

## ECFS Diagnostic Network Working Group

12<sup>th</sup> Annual Meeting, Warsaw, 12-14<sup>th</sup> February 2015

### Meeting report

#### Welcome and opening

The meeting started on Thursday, 12th February with a welcome from Dr. Maciejewski, head of the Institute of Mother and Child in Warsaw. Afterwards, Dorota Sands, president of Polish Cystic Fibrosis Society, gave an overview of CF in Poland. Then, Nico Derichs, DNWG coordinator, welcomed everyone and the meeting was opened.

#### CFTR 2015 (Chair: Kris de Boeck, Leuven)

The first session started with a presentation from Piotr Zielenkiewicz (Warsaw, Poland) about "CFTR models". Afterwards, Aleksander Edelman (Paris, France) gave an overview on "CFTR modulators - state of the art", concluding that combinations of correctors and potentiators will be necessary to treat patients with f508del mutation, that chronic CFTR modulator treatment might have unwanted effects, and that a new class of correctors might prevent pathologic interactions in the cell. Then, the 1st *Young Investigator* Robert Rauscher (Potsdam, Germany) presented his work on "Global Adaption to CFTR misfolding in Cystic Fibrosis Bronchial Epithelial cells". This talk was followed by Zoya Ignatova (Potsdam/Hamburg, Germany) reporting about her work on "Diagnostic implications of "silent" CFTR mutations", stating that silent mutation still have an impact on the processing of proteins and their function. The session was closed by Michael Wilschanski (Jerusalem, Israel) speaking about Primary Sclerosing Cholangitis (PSC) as a possible CFTR-related disorder. He introduced a current study investigating patients with PSC concerning CFTR function and genetics.

#### "Graduating from CFSPID: CF Newborn Screening challenges" (Chair: Kevin Southern, Liverpool and Dorota Sands, Warsaw)

The 2<sup>nd</sup> day was opened by Kevin Southern giving an introduction and overview of Cystic Fibrosis Screen Positive Inconclusive Diagnosis (CFSPID) evaluation and management. He explained the development of the term CFSPID and the consequences for these infants. Afterwards, Mariusz Ołtarzewski (Warsaw, Poland) reported about the different protocols of NBS in Poland. He stated that the current protocol consisting of IRT, DNA and extended genetic analysis has a positive predictive value of 25% and that 19% of the identified mutations are not classified in CFTR2. He concluded that Poland needs to reduce the number of false positive results, but still genetic sequencing currently seems to be the suitable method for the Polish heterogeneity. Then, Caroline Raynal (Montpellier, France), spoke about CFTR mutation interpretation challenges in CF NBS. She gave examples of a broad spectrum of mutations and pointed out the high amount of variants with unknown clinical consequence in France that causes difficulties in interpretation. Afterwards, the 2nd *Young Investigator*, Joanna Jaworska (Warsaw, Poland) presented her work on "Bilateral sweat test results after CF NBS in Poland". In the presented study, sweat test results in newborns obtained with 2 different methods - conductometry and chloride titration - were compared, with a high agreement between the two methods. Additionally, new cut-off values for sweat test after positive NBS were suggested. Afterwards, the 3rd *Young Investigator*, Justyna Milczewska (Warsaw, Poland), talked about "Genotypes of infants with inconclusive diagnosis after Polish CF NBS". She pointed out that the mutations D1152H and F1052V, classified as mutations with varying clinical consequence in CFTR2, as well as IVS8-5T+TG(12), causing CF or CFTR-related disorder according to CFTR2, occurred frequently in this group. She concluded that extended genetic analysis in CF NBS leads to the detection of more uncertain cases with unclear CFTR genotypes. The next presentation was given by Milan Macek (Prague, Czech Republic) on "Challenges associated with the utilisation of next generation sequencing in CF diagnostics: how to assign disease liability of variants detected within the CFTR gene, and beyond". He underlined that this new technique need to be used responsibly and

that indicating physicians should be aware of blurred boundary between research and diagnostics. After a short break, Jürg Barben (St. Gallen, Switzerland) talked about "Sweat testing in the era of newborn screening". He presented an overview of results and failure rates with 2 different methods in Switzerland. In the end he concluded that conductivity testing is a suitable tool for sweat testing after newborn screening, although in Switzerland they continue to do both methods. Jarosław Walkowiak (Poznan, Poland) referred about "Fecal elastase as a noninvasive marker of pancreatic function". He underlined that fecal elastase is a good marker to longitudinally follow up on pancreatic function in CF patients, because the pancreatic status cannot be predicted by the genotype. Afterwards, Waldemar Tomalak (Rabka-Zdroj, Poland) introduced "Impulse oscillometry in the evaluation of CF". The method is used as a measure of changes in the small airways and is especially useful in children, since only passive cooperation is needed. He showed also a strong correlation to spirometry. However, the method is still not very popular and lacks of reliable reference values. The session was ended by Anne Munck (Paris, France) explaining the "Rationale for R117H being removed from the CFTR mutation panel in France". According to the results to date, the removal of R117H would increase the positive predictive value of the NBS to 27%. However, the mutation will remain in the 2nd test panel, so that symptomatic patients with 1 other mutation will still be diagnosed.

