Study No. IND/SUI/14/037

FIELD TRIAL REPORT

EFFICACY AND SAFETY STUDY OF RECOMBINANT PORCINE CYSTICERCOSIS VACCINE (TSOL18 VACCINE) IN PIGS

SPONSORED BY

INDIAN IMMUNOLOGICALS LIMITED

GACHIBOWLI, HYDERABAD-32.

CONDUCTED BY

S.P.V.N.R.T.S.U.FOR VETERINARY, ANIMAL AND FISHERY SCIENCES

(FORMERLY SRI VENKATESWARA VETERINARY UNIVERSITY)

RAJENDRANAGAR, HYDERABAD, TELANGANA

INDEX

SYNOPSIS OF STUDY

Study Title: Efficacy and safety study of recombinant porcine cysticercosis vaccine (TSOL18 Vaccine) in Pigs.

Study Objective: The primary objective of the study is to determine the efficacy and safety of Porcine Cysticercosis Vaccine (TSOL18 vaccine), produced as a GMP batch suitable for commercial use, in the prevention of cysticercosis caused by *Taenia solium* in Pigs. Efficacy will be determined by serum IgG antibody responses as determined by specific ELISA. Safety will be determined by clinical observations including rectal temperatures and injection site observations.

Test Vaccine: Recombinant porcine cysticercosis vaccine (TSOL18 vaccine) manufactured by Indian Immunologicals Ltd. (IIL)

Study Center: Instructional Livestock Farm Complex
College of Veterinary Sciences
S.P.V.N.R.T.S.U.for Veterinary, Animal and Fishery Sciences
(Formerly Sri Venkateswara Veterinary University)
Rajendranagar, Hyderabad-30, Telangana, India.

Sponsor: Indian Immunologicals Limited
Rakshapuram, Gachibowli, Hyderabad-32
Telangana, India.

Principal Investigator: Dr. T. Raghunandan, Ph. D.
Professor and Head
Instructional Livestock Farm Complex
College of Veterinary Sciences
S.P.V.N.R.T.S.U.for Veterinary, Animal and Fishery Sciences
(Formerly Sri Venkateswara Veterinary University)
Rajendranagar, Hyderabad-30
Telangana, India.

ABBREVIATIONS

%	Percent
^{0}C	Degree celsius
µg	Microgram
CP	Control product
CPCSEA	Committee for the purpose of control and supervision of experiments on animals
DCF	Data capture form
DPV	Day post vaccination
ELISA	Enzymed linked immune sorbent assay
GMP	Good manufacturing practices
GMT	Geometric mean titre
IgG	Immunoglobulin G
IIL	Indian Immunologicals Limited
IVP	Investigational veterinary product
MBP	Maltose binding protein
nm	Nanometer
OD	Optical density
OIE	Office International des Epizooties (World Organization for Animal Health)
PBST	Phosphate buffered saline with tween 20
q.s	Quantum sufficit
S.P.V.N.R.T.S.U	Sri P.V. Narasimha Rao Telangana State University
TMB	Tetramethylbenzidine
VICH	International Cooperation on Harmonization (Veterinary)
WHO	World Health Organization

DECLARATION BY THE INVESTIGATOR

This study was conducted in accordance with the ethical principles as per Good Clinical Practice guidelines and Ethical Guidelines for Biomedical Research on animal subjects as per International Committee on Harmonization (Veterinary) (VICH) and Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA).

Principal Investigator

18/8/15

Professor & Head
Instructional Livestock Farm Complex
College of Veterinary Science
Rajendranagar, Hyderabad-500 030.

Dr. T. Raghunandan
Professor and Head
Instructional Livestock Farm Complex
College of Veterinary Sciences
S.P.V.N.R.T.S.U.for Veterinary, Animal and Fishery Sciences
Rajendranagar, Hyderabad-30
Telangana, India.

