Statistical analysis

A linear mixed model was used to evaluate the evolution of FEV1 %, quantify the yearly rate of change and verify if the FEV1 progression depended on baseline characteristics by including interaction terms in the model. Nonlinear evolution of FEV1 as a function of age was allowed in the model using restricted cubic splines. The approach is similar to the model proposed by Szczesniak et al, but they did not include slope as a random effect. (1)

Restricted cubic splines with five knots (at percentiles 5, 27.5, 50, 72.5, and 95) were used (2) to allow nonlinearity in the relation between age and FEV1 (%). This approach allows capturing in a flexible way deviation from linearity dividing age into intervals (defined by the knots) using only a limited number of extra parameters (using 5 knots implies 3 extra parameters).

The yearly change in FEV1 (%) - due the nonlinear relation not being constant – was calculated as the derivative of the function used to model age. The delta rule was used to calculate a 95% confidence interval (CI) for the yearly rate of change. Note that this approach has the advantage that no strong assumptions were made on the relation between age and the rate of change. For example, a regression model with only linear slopes assumes a constant rate, a regression model adding a quadratic term assumes that the rate of change evolves linearly over time(1).

It was evaluated if each of these variables (gender, socio-economic status of country, age at diagnosis, neonatal screening, sweat test value, meconium Ileus and presence mutation) moderated the rate of FEV1(%) change. To this purpose, each variable was added in the model allowing an interaction with age. Not only the parameter for the linear slope, but also the parameters for the splines were included in the interaction. Note that allowing an interaction between a variable and age in the model for the mean FEV1(%) is equivalent to allowing a main effect of this variable on the rate of change. To have a uniform approach in analysis and reporting of results, the continuous moderators age at diagnosis and sweat test value have been categorized. For age at diagnosis: <1year, 1-10yrs, 11-18 yrs and >18yrs. For sweat test value, the patients were divided into four groups based on the quantiles (<Q1, Q1-median, median-Q3 and >Q3).

Results were reported from models for each moderator separately ('univariable model') and from a multivariable model containing all specified potential moderators. Models were based on all available cases. Differences between subjects with and without complete moderator information were evaluated. Given the large amount of missing values for sweat test value, a multivariable model without sweat test value as moderator was additionally considered.

Extending the models with a random country effect was computationally not feasible. A simplified analysis however, without random slopes, revealed that the random effect of country was negligible (intra-class correlation (ICC)<0.02). In models for each moderator separately, country was therefore included as a fixed effect. However, since combining country economic status with country as fixed effects yields an over-parameterized model, country was not included as effect in models containing country status as moderator.

Predicted mean evolution and mean rate of change

Plots with predicted mean profiles and predicted mean rate of change were given for the 6-50 age. For the mean evolution of FEV1(%) a distinction was made in how each country was weighted in the calculation of the mean: each country having the same weight or each country having a weight equal to the number of included subjects. Note that the derivative of the mean evolution does not depend on this choice, hence a difference in the country weights does not impact the mean rate of change.

Software

All analyses have been performed using SAS software, version 9.4 of the SAS System for Windows. Copyright © 2002 SAS Institute Inc. SAS and all other SAS Institute Inc. product or service names are registered trademarks or trademarks of SAS Institute Inc., Cary, NC, USA.