Multicentre external quality control evaluating universal 16S polymerase chain reaction (PCR) in the diagnosis of bone and joint infections

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Auteur	Plouzeau, Chloé [1], Domelier, A.-S. [2], Héry-Arnaud, Genevieve [3], Bémer, Pascale [4], Bret, Laurent [5], Burucoa, Christophe [6], Cognee, A.-S. [7], Cottin, Jane [8], Gibaud, Sophie [9], Guinard, J. [10], Jolivet-Gougeon, Anne [11], Kempf, Marie [12], Lemarié, Carole [13], Quentin, R. [14], Tande, Didier [15], Vincent, P. [16], Corvec, Stéphane [17]
Pays	Allemagne
Ville	Berlin

Résumé en anglais	**Objectives:** During a multicentrer French Study performed to assess the contribution of 16S PCR in the diagnosis of prosthesis osteoarticular infections, 300 patients were included from December 2009 to April 2012. An external quality control (QC) was considered essential due to the diversity of molecular equipment for each laboratory. Three sets were held, for each including 4 bacterial DNA extracts (E) and 4 crushed osteoarticular deep samples (S). **Methods:** Extraction: 0,2 ml of pretreated S (PK, 37 °C, 3h) with elution in 0.1 ml. Four laboratories used Qiagen manual extraction and 3 others used automated extraction 1 MagnaPur Roche, 1 Easy Mag, BioMérieux and 1 iPrep, Invitrogen. Real time 16S PCR with SybrGreen was performed with degenerate primers amplifying 658pb followed by sequencing. In the 7 centers, PCR thermocyclers used were 2 MX 3000p Agilent, 1 Roche Light Cycler, 1 Abi 7900 and 1 Applied Step one plus, 1 Smartcycler Cepheid,1 Biorad Chrono 4 and for PCR premix, Takara premix exTaq, Applied, Promega and Biorad were used. **Results:** 168 QC were sent and 160 responses were analyzed (1 laboratory did not participate in the first QC series). Expected results were obtained in 97.5% for Extracts and 95% for Samples. Sensitivity and Specificity were 100 and 90% for E and 93.3 and 100% for S. Ct standard deviations (SD) for E were from 1 to 9 while SD was 2 to 7 for S. For centers using the same premix, the results were closer, SD 0.5 to 1.5 (3 Ct gap max). For S, no influence of extraction system was observed. **Conclusion:** If extraction system had no influence, premix seems to be the most important factor influencing the value of threshold. This QC demonstrates the possibility to obtain good and homogeneous results by using the same 16S PCR in laboratories with different equipments for molecular bone and joint infection diagnosis.
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