Study Dates:
Start date: 06/04/2015
End date: 24/06/2015

INTRODUCTION AND BACKGROUND

Disease:

Cysticercosis in various farm and wild animals and in humans is caused by larval cestodes of Taenia spp. tapeworms. Carnivore definitive hosts (humans, dogs, and wild canids) harbor adult tapeworms in the small intestine. Porcine cysticercosis and human neurocysticercosis are caused by *Taenia solium*. It is found principally in Mexico, Central and South America, sub-Saharan Africa, non-Islamic countries of Asia, including India and China in regions with poor sanitation and free-ranging, scavenging pigs. *Taenia solium* segments, however, are often passed in chains and flies are unimportant in dissemination. Eggs are immediately infective when passed. Animals acquire infection from ingestion of food or water contaminated with sticky eggs, ingestion of segments or faeces containing eggs. It is possible that pigs also acquire *T. solium* by coprophagy of the faeces of pigs that have eaten segments. Humans may be infected with *T. solium* by eggs on vegetables, in water, etc., that have been contaminated by faeces, or food contaminated by dirty hands, by faeco–oral transmission or through retroperistalsis and hatching of eggs internally. Disease clusters where a human carrier exists. Routine diagnosis of taeniasis continues to be mainly based on the morphology of the adult tapeworm and the presence of eggs or segments in the faeces of infected definitive hosts (OIE Terrestrial Manual 2014).

Taeniasis due to *T. solium* is usually characterized by mild and non-specific symptoms. Abdominal pain, nausea, diarrhoea or constipation may arise 6–8 weeks after ingestion of the cysticerci when the tapeworms become fully developed. In the case of cysticercosis due to *T. solium*, the incubation period is variable and infected people may remain asymptomatic for years or may develop visible or palpable subcutaneous nodules. When cysts are recognized by the host following spontaneous degeneration or after treatment, an inflammatory reaction may occur. Neurocysticercosis is associated with a variety of symptoms and signs depending on the number, size, stage and location of the pathological changes as well as the host's immune response and the parasite's genotype, but it can also be clinically asymptomatic. Symptoms may include chronic headaches, blindness, seizures (epilepsy if they are recurrent), hydrocephalus, meningitis, dementia and symptoms caused by lesions occupying spaces of the central nervous system (WHO Fact sheet no. 376).

Control of Neurocysticercosis

Transmission of *T. solium* has decreased or been eliminated in much of the first world over the last century as a result of improvements in general sanitation and public health. However, similar improvements in public living conditions are unlikely to occur in the short term in much of the developing world where cysticercosis is highly endemic, and other parasite control measures are required to have a more immediate impact on the prevalence of this disease. Neurocysticercosis is potentially eradicable since inexpensive and highly effective anthelmintics are available that could potentially be used to eliminate human tapeworm infections. To date, these measures have not achieved sustainable control of *T. solium* transmission. One major barrier of the use of anthelmintics to control *T. solium* is that despite the elimination of tapeworm carriers by mass treatment of the population with drugs to remove tapeworms, pig infection will remain. This reservoir of infection can readily establish new tapeworm infections of humans and renewed disease transmission (Armando *et al.*, 2005).

Vaccination

Lightowlers (1999, 2010) emphasized the comparative advantage of vaccination of pigs over vaccination of human population, for prevention of *T. solium* transmission and source of *T. solium* infection.

Gonzalez *et al.* (2005) and Lightowlers (2010) found recombinant TSOL18 antigen to be effective in the prevention of *T. solium* infection in pigs. A similar result was obtained by Cai *et al.* (2008).

Indian Immunologicals Limited (IIL) has produced TSOL18 vaccine using a more commercially-suitable procedure using *Pichia* expression and this has been shown to be particularly immunogenic when used in combinations with an oil adjuvant (mineral oil). The primary course of this vaccine is given as two doses (150 µg TSOL18 /dose), 3-4 weeks apart. The following studies have established the safety and efficacy of the vaccine.

1. Safety study conducted at University of Zaragoza, Spain:

The TSOL18 vaccine containing 150 µg recombinant protein and Montanide ISA206V adjuvant in 1 mL was shown to be safe in growing pigs from 8 weeks of age. This was further confirmed when given at 2X dose (300 µg) on 4 occasions at 7 to 14 day intervals and at 5X dose (750 µg, single dose) with Montanide ISA206V. Following treatment administration

there was transient, mild pyrexia which resolved within 24 hours and mild injection site reactions which resolved within 7 days. There were no significant adverse clinical or pathological effects of TSOL18 with ISA 206. All pigs given two doses of *Pichia* TSOL18 + ISA 206 developed serum IgG TSOL18 titres ≥ 500. It was concluded that the IIL manufactured TSOL18 vaccine containing 150 µg recombinant protein (expressed in *Pichia pastoris*) with ISA206 adjuvant was safe for use in growing pigs.

