

Fluorescence microscopy has been used to measure phenotypic responses of bacterial cultures to antibiotics, either by detecting the effects of antibiotics on cell growth or by detecting cell lysis in agarose microgels (Tamayo et al. 2009; Santiso et al. 2011). Cell lysis-based methods can rapidly detect bacteria that are phenotypically resistant to antibiotics and measure their level of sensitivity (Pulido et al. 2013). The bacteria are incubated with different concentrations of the antibiotics to be tested and immobilized in microgels on microscope slides. Assays can be tailored to detect resistance to antimicrobial agents that affect bacterial cell walls or to those that damage DNA. For detecting resistance to antibiotics affecting peptidoglycans in cell walls, a lysis solution is selected that will lyse only the bacteria whose cell walls have been damaged (Santiso et al. 2011). Following removal of the lysis solution, the bacteria in the microgels are stained using a DNA-staining fluorescent dye, such as SYBR Gold. Resistant cells are not lysed and show normal DNA in the nucleoid. In contrast, the susceptible bacteria are lysed, resulting in spreading of the DNA (Santiso et al. 2011). Other cell lysis methods have been used to determine the sensitivity of bacteria in agarose microgels to ciprofloxacin and other fluoroquinolones that damage the DNA, inducing double-stranded breaks and causing DNA fragmentation (Tamayo et al. 2009). After exposure to ciprofloxacin, the cells are embedded in a microgel and treated with a stronger lysis solution that destroys protein and removes the cell wall and membrane. The lysis solution is removed, and the bacteria are stained with a fluorescent dye and visualized (Tamayo et al. 2009). In antibiotic-resistant cells, the DNA appears intact in the nucleoid, but in sensitive cells, the DNA becomes fragmented and appears to have diffused out of the nucleoid. Quantitative fluorescence microscopy with bacterial cytological profiling has been used for identifying β -lactam-resistant (including methicillin) and daptomycin-resistant strains of *S. aureus* within one to two hours by measuring changes in cellular structure induced by antibiotic exposure (Quach et al. 2016). The increased length of sensitive Gram-negative cells induced by ceftazidime has been measured by fluorescence microscopy, even without cell lysis, by staining with SYBR Gold, followed by digital image analysis (Otero et al. 2017).

A miniaturized *lab-on-a-chip* can be fabricated from various materials and substituted for agar as a support to grow bacteria for rapid testing. For instance, porous aluminum oxide ceramic chips have been used to detect trimethoprim-resistant strains of the *Enterobacteriaceae* in two to three hours and rifampicin/isoniazid-resistant strains of the slow-growing *Mycobacterium* spp. in three days (Ingham et al. 2006, 2008). Antibiotic-sensitive bacteria can be detected in tiny compartments on a chip by comparing their growth with and without antibiotics after only one hour (Besant et al. 2015). The reduction of a dye, resazurin, shows growth, and differential pulse voltammetry distinguishes the oxidized and reduced forms of the dye. Another lab-on-a-chip, which uses a