste thresholds played a key role in the early use of plants to treat disease. Ferreira Junior et al. (2015) argue that, during human evolution, people who were genetically more sensitive to the perception of bitter taste were more likely to associate the taste of a plant with its medicinal properties and that they were possibly shamans or holders of vast knowledge on plants used for food and medicine. Nowadays, it is likely that these social roles would still be related to the bitter taste threshold, as this feature would facilitate the performance of their role in local medical systems. Heinrich (1994) reported that local specialists from Sierra Mixe in Oaxaca (Mexico) use taste and smell as criteria to decide which plant is useful for treating a particular disease. The authors argue that these organoleptic characteristics may guide specialists in the search for new medicinal plants.

In this study, we aimed to understand whether the biological base of chemosensory perception plays a role in the selection of medicinal plants in local populations in northeastern Brazil. Specifically, we are choosing the Bitter Taste Identification Threshold - which represents the phenotypic expression of the *TAS2R* gene - as our biological based measure to test some hypotheses related to the medicinal plant selection process. To the best of our knowledge, this is the first study to measure the bitter taste threshold *in situ* and to relate it to the selection of medicinal plants. We investigated the bitter taste threshold due to the large amount of knowledge in the literature on evolutionary, physiological, behavioral and genetic aspects of bitter taste perception. In addition, the bitter taste threshold correlates with the threshold of other tastes (Chang et al. 2006; Gent and Bartoshuk 1983; Pasquet et al. 2002).

We hypothesized that the biological base of chemosensory perception influences in assigning the social role of the local expert. We consider as experts those locally recognized as having extensive knowledge on medicinal plants. We expect people who play this social role to have lower thresholds of bitter taste than non-specialists. Alternatively, we hypothesize that the biological base of chemosensory perception influences in the habit of being an active experimenter of medicinal plants. In our study, the active medicinal plant experimenter is that individual who, regardless of his social role in the medical system, usually tastes new plants to identify their medicinal potential. This hypothesis does not nullify the first, since local experts and experimenters may use similar biological mechanisms in plant selection to treat diseases. Thus, we expect that an active experimenter of medicinal plants will present a lower bitter taste

threshold compared to a non-experimenter. We also hypothesized that the chemosensory perception of taste influences on the selection of medicinal plants by taste. We expect an inverse relationship between the bitter taste threshold and the citation number of bitter and tasteful plants, that is, the higher the sensitivity of an individual, the more tasteful plants they cite. Finally, we hypothesized that individuals with lower bitter taste thresholds would assign more therapeutic targets to plants. We also expect an inverse relationship between our variables, that is, the higher the sensitivity of experimenters, the more therapeutic targets they will cite to the plants.

Material and Methods

Study area

Our study involves an experiment that usually takes place in reserved lab rooms or research centers. However, this is an *in situ* study conducted in two rural communities in northeastern Brazil. We selected communities that are already part of a larger project, coordinated by researchers from the National Institute of Science and Technology - Ethnobiology, Bioprospecting and Nature Conservation (www.inctethnobia.com/), to which this study is linked. The purpose of INCT is to meet the demand for systematic studies on the nature-society interface related to ethnobiology, bioprospecting and nature conservation.

We selected the local communities Sítio Igrejinha and Sítio Muquém (here referred to as Igrejinha and Muquém, respectively), located within the Catimbau National Park (Figure 1). The PARNA Catimbau covers approximately 62,000 hectares and is located in the municipalities of Buíque, Ibirimirim and Tupanatinga, in the state of Pernambuco, approximately 300 km from Recife (state capital). The vegetation is typical of Caatinga, a seasonally dry forest with semi-arid climate (BSH according to the Koppen classification). The region has an average temperature of 23°C and annual average rainfall ranging from 486 mm to 975 mm. PARNA Catimbau does not have a management plan yet, even though it was created since 2002 (a decree of December 13, 2002). There are still human populations living inside the park, distributed among 11 communities, even though this category of Conservation Unit does not permit the presence of human populations or private areas.

