European Regulatory Framework and Practices for Veterinary Leptospira Vaccine Potency Testing

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Synopsis

• **Role of**
  - National Authorities
  - the EU
  - the Ph.Eur.

• **Organization & Products of the Ph.Eur.**

• **The Pharmacopoeia**
  - Elaboration
  - Content
  - Leptospira vaccine monographs

• **Recent developments in Leptospira vaccine testing**
The European Pharmacopoeia

- 1964: Convention on the elaboration of a European Pharmacopoeia

- Today:
  - 36 member states + the European Union
  - 24 observer countries and international organisations including World Health Organization (WHO)
European Union (EU)
Its legal status

• Lays down common, compulsory quality standards for all medicinal products in Europe, i.e. raw materials, preparations, dosage forms, containers

• Mandatory at the same date in 36 states (CoE) and the EU

• National pharmacopoeias to cover subjects of solely national interest

• 1975: Mandatory status reinforced in the EU pharmaceutical legislation for the EU/EEA member states (Directives 2003/63/EC, 2001/83/EC, 2001/82/EC)
European Pharmacopoeia (Ph. Eur.)

Organization of the Ph. Eur.

- Commission

European Regulatory Framework and Practices for Veterinary Leptospira Vaccine Potency Testing
Ph. Eur. Commission

- One delegation per member state or observer
- 36 Member States plus a delegation from the EU (a representative from DG Health & Consumer and the EMA);
  24 Observer countries and World Health Organization (WHO).
- Persons come from health ministries, health authorities, pharmacopoeias, universities, or industry and are appointed by the national authorities on the basis of their expertise.
- Three sessions a year; texts are adopted by unanimous vote.
European Pharmacopoeia (Ph. Eur.)

Organization of the Ph. Eur.

- **Commission**

- **Expert groups**
  - Biologicals methods and statistical analysis
  - Organic chemistry - Synthetic products
  - Veterinary sera and vaccines
  - …..

- **Technical Secretariat (EPD)**
European Pharmacopoeia (Ph. Eur.)

“Products”

- European Pharmacopoeia
  - Book
  - On-line (http://online.edqm.eu)
  - USB-stick
European Pharmacopoeia (Ph. Eur.)

“Products”

• European Pharmacopoeia
  → Book
  → On-line (http://online.edqm.eu)
  → USB-stick

• PHARMEUROPA
  → On-line (http://pharmeuropea.edqm.eu)

• Knowledge database
  → On-line

• Reference Standards
  → Biological Standardisation Committee
The Vision of the EDQM
- A Directorate of the Council of Europe created in 1984
- A leading organisation that protects public health by enabling the development, supporting the implementation and monitoring the application of quality standards for safe medicines and their safe use.
- Develops also guidance and standards in the areas of blood transfusion, organ transplantation and consumer health issues.

What's new at the EDQM?

Latest News
12 September 2012
11th Symposium on Pharmaceutical Reference Standards
The Symposium brought together 146 experts from 27 countries representing authorities, pharmacopoeias, international organisations and industry. Proceedings available here.
10 August 2012
CEP July Monthly Report now available
The DCEP publishes on a monthly basis a report containing some relevant figures relating to its main activities.
09 August 2012
EDQM Publishes Annual Report for the year 2011
The report outlines its different activities and provides overview of achievements and results from 2011.

Focus

Biologics
The European Pharmacopoeia has its eye on the future.

In the spirit of continuous improvement, the Ph Eur Commission is committed to embracing the current technological and regulatory trends, but also appropriately paves the way for the future. In response to this, the EDQM has undertaken an extensive consultation among regulatory authorities and Expert Groups.

Read more on the European Pharmacopoeia

Upcoming Events
27-29 September
IPU/EDQM/WHO Technical Conference “Current Challenges in Global Regulatory Compliance”
Mumbai, India

3-8 October 2012
FIP Centennial Congress
Amsterdam, The Netherlands

13 October 2012
European Day for Organ Donation & Transplantation
Elaboration of a Ph.Eur monograph

The **European Pharmacopoeia Commission** decides to elaborate/revise a monograph

- **Group of experts**
  - a rapporteur prepares a draft monograph, which is evaluated by the experts

**PHARMEUROPA**

- the draft monograph is published for public enquiry, which lasts 3 months

The **national pharmacopoeia authorities** process the comments received on the draft

- The **technical secretariat (EPD)** compiles the comments

The **group of experts** examines the comments and revises the draft monograph accordingly

