CURRICULUM VITAE

ALENA DABRAZHYNETSKAYA

NCI-Frederick
Gene Regulation and Chromosome
Biology Lab, Bldg. 539, Rm. 204
PO Box B, Frederick
MD 21702
Phone: 301-846-1256
Fax: 301-846-6988
e-mail: elenad@ncifcrf.gov

EDUCATION:

1995 - Ph.D. in Genetics and Biotechnology. *Theses:* "Regulation of aromatic amino acids biosynthesis in the obligate methylotrophic bacteria *Methylobacillus mucogenes*". Belarus State Institute of Genetics and Cytology, Minsk, Belarus.

1987 - Diploma in Biology, with honour (Equivalent of the US Master in Science degree). Theses: "Regulation of phenylalanine biosynthesis in the obligate methylotrophic bacteria *Methylobacillus M75"*. Belarus State University, Department of Microbiology, Minsk, Belarus.

EMPLOYMENT HISTORY:

08/2001 – ***present.*** – Visiting Fellow. Cell Cycle Regulation Section, Gene Regulation and Chromosome Biology Laboratory, National Cancer Institute - Frederick, NIH, Frederick, MD, USA.

08/2000-08/2001 – Docent (Equivalent of USA Associate Professor), Department of Genetics and Biotechnology, Belarus State University (BSU), Minsk, Belarus.

07/1998-08/2000 - Assistant Professor, Department of Genetics and Biotechnology, BSU, Minsk, Belarus.

02/1996-07/1998 – Senior Scientist, Molecular Genetics of Bacteria Laboratory, Department of Microbiology, BSU, Minsk, Belarus.

05/1992-02/1996 - Research Scientist, Molecular Genetics of Bacteria Laboratory, Department of Microbiology, BSU, Minsk, Belarus.

08/1987-05/1992 - Junior Researcher, Molecular Genetics of Bacteria Laboratory, Department of Microbiology, BSU, Minsk, Belarus.

MAJOR SCIENTIFIC PROJECTS:

Project: Partition specificity determinants in the pMT1 virulence plasmid of *Yersinia pestis* (2001-present). The genetic determinants that could be essential for species specificity of the pMT1 plasmid partition are identified in the *parS* centromere analog site. We conclude that the Box B sequences determine the specificity in at least three plasmid species (P1, P7 and pMT1), and the contact between these bases and ParB constitute a special mechanism for species determination. The specificity of the DNA site can be completely switched by changing as little as one base. Technology and methods used: high-throughput and routine cloning, *in vivo* and *in vitro* recombination technologies, transformation, λ (including λInCh shuttle system) and P1 transduction, restriction endonuclease digestion and ligation, single- and multi-site directed mutagenesis, genetic mapping, hybrid protein gene expression, fusion genes technology, DNA and

RNA purification, electroporation, gel-electrophoresis, gene construction analysis (Gene Construction Kit 2.5.6), PCR (PTC-100), DNA sequencing (Finch-Suite Distribution v.2.7.0), time-lapse fluorescent microscopy (Nikon eclipse E600 microscope), flow cytometry (Bryt SH, Bio-Rad), sequence alignment (NCBI-BLAST, BLAST Network Service on ExPASy, PILEUP from the Wisconsin package of program).

Project: Biogenesis of pyoverdine in *Pseudomonas putida* (1998 – 2001). PI. Pyoverdine, the green-yellow fluorescent pigment of *Pseudomonas* bacteria, is a product of oxidative deamination of L-phenylalanine. L-Phe plays the role of an aromatic precursor in the quinoline chromofore synthesis. Technology and methods used: protein purification, aromatic pathway intermediates and biochemical precursors organic synthesis, gene analysis, chemical and transposone mutagenesis, enzyme analysis, HPLC column and thin layer chromatography, [^{32}P] binding to L-Phe analysis, lyophilization.

