PROCEEDINGS & ABSTRACTS

PORCINE MEDICINE
POSTER SESSION ABSTRACTS
HIGH PREVALENCE AND GENETIC VARIABILITY OF PORCINE ASTROVIRUSES IN PIGS ON BACKYARD HOLDINGS IN CROATIA

D. Brnić1, L. Jemeršić1, T. Keros1, J. Prpić1, T. Bedekovič1, I. Lojkić1, N. Krešić1, B. Roić1, A. Jungić1, V. Starešina2

1 Virology Department, Croatian Veterinary Institute, Zagreb, Croatia
2 Department of Microbiology and Infectious Diseases, Faculty of Veterinary Medicine, Zagreb, Croatia

Topic: 10. Porcine Medicine / Emerging and Re-Emerging Diseases

Astroviruses comprise a group of emerging, primarily enteropathogenic RNA viruses, distributed worldwide in numerous species of mammals and birds. Recently they have been acknowledged to have extra-intestinal implications in humans, mink and pigs. Porcine astrovirus (PoAstV) is a highly prevalent pathogen of domestic pigs. Its vast genetic diversity has been recognized, with strains being assorted into five distinct lineages, possibly representing individual species. In the present research we have collected 48 composite fecal samples originating from 11 small backyard pig holdings in Croatia. Samples were distributed over six age categories of healthy domestic pigs and were processed by using a pan astrovirus molecular approach. We report high astrovirus prevalence (83.3%) in pigs bred on backyard holdings, with all age categories being affected. Furthermore, PoAstV is present in all tested holdings. Molecular characterization and phylogenetic analysis of selected astrovirus strains indicate the presence of four PoAstV lineages, with lineage 3 detected for the first time outside of North America. Lineage 2 was found to be the most prevalent among PoAstV strains in Croatian domestic pigs. Additionally, representatives of lineage 2 and 4 suggest the presence of potential sub lineages. We conclude that PoAstV is a highly prevalent and potentially persistent enteropathogen in domestic pigs bred on backyard holdings in Croatia, but their clinical significance remains doubtful. By detecting the circulation of four potential lineages, our results support previous knowledge of the immense genetic variability of PoAstV. Furthermore, this is the first evidence of PoAstV lineage 3 circulation outside of North America.
Climatic factors associated with peripartum sows died for 4 weeks during hot and humid season or cold season

R. Iida, Y. Koketsu

School of Agriculture, Meiji University, Kawasaki, Japan


The objective of this study was to quantify associations between climatic factors and occurrences of death 4 weeks before or after farrowing. The study analyzed records of 93,837 female pigs entered into 98 commercial herds in Japan from 2003 to 2007. Average daily maximum (HT) and minimum temperature (LT) for the 4 weeks before due date and after farrowing were obtained from 21 weather stations, and were coordinated with respective performance data. Two-level mixed-effects models were applied to two separate datasets. One comprised females having due dates during the hot and humid season (Jun. to Sep.), and the other comprised females having due dates during the cold season (Dec. to Mar.). Overall, 1.4 and 2.8% of females died during the 4 weeks before and after farrowing, respectively. Mean (range) HT and LT were 28.7°C (13.4 to 39.8°C) and 1.6°C (-14.8 to 17.6°C), respectively. In the hot and humid season, deaths of parity 0-3 pregnant pigs increased by 1.27-1.52 times per 5°C increase in HT (P < 0.05). Deaths of mated parity 0-3 sows after farrowing increased by 1.32-1.39 times (P < 0.05). In the cold season, deaths of mated parity 4 and 5 sows before and after farrowing increased by 0.22 and 0.21%, respectively when LT decreased from 5°C to 0°C (P < 0.05). There was no association between deaths and HT or LT in any other parity group. Therefore, increased HT and decreased LT can increase the risk of female pig death during the peripartum period.
LOW RETENTION RATE BY PARITY 3 ASSOCIATED WITH GILT DEVELOPMENT, REPRODUCTIVE PERFORMANCE AND CULLING PATTERN OF SOWS IN SWINE HERDS

