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ABSTRACT BOOK

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PC-82405-18 A rapid sputum PCR-SSCP test for guiding pyrazinamide use in tuberculosis therapy

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Rationale: Pyrazinamide susceptibility testing is usually too slow to guide initial therapy, so that many patients with pyrazinamide-resistant tuberculosis receive ineffective pyrazinamide therapy.

Objectives: To optimize and evaluate a rapid molecular test for tuberculosis drug susceptibility to pyrazinamide.

Methods: Tuberculosis polymerase chain reaction-single strand conformational polymorphism (PCR-SSCP) was optimized to test for mutations causing pyrazinamide resistance directly from sputum samples and *Mycobacterium tuberculosis* isolates. The reliability, sensitivity and specificity of PCR-SSCP in sputum ($n = 65$) and isolates ($n = 185$ from 147 patients), was compared with the Bactec-460 microbiological test ($n = 139$) that was considered to be the gold standard. Pyrazinamide susceptibility testing for all 185 isolates was also compared with the Wayne biochemical test, with DNA sequencing for *pncA* mutations that cause pyrazinamide resistance and with traditional microbiological susceptibility testing in duplicate broth cultures containing pyrazinamide.

Results: PCR-SSCP provided interpretable results for 96% (46/48) of smear positive, 76% (13/17) of smear negative sputum samples and 100% of *M. tuberculosis* isolates. PCR-SSCP had pyrazinamide susceptibility testing sensitivity and specificity of 88% and 93% for sputum and 89% and 95% for isolates. For isolates that yielded concordant results in conventional broth culture, sensitivity and specificity were 95% and 92%, compared with 88% and 100% for Wayne biochemistry. All 34 negative-controls were PCR-SSCP negative. Material costs were \$1.36 per test and PCR-SSCP took one day, compared with 3–7 weeks for the other techniques.

Conclusions: PCR-SSCP is a rapid test that indicates which patients should receive pyrazinamide from the start of therapy, potentially preventing months of inappropriate treatment.