### **"Standardising CFTR Biomarker for CF Diagnosis and Clinical Outcome" (Chair: Nico Derichs, Berlin)**

The next session was opened by Natalia Cirilli (Ancona, Italy), who introduced a new DNWG project: "Development of an ECFS sweat test guideline: Overview of current practice in Europe". She presented the research plan, and in the end, many attendants signaled their willingness to contribute to the project. She was followed by Francois Vermeulen (Leuven, Belgium), speaking about "Optimizing sweat test frequency in clinical trials". He pointed out that the variability of sweat test had not been assessed formerly, although as biomarker of CFTR function endpoint in clinical trials variability is a major factor in power calculations and design of clinical trials. Data from the placebo arm of the phase III trial of VX-770 in subjects with a G551D mutation were analyzed. He concluded that sweat test has a high success rate, that sweat chloride has substantial variability and that the design of a trial could be optimized by 4, perhaps 8 cross-over and repeated measurements. Then, Isabelle Sermet-Gaudelus (Paris, France) presented the current state of the ECFS NPD SOP multicenter validation study. 15 centers have already been certified for NPD by the ECFS CTN. Results from the certification process were shown, and the attendants were encouraged to send in NPD tracings fulfilling the described standardization criteria for the ongoing NPD validation study in order to establish reference values for the ECFS NPD SOP. Then the 4th *Young Investigator*, Denise Peserico (Verona, Italy) presented her work on "Developing a multicenter protocol for ratiometric evaluation of B adrenergic/cholinergic sweating in human sweat glands". She explained the method which is comparing the CFTR-dependent adrenergic stimulated sweat secretion with the non-CFTR dependent cholinergic sweat secretion and described the implementation of the method in Verona. She concluded that the test is reproducible and feasible and might be used to evaluate CFTR modulator therapy effects or support diagnosis in controversial cases. Afterwards, the 5th *Young Investigator* Jan Nowak (Poznan, Poland) reported on "Optical coherence tomography of the labial salivary glands in cystic fibrosis". He concluded that a lower surface density of lower lip's LSGs was a previously unrecognized CF-related pathology. However, the difference was not significant, which makes the OCT of the LSG unsuitable as a diagnostic test. The last talk of the day was given by Malena Cohen-Cyberknoh (Jerusalem, Israel) about "CFTR dysfunction in Primary Ciliary Dyskinesia?" CFTR function was analyzed by sweat test and NPD in PCD patients. She concluded that PCD patients have elevated sweat chloride tests, suggesting reduced CFTR activity in a non-respiratory organ, and that therefore, it is important to perform NPD in patients with respiratory symptoms and abnormal or borderline sweat test in order to differentiate between PCD and CF and to confirm the diagnosis with genetic analysis. She suggested to add PCD to the list of diseases causing false-positive sweat tests. Finally, she emphasized to further investigate decreased CFTR function in PCD patients since CFTR modulators might be beneficial to these patients.

## **"Journey from Genotype to Phenotype (and retour)" (Chair: Michael Wilschanski, Jerusalem)**

Carlo Castellani (Verona, Italy) started on the last day with his talk on "Present developments in CFTR2, compound heterozygous newborns and sweat test". He introduced the upgrading of the CFTR2 dataset, explained the CFTR2 website redesign and asked for feedback on developments of the website and terminology. Afterwards, Elke De Wachter (Brussels, Belgium) spoke about "R75Q = Non CF-causing mutation?" She introduced a case of a patient with clinical CF features, normal sweat test, an abnormal NPD and R75Q mutation in trans with a CF-causing mutation, raising the question whether R75Q might be a CF-causing mutation. Afterwards, Sheila Scheinert (Berlin, Germany) gave an update on the DNWG project "CFTR3: Personalised characterization of rare CFTR mutations". CFTR3 is supposed to be a multicenter project characterizing non-CFTR2 mutations clinically and functionally in an open-access database. The session was completed by a Pro-Con debate on the topic "Patients taking CFTR modulators do not have CF anymore". The Pro side was presented by Hugo de Jonge (Rotterdam, Netherlands). He argued that patients with CRMS, CFTR-RD or CFSPID do not have CF, although they have a lowered level of CFTR function. CFTR modulators could increase CFTR function to a level of CFTR-RD and therefore the patients would no longer have CF. He underlined that at 10% of CFTR function the threshold for the transition from CF to non-CF would be reached, which could also be achieved by CFTR modulator therapies. Finally, he admitted that further clinical and pre-clinical research is needed to finally proof that CFTR modulators like Ivacaftor can "cure" CF. The con side was represented by Kris De Boeck (Leuven, Belgium). She argued that e.g. CF patients under CFTR modulator therapy still need all their regular medication and therefore are not cured from CF.

Finally, DNWG coordinator Nico Derichs (Berlin, Germany) gave an overview of the meeting and current as well as future DNWG projects. He thanked the hosts, and closed the meeting.

We thank all the speakers and participants for their contribution to this interesting and constructive meeting. We are looking forward to the next ECFS Diagnostic Network Working Group Meeting 2016 in London!

24 March 2015

Sheila Scheinert – DNWG Assistant  
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