R. Iida, M. Kaneko, Y. Koketsu
School of Agriculture, Meiji University, Kawasaki, Japan


The objective of this study was to characterize swine herds having a low retention rate (LRR herds). Questionnaires were sent to 115 herds that use the same recording system. Data from 80 completed questionnaires were coordinated with the reproductive data for relevant individual females, which included records of 30,759 females and lifetime records of 26,254 females. Herds were categorized into LRR herds and ordinary herds on the basis of the lower 25th percentile of retention rate by parity 3. Two-level mixed-effects models were applied to the data. A cox proportional hazards model was used to compare hazards for culling in females by the herd group. Mean (± SEM) retention rates by parity 3 in LRR herds and ordinary herds were 56.1 ± 1.07 and 73.7 ± 1.02%, respectively. The proportion of females culled for reproductive failure by parity 3 in LRR herds was 67.3%, whereas in ordinary herds it was only 52.2%. Retention rate at parity 0 was lower in females that received restricted feeding as prepubertal gilts compared with those not on a restricted diet (P < 0.05). Additionally, culling hazards increased more rapidly from 20 weeks after first mating in females that received restricted feeding as prepubertal gilts than those not on a restricted diet (P < 0.05). In conclusion, females in LRR herds were at higher risk of culling due to reproductive failure from parity 0 to 3 than those in ordinary herds. Additionally, unrestricted feeding during prepubertal periods is recommended to improve sow longevity.
FACTORS ASSOCIATED WITH FARROWING CRATE UTILIZATION EFFICIENCY ON JAPANESE COMMERCIAL SWINE FARMS

Y. Koketsu1, R. Iida1, D. Polson2, G. Dial3

1 School of Agriculture, Meiji University, Kawasaki, Japan
2 Research and Development, Boehringer Ingelheim Vetmedica, Inc., Ames, United States
3 Swine group, Greenleaf Agribusiness Group LLC., Williams, United States


The objective of this study was to examine the within-farm variability of sow flow associated with low farrowing crate (FCRT) utilization efficiency. A questionnaire survey was undertaken to obtain information about FCRT utilization from 115 Japanese commercial swine farms. Questionnaire data from the 94 farms that responded were coordinated with the respective reproductive performance data. A set of two time-plot data for the numbers of lactating sows and serviced females in 12 four-weekly periods during a year was created for each farm to obtain the means and coefficients of variation (CV, %). Regression analysis was performed to quantify the association between the two measurements. Mean (± SEM) litters per FCRT per year and CVs of the number of lactating sows and serviced females in a year were 9.86 ± 1.28 litters, 10.58 ± 0.66% and 12.82 ± 0.79%, respectively. A higher number of litters per FCRT per year was associated with a lower CV% for the number of lactating sows per year (P < 0.05). The lower CV% for the number of lactating sows was associated with a lower CV% for the number of females served 16 weeks previously and a higher annual farrowing rate (P < 0.05). The higher number of litters per FCRT per year was associated with higher female inventories per FCRT (P < 0.05). In conclusion, higher FCRT utilization efficiency was related to lower within-farm variability of female pig flow. Also, improving FCRT utilization efficiency appeared to increase female inventories per FCRT.
**EPIDEMIOLOGICAL STUDY OF LAWSONIA INTRACELLULARIS OF SHIPMENT PIGS IN JAPAN**

M. Sueyoshi¹, A. Hirata², M. Tsurita², D. Miyayama², Y. Sasaki², R. Uemura²

¹ Center for Animal Diseases Control, University of Miyazaki, Miyazaki, Japan
² Veterinary Medical Science, University of Miyazaki, Miyazaki, Japan

**Topic:** 10. Porcine Medicine / Infectious Diseases

**Introduction** *Lawsonia intracellularis* is the main pathogen of porcine proliferative enteropathy (PPE). PPE occurs in pig production countries of the world. PPE causes a big economic loss to cause a growth delay. *L. intracellularis* is bacteria in the obligate cell, and the isolation in the agar medium is so difficult. The epidemiology investigation in Japan is not yet clear. In this study, we carried out the survey of *L. intracellularis* in Kyushu District of Japan. In Kyushu District, 34% of Japanese pigs are bred.

**Materials and method** The sera of 1,362 shipment pigs (3-30 heads/farm) of 96 farms of Kyushu District were examined. The sera of pigs carried in to the slaughterhouse of Miyazaki were collected. The pig was carried in from Miyazaki, Kumamoto, Oita, Fukuoka, Kagoshima, and Nagasaki. The indirect fluorescent antibody method (IFA) and ELISA (bioScreen Ileitis Antibody ELISA kit) were used for this examination.

