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ABSTRACT

The porcine circovirus type 2 and the porcine reproductive and respiratory syndrome virus are the major determinant of the swine respiratory disease complex (PRDC-Porcine respiratory disease complex).

The porcine respiratory disease complex (PRDC) is a multifactorial syndrome that implies allegedly involvement of many etiological agents. Therefore, early detection and prevention of co-infections are important aspects in managing PRDC. The swine youth is the most severely affected given the morbidity can be up to 70%. The porcine circovirus type 2 and PRRS virus are the main etiological agents of PRDC, but they can be adjoined by the Swine influenza virus (SIV), Porcine respiratory coronavirus, *Mycoplasma spp.*, *Actinobacillus pleuropneumoniae*, *Streptococcus suis* and *Pasteurellas pp.*

The paper is made of 265 pages and it is divided into two parts (according to required criteria) and eight chapters.

The first part (Chapters I and II) delivers the current level of knowledge and data from the international bibliography on porcine circovirus type 2 and on the porcine reproductive and respiratory syndrome virus.

Part two (chapters III, IV, V, VI, VII, VIII and IX) is dedicated to own research and each chapter is structured into sub-chapters that comprises the materials and methods used, obtained results and their debate and partial conclusions.

Chapter III present the aim and the objectives of PhD thesis.

Chapter IX contains the main conclusions that have been drawn following the extensive research and study of recommendations to farmers; and the bibliography includes 423 references.

The first part of the doctorate thesis is a summary of the data currently available in the international literature as respect to porcine circovirus type 2 infection and porcine reproductive and respiratory syndrome virus.

Chapter I is a bibliographic study on the results of national and international research regarding the infection with porcine circovirus type 2, with reference to the history and etiology of the infection, epidemiological characteristics of the virus, pathogenesis of circoviral infection,



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PCV2 associated disease complex (PCVAD), co-infections, laboratory diagnosis, prevention and control of diseases associated to PCV2 infection, surveillance and control measures as well as the data on the presence of the porcine circovirus in wild boar.

Chapter II contains bibliographical data on the porcine reproductive and respiratory syndrome virus with reference to its historical background, etiology, epidemiological characteristics and PRRS pathogenesis, clinical and morpho-pathological picture of the infection, diagnosis, prevention and control of the infection applied measures.

The premise of carrying out the research was to conduct epidemiological investigations on the swine circovirus and on the porcine reproductive and respiratory syndrome to determine the presence and the prevalence of PCV2 and PRRSV in pig livestock from Moldavia and Southern Romania and the presence of clinical manifestations was not a selection criteria of the herds. It has been pursued whether is possible to identify the two infections in pig herds which haven't shown any clinical signs for any of the two diseases, or on the contrary to which extent PCV2 is found in a PRRSV outbreak and vice versa.

It was also interesting to discover to what extent the persistence of PRRS infection is influenced by the strict compliance with the measures to combat the syndrome.

In order to achieve these goals there were performed serological tests using ELISA method; it was also effectuated detection of PCV2 and PRRSV viral antigens by PCR reaction (Real-Time, classic and nested) and subsequently there were studied the sequences obtained and their phylogenetic analysis.

In the second part of the study, organised in seven chapters, the research was focused on seroprevalence of PCV2 infection and anatomoclinical aspects in pigs (chapter IV), on seroprevalence of PRRSV infection and anatomoclinical aspects in pigs (Chapter V), on detection of viral antigens of the porcine circovirus and porcine reproductive and respiratory syndrome (chapter VI), on histopathological diagnosis in porcine circovirus and porcine reproductive and respiratory syndrome (chapter VII) and on correlation of infections caused by PCV2 and PRRSV (chapter VIII).

In chapter IX are listed the main conclusions drawn from the results consecutive to clinical, morpho-pathological and laboratory examinations used in diagnosing both infections.



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The results of investigations on the seroprevalence of PCV2 infection (Chapter IV) register differences in each livestock studied, but it was observed a high prevalence (65.68%) of IgG antibodies in comparison with IgM antibodies ratio which were detected in a much lower percentage (4.22%). The positivity ratio at the end of Real-Time reaction for the detection of PCV2 was of 35.30%. Following clinical and necropsy examinations, it was found that the lack of clinical signs and lesions do not exclude the possibility of diminished numbers of antibodies and/ or antigen detection in clinically healthy animals but PCV2 virus carriers, which are responsible for the transmission and spread of the PCV2 infection.

The results of investigations on the seroprevalence of PRRSV infection (Chapter V) showed a reduced presence of PRRSV infection as a small percentage of anti-PRRSV antibodies detected in the serum (2.60%) and of viral antigen one (8.32%). It was proven that PRRSV virus may be present in a herd to which no eradication measure was applied at the time of an outbreak diagnosis both by clinical and unapparent means. As in PCV2 infection it was proven that the lack of clinical signs and lesions picture do not exclude the possibility of diminished numbers of antibodies and/or antigen detection in clinically healthy animals but virus carriers, which are responsible for the transmission and spread of the PRRSV infection.

The research on the detection of viral antigens in porcine circovirus and porcine reproductive and respiratory syndrome (chapter VI) pursued the identification of viral antigens by PCR amplification of the samples corresponding to positive results after serological testing. Analysing the results it has been noticed an increased sensitivity of Real-Time PCR technique compared to the classical one for both infections. In a livestock clinically healthy it was diagnosed PCV2 subclinical infection and persistent infection with PRRSV by classical PCR amplification.

For the PCV2 isolated sequences following investigations it was observed the interrelation from phylogenetic point of view with a strain isolated in China, respectively with a PCV2 strain isolated from the same area in wild boar. In PRRSV sequences case it was noticed their interrelation with isolated strains from the same area, in domestic pigs.

The histopathological examination highlighted specific lesions to chronic form for both infections, confirmed serologically by Real-Time PCR, too to a herd which developed a PRRSV



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episode and eradication measures were not followed at the time of the outbreak discovery (chapter VII).

The correlation of infection of porcine circovirus type 2 and of porcine reproductive and respiratory syndrome virus is tackled in Chapter VIII and the most interesting aspect is the presence of double infection with PCV2 and PRRSV in a clinically healthy livestock which is explained in PCV2 case by a sub-clinical infection (in which specific antibodies have been identified concomitantly with antigen detection in tissues), and a PRRSV persistent infection which caused only a weak response of neutralizing antibodies and lack of virus elimination from tissues during ample viral replication.

The double infection with PCV2 and PRRSV was also highlighted in an outbreak of PRRSV and possibly the etiologic agent to have acted first to have been PCV2 due to its immunosuppressive effect. In conclusion, this double PCV2 and PRRSV infection can be found in both livestock which shows clinical manifestation and unapparent ones; and by conducting regular tests (Real-Time PCR) for the two infections contributes to the control of new episodes outbreaks.