The draft is proposed to the **European Pharmacopoeia Commission**

- adopts
- does not adopt

**EUROPEAN PHARMACOPOEIA:**

- the monograph is published about 6 months after adoption
European Pharmacopoeia (Ph. Eur.) → legal requirements

• General Notices
• Monographs
  • Vaccines for Veterinary Use (0062)
  • Vaccine specific monographs
• Supplementary texts
  • 2.6.1. Sterility
  • 5.2.4. Cell cultures for the production of veterinary vaccines
  • 5.2.6. Evaluation of safety of veterinary vaccines and immunosera
  • 5.2.7. Evaluation of efficacy of veterinary vaccines and immunosera
  • ....
CANINE LEPTOSPIROSIS VACCINE (INACTIVATED)

Vaccine leptospirosis caninae inactivatum

1. DEFINITION
Canine leptospirosis vaccine (inactivated) is a preparation containing an inactivated vaccine of Leptospira interrogans serovars canicola, pomona, australis, and icterohaemorrhagiae, and/or serovars Icterohaemorrhagiae, canicola, autumnalis, and icterohaemorrhagiae, and/or serovars icterohaemorrhagiae, canicola, autumnalis, and icterohaemorrhagiae. The vaccine is administered to dogs of the minimum age to be recommended. The vaccine administered to each dog is of minimum antigen content and/or potency.

2. PRODUCTION

2.1. PREPARATION OF THE VACCINE
The seed material is cultured in a suitable medium; each strain is propagated and harvested separately. The harvested cultures are concentrated and inactivated by a suitable method. The antigen may be further concentrated and/or inactivated by a suitable method. The vaccine may be further purified and/or inactivated by a suitable method.

2.2. CHOICE OF VACCINE COMPOSITION
The antigen is shown to be satisfactory with respect to safety and efficacy. The product is not intended for use in dogs for which it is intended.

3. RATCH TESTS
3.1. Identification. When injected into healthy animals that do not have specific antibodies against the principal serovars of L. interrogans (icterohaemorrhagiae, canicola, and autumnalis), or have been obtained from a regularly inspected source. Administration of the vaccine to dogs by the intramuscular route into a suitable quantity. The vaccine is administered to dogs at a suitable time after the onset of the infection. The vaccine is administered to dogs at a suitable time after the onset of the infection.

4. REACTIVITY

4.1. Protection. When injected into healthy animals that do not have specific antibodies against the principal serovars of L. interrogans (icterohaemorrhagiae, canicola, and autumnalis), or have been obtained from a regularly inspected source. Administration of the vaccine to dogs by the intramuscular route into a suitable quantity. The vaccine is administered to dogs at a suitable time after the onset of the infection. The vaccine is administered to dogs at a suitable time after the onset of the infection.

5. STABILITY

5.1. Stability. When injected into healthy animals that do not have specific antibodies against the principal serovars of L. interrogans (icterohaemorrhagiae, canicola, and autumnalis), or have been obtained from a regularly inspected source. Administration of the vaccine to dogs by the intramuscular route into a suitable quantity. The vaccine is administered to dogs at a suitable time after the onset of the infection. The vaccine is administered to dogs at a suitable time after the onset of the infection.

See the information section on general monographs (cover page).
Canine leptospirosis vaccine (inactivated)

Batch potency test *(Hamster Test)*

It is not necessary to carry out the Potency test (test in dogs) for each batch of the vaccine if it has been carried out using a batch of vaccine with a minimum potency. Where the test is not carried out, an alternative validated method is used, the criteria for acceptance being set with reference to a batch of vaccine that has given satisfactory results in the test described under Potency. The following tests may be used.

2-3-1-1. *For vaccines with or without adjuvants.* If leptospira from more than one serovar (for example L. interrogans serovar canicola and serovar icterohaemorrhagiae) has been used to prepare the vaccine, carry out a batch potency test for each serovar against which protective immunity is claimed on the label. Use for the test 10 healthy hamsters not more than 3 months old, that do not have antibodies against the principal serovars of L. interrogans (icterohaemorrhagiae, canicola, grippotyphosa, sejroe, hardjo, hebdomonadis, pomona, australis and autumnalis) and which have been obtained from a regularly tested and certified leptospira-free source. Administer 1/40 of the dose for dogs by the subcutaneous route to 5 hamsters. Maintain 5 hamsters as controls. Challenge each hamster after 15-20 days by the intraperitoneal route with a sufficient quantity of a virulent culture of leptospirae of the serovar against which protective immunity is claimed on the label. The vaccine complies with the test if not fewer than 4 of the 5 control hamsters die showing typical signs of leptospira infection within 14 days of receiving the challenge suspension and if not fewer than 4 of the 5 vaccinated hamsters remain in good health for 14 days after the death of 4 control hamsters.
Canine leptospiroisis vaccine (inactivated)

Batch potency test (ctd.)