**Result** The positive rate of *L. intracellularis* antibody of the farm was 100% (96/96) by IFA and ELISA. The positive rate of *L. intracellularis* antibody of the pig was 88% (1,202/1,362) by IFA, and 87% (1,184/1,362) by ELISA.

**Discussion** *L. intracellularis* spread over the pig farms of Kyushu District widely. The disclosure rate of PPE in the meat inspection center is reported with 0.27% in Japan. Therefore, a lot of pigs were regarded as a subclinical infection in Japan. Based on these, it was suggested that the inapparent economic loss by growth delays in the *L. intracellularis* infection occurred.
INHIBITION REPLICATION OF PRRSV ON MARC 145 CELL CULTURE USING GLYCYRRHIZINIC ACID AQUEOUS SOLUTIONS

Z. Urban\textsuperscript{1}, A. Jiménez\textsuperscript{2}, S. Mendoza\textsuperscript{2}, H. Ramírez\textsuperscript{3}, A. Ciprián\textsuperscript{2}, D. Quintanar\textsuperscript{1}

\textsuperscript{1} Laboratorio de Investigación y Posgrado en Tecnología Farmacéutica, Facultad de Estudios Superiores Cuautitlan-UNAM, Cuautitlan Izcalli, Estado de México, Mexico
\textsuperscript{2} Laboratorio de Virología y Microbiología de Enfermedades Respiratorias del Cerdo, Facultad de Estudios Superiores Cuautitlan-UNAM, Cuautitlan Izcalli, Estado de México, Mexico
\textsuperscript{3} Laboratorio de Virología, Facultad de Medicina Veterinaria y Zootecnia-UNAM, Distrito Federal, Mexico

\textbf{Topic:} 10. Porcine Medicine / Infectious Diseases

Glycyrrhizinic acid (GA), is the most important saponin of licorice root which has been used as anti-inflammatory, anti-ulcer, anti-tumor, anti-viral, etc., for many years. It has been demonstrated its antiviral activity against several viruses, e.g. herpes simplex virus, Newcastle virus, Epstein-Barr virus, Hepatitis virus, SARS-coronavirus, flaviviruses, influenza A virus, pseudorabies virus and human immunodeficiency virus.

The aim of this research was to study the effect of GA solutions on uninfected and PRRS virus-infected cells in culture. Solutions of GA at different concentrations (1-30 mg/ml and 0.1-0.9 mg/ml) were tested on MARC 145 cells. Trypan blue dye exclusion and MTT assay were performed in order to obtain the GA concentration which reduced cell viability to 50% (CC\textsubscript{50}) and the concentration that inhibited the cytopathic effect to 50% (EC\textsubscript{50}). The viral titer decreased two logarithms compared to the titer obtained in the first viral titration without GA treatment. EC\textsubscript{50} (0.5 mg/ml) was evaluated against viral dilutions from $10^1$ to $10^6$ TCID showing an inhibition of the cytophatic effect for all tested concentrations. Although the selectivity index (CC\textsubscript{50}/EC\textsubscript{50}) was relatively low (1.73), GA showed reduction of PRRS replication \textit{in vitro}. The results are very important because they represent the first step to establish an option to treat PRRS infections.
MOLECULAR CHARACTERIZATION OF SPIKE, ORF3 AND MEMBRANE GENE SEQUENCES OF RECENT PORCINE EPIDEMIC DIARRHEA VIRUSES IN KOREA


Animal Disease Diagnostic Division, Animal and Plant Quarantine Agency, Anyang, South Korea

Topic: 10. Porcine Medicine / Infectious Diseases

Porcine epidemic diarrhea virus (PEDV) is the etiological agent of enteropathogenic diarrhea in piglets of Korea. PEDV has been consistently detected in sporadic areas since 1993. To better understand the extent of genetic variation of PEDV in Korea, 50 PEDV isolates from diarrhea and intestines from various pig farms from different provinces in Korea between the years 1998 and 2013 were sequenced and compared for spike, ORF3 and membrane with reference strains reported from other countries.

