

NEW METHODS FOR THE QUALITY CONTROL OF CULTURE MEDIA FOR MYCOBACTERIA

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Purpose: The existing recommendations for the quality control of culture media for mycobacteria are poorly conceived. We analysed the possibility to characterize culture media regarding their potency for stimulation of mycobacterial growth by analysing the growth of tubercle bacilli in different media with the help of the BacT/Alert 3D system (3D system). Furthermore we characterized the potency of supplements for the suppression of contaminants.

Methods: For the analysis, we removed the original medium of the MP bottles of the 3D system and replaced it by 10 ml of the medium under investigation. In the case of characterizing solid culture media we used the unspiced media. The bottles were then inoculated with test strains (*M. tuberculosis* H37Rv, *M. bovis* IMM100) and incubated in the 3D system. The following parameters were recorded and calculated, resp.: time to detection, generation time, slopes of the growth curves of the 3D system in defined sections. The potency of supplements for inhibition of contaminants was characterized by determination of their MIC values for a set of defined test strains of bacteria (Gram+, Gram-) and yeasts.

Results: The mentioned growth parameters allow the recognition of growth stimulating effects of substrates (e.g. glycerol for *M. tuberculosis*, pyruvate for *M. bovis*), or growth protracting effects of omitted ingredients (e.g. serum in Kirchner medium), and allow the comparison of growth stimulating effects of supplements (e.g. OADC versus serum). The patterns of MIC values of widely-accepted antibiotic supplements for mycobacterial media for the test strain panel correlate with the known features of these supplements in practice.

Conclusion: The described methods are able to compare objectively the quality of media and supplements for cultivation of mycobacteria. Furthermore the presented method for growth characterization facilitates the optimization of culture media for mycobacteria.