EVALUATION OF PORCINE IMMUNE RESPONSES AMONG DIFFERENT GENETIC GROUPS

JEEBA K. GEORGE

Thesis submitted in partial fulfilment of the requirement for the degree of

Master of Veterinary Science

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University, Thrissur

2005

Department of Animal Breeding and Genetics
COLLEGE OF VETERINARY AND ANIMAL SCIENCES
MANNUTHY, THRISSUR-680651
KERALA, INDIA

DECLARATION

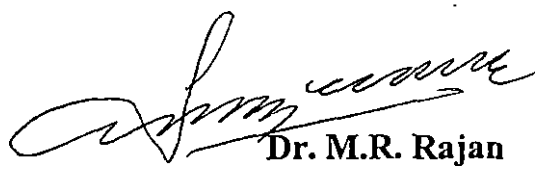
I hereby declare that this thesis entitled **"EVALUATION OF PORCINE IMMUNE RESPONSES AMONG DIFFERENT GENETIC GROUPS"** *is a bonafide record of research work done by me during the course* of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Mannuthy
19.08.05

JEEBA K. GEORGE

CERTIFICATE

Certified that the thesis entitled **"EVALUATION OF PORCINE IMMUNE RESPONSES AMONG DIFFERENT GENETIC GROUPS"** is a record of research work done independently by **Dr. Jeeba K. George**, under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.



Dr. M.R. Rajan
(Chairman, Advisory Committee)
Associate Professor and Head
Centre for Pig Production & Research
College of Veterinary
and Animal Sciences, Mannuthy

Mannuthy
19/8/05

CERTIFICATE

We, the undersigned members of the Advisory Committee of **Dr. Jeeba K. George**, a candidate for the degree of Master of Veterinary Science in *Animal Breeding and Genetics*, agree that the thesis entitled **"EVALUATION OF PORCINE IMMUNE RESPONSES AMONG DIFFERENT GENETIC GROUPS"** may be submitted by Dr. Jeeba K. George, in partial fulfilment of the requirement for the degree.

30/4/05

Dr. M.R. Rajan
(Chairman, Advisory Committee)
Associate Professor and Head
Centre for Pig Production & Research
College of Veterinary and Animal Sciences, Mannuthy

30/9

Dr. K.V. Raghunandanan
(Member, Advisory Committee)
Associate Professor and Head
Department of Animal Breeding &Genetics
College of Veterinary and
Animal Sciences, Mannuthy

30/9

Dr. P. Nandakumar
(Member, Advisory Committee)
Associate Professor and Head
University Goat and Sheep Farm
College of veterinary and
Animal Sciences, Mannuthy

30/9

Dr. K. Vijayakumar
(Member, Advisory Committee)
Assistant Professor
Department of Veterinary Epidemiology and
Preventive Medicine
College of Veterinary and Animal Sciences, Mannuthy

30/9/05

External Examiner

Dr. C. S. Nagarajan
Associate Professor
Dept of ABG
Veterinary College
Bangalore-24

ACKNOWLEDGEMENTS

To the greatest degree, I am indebted to Dr. M.R. Rajan, Associate Professor and Head, Centre for Pig Production and Research and Chairman of the advisory committee for his meticulous guidance, valuable suggestions, keen interest, incessant support and encouragement and timely advice during the entire study period.

I would like to express my sincere and heartfelt gratitude to Dr. P. Nandakumar, Associate Professor and Head, University Goat and Sheep Farm for his inspiring advice, valuable suggestions and encouragement during the course of my study.

I am very much obliged to Dr. K. Vijayakumar, for his invaluable guidance, wide spread suggestions and help rendered as a member of advisory committee.

I wish to place on record my sincere thanks to Dr. K.V. Raghunandanan, for his priceless advice, comprehensive suggestions and support throughout the study.

I take great pleasure in thanking Dr. A. Kannan and Dr. K. Shyama, Assistant Professors, Centre for Pig Production and Research, for their generous support extended to me during the course of this study.

I am extremely thankful to Dr. M. Mini, Assistant Professor, Department of Microbiology for the help rendered in conducting lab works.

I am thankful to my respected teachers Dr. Stephan Mathew, Dr. K. Anilkumar, Dr. K.C. Raghavan, Dr. T.V. Aravindakshan, Dr. K.A. Bindu and Dr. A.P. Usha for their help.

