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### Area of research:

- 1. Enzymes of lipoxygenase cascade and plants protection proteins;
- 2. Molecular mechanisms of plant-microbe interactions;
- 3. Adaptive strategies of microorganisms associated with the formation of structured bacterial populations.

#### Main results:

- 1. For the first time there was obtained the model of the pathological changes of plant tissues and *Pectobacterium atrosepticum* cell differentiation during colonization of *Nicotia na tabacum* plants. A new type of the structured microbial communities which we referred to as bacterial embols was identified. During bacterial diseases the embols were formed in the plant xylem vessels causing plant wilt symptoms.
- 2. The technique for quantitative definition of transcriptional activity of gene loci differentially estimating the RNA synthesis from sense and antisense strands of DNA has been developed. According to this technique we have shown that the ratio of sense and antisense transcripts changes significantly depending on the growth phase of cell population of *Pectobacterium atrosepticum*.

The data obtained enrich our understanding about bacterial regulatory networks associated with regulatory RNAs.

3. An understanding of the adaptive strategies of gramnegative and grampositive bacteria ( *Erwinia carotovora* 

Escherichia coli

Xanthomonas campestris

Azospirillum brasilense

Salmonella enterica

Bacillus subtilis

) to substrate starvation has been developed. It has been established that under starvation conditions the magnitude of population increase or decrease (depending on initial population density), stabilizing at 10

CFUs/ml. Quorum sensing is shown to take part in this process. It has been demonstrated that bacteria is able to increase their numbers in absence of carbon and phosphor sources no less than three order of magnitude at the expense of formation of cells with reduced size and atypical morphology.

- 4. The sites determining the mechanism of catalytic action of CYP74 enzymes have been revealed. As a result of single amino acid replacement in the revealed sites the conversions of enzymes of different functional groups have been made: tomato allene oxide synthase (dehydrases) LeAOS3 into hydroperoxyde lyase (isomerases), flax divinyl ether synthase LuDES into allene oxide synthase.
- 5. We have developed the model of interaction of substrates (fatty acids and complete lipids) with active centers of maize (ZmLOX3) and soya (GmLOX1) lipoxygenases. According to the model, the substrate targeting is not defined by the positional (9- or 13-) specificity of lipoxygenase reactions, but determined by the structural and functional enzymes features.
- 6. For the first time the participation of (7*S*)-hydroperoxide of hexadienoic acid in lipoxygenase cascade was shown. Under the action of tobacco divinyl ether synthase and maize allene oxide synthase (7 S)-hydroperoxide converts into divinyl ether (?) and alpha-ketol, respectively.
- 7. The model of P450 cytochromes molecular phylogeny was obtained according to which the CYP74 family was originated before the divergence of the last common ancestry of eukaryotes and could participate in initial steps of P450 cytochromes evolution.

The members of the Laboratory participate in joint investigations with other departments of the Institute. The foreground task of the Laboratory is to render assistance for conducting genomeand transcriptome-wide studies and using recombinant proteins in biochemical and biophysical experiments.

## Main publications:

1. Grechkin A.N., Mukhtarova L.S., Latypova L.R., Gogolev Y., Toporkova Y.Y., Hamberg M. Tomato CYP74C3 is aMultifanctional Enzyme not only Synthesizing Allene Oxide but also Catalyzing its Hydrolysis and Cyclization // Chembiochem, 2008. – V. 9. – N.15. – P249-2505.

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- 8. Gogolev Yuri V., Gorina Svetlana S., Gogoleva Natalia E., Toporkova Yana Y., Chechetkin Ivan R., Grechkin Alexander N. Green leaf divinyl ether synthase: Gene detection, molecular cloning and identification of a unique CYP74B subfamily member // Biochimica et Biophysica Acta, 2012. Vol. 1821. P. 287 294.
- 9. Grechkin AN, Lantsova NV, Toporkova YY, Gorina SS, Mukhitova FK, Khairutdinov BI. Novel Allene Oxide Synthase Products Formed via Favorskii-Type Rearrangement: Mechanistic Implications for 12-Oxo-10,15-phytodienoic Acid Biosynthesis. ChemBioChem., 2011. Vol. 12(16). P. 2511-2517.
- 10. Nadezhda Tarasova, Vladimir Gorshkov, Olga Petrova, Yuri Gogolev. Potato signal molecules that activate pectate lyase synthesis in Pectobacterium atrosepticum SCRI1043. World Journal of Microbiology and Biotechnology. DOI 10.1007/s11274-013-1281-9.
- 11. Petrova O.E. Stress response in Pectobacterium atrosepticum SCRI1043 under starvation conditions: adaptive reactions at a low population density / O. Petrova, V. Gorshkov, A. Daminova, M. Ageeva, L.N. Moleleki, Y. Gogolev // Research in Microbiology. 2014. V. 165. P. 119–127.
- 12. Gorshkov V. Dissociation of a population of *Pectobacterium atrosepticum* SCRI1043 in tobacco plants: formation of bacterial emboli and dormant cells. / V. Gorshkov, A. Daminova, M. Ageeva, O. Petrova, N. Gogoleva, N. Tarasova, Y. Gogolev // Protoplasma. 2013. Published online DOI: 10.1007/s00709-013-0546-3.
- 13. Toporkova Y.Y. Structure-function relationship in CYP74 family: conversion of divinyl ether synthases into allene oxide synthases by site-directed mutagenesis / Y.Y. Toporkova, V.S. Ermilova, S.S. Gorina, L.S. Mukhtarova, E.V. Osipova, Y.V. Gogolev, A.N. Grechkin //

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