In Igrejinha and Muquém there are approximately 56 and 20 families, respectively, living mainly from subsistence farming and goat raising. The population of Igrejinha and Muquém depends on natural resources for some basic demands such as the use of woody

resources as plant fuels and for the construction of houses and fences, the collection of food plants and, above all, the use of plants to treat diseases. These populations have already experienced several conflicts with the management of PARNA Catimbau due to their dependence on natural resources to meet their subsistence demands. Because of this conflict, there is a fear among the people of the region to share information with researchers about the use of natural resources, especially hunting and woody resources. In addition, the population also has to deal daily with water shortages, which is a common problem in various locations in Brazil's semiarid region.

Both communities do not have a school, community association, health center or hospital. Health care is provided in Vila do Catimbau (a small village of 2,240 inhabitants (IBGE 2000) which is the main access to PARNA Catimbau and is 12 km from city center Buíque and approximately 15 km from the surrounding communities) or in the city center of Buíque. Thus, they are inaccessible to many families who do not have the financial resources to move frequently to these places. Medicinal plants end up being the most accessible resource for primary health care. These communities were chosen because of their receptiveness to the research group of the Laboratory of Ecology and Evolution of Social-ecological Systems that has been conducting studies at PARNA Catimbau since 2017. The communities also have a history of using medicinal plants to treat various diseases.

Ethical Aspects

We conducted our study following the rules and guidelines of the National Health Council (Resolution 466/12), through the Research Ethics Committee (CEP) of the University of Pernambuco, which gave a favorable opinion for the execution of the research (CAAE: 89890917.1.0000.5207). Our study was also appreciated by the Biodiversity Authorization and Information System (SISBio), which granted authorization to undertake the research at PARNA Catimbau (No. 55107), as it is a Conservation Unit.

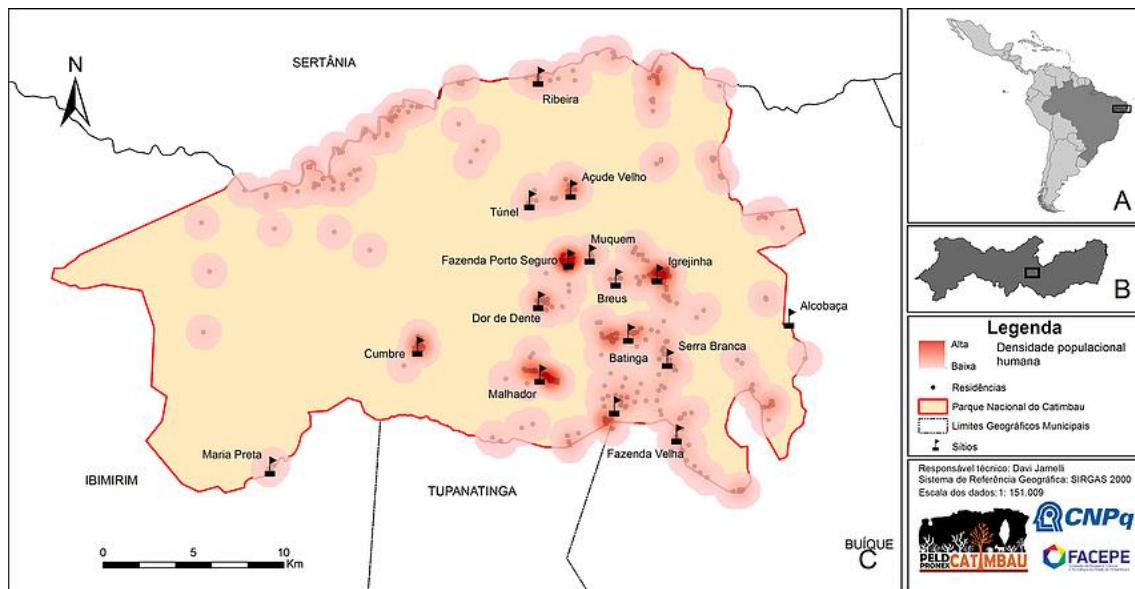


Figure 1. Location of the communities Sítio Igrejinha and Sítio Muquem in the Catimbau PARNA. (Source: PELD Catimbau / <https://www.peldcatimbau.org/>).

