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Cover Page Footnote

I thank Amalia, Aurora, and Maria Luisa Astorga, Horacio Liñeiro, Manuel Monroy, Laura Monti, and Gary Nabhan for their help while in the Seriland. I am deeply in debt with Arli De Luca and Alberto Burquez who have long heard and helped polish the idea that aposematism can effect human cognition. Thanks to Diogo Samia for providing me with the R code for calculating COI and to Dr. Marco Augusto Miranda-Ackerman for assisting me through the use of R. The final manuscript received helpful comments from Lisa L. Price, precise edits from Cathy Moser Marlett and kind and thoughtful comments from anonymous reviewers. I thank the support of El Colegio de Michoacán and Jesus Medina, Research Assistants who greatly helped with the minute details of the final manuscript.

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Crib Notes

Aposematism: An Alternative Hypothesis on How We Discover Medicinal Organisms

Nemer E. Narchi

ABSTRACT

The most widely accepted theories as to how primates select medicinal organisms are based on smelling and tasting bitter—and most of the times toxic—plants (Etkin and Ross 1982; Johns 1990). In primates, however, the senses of smell and taste are not as dominant as is vision. I present a perspective claiming that aposematism, the conspicuous coloration displayed by toxic organisms, may play an important role in human recognition of medicinal organisms. This paper represents an open invitation for ecological anthropologists and ethnobiologists to generate empirical data that can support the aposematic hypothesis of medicine selection.

INTRODUCTION

Early in the academic life of anthropologists, introductory courses and textbooks suggest vision as being the most prominent of the senses used by primates (e.g., Kottak 2008:136; Relethford 1997:153-154). Such ideas are a mere reflection of the general trend in research, namely that the adaptive significance of primate color vision is seen almost exclusively in terms of foraging advantages (Miller 2002:59).

In this paper, I examine the idea that primate color vision, namely binocular stereoscopic trichromic vision, is not only a successful adaptation to facilitate hand-eye coordination for object manipulation or the recognition of ripe fruits and tender leaves (Melin et al. 2013), but a broader and more powerful adaptation capable of sending large chunks of environmental information about the existence of secondary metabolites in the surrounding environment. In doing so, I propose that aposematism, a conspicuous display that organisms possessing chemical defenses use as a warning signal to would-be predators (Poulton 1890), enables humans, and perhaps other primates, to select, conceptualize, and use medicinal organisms. I provide empirical examples from marine and terrestrial environments. Finally, I sketch a plausible methodology to test the hypothesis that there is a visual detection of medicinal organisms

WITH BITTER HERBS THEY SHALL EAT

Ethnobiologists have long struggled to explain how different human societies independently developed local pharmacopoeias (Berlin and Berlin 1994:242). However it is a highly supported claim that humans use perceptual givens to discern and organize biodiversity (Berlin 1992:9).

Organizing biodiversity is a quotidian practice that shapes human behavior and decision-making processes by means of linking schemas, models, and contingencies (Gragson and Blount 1999:xv). In this regard, organoleptic qualities, mainly smell and taste, are used to construct schemas that play an important role in the primal selection of medicinal plants around the world (*cf.* Johns 1990; Huffman 2001; Bennett 2007).

Biologically active ethnomedicines are detected by the characteristic smell and taste of the compounds that give them their bioactivity (Johns 1990:160). The smell-taste explanation for detecting medicinal organisms helps us understand why medicinal plants with little or no nutritional value are domesticated and retained in gastronomic traditions. However useful, the smell-taste hypothesis is problematic when medicinal organisms are not readily accessible. Such is the case with marine organisms, since their collection, with the sole purpose of smelling or tasting them, would involve a comparatively higher energy investment plus potential physical damage.

Trying to smell or taste an organism while being exposed to tidal flows and wave action may well be a hard task to accomplish. It seems that a taste-smell theory for marine ethnomedicine is rather implausible in such settings. However, what would be the outcome if the human brain were equipped with another intuitive and practical strategy to identify organisms rich in bioactive compounds, namely color vision?

To make it clear, let's portray ourselves trekking in search of medicinal plants along with a knowledgeable collaborator. The first thing our collaborator does when faced with a promising medicinal plant is to visually recognize the plant, pluck a leaf, tear it into pieces, begin smelling it and then eat it. It is a see, smell, and taste process of recognition.

APOSEMATISM, BIOACTIVE COMPOUNDS, AND HUMAN INGENUITY

The adaptive coloration of living organisms serves three major functions: concealment, disguise, and warning (Cloudsley-Thompson 1999). Aposematism, the conspicuous coloration that organisms possessing chemical defenses use as a warning signal to would-be predators (Lev-Yadun 2009; Terrick et al. 1995), is a well-documented phenomenon in nature, and a widespread trait in invertebrates, reptiles, and amphibians.