Phylogenetic analysis of spike region revealed that most Korean isolates were divided into two groups (G1 and G2) with two subgroups (G2-1, -2). An analysis of the homology of spike genes showed genetic diversity, not only among Korean isolates, was 91.9-99.8% and 90.4-99.7% homology at the nucleotide (nt) and amino acid (aa), respectively, but also between Korean and other countries including reference strains was 92.2-99.3% nt and 90.8-99.2% aa homology. Furthermore, sequence homology in spike genes showed a tendency to be relatively low in years. The Korean isolates showed 92.8-98.3% sequence homology with sequences of vaccine strains used in Korea, which corresponded to 91.0-98.0% homology at the amino acid level. In the ORF3 region, cell adaptation strains and some field strains showed large deletion. Since the deleted regions were varied in those strains, the deletion of ORF3 in any place might indicate the cell adaptation of field strain, not a reduction of the virulence. The membrane sequences appeared more conserved than other regions. This report may contribute to understanding the nature of the current porcine epidemic diarrhea in Korea.
ANTIMICROBIAL ACTIVITY OF STALOSAN® F AGAINST BRACHYSPIRA HYODYSENTERIAE USING IN VITRO TEST APPLICATIONS

V. Vandenbroucke¹, F. De Jaeger²

¹ Department of Bacteriology, Dierengezondheidszorg Vlaanderen, Torhout, Belgium
² R&D, INVE België nv, Baasrode, Belgium

Topic: 10. Porcine Medicine / Infectious Diseases

Brachyspira hyodysenteriae is the etiological agent of swine dysentery, a major threat in the pig industry with severe economic impact. In the prevention, management procedures and hygienic measures are of great importance. Stalosan® F is a chemical disinfectant indicated for use in animal housing to reduce the number of microorganisms. The effectiveness of Stalosan® F on Lawsonia intracellularis has been described. However, no data are available on the effect of Stalosan® F on B. hyodysenteriae.

In this study the effect of Stalosan® F on the survival of B. hyodysenteriae was examined using in vitro test assays. The typical swarming aspect of B. hyodysenteriae combined with strong hemolysis on blood agar plates was used to examine the effect of Stalosan® F on the growth of B. hyodysenteriae. In a first experiment Stalosan® F was evenly applied on selective culture media previously inoculated with B. hyodysenteriae, after which the growth of B. hyodysenteriae was examined. In a second experiment, Stalosan® F was added to dilutions of B. hyodysenteriae. The bacterial suspensions were incubated for 6h at room temperature after which they were inoculated on selective culture media. In both experiments, application rates of Stalosan® F corresponding to 50g/m² were used.

When exposed to Stalosan® F, an inhibition of the growth of B. hyodysenteriae was seen in both experiments. These in vitro results indicate that Stalosan® F could be considered as an useful disinfectant in the framework of hygiene and sanitation practices to help control swine dysentery.
CONFIRMATION BY POLYMERASE CHAIN REACTION (PCR) OF SALMONELLA SPP. IN PIGS

R. P. Schocken-Iturrino

State Public University

**Topic:** 10. Porcine Medicine / Infectious Diseases

The identification of *Salmonella* spp. in pigs is considered an economic problem and of public health. Pigs are asymptomatic carriers, hindering the precise diagnosis of infection and facilitating the dissemination of the agent. The objective was to confirm *Salmonella* spp. in pigs using PCR in rectal swabs from 84 animals coming from slaughterhouse in the region of Ribeirão Preto, SP, Brazil. The swabs were placed in tubes with peptone water 0.1%, incubated at 42 °C for 24 hours. Aliquots 1 mL were transferred to tubes with Selenite and Rappaport broth incubated for 24 hours at 37 °C. Both were streaked onto MacConkey and XLD agar plates, incubated in the same conditions. For confirmation, bacterial DNA was extracted using Marmur (1961) method. The PCR for the gene *invA* (Oliveira *et al.*, 2002) followed one cycle of amplification (95 °C for 5 minutes), 35 cycles (94 °C for 1 minute, 60 °C for 1 minute and 72 °C for 1.5 minutes), and 1 cycle (72 °C for 10 minutes). Eighteen samples (21.43%) were positive for *Salmonella* spp.. Detection of *Salmonella* spp. in pigs is considered a risk factor for the industry, may cause contamination during slaughter, evisceration and cutting carcasses, since the manufacturing process is fast and there is no possibility of and effective control of this agent. It was concluded that the PCR technique is effective in confirming *Salmonella* spp. using rectal swabs in pigs, and may avoid health hazard for consumers.
PRELIMINARY STUDY OF THE ANTIVIRAL ACTIVITY OF GLYCIRRHICIC ACID ON THE AUJESZKY DISEASE VIRUS