A note of special thanks to Dr. Smitha P.S. for her untiring help and co-operation.

A special word of thanks to Mr. T.K. Haridasan and Mr. C.G. Varghese, Farm Assistants and Smt.Joicy.T.John, Technical Officer (Stati.), Centre for Pig Production and Research for their assistance.

I sincerely acknowledge the help rendered by my friends Dr. Anton Roseline, Dr.Dhannia, Flamy, Lali, Winny, Divya P.D., Veena, Ninu and Divya, K.N.

I remember and acknowledge my colleagues Dr. Naicy Thomas, Dr. Shymaja Uthaman, Dr. Amrita Susan, Dr. Suprabha, Dr. Bindu Mathew, Dr. Chitra, Dr. Kowsigraj, Dr. Jeeva, Dr. Sajeev, Dr. Seena, Dr.Jaibi, Dr. Sarad, Dr. Ariprasath and Dr. Preethy.

I am also thankful to the staff of Centre for Pig Production and Research and University Goat and Sheep Farm for their help and concern towards my study.

I offer my sincere thanks to all the staff of the centre for Advanced Studies in Animal Genetics and Breeding for their kind co-operation.

I sincerely acknowledge Kerala Agricultural University for awarding me the Junior Fellowship for post graduate study.

I appreciate with thanks, Mr.O.K.Ravindran, M/s. Peagles, Mannuthy for his neat and prompt typing of this thesis.

I express my heartfelt love towards my beloved parents and family members for their constant encouragement.

Above all I kneel before God for all He has given and .....

JEEBA K. GEORGE

CONTENTS

LIST OF TABLES

Introduction

LIST OF FIGURES

Introduction

1. INTRODUCTION

The pig serves as a large animal model for several human diseases and may be used in future for xenotransplantation. Increased disease resistance through improved general immune capacity would prove beneficial for the welfare and productivity of farm animals. Thus the porcine immune system is the subject of interest both in basic and applied research. Consequently, the elaboration and standardization of reagents for studies of immune functions in the pig has emerged rapidly during recent years. However, a comprehensive elucidation of the basis for variation in immune functions requires further knowledge of their genetic regulation.

The major source of loss in pig production has attracted little genetic research. The existence of genetic variation in immune responsiveness within and between breeds has only recently been demonstrated. A selection on a BLUP index for high or low immune responsiveness was successful in creating a genetic difference. However, a line with higher immune responsiveness would be expected to show a correlated reduction in lean growth every time the immune system was triggered.

In every animal species, there is a stretch of related genes known as the major histocompatibility complex (MHC). The MHC is home to array of genes which perform the critical tasks of helping the body to distinguish between things that are parts of itself and other intruding antigens. Because this ability is the crux of disease resistance, and because disease resistance is one of the hottest targets for genetic improvement in swine, MHC is a prime target of gene mapping efforts. By identifying markers linked to genes imparting superior ability to fight off invaders we can breed more highly resistant pigs, thereby reducing economic, environmental and health costs of combating disease.

Several studies have shown that genetic polymorphism within the MHC influences variation in immune functions and/or disease resistance. However, non-MHC, and a polygenic control of quantitative immune response traits have been confirmed in several experimental studies. In pig, additive genetic variation has been documented for a number of immune traits, e.g., antibody response, proliferative and cytokine responses of mononuclear cells, delayed-type hypersensitivity reactions and total and differential leukocyte counts. Medium-to-high heritabilities (h^2 = 0.3-0.8) have been estimated for several of the immune traits suggesting a large genetic impact.

Linkage mapping using dense genetic maps is a straight forward approach to locate genes that control traits and inherited diseases with a monogenetic inheritance. The same approach has also successfully been used to identify quantitative loci (QTLs) that control various polygenic traits in different organisms. Regarding immune functions, several QTLs influencing antibody response have recently been identified in mice. Detailed linkage maps of the porcine genome are now available. It is possible to identify QTLs for immune capacity in pig populations by genome analysis.

So far, genetic researchers have mapped 100 genes and 700 micro satellites in pig genome. Inside knowledge of the pig genome already is yielding practical payoffs in the case of porcine stress syndrome (PSS).

