# Diatomic compounds in the soils of bee-farm and nearby territories in Samarskaya oblast

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**Abstract.** This article sheds light on the role diatomic algae in soils play in the assessment of bee farm and nearby territories in four soil and landscape zones in Samarskaya Oblast. The community of diatomic algae is characterized by low species diversity, of which 23 taxons were found. The most often found species are represented by *Hantzschia amphioxys* (*Ehrenberg*) Grunow in Cleve & Grunow and *Luticola mutica* (*Kützing*) D.G.Mann in Round et al. The maximum of phyla (18) were found in the buffer (transient) zone; in the wooded steppe zone, 11 species were recorded; in the steppe – 2 species, and in the dry steppe zone no species of diatomic algae were found. The qualitative and quantitative characteristics of diatomic algae communities in various biotopes depend on the natural and climate features of a territory and the degree of the anthropogenic impact on the soil and vegetation, which is proved by the fact that high species wealth signifies that the ecosystem is stable and resilient to the changing conditions in the environment, while poor algal flora is less resilient due to the lower degree of diversity.

## 1 Introduction

Beekeeping is a basic branch of agriculture with close connections to crop farming. A key condition for its intensification is the presence of wild and cultivated plants growing on soils that are not contaminated with ecotoxicants [1, 2]. Nowadays, however, bee farms are usually located in residential (inhabited) areas, that often act as contamination centers. They are especially harmful to soils due to the contamination of stationary sources with ecotoxicants, the use of cars, etc [2–4]. Setting up bee farms without considering the ecological safety in the area is one of the key factors causing honey bee diseases (*Apis mellifera*) because many ecotoxicants have cumulative properties and are able to impact metabolic cycles. They make up highly toxic metallic organic compounds that reduce the resistant abilities of organisms. This leads to an increase in disease rate, colony collapse, and population decline [3].

All of the abovesaid makes the research of the ecological conditions of soils as a habitat and substrate for honey plants and bees relevant these days. One of the directions of theoretical and applied research in this respect is the assessment of the environmental position of bee-farm and nearby soils using the bioindication method. Currently, various organisms are used as bioindicators, including hydrobionts, insects, honey products, diatomic algae (*Bacillariophyceae*), etc. According to specialists [5], diatomic algae are an integral part of the land ecosystem. They belong to the first link of environmental food chains and take an active part in the cycle of matter. Diatomic algae play a special role in habitats with extreme conditions [6, 7].

The goal of the work is the study of the diversity of diatomic algae communities in bee-farm and nearby soils within the soil and landscape zones of Samarskaya oblast [8].

## 2 Materials and Methods

Samarskaya oblast is located in the South-East of the European part of Russia, between 47°55′ and 52°35′ east longitude and 51°47′ and 54°41′ of north latitude. According to the landscape and climate zoning principles, four soil and landscape zones can be found there (Fig. 1). The wooded steppe is located on typical, leached, and degraded black soil (chernozem); grey forest soils, rendzina soils, and floodplain soils. The vegetation there is represented by deciduous and coniferous forests, intermingled with meadow steppe, floodplains, and marshes. The woodland amounts to 22–26 %. The buffer zone (transient from the wooded steppe to steppe) is located on the same black soils as the wooded steppe, but also on alkaline soils, grey forest soils, and flood plains with mixed vegetation. The steppe zone is located on southern and common black soils, dark brown soils, alkaline soils, and flood plains with almost no woodland and a great abundance of feather grass, mixed herb, and fescue steppes, shrubland, stony and sandy steppes. The woodland amounts to 1.5 %. The dry steppe zone is located on southern black soils, dark brown

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soils, alkaline soils, and flood plains with narrow silvicultural strips.


**Fig. 1.** Soil and landscape zones in Samarskaya oblast: I – wooded steppe, II – transient zone from wooded steppe to steppe (buffer zone), III – steppe zone and IV – dry steppe zone

The research material is represented by 28 mixed soil samples. Sampling and diversity analysis was carried out with classic algological methods [6, 9]. The technical preparation of samples was done using the standard procedure [7]. To prepare the specimen, the valves were cleaned of protoplasts by boiling in concentrated nitric acid and put into Naphrax™ resin [10]. To identify single species and in-species taxons, we used a visual abundance estimate expressed in points on a modified Kolbe-Visloukh scale [11]. The dominating or main compound was made up of species and in-species taxons of 3-6 points abundance. Diatomic algae with 6 points counted as dominant, subdominants had 4–5 points, and those with 3 points were deemed companion species. The identification was performed using foreign guides [12–16]. To assess the sustainability of the development of diatomic algae compounds, we used the frequency index. The species frequency was calculated using the following formula:

$$C = \frac{n}{N} \times 100 \,, \quad (1)$$

where C is the frequency in %; n is the number of samples in which the species was found, and N is the total number of samples.