2. Efficacy Study conducted in Peru:

The vaccinated animals given two doses of *Pichia* TSOL18 + ISA 206 vaccine developed adequate titres (≥ 500) of TSOL18-specific IgG. Two doses of *Pichia* TSOL18 vaccine were found to be highly effective in the prevention of *T. solium* cysts and substantially reduced the burden of *T. solium* infection in pigs by >94% compared with control animals. Furthermore, infection did not establish (no cysts in tissues) in >90% of vaccinated pigs compared with 40% of control pigs. The results confirm that two doses of TSOL18 vaccine given three weeks apart are safe and effective.

Safety and Efficacy study conducted at Instructional Livestock Farm Complex, Hyderabad, India:

The present study was undertaken to determine the efficacy and safety of recombinant porcine cysticercosis vaccine (TSOL18 vaccine) produced by IIL, in the prevention of cysticercosis caused by *Taenia solium* in pigs.

STUDY OBJECTIVE:

The primary objective of the study is to determine the efficacy and safety of Porcine Cysticercosis Vaccine (TSOL18 vaccine), produced as a GMP batch suitable for commercial use, in the prevention of cysticercosis caused by *Taenia solium* in Pigs. Efficacy will be determined by serum IgG antibody responses as determined by specific ELISA. Safety will be determined by clinical observations including rectal temperatures and injection site observations.

MATERIAL AND METHODS

Study design:

This study was designed to assess the efficacy and safety of recombinant porcine cysticercosis vaccine (TSOL18 vaccine) in healthy pigs of at least 2 months of age and not older than 2 years which are serologically negative (serum TSOL18 antibody titre <500) for porcine cysticercosis. The animals were enrolled based on inclusion and exclusion criteria and allocated to Treatment Group T1 (Adjuvant control) or T2 (TSOL 18 vaccine) based on randomization plan. The T1 and T2 Groups were administered 2 doses (1 ml/dose) of Adjuvant control and TSOL 18 vaccine, respectively, 3 weeks apart through intramuscular route. Following booster administration, the study animals were observed for 28 days. The total duration of the study was 49 days.

Animal Selection, Groups and Treatments:

The animal selection was based on the inclusion and exclusion criteria of the study. Only the animals that complied with the following inclusion and exclusion criteria were enrolled for the study.

Inclusion Criteria:

- Healthy animals
- Amenable to handling
- Animals those were weaned for a minimum of 2 weeks before enrollment
- Animals those were at least 2 months of age and not knowingly older than 2 years
- Serologically negative for porcine cysticercosis
- All routine vaccinations and medications completed at the beginning of the study and at least 3 weeks prior to day 0 (day of vaccine or adjuvant control administration)
- Animals were moved to the study site for at least one month prior to selection
- Animals which were always kept indoors and not been allowed to roam free
- No possibility of contact with human faeces

Exclusion Criteria:

- Unhealthy, injured and/or under-weight animals
- All animals with hernias
- Any animal with an unacceptable growth rate

- Animals that were serologically positive for porcine cysticercosis in the acclimatization period.

In all, 100 seronegative Large White Yorkshire pigs in the age group of 2 months up to 2 years were enrolled for the trial. The grouping and treatments were as follows.

Treatment Group	No. of Animals and Age groups	Treatment administration
Treatment-1 (Adjuvant Control Group)	Total animals: 50 20 animals of 2-4 months; 30 animals of >4 months up to 2 years	One ml of adjuvant control by intramuscular route.
Treatment-2 (TSOL 18 Vaccine Group)	Total animals: 50 20 animals of 2-4 months; 30 animals of >4 months up to 2 years	One ml of TSOL 18 vaccine by intramuscular route.

Site of treatment administration:

The injection site for administration of the TSOL18 vaccine/Adjuvant control was on the left lateral neck muscle mass approximately 5-10 cm ventral to the dorsal surface of the neck and 5-10 cm posterior to the base of ear. A new needle and syringe was used for each pig.