Most quantitative traits are controlled by many hundreds of genes, each with a small effect. A gene with a large effect such as halothane gene is very much the exception. Nevertheless much research is now under way to identify possible genes with useful effects on performance. The function of most of the genes so far detected is unknown. They may however be situated on the chromosome close to a gene that does affect performance for e.g., growth rate but for which no DNA test exists. The DNA tested gene is known as a marker,

because it marks a section of chromosome affecting performance. Possible markers have been reported for all the important traits, and many have been mapped.

In the process of marker assisted selection, DNA testing for the marker can be used to increase the frequency of the QTL and lead to an improvement in a production trait. The main benefit would be in traits such as meat quality or disease resistance which are difficult or expensive to measure in the live pig.

Current thinking is that the interaction of genes with each other is probably more important than originally recognized, so the implications of changing the frequency of any gene with a large effect may be difficult to predict from one population or even family to the next. A further difficulty is that the information from hundreds of markers of small effect may be difficult to assemble. At this stage the use of markers is risky, whereas BLUP selection is already proven and cost-effective.

The success of a genetic improvement programme dependens on its use by both seed stock and commercial swine producers. No genetic progress can be initiated if seed stock producers do not utilize accurately identified, genetically superior animals in their breeding programme. Researchers are trying to accelerate genetic improvements through molecular genetics, to evaluate the role of immune response during the development of the animal and to optimize factors related to herd management such as feeding and housing. Efficiency of production can also be improved by a better knowledge of the mechanisms involved in the regulation of immune responses.

Meantime it is important to realize that modern selective breeding programmes have been successful, cheap and safe, and that for the present there is little pressure to introduce biotechnology. The swine industry should use this

period wisely to carefully evaluate its positioning in relation to the new technologies and the public.

The present study was undertaken in three genetic groups namely Desi, Large White Yorkshire and Duroc x Large White Yorkshire with the following objectives.

1. To assess the magnitude of humoral and cell-mediated immune responses to specific antigens in pigs.
2. To compare immune responsiveness in different genetic groups of pigs.
3. To analyze the relationship of immune response with growth, occurrence of diseases and mortality among littermates.

Review of Literature

2. REVIEW OF LITERATURE

2.1 PORCINE IMMUNE SYSTEM

Splichalova *et al.* (2004) determined TNF-alpha, IL-10, IL-1 beta and IFN-gamma in amniotic fluids by ELISA and the expression of cytokines and some other inflammatory markers by immunohistochemistry.

Ficinska *et al.* (2005) reported that pseudorabies virus (PRV) g D protein contains a functional endocytosis motif (YRLL) in its cytoplasmic domain that drives spontaneous endocytosis of g D from the cell surface early in infection and that acts in concert with the endocytosis motif in g B to contribute to efficient internalization of antibody - antigen complexes in PRV - infected monocytes.

Experiments in a full thickness porcine wound model showed that rejection of glycerol treated allogenic skin grafts was up to six days delayed. Viable, untreated allogenic skin graft were rejected predominantly by CD8 positive T cells whereas in the glycerol treated grafts the influx of host cells was lower and the majority of the cells were macrophages (Richters *et al.*, 2005).

Yamamoto *et al.* (2005) isolated pig TRA clones from c DNA libraries of total RNA from two different sources, the thymus of a 1-month-old Large White strain pig and the peripheral blood lymphocytes of a 5-month-old clawn strain pig. Among 103 complete TRA c DNA clones from both sources, 33 different TRA V genes were identified.

Yang *et al.* (2005) proposed a strategy to study the structure of CD3 molecule, expressed specifically at the surface of porcine gamma delta - T cells. The results suggested that there are differences in the antigenicity, signal

transduction potentials and probably structural difference, between the CD3 molecule, expressed at the surface of alpha beta- and gamma delta - T cells.

2.2 CELL-MEDIATED IMMUNE RESPONSE

The chromium release test (CRT) was used to assess cell-mediated immunity to syngenic chemically induced tumors in guinea pigs (Miller and Blazkovec, 1979). In the strain 2 guinea pigs with diethyl nitrosamine (DEN) - induced hepatocarcinomas, 2 lines, line 1 and line 10 are antigenically distinct by the CRT but the two MCA-induced tumors have specific antigens as well as a common cross reactive antigen.