Taxons whose species count was above average were deemed dominant.

## 3 Results and Discussion

The research we carried out allowed us to identify 23 species and in-species taxons, belonging to 2 classes, 6 orders, 9 families and 11 genera of diatomic algae: *Achnanthes lanceolata* (Brébisson ex Kützing) Grunow in Van Heurck, *Amphora ovalis* (Kützing) Kützing, *Gomphonema parvulum* (Kützing) Kützing, *Gomphonema angustatum* (Kützing) Rabenhorst, *Hantzschia amphioxys* (Ehrenberg) Grunow in Cleve & Grunow, *Hantzschia abundans* Lange-Bertalot, *Hantzschia amphioxys* f. *capitata* Otto Müller, *Humidophila contenta* (Grunow) Lowe, Kociolek, J.R.Johansen, Van de Vijver, Lange-Bertalot & Kopalová, *Luticola cohnii* (Hilse) D.G.Mann in Round, Crawford & D.G.Mann, *Luticola mutica* (Kützing) D.G.Mann in Round et al., *Luticola ventricosa* (Kützing) D.G.Mann in Round, Crawford & Mann, *Navicula minima* Grunow in van Heurck, *Nitzschia perminuta* (Grunow) M. Peragallo et al. (Fig. 2).


**Fig. 2.** Species frequently found on the explored territories in Samarskaya oblast: 1 – Amphora ovalis, 2 – Hantzschia amphioxys, 3 – Hantzschia amphioxys f. capitata, 4 – Luticola mutica, 5 – Pinnularia borealis, 6 – Pinnularia microstauron.

The leading taxons according to the number of species and in-species taxons are as follows: the *Bacillariophyceae* class, the *Naviculalles* order, the *Bacillariaceae* family, and the *Pinnularia* genus (Table 1).

**Table 1.** The leading taxonomic ranks in Samarskaya oblast according to the number of species and in-species taxons

| Class | Order | Family | Genus |
|---|---|---|---|
| *Bacillariophyceae* (22) | *Naviculales* (12) | *Bacillariaceae* (6) | *Pinnularia* (5) |
| | *Bacillariales* (6) | *Diadsmidaceae* (5) | *Luticola* (4) |
| | | *Pinnulariaceae* (5) | *Hantzschia* (3) |
| | | | *Nitzschia* (3) |

Note. The number in brackets represents the number of species

Overall, the results show a rather low species diversity of diatomic algae communities in the soils of the explored territories. The results show the uneven zone distribution of taxonomic ranks of diatomic algae in Samarskaya oblast (Table 2).

**Table 2.** The taxonomic wealth and diversity of diatomic algae compounds in various zones of Samarskaya oblast

| Indicators | WS | BZ | ST | DS |
|---|---|---|---|---|
| No. of classes | 1 | 2 | 1 | - |
| No. of orders | 3 | 6 | 2 | - |
| No. of families | 6 | 7 | 2 | - |
| No. of genera | 7 | 9 | 2 | - |

The number of species and in-species taxons of algae gradually reduces going north to south. The maximum number of taxonomic groups is found in the buffer zone. Then this index drops from the wooded steppe to the steppe zone. The soils in the bee-farm lands in the wooded steppe zone were less diverse in terms of diatomic algae. The steppe soils took the third place in algae diversity. In the last zone, i.e. the dry steppe, no specimen was found.

For the majority of diatomic algae species and in-species taxons (91 %), found in bee-farm lands, the frequency rate is between 4 and 18 %. Only two species have a frequency rate of over 30 %, and these species are deemed leading: *Hantzschia amphioxys* (*Ehrenberg*) Grunow in Cleve & Grunow (68 %) and *Luticola mutica* (*Kützing*) D.G. Mann in Round et al. (36 %). These algae are the least susceptible to shifting environmental factors. Many researchers pointed out that the overall spread and constant occurrence of *Hantzschia amphioxys* in various ecosystems is a sign of its 'cosmopolitanism' and high resilience to external impacts [6].

