

VIII INTERNATIONAL CONFERENCE

29-31 March 2023



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**«Advanced Agritechnologies, Environmental Engineering and Sustainable Development»**

**AGRITECH-VIII 2023**

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**«Assessment (*in vitro*) toxicity of small molecules of plant origin»**

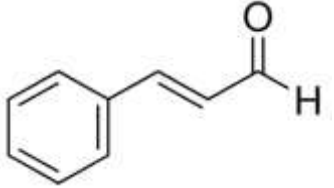
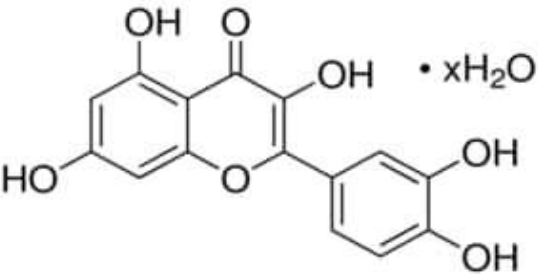
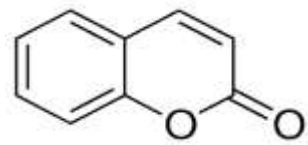
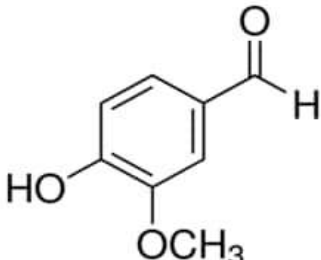
*Ludmila Vlasenko, and Kseniya Atlanderova*

# Problem statement

- Phytochemicals have become the subject of in-depth study in recent years. The special properties of these compounds determine the possibility of their use in various fields of human activity, including the pharmaceutical, cosmetic, agricultural and food industries.
- However, this makes it necessary to conduct appropriate tests and determine their danger, which can also be useful in justifying the use of small molecules of plant origin as feed additives.

Therefore, the goal of this work was to determine the toxicity of various chemically synthesized small molecules of plant origin using the *E. coli* MG1655 pXen7 *lux*-biosensor and the *Stylonychia mytilus* test system as test objects.



Test substances	Chemical formula	Structural formula	Molar mass
Trans-cinnamaldehyde (cinnamaldehyde), 99 %, C80687	$C_9H_8O$		132.16 g/mol
Quercetin hydrate (quercetin), $\geq 95$ %, 337951	$C_{15}H_{10}O_7 \cdot xH_2O$		302.24 g/mol
Coumarin, $\geq 99$ %, C4261	$C_9H_6O_2$		146.14 g/mol
Vanillin, 99 %, V1104	$C_8H_8O_3$		152.15 g/mol

**Table 1.** General characteristics of small molecules of plant origin.

# Materials and methods

## Bioluminescent test

A laboratory luminescent strain *E. coli* K12 MG1655 (pXen7) is used in the work, which was obtained by transforming cells of the host strain with a hybrid plasmid pUC18 with an embedded *EcoRI* DNA fragment containing the *lux*-CDABE genes of the soil microorganism *Photorhabdus luminescens* ZM1.

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The calculation of the toxicity index (TI) was carried out according to the equation:

$$TI = (RLU_{K_0} \times RLU_{O_n}) / (RLU_{K_n} \times RLU_{O_0}),$$

where  $RLU_{K_0}$  and  $RLU_{O_0}$  – luminescence values of control and experimental samples at the 0-th minute of measurement,  $RLU_{K_n}$  and  $RLU_{O_n}$  – glow values at the n-th minute of measurement.

## Protozoa test

The ciliates *Stylonychia mytilus* were used as a test object, which were grown on Lozin-Lozinsky medium at 23°C.

The cessation of the movement of protozoa, the violation of their integrity, which ultimately led to cell lysis, indicated the toxic effect of the substances under study.



# Results

Test materials	60 minutes	120 minutes	180 minutes
Trans-cinnamaldehyde	<0.78	<0.78	<0.78
Quercetin hydrate	-	-	-
Coumarin	3.10±0.28	2.80±0.25	2.70±0.21
Vanillin	2.90±0.54	2.50±0.17	2.40±0.19

Notation: «-» no toxic effect.

**Table 2.** The values of the toxicological parameter EC50 (mM) determined for the studied small molecules of plant origin in assessing their effect on the *lux*-biosensor *E. coli* K12 MG1655 pXen7

## Conclusion

The obtained data on the toxicity of small molecules of plant origin can be further used for the targeted development of new and safe components for their inclusion in the diet of animals (including cattle) as an alternative to feed antibiotics.

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