A. Jimenez¹, Z. Urban², H. Ramirez³, E. Hernandez¹, D. Quintanar², S. Mendoza¹

¹ Laboratorio de Virología y Microbiología de Enfermedades Respiratorias del Cerdo, Facultad de Estudios Superiores Cuautitlan, UNAM, Estado de Mexico, Mexico
² Laboratorio de Investigación y Posgrado en Tecnología Farmaceutica, Facultad de Estudios Superiores Cuautitlan, UNAM, Estado de Mexico, Mexico
³ Laboratorio de Virologia, Facultad de Medicina Veterinaria y Zootecnia, UNAM, Distrito Federal, Mexico

Topic: 10. Porcine Medicine / Infectious Diseases

The importance of eradicating Aujeszky’s disease lies on three aspects: Sanitary. Due the persistence of the virus for prolonged periods after an outbreak. Economic, as it causes important economic losses, as well as secondary bacterial infections and increased expenses caused by treatments and vaccination; and Commercial, since the presence of the disease limits the possibility of exporting pigs or pig carcasses’. The objective was to evaluate *in vitro* glycyrrhic acid (GA) in Aujeszky’s Disease Virus (ADV) infected cells. ADV was titrated in MDBK cell line. Concentrations of GA from 0.1mg/ml to 0.9mg/ml were prepared and tested for cytopathic effect. The monolayers were infected with 1 LD₅₀ of ADV, 24 hours after infection; the cells were treated with GA, and were evaluated by microscopy at different time intervals post-treatment. The maximum non-toxic dose of GA was 0.8mg/ml. The viral titer was 10⁵.⁴⁶ while in the infected cultures treated with the GA the viral titer was lower, 10³.²⁴. With constant virus, the monolayers treated with the drug survived the 72 hours without change. Both the reduction of titer and the protection of the monolayers by the GA indicate an inhibitory effect; however the mechanism of virus inhibition was not determined at this time. In this trial, the inhibitory effect was observed from 0.1 mg/ml. With the Trypan blue and MTT assays, the cell viability was higher with the GA. The GA had an inhibitory effect on the ADV both in a titration and against constant virus.
SANITARY QUALITY OF THE NATIVE BREEDS OF PIGS IN FAMILY AGRICULTURE FROM FEDERAL DISTRICT, BRAZIL.

G. Rocha, R. Camargo, R. Lourinho, S. Perecmanis

Faculty of Agriculture and Veterinary Medicine, Universidade de Brasília, Brasilia, Brazil

Topic: 10. Porcine Medicine / Zoonotic Diseases

Increasing worldwide ecological awareness highlights the importance of biodiversity and the preservation of domestic animal breeds at risk of extinction. The domestic pig has a prominent place in family subsistence throughout Brazil since colonial times. The project "Breeding and Conservation of National Pigs", was carried out by a partnership between the Brazilian Agricultural Research Corporation (Embrapa), Enterprise Technical Assistance and Rural Extension of Federal District (Emater) and the University of Brasília (UnB). The aim was to promote preservation of genetic heritage and the social value of naturalized pigs in family farming. Along a year, family farms of rural centers of the Federal District were surveyed and 164 of swine fecal samples were processed to evaluate the sanitary quality of the breeding. Approximately 94% of properties have not sewage, 88% have no sanitized water, 63% of water was obtained from wells and only 10% of treated water was used for facilities cleaning. Among 27% farmers who used kitchen waste to feed pigs 97% do not boiled it to offer the animals. Pathogenic *Escherichia coli* samples, *Salmonella Cerro*, *Salmonella Madelia* and *Salmonella Schwarzengrund* samples were isolated. The most prevalent parasites were: coccidia (Apicomplexa), *Balantidium coli* (19% each one), strongyles helminthes (Strongylida - 18%), *Ascaris suis* (16%) and *Strongyloides ransomi* (14%). The lack of hygiene, the incorrect destination of human and animal wastes, the absence of preventive measures and also an adequate technical guidance and information were the main factors for the occurrence of microbial and parasitic infections diagnosed in this study.
SERO-PREVALENCE OF PORCINE PSEUDORABIES IN TAIWAN IN 2011