The dominant species set was small and it included specimens from four families (*Bacillariaceae, Catenulaceae, Diadesmidaceae, Gomphonemataceae*). Five species were dominant (*Amphora ovalis, Gomphonema parvulum*, *Hantzschia abundans*, *Hantzschia amphioxys* f. *capitata*, *Luticola mutica*), while no subdominants were found. The set of companion species included two taxons (*Luticola ventricosa, Nitzschia fonticola)*. 16 diatomic algae species showed only single or sporadic occurrences. To see the changes in the diatomic algae set structures, we analyzed the floral structures of all zones.

***Wooded steppe zone.*** The total number of species recorded: 11. Species of six families played a significant role in the formation of the algae set. The first and the second places went to two families *Pinnulariaceae* и *Bacillariaceae*, consisting of three species, and the third place went to the *Diadesmidaceae* family, comprising two taxons. The rest of the families comprised 27 % of all species diversity. In this zone, different administrative districts were investigated and they all have different soils, such as Syzranskiy district (near Syzran and Oktyabrsk – various black and dark brown soils), Shentalinskiy district (grey forest soils), Volzhskiy districts (near Samara – leached and degraded black soils, grey forest soils, flood plains), Stavropolskiy district (near Togliatti – typical black and grey forest soils), Krasnoyarskiy district (typical black, meadow black and flood plain soils), Isaklinkskiy (typical black soils), Pokhvistnevskiy district (leached and degraded black soils, grey forest soils, and flood plain soils), as well as a plot of land near the Samarskaya Luka National Park, spanning the territories in Volzhskiy, Stavropolskiy and Syzranskiy districts (various black and rendzina soils).

The highest number of species was found in bee farming territories in Stavropolskiy district. The species diversity of diatomic algae is characterized by the presence of algae families *Diadesmidaceae* and*Bacillariaceae* and is due to the wetness of vegetational cover [5] of grey forest soils. This led to the increase in algae community diversity due to the development of the *Pinnularia* genus specimens that are often found in forest ecosystems.

The least diatomic species diversity – only two species (*Hantzschia amphioxys, Luticola mutica*) – was found in the Samarskaya Luka National Park, which is explained by the large proportion of stony steppe there, which is formed on stony outcrops with poor, washed-off soils that often have a heavy texture that is unsuitable for the growth and the development of algae. In other samples, the number of algae species varied from one to three due to the anthropogenic impact and the changes in natural wooded steppe vegetation. The high concentration and active operation of oil refining facilities, chemical plants, and other industries in the explored territories lead to the depletion of natural resources, contamination of biosphere with production wastes, destruction of natural ecosystems, and, as a consequence, to the decrease in algae diversity.

In the soil samples from the wooded steppe zone, only *Hantzschia amphioxys*, *Hantzschia amphioxys* f. *capitata* Otto Müller and *Luticola mutica* developed in abundance, which is due to the resilience of these specimen to various environmental factors.

***Buffer zone.*** Soil samples here show the greatest number of algae species and in-species taxons – 18 (78 % of the species diversity). The number of species increased due to the ecotone effect that conditioned the high level of biodiversity. The increase was implemented due to the natural introduction of wooded steppe and steppe species [17, 18].

In this territory, the *Bacillariaceae* family (five taxons) holds the first place, then comes the *Pinnulariaceae* family (four species), then the *Diadesmidaceae* family with three species and the *Gomphonemataceae* with two taxons. In this zone, plots with different soils were chosen for analysis, belonging to four administrative districts: Kinel-Cherkasskiy district (various black soils), Kinelskiy district (black soils, soil gardens and flood plains), Privolzhskiy district (common black soils), Bezenchukskiy district (various black soils) and two urban districts (Novokuybyshevsk – common black soils and Chapayevsk – common, southern and flood plain black soils). The number of species in samples varied greatly depending on the location and it was from 1 to 14 species. Despite the anthropogenic load, the samples collected from the bee farms near Novokuybyshevsk showed high diversity – 14 algae species (78 %) were found in them. In this biotope, such species as *Hantzschia amphioxys*, *Gomphonema parvulum* (Kützing) Kützing and *Amphora ovalis* (Kützing) Kützing reached the mass development levels. The companion species were represented by *Luticola mutica* and *Nitzschia fonticola* (Grunow) Grunow in Van Heurck.

A decent species diversity (six species or 34 %) was detected in samples from Kinelskiy district. The dominating species are *Hantzschia amphioxys* and *Luticola mutica*. The companion species is represented by *Hantzschia amphioxys* f. *capitata. P.* In the soils from bee farms in Kinel-Cherkasskiy district, species from the *Pinnulariaceae* family were found. Other districts showed poor algae development. Only *Hantzschia amphioxys* could be found everywhere. The uneven distribution of diatomic algae in the soils of this zone is explained by the high concentration of underground resources and industries. The presence of diverse communities of diatomic algae near Novokuybyshevsk is due to the fact that the further from the source of contamination you get, the more diverse and numerous the ecosystems are [18, 19].

