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The Risk Assessment of Threats from Biological Objects in Environmental Safety

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Summary. Necessity to present results of scientific research in an accessible form for proper interpretation by public and environmental safety experts has been shown. Myxomycetes ecology and metabolism to verify their safety for human health has been evaluated as an example. Bayesian methods for probability analysis is proposed to be used for risk assessment. The "Environmental safety passports of species" are recommended for evaluation of impact of each species on the environment, humans and other living organisms.

Key words: environmental safety, risk assessment, myxomycetes, ecological passport.

INTRODUCTION

Improvement of methods for evaluation and display of environmental risk is necessary for operational changes analysis in the environment, to show the dynamics and predict the possible consequences on the health of the population. Living organisms play a key role in the transformation in nature, they are able to enhance and reduce the impact of negative factors on the environment, human and other natural objects [6]. Also living organisms can be of danger for environment or people health so it is very important to make correct evaluation of threat from them. The problem is that biological scientific information not always has correct interpretation in ecological safety study. For example, the article about dangerous properties of slime mold for human health [13] was surprising for all scientists who study myxomycetes. Slime molds (myxomycetes) are fungus-like protozoa, which is mainly associated with remains of woody microhabitats [4], but was never found in human body. That article has not presented verification of reliable proof base about pathogenic internals of myxomycetes, so scientists are unlikely to perceive such information seriously. But because these organisms are poorly-known by public, people were very amazed by the idea about a new causer of "all illness" and wrong information is quickly spread through mass-media and Internet. As a result some patients take treatment against "very dangerous" myxomycetes, insist to found the real cause of their illness and have a proper treatment. This example shows the necessity to present results of scientific research in an accessible form for public display in order to avoid ambiguity. The proper interpretation of investigations is especially important in environmental safety, because it may affect the accuracy of decision-making measures to ensure the safety of the population and environment.

PURPOSE OF WORK

The objective of this study is found the way of evaluation of threats from biological objects to environment, human and other living organisms. According with this purpose, myxomycetes are used as a model organism to find the way of risk assessment of environmental safety threats. For assessment of their danger for human health Bayesian methods are used for probability analysis that studies their distribution, ecology and metabolism.

MATERIAL AND METHODS

Material for this study is result of myxomycetes research for more than 20 years in their native habitats in Ukraine: 1643 samples from field collections, herbarium of M.G. Kholodny Institute of Botany NAS of Ukraine and literature sources about distribution of myxomycetes in Ukraine (278 species total) [4]. There are near a thousand species of myxomycetes total known in over world.

Bayesian rule is used for analysis probability to find species of myxomycetes on reputed substratum used:

$$P(M \mid E) = \frac{P(E \mid M)}{\sum_m P(E \mid M_m) P(M_m)} * P(M), \quad (1)$$

where:
$|$ – a conditional given probability,
E – the evidence corresponds to new data that were not used in computing the prior probability,
M – event represented model,
$P\ (E \mid M)$ – the conditional probabilities are specified to define the models,
$P\ (M_m)$ – the degree of belief in M_m.

For discover possibility to find myxomycetes on before unknown substratum use additive Laplace smoothing is a technique used to smooth categorical data. Given an observation x = (x_1, …, x_d) from a multinomial distribution with N trials and parameter vector $\theta = (\theta_1, \ldots, \theta_d)$, a "smoothed" version of the data gives the estimator:

$$\hat{\theta}_i = \frac{x_i + \alpha}{N + \alpha d} (i = 1, \ldots, d), \quad (2)$$

where:
$\alpha > 0$ is the smoothing parameter ($\alpha = 0$ corresponds to no smoothing). Additive smoothing is a type of shrinkage estimator, as the resulting estimate will be between the empirical estimate x_i / N, and the uniform probability $1/d$ [18].

The control action generally is represented in the form of a functional:

$$R = F\left(x_i, \nu_i, \frac{dx_i}{dt}, .., \frac{d\nu_i}{dt}, .., \int x_i dt, .. \int \nu_i dt\right), \quad (3)$$

where:
x – a controlled variable,
ν – parameter of external influences,
dx/dt and dv/dt – derivatives on a time from parameters of regulating and external influences, accordingly, that is velocities of a modification of parameters,
integrals on a time from parameters of regulating and external influences, that is the accumulated modifications of parameters [14].