Study procedures:

The study animals were identified with dual tagging each with unique identification. The animals were acclimatized for the period of 1 month before administration of vaccine or adjuvant control treatment. No concurrent drugs interacting with immune system (glucocorticoids, antibiotics, etc.) were used throughout the study period. Animals were housed in different pens in groups, as per the randomization plan following randomised block design where block is a pen, and pigs within a pen are ranked on basis of sex, age and estimated body weight and only then randomly allocated to treatments. Each pen contained about 10 animals belonging to both treatment groups, T1 and T2. Throughout the acclimatization and study period, the animals were kept in confinement and not allowed to roam outside their pens and courtyards. Standard husbandry practices for feeding, watering and management were followed. A nutritionally adequate diet in adequate quantities was provided. *Ad libitum* clean water was made available all the time.

Blood samples were collected from anterior vena cava in serum vacutainer tubes (Beckton Dickinson, USA). Sera samples from the animals were screened for TSOL 18 antibody titres using indirect ELISA method. Healthy animals which were found seronegative by indirect ELISA were selected for the study. At day 0, the T1 group animals were administered with 1 ml of Adjuvant control through intramuscular route and T2 group animals were administered 1ml of TSOL18 vaccine. Access to study information was partially restricted. Personnel involved in clinical examinations and laboratory ELISA assays were blinded. The personnel dispensing and administering the IVP and CP were not blinded. The non-blinded persons did not participate in any study procedures apart from those associated with the random allocation of animals to groups and IVP/CP administration.
On 21 DPV, the T1 and T2 group animals were administered with booster dose of Adjuvant control and TSOL18 vaccine, respectively, through intramuscular route.

Blood samples were collected on -30, -1, 20, and 35 DPV for antibody response by indirect ELISA. For safety evaluation, all the animals were observed for local and / or systemic reactions after vaccination and during the follow-up period.

Rectal temperatures, clinical observations and injection site observations were made 1 to 3 h before treatment administration and 4 to 8 h after administration and also on day 1, 3, 7, 14, 22, 24, 28, 35 and 49 as per study schedule. General health observations were performed on all days unless clinical observations were performed on the same day. The data were captured on relevant data capture forms (DCF).

Summary of activities:

Study day	Activity details
Prior to selection	Animal identification with two ear tags, each tag containing unique identification number.
-30	Test bleeding for screening of porcine cysticercosis antibodies by indirect ELISA method.
-15 to -13	1. Clinical examination of experimental pigs. 2. Animal selection based on inclusion and exclusion criteria. 3. Enrollment of animals for the study: Data recording of age, breed, sex and body weight of study animals. 4. General health observations for each individual pig on daily basis on the day after enrolment and continuation till the end of the study excluding days when clinical observations were made.
-14 to -5	Randomization and animal grouping.
-1	Test bleeding for antibody titre estimation by indirect ELISA method.
0	1. TSOL18 vaccine or Adjuvant control administration by intramuscular route in the neck region as per randomization plan. 2. Recording of clinical observations, injection site observations, rectal temperatures, 1 -3 hours before and 4 to 8 h after TSOL18 vaccine or Adjuvant control administration.
1, 3, 7, 14	Clinical observations. Injection site observations. Rectal temperatures.
20	Test bleeding and antibody titre estimation by indirect ELISA method.
21, 22	1.Administration of booster dose of TSOL18 vaccine or Adjuvant control intramuscularly in the neck region based on randomization plan. 2.Recording of clinical observations, injection site observations and rectal temperatures, 1 -3 hours before and 4 to 8 h after TSOL18 vaccine or Adjuvant control administration.

22, 24, 28. 35, 49	Clinical observations. Injection site observations. Rectal temperatures.
35	Test bleeding and antibody titre estimation by indirect ELISA.
49	End of study.
All days	Daily general health observations during the study period.

Test vaccine details:

Name:	Recombinant porcine cysticercosis vaccine (TSOL18 vaccine) (Test Vaccine)	
Composition:	*Taenia solium* oncosphere antigen (TSOL 18)	150µg
	Thiomersal	0.01%
	Phosphate buffer	q.s
	Mineral oil used as adjuvant	
Indications:	For the active immunization of pigs against porcine cysticercosis	
Storage and transport:	The vaccine was stored and transported between 2°C and 8°C.	