***Steppe zone.*** The smallest diatomic algae set was found here. The reason is the plot's location: it is the true (feather grass and fescue) steppe zone, undergoing intensive grazing load, which leads to soil compaction, grass cover exhaustion, and stream conditions disruption. Besides, this territory features operating mining companies and associated gas treatment facilities, metal works, etc. Within the zone, plots in three districts were selected for the analysis: Alekseyevskiy (southern black soils, dark brown soils, and salt gardens), Khvorostyanskiy (southern black soils) and Neftegorskiy (near Neftegorsk – southern and common black soils). A total of two species of the families *Diadesmidaceae* and *Bacillariaceae* were found. Both species – *Hantzschia amphioxys* and *Luticola mutica* – were among the dominants in Alekseyevskiy district. In Khvorostyanskiy and Neftegorskiy districts, algae were not found. Apart from the impact of anthropogenic-origin ecotoxicants, the decrease of diversity in steppe communities is due to the indirect influence of topsoil disturbance because of grazing [5]. Some authors claim [20] that topsoil compaction, poor turfness, and grass cover sparseness on pastures have a positive effect on the development of other organisms, e.g. cyanobacteria, a competitor of diatomic algae. The suppression of diatomic algae in short grass and feather grass communities might be caused by other factors, such as dense turf and thick grass stand (90-100 %).

***Dry steppe zone.*** In this territory, sampling was performed in the bee-farms of the Bolshechernigovskiy district. No diatomic algae were found here. This is mostly due to the presence of several large operating oil prospecting and refining facilities, as well as companies mining oil shale, mineral construction materials, sand, and gravel. All of the above create adverse conditions for the development of diatomic algae.

## 4 Conclusion

The changes in the floristic composition of higher plants, soils, and environmental conditions going from the north to the south of Samarskaya oblast are reflected in the changes of species diversity and diatomic algae set structure. Herewith, the highest number of diatomic algae species and in-species taxons were found in the buffer zone, in the Novokuybyshevsk urban district, while to the north and the south of the buffer zone, the number of species was reduced. In the northern wooded steppe zone (Stavropolskiy district), the number of species reduced two-three times, and in the southern portion of the oblast, the species composition reduced more dramatically. In the steppe zone (Alekseyevskiy district), the number of species reduced seven times, and in the dry steppe zone (Bolshechernigovskiy district) no algae were found.

The analysis of family ratio showed that in the wooded steppe zone the *Bacillariaceae* and *Pinnulariaceae* families were dominant, in the buffer zone – the *Bacillariaceae*, *Pinnulariaceae* and *Diadesmidaceae*, families, and in the steppe zone – the *Bacillariaceae* and *Diadesmidaceae*. However, in all of the districts, the leading species were represented by two: *Hantzschia amphioxys* and *Luticola mutica*, except for the dry steppe zone where no diatomic algae were found. The results of the taxonomic structure analysis for diatomic algae sets show significant discrepancies in the number of orders, families, genera, and species, which, first of all, signifies a close connection between the algae communities and environmental conditions.

The research performed showed that the most resilient and stable sets of diatomic algae in soils are the communities of Stavropolskiy, Kinel-Cherkasskiy, and Kinelskiy districts, as well as Novokuybyshevsk urban district. The algae communities of Syzranskiy, Kuybyshevskiy, Krasnoyarskiy, Pokhvistnevskiy, Volzhskiy, Privolzhskiy, Bezenchukskiy, and Alekseyevskiy districts, the Samarskaya Luka National Park and Chapayevsk urban district proved to be less resilient in terms of the number of species. The minimum number of algae species were found in Shentalinskiy, Isaklinskiy, Khvoroskyansky and Neftegorskiy districts. In the Bolshechernigovskiy district in the dry steppe zone, no diatomic algae were found.

The results of environmental monitoring of soil and landscape zones in Samarskaya oblast (wooded steppe, buffer/transient, steppe, dry steppe) showed that the soils of bee-farm and nearby areas, where ecotoxicants were recorded officially, have the least of diatomic algae taxonomic groups or none at all.

Therefore, the methodology presented in this work allows evaluating the environmental position of the soil by using soil diatomic algae, irrespective of the geographic location of the bee farm in question. It also provides for the timely decision making to prevent bee farms being set up in environmentally adverse areas and thus avoid the collapse of evolutionary genetic bases of populations, including honey bees.