RESULT OF RESEARCH

Myxomycetes (slime molds) in different stages of their life cycle are presented as spores, myxamebaes, flagellate cells, plasmodium and fruiting bodies. Myxomycetes tend to be rather inconspicuous in their occurrence. This stage also includes myxamebaes and flagellate cells. From plasmodium after a period of feeding and growth then fruiting bodies develop in a drier and more exposed location. Fruiting bodies of most species are relatively ephemeral and do not persist in nature for very long. These fruiting bodies containing numerous spores which can be dispersed by wind and will eventually germinate and develop into a plasmodium under suitable conditions. Myxomycetes spend a portion of their life cycle in a state where le their very presence in a given habitat can be exceedingly difficult if not impossible to determine.

Analyses of substratum where fruiting bodies of myxomycetes in Ukraine were find show the following distribution: 42% - on destroying dead wood (xylophilic), 20% - on forest floor litter (litterophilic or deciduophilic), 20% - on bark of living trees (corticolous), 7% - on mosses (bryophilic) and 2 % – on living herbal plants (herbophilic). Sometimes myxomycetes fruiting bodies (until 1% of total biota) possible to found on dung of herbivorous animals (coprophilic), on fungi fructifications (mycophilic), on lichen (lichenophilic), and even on soil or stone (geophilic). Also 8% of myxomycetes species belong to nivicolous ecological group, occurring at the edge of melting snow in mountain high altitude in the Carpathians in early spring [5]. This ecological group divides by phenology and physiology, but substratum preference show nivicolous myxomycetes as deciduophilic and herbophilic. The diagram of myxomycetes fruiting bodies distribution by type of substratum was made according with these data (fig. 1). Nivicolous myxomycetes were divided equally between deciduophilic and herbophilic, then substratum groups of herbophilic and bryophilic were integrated.

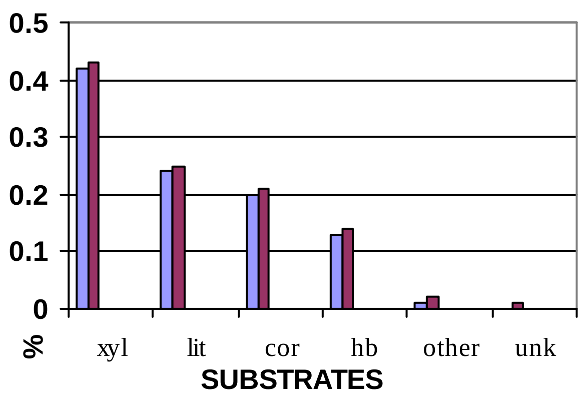

Fig. 1. The distribution of myxomycetes fructifications by type of substratum: xyl – xylophilic, lit - litterophilic (deciduophilic and half of nivicolous), cor – corticolous, hb - herbophilic (includ bryophilic and half of nivicolous), other - coprophilic, mycophilic, lichenophilic, geophilic et al.; unk – unknown substratum up to now; ▫ – real data; ▫ – data with additive Laplace smoothing

As we can see, myxomycetes sporocarps have never been found on or inside human body. Additive Laplace smoothing (2) gives 1% probabilities to find myxomycetes on before unknown substratum. Also the same frequency added to the each substratum types where myxomycetes sporocarps were really found. Calculation according to Bayesian analysis (1) gives around 10^{-5} probability for all unbelievable substrates for myxomycetes. This probability includes volcano, vacuum, human body and other millions of habitats, where fruiting bodies of myxomycetes were never found before. Each of hypothetical microhabitats reduces such events probability by exponentially. So chance to find myxomycetes sporocarps on or inside human organism is negligibly small. Even this tiny chance can't be used, because for creating fruiting bodies myxomycetes need more or less dry substratum and sun shine, but inside human body there are no suitable conditions for this. Any organism can't become parasitic without effective reproducing strategy inside of host body, so myxomycetes can't be dangerous for people. For hundreds of years of myxomycetes study only a few very exotic cases were registered of finding myxomycetes sporocarps on living animals: *Physarum pusillum* on lizard, *Arcyria stipata* on snail shell and *Diderma effusum* on aquarium fish. The *Corytophanes cristatus* belongs to the lizard who most part of their life don't move waiting for prey and as a result on the skin of this animal not only myxomycetes sporangia is found, but also moss and other vegetation [21]. Sporangium of *Arcyria stipata* on shell can't make damage to snail, because it even doesn't touch the body of snail. Phaneroplasmodia of *Diderma effusum* were observed feeding on green algae and diatoms submerged in water in an aquarium as result of a laboratory experiment. This species submerged in water and growing on agar culture. Portions of the plasmodium were also observed adhering to the skin surface of an eel-like fish in the aquarium [19]. All these exceptional cases only prove the rule that myxomycetes don't have a habit to create fruiting body on living animals and it is absolutely impossible for them to do it inside human body.

