

RESEARCH ARTICLE

Bifurcations and neutrality in the biosynthesis of plant flavonoids

Ivanov Vyacheslav^{1*}, Shcherbakov Arkadiy², Usmanov Iskander²

¹Department of Pedagogical Sciences, Nizhnevartovsk State University, Lenina St. 56, Nizhnevartovsk, Russia; *karatazh@mail.ru

²Department of Biological Sciences, Institute of Genetic and Tissue Engineering, Mendeleeva St. 9, Ufa, Russia

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Abstract

During field ecological expeditions, plant samples were collected from oligotrophic bogs in Western Siberia and the steppes of the Southern Trans-Urals. HPLC spectra of flavonoid biosynthesis of wild plants were analysed. It was shown for the first time that, irrespective of the plant species and habitats, biosynthesis of flavonoids has a fractal structure. High species specificity of fractal structures of flavonoid biosynthesis has been shown.

Analysis of the bifurcations of the metabolic pathways of flavonoid biosynthesis showed the possibility of biosynthesis of some substances by alternative pathways. It was shown for the first time that, irrespective of the plant species and habitats, biosynthesis of flavonoids has a fractal structure. The possibility of hit compound several of metabolic pathways at some common point (substance) has experimentally shown the effectiveness of neutral mechanisms.

Keywords: Western Siberia, bogs, southern trans-urals, steppes, flavonoids, biosynthesis of flavonoids, bifurcations, strange attractors, fractal object, multifractal

Introduction

It is known that when adapting to particular habitat conditions, the plant shows the principle of plurality of primary adaptive mechanisms: it triggers as many mechanisms as it can afford from the point of view of resource allocation. Most of the physiologically significant metabolites in plants are represented not by one, but by a certain list of closely related compounds (many hormones, pigments of photosynthesis, etc.) (Tilman 2004; Hubbell 2006; Lambers et al. 2008; Mc Gill 2010; Rosenberg 1984, 2013; Petrusa et al. 2013).

This list fully includes biologically active plant compounds. However, the general principles of ensuring such a polyvariety of physiological mechanisms, in relation to biologically active plant compounds, have been poorly understood until now.

Flavonoids are the most important elements of the non-specific antioxidant and metal-chelating systems of plants. They belong to specialized plant metabolites, and also have high biological activity (Mierziak et

al. 2014). At the same time, scientists have no clear understanding of how particular growing conditions led to the accumulation of this very spectrum of flavonoids in certain plant species, and not another one, as in the case with alkaloids. The studies having been conducted by us for more than 15 years also did not reveal stable correlations between the accumulation of flavonoids and the numerical values of environmental factors: temperature, moisture content, the content of soil elements, etc. in the studied species. It made us pay close attention to the structure, organization, and general logic of the functioning of the metabolic pathways of the biosynthesis of these compounds. In this regard, we have studied the structure, organization and general logic of the functioning of the metabolic pathways of biosynthesis of these compounds (Usmanov et al. 2015, 2016, 2017, 2019, 2020, 2021; Mavletova-Chistyakova et al. 2017).

Materials and Methods

Samples were collected in ecologically contrasting communities of Western Siberia and the steppes of

the South Trans-Urals. Each of the areas is ecologically homogeneous. The raised bogs of Western Siberia were represented by *Oxycocco-sphagnetea*. The team studied the dominant species of *Oxycoccus palustris* Pers., *Chamaedaphne calyculata* L., and *Andromeda polifolia* L. The true steppes of the South Trans-Urals were represented by the community of *Festuco-Brometea*, where the dominant species are *Juniperus sabina* L., *Glycyrrhiza korshinskyi* Grig., and *Achillea millefolium* L.

Spectra of flavonoids and physico-chemically similar compounds in the studied plants were determined by High-Performance Liquid Chromatography (HPLC). Comparative analysis of HPLC spectra was carried out by the method of principal components.

Results

In a comparative analysis of various literature sources, it was found that the metabolic pathways of the main classes of flavonoids, in general, represent a kind of tree with switching points (bifurcations). Analysis of the metabolic pathways showed that the total number of alternative biosynthetic variants of the same non-glycosylated flavonoid can reach 5 options. In addition, it should not be forgotten that most flavonoids are present in plants in the form of various kinds of glycosides. At the same time, the transformations associated with the addition and disconnection of the carbohydrate component represents an additional metabolic system, for the operation of which enzymes are not even always necessary. Thus, plants have a very flexible system of flavonoid biosynthesis. Theoretically, the possible number of synthesized compounds in the operation of this system, taking into account all possible options of transformations, reaches many millions (Fig. 1) (Harborne 2013; Usmanov et al. 2019-2021; Scherbakov et al. 2021).

At the same time, the solution of the same problem (for example, antioxidant protection) in the operation of

this system can be solved by the simultaneous inclusion of many alternative methods.

During the research, questions arose, what is the logic of such a multi-component system, are the spectra of flavonoids that synthesize plants in certain habitats random or non-random?