2-3-1-2. For vaccines with or without adjuvants. A suitable validated sero-response test may be carried out. Vaccinate each animal in a group of experimental animals with a suitable dose. Collect blood samples after a suitable, fixed time after vaccination. For each of the serovars present in the vaccine, an in vitro test is carried out on individual blood samples to determine the antibody response to one or more antigenic components which are indicators of protection and which are specific for that serovar. The criteria for acceptance are set with reference to a batch of vaccine that has given satisfactory results in the test described under Potency.

➢ Serological Test

2-3-1-3. For vaccines without adjuvants. For each of the serovars present in the vaccine, a suitable validated in vitro test may be carried out to determine the content of one or more antigenic components which are indicators of protection and which are specific for that serovar. The criteria for acceptance are set with reference to a batch of vaccine that has given satisfactory results in the test described under Potency.

➢ Antigen Quantification Test
Bovine leptospirosis vaccine (inactivated)

Batch potency test (*Serological Test*)

It is not necessary to carry out the Potency test (test in cattle) for each batch of vaccine if it has been carried out using a batch of vaccine with a minimum potency. Where the test is not carried out, an alternative validated method is used, the criteria for acceptance being set with reference to a batch of vaccine that has given satisfactory results in the test described under Potency. The following test may be used.

For each of the serovars for which protection is claimed, the antibody response from vaccinated animals is measured. Use not fewer than 12 guinea-pigs weighing 250-350 g that do not have antibodies against *L. borgpetersenii* serovar hardjo and the principal serovars of *L. interrogans* (icterohaemorrhagiae, canicola, grippotyphosa, sejroe, hardjo, hebdomonadis, pomona, australis and autumnalis) and that have been obtained from a regularly tested and certified leptospira-free source. The dose to be administered to the guinea-pigs is that fraction of a cattle dose which has been shown in the validation studies to provide a suitably sensitive test. Vaccinate each of 10 guinea-pigs with the suitable dose. Maintain not fewer than 2 guinea-pigs as controls. At a given interval within the range of 19-23 days after the injection, collect blood from each guinea-pig and prepare serum samples. Use a suitable validated method such as a micro-agglutination test to measure the antibodies in each sample.

The vaccine complies with the test if antibody levels are equal to or greater than those obtained with a batch that has given satisfactory results in the test described under Potency and there is no significant increase in antibody titre in the controls.
EDQM Workshops

on Alternatives to the Leptospira Vaccine potency test with participants from

• Licensing authorities
• Official Medicines Control Laboratories
• Industry
• Academia
• OIE reference laboratory

1999: Participants from Europe and U.S.A.
2012: Participants from Europe
Conclusions   workshop 1999

• Ph.Eur. monograph is outdated
• Hamster Potency test has deficiencies
• Alternative methods should be based on efficacy tests in the target species
  • Monographs revised, Conclusions integrated as “door openers”

• Working group should be created
  • sharing knowledge and
  • to coordinate efforts to replace the hamster test
Conclusions  workshop 2012

• LPS immunodominant
  • candidate antigen for antigen quantification
  • due to antigen variation no common reference material
  • mAbs may be obtained from Royal Tropical Institute Amsterdam (NL)
  • 2 manufacturers have CA approved vaccine on the market, tested for potency with an *in vitro* test

• LipL32 no evidence for protection
Conclusions  workshop 2012

• *In vitro* assay may be used to assess
  • Antigen quantification
  • Potency
  • (Stability)

• No single, universal alternative method
  • due to the complexity of the vaccines
    → number of serotypes, number of serovars
    → specific antigens as protective agents
    → combinations
    → presence/absence of adjuvants
Conclusions workshop 2012

- Consistency approach may reduce final product testing

- EDQM might provide standard mAbs for ELISA test

- Press release
  http://www.edqm.eu/medias/fichiers/edqm_workshopleptospirosis_vaccine_batch_potency_t.pdf
Further steps

✓ Information of Ph. Eur. Commission
  • Revision of the monograph for vaccines for dogs
    • On the agenda of expert group 15V
  • Develop guidance on implementation of consistency approach
✓ Update of EPAA
✓ Update of EMA/CVMP/IWP
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