C.Y. Yang¹, P.C. Yang¹, M.T. Chiou²

¹ Division of Animal Medicine, Animal Technology Institute Taiwan, Chunan, Miaoli, Taiwan
² Department of Veterinary Medicine, National Pingtung University of Science and Technology, Pingtung, Taiwan

Topic: 10. Porcine Medicine / Infectious Diseases

Pseudorabies (PR) virus is an un-eradicated swine disease in Taiwan, causing agent of respiratory and reproduction failure disease. PRV inactivated vaccine and glycoprotein E-deleted vaccine are major vaccine types. In 2011, a survey was conducted to estimate herd prevalence of Pseudorabies. A total of 273 randomly selected herds were included in the epidemiology survey of PRV was examined using stratified random sampling method. The sample size of 15 pigs per herd was tested that is intended to detect PRV at 95% confidence interval with a 20% infection rate. Pigs over 5 month of age were randomly selected and bled for testing Glycoprotein E (gE) Enzyme linked immunosorbent assay (ELISA). The overall herd sera prevalence of these swine herds was 38.9% (106/273). Among 124 swine herds vaccinated with PRV gE-deleted vaccine, gE antibodies were detected in 59 herds. Among 118 unvaccinated herds, seroprevalence of PRV gE antibody is 13.6%, which could be the real field infection rate of PRV. The field seroprevalence 13.6% is lower than first reported 96% in 1994 and another reported 37.7% in 2008. In this study, 16 out of 273 pig herds is vaccinated with PRV inactivated vaccine, it is helpless to differentiate antibodies between vaccinated pig and infected pigs. In the low prevalence stage, vaccine chooses is a key step to eradicate PRV.

Table 1. Seroprevalence of tested pigs in Taiwan

<table>
<thead>
<tr>
<th>Vaccination status</th>
<th>No. of sero-positive herds</th>
<th>No. of sero-negative herds</th>
<th>No. of herds tested</th>
<th>Herd prevalence, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unvaccinated</td>
<td>16</td>
<td>102</td>
<td>118</td>
<td>13.6</td>
</tr>
<tr>
<td>Vaccinated with PRV gE(-)</td>
<td>59</td>
<td>65</td>
<td>124</td>
<td>47.6</td>
</tr>
<tr>
<td>Vaccinated but vaccine type</td>
<td>16</td>
<td>0</td>
<td>16</td>
<td>100</td>
</tr>
<tr>
<td>Vacinnated but vaccine type</td>
<td>15</td>
<td>0</td>
<td>15</td>
<td>100</td>
</tr>
<tr>
<td>Total</td>
<td>106</td>
<td>167</td>
<td>273</td>
<td>38.9</td>
</tr>
</tbody>
</table>
ID: 408

EFFECTS OF ZEARALENONE ON GROWTH, SERUM BIOCHEMISTRY AND STEROIDOGENESIS IN MALE PIGLETS

Y. H. Cheng¹, Z. W. Liao², D. W. Chen²

¹ Biotech and Animal science, National I-Lan University, I-Lan, Taiwan
² Research and Development Section, Liferainbow Biotech. Co., Ltd, I-Lan, Taiwan


Zearalenone (ZEA) is an estrogenic mycotoxin biosynthesized by Fusarium spp. The toxic effects are associated with hyperestrogenism and multi-reproductive disorders in animals. Moreover, swine is the most sensitive animal of the domestic livestock and poultry to ZEA. The aim of this study was try to examine after feeding diets which contain defined levels zearalenone (0, 0.5 and 1 ppm) in weaned piglets for 3 weeks. Our results showed that there are no significant effects on the growth, testes weight and serum biochemistry in weaned piglets while the diets containing 1 ppm ZEA (P > 0.05). However, the transcription level of the key steroidogenic enzymes including 3β-HSD and P450scc were up-regulated by ZEA, and in dose-dependent manner (P < 0.01 and 0.05, respectively), but no significant expression was observed in StAR and aromatase. In conclusion, it can be assumed that the ZEA concentration affects gonad’s gene expression arised in progestagens superfamily but did not involve in interfering biosynthesis of downstream sex hormone as testosterone, further testicular proteomics assays will be performed.
EEFFECTS OF CORDYCEPS MILITARIS APPLIED IN ANTI-INFLAMMATION AND ANTI-PRRSV REPLICATION IN PIGS.