## References

1. M.D. Yeskova, *The biological bases of bee farming*: study guide (RGAZU, Moscow, 2010), 175 p.
2. N.Ye. Zemskova, *Morphological assessment of the honey bee in Samarskaya oblast in introgressive and anthropogenic aspects* (Doct. Dissertation thesis) (Editor. and Publ. Depart. of Samara State Agrar. Univer., Kinel, 2017), 38 p.
3. S.A. Pashayan, *Ecological and biological bases determining bees' resistance to diseases* (Doct. Dissertation thesis) (Yekaterinburg, 2012), 38 p.
4. V.N. Sattarov, V.R. Tuktarov, I.M. Borisov, et al., *The impact of stationary ecotoxicant sources on the*

*honey bees' habitat in the Republic of Bashkortostan*, Beekeep. **2**, 8–9 (2011)

5. A.I. Fazlutdinova, R. R. Kabirov, *Soil diatomic algae of the Southern Urals* (Gilem, Ufa, 2013), 128 p.
6. M.M. Gollerbakh, E.A. Shtina, *Soil algae* (Nauka, Leningrad, 1969), 228 p.
7. *Diatomic algae of the USSR (fossilized and modern)*, ed. by A.I. Proshkina-Lavrenko, vol. 1 (Nauka, Leningrad, 1974)
8. State Report on the Environment Conditions and Natural Resources in Samarskaya oblast for 2018, **29**, (2019), Samara. Retrieved from: http://www.priroda.samregion.ru/external/priroda/files/c_116/Doklad_ob_ekologicheskoj_situatsii_na_territorii_Samarskoj_oblasti_-_2018_god.pdf
9. F.Kh. Khaziyev, R.R. Kabirov, *Quantitative methods of soil and algological research* (BFAN, USSR, Ufa, 1986), 172 p.
10. F. Acker, B. Russell, E. Morales, *Preparation of diatom slides using Naphrax™ mounting medium*, PCER, ANSP, Protocol, **13-49**, 41–54 (2002)
11. A.S. Stenina, *Diatomic algae (Bacillariophyta) in lakes of eastern Bolshezemelskaya tundra* (Komi Sci. Center of Ural Branch of the Russ. Acad. of Sci., Syktyvkar, 2009), 176 p.
12. H. Ettl, G. Gartner, *Syllabus der Boden-, Luft-, und Flechtenalgen* (VEB Gustav Fischer Verlag, Stuttgart, 1995)
13. K. Krammer, H. Lange-Bertalot, *Bacillariophyceae, 1: Naviculaceae Sußwasserflora von Mitteleuropa* (VEB Gustav Fischer Verlag, Stuttgard; New-York, **2**, 1986), 876 p.
14. K. Krammer, H. Lange-Bertalot, *Bacillariophyceae, 2: Bacillariaceae, Epithemiaceae, Surirellaceae Sußwasserflora von Mitteleuropa* (VEB Gustav Fischer Verlag, Jena, **2**, 1988), 536 p.
15. K. Krammer, H. Lange-Bertalot, *Bacillariophyceae, 3: Centrales; Fragilariaceae, Eunotiaceae Sußwasserflora von Mitteleuropa* (VEB Gustav Fischer Verlag, Stuttgard; Jena, **2**, 1991a), 576 p.
16. K. Krammer, H. Lange-Bertalot, *Bacillariophyceae. 4: Ahnanthaceae, Kritische Erganzungen zu Navicula (Lineolatae) und Gomphonema Gesamtliteraturverreichnis Sußwasserflora von Mitteleuropa.* (VEB Gustav Fischer Verlag, Stuttgard; Jena, **2**, 1991b), 437 p.
17. A.S. Stenina, B.Yu. Teteryuk, Ye.N. Patova, *Plant communities of coastal ecotones in the Vangyr river valley in the Nether-Polar Urals,* Writings of Komi Sci. Center of Ural Branch of the Russ. Acad. of Sci., **165**, 20–36 (2001)
18. M.Yu. Sharipova, *Algocoenoses of ecotones: ecology, floristic composition, structure as exemplified by the Southern Urals region* (Cand. Dissertation thesis) (Ufa, 2006), 37 p.
19. E.A. Shtina, G.M. Zenova, N.A. Manucharova, *Algological monitoring of soils,* Eurasian Soil Sci., **12**, 1449–1461 (1998)
20. L.N. Novichkova-Ivanova, *Soil algae in the phytocoenoses in the Sahara-Gobi desert region* (Nauka, Leningrad, 1980), 256 p.