Control product details:

Name:	Adjuvant control	
Compostion:	Mineral oil used as adjuvant	
	Thiomersal	0.01%
	Phosphate buffer	q.s

Mode of administration of TSOL 18 vaccine/ Adjuvant Control:

- To the animals in T2 group, 1 ml of TSOL18 vaccine was administered intramuscularly through an area of clean and dry skin on the left lateral neck muscle

mass approximately 5-10 cm ventral to the dorsal surface of the neck and 5-10 cm posterior to the base of ear.

- To the animals in T1 group, 1 ml of adjuvant control was administered by intramuscular route through an area of clean and dry skin on the left lateral neck muscle mass approximately 5-10 cm ventral to the dorsal surface of the neck and 5-10 cm posterior to the base of ear.

Assessment after vaccination:

Efficacy

Efficacy of the TSOL18 vaccine was estimated by determining the IgG antibody response of recombinant porcine cysticercosis vaccine (TSOL18 vaccine) on 20 and 35 DPV by indirect ELISA. The pre and post vaccination antibody titres were compared to evaluate efficacy of vaccine in eliciting immune response and increasing the antibody titres above the protective level (≥500 IgG).

Safety

Animals were monitored for safety for the entire duration of the study. Rectal temperatures, clinical observations and injection site observations and general health observations were performed as per study schedule.

Test Procedures:

The serum samples collected from the study pigs were identified with label and stored at -20^{0}C in deep freezer until testing. The sera samples were subjected to indirect enzyme linked immunosorbent assay (i-ELISA) for the characterization of antibody responses induced by TSOL 18 vaccine.

Enzyme-linked immunosorbent assay:

TSOL18-specific antibody titres were determined using enzyme-linked immunosorbent assay as per Assana *et al.* (2010) using purified TSOL18-MBP as antigen. ELISA plates (Nunc, maxisorp, cat.No. 442404) were coated overnight at 4^{0}C with TSOL18-MBP antigen in carbonate buffer (pH 9.6, 100 µl/well). The plates were washed thrice with phosphate-buffered saline containing 0.05% Tween 20 (PBST) and blocked by incubation with 200µl/well of blocking buffer (1% Sodium caseinate in PBST) for 1 h at 37^{0}C. Plates were emptied and washed thrice with PBST. The serum samples were diluted to 1:20 in blocking

buffer and incubated at 2-8^{0}C for 2 hrs. 50µl of test serum samples serially diluted in blocking buffer were added and incubated at 37^{0}C for 1 hour after which the plates were washed thrice with PBST and incubated with 50µl/well of rabbit anti-porcine IgG HRPO conjugate (1 in 8000 in blocking buffer) at 37^{0}C for 1 hour. Then the plates were washed thrice with PBST and two times with distilled water. Chromogen / substrate solution (TMB and H_2O_2) was added (100 µl/well) and incubated at 37^{0}C for 30 min. in dark. The reaction was stopped by addition of 50 µl/well of 1.25N H_2SO_4 solution to each well. Optical densities (OD) were measured at 450 nm using a microplate reader.

Interpretation of results:

1. Samples are considered seropositive/seroprotective, if the titres are ≥500
2. Titres were calculated as the reciprocal of the highest dilution at which the sera had an OD of ≥1.0.
3. Titre values of serum samples were calculated by four parameter curve fitting using the statistical software, Sigmaplot 12.5.

RESULTS

Efficacy Evaluation:

Serological assessment:

The serological response induced by recombinant porcine cysticercosis vaccine (TSOL18 vaccine) was determined by indirect ELISA and expressed as geometric mean titres (GMT). All the animals were seronegative on day-1, with geometric mean titre of 181 in vaccinated group (T1) and 201.8 in control group (T2). The geometric mean titre on day 20 was 479, which increased to 2335.4 by day 35 in T2 group animals following booster vaccination on day 21. In the vaccinated group (n=48), there was 39.58 % seroprotection on day 20 which increased to 97.9% seroprotection by day 35 (47 out of 48 animals seroprotected), compared to day -1. All of the T1 group animals (adjuvant control, n=49) remained seronegative.

