

Treatment of tuberculosis (TB) takes at least six months for drug sensitive cases and up to several years for cases caused by drug (antibiotic) resistant *Mycobacterium tuberculosis* bacterium. Globally, and in Eastern Europe as one of the hot spots, drug resistant TB is responsible for high mortality and high healthcare costs. Monitoring of treatment progress is essential for successful treatment outcome but methods for monitoring are limited at present.

The purpose and aims

The purpose of this project has been to strengthen TB management through establishing international research collaboration between countries with different TB burden. We had following specific aims:

- i) To explore metabolites that can serve as biomarkers for monitoring of TB treatment

Approach: small organic molecules from blood – blood metabolites - are promising biomarkers for treatment monitoring of TB. Broad spectrum of metabolites has been analyzed simultaneously by metabolomics. This technique uses chromatography and mass spectrometry together with multivariate analysis and other statistical methods to provide comparative analysis of metabolites in patient samples during treatment

- ii) To identify *M. tuberculosis* strains currently circulating in Ukraine and their drug resistance profiles

Approach: *M. tuberculosis* drug resistance and transmission of different strains in southern Ukraine was studied using genomics whereby new generation sequencing was used to identify strains and mutations of drug resistance. Results of genomics were analyzed together with other conventional methods to be able to follow the trends for drug resistance

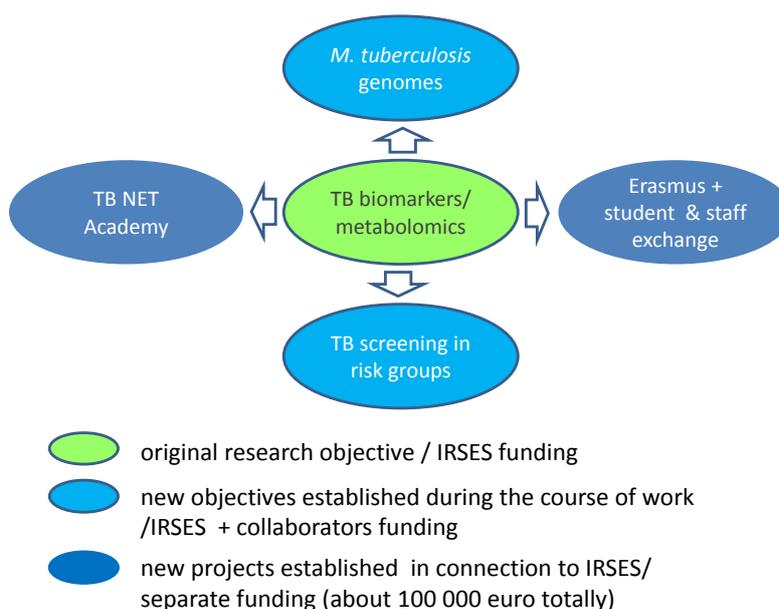
Results

Over ten young researchers and over ten experienced researchers/clinicians were exchanged between Sweden, Spain and Ukraine and trained with focus on diagnostic methods and omics research. Several international workshops were organized. This became a base for a unique consortium that operates now at the cross-section of TB epidemiology, microbiology and analytical chemistry.

65 TB patients and 15 healthy subjects from Ukraine volunteered to participate in the metabolomics study. Our data

showed that metabolome of these TB patients differed from the one of healthy volunteers. We also found significant differences in the metabolome of patients with poor response to treatment comparing to those with good treatment progress. This finding may open the possibility of the development of a laboratory test for treatment monitoring. We plan to validate these results in a bigger study.

In cooperation with colleagues from Germany, we performed genomic DNA sequencing of 186 Ukrainian clinical strains of *M. tuberculosis*. Analysis showed that a particular strain of highly drug-resistant *M. tuberculosis* (pre-XDR-TB) is transmitted within the Ukrainian population. The share of



Structure of IRSES TB-prognosis & connected projects established over reporting period

this strain is over 10% of all drug-resistant strains analyzed in our study. This finding challenges the existing standard treatment regimen in Ukraine and poses a question of alternative treatment strategies.

Additionally, we conducted a smaller questionnaire survey that showed that among the risk groups for TB only HIV-positive subjects are targeted by TB screening. This finding shows that better programs should be developed for targeting risk groups like internally displaced people, for example.

Social & economic impact

We believe our project has created a powerful social and economic impact especially in Ukraine. Ukraine is one of top 20 countries by number of drug-resistant TB. One year after our project had started this country was shaken by revolution, economic crisis and military conflict. The salaries of the state employees (researchers/clinicians) dropped drastically in value what pushed people to look for jobs outside of research/TB healthcare. In these difficult times, travel funding from Marie Curie Actions and exchanges to Spanish and Swedish institutions provided a certain encouragement and support for Ukrainian colleagues and kept them in the field of research.

We also linked our consortium to TB NET, a large European network for clinical TB research. By joint effort of TB NET and partner institutions of TB-prognosis, we organized in 2016 an Academy for Young TB Researchers in Odessa, Ukraine. German Government funded this initiative and 20 young researchers from across Europe and Ukraine came to Odessa for 4 days training in clinical TB and career coaching led by prominent European TB experts. In connection to this training, there was a press conference and “Cycling against TB” action. This was a team-building for TB researchers, an integration of Ukrainian colleagues in European networking against TB and an outreach to the public and officials in one of the major Ukrainian cities: <https://www.youtube.com/watch?v=a1FveD9aLqc&t=28s>

As a spin-off from the IRSES project we also developed Erasmus + collaboration in Life Science teaching with focus on clinical and molecular microbiology between Odessa National University and Umea University: <https://www.youtube.com/watch?v=zuoOHzhPT1s&t=19s>

Our scientific findings can improve TB management routines and increase treatment success, which is good for people and may lower the healthcare costs. Our networking and educational achievements help to single researchers and institutions to raise expertise and improve their work conditions.

Selected publications and presented posters/talks

- 1) Tackling the MDR-TB epidemic in Ukraine: every little helps ... and much more needed Burman M *et al* J Public Health (Oxf) 2017. doi: 10.1093/pubmed/fox014.
- 2) Multidrug-resistant tuberculosis in Ukraine at a time of military conflict; Dudnyk A *et al*; Int J Tuberc Lung Dis. 2015;492-3. doi: 10.5588/ijtld.14.0940.
- 3) The 26th ECCMID, 2016; Abstract, ID 4251; Spoligotypes of *Mycobacterium tuberculosis* isolated from tuberculosis patients in Odessa, Ukraine; Nikolaevskaya E, Pavlovska O, Molina B, Brännberg P, Marynova I, Krylova K, Ivanitsa T, Filuk V, Lacoma A, Dominguez J, Rzhepishevskaya O, C Prat C.
- 4) The Congress of the European Society for Translational Medicine, Prague, 2016 Poster. Evaluation of treatment progress in tuberculosis patients based on their serum metabolite profiles; Hedenström M, Prat C, Gerasimova N, N Dovga N, Sokol D, Limanska N, Nikolaevskaya E, Stokich V, Sjöstedt A, Dominguez J, Antti H, Rzhepishevskaya O
- 5) the 10th International Conference on the Pathogenesis of Mycobacterial Infections; August 2017, Stockholm, Sweden; Oral presentation; Genomic sequencing provides new ideas for tuberculosis management in Ukraine and identifies a pre-XDR outbreak; Merker M, Nikolayevskaya E, Kohl T, Molina B, Pavlovska O, Brännberg P, Dudnyk A, Stokich V, Marynova I, Philippova T, Prat C, Sjöstedt A, Dominguez J, Rzhepishevskaya O, Niemann S