Recruitment of participants and data collection

We conducted visits in all homes of the two communities (16 in Muquem and 51 in Igrejinha) to invite people over 18 years old to participate in the study. We explained to each participant the purpose of the research and those who agreed to participate were invited to sign the Informed Consent Term (ICT). We performed data collection in two steps, not necessarily consecutive. One of the steps was the use of the free listing technique (see Albuquerque et al. 2014) to record the medicinal plants known to each participant (Figure 2a). We carried out this step with other researchers who are also associated with the INCT, as well as gathering other information about the use of natural resources in the region. The guiding question of the technique was "which medicinal plants do you know?" Subsequently, we asked on the therapeutic indications of each plant and its taste attributes. This was a long phase, which took place between January and November 2017, and involved all 11 communities located within PARNA Catimbau. The total number of participants who agreed to participate in this phase were 32 (84.21% of the population over 18 years old) people in Muquem and 84 (75% of the population over 18 years old) people in Igrejinha. This was the total number of people we invited to participate in the second phase of the survey in both communities. The second stage of data collection involved the execution of the experiment on bitter taste identification threshold, designed to determine how sensitive each participant is to the perception of bitter taste, in addition to identifying local experts and active experimenters in the communities.



Figure 2. Illustration of procedures performed in local communities in northeastern Brazil. A. Free listing of medicinal plants; B. Logistics to transport the materials of the bitter taste threshold experiment; C. Participant of the study during bitter taste threshold experiment. (Photos: first author).

We measured the bitter taste identification threshold using phenylthiocarbamide (PTC) solutions diluted in distilled water. PTC is a synthetic compound and is not found in nature (Tepper 1999), but the ability to detect PTC is correlated with the ability to test other bitter compounds that occur in nature (Bartoshuk et al. 1988; Drewnowski 2009; Wooding et al. 2004). We acquired PTC in its commercial form from “ACS Científica”. Dilutions were prepared according to the method of Harris and Kalmus (1949). Firstly, we prepared a 13% PTC solution that was our stock solution and from it we made 13 serial dilutions at a 1:2 ratio. Thus, the most concentrated solution, “C1”, had 1300 mg/L while the most diluted concentration at C14 was 1.6 mg/L. The solutions were prepared at the Laboratory of Applied Ecology and Phytochemistry, Federal University of Pernambuco. All solutions were stored at 4°C in amber flasks and brought to room temperature prior to use. We pre-tested the bitter taste threshold experiment to validate concentrations, test possible dynamics, and get an idea of how long each procedure would take. The pre-tests were conducted with students from the Petrolina Campus of the University of Pernambuco.

Before the start of the experiment we ensured that the participant was fasting for at least 1 hour. When this did not happen, we sought to schedule visits, advising participants to keep fasting until the scheduled time. Additionally, we made sure that the participant was healthy,

did not receive prescription drugs or received medical treatment, and, in the case of women, was not pregnant or nursing. The experiments were conducted individually, avoiding distractions and influences from others as much as possible.

To determine the threshold of identification of bitter taste we followed the procedures proposed by Mennella et al (2005). We offered 5ml of PTC test solution in 50ml disposable cups. The first cup offered contained only distilled water as our control measure. The second cup offered contained solution C14 (0.16 mg/L). Participants were instructed to bring the contents of the cup to their mouths, to rinse and spit, never swallowing (Figure 2c). When trying the cup with the first solution, we asked, "Does it taste like water, yes or no?" If the participant answered "yes", the procedure continued with the offer of the next solution (C13 = 0.065 mg/L), in ascending order of concentration. If the participant answered "no", we would ask "do you feel any taste?". If the participant answered yes (it had a taste), we would ask what the taste of that solution was like. If they stated that the solution had a bitter taste, the test would be interrupted, and that concentration would be considered his bitter taste identification threshold. If the participant could not identify the bitter taste, we would offer the next solution, in ascending order of concentration, until it reached the concentration where the bitter taste in the solution could be identified. Participants always rinsed their mouth with distilled water before tasting each new concentration. The total number of participants who agreed to participate in the experiment were 27 people in Muquém and 9 people in Igrejinha. We conducted the bitter taste threshold experiments between October 2018 and January 2019.