Table 1: Geometric Mean Titres of TSOL 18 antibodies at different days after vaccination

Treatment Group	Day -1(GMT)	Day 20 (GMT)	Day 35 (GMT)
Adjuvant Control group (T1)	201.8	281	278.1
TSOL 18 vaccine group (T2)	181	479	2335.4

Table 2: Percent seroprotection for TSOL 18 vaccinated animals on different days:

Group	Percent seroprotection on different days		
	Day -1	Day 20	Day 35
Adjuvant control group (T1)	0	0	0
TSOL 18 vaccine group (T2)	0	39.58	97.92

Fig 1: Geometric Mean titres at different days after vaccination

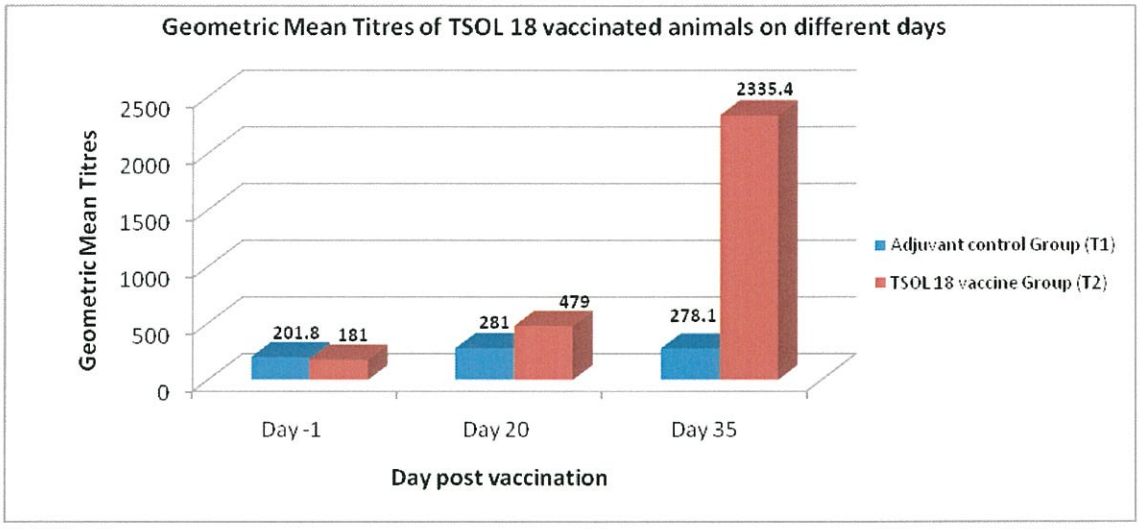
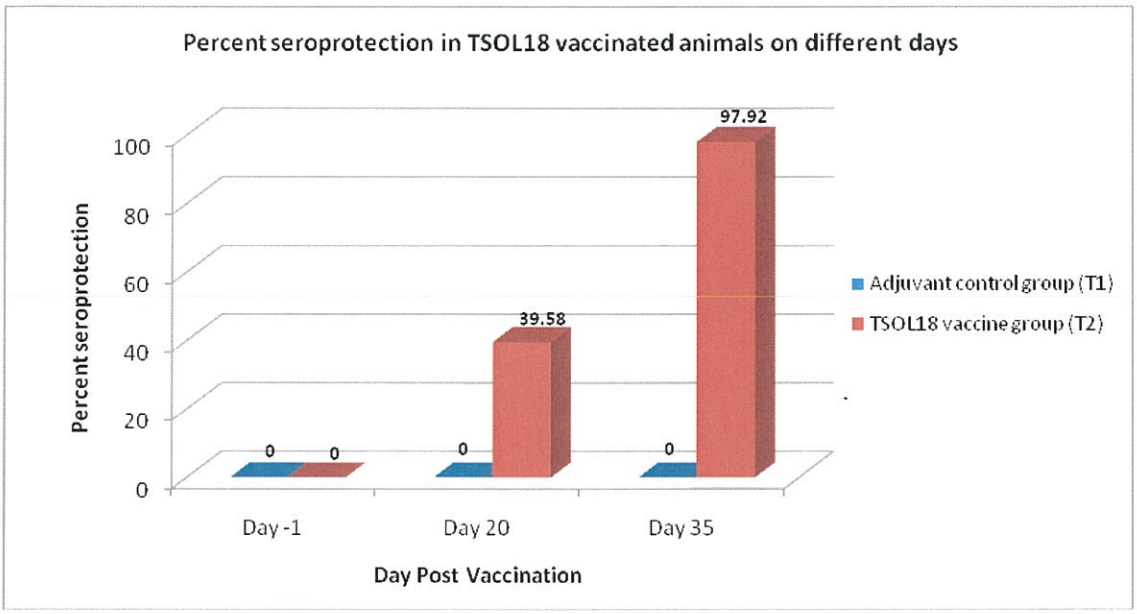


