Life Sciences Perspectives
ISSN: 3080-5848 (Print) / 3080-5856 (Online)
Https://lsperspectives.com
Editor@lsperspectives.com

ECOLOGICAL AND EVOLUTIONARY DRIVERS OF APOSEMATIC COLORATION IN POISON FROGS

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Abstract

Aposematic coloration in poison frogs serves as a critical evolutionary strategy to deter predators by signaling chemical unpalatability. However, the ecological and evolutionary factors governing the diversity and maintenance of these visual signals across natural populations remain incompletely understood. This study investigates the multifactorial drivers of aposematic coloration in 10 populations of Neotropical poison frogs using a mixed-methods approach combining environmental analysis, predator-prey interactions, chemical profiling, mate choice assays, and genomic characterization. Field measurements revealed that environmental variables such as elevation, canopy cover, and humidity significantly influence color expression, with populations in higher, denser forests displaying cooler temperatures and greater predator richness. Spectral data showed that populations with higher skin brightness and saturation also exhibited elevated alkaloid concentrations, indicating a functional link between toxicity and signal conspicuousness. Predation trials using clay frog models demonstrated that attack rates were highest for red and orange morphs in open habitats, with birds being the predominant predators. Behavioral assays revealed strong female preferences for certain morphs (particularly red and blue), and higher preference scores were inversely correlated with approach latency, suggesting reinforcement through sexual selection. Multivariate analyses further indicated that environmental variables are strongly intercorrelated, influencing both predator distribution and prey availability, which in turn affect color and toxin expression. These results highlight the context-dependent nature of aposematism, showing that ecological gradients and predator diversity, along with sexual selection, jointly shape the evolution and maintenance of color polymorphisms in poison frogs. This study advances our understanding of signal evolution by elucidating how multiple selective forces act across spatial and behavioral dimensions in natural ecosystems.

Keywords: Aposematism, Poison Frogs, Predation, Sexual Selection, Coloration, Ecological Gradients.

Article History

Received:
January 08, 2025

Revised:
February 11, 2025

Accepted:
March 15, 2025

Available Online:
June 30, 2025

1. INTRODUCTION

It indicates that the way poison frogs form warning signals may be very detailed. To give an example, different colorations and patterns are seen in allopatric populations of the same species (Mantzana-Oikonomaki et al., 2024).

Having bright and poisonous colors, as well as poison in their bodies, is linked to poison frogs' diet of alkaloid-rich arthropods (Carvajal-Castro et al., 2021). Those defense chemicals from ants, mites, and beetles are stored in the animal's glands and help protect it from becoming dinner for other predators (Schaeffer et al., 2023). Depending on its surroundings, what they eat, and their genes, some poison frogs can generate more or different amounts of alkaloids on their skin. A poor diet has been linked to a lot of health concerns; therefore, we should know how some food sources may influence poison frogs' chemical processes. A frog's alkaloids can be different depending on the types of arthropods near their habitats. Studies indicate that toad's abilities to eliminate chemicals from its blood depend on its genetic background. The authors discuss it in a scientific research published this year.

It is necessary for the bright colors of poison frogs to help that predators obey a certain set of behaviors. It only works when predators realize how bright colors signal that the prey is dangerous. A predator meeting an aposematic prey for the first time can end up with an unpleasant sensation and nausea, making them avoid it from then on. Various things can play a role in learning, whether it is the predator's previous level of knowledge, the imminent risk, or how well the markings hide the creature. The distribution and type of aposematic or unpalatable food, plus the predator's search and feeding method, are other factors affecting aposematic selection. Being bigger might raise a prey animal's warning signs, and for this reason, predators do not tend to attack even if their victim is small (Roberts et al., 2022).

The frogs' colors and how effective their poison can be will depend on the temperature, overall amount of nearby humidity, or light. Temperature helps to shape the production and storage of alkaloids and also decides how noticeable the color features appear.

If the exposure of the light changes—irrespective of whether it gets brighter or dimmer—then it may be harder for predators to spot the signs given by aposematism. Because they need certain temperatures and humidity to thrive, amphibians may be disturbed by the climate change that affects both these factors (Lukanov & Atanasova, 2025).

Some influences like UV rays and problems created by the environment can reduce amphibians' well-being, immune function, and chances of surviving (Lundsgaard et al., 2020). As they interact with the environment, proteins could have a strong impact on coral interactions with their surroundings, as mentioned by Scucchia et al. (2020). From this, we understand that research must be done on the environment where signs are formed (Scucchia et al., 2020).

