

Philipe Anochie

Dr. Anochie is a Microbiologist/ Research fellow at the National Tuberculosis Reference Laboratory of the Nigerian Institute of Medical Research. His research interest is on the study of genomics, pathogenesis and drug resistance in Mycobacterium tuberculosis isolates. Dr. Anochie is the first recipient of the ISID/ESCMID Fellowship.

The ISID and the European Society of Clinical Microbiology and Infectious Diseases (ESCMID) are co-sponsoring the ISID/ESCMID Fellowship to enable young researchers from developing countires to come to Europe for training. More info can be found at www. isid.org/grants/grants.shtml

ISID/ESCMID Fellowship Report

Philipe Anochie, Nigeria

The ability to discern the molecular "fingerprint" (genotype) of *Mycobacterium tuberculosis* isolates has revolutionized the understanding of the transmission of tuberculosis. Increasing levels of drug resistance are threatening to erode the medical advances of recent decades. The social causes contributing to the spread of anti-tuberculosis resistance are somehow paradoxical. In some settings – especially in resource limited countries – the under-use of drugs or the use of low quality drugs encourages the development of resistance. In wealthy countries, resistance is the opposite reason – the over use of drugs, that is driving the emergence of resistance. Regardless of where drug resistance originates, globalization, increased travel and trade ensure that these strains quickly travel elsewhere. The DNA fingerprinting technology (genotyping) is useful to track down the worldwide spreading of resistant strains.

Health authorities in many countries are confronted with a serious challenge of *Mycobacterium tuberculosis* that are resistant to anti-mycobacterial drugs. Dramatic outbreaks of multi-drug resistant tuberculosis (TB) among institutionalized HIV-infected patients have focused international attention on this problem. There is therefore an urgent need to thus generate reliable and internationally comparable data on TB drug resistance in these countries. This comparable data will not be obtained without the genotypic analysis of the *Mycobacterium tuberculosis* isolates, which will ensure the elimination of nosocomial transmission and laboratory error.

Mycobacterium tuberculosis genotyping is essential to investigate and confirm transmission, and to confirm and exclude laboratory contamination by genotyping isolates for every new TB case to improve the efficiency of tuberculosis control, decrease the length of unnecessary treatment among patients with false-positive cultures, identify previously unknown links among genotypically clustered patients and unidentified clusters of transmission. Genotyping will also more rapidly and efficiently determine the extent and dynamics of ongoing transmission to focus programme interventions for specific areas and populations, assess tuberculosis transmission in outbreaks to refine contact investigations, identify nosocomial transmission not identified by conventional methods and also identify false-positive cultures so that clinicians could be notified on diagnostic errors quickly and prevent unnecessary tuberculosis treatment.

With an ISID/ESCMID Fellowship, Dr. Anochie Philip recently concluded a study on the genotype analysis of *Mycobacterium tuberculosis* isolates at the Emerging Bacteria Pathogens Laboratory of the San Raffaele Scientific Institute, Vita – salute San Raffaele University, Milan, Italy under Dr. Daniela Maria Cirillo, the Head of the laboratory. The laboratory focuses its activities on the study of major problems related to the issue of drug resistance in *Mycobacterium tuberculosis* strains: the biology and resistance mechanisms of multi drug resistance (MDR). The laboratory for tuberculosis control for the international activities performed in this field. Dr. Cirillo is the coordinator of two EU funded projects (2008-2013): "TB PAN – Net – Pan-European network for the study and clinical management of drug resistant tuberculosis", and "TM – Rest – A new platform for fast molecular detection of multi drug resistant (MDR) and extensive drug resistant (XDR) strains of *Mycobacterium tuberculosis*."

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During this time, Dr. Anochie studied the principal and applications of molecular techniques for tuberculosis diagnosis and genotyped more than 300 *Mycobacterium tuberculosis* isolates sing the following *Mycobacterium tuberculosis* genotyping methods:

- Geno Quick[®]MTB: Rapid molecular genetic assay for the direct detection of the *Mycobacterium tuberculosis* complex from patient specimens.
- Geno *Quick*[®]MTBC: Molecular genetic assay for the differentiation of the *Mycobacterium tuberculosis* complex from cultured material.
- Geno *Quick[®] Mycobacterium* CM: Molecular genetic assay for the identification of the clinically most relevant Mycobacterial species from cultured material.
- Geno *Quick*[®]MTBDR *plus*: Molecular genetic assay for the identification of resistance to rifampicin and/or isoniazid of the *Mycobacterium tuberculosis* complex.
- Geno *Quick®*MTBDR*sl*: Molecular genetic assay for the identification of resistances to floroquinolones aminoglycosides/cyclic peptides, and ethambutol of the *Mycobacterium tuberculosis* complex.
- Spoligotyping: A PCR-based method to simultaneously detect and type *Mycobacterium tuberculosis* complex bacteria for epidemiological studies.
- BACTECTM MGITTM 960 SIRE test for the antimycobacterial susceptibility testing of *Mycobacterium tuberculosis* for first and second line drugs.
- Capilia TB-NEO for the detection of *Mycobacterium tuberculosis* complex from patient specimens.
- PCR and reverse hybridization assays.
- DNA inactivation and extraction from positive MGIT tubes.
- DNA inactivation and extraction from positive LJ tubes.
- DNA inactivation and extraction from sediments.
- Chemical DNA extraction.
- MIRU-VNTR_{plus} Mycobacterium interspersed repetitive units variable number tandem repeats; a strain identification and differentiation protocol for the epidemiological study of *Mycobacterium tuberculosis*.
- GT-BLOT 20 automated equipment for reverse hybridization assays.

These tuberculosis genotyping techniques will be applied in local tuberculosis research and training programmes in Nigeria.

This three months' training provided me the opportunity to learn not only the tuberculosis genotyping techniques but also laboratory organization and administration techniques. I am now a new man scientifically. I thank ISID/ESCMID for giving me this fantastic and unique opportunity as well as Dr. Cirillo and all the Emerging Bacterial Pathogens laboratory staff for their kindness and support.