A modified passive haemagglutination (PHA) test was developed by Equchi *et al.* (1988) to detect antibodies to *Taylorella equigenitalis* in the sera of mares. A PHA test using horse red blood cells as indicator and an antigen prepared from *T. equigenitalis* by sonication following treatment with hyaluronidase was the most satisfactory in terms of sensitivity and specificity.

Raszyk and Pillich (1990) conducted a simple skin test in piglets and the dermal reaction was evaluated 20 minutes, 24 and 48 hours after the phytohaemagglutinin administration. Piglets that had papule larger than 15 mm 20 minutes after the antigen administration was regarded as piglets susceptible to states of allergy.

CMI was assessed by skin sensitivity testing, graft versus host (GVH) reaction and T lymphocyte count (Singh *et al.*, 1990). They observed highly significant reductions in CMI indicated by diminished skin sensitivity, GVH reactions and T lymphocyte counts.

Ekkel *et al.* (1995) described two experiments to standardize the phytohaemagglutinin (PHA) skin test as an indicator of lymphocyte reactivity in pigs after exposure to stressful situations in practical pig husbandry. They concluded that frequent measurements are preferable to single measurements when the effects of stress on immunological processes are studied.

Samarineau *et al.* (2000) conducted a study on cell-mediated immune response in swine and also detected the level of gamma interferon in swine lymphocyte culture, stimulated with phytohaemagglutinin.

Pre-natal maternal stress during late gestation is able to impair both humoral and cellular immune function in suckling piglets was reported by Tuchscherer *et al.* (2002). They also observed that gestational stress in pigs might affect the ontogeny of foetal immune system with consequences on the susceptibility to disease, and immune responsiveness to stressful stimuli of the offspring.

Brown (2005) reported that immunological competence of recipients of hematopoietic cell transplantation does not correlate with the administration of non-corticosteriod immuno suppressive agents. This apparent paradox reflects the unique and dynamic conglomeration of factors that affect immune reconstitution after haematopoietic cell transplantation.

Huang *et al.* (2005) conducted studies about cellular immunity against the measles virus. This study surveyed cellular immunity after measles, mumps and rubella combined vaccine (MMR) immunization. They concluded that for a better understanding of the durability of vaccine induced immunity and in order to establish the most appropriate immunization schedule, long term and large-scale prospective studies of measles - specific sero epidemiology and cellular immunity will be needed.

Mazumdar *et al.* (2005) demonstrated a correlation of humoral and cell-mediated immunity with protection against visceral leishmaniasis in hamsters.

2.3 HUMORAL IMMUNE RESPONSE

A role for specific cellular as well as humoral immunity has been suggested in experimental adrenalitis by Ischizawa and Daniels (1980). They observed histopathologic changes in adrenal glands correlated better with cell-mediated immune parameters than with specific antibody titres and cell-mediated mechanisms may be the more important factor in pathologic lesions of experimental adrenalitis.

A whole blood technique was used to assess the response of pig peripheral blood lymphocytes to phytohaemagglutinin by Jensen and Christensen (1981). PHA concentration giving maximum stimulation response, as calculated by a dose response function was found to vary depending on genetic variation among the pigs with an estimated heritability of 0.38.

Meier and Drake (1982) developed antibodies combined with a cell surface marking technique to facilitate scanning election microscopic (SEM) identification of quail cells in chimeras. They demonstrated that rabbit antibodies prepared by injection of stage 4 quail primitive streaks can be used to specifically label quail epiblast and mesoblast cells, providing markers for atleast two germ layers.

Hemagglutinating properties of *Haemophilus paragallinarum* serotype 2 and serotype C against freshly collected and glutaraldehyde (GA) - fixed chicken RBC were investigated by Sawata *et al.* (1982). They found that different from serotype 1, the non-treated organisms of serotype 2 and serotype C lack hemagglutinating activity.

Cox and Mc Auliffe (1983) reported that suppressor cells induced by injections of rat RBC are effective in preventing autoantibody production induced by rat bromelain treated (brom) RBC and vice versa.

Nesterenko *et al.* (1983) found that antisera did not inhibit the immune response of lymphocytes from unimmunized rabbits to SRBC or their proliferation in mixed lymphocyte culture.

Studies were performed on the behaviour of cutaneous delayed - type hypersensitivity (DTH) in guinea pigs in which macrophage disappearance reaction (MDR) was induced by Ochiya *et al.* (1983). The culture supernatants of macrophages incubated with the MIF fraction showed the ability to suppress reactions of cutaneous DTH, PHA (phytohaemagglutinin) and skin reactive factor.