Conspicuous organisms create or sequester diverse toxins aimed at deterring predators (Savitzky et al. 2012). It is very likely that aposematic patterns of pigmentation are a direct result of complex biosynthetic pathways (Bandaranayake 2006) that blend in associate coloration, toxicity and diet specialization in noticeable but still poorly described manners (Santos et al. 2003).

Humans are very good at recognizing aposematism; a number of dangerous organisms appear conspicuous and colorful to us (Inbar and Lev-Yadun 2005). Perhaps one of the best examples of detection and use of aposematic organisms in local ecological knowledge is that of *Phyllobates terribilis*, the poison-dart frog (Saporito et al. 2007). First reported scientifically in 1824, the use of *P. terribilis*' poison as an arrow primer still survived as a common practice among Embera and Noanamá people in Colombia as recently as 1978 (Myers et al. 1978).

P. terribilis is considered to be the most toxic of the Colombian poison-dart frogs as it has about 1000 µg of batrachotoxins per adult frog skin (Daly et al. 2005). Batrachotoxins are unique alkaloids first characterized from the skin extracts of Dendrobatidae frogs. Five of the species in the Dendrobatidae family (genus Phyllobates) are known to carry these toxins, while simpler lipophilic alkaloids are known to occur in the skin of frogs in Dendrobates, Epipedobates, and Minyobates genera (Daly1998). In skin extracts from *P. terribilis,* three major alkaloids are present; batrachotoxin, homobatrachotoxin and batrachotoxinin A (Tokuyama and Daly 1983). Batrachotoxin is among the most potent natural products known to date (Dumbacher et al. 2004). The toxin irreversibly affects sodium channels which are essential for the transmission of electric signals, causing nerve and muscle depolarization, fibrillation, arrhythmias, and heart failure (Bosmans et al. 2004).

There are two known procedures used to obtain the frog's poison. In the most commonly known method, the frogs are impaled on a stick and held close to a fire while scraping the dart tips against the animal's skin. In a second method, the frogs are held down with a stick without being killed. In this method, fire is used only for drying the dart tips after they were impregnated with the toxin (Myers et al. 1978).

It is widely known that plants (e.g., Schultes and Hoffman 1979), minerals (e.g., Yang and Guan 2008), and terrestrial fauna (e.g., Costa Neto 1999) are well known sources of medicine for rural and indigenous people. However, these have not been the only source of medicine for humans throughout history. Lev (2007) and Voultsiadou (2010), among others, have provided substantial evidence to affirm that humans have given a medicinal use to marine algae, invertebrates, and fish for at least 5,000 years.

Marine ethnomedicines are found in intertidal and shallow subtidal environments (see Narchi 2013) where they lie in plain sight. Aposematism has been well documented for marine invertebrates (Ritson-Williams and Paul 2007), including ascidians (Young and Bingham 1987), echinoderms (Jones and Mallefet 2010), gastropods (Rosenberg 1989), flatworms (Ang and Newman 1998), and zoanthids (West 1976). Aposematism has also been documented to occur in fish (Smith 1992). It should not be surprising if aposematism is a common characteristic of marine ethnomedicines.

Marine chemists and natural product researchers sometimes use a selection criteria that closely resembles aposematic cues. In an interview for UCSD´s Exploration Magazine, professor John Faulkner said: "So if we go out onto the reef and we see something that looks like a large chunk of food –poorly protected, soft bodied, and easy to grab- and nothing is eating it, then we assume it has chemical protection" (Howard 1997:14).

CONSPICUOUSNESS IN MARINE ORGANISMS RICH IN BIOLOGICALLY ACTIVE METABOLITES

Many cultures, past and present, use marine medicines (Narchi et al. 2015a). Among these, the modern Seris (Comcaac), a fishing society native to the Central Gulf Region of the Sonoran Desert, still practice an extensive use of marine resources (Bertsch and Marlettt 2011). Seri marine pharmacopoeia is comprised of vertebrates, macro-invertebrates, macro algae and sea grasses, including at least 22 organisms (Narchi et al. 2002).

Previous research shows that Seri marine medicinal knowledge is at least as successful in detecting bioactive metabolites as is sampling using a chemical ecology approach[1] (Narchi 2003). Crude extracts of 16 marine organisms used in Seri medicine display bioactivity against brine shrimp in lethal concentrations (LC_{50}) between 0.0158 $^{mg}/_{ml}$ and 7.94 $^{mg}/_{ml}$. These results include bright red (*cheel*) extracts obtained from *Rhizophora mangle* (LC_{50} 0.0158 $^{mg}/_{ml}$) and *Maytenus phyllanthoides* (LC_{50} 0.123 $^{mg}/_{ml}$) (Narchi 2011).