Figure 2: Percent seroprotection for TSOL 18 vaccinated animals at different days



Safety Evaluation:

Adverse reactions

The adverse reactions were categorized as local reactions and systemic reactions.

Local adverse reactions

The animals were observed for local pain, swelling, rashes, skin eruption, sloughing of mucous membrane, redness at administration site, local reaction etc. Details of observations are mentioned in Table 3.

Systemic adverse reactions

The animals were observed for fever, loss of appetite (anorexia) and restlessness etc. which are listed in Table 3.

Table 3: Summary of adverse reactions in different Groups

Adverse reactions	Adjuvant control group (T1) [N=49] n (%)		TSOL 18 vaccine group (T2) [N=48] n (%)	
	Primary dose	Booster dose	Primary dose	Booster dose
Local:				
Local pain	0 (0)	0 (0)	0 (0)	0 (0)
Swelling	3 (6.12%)	0 (0)	0 (0)	6 (12.5%)
Redness at administration site	0 (0)	0 (0)	0 (0)	0 (0)
Systemic:				
Fever	3 (6.12%)	0 (0)	4 (8.33 %)	2 (4.2%)
Anaphylactic reactions	0 (0)	0 (0)	0 (0)	5 (10.4%)
Loss of appetite	0 (0)	0 (0)	0 (0)	0 (0)
Restlessness	0 (0)	0 (0)	0 (0)	0 (0)
Systemic reactions (Diarrhea)	0 (0)	0 (0)	0 (0)	0 (0)

N – No. of animals in the study group; n- No. of animals showing AEs

In the T1 group (Adjuvant control group), 3 out of 49 animals (6.1%) showed swellings at the site of injection during primary vaccination and in the T2 group (TSOL18 Vaccine group), 6 out of 48 vaccinated animals (12.5%) showed swellings at the site of injection during booster administration. These swellings resolved within 2-3 days. In T1 (Adjuvant control group), 3 animals out of 49 animals (6.1%) showed fever on primary vaccination; whereas in T2 (TSOL18 Vaccine group), 4 out of 48 animals (8.33 %) on primary vaccination and 2 out of 48 vaccinated animals (4.2%) on booster vaccination exhibited fever.

After TSOL 18 vaccine booster dose administration, 5 out of 48 vaccinated animals experienced mild to moderate anaphylactic reactions. The pigs showed the symptoms of collapse, convulsion, shallow and rapid breathing and hyperemia of extremities. The symptoms resolved within 5-10 minutes with sprinkling of water on the animals and without any medical intervention.

Serious Adverse Reactions: No Serious adverse reactions were recorded in the study.

DISCUSSION AND CONCLUSION

A study was undertaken to evaluate the efficacy and safety of recombinant porcine cysticercosis vaccine (TSOL18 vaccine) administered through intramuscular route in pigs. The vaccine has been manufactured by Indian Immunologicals Limited.

In efficacy evaluation of the present study, the Geometric Mean TSOL 18 antibody titres in the vaccinated animals increased by 20 DPV and reached peak by 35 DPV compared to day -1 geometric mean titres. With regard to seroprotection, 39.58% of the vaccinated animals showed seroprotection on 20 DPV compared to day -1, when the animals were all seronegative. By 35 DPV, 97.92 % of the vaccinated animals were seroprotected following booster dose administration of the TSOL 18 vaccine on day 21, indicating the booster effect. These findings are in agreement with the results obtained in an immunogenicity study conducted by University of Zaragoza, Spain in which the animals of approximately 10 weeks age, which received two doses of vaccine developed titres ≥500.

