Commutability of the World Health Organization International Standard for Human Cytomegalovirus: Standard or Assay

Jacqueline F. Fryer, Clare L. Morris, Neil M. Almond, Philip D. Minor

Division of Virology, NIBSC, Medicines and Healthcare Products Regulatory Agency, South Mimms, Potters Bar, Hertfordshire, United Kingdom

The development and widespread use of calibrants that are traceable to a higher-order standard enable the results of diagnostic tests performed by different laboratories or at different times to be compared effectively. However, for this to be the case, then it is essential that all reference materials are calibrated appropriately and perform in a similar manner to a patient sample. In 2010, the first WHO international standard (IS) for human cytomegalovirus (CMV) was established as a high-order reference material. Its intended use is to calibrate secondary reference materials used in CMV nucleic acid amplification techniques (NAT) in international units (IU) (1). The CMV IS comprises laboratory-grown virus formulated in a Tris-HCl buffer to enable dilution in the matrix appropriate to the assay (1, 2). This was designed to control for matrix effects and ensure that the standard would be commutable, although at the time of establishment, this was not assessed formally. In their recent study, Jones et al. report on their studies of the commutability of the CMV IS with plasma and whole-blood specimens using the Abbott RealTime CMV assay (3). The authors report that the CMV IS behaves differently when diluted in plasma and whole-blood matrices and extracted using the respective protocols for these matrices, whereas patient samples do not. This matrix effect appeared to reduce the signal for the IS when diluted in whole blood by 0.4 log IU/ml compared with the plasma extraction procedure in this assay platform. The authors conclude that the IS is not commutable.

Commutability of a reference standard is dependent not only on the material itself but also on the assay in which it is applied. In particular, in quantitative assays where internal calibrators are included, the nature of these calibrators and the way that they are processed will impact the outcome of the assay (1). In the reported assay, the plasmid-based calibrator used in both plasma and whole-blood processes was calibrated against the IS in plasma alone, rather than in both matrices as recommended (2). This is likely to have contributed to the differing results observed for the patient samples and the IS when diluted in each matrix. If the calibration for the whole-blood process been performed against the IS in whole blood, then the stated conversion factor would have been lower, but comparable with those of other studies (4, 5).

In order to fully establish the commutability of reference materials, large-scale studies using multiple assay platforms in many laboratories are required (6, 7). We are undertaking studies to investigate the commutability of the IS for CMV NAT and are collaborating in studies organized by others. The results will contribute to this active debate and clarify the benefit and limitations

of specific reference materials in the standardization of assays and data that are designed to assist the clinical management of CMV. Until that time, it will not be possible to fully establish whether the different results observed in the article are due to the IS or the assay used.

REFERENCES

1. Fryer JF, Heath AB, Minor PD, Collaborative Study Group. 2016. A collaborative study to establish the 1st WHO international standard for human cytomegalovirus for nucleic acid amplification technology. *Biologicals* 44:242–251. <http://dx.doi.org/10.1016/j.biologics.2016.04.005>.
2. World Health Organization. 10 September 2014. 1st WHO international standard for human cytomegalovirus for nucleic acid amplification techniques, NIBSC code: 09/162, instructions for use, version 6.0. World Health Organization, Geneva, Switzerland. <https://www.nibsc.org/documents/ifu/09-162.pdf>.
3. Jones S, Webb EM, Barry CP, Choi WS, Abravaya KB, Schneider GJ, Ho SY. 2016. Commutability of cytomegalovirus WHO international standard in different matrices. *J Clin Microbiol* 54:1512–1519. <http://dx.doi.org/10.1128/JCM.03292-15>.
4. Schnepf N, Scieux C, Resche-Riggon M, Feghoul L, Xhaard A, Gallien S, Molina JM, Socié G, Viglietti D, Simon F, Mazon MC, Legoff J. 2013. Fully automated quantification of cytomegalovirus (CMV) in whole blood with the new sensitive Abbott RealTime CMV assay in the era of the CMV international standard. *J Clin Microbiol* 51:2096–2102. <http://dx.doi.org/10.1128/JCM.00067-13>.
5. Furione M, Rognoni V, Cabano E, Baldanti F. 2012. Kinetics of human cytomegalovirus (HCMV) DNAemia in transplanted patients expressed in international units as determined with the Abbott RealTime CMV assay and an in-house assay. *J Clin Virol* 55:317–322. <http://dx.doi.org/10.1016/j.jcv.2012.08.017>.
6. Hayden RT, Preiksaitis J, Tong Y, Pang X, Sun Y, Tang L, Cook L, Pounds S, Fryer J, Caliendo AM. 2015. Commutability of the first World Health Organization international standard for human cytomegalovirus. *J Clin Microbiol* 53:3325–3333. <http://dx.doi.org/10.1128/JCM.01495-15>.
7. Preiksaitis JK, Hayden RT, Tong Y, Pang XL, Fryer JF, Heath AB, Cook L, Petrich AK, Yu B, Caliendo AM. 15 June 2016. Are we there yet? Impact of the first international standard for cytomegalovirus DNA on the harmonization of results reported on plasma samples. *Clin Infect Dis* 63:583–589. <http://dx.doi.org/10.1093/cid/ciw370>.

Citation Fryer JF, Morris CL, Almond NM, Minor PD. 2016. Commutability of the World Health Organization international standard for human cytomegalovirus: standard or assay. *J Clin Microbiol* 54:3073. doi:10.1128/JCM.01706-16.

Editor: A. J. McAdam, Boston Children's Hospital

Address correspondence to Jacqueline F. Fryer, Jacqueline.Fryer@nibsc.org.

For the author reply, see doi:10.1128/JCM.01725-16.

© Crown copyright 2016.