

ing clustered by cgMLST analysis resulting in 19 (9.0%) clustered isolates by cgMLST. By clustering analysis with the distance  $\leq 12$  SNPs, 18 isolates clustered into 7 clusters. With the 1 SNP cut-off, three clusters with a total of seven strains were found and these were similarly clustered also by cgMLST and conventional genotyping analysis.

A reliable prediction of drug susceptibility can be obtained with WGS combined with data analysis with software tools. In routine practice, *M. tuberculosis* isolates can be screened with WGS for mutations associated with drug resistance, and only resistant strains confirmed with the MGIT system. Compared to conventional genotyping methods, WGS analysis is more discriminatory, reducing the risk of false clustering and unnecessary contact tracing.

6.49

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#### SINGLE NUCLEOTIDE POLYMORPHISMS IN *hsp65* AND MACPPE12 GENES OF MYCOBACTERIUM AVIUM subsp. HOMINISSUIS

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*Mycobacterium avium* subsp. *hominissuis* (MAH) represents a group of environmental bacteria known as opportunistic pathogens of animals and humans, especially HIV positive. Polymorphisms in *hsp65* and MACPPE12 genes are used for identification, intra-subspecies differentiation and phylogenetic studies of MAH populations.

The aim of our study was to identify single-nucleotide polymorphisms (SNPs) in *hsp65* and MACPPE12 genes of clinical isolates from Russian patients with pulmonary and disseminated mycobacteriosis and to assess phylogenetic relationships of geographically distant MAH populations.

The sequence analysis of the 3'-portion of the *hsp65* gene and MACPPE12 gene was applied for 40 MAH strains isolated from humans with mycobacteriosis (including 19 HIV-positive) in St. Petersburg, Russia (2008–2011). The nucleotide sequences were aligned to the reference genome of *M. avium* subsp. *hominissuis* 104 (NC\_008595.1).

In total, the 40 MAH strains were classified into three different *hsp65* sequevars: code 1, code 2 and code 3. The majority of MAH strains (72.5%) belonged to code 1, the same sequevar as for MAH strain 104. The code 2 and code 3 included 3 (7.5%) and 8 (20%) strains, respectively. The largest *hsp65* sequevar code 1 has observed only in 4.7% of isolates from Japan and absent in Korean human isolates. The sequevars code 1 and code 2 predominated among MAH strains in the USA, Canada, Belgium.

The sequence analysis of the MACPPE12 gene revealed 20 SNPs grouped into nine sequevars at the nucleic acid level: NA01, NA02, NA03, NA06, NA10, NA13, NA14, NA19, and NA\_Rus01. Among 20 SNPs eight were nonsynonymous resulting in seven sequevars at the amino acid level: AA01, AA02, AA04, AA07, AA08, AA13, and AA\_Rus01. The sequevar AA02 consisted of three different NA variants with synonymous SNPs profiles: NA02, NA03, and NA06. Half of the MAH strains belonged to the sequevar AA02 (type NA02). The predominant cluster AA02 (type NA02)/code 1 and the unique variant AA\_Rus01 (NA\_Rus01) were identified among MAH strains from Russia. The present study demonstrated the prevalence of the sequevar AA02 in MAH strains isolated from humans in Russia, Japan, and Korea.

Thus, we confirmed the relative conservativeness of the nucleotide sequence of the *hsp65* gene but the polymorphism of the MACPPE12 gene. A comparative analysis of the SNPs profiles of the *hsp65* and MACPPE12 genes allowed to identify differences and similarities between geographically distant populations of MAH, which highlighted the variability of the global population of *M. avium* species.

6.50

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#### MOLECULAR EPIDEMIOLOGY OF TUBERCULOSIS IN ALBANIA (2006–2011)

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Tuberculosis (TB) epidemics in Albania has been stable over the past years with a gradual decreasing incidence (from 18.7 to 14.8 per 100 000 inhabitants, in the period 2001–2016) with a slight deterioration in 2013 (16.8 per 100 000 inhabitants). First insight data (2008) on TB molecular epidemiology showed a moderate Recent Transmission Index (RTI) (28%) and a high level of genetic diversity.

We aimed with this study to better understand the correlation of ubiquitous and autochthonous *Mycobacterium tuberculosis* complex (MTBC) genotypes with available demographic and epidemiologic data over a six-year period, in Albania.

MTB strains isolated in Albania (n = 745, 1 isolate per patient) between 2006 and 2011 were analyzed by spoligotyping and MIRU-VNTR typing by 24 loci scheme. The data obtained were compared with MIRU-VNTRplus database. Using molecular typing 486 (65.23%) isolates (patients) were distributed into 113 clusters and the remaining 259 (34.77%) isolates had a unique pattern. The cluster sizes ranged from 2 to 21 isolates per cluster. RTI ((nc – c)/n) resulted 50.07%. The most predominant lineages were Ghana (28.59%), Haarlem (19.73%) UgandaI (18.79%), LAM (7.11%), Ural (5.64%), TUR (3.89%) and Caprae (3.49%). Other lineages identified were Cameroon (1.74%), X (1.48%), S (1.21%), Bovis (0.54%), Delhi/CAS (0.54%), Beijing (0.4%) and West African 2 (0.13%). This study highlighted the predominance of five shared spoligotypes: ST 53 (T1) (n = 166, 22.28%), ST 4 (LAM3 and S/convergent) (n = 39, 5.23%) and ST 42 (LAM9) (n = 38, 5.10%) ST 613(T1) (n = 37, 4.97%) and ST 47 (H1) (n = 35, 4.70%). Of the unknown spoligotype signatures three were more frequent than others (4.70%, 3.22%, 3.22%), their origin and historical link to other genotypes is yet unknown. Among the MLVA MtbC15-9 types, MLVA 15411-85 and MLVA 15419-69 (both of unknown spoligotype signatures) resulted the predominant types involved in recent transmission in Albania (two biggest clusters identified with 21 and 19 identical isolates respectively).

In conclusion, MTBC genetic population in Albania is highly heterogenous. TB epidemics in Albania is fueled mostly by evolutionary-recent lineages. It is largely dedicated to recent transmission (50.07%). Autochthonous genotypes result linked to the 2 biggest clusters identified. One of them is found exclusively in Tirana (MLVA 15411–85). The new MTBC genotypes will require further molecular characterization.