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Letters to the Editor

Towards "Molecular Esperanto" or the Tower of Babel? (The Need for Harmonization of Techniques for Genotyping Clinical Isolates of *Pseudomonas aeruginosa* Isolated from Patients with Cystic Fibrosis)

We read with interest the letter of Armstrong et al. (D. Armstrong, S. Bell, M. Robinson, P. Bye, B. Rose, C. Harbour, C. Lee, H. Service, M. Nissen, M. Syrmis, and C. Wainwright, Letter, J. Clin. Microbiol. **41:**2266-2267, 2003) describing the occurrence of a clonal type (pulsotype I) of *Pseudomonas aeruginosa* at five cystic fibrosis (CF) centers along the east coast of Australia. This letter strongly asserts that this clonal type has spread to CF patients, not from either the hospital or external environments, but by person-to-person transmission. This letter also describes some of the recent events that have led to advances in molecular typing techniques and thus to the identification of a dominant and hypertransmissible clonal strain of *P. aeruginosa* at several CF centers. What this letter also demonstrates to the CF microbiology community is that there is an urgent requirement for the standardization and harmonization of genotyping techniques, both nationally and internationally, where several methods presently exist for strain characterization (for a detailed review of *P. aeruginosa* typing, see reference 15).

Armstrong et al. (letter) cite several previous studies employing pulsed-field gel electrophoresis (PFGE) typing of *P. aeruginosa*. A methodological comparison highlighting technical variations in previous PFGE typing studies of *P. aeruginosa* is detailed in Table 1. Thus, Armstrong et al. were correct and diligent in comparing interlaboratory variability in deciding the relatedness of isolates from five CF centers, when more than one microbiology laboratory was involved in strain analysis.

Recently Jones et al. (9) have stressed the importance of microbiological surveillance through molecular fingerprinting, namely, PFGE typing, in order to ascertain the extent of cross-infection of *P. aeruginosa* among CF patients. PFGE is no longer a method solely restricted to epidemiological typing research studies, but is one which is now used in many clinical diagnostic laboratories to aid with routine epidemiological analysis. Laboratories which are introducing such techniques for the analysis of their *P. aeruginosa* subtypes should adopt protocols that will yield data for their isolates which can be compared with data from other CF centers. This strategy has been successfully applied for typing enteric bacterial pathogens through the PulseNet system (www.cdc.gov/pulsenet). The exchange of typing data obtained by employing standardized and harmonized methods, via an electronic medium, as has been developed recently by the ESF Network for Exchange of Microbial Typing Information (http://lists.nottingham.ac.uk/mailman/listinfo/enemti), would facilitate our understanding of the molecular epidemiology of *P. aeruginosa* in CF patient populations.

Consensus is thus needed in order to ascertain if such differences, for example, different suppliers of restriction enzymes, internal standards, pulse field parameters, etc., are significant or not. If the effects of such differences are not known, then a feasibility study involving several centers needs to be undertaken to resolve these technical anomalies, so that a robust method may be adopted by the CF microbiology community. If it felt that such a trial is unnecessary, then it will be a relatively easy task to define a consensus PFGE protocol that will gain widespread acceptance.

The real treasure to the CF microbiology community of adopting standardized and harmonized genotyping protocols such as PFGE is that it will allow us to identify transmissible types locally as well as globally and provide a basis for the examination of virulence in common genotypes. If we fail to attempt to learn a form of "molecular Esperanto" in terms of speaking a common language of harmonized and standardized methodology, we will potentially suffer, as befell the Israelites in Old Testament times with the Tower of Babel (Genesis 11), from misunderstanding. Indeed, if we cannot communicate with each other, we can hardly cooperate with each other.

TABLE 1. Comparison of PFGE typing methodologies

Reference for method	Reference for method modified	Restriction enzyme(s) employed	Electrophoresis parameters
1	7	*Spe*I	0.5–25 s, 20 h
	3		30–60 s, 4 h, 6 V/cm
12	17	*Spe*I	5–35 s, 11°C, 24 h, 6 V/cm
14	13	*Spe*I	5–90 s, 24 h, 13°C
5	4	*Xba*I, *Spe*I	2–28 s, 20 h, 14°C at 200 V
9	ND[a]	*Xba*I, *Spe*I	ND
6	10	*Dra*I, *Xba*I	ND
16	ND	*Spe*I	5.3–34.9 s, 14°C, 20 h at 100–130 mA, 6 V/cm
2	10	*Dra*I, *Xba*I	ND
	18		
	11		
8	10	*Dra*I, *Xba*I	3–15 s, 5.6 V/cm

[a] ND, not described.

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