A 51 Cr release microhemolytic complement assay is described by Poutrel and Caffin (1983) to detect hemolytic complement activity in bovine milk. CH 100 titer was determined by difference of counting between heated and unheated diluted whey samples from a standard linear regression. Comparative hemolytic values throughout lactation were established for the first time and confirmed the improved sensitivity of the assay.

Sensitive methods of an enzyme linked immunosorbent assay (ELISA) using red blood cells have been developed and were applied by Asahi *et al.* (1984) to detect antisheep red blood cell heterophile antibodies present in sera of *Schistosoma japonicum* (SJ) - infected mice. They concluded that heterophile antibody response is not a consequence of a specific immune response directed to the antigens of SJ parasites, since absorption of the heterophile antibody with SJ adult worms or an egg preparation did not reduce the heterophile antibody level.

The emigration of labeled thymus cells in the pig was studied directly in blood draining the large right distal cervical lobe of the thymus after controlled labeling with FITC by Binns *et al.* (1988). Surface marker studies showed that the surface phenotypes of the emigrants differ from both typical thymus and peripheral blood lymphocytes. The emigration of thymic cells is discussed in relation to its implications for the turnover of known functional peripheral T-cell populations.

The passive haemagglutination, enzyme linked immunosorbent assay and indirect fluorescent antibody tests were applied to study the non-specific reactions in experimentally infected guinea pigs and tuberculin positive bovines by Hammam *et al.* (1989). They were able to observe that the use of both absorbed sera and antigen raised the specificity of PHA and ELISA to 100 per cent and reduced the sensitivity of ELISA, IFA and PHA by 14, 27 and 29 per cent respectively.

van der Meijden *et al.* (1989) conducted a study in which BCG was administered intravesically in guinea pigs to investigate immune response and found that in the spleen no differences were observed after BCG administration regarding the number of cells present and in terms of the MHC class II (1a) antigen expression of the leukocytes.

Cerrone and Kuhn (1991) carried out limiting dilution analysis for both determining the frequency of cell subpopulations elicited during immune responses as well as for the analysis of immunoregulatory circuits. They suggested that the urease micro ELISA should be amendable for use with antigens not readily conjugated to an indicator RBC, and should be useful in those situations where determination of the antibody subclass (es) produced by responding micro- cultures is desired.

Three models were used to test the hypothesis that interspecific pregnancy failure between the sheep and goat is due to a species - specific maternal antibody response by Mac Laren *et al.* (1992). It was observed that a maternal cytotoxic antibody response to species - specific antigen (s) contributed failure of hybrid or ovine pregnancy in does.

Rao *et al.* (1992) described T 560, a tissue culture adapted B lymphoma derived from the gut associated lymphoid tissue (GACT) of a (B10 x B10, H - 2a H - 4b) F1 hybrid mouse. These T560 bind bromelain - treated mouse RBC (Br MRBC) in a PC chloride inhibitable manner but do not bind SRBC, OX RBC (ORBC) or TNP-ORBC.

Weaned pigs exposed daily to either unpredictable draught or intermittent unpredictable draught showed different lymphocyte blastogenic response after mitogenic stimulation with phytohaemagglutinin and concanavalin (Scheepens *et al.*, 1994).

Takemoto *et al.* (1994) performed the antigenicity tests of Tazobactam/piperacillin (TAZ/PIPC), tazobactam (TAZ: beta-lactamase inhibitor) and piperacillin (PIPC: penicillin antibiotic) in mice and guinea pig. The results of the study confirmed that TAZ/PIPC, TAZ or PIPC had no immunogenicity and allergenicity in either passive cutaneous anaphylaxis (PCA) test using BALB/C and C3H/Hc mice or in PCA test using guinea pigs.

van Heugten *et al.* (1994) measured antibody response to sheep red blood cells in weaned piglets allotted to 12 dietary treatments. Antibody response to SRBC and serum IgM and IgG concentrations were not affected by dietary treatments.

Through interval mapping using a least-squares method, four QTLs with significant effects were identified by Inger *et al.* (1998). The results of the study conclusively showed that it was possible to identify QTLs for immune capacity traits in pigs.

Kasukawa (2001) reviewed first the history of detection for auto-antibodies and the methods to detect the circulating immune complexes.