We sought to identify active medicinal plant specialists and experimenters among the people who participated in the first stage of the research. We identified the specialists through local legitimation of this social role by asking each participant who they consider to have extensive knowledge of medicinal plants. Thus, we considered as a specialist the participant who was cited by at least 25% of the total sample (nine people). The dataset showed a very clear threshold, which separates some individuals (eight) from the rest of the sample that received citation. We identified eight experts in our sample. We needed to innovate to measure the habit of an active experimenter as there is no validated method in the literature. Active experimenters were identified by self-declaration through the following objective question: "Do you have the habit of tasting new plants to find out if they are medicinal?" To make sure that the participants understood the question, we asked a second question with a brief contextual example: "There are curious people who like to test new plants. Do you have this habit? We then consider as an experimenter the participant who held the affirmative answer after the second question. We identified seven experimenters in our sample. Although we cannot rule

out the possibility of a type of error due to the sample size, we sought the independence and randomness between the samples with methodological rigor, ensuring the quality of the data obtained.

Data analysis

We tested the normality of our data using the Kolmogorov-Smirnov test and found that none of the variables that would be tested had a normal distribution. Thus, we chose to use only nonparametric tests in all analyzes. We applied the Mann-Whitney test to test the hypothesis that there is a biological basis in assigning the social role of medical systems specialist. We opted for the Mann-Whitney test because it is able to determine if samples from both groups (experts and non-specialists) were selected from populations that have the same distribution. We used the same test to see if there were differences in the bitter threshold between people who have the habit of tasting medicinal plants (experimenters) and those who do not have this habit (non- experimenters).

We used the simple linear regression test to verify if there is a relationship between the number of citations of plants with taste and the threshold of bitter taste. We opted for this test because it is able to verify if two variables are related. For this, we counted the plants that were cited by each participant with some attribute of taste, even if it was a comparative taste (e.g. mint taste, earth taste, etc.) or when the participant stated that the plant had an unknown taste. We did the same procedure to check whether there is a relationship between the number of citations of bitter and astringent plants and the taste threshold. We have decided to analyze the bitter and astringent tastes together because there is already evidence in the literature that many bitter-tasting secondary compounds also cause a sense of astringency in the mouth, such as phenols and alkaloids (Shepard Jr. 2004). Most likely, the plants that were cited as astringent by the participants also have a bitter taste.

We used the simple linear regression test to verify if there is a relationship between the versatility attributed to plants and the bitter taste threshold. We then calculated the average versatility for each participant by dividing the sum of the number of therapeutic targets cited by the number of plants cited. For this analysis, we selected only the most popular plants, avoiding possible idiosyncrasies from the free lists. This selection was based on the citation frequency of plants, selecting those that were present in at least 25% of the lists. This was based on the evidence found in the literature. Morales and Ladio (2009) showed that the most frequently used species in Lake Rosario are those that have more detailed taste descriptions to

distinguish them. Medeiros et al. (2015) showed an association between plant taste and therapeutic indications only when they made a similar cut, based on popularity.

We performed again the same analyzes to test our hypotheses (except hypotheses involving experts and experimenters) using this selection of most popular plants. We performed all tests using R software, version 3.2.4 (R Core Development Team 2010). Table 1 presents a compilation of the ideas we tested, with the data and tests that were used.

Table 1. Hypotheses tested to evaluate the role of the bitter taste threshold in the selection of medicinal plants in local communities in northeastern Brazil.