Sporocarp stage is short and ephemeral for slime molds. Most part of their life cycle myxomycetes exist as vegetative stage presented by mobile free-living plasmodium and as spores. Myxomycetes are fundamentally terrestrial organisms and they have a significant impact on the species diversity of soil microorganisms. Plasmodium typically lives in cool and shady moist places and feed on bacteria, protozoa, yeast cells, fungi, organic remains, and etc. It is very inconspicuous in their occurrence inside destroyed wood and in soil and it is rather difficult to find plasmodium in nature. Plasmodium can't crawl into human body itself being unnoticed, only if people eat them especialy. In Mexico people prepare exotic food from big plasmodium *Fuligo septica* [16]. In 4 Amazonian communities of Ecuador mythical, food, medicinal and recreational use of 3 myxomycetes species recorded: *Arcyria denudata, A. cinerea* and *Lycogala epidendrum.* They are used *Lycogala epidendrum* medicinally by rubbing the myxomycetes aethalia in skin infections and also they call this species "fungus for eating" and is eaten raw [3]. The plasmodium can't live and give fructification in human body, because it is quickly destroyed by enzymes. On myxomycetes feed insects, fungi, slime and invertebrates. Some spores go through beetles guts and it is example of mutual advantage, because it help to myxomycetes distributions. But slime molds spores never grow up in guts of living animals [10]. A few myxomycetes spores can be got by breathing with air, but human immune system has very strong protection against bioactive metabolites of myxomycetes. It was confirmed by allergy prick testing, which shows sensitization to myxomycetes [7]. But myxomycetes can't become strong allergen, because concentration of their spore in air is so low, that it is very difficult to inbreathe enough number of spores to cause allergy reaction. The best focus group for study allergy from myxomycetes is myxomycetologists, people who permanently work with these organisms in the field and in laboratories. Preliminary study of their allergy reaction on myxomycetes spores found is only one real case of sensitization. It confirms that myxomycetes have no chance to become real dangerous allergen.

Almost 100 natural compounds were described from myxomycetes including their chemical structures and biological activities: lipids, fatty acid amides and derivatives, alkaloids, amino acids and peptides, naphthoquinone pigments, aromatic compounds, carbohydrate compounds, terpenoid compounds [2]. Myxomycetes plasmodia and sporocarps have biological activity against micropathogens *Candida* [8]. Red colored sporophores of *A. denudata* contain major pigments alkaloids arcyriarubins and arcyriaflavins, which are new lead structures for the synthesis of biologically active substances and show highly potent biological effects. Arcyriaflavin derivatives have antimicrobial activity against *Bacillus cereus* and antitumor activity against leukemia cells [2]. Arcyriaflavin analogues are currently being evaluated in human clinical trials as anticancer drugs [15].

All these dates show that myxomycetes can be classified to the group of the living organisms with lowest danger probability for human health.

DISCUSSION

This research is to show that myxomycetes are not dangerous for humans. Also on this example we can see that even such innocuous organisms can be under distrust. Such situation is not unique only for myxomycetes and can happen with any living organisms. Wrong evaluation of dangerous level can have unpredictable and far-reaching effects on safety of human, other living organisms and on environment in general. It shows necessity of scientific information to be presented in the way which can be understandable for all stakeholders of groups to avoid improper interpretations. For this reason we propose to implement a system of safety certification of all living organisms. The myxomycetes were proposed to be used as model objects to develop patterns of environmental safety passports of species. The reason of such a choice is justified by morphology, physiology, biochemistry and phylogeny of myxomycetes and by their wide distribution in different habitats of different regions of the world.