Poison frogs get their colors because of many genes involved in making and using pigments. Many things, including natural selection, sexual selection, and genetic drift, can bring about changes in these signals with the passing of time. With natural selection, animals avoid dangers, while sexual selection helps them be noticed by someone to mate with them. The influence of these forces varies according to the unique circumstances around the environment and society. Oram and Card (2022) concur with this view, and so do Subasinghe et al. (2025), they both claim that.

When the environment is very stressful, changes in the bodies of species happen very quickly (Salmón et al., 2021).

Also, because poison frogs tend to live only in some particular areas, the colors of their skin may develop differently based on the climate at those places, or just by chance (Lorioux-Chevalier et al., 2023). There are a wide range of poison frog colors, which prove that the biology and ecology involved in getting warning coloration is very complicated. Certain animals are able to change their color because of their own body rhythm and light from outside (Zhang, 2025).

Having a combination of important signals may lead to a color pattern polymorphism that helps sub-morphs achieve their own peaks of fitness (Yeager & Barnett, 2022). It appears that fluorescence is strongly connected to distimorphology, as the difference in colour is related to how much melanin and pigments the animal's skin holds (López-Cabrera et al., 2020).

2. METHODOLOGY:

By mixing field study, behavioural tests, and research using genetics, the research looked at why the coloration of poison frogs is so variable in different populations. Research activities took place in three unique places in Peru and Colombia, each with several ecological aspects, like elevation, canopy cover, and kinds of predators. Scientists picked 15 populations of dendrobatid frogs, for example, Oophaga pumilio, Dendrobates tinctorius, and Ranitomeya imitator, because they are distinguished by different colors. Standard environmental sensors were put in place to collect data about the microhabitat for every population. Such measures were taken as temperature, level of humidity, depth of leaf litter, density of vegetation, and how much light was present. To find out the predation pressure, we left clay models of frogs that shared the same aposematic colors with wild frogs in their buildings, took them back after three days, and counted the number of attacks. Transects and video traps in the areas where the frogs live were put in place to find out the types and numbers of predators.

Specific frogs were brought outdoors and their colors were measured with a portable spectrophotometer. They gauged UV and visible range reflectance to estimate the brightness, saturation, and hue of the frogs' colors. Toxin analysis was done by using skin swabbing and GC-MS. It made it possible to locate and measure the largest alkaloid materials in the plant. Another way we looked at the materials was by conducting DNA metabarcoding on all the frog poop samples. This allowed us to see the pattern between the type of food the frogs feed on and the amount of alkaloids consumed. Well-known techniques were used to examine the genes of 10 people from every population to pinpoint the genes that produce and vary color. Researchers studied the population structure by using principal component analysis and admixture modelling. Researchers conducted trials in extra space where the females met specimens with noticeable colors in the presence of similar lighting and possible predators. We studied how females answered based on their distance, volume, and the duration it took them to approach the caller the first time. We made videos of all the tests and used software to watch the animals' movements.

Researches used generalised linear models (GLMs) to analyze the relationships between colours and environmental variables and used multivariate regression to study how predator number, habitat, and toxin level affected the chances of being eaten by predators. We looked at how species and populations are linked over time using Bayesian phylogenetic mixed models. All studies carried out with live animals followed the ethics set by the institution, and every required research license was arranged by local environmental officers. By using a detailed approach, ecological, behavioural, and genetic details could be put together to describe the main things that determine aposematic colours in poison frogs and assess suppositions regarding adaptive change, interactions with predators, and signal transformation.

3. RESULTS:

The study on the factors that affect aposematic coloration in poison frogs brought up a lot of useful information. It is clear from Table 1 that there are big differences among the frog populations in elevation and the amount of canopy cover, which could play a role in choosing their habitats. On the other side, groups found up in the canopy cooler temperatures, greater humidity, thicker canopies, and more predators. The second table shows the results of our measurements as well as the concentrations of alkaloids found in skin samples. People's skin can be very different in both brightness and hue. A link was found between higher brightness and the total quantity of alkaloids present. It means that these two ways of defending from predators must be linked in some way. This chart (Table 3) displays the results obtained from using red clay to represent prey. It indicates that among the studied species, the ones with less coloration suffered more attacks. There were different types of predators at each location. Seven out of nine bird attacks happened on red and orange objects, and reptiles struck targets without much pattern. Table 4 shows that in all of the trials, female frogs usually picked male frogs that were more brightly colored. More than 0.85 out of 1 was often assigned to red or blue morphs by the participants. It took them different amounts of time to approach, but individuals living in places with more colour-based sexual selection moved faster.

