408/2014

This is to certify that I, Marco Antônio Rochadel, Official Public Translator, designated and installed in Office according to The Official Gazette of June 23, 1982, page 5428, have received and translated, to the best of my knowledge and belief, a document with the following contents:

Ministério da Ciência e Tecnologia

GOVERNO FEDERAL BRASIL PAÍS RICO É PAÍS SEM POBREZA

Ministry of Science, Technology and Innovation – MCT
National Biosafety Technical Commission – CTNBio
Office of the Executive Secretary

comissão técnica nacional CTNBio de biossegurança

SPO, Área 05, Quadra 03, Bloco B, Térreo, Salas 08 a 10
70610-200 Brasília, Distrito Federal, ☎ +55 61 3411 5516 • +55 61 3317 7475

Technical Opinion no. 4090/2014

Proceedings:	01200.004713/2013-81
Applicant:	Laboratórios Vencovarma do Brasil Ltda.
CQB:	349/1723
Proton:	41912/2013
Matter:	Commercial Release of Inactivated Vaccine against Porcine Circovirus PRO-VAC CIRCOMASTER.
Previous Extract:	3814/2013, published on 10.14.2013.
Meeting:	173rd CTNBio Regular Meeting held on June 05, 2014
Decision:	**GRANTED**.

CTNBio, following examination of the proceedings related to the request for Technical Opinion on biosafety of a product intended for commercial release, reached a conclusion favorable to granting the request on the terms of the within Technical Opinion.

Regarding the competences provided by Law no. 11105/2005 and Decree no. 5591/2005, the Commission found that this request complies with CTNBio rules and applicable legislation

aimed at securing environment, agriculture, human and animal health biosafety.

TECHNICAL OPINION

ABSTRACT: The person legally in charge of the institution requested CTNBio a technical opinion related to biosafety involving the commercial release of Inactivated Vaccine against Porcine Circovirus named PRO-VAC CIRCOMASTER. The vaccine refers to a suspension containing, as active components, a minimum of 200 micrograms of antigen ORF2 of Porcine Circovirus type 2 (PCV2) (subunit vaccine) per dose of 1 ml. Protein ORF2 of PCV2 is produced in an expression system of baculovirus in an insect cell and used as antigen in this vaccine. The baculovirus was genetically modified to produce gene ORF2 of PCV2. Antigen ORF2 is produced in the cytoplasm of insect cells infected by recombinant baculovirus and secreted in the cultivation medium. After being produced, the antigen is purified and qualified for formulation. The structural protein of the modified ARF2 was selected as the vaccine antigen for being immunodominant and for inducing neutralizing antibodies that showed to be protective.

1. General Information.

The ORF2 recombinant protein of Porcine Circovirus type 1 (PCV2) will be produced in baculoviruses and secreted to the culture medium. There is no evidence of infection by PCV2 in other animal species and human beings and the heterologous baculovirus system is specific for insect cells. All the production shall be inactivated before its use in the form of vaccine for animal use.

The final product has inactivated baculovirus and ORF2 vaccine antigen, therefore, according to the National Biosafety Legislation, such product is a derivative (Article 3 of Law no. 11105/05).

2. GMO Description.

GMO is a baculovirus containing gene ORF2 of virus PCV2 (*Porcine Circovirus* – type 2) isolated by Komipharm International Co. Ltd. from an aviary in Korea. Gene ORF2 is a viral protein from the virus capsid and its use promotes the production of neutralizing antibodies. The

baculovirus used is the PvI1392/1393.

3. Product Biosafety

Analysis according to Ruling Resolution no. 5, of March 12, 2008, Annex III

PCV was first identified in 1974, as a contaminant of pig kidney cell cultures, PK-15. The first isolation of PCV-2, which is antigenic and genetically distinct from PCV-1 (held as non-pathogenic) was only isolated in 1996, from animals affected by the Porcine Multi-systemic Wasting Syndrome. PCV1 and PCV2 are small viruses, with about 17 nm of diameter, of icosaedric morphology and devoid from envelopes. Their nucleic acid is constituted by single strand DNA, with about 1759 nucleotides, of negative polarity and with covalently closed circular structure.