Lessard *et al.* (2002) evaluated humoral and cellular immune responses of piglets after castration at different ages. They found that age of first immunization significantly influenced the ability to raise antibodies to the injected antigen.

Biberthaler (2005) stated that peripheral blood monocytes play a critical role in immune response. Genome - wide mRNA expression patterns in circulating peripheral blood monocytes were also studied.

Chow *et al.* (2005) depicted the complex biology of immune response. They also developed a mathematical model incorporating major elements of the acute inflammatory response in C57B1/6 mice.

According to Hoesel *et al.* (2005) at the onset of sepsis, PMN are important in regulating the levels of bacteremia, whereas after the onset of sepsis, as they lose innate immune functions, their presence is associated with higher levels of bacteremia and intensified organ dysfunction. They also demonstrated the harmful effects of neutrophils (PMN).

Molina (2005) emphasized in their study the mechanisms involved in mediating the stress response and their role in modulating immune function during and after traumatic injury.

2.4 ASSOCIATION WITH ECONOMIC TRAITS

The high and low susceptible bloat herds at Ruakura have been compared for body composition (Carruthers and Morris, 1988), food intake, rib girth and milk yield (McIntosh *et al.,* 1988).

2.4.1 Litter Traits

Rajan (2002) conducted comparative study of immune responsiveness in Desi and Large White Yorkshire piglets. The litter size at birth, litter size at weaning, pre-weaning mortality percentage were 11.453 ± 0.240, 9.107 ± 0.279, 18.38 ± 2.163 for Large White Yorkshire piglets and 8.907 ± 0.219, 7.520 ± 0.697, 16.663 ± 1.974 for Desi piglets respectively. He also estimated the sire effect and heritability of immune responsive traits.

Sivaraman *et al.* (2005) evaluated litter traits and estimated their genetic and non-genetic parameters in a broiler dam line. Phenotypic correlations among body weights were high and positive but were very low between body weights and most immunological traits. Genetic correlations of body weights were positive and medium to high with serum IgG.

2.4.2 Growth

Meeker *et al.* (1987) conducted a three-breed diallele crossbreeding experiment to estimate general combining abilities of swine breeds and heterosis for humoral immune response to pseudorabies virus and atrophic rhinitis vaccines. The relationship between humoral immune response to PR vaccine and growth traits were similar to that observed for *B. bronchoseptica* vaccine. Immune response to both antigens was not associated with back fat thickness.

The effects of L-carnitine on porcine fetal growth traits and the IGF system were determined by Waylan *et al.* (2005). The results suggested that L-carnitine supplemented to gestating sows altered the IGF system and may affect fetal growth and development.

2.5 DISEASE INCIDENCE AND MORTALITY

Lillehoj (1991) reported that resistance to *Eimeria tenella* is due to both MHC - associated and non-MHC - associated genes. They also assessed the role of MHC genes in resistance to other species of *Eimeria.*

Variation in resistance to *Pasteurella multocida* was reported by Lamont (1998). In a cross between lines he found difference in survival following intramuscular inoculation. Comparing segregating MHC haplotypes he found small but statistically significant difference in survival between MHC haplotypes.

The relationship between level of disease resistance and production under constant infection pressure was noticed by van der waaiji *et al.* (2000).

Visscher *et al.* (2002) proved that diseased incidence and immunological traits are the two important criteria for the selection of healthy pigs.

Materials and Methods

3. MATERIALS AND METHODS

3.1 ANIMALS

The study was carried out on 108 piglets of either sex, thirty six each belonging to Duroc x Large White Yorkshire, Large White Yorkshire and Desi within the age group of 2 - 3 months, maintained at Centre for Pig Production and Research, College of Veterinary and Animal Sciences, Mannuthy. Animals were under the similar management conditions. Piglets of each genetic group were sired by six boars.

3.2 TRAITS MEASURED

The litter traits studied were birth weight, litter size at birth, weaning litter size and body weight at weaning.

3.3 CELL-MEDIATED IMMUNE RESPONSE

The cell-mediated immune response was assessed by noting cutaneous response to the intradermal injection of phytomitogen, phytohaemagglutinin (PHA-M).