These organisms combine inherent features of fungi and protozoa, but now there is general agreement that their true position is among the protozoa. However they are covered by the same nomenclatural code, and are listed in mycology's main reference work. So it is give possibility to work out of certification structure for both kingdoms.

It is possible to see most myxomycetes species by naked eye, but special equipment is needed for studying details of their morphology and for discovering species of the size less than 1 mm. So they have intermediate position between macroscopic and microscopic organisms. This factor also supports choosing myxomycetes as a model organism for certification, because it helps to find the universal common model for certification.

Myxomycetes are easy to be cultivated in a laboratory on agar and other substrates. This gives possibility to study their reactions on different influences. Their full life cycle takes rather short time and transformation from plasmodium to mature sporocarps can last for just a few days. These features make myxomycetes excellent model objects, which are very often used for a wide range of scientific researches.

Myxomycetes occur in all spheres of nature on different stage of their life cycle: soil, air and less in water. Study DNA from soil shows that myxomycetes (including protostelids and dictyostelids) are dominating group of protists in this habitat, where they are presented as spores, plasmodium and myxamoebas. DNA analyses show also presence of myxomycetes in water, probaply in stage of spores [17]. Lower atmospheric levels with vegetation give best condition for the development of myxomycetes fruting body and to distribute their spores by air. So data for environmental certification of slime molds can be extrapolated to other organisms living in soil, air and even water. Myxomycetes are distributed nearly in all terrestrial ecosystems worldwide from Antarctica to desert. Approximately 30% of the species are cosmopolitan, but they are the most common in temporary zones of North Hemisphere, where moisture and decaying organic matter are available [17]. Temperature and humidity are the main factors that regulate myxomycetes distribution and abundance. They are widespread in various habitats in the world, which is also a significant argument for choice slime molds as a model object for environmental certification.

The role of myxomycetes role in ecosystems is not studied enough yet. They help to keep bacterial levels and are important for nutrient cycling and forest productivity. Slime molds have ability to decompose plant matter and absorb nitrogen and heavy metals. This shows their potential use for bioremediation and pollutant removal. Myxomycetes can be used as air quality indicators of natural ecosystems and urban areas. Also they are indicators for evaluation of forest ecosystem condition. As we can see from this research, myxomycetes are not dangerous for human health and it gives possibility to discover their internals, which can be useful for people.

Myxomycetes are excellent model for genetic, biochemistry, biophysics, bionic, bioengineering and other researches. For example, *Physarum polycephalum* is an extensively studied system in biophysics. The plasmodial stage is of particular interest, since it exhibits, despite the relatively simple organization of this unicellular organism given an external stimulus, the plasmodium optimizes its cell shape, vein network and growth with respect to transport efficiency, robustness with respect to link deletion and avoidance of unfavorable conditions. Also remarkable phenomenon is the synchronization of the contraction patterns in the tubular vein network that generates shuttle streaming to distribute nutrients efficiently throughout the organism. From the perspective of biophysics it is natural to consider these phenomena in the framework of self-organized complex systems [11].

The plasmodium of *Physarum polycephalum* changes its shape as it crawls over a plain agar gel and, if food is placed at two different points, it will put out pseudopodia that connect the two food sources. Plasmodia were able to find the shortest path between two food sources placed at the exits of a labyrinth. Here we show that this simple organism has the ability [9].

Plasmodium creates networks to connect food sources with incredible efficiency to reproduce public transport networks on the scale of a petri dish. *Physarum polycephalum* forms networks with efficiency, tolerance and cost comparable to those of real-world infrastructure networks (Tokyo rail system). The core mechanisms needed for adaptive network formation can be captured in a biologically inspired mathematical model that may be useful to guide network construction in other domains [20].

Recently an interest in technological developments started to move away from solid materials to soft matter implementations and bio-inspired and hybrid implementations: bio-mimetic sensors which employ a conductive fluid encapsulated in elastic container and use deformation of the elastic container in transduction, carbon nanotube filled elastomers, polymer hair cell sensors. An experimental laboratory implementation of *Physarum polycephalum* was designed based on tactile bristles. The plasmodium responds to repeated deflection of bristle by an immediate high-amplitude spike and a prolonged increase in amplitude and width of its oscillation impulses. The signal strength of myxomycetes tactile bristle sensor averages near six for an immediate response and two for a prolonged response [1].