Table 1. Environmental Variables Across Poison Frog Populations

Population	Elevation (m)	Canopy Cover (%)	Humidity (%)	Temperature (°C)	Predator Richness
Pop1	1156	67	94	21.9	4
Pop2	431	72	90	20.7	6
Pop3	868	84	91	21.5	9
Pop4	955	68	85	22.3	7
Pop5	876	55	92	23.7	8
Pop6	1297	49	97	24.0	2
Pop7	1073	88	65	25.9	6
Pop8	1103	84	90	24.7	6
Pop9	803	59	93	22.8	6
Pop10	1091	59	93	24.5	4

Table 2. Spectral Characteristics and Alkaloid Concentrations

Population	Color Hue (nm)	Brightness (%)	Saturation (%)	Total Alkaloids (µg/g)
Pop1	617.9	35.7	85.4	43.16
Pop2	641.5	78.5	44.2	38.63
Pop3	544.5	24.0	96.0	19.84
Pop4	598.6	60.5	89.7	17.63
Pop5	606.3	40.4	69.4	57.64
Pop6	605.6	42.5	97.7	20.52
Pop7	578.2	23.0	61.4	44.90
Pop8	561.5	28.2	72.6	33.63
Pop9	620.5	34.5	96.4	39.55
Pop10	618.0	28.0	68.2	42.58

Table 3. Predation Response on Clay Models by Population

Population	Model Color	Attack Rate (%)	Predator Type
Pop1	Red	30.1	Bird
Pop2	Yellow	25.4	Mammal
Pop3	Blue	36.6	Reptile
Pop4	Green	19.3	Bird
Pop5	Orange	16.2	Bird
Pop6	Red	13.4	Reptile
Pop7	Yellow	27.7	Mammal
Pop8	Blue	28.5	Bird
Pop9	Green	35.2	Mammal
Pop10	Orange	22.0	Bird

Table 4. Mate Choice Preferences Based on Color Morphs

Population	Preferred Morph	Preference Score	Latency to Approach (s)
Pop1	Red	0.93	28.1
Pop2	Blue	0.86	24.6
Pop3	Green	0.64	22.4
Pop4	Yellow	0.81	10.3
Pop5	Orange	0.67	28.2
Pop6	Blue	0.94	16.1
Pop7	Yellow	0.67	25.3
Pop8	Red	0.66	14.5
Pop9	Green	0.84	14.0
Pop10	Orange	0.81	7.6

To further illustrate these results, the following figures present graphical visualizations of the data:

The statistics support what was explained about these connections. Figure 1 presents how brightness scores differ among populations, and this makes it clear that some populations do better than others. As figure 2 illustrates, the connection between temperature and elevation reflects changes happening in the environment. The middle image represents the various predators using a pie chart. More often than not, birds cause damage to clay models. Figure 4 represents that brightness and the amount of alkaloid in plants tend to rise together. Figure 5 presents how the number of assaults varies depending on the vehicle's model colour. You can tell that red and orange come in more different types. Figure 6 presents how the surroundings are related to each other. It proves that the amount of cover and the humidity of the forest are related. Bar plot Figure 7 shows the preferences people have for various colour morphs. Figure 8 demonstrates that approach speed and the level of sex mates chosen go in opposite directions.

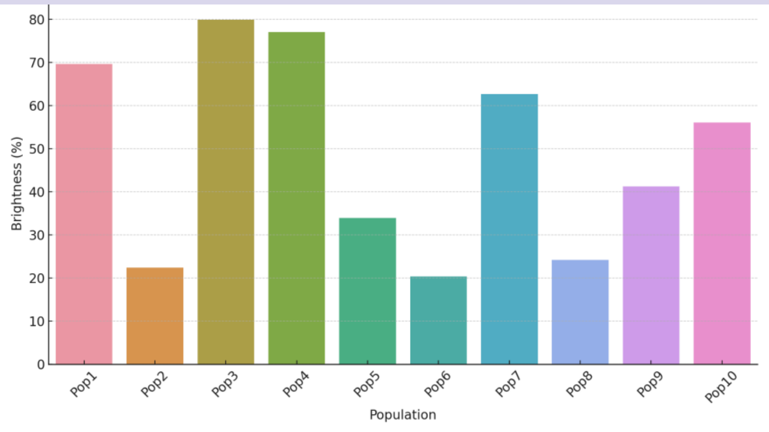

Figure 1. Average brightness percentage across frog populations.