In other cultures, red (blood-like) color is often mentioned as the sole efficacious property in treating illnesses (Price and Narchi 2015), many times following a doctrine of signatures (Dubick 1986). While documenting the role of senses in medicinal plant selection and therapy in two Amazonian societies, Matsigenka and Yora, Shepard (2004) noted the salient role that red and purple Acanthaceae, Gesneriaceae, and Nyctaginaceae plants have in the pharmacopoeia of both societies.

Shepard (2004) explained that red latex was related to blood and used against blood-related ailments. He provided no further explanation for the purple plants other than highlighting the importance of visual cues for sensorial ecology. In contrast, Seri use of medicines has no explicit explanation as to the mechanisms and/ or doctrines explaining the use of remedies; these are simply used "because they work" (Felger and Moser 1974: 415).

PLANTS AND APOSEMATISM

Plant aposematism has been largely ignored in research (Lev-Yadun 2009). In arid regions, physical structures spinescence, spines, prickles, and trichomes are common means of antiherbivory in thousands of species (Halpern et al. 2007); however, chemical defenses are also common for plants living in such regions. It seems, therefore, that resource availability determines the amount and type of plant defense (Coley et al. 1985). Similarly, conspicuous plants receive less damage due to predation (Strauss et al. 2015), suggesting plant aposematism.

Plant aposematism can easily be observed in deserts where some plants keep their green foliage throughout the year (Lev-Yadun and Ne'eman 2004). In such a regard, Seri pharmacopoeia offers numerous examples of plant aposematism. Many estuarine and desert plants used as medicine (see Narchi et al. 2015b) display a startling and easily distinguishable green color that contrasts with the desert background. One good example is *Ambrosia salsola* (Figure 1), called by the Seris *caasol cacat*, literally "bitter *caasol*."

FIGURE 1. *Ambrosia salsola*, called by the Seri *caasol cacat*. A tea made with the stem is used to heal swollen parts of the body and respiratory ailments. The tea can also be used as a topical solution against skin infections and as a wound cleanser. In recent years, Seri women use *A. salsola* as raw material to create medicinal soaps which they sell to outsiders.

A METHOD LINKING APOSEMATISM WITH ETHNOMEDICINES

The association between aposematic coloration and toxicity is noticeable, yet poorly described to the present date. Nonetheless, it is clear that the link between toxicity and aposematism results from the interactions between complementary morphological and physiological processes. Therefore, one can hypothesize that conspicuous ethnomedicines should exhibit some kind of correlation linking their chromatic contrast and their toxicity. The following section describes a simple method to test that hypothesis.

Measuring aposematism

A number of publications deal with how to measure aposematism (e.g., Prudic et al. 2007 and references therein). Many of the methods portrayed in these publications are not easily accessible to the average ethnobiologist, who would need an easy-to-carry spectrophotometer, illumination devices, analysis software, and the proper training to operate all of the above stated. Nonetheless, Samia and Francini (2015) have developed an affordable method that allows for the detection of aposematism by combining standardized field photographs with a post hoc software analysis. The method uses a fixed film ISO (800) and aperture priority (f5.6) to then determine the degree of animal-substrate contrast expressed in the form of a color-overlapping index (COI).

Measuring toxicity

There is a plethora of accessible and easily performed toxicity tests. While in the field I have used a simplified version of the brine shrimp test (see Sam 1993). The basic idea behind the test is that brine shrimp are exposed to successive dilutions of the extracts obtained from medicinal organisms. After 24 hours the analyst counts how many shrimps survived the test. Dead/alive quantities are plotted on the Y axis against the known concentrations of the extract (X). Dead/alive curves intersect and the analyst observes the X value for the intersection. The X value under the intersection (Figure 2) will be considered as the concentration at which 50 percent of the brine shrimp population is dead (LC_{50}). Researchers not willing to undergo the burden of performing a field LC_{50} test can obtain toxicity data by logging into a natural product database such as NAPRALERT (http://napralert.org/).