Question	Data	Test
1. Is there a biological basis in assigning the social role of medicinal plant experts in medical systems?	Bitter taste threshold of experts and non-specialists	Mann-Whitney
2. There is a biological basis in the habit of being an active experimenter of medicinal plants.	Bitter taste threshold of experimenters and non-experimenters	Mann-Whitney
3. Individuals with lower bitter taste threshold cite more tasteful plants	Bitter taste threshold and number of tasteful plants cited	Simple linear regression
4. Individuals with lower bitter taste threshold cite more plants with bitter taste	Bitter taste threshold and number of plants with bitter taste cited	Simple linear regression
5. Individuals with lower bitter taste thresholds indicate more therapeutic targets for plants	Bitter taste threshold and average participant versatility	Simple linear regression

Results

Participant Profile and Descriptive Results

Our sample consisted of 19 women and 17 men, aged between 18 and 81 years old, averaging 49. All participants identified bitter taste at one of the different concentrations, so

our sample had no non-tasters (individuals who cannot detect chemical stimuli of taste). More than half of the participants identified bitter taste in the first six concentrations, with C10 (2.54 mm/L) and C9 (5.08 mm/L) concentrations being the ones with the highest frequency (41.66% of the total).

Medicinal plants and their taste attributes

We cited 97 medicinal plants indicated to treat 141 distinct therapeutic targets. *Ximenia americana* L. was the most cited plant, reported by 71.4% of participants, followed by *Myracrodruon urundeuva* Allemão (54.3%), *Myroxylon peruiferum* LF (51.4%) and *Hymenaea courbaril* L. (48.57%). The most popular medicinal species and their respective therapeutic targets and taste attributes are listed in Table 2. Bitter taste had the highest number of citations (28.25%), followed by astringent (23.8%), "tasteless" (9.04%) and sweet (3.71%). Among the most popular plants, all had some taste attribute.

Hypothesis results

Our results showed that the bitter taste threshold does not play a key role in assigning the social role of local specialists and selecting medicinal plants. We refute the hypothesis that the biological base of chemosensory perception influences in assigning the social role of local expert, since local experts ($n = 8$) are no more sensitive than non-experts ($n = 28$) in the perception of bitter taste, as we expected ($W = 92.5$; $p = 0.464$). We also refute the hypothesis that the biological base of chemosensory perception influences in the habit of being an active experimenter of medicinal plants, since there are no differences in the bitter taste threshold of active experimenters ($n = 7$) and non-experimenters ($n = 29$) of medicinal plants ($W = 123.5$; $p = 0.384$).

We refute the hypothesis that individuals with lower bitter taste thresholds cite more plants with taste (regression $F=0.325$, $p=0.578$). We also refute the hypothesis that individuals with lower bitter taste thresholds cite more plants with bitter taste (regression $F=1.049$, $p=0.3140$), as well as the hypothesis that individuals with lower bitter taste threshold indicate more therapeutic targets for the plants (regression $F=1,157$, $p=0.696$). Our results showed that differences in the number of tasting plants citations, citations of bitter tasting plants and the versatility attributed to plants are not related to the bitter tasting threshold. When we analyzed the same variables (number of citations of plants with taste and bitter taste) only among the most frequent plants, we also found no relationship with threshold of bitter taste.

Discussion

We do not corroborate the hypothesis that the biological basis of chemosensory perception is important to assign the specialist's social role in medical systems, given that local specialists did not present significant differences of bitter taste threshold from non-specialists. Thus, it is likely that other mechanisms are related to the attribution of this social role nowadays.