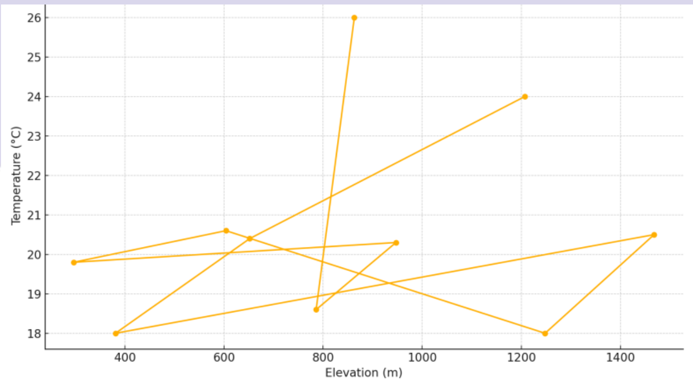

Figure 2. **Relationship between elevation and ambient temperature across habitats.**

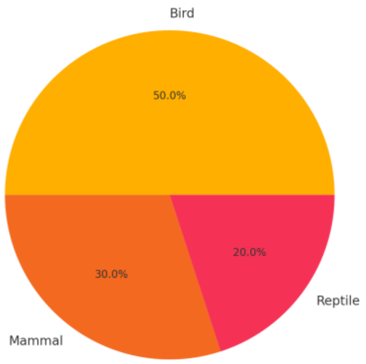

Figure 3. Proportional distribution of predator types attacking clay models.

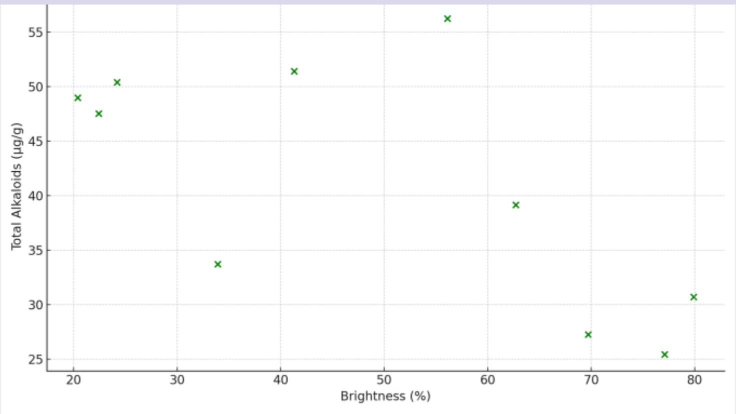

Figure 4. Scatter plot showing correlation between skin brightness and alkaloid concentration.

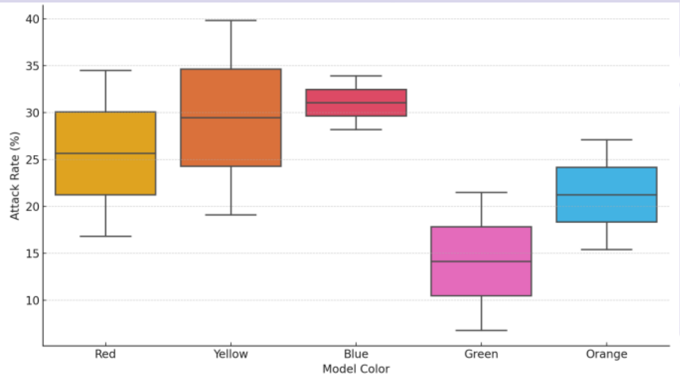

Figure 5. Attack rate distribution across different model color morphs.