Myxomycetes use an externalized spatial memory to navigate in complex environments. *Physarum polycephalum* constructs a form of spatial memory by avoiding areas it has previously explored. This mechanism allows the slime mold to solve the U-shaped trap problem — a classic test of autonomous navigational ability commonly used in robotics requiring the slime mold to reach a chemoattractive goal behind a U-shaped barrier. Drawn into the trap, the organism must rely on other methods than gradient-following to escape and reach the goal. The spatial memory enhances the organism's ability to navigate in complex environments. It is a unique demonstration of a spatial memory system in a nonneuronal organism, supporting the theory that an externalized spatial memory may be the functional precursor to the internal memory of higher organisms [22].

All these examples show that myxomycetes are very popular model objects in various researches. Their unique characteristics give possibility for slime molds to be used for working-out with structure of "Environmental safety passports of species" as a biological system model. This structure can be adapted for certification of other groups of organisms, in accordance with their morphology and characteristics of metabolism.

ENVIRONMENTAL SAFETY PASSPORT OF SPECIES

"Environmental safety passports of species" (ESPS) are necessary for effective monitoring and management of environmental safety. Implementation of species certification will increase the level of control in the system of ecological safety and effectiveness of protection actions for environment and humans. The main purpose of the introduction of passports is to systematize scientific information for determination of effect of this species on the environment, humans and other living organisms. The new system will enhance control level of species metabolism and improve the effectiveness of environmental protection. ESPS is a comprehensive document which is necessary for all species of living organisms. Such passport describes relationship between these species, other organisms and human with evaluating threatens from all sides. There are also analyses of impacts of abiotic factors from habitat to species vital activity to show range of tolerance, optimum, minimum and maximum value of each factor. The conclusion will be made about real and potential risks associated with this species for other organisms, human and environment. The recommendations will be designed for reduction or prevention of such risks. Also threats will be evaluated for this species from other organisms, human and different environmental factors. According to them the strategy plan of species protection will be developed.

ESPS summarize data of scientific researchers from different fields of science in any accessible way. Certification will provide the most comprehensive information about safety aspects of the species from reliable sources of information. All available data about each species of living organisms will be conveniently structured in such passport. ESPS will have an open structure for addition of new data from experts. In such way scientific information becomes available to a wide range of stakeholders and it helps to avoid wrong interpretations and systematization of environmental safety will be reinforced.

This minimizes the possibility of incorrect interpretation of scientific data and the conditions for constructive interaction between scientists and managers.

Introduction of ESPS will improve the system of environmental safety for the effectiveness of measures enhancement to protect the nature and people.

CONCLUSIONS

1. Correct evaluation of threats from biological objects to environment, human and other living organisms is very important for environmental safety.

2. Analysis of myxomycetes habitats, ecology, distribution and metabolism show that these organisms can't be really dangerous for health of people.

3. In environmental safety Bayesian methods for probability analysis is perspective methodology for evaluation of risk assessment of threats from biological objects to environment, human and other living organisms.

4. For proper interpretation of results of biological research in environmental safety the implementation of "Environmental safety passports of species" is proposed, which include veracious scientific information about these organisms and evaluation of risk probability.

5. Myxomycetes are suitable for being used as a model organism in process of development of "Environmental safety passports of species" structure.

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ОЦЕНКА РИСКА УГРОЗ СО СТОРОНЫ БИООБЪЕКТОВ В СФЕРЕ ЭКОЛОГИЧЕСКОЙ БЕЗОПАСНОСТИ

Аннотация. Обоснована необходимость предоставления результатов научных исследований в форме, доступной для правильной интерпретации общественностью и экспертами в сфере экологической безопасности. В качестве примера была доказана безопасность миксомицетов для здоровья человека, путем анализа особенностей их экологии и метаболизма. Для оценки рисков предложено использовать Байесовские методы анализа вероятности возникновения угроз. Рекомендовано внедрение «Паспортов экологической безопасности видов» для оценки воздействия каждого вида на окружающую среду, человека и другие живые организмы.

Ключевые слова: экологическая безопасность, оценка рисков, миксомицеты, экологический паспорт.