The molecular structure and the folding of the whole Cystic Fibrosis Transmembrane Conductance Regulator (CFTR): corrector sites

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Background and rationale. CFTR is an anionic channel expressed in epithelia whose mutations cause cystic fibrosis, but its structure is still unknown.

Hypothesis and objectives. We explored a set of structural parameters of the wild type and mutated (F508del) CFTR by physical methods.

Essential methods. WT and F508del CFTR were over-expressed in yeast, solubilised in the detergent LPG-14 and purified. The detergent-CFTR complexes were studied by SAXS techniques using a solvent of variable density.

Results. The final result of the study is the numerical value of a set of parameters: molecular mass, volume and radius of gyration, average electron density and second moment of the electron density fluctuations inside the particles. It is also shown that in the complex the centres of gravity of CFTR and of the detergent are displaced relative to each other. The analysis of these parameters led to the determination of the size and shape of the volumes occupied by protein and by detergent in the complex. WT-CFTR to be an elongated molecule (maximum diameter ~12.4nm) which spans a flat detergent micelle. The distance distribution function, P(r), confirms that the WT-CFTR is elongated and with an inhomogeneous electronic density. The F508del-CFTR molecule is also elongated (maximum diameter ~13.2nm), but the associated detergent micelle hides a larger surface, plausibly related to an increased exposure of hydrophobic portions of the mutated protein. The corresponding P(r) is consistent with the presence of well defined domains, probably linked by flexible regions.

Significance.These differences suggest that the full-length mutant F508del-CFTR has a detectably different conformation, in contrast to the minor differences observed for the isolated F508-containing domain. We interpret the data in terms of an incomplete post-translational assembly of the protein domains. It follows that the use of WT molecular models to find better corrector must be reconsidered.

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