Y. H. Cheng¹, Z. K. Wang¹, Z. W. Liao², C. K. Yang²

¹ Biotech and Animal Science, National I-Lan University, I-Lan, Taiwan
² Research and Development Section, Liferainbow, Co. Ltd., I-Lan, Taiwan

Topic: 10. Porcine Medicine / Infectious Diseases

*Cordyceps militaris* is a rare and exotic Chinese medicinal mushroom. Recently, it was reported that the metabolites such as cordycepin in fermentative products of *Cordyceps militaris* showed an anti-inflammatory activity. The objectives of this study were to extract cordycepin from *Cordyceps militaris* solid-phase fermentative, and determine the anti-inflammation mechanism, PRRS virus replication by hot water extract from *Cordyceps militaris* (CMHW) in porcine alveolar macrophage (PAM). Our results demonstrate that CMHW inhibit COX-2 expression by block the MAPK and IKK cascades via suppress the phosphorylation in JNK, p38 and IkB signal pathway, ultimately inactivate transcription factor of AP-1 and NFkB. In another stimulation experiment, PRRSV infection alone or combined with bacterial infection in PAM model, CMHW could significantly reduce pro-inflammatory cytokines expression, such as, TNF-α, IL-1β, IL-6, and inhibit PRRSV replication. CMHW elucidate inhibiting the transcription factor activation by suppressing upstream signaling events, and it could be applied in anti-virus and anti-bacterial infection application in pigs.
ESCHERICHIA COLI ENTEROTOXINS GENES DETECTION IN SWINE COLIBACILLOSIS OUTBREAK FROM A BRAZILIAN FARM

S. Perecmanis¹, G. C. da Rocha¹, V. O. Drummond¹, H. H. de Andrade¹, M. L. Tokatjian¹, L. F. I. de Souza¹, C. G. Freitas², R. T. Moreira², E. F. Gaudencio¹, A.S. Silva¹

¹ Faculty of Agronomy and Veterinary Medicine, Brasilia University, Brasilia, Brazil
² IFB, Brasilia Federal Institute, Brasilia, Brazil

Topic: 10. Porcine Medicine / Infectious Diseases

Colibacillosis is a major cause of illness and death in pigs and presents itself in two clinical forms, the neonatal colibacillosis and the post-weaning coliform gastroenteritis. The following report occurred on a farm in the Federal District, Brazil in a batch of 8 pigs aged between 1 month and 2 months old, with characteristics of the post- weaning colibacillosis after the start of feeding with swine ration. All 8 animals in batch had diarrhea, watery or pasty. Swine feces samples were collected from seven animals and one animal was sent for necropsy after death. Were isolated colonies of E. coli in all samples and submitted them to polymerase chain reaction to detect enterotoxin genes. The thermostable B toxin gene was detect in two strains and intimin gene into other two strains. One strain of E. coli positive with was positive for the intimin gene was also positive for Shiga like toxin 2e. In parasitological examinations were identified oocysts (+) in one animal, demonstrating a concomitant infection with E. coli. The necropsied animal showed microscopic findings compatible with sepsis and colibacillosis lesions and granulomatous abscesses including serous various organs. The clinical and laboratory diagnosis associated with the results of the necropsy clearly showed the condition as colibacillosis.
GENETIC DIVERSITY OF TORQUE TENO SUS VIRUS IN PIGS FROM BRAZIL


Department of Preventive Veterinary Medicine and Animal Health, College of Veterinary Medicine, University of São Paulo, São Paulo, Brazil