According to modern concepts, multicomponent systems can theoretically have three options of structural organization with a wide range of transition states (Gelashvili et al. 2013):

- An object of regular nature. The spatial structure and general logic can be described by relatively simple equations, comparable in complexity to the equations of Euclidean geometry. Knowing the properties of one of the parts of this system a priori allows predicting with high accuracy the properties of the other parts and the entire system, in general. If an object has self-similarity in a wide range of levels of the organization, it is usually referred to as a regular fractal.
- An object of multifractal nature is an inhomogeneous fractal object, for the complete description of which, unlike ordinary fractals, a whole range of equations is required, the number of which can go to infinity. However, in general, the property of self-similarity in a wide range of levels of the organization is preserved for such an object.
- An object of chaotic nature. Self-similarity is expressed slightly, the properties of the object are described mainly by the equations of statistics and probability theory.

Iudin and Gelashvili proposed an algorithm for testing the properties of multicomponent systems in ecology for compliance with the principles of fractal formalism (Gelashvili et al. 2013). It was found that this algorithm can be successfully applied to solve problems of ecological plant physiology. The use of this technique allowed us to check for compliance with the fractal logic

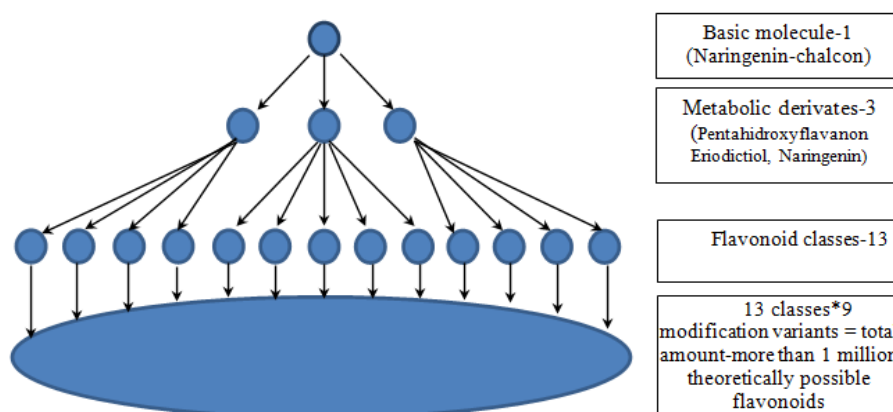


Figure 1. Basic steps of principles of flavonoid diversity forming.

of large data sets containing individual characteristics of plants by morphological, biochemical, and physiological parameters.

In our research, it is shown for the first time that, regardless of the species and habitat, plant metabolism growth biochemical and physiological indicators (including the functioning of the system of flavonoid biosynthesis pathways) in most cases has the properties of a fractal object (Usmanov et al. 2018-2021; Ivanov et al. 2016, 2019).

Such results could be analysed from the standpoint of the neutrality theories. In the molecular neutrality theory model by Kimura (1983), neutrality is a random set of the existing genes, the choice of which has a weak correlation with the external conditions. The neutrality theory assumes the existence of a “general population” that can produce various combinations of “responses”. Similar reasoning, later proven by facts, was the basis of the plant community models. In that case, neutrality is the random introduction of this or that species from the total flora list of the region to the community. The applicability of the neutrality theory was not tested on the level of metabolic pathway analysis (Gaston et al. 2005).

In the process of multi-years research, it was found that the deterioration of habitat conditions leads to a decrease in the options available for plants to include alternative protective mechanisms. As a result, the similarity between plants in the implementation of adaptation mechanisms increases, and the entire array of individual plant data under such conditions begins to show a tendency to turn into a regular fractal (a sharp increase in the correlation between individual plant indicators while maintaining self-similarity).

The opposite is also true: in the case of insufficient stressful external conditions, plants can include many alternative ways of survival (including through the synthesis of alternative lists of metabolites). At the same time, the diversity in the individual reactions of plants increases, but the property of self-similarity remains. Thus, the array of individual plant data begins to show a trend of turning into a multifractal.

Our research has shown that fractal analysis can be successfully applied to assess the level of stress load on plants in any habitat. This method is universal due to the following features:

- As a basis for calculations, any individual plant parameters can be used: growth, morphological, biochemical, and physiological
- Any species that are convenient for research can be used as marker species

Thus, it is established that the possibility of passing several metabolic pathways through a certain common point (substance) is the key to the high efficiency of neutral mechanisms in the formation of a pool of flavonoids by a plant in various living conditions.

The selected species complexes should be considered as regional products that have specific biological activity in response to the habitation of plants in particular environmental conditions.

Conclusion

For all studied species, it was shown that the aggregates of synthesized flavonoids (chromatograms) possess the properties of a “strange attractor”. This will make it possible to study the formation and transformation of the boundaries of phase spaces, as well as to build models for the movement of attracting centres and strange attractors in general under changing environmental conditions.

The complex of studies carried out contains fundamentally new information about the biological organization of the biosynthesis of flavonoids. It is expedient to consider the selected species complexes as regional products with specific biological activity.

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