The present study results are similar to the results obtained in another study conducted at College of Veterinary Medicine, Universidad Nacional Mayor de San Marcos, Peru. The vaccinated animals given two doses of *Pichia* TSOL18 + ISA 206 vaccine developed adequate titres (≥500) of TSOL18-specific IgG. The results confirm that two doses of TSOL18 vaccine given three weeks apart were safe and effective.

In safety evaluation, there were some mild local and systemic reactions like swellings at the site of injection or fever observed in some animals. However, these symptoms disappeared after 2-3 days without any treatment.

Similar symptoms were observed in a safety study conducted in pigs at University of Zaragoza, Spain. Following treatment administration, there was transient mild pyrexia which resolved within 24 hours and mild injection site reactions which resolved within 7 days.

Some animals showed mild to moderate anaphylactic reactions following booster dose administration of the vaccine which resolved within 5-10 minutes without any medical intervention. The possible cause for anaphylactic reactions could be individual animal response (idiosyncratic drug reaction) to the vaccine.

Anaphylactic reactions in vaccinated pigs have earlier been reported by some authors for other vaccines. Turnquist *et al* (1993) reported a case of anaphylactic or anaphylactoid type reaction. Six hundred pigs were immunized with two doses of bacterins, reactions followed the second vaccination. Reactions were seen within 1.5 hours. Thirty six animals were found dead the next day. The signs included: hyperventilation, vomiting, urticaria, loose faeces. However, in the present study, no such findings were observed as only mild reactions occurred without any serious adverse reactions or deaths.

Overall, the efficacy and safety study of recombinant porcine cysticercosis vaccine (TSOL18 vaccine) in pigs revealed that the vaccine is efficacious in eliciting excellent antibody response and seroprotection in almost all the experimental animals during the trial period of 49 days when administered by intramuscular route. The vaccine is safe barring some mild local and moderate systemic reactions, which resolved on their own without any medication.

It can be concluded that the recombinant porcine cysticercosis vaccine (TSOL18 vaccine) manufactured by Indian Immunologicals Limited is efficacious and safe in pigs aged 2 months up to 2 years when administered through intramuscular route under field conditions.

LIST OF REFERENCES

Armando, E.G., Charles, G. G., Dylan, B., Robert, H. G., Victor, C. W. T., Hector, H. G., Manuela V., Marshall W.L.,2005. Short Report: Vaccination of Pigs to control Human Neurocysticercosis. Am. J. Trop. Med. Hyg., 72(6), 2005, pp. 837–839

Assana, E., Gauci, C.G., Kyngdon, C.T., Zoli, A.P., Dorny, P., Geerts, S., Lightowlers, M.W., 2010. Antibody responses to the host-protective *Taenia solium* oncosphere protein TSOL18 in pigs are directed against conformational epitopes. Parasite Immunol. 2010 32(6):399-405.

Cai, X., Yuan, G., Zheng, Y., Luo, X., Zhang, S., Ding, J., Jing, Z., Lu, C., 2008. Effective production and purification of the glycosylated TSOL18 antigen, which is protective against pig cysticercosis. Infect. Immun. 76,767–770.

Gonzalez, A.E., Gauci, C.G., Barber, D., Gilman, R.H., Tsang, V.C., Garcia, H.H., Verastegui, M., Lightowlers, M.W., 2005. Vaccination of pigs to control human neurocysticercosis. Am. J. Trop. Med. Hyg. 72, 837–839.

Lawson, J.R., Gemmell, M.A., 1990. Transmission of taeniid tapeworm eggs via blowflies to intermediate hosts. Parasitology, 100: 143-146.

Lightowlers, M.W., 1999. Eradication of *Taenia solium* cysticercosis: a role for vaccination of pigs. Int. J. Parasitol. 29, 811–817.

Lightowlers, M.W., 2010. Eradication of *Taenia solium* cysticercosis: a role for vaccination of pigs. Int. J. Parasitol. 40, 1183–1192.

OIE Terrestrial Manual 2014.

Turnquist et al 1993 Naturally occurring systemic anaphylactic and anaphylactoid reactions in four groups of pigs injected with commercially available bacterins Susan E. Turnquist, Guy Bouchard, John R. Fischer J Vet Diagn Invest 5:103-105 (1993)

World Health Organization. Media centre: Taeniasis/cysticercosis: Fact sheet no. 376. http://www.who.int/mediacentre/factsheets/fs376/en/