Topic: 10. Porcine Medicine / Infectious Diseases

Torque teno sus virus (TTSuV) is genetically very distinct virus infecting pigs and boars around the world. TTSuV is classified according to ICTV (2012) within the Anelloviridae family, genus Iotatorquevirus with two species described: Torque teno sus virus 1a (TTSuV1a) and Torque teno sus virus 1b (TTSuV1b); and another genus Kappatorquevirus with even now one species described: Torque teno sus virus k2 (TTSuVk2). The genome of TTSuV contains three open reading whereas ORF1 is the largest codify the capside protein and rolling circle replication motif (ORF1). Lung samples from five different pigs from Brazil herds had 1929 bp (TTSuV1) and 1890 bp (TTSuVk2) of the DNA amplified, cloned into pTZ57R/T vector and sequenced. The aim of this study was to investigate the genetic diversity of the virus in samples of pigs. Aminoacids sequences from nine TTSuV1 and 5 TTSuVk2 clones were recovered and aligned together with reference sequences available on Genbank (Figure 1 and 2). The mean divergence with TTSuV1 sequences was 66% and 26% with TTSuVk2. Three clones were classified within TTSuV1a and six within TTSuV1b species. Sample AM66 showed to be infected with both TTSuV1 species and different genotypes were found within TTSuV1b species. Likewise sample AM65 showed to be infected with both TTSuV1 species and further with different genotypes within TTSuVk2. There was no correlation between infection with certain species or genus of TTSuV and the presence of organ lesion. In conclusion both TTSuV genus and species are disseminated in pig herds exhibiting great genetic diversity.

Figure 1. Aminoacid phylogenetic tree of the TTSuV1 ORF1 aminoacid sequences constructed by neighbour-joining algorithm and the K21 model with 1,000 bootstrap replicates. AB075001 TTSuV 1a-TTSuV1 and AK282899 TTSuV 1b represents the prototypes of TTSuV1a and TTSuV1b species respectively. Samples recovered in this study are marked with the symbol F. AM65 clones C2 and C3; AM66 clones C1, C2, C3, C4 and C5; AMF1 clone C2; AME36 clone C2.
Figure 2. Amino acid phylogenetic tree of the TTSuV2/3 ORF1 amino acid sequences constructed by neighbour-joining algorithm and the HKY model with 1,000 bootstrap replicates. AVB23992 TTSuV2p represents the prototype of TTSuV2 genus. Samples recovered in this study are marked with the symbol ▼. AM85 clones C2 and C4, AM89 clones C2, AM14 clones C2, AM219 clone C4.
COMPARISON OF THE TTSUV1 AND TTSUVK2 VIRAL LOAD IN PCV2 VACCINATED AND NON VACCINATED PIGS


Department of Preventive Veterinary Medicine and Animal Health, College of Veterinary Medicine, University of São Paulo, São Paulo, Brazil

Topic: 10. Porcine Medicine / Infectious Diseases

Torque teno sus virus (TTSuV) is classified according to ICTV (2012) within Anelloviridae family, genus Iotatorquevirus (TTSuV1) and Kapatorquevirus (TTSuVk2). The pathogenesis of TTSuV nowadays is not clear but both of them were linked to disease or responsible for triggering clinical signs of porcine circovirus associated diseases (PCVAD). An increased TTSuV prevalence was described in porcine respiratory disease complex (PRDC) in interstitial pneumonia and broncho-interstitial pneumonia affected pigs. The same way an increase of TTSuV viral load was related in pigs with PCVA. Since vaccination against PCV2 is able to decrease viral load and hence immunosuppression and clinical disease onset, the same hypothesis was tested for TTSuV1 and TTSuVk2. Two hundred and fifteen lung samples had the TTSuV1 and TTSuVk2 DNA quantified by real time PCR technique with primers already described in the literature. Statistical analysis was performed based on nonparametric statistics, using the Mann–Whitney U-test. The median viral loads to vaccinated groups were 6.9x10^5, 1.3x10^4 and 7.7x10^5 while for non-vaccinated were 1.3x10^6, 4.3x10^4 and 2.4x10^6 for TTSuV1, TTSuVk2 and TTSuV1+TTSuVk2 respectively (Figure 1). There was no statistically significant difference between TTSuV viral loads from PCV2 vaccinated and non-vaccinated animals; TTSuV1 (p=0.58), TTSuV2 (p=0.49) and coinfection (p=0.54). Vaccination against PCV2 showed no influence on the dynamics of TTSuV infection.

Fig1: Median viral load of TTSuV1, TTSuVk2 and TTSuV1+TTSuVk2.