Project: Regulation of aromatic amino acid biosynthesis in the obligate methylotrophic bacteria *Methylobacillus mucogenes* (1992-1998). Control of shikimate pathway in the *M. mucogenes* occurs via allosteric regulation all of thee DAHPS-isozymes by L-Phe, L-Tyr and L-Trp . L-tryptophan biosynthesis is controlled through trpE-, trp-D and trpC-genes repression. L-phenylalanine and L-tyrosine biosynthesis regulations occur by CM-repression and PDT-retroinhibition. Technology and methods used: protein purification, organic synthesis of aromatic intermediates and biochemical precursors, gene analysis, chemical and transposone mutagenesis, enzyme analysis, ultrasound desintegration, gel-electrophoresis, HPLC column and thin layer chromatography, GLC-chromatography.

FUTURE PLANS:

Investigate the molecular mechanism of the species determination within the P1*par* family of plasmid partition elements. Particularly, determine the molecular mechanism of DNA-protein and protein-protein interaction in *par* system of the pMT1 large virulence plasmid of *Yersinia pestis* that could be essential for deep tissue invasion of this strong human pathogen.

The main goal of the project: To identify the critical information (discriminator recognition sequence) within the C-terminal domain of the ParB protein in the pMT1 plasmid of *Y.pestis*;

Expected results:

- Genetic mapping of the pMT1*par* region.
- To localize the discriminator sequences within the pMT1 parB gene.
- To determine the target contact between ParB protein and the *parS* site.
- To identify the ParA-ParB protein-protein interaction determinants.
- To define the special mechanism of specificity in the pMT1 virulence plasmid of *Y. pestis*.

COMPUTER SKILLS:

Mac OSX, Windows OS (Microsoft Office X), Internet database search, Free Hands X, Power Point, Adobe Photoshop 7, Adobe Acrobat 6.0, Gene Construction Kit 2.5.6 (Textco, Inc.), Canvas 9.0, OpenLab software (Improvision, Lexington, MA), WinBrite 2.03 software program (Bio-Rad, Hercules, CA), Network Service on EXPASy (ExPASy Proteomics tools, Switzerland), Finch-Suite Distribution v 2.7.0 (Geospiza Inc.)

PROFESSIONAL SKILLS:

High-throughput and routine cloning, *in vivo* and *in vitro* recombination technologies, transformation, λ and P1 transduction, single- and multiple site directed mutagenesis, chemical and transposone mutagenesis, genetic mapping, hybrid protein gene expression, DNA and RNA purification, electroporation, gel-electrophoresis, restriction endonuclease digestion and ligation, gene construction analysis (Gene Construction Kit 2.5.6), PCR (PTC-100), DNA sequencing (Finch-Suite Distribution v 2.7.0), time-lapse fluorescent microscopy (Nikon eclipse E600 microscope), flow cytometry (Bryt SH, Bio-Rad), sequence alignment (NCBI-BLAST, BLAST Network Service on ExPASy, PILEUP, protein purification, enzyme analysis, ultrasound desintegration, HPLC column and thin layer chromatography, GLC-chromatography, lyophilization.

INVITED TALKS:

A mechanism of protein-DNA recognition in plasmid segregation. Scientific seminar at Dept. of Molecular Genetics and Microbiology, University of Massachusetts School of Medicine. Worcester, January 28, 2005.

A mechanism of protein-DNA interaction in plasmid partition. Scientific seminar in Functional genomics and Molecular medicine Research Group, Dept. of Physiology, University of Maryland Medical School, Baltimore, December 10, 2004.

A novel mechanism for changing the specificity of protein-DNA recognition during evolution. NCI-Frederick Interdisciplinary Retreat. Rocky Gap, October 25-27, 2004.

MEETINGS AND CONFERENCES:

Dabrazhynetskaya A. A novel mechanism for changing the specificity of protein-DNA recognition during evolution. NCI-Frederick Interdisciplinary Retreat. *Poster presentation.* Rocky Gap, October 25-27, 2004.