According to Wiley and Allen (2009), the way in which attribution of the social role of local experts varies between cultures is related to an individual vocation or spiritual heritage. Among the Luo, one of Kenya's largest ethnic groups, one can train an apprentice to become a local expert's successor by developing an herbal knowledge base used to treat a wide variety of therapeutic targets (Wiley and Allen 2009). To be considered a good candidate, among other characteristics, the learner needs to be intelligent, have a good memory, have a good heart and have a willingness to listen and learn. In different regions of Tanzania, the social role of healers, herbalists, midwives, and other types of local specialists may have different paths, such as inheritance within the family, initiation by ancestral spirits, the experience of being cured by the use of medicinal plants or even by their own decision after a period of learning (Prince and Geissler 2001). Based on this evidence, it can be seen that the social role of local medicinal plant specialist can be culturally assigned and not clearly rooted on the chemosensory perception of taste. Future studies could test the role of the bitter taste threshold in other medical contexts and systems, also considering the existence of this diversity of social role assignment as a local specialist. We must also consider that the selection of medicinal plants to form pharmacopoeia is not guided only by the taste (Bennet 2007; Hart 2005).

We believed that even if the attribution of the social role of local expert was culturally legitimated, it had the biological basis of the bitter taste threshold, as this characteristic could favor the selection of medicinal plants and, therefore, the performance of their role in the medical system. Based on our findings, it is likely that these social roles are currently unrelated to the bitter taste threshold. If we think about a hypothetical scenario, a low threshold of bitter taste may have been important in our evolutionary past, when humans began to use medicinal plants to treat diseases, highlighting individuals in the population who could make more associations between plant taste and the symptoms of the diseases that could be treated from it. This may have brought recognition among peers and, later, the confidence of the population in the search for this individual to treat their diseases. Subsequently, recognition and trust in these individuals came to be designated and maintained by cultural mechanisms such as cultural transmission and symbolic and religious aspects.

Our findings also suggest that the habit of experiment medicinal plants does not have a biological basis. The process of experimenting with new medicinal plants can be risky, especially when it involves plants with bitter taste that may have high toxicity (Glendinning 1994). Therefore, an experimenter with a low bitter taste threshold could make it easier to identify the tastes of medicinal plants and still avoid ingesting toxic compounds. Thus, regardless of the social role in medical systems, the bitter taste threshold is not related to this profile of individuals who usually try medicinal plants. Ferreira Junior et al. (2015) argue that, in our evolutionary past, individuals who were more sensitive in the perception of bitter taste acquired knowledge about medicinal plants through experimentation, associating the taste of a plant with its medicinal property. Later this knowledge was culturally transmitted to other individuals.

Plant selection by experimenting and identification of bitter taste may be more relevant in populations living in unstable environments and dealing, for example, with local plant extinction or the frequent presence of new diseases. There is evidence that in unstable environments, the process of individual knowledge production is favored (Richerson and Boyd 2005), and that in stable environments individuals more often acquire information through different channels of cultural transmission (Richerson and Boyd 2005; McElreath and Strimling 2008). Thus, in nomadic and semi-sedentary societies, such as hunter-gatherer societies, which deal with new environments more often than sedentary societies, one would expect to observe the relationship between the habit of tasting new plants to identify their medicinal potential and a low bitter taste threshold. In these social contexts, the ability to detect bitter taste would be crucial in identifying the pharmacological and toxic potentials of plants.

However, Soldati et al. (2015) showed that in the populations of three traditional communities in northern Minas Gerais - Brazil, the process of individual knowledge production through experimentation was not favored in situations of social and environmental instability. The authors recorded a low expression of the experimenting process that could be related to the confidence in the use of previously known and validated plants, to the detriment of the risk of tasting with new plants. If there is a low proportion of experimenters (as in the population studied here) and also a low frequency of this behavior, understanding whether the process of tasting medicinal plants is related to the bitter taste threshold presents a challenge to current science.