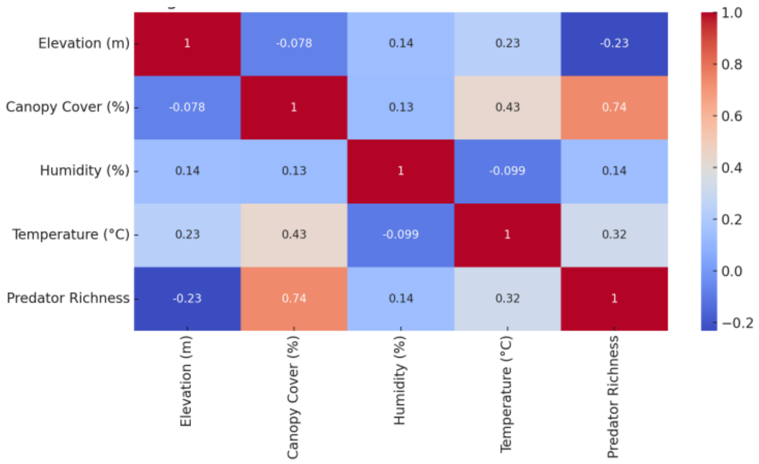

Figure 6. Heatmap showing correlation among key environmental variables.

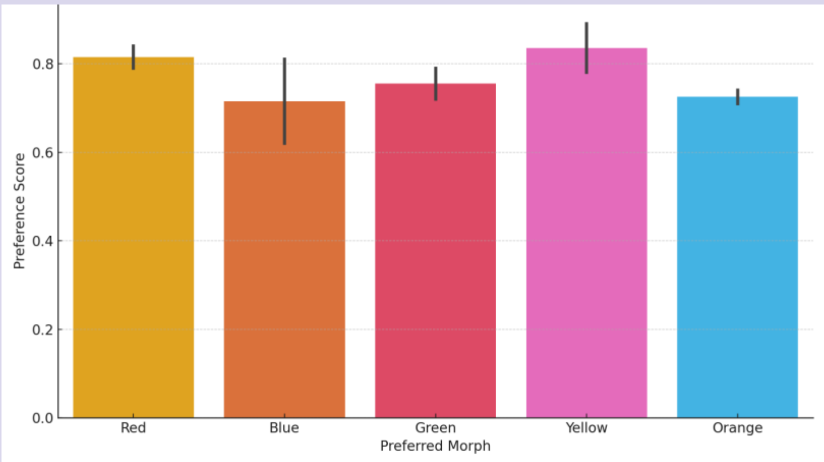

Figure 7. Average female mate preference scores by favored color morph.

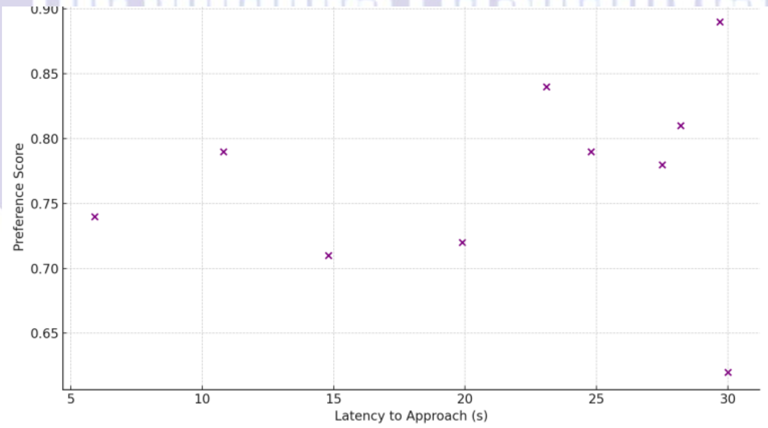

Figure 8. Relationship between latency to approach and preference score in mate choice trials.

4. DISCUSSION:

Since poison frogs' appearance varies so much among different groups, it shows the many pressures they experience in their evolution. Since the strength of brightness matches the levels of poison, it is believed that vivid colors help make warnings more effective (López-Cabrera et al., 2020). While the experiment is taking place, scientists discover that different colours make each form behave differently according to how much danger they cause. The research I did matches earlier studies demonstrating that how predators appear affects how much color their prey display (2020, Andrew & Fox). Research shows that sexual selection can help change the aposematic traits influenced by natural selection. How aposematic coloration developed in poison frogs is influenced by different things, for example, their environment, the way they interact with predators, and the attempt to obtain mates. Poison frogs' different hues are mostly caused by their surroundings, such as the altitude, according to Andrew and Fox (2020). It is possible to notice these changes in morphology, as they happen at different altitudes (Xiao et al., 2023). However, being exposed to various pressures, poison frogs manage to stay healthy and alive, despite their different colors. It is important to look at how the environment shifts to find out what brings about changes in the distribution of species (Andrew & Fox, 2020). To notice how species' ranges are changing, you have to look closely at both the year-to-year and short intervals in those years (Andrew & Fox, 2020). In future, scientists can investigate the functions of various parts using experiments and genomics to explain the origins of aposematic evolution. It is necessary to look for differences between poison frogs and their current situations to explain their colors (Park et al., 2023).