Gene ORF2 was isolated by the company Komipharm International Co. Ltd. from the virus PCV2 (*Porcine Circovirus* – type 2) that codifies ORF2, the capsid protein of virus PCV2, which serves as a vaccine antigen. Protein ORF2 of PCV2 was produced in a baculovirus expression system of an insect, Sf9 – *Spodoptera Fugiperda 9*. Antigen ORF2 is secreted to the cultivation medium. The protein is purified and used to formulate the vaccine, which is inactivated. Description of the whole methodology and genetic maps were adequately presented.

Baculoviruses are used as heterologous systems for production of recombinant proteins in laboratories. They are specific for insect cells and pose no risk to other animals or human beings. The vaccine is based on a protein of capsid PCV2 and has no apparent risk in the form it was expressed.

The whole production process involves the virus inactivation. Innocuity tests performed in vaccinated animals fail to reveal any deleterious effect such as suppuration, local necrosis, fever, death or any type of abnormal reaction. Innocuity tests were also held in pregnant females within the four first hours and at the 21st day from vaccination, and failed to display changes in vaccinated females and their litters. The information was displayed at the

company's report.

According to the Ministry of Health Biologic Agents Risk Classification, 2nd edition – 2010, the Circovirus belongs to Risk Class 1.

4. Environment Safety

The vaccine PRO-VAC® CIRCOMASTER will be marketed by Vencofarma do Brasil Ltda. under a commercial agreement with Komipharm International Co. Ltd. Vencofarma was established in 1986 and markets bacterial and viral vaccines as well as serums and other products. It is described that the vaccinated animals will be maintained in controlled areas under the supervision of technicians and veterinarians, and risks for the environment are not expected. Besides, it was described that protein ORF2 is present in other vaccines without posing any risks. All materials will be inactivated before they are used in animals.

5. Post-Commercial Release Plan

Monitoring of the vaccine shall be performed by Vencofarma and regulatory bodies. The company commits itself to perform a statistical analysis of the number of doses sold and the number of complaints received by its Customer Services and publish a comparative yearly report to be delivered to CTNBio.

6. CTNBio Final Opinion

Once conditions described by the protocol and biosafety measurements contained in the process are met, the activity is not potentially a cause of significant degradation to the environment and human health. Therefore, we are favorable to the commercial release of this inactivated vaccine.

7. Bibliography

Ye Y, Cheng X, Zhang J, Tong T, Lin W, Liao M, Fan H. Induction of robust immunity response in mice by dual-expression-system-based recombinant baculovirus expressing the capsid protein of porcine circovirus type 2. Virol J. 2013 Oct 28;10(1):316.

Trible BR, Kerrigan M, Crossland N, Potter M, Faaberg K, Hesse R, Rowland RR. Antibody recognition of porcine circovirus type 2 capsid protein epitopes after vaccination, infection, and disease. Clin Vaccine Immunol. 2011 May;18(5):749-57. doi: 10.1128/CVI.00418-10.

Fan H, Ju C, Tong T, Huang H, Lv J, Chen H. Immunogenicity of empty capsids of porcine circovirus type 2 produced in insect cells. Vet Res Commun. 2007. May;31(4):487-96.

Blanchard P, Mahé D, Cariolet R, Keranflec'h A, Baudouard MA, Cordioli P, Albina E, Jestin A. Protection of swine against post-weaning multisystemic wasting syndrome (PMWS) by porcine circovirus type 2 (PCV2) proteins. Vaccine. 2003 Nov 7;21(31):4565-75. PubMed PMID: 14575769

Dr. Edivaldo Domingues Velini

CTNBio President

In Witness Whereof, I have hereunto set my hand and seal in this City of Brasília, Federal District, Brazil, this Thursday, August 21, 2014.

Fees according to

Official Gazette of 04/15/2011 Marco Antônio Rochadel

Page 73 R$ 270.00 Public Translator