3.3.1 PHA Skin Test

With suitable modifications, *in vivo* cutaneous response to PHA-M was assessed as described by Wilkie *et al.* (1991). The skin on the base of left ear was clipped and disinfected with 70 per cent alcohol. Using a Harpender skin fold caliper, the double fold skin thickness was measured in millimeters. Phytohaemagglutinin-M (PHA-M) (GIBCO BRL, USA) was dissolved in sterile saline and diluted to contain 50 μg in 0.1 ml. The prepared solution was kept

frozen at -20°C until half an hour before use. The solution was taken out and thawed to room temperature. Using a 24-gauge needle 0.1 ml of this solution was injected intradermally at the base of left ear. As a control, 0.1 ml of sterile saline was injected at the base of right ear. At 24, 48 and 72 hour post-injection double fold skin thickness was measured and the increase in skin thickness was calculated.

3.4 HUMORAL IMMUNE RESPONSE

3.4.1 Test Antigen

As sheep red blood cells are complex and apparently harmless antigens, were chosen as test antigen. One hundred milliliters of blood was collected from a healthy sheep in double the quantity of Alsever's solution and the red blood cells were washed thrice in sterile phosphate buffered saline (PBS 0.01 M, pH 7.2) by the method of repetitive centrifugation (1500 rpm for 10 minutes). The cells were suspended in a final concentration of 20 per cent (v/v) in fresh sterile PBS and stored at 4°C in sterile glass containers until used.

3.4.2 Antigen Administration

The test antigen was injected intravenously through the marginal ear vein at the rate of 0.5 ml per kilogram body weight using 24-guage hypodermic needle.

3.4.3 Blood Sourcing for Serum

Using a 18-gauge needle, blood samples were taken from the right external jugular vein. The puncture site was the deepest point of the jugular groove formed by the medial sternocephalic and lateral brachiocephalic muscles and in between the angle formed by anterior border of sternum and clavicle. Sera

were separated from blood samples collected at days zero, seven, fourteen and twenty-one days post-inoculation and stored in sterile storage vials at -20°C until used.

3.4.4 Microhaemolytic Test

Microhaemolytic test to assess the serum antibody titre to sheep RBC was performed as per Hines (1988). The test was carried out in 96 welled microtitre plate (Laxbro, Pune).

1. To each well in the first row of the microtitre plate 50 µl of Phosphate Buffered Saline (pH 7.2) was added.
2. To the first well 50 µl of neat serum was added.
3. Mixed and transferred 50 µl to the second well.
4. Serial double fold dilutions of serum were made and 50 µl of the diluted serum was discarded from the last well.
5. Added 25 µl of 20 per cent suspension of sheep RBC in PBS followed by 20 µl of fresh guinea pig serum to each well.
6. The plate was incubated at 30°C for 4 hours before taking the reading.

Control

1. RBC control

25 µl of 20 per cent sheep RBC suspension + 70 µl of PBS.

2. Positive control

50 µl of known positive serum + 25 µl of 20 per cent sheep RBC + 20 µl of fresh guinea pig serum.

3. Negative control

50 µl of known negative serum + 25 µl of 20 per cent sheep RBC + 20 µl of fresh guinea pig serum.

The extent of haemolysis was read as follows.

0 - All cells settled at bottom intact and supernatent was clear.

1 - Nearly 20 per cent of cells were lysed. Supernatent was reddish coloured.

2 - Nearly 50 per cent of the cells were lysed. The intact cells formed a small button or ring at bottom. Supernatent was red.

3 - Nearly 90 per cent of the cells were lysed. Supernatent was bright red which on shaking became cloudy.

4 - All cells were lysed. Whole liquid was bright red and retained the brightness even after shaking.

The antibody titre was recorded as reciprocal of highest dilution of serum giving the reaction to the extent of two.

3.5 STATISTICAL ANALYSIS

3.5.1 Least Square Analysis

Least square analysis using the LSW-LMW package (Harvey, 1987) was performed to assess the influence of breed, sex, sire, litter traits, growth, disease occurrence and mortality among piglets on immune response traits.

In an attempt to distinguish the effect of breed, sex, sire and weaning body weight classes, least square analysis was performed on immune responses (Model 1).

Table 3.1 Weaning body weight classes

Class	Frequency
6.0-7.5	31
7.5-9.0	25
9.0-10.5	25
10.5 – 12.0	17
12.0 – 13.5	10

Model 2 was used to assess the influence of litter traits on immune responses. Model 3 was used to test the influence of disease occurrence and pre-weaning mortality of littermates on immune responses.