Dabrazhynetskaya, A. Partition specificity determinants in the pMT1 virulence plasmid of *Yersinia pestis. Poster presentation.* NCI-Frederick Spring Research Festival. Frederick, May 14-15, 2004.

Dabrazhynetskaya A. and Austin, S. Species specificity of the ParA, B type DNA partition proteins. *Poster presentation.* Keystone Symposia. Bacterial Chromosomes. Santa Fe, New Mexico, February 7-14, 2004.

Dabrazhynetskaya, A. Specificity determinants of the pMT1 plasmid centromere analog. 2003. NCI-Frederick Interdisciplinary Retreat. ***Oral*** *and poster presentations.* Rocky Gap, October, 2003.

Dabrazhynetskaya, A. 103rd General Meeting. American Society For Microbiology. Washington, DC, May 18-22, 2003.

Dabrazhynetskaya, A. The role of the centromere analog site of the pMT1 virulence plasmid. *Poster presentation.* 2003 Gordon Research Conference on Chromosome Dynamics. Tilton School, New Hampshire, August 17-23, 2003.

Dabrazhynetskaya, A. Putting plasmid partition genes under the control of the *E.coli* chromosome. *Poster presentations.* 2002 NCI-Frederick Interdisciplinary Retreat. Rocky Gap, October, 2002.

HONORS, AWARDS and GRANTS:

Fellowship from **National Institute of Health** in General Genetics, 2001-2006.
Grant from **National Academy of Science**, Belarus, Minsk. 2000-2001 (**PI**).

PATENTS ISSUED:

Maksimova N.P., **Dobrozhinetskaya E.V.,** Fomichev YuK. (1991). Bacterial strain *Methylobacillus sp. M75* used as test-culture for methanol and methylamine identification. **Patent of Belarus**. #1973. BY C1. C 12N 1/20, C12Q 1/06.

TEACHING EXPERIENCE:

Course: **Molecular basis of replication, reparation and recombination** (lectures and laboratories for undergraduate students), BSU, Department of Genetics and Biotechnology, 2000-2001.

Course: **Molecular genetics of bacteria** (lectures and laboratories for undergraduate students), BSU, Department of Genetics and Biotechnology, 1999-2001.

Course: **Selection of industrial strains** (lectures and laboratories for undergraduate students), BSU, Department of Genetics and Biotechnology, 1998-2001.

ACADEMIC ADVISORY FOR:

Kuleshova, V. - Diploma theses: "The role of phenylacetamide in the biogenesis of the quinoline chromophore of *Pseudomonas putida.* Department of Genetics and Biotechnology, BSU, Minsk, Belarus, 2001.

Dobrosotskih, S. – Diploma theses: " Pioverdine biogenesis in the *Pseudomonas putida*: L-phenylalanine is an aromatic precursor of quinoline chromofore synthesis". Department of Genetics and Biotechnology, BSU, Minsk, Belarus, 2000.

Ivanovskaya, T. – Diploma theses: "Regulation of tryptophan biosynthesis in the obligate methylotrophic bacteria". Department of Genetics and Biotechnology, BSU, Minsk, Belarus, 1999.

PROFESSIONAL SERVICE:

Proposal Reviewer: National Academy of Science Foundation, Industrial Biotechnology Program. 2000-2001.

TRAINING, COURSES AND LECTURES:

Specialized NCBI Classes "Gene Expression Resources at the NCBI". Ft. Detrick, Frederick, March 4, 2005.

Training in real time PCR (Bio-Rad, GRCBL, November, 2004)

Specialized NCBI Classes "NCBI'S BLAST Quick Start". Ft. Detrick, Frederick, August 10, 2004.

Training in flow cytometry (Bryt SH, Bio-Rad, GRCBL, March, 2004)

Training in time-lapse fluorescent microscopy (Nikon eclipse E600 microscope, GRCBL, November, 2002)

AFFILIATION:

Member of Belorussian Society for Genetics (1999-2001).
Member of Belorussian Society for Microbiology (1996-1999).