We do not corroborate the hypothesis that the bitter taste threshold is related to a higher number of citations of bitter plants and plants with other tastes. Although many studies report that taste is used by traditional populations to select plants for medicinal use, the causal

relationship is unclear (Casagrande 2000; Heinrich et al. 1992). Heinrich et al. (1992) reported that astringent-tasting plants are generally useful in treating dysentery and diarrhea in the Mixe ethnic group (Oaxaca, Mexico). However, not all plants that treat these therapeutic targets are astringent. Casagrande (2000) reports similar evidence indicating that even though bitter-tasting plants are widely used to treat gastrointestinal disorders, bitter tasting alone is not sufficient to predict the use of a medicinal plant to treat a disease. Another explanation to not having our hypothesis corroborated is that perhaps plant taste is just a mnemonic resource, not a determinant of its usefulness, in treating therapeutic targets (Casagrande 2000; Medeiros et al. 2015). According to Medeiros et al. (2015), taste, as well as other plant characteristics (smell, texture, color), can be understood as artifact of remembrance and association, and association between taste attributes and therapeutic indication would serve to remind people of the therapeutic indication of the plant.

We do not support the hypothesis that the bitter taste threshold is related to the versatility attributed to plants. We believed that individuals who are more sensitive to bitter perception could perceive the peculiarities of taste determined by the diversity of chemical compounds and thus make more associations between the perceived taste of plants and their therapeutic indications. Some participants in our study even reported that some plants have different tastes even if they are classified as bitter, that is, there are peculiarities in the bitter taste of plants. However, this has not resulted in a greater number of therapeutic indications for them nor is it related to the bitter taste threshold. Also, for this set of plants, people reported the taste attribute only as “bitter”. This brings us to a difficulty in studies of taste perception which is the linguistic dimension of taste. The scientific classification of five primary tastes (bitter, sweet, salty, sour and umami) is in many cases limited to represent local taste denominations. The Hindi vocabulary, for example, consists of six basic tastes: sweet, sour, salty, bitter, spicy and astringent (Gollin 2004). In our study, we have registered local denominations for tastes that do not fit the five basic tastes of Western science, such as “nauseated”, “pampered” and “different”. These terms probably hide a much greater diversity of perceived chemical stimuli than those expressed verbally.

The linguistic dimension of taste is also important for information sharing within the local medical system. According to Osawa and Ellen (2014), even if people taste the same stimulus of taste, how it will be classified is influenced by the fact that people know the terms of taste to describe what they have tasted. Additionally, to what extent they have opportunities to share and compare taste experiences in order to agree at a pattern of the use of terminology. Thus, it is possible that a person can perceive a taste but cannot express anything other than the

terms used in their culture. It is possible, then, that many taste sensory experiences are not shared in a population, being limited by local terminological diversity to classify the types of tastes.

Finally, it is possible that other factors, especially cultural ones, may help to understand the role of the bitter taste threshold in the selection of medicinal plants. Some authors have already stressed the importance of culture in understanding the biological process of taste perception (Brett and Heinrich 1998; Casagrande 2000; Hladik et al. 2002). According to Brett and Heinrich (1998), the perception of chemosensory stimulus is a physiological process, and its interpretation, evaluation and validation is determined by culture. For Shepard Jr (2004), sensations can be understood as biocultural phenomena rooted in human physiology, however, they are also constructed through individual and cultural experiences.

Study Limitations

Our sample did not have the planned size due to the difficulties faced to carry out this type of study in non-laboratory conditions beyond the population conflict with the current management of PARNA Catimbau. One of the challenges faced was the concern that individuals had to participate in the research because they thought it could endanger their health. In fact, the bitter taste threshold experiment is a bit invasive, requiring the participant to taste a liquid that is unknown by them. Thus, our sample was smaller than that commonly found in laboratory studies, which have a larger number of volunteers. This diminished the power of our analyzes.

It should be noted that the proportion of specialists is lower than that of non-specialists in traditional populations, and our sample comprised only eight specialists. A low proportion of experimenters is also expected as tasting new plants by trial and error carries risks to individuals (Soldati et al. 2015). Finally, even if we have a good sampling, we do not control the degree of kinship among the participants which may influence the phenotypic expression of chemosensory taste perception. Future studies could try to map the degree of kinship between individuals to reduce possible bias in the interpretation of their findings.