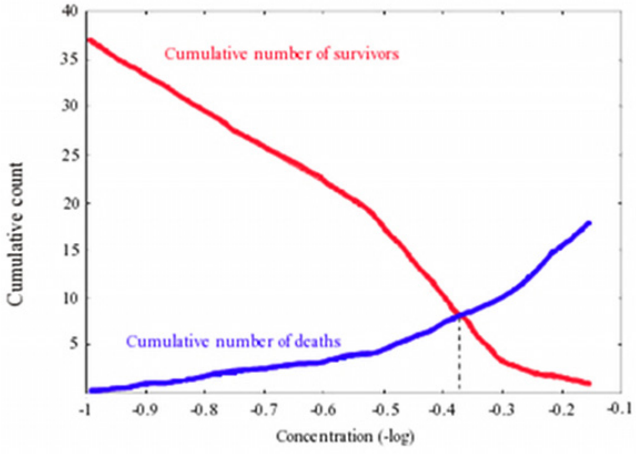

FIGURE 2. LC_{50} estimation following the Reed - Muench method. The two curves intersect at the point where 50% of the population has died.

Pilot results

The links between toxicity and aposematism can be presented in the form of a table (Table 1). If aposematism is a true reflection of the chemical defenses possessed by a given organism, some kind of arithmetic correlation (r) should be expected.

Species	Mean COI	Toxicity (LC_{50}) $^{mg}/_{ml}$
Echinometra vanbrunti	0.61	0.0158
Rhizophora mangle	0.534	0.05
Heliaster kubinjii	0.948	0.7244
Eurithoe cf. complanata	1.71	1.548

TABLE 1. Color-overlapping indices and toxicity values (LD50) for five Seri marine medicines[2].

FINAL REMARKS

Decades of ethnopharmacological research have pushed forward the idea that smelling-tasting plants and animals are preponderant mechanisms for medicine selection in primates. This article provides a different framework for the selection of ethnomedicines by promoting a broader sensorial input that relies heavily on visual cues. Primates are the most visually oriented mammalian order (Heesy 2009) and their color discrimination ability exceeds that of other mammals (Melin et al. 2013). Such an adaptation should be advantageous beyond food selection and object manipulation.

I think that the visual evaluation of the living world provides humans and other primates with immediate information about the chemical properties of their surrounding biota. I have suggested aposematism as a plausible ecological mechanism for the visual detection of chemical properties. Example data presented in this paper supports the idea that there is a consonance between brightly colored organisms and their toxicity. These organisms have long been used as ethnomedicinal ingredients in Seri pharmacopoeia, and while the aposematic rationale is particularly clear for marine ethnomedicines, it should also be applicable to conspicuous botanicals and fungi.

One of the main postulates of this article is that human beings are constrained in very similar ways in their conceptual recognition of potentially medicinal plants through a sensory evaluation of their surroundings. The sensory evaluation of medicinal plants should be understood as a complex biocultural process affected by culture and individual experience (Shepard 2004). However, these perceptual givens are firmly rooted in a biological reality that, at least from the results of pilot data, is perceptually unambiguous. Researching the links between medicinal organisms and their conspicuousness can offer deep insight into our species' environmental adaptation capabilities. The only thing we need to do is to generate enough data to unfold the links between toxicity, conspicuousness, and human cognition.

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I thank Amalia, Aurora, and Maria Luisa Astorga, Horacio Liñeiro, Manuel Monroy, Laura Monti, and Gary Nabhan for their help while in the Seriland. I am deeply in debt with Arli De Luca and Alberto Burquez who have long heard and helped polish the idea that aposematism can effect human cognition. Thanks to Diogo Samia for providing me with the R code for calculating COI and to Dr. Marco Augusto Miranda-Ackerman for assisting me through the use of R. The final manuscript received helpful comments from Lisa L. Price, precise edits from Cathy Moser Marlett and kind and thoughtful comments from anonymous reviewers. I thank the support of El Colegio de Michoacán and Jesus Medina, Research Assistants who greatly helped with the minute details of the final manuscript.

NOTES

[1] Chemical ecology tries to understand species dispersal and distribution by means of chemical interactions between species and between these species and their environment (Bakus et al. 1986). In the caves of Punta Banda, Baja California, sponges, anemones, tunicates, echinoderms, and crustaceans use secondary metabolites to compete for space by digesting, fouling, or poisoning their competitors.

[2] Pilot data was obtained using my photographic archive. The sample is limited to four different organisms that adhere to the following selection criteria: a) organisms should be photographed in their natural surroundings and b) the camera set at ISO 800. Aperture priority was not considered part of the criteria as it is inconsistent throughout the archive. Therefore, the data should be considered as preliminary. To calculate an average COI, I captured the organism's image inside an equilateral triangle. Three different areas of the background were selected per picture and correspond to the vertices in the triangle. The color information of each area was fed independently to the COI function along with a single area comprising the animal's body.

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