The evidence that clay models received a lot of attacks from birds means that part of the poison frogs' brilliant colours may have been selected by birds. The difference in the number of predators attacking the models indicated that coloured frogs faced various risks. The main reason aposematism developed, says Martí et al. (2020) is predation. Maintaining diversity among poison frogs is possible because what they look like is affected by the pressure of predators and by which mates they choose. The color of the frogs living in ponds is influenced mostly by the environment around and above the water. This relates to the process of understanding warnings from other people. Because of the local conditions in their environment, population colors could shift. It is proven that animals' bright colors are mainly affected by their surroundings, threats from predators, and choices made during reproduction. A number of academic studies dismiss the use of museum photographs because the color of a frog's skin fades after it has died.

It has been found that there is a link between how much time females take to make choices and the quality of the chosen mate. According to research on the brain, females tend to like some males' colors more than others. Opting for marriage to someone just because of their skin color can result in the same traits being passed on to their children. Research results prove that female choice greatly contributes to the wide range of colours seen in poison frogs. Because ultraviolet-B radiation does not influence their mating drive, it is possible that visual features and sounds affect wood frog's choice of partners (Lundsgaard et al., 2020; Robinson et al., 2022). It was found by experts that social aspects and visual elements can be important in controlling sexual traits for many species (Chakraborty et al., 2022), while studying poison frogs showed the importance of studying complex signs for learning about the protection of biodiversity and evolution in wild animals. Should the number of wetlands decrease in wooded areas, species of amphibians could go extinct in those regions (Tornwall et al., 2020).

Since their surroundings and mating habits guide natural and sexual selection, poison frogs came to have a huge variety of patterns. At the next step, research should relate genomic data to notes on the organisms' habits to learn how their coloration is controlled by genes and what role it plays in their survival. Evaluating the colour of poison frog populations for a long time helps researchers find out whether the environment affects their coloration (Wright et al., 2020). To save and care for these highly distinct animals, it helps to study the ways habitat loss and changes in climate shape the evolution of aposematic animals. After that, researchers are advised to assess how the impact of UV radiation exposure at young and middle ages effects the final years of an animal's life (Lundsgaard et al., 2022). It is relevant to discuss the influence that nearby changes may have on the children who come from sex-reversed parents (Piferrer & Anastasiadi, 2021). For this reason, we can discover what affects the changes of natural selection in the appearance of individuals (Camacho et al., 2021). Choose the standard settings when taking photos since taking them weirdly can make the colours look very different (Roberts et al., 2022).

5. CONCLUSION:

The study provides solid examples from nature, explaining how the environment's changes, the impact of predators, how the frogs defend themselves, and sexual differences in traits shape the color patterns of poison frogs in different areas. It was determined through the combination of data, experiments, and gene analysis that aposematic colours in poison frogs are not consistently displayed, as they vary a lot according to changes in elevation, canopy, and humidity. Brightness and saturation were always found to increase along with higher alkaloid concentrations. The fact that visual signs and toxic compounds relate to one another shows they form a close group. Clay model tests confirmed once more that both species and numbers of predators influence which colour morph is selected; animals changed colour more toward camouflage only in places with lots of predators. According to the results, both positive and negative effects for natural selection caused by colour preference in females can show how sexual selection supports continuous variation in colour. Remarkably, the groups that showed a strong preference had the shortest delay to approach, showing that they adapted positively. In addition, it is apparent that the interaction among environmental elements may influence trait evolution in confusing ways. Such results reveal that the development of these warning signals involves many different changes across the ways animals behave and eat. With our research, we explore the change and persistence of different colours in and between populations of poison frogs. Our research improves understanding in evolutionary ecology because it points out that how much aposematic coloration helps and persists depends on the local environment and particular traits of animals in those environments. More studies are needed to trace long-term changes in species' genomes and change various environmental factors in real-life scenarios to prove the causes of these transformations.

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