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CAPÍTULO 4: CONSIDERAÇÕES FINAIS

4.1 PRINCIPAIS CONCLUSÕES

Nossos achados corroboram um amplo conjunto de evidências de que determinados gostos das plantas estão associados a determinados sistemas corporais. No entanto, as associações entre gosto e grupo de doenças variou entre as cinco comunidades, inseridas num contexto ecológico semelhante. Isso possivelmente indica que grupos humanos podem fazer interpretações distintas do gosto das plantas e que o contexto cultural pode ser mais relevante para as associações entre gosto e finalidade terapêutica de plantas medicinais que outros fatores, como a percepção quimiossensorial de gosto.

Apesar da literatura apresentar uma argumentação teórica indicando que a percepção quimiossensorial de gosto amargo poderia influenciar aspectos da seleção de plantas medicinais, não corroboramos essas ideias em nosso estudo empírico. Nossos achados mostram que especialistas locais e experimentadores ativos de plantas medicinais não são mais sensíveis na percepção de gosto amargo, e que pessoas mais sensíveis na percepção de gosto não reconhecem maior número de plantas por meio do gosto ou atribuem mais indicações terapêuticas para as plantas. Isso indica que embora o aparato biológico seja necessário para o processo da percepção de gosto, nosso estudo apresenta uma forte evidência de que a seleção de plantas por meio de gosto provavelmente depende de outros fatores, como o contexto cultural das sociedades estudadas.

4.2 CONTRIBUIÇÕES TEÓRICAS E/OU METOLÓGICAS DA TESE

Nosso estudo ajuda a compreender que a percepção quimiossensorial de gosto amargo não influencia alguns aspectos da seleção de plantas medicinais por meio do gosto. Embora haja uma boa argumentação teórica na literatura relacionando essa base biológica ao início do uso de plantas medicinais, provavelmente nos sistemas médicos atuais essa característica não é mais relevante que aspectos culturais e individuais.

Além disso, utilizamos uma metodologia que é simples em relação as outras existentes na literatura para medir a percepção quimiossensorial de gosto amargo. Embora possua alguns desafios logísticos, especialmente na coleta de dados *in situ*, é possível incorporar esses métodos em estudos de etnobiologia para melhor entender aspectos da seleção de plantas medicinais por meio do gosto.

4.3 PRINCIPAIS LIMITAÇÕES DO ESTUDO

O fato de o teste de limiar de gosto amargo ser invasivo, levou muitas pessoas a declinar do convite para participar da pesquisa, e isso influenciou o tamanho de nossa amostra. Caso conseguíssemos uma amostra maior poderíamos englobar uma maior quantidade de variação do fenômeno. Além disso, não foi possível mapear o grau de parentesco entre os participantes da pesquisa para identificar se isso estaria influenciado de alguma forma nossos resultados.

Outra limitação é o fato de utilizarmos as classificações êmica para registrar os atributos de gosto citados pelos participantes pois a diversidade de gosto foi diluída na grande diversidade linguística local para designá-los. No entanto, sugerir que o participante enquadrasse os atributos citados dentro das categorias de cinco gostos básicos poderia superestimar alguns itens que localmente não fazem parte do mesmo conjunto de atributos de gosto, e isso poderia enviesar fortemente os resultados.

4.4 PROPOSTAS DE INVESTIGAÇÕES FUTURAS

Futuros estudos poderiam buscar entender se a percepção quimiossensorial de gosto influencia a seleção de plantas por populações sedentária e semisedentárias, uma vez que essas comunidades costumam lidar com ambientes que variam no tempo e espaço. Além disso, um estudo que abranja maior número de comunidades poderia aumentar a representatividade de especialistas locais na amostragem, permitindo realizar uma análise mais robusta entre a atribuição desse papel social e a percepção de gosto amargo.

4.5 ORÇAMENTO

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R\$440. Os custos acima estão ligeiramente subestimados por não incluírem o tempo gasto com preparação logística ou com planilhamento, digitalização de entrevistas e organização de banco de dados.

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