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**Comments on the proposed revision of European Pharmacopoeia General Chapters 2.6.24 [PA/PH/Exp. 15 V/T (11) 93 ANP] and 2.6.25 [PA/PH/Exp. 15V/T (11) 94 ANP]**

People for the Ethical Treatment of Animals (PETA) represents more than three million members and supporters who are concerned about the use of animals in experimental procedures, and are especially troubled in instances where a non-animal-based replacement is available, provides valid data, has been proven accurate, yet is not used. As part of our ongoing work to help reduce the use of animals in these instances, the PETA International Science Consortium (PISC) works with regulators, companies and scientists to promote the implementation of validated humane methods.

PISC supports the amendment of European Pharmacopoeia (Ph. Eur.) General Chapters 2.6.24 and 2.6.25 to further promote the use of alternative methods, specifically PCR and other nucleic acid techniques, when virus neutralization and consequently egg yolk sac inoculation are not possible. This option allows for the use of non-animal methods with the agreement of the competent authority, following validation for specificity and sensitivity against codified tests.[1] So that these techniques may be more fully put into use, PISC recommends that Ph. Eur. commit to further work to support the wider implementation of these non-animal approaches to vaccine purity testing.

Requirements of the Ph. Eur. for avian viral vaccine purity testing are confusing, and can be interpreted differently by manufacturers and licensing authorities. The result is that, currently, manufacturers are obliged to carry out full testing of starting materials, master seeds and final product batches using methods that uniformly require the use of birds or tissue components derived from specific pathogen free (SPF) flocks.**Error! Bookmark not defined.** These tests require embryonated eggs from SPF flocks, chicken embryo kidney cells from SPF flocks, and serological tests directly carried out on two-week old SPF chicks.[2] The production and maintenance of SPF flocks themselves require the quarantine of at least three generations of chickens who are subjected to frequent testing to assess the presence or absence of pathogens of concern.[3] Vaccine manufacturers have estimated that these testing practices are responsible for the use of as many as 51,000 birds each year.[4]

To provide a forum for discussion of extraneous agents testing issues, the International Association for Biologicals (IABS) and the International Federation for Animal Health Europe (IFAH-Europe) hosted an October 2009 workshop to review and evaluate the current procedures and purity standards of veterinary vaccines. At this workshop, participants agreed on the need for practical tests that are less complicated and less expensive, and which would reduce the use of tests that require SPF chicken flocks. PCR and nucleic acid tests were highlighted at this workshop for their potential to replace the use of currently required tests at a lower cost, with greater speed and a comparable—and sometimes superior—degree of sensitivity.

Although Ph. Eur. currently allows the use of nucleic acid amplification techniques that have been validated against codified purity tests for sensitivity and specificity, workshop participants noted that comparison of PCR-based tests to codified tests is difficult because many of these codified tests have not themselves been validated.[5] Several approaches to remedying this situation have been proposed, and PISC recommends that Ph. Eur. give them further consideration:

1. To facilitate the use of PCR-based methods in purity testing, there is a strong need for PCR reference materials for the list of avian viruses for which testing is mandated in General Chapters 2.6.24 and 2.6.25. We urge Ph. Eur. to commit to the development of PCR reference materials for the list of avian viruses which must be tested for as potential extraneous agents. Availability of these reference preparations would help authorities and manufacturers evaluate and compare the suitability of newly developed PCR methods with regard to their sensitivity.[6] Test methods currently mentioned in these chapters for which no validated protocols and no reference standards are available should be deleted.[5] This approach will ultimately facilitate future harmonization efforts, as the United States Department of Agriculture (USDA) Center for Veterinary Biologics (CVB) is considering the use of PCR as a potential approach to avian viral vaccine purity testing.[7] We continue to support the harmonization of PCR-based testing guidelines with the International Cooperation on Harmonisation of Technical Requirements for Registration of Veterinary Medicinal Products (VICH), as noted in this letter.

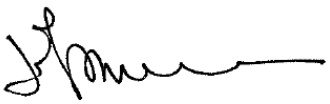
2. PISC also recommends that Ph. Eur. include reference to PCR assays that have already been developed and at least partially validated for use in place of current codified tests. The Institute of Virology and Immunoprophylaxis (IVI) and the Paul-Ehrlich-Institut (PEI) have jointly validated PCR assays for use in testing for a variety of extraneous agents, including avian leucosis viruses, avian orthoreovirus, infectious bursal disease virus, infectious bronchitis virus, Newcastle disease virus, infectious laryngotracheitis virus, influenza A virus, Marek's disease virus, turkey rhinotracheitis virus, egg drop syndrome virus, fowl adenovirus, chicken anemia virus and avian encephalomyelitis virus.[2,8,9,10,11,12,13] These viruses represent almost half of the extraneous agents listed in General Chapter 2.6.24 for which live bird inoculation testing is required. Although PCR-based tests for these viruses are not considered fully validated for pharmacopoeial reference, methods for their validation have been demonstrated and published in the literature. Referencing the availability of these methods in the revised General Chapters will assist vaccine manufacturers in the in-house validation process required for their use. In the case of extraneous agents that have documented incompatibilities with the codified tests—for example, reovirus, poxvirus and Newcastle disease virus[14]—we recommend that these general chapters specifically mention pathogens for which this justification is already known. Wherever possible, Ph. Eur. should collaborate with Official Medicines

Control Laboratories (OMCL) in order to more fully validate the use of PCR-based extraneous agent tests.[15]

3. While the quality of starting materials and final products have been increased by the introduction of quality systems such as Good Manufacturing Processes (GMP) into the manufacturing process, purity testing provisions of the Ph. Eur. have not been adjusted accordingly. Workshop participants encouraged Ph. Eur. to recognize the need to reduce extraneous agent tests, in particular those on finished products involving experiments using animals, when sufficient upstream measures to guarantee viral safety are in place.[16] PISC echoes this recommendation to increase the use of risk-benefit assessments in regulatory policies in place of finished product testing where GMP considerations have been implemented.

We look forward to the successful revision of the Ph. Eur. to more fully support the use of non-animal methods, and we ask that EDQM keep us informed of any further plans to revise these general chapters. Please feel free to contact me by phone at (310) 437-8003, or by email at JeffreyB@peta.org, if you have any questions on this important matter.

Sincerely,



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