Model 1

Y_{ijkl} - $\mu + b_i + Sx_j + Sr_k{:}b_i + Wt_l + e_{ijkl}$ where

Y_{ijkl} - immune response of Y_{ijkl}^{th} piglet

μ - population mean

b_i - effect of i^{th} breed (i = 1,2,3)

Sx_j - effect of sex of piglets (j = 1,2)

$Sr_k{:}b_i$ - effect of k^{th} sire nested in i^{th} breed (k = 1,2 - - 18)

Wt_l - effect of l^{th} body weight class (l = 1,2, - - 5)

e_{ijkl} - error

Model 2

Y_{ijklpq} - $\mu + b_i + Sx_j + Sr_k:b_i + Wt_l + Sb_p + Sw_q + e_{ijklpq}$ where, all terms are as defined in model 1 except

Sb_p - litter size at birth (p = 7-----14)

Sw_q - litter size at weaning (q = 6-----10)

Model 3

Y_{ijklmn} - $\mu + b_i + Sx_j + Sr_k:b_i + Wt_l + Dg_m + Mt_n + e_{ijklmn}$ where, all terms are as defined in model 1 except

Dg_m - occurrence of enteritis (0,1)

Mt_n - pre-weaning mortality among littermates (0,1)

3.5.2 Correlation

The correlations of immune responses with birth weight, weaning body weight, and pre-weaning mortality among littermates were estimated. The phenotypic correlations between immune response traits were worked out.

Cross covariance analysis (Hazel, 1943) was used for estimation of correlation.

3.5.3 Heritability

Heritabilities of immune response traits were estimated by paternal half-sib correlation anlaysis (One Way Layout, Unbalanced Design).

Results

4. RESULTS

4.1 LITTER TRAITS

Different litter traits in Desi, Large White Yorkshire and Duroc x Large White Yorkshire are compared in Table 4.1.

The highest birth weight, body weight at weaning, litter size at birth and weaning were recorded in Duroc x Large White Yorkshire, medium in Large White Yorkshire and least in Desi. The least square mean for birth weight, weaning body weight, litter size at birth, litter size at weaning had probability values of $P = 0.0042$, $P = 0.0093$, $P = 0.0081$, $P = 0.0068$ respectively. The litter traits were highly significantly different in different genetic groups ($P < 0.01$).

4.2 CELL-MEDIATED IMMUNE RESPONSE

Cutaneous response to intradermal injection of PHA-M in Desi, Large White Yorkshire and Duroc x Large White Yorkshire are presented in Table 4.2.

The overall pre injection skin thickness in Desi piglets was 3.273 ± 1.150 with a range of 2.80-3.80 (mm). Cutaneous response to PHA-M at 24 hour post-injection as evidenced by increase in skin thickness had a mean value of 2.405 ± 0.805 (2.20-3.00). The mean value for cutaneous response to phytohaemagglutinin at 48 hour was 1.520 ± 1.068 (1.10-1.90). The 72 hour increase in skin thickness in response to PHA-M had mean of 1.251 ± 0.993 (0.50-1.80).

In Large White Yorkshire, the zero hour skin thickness measured had mean of 3.357 ± 0.833 (3.1 to 3.5). The 24 hour increase in skin thickness in

Table 4.1 Comparison of litter traits in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets

Traits	Desi		Large White Yorkshire		Duroc x Large White Yorkshire	
	Mean	SE	Mean	SE	Mean	SE
Birth weight(kg) P=0.0042**	0.806	0.312	1.631	0.221	1.890	0.126
Body weight at weaning (kg) P=0.0093**	8.316	0.136	11.106	0.126	14.312	0.136
Litter size at birth P=0.0081**	9.316	0.531	12.306	0.312	14.132	0.432
Litter size at weaning P=0.0068**	6.313	0.238	10.112	0.112	10.136	0.216

** -Significant at 1% level

Table 4.2 Cutaneous response to intradermal injection of phytohaemagglutinin in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets

Breed	Sex	N	Zero hour skin thickness (mm)			Post-injection increase in skin thickness (mm)								
						24 hour			48 hour			72 hour		
			Range	Mean	SE	Range	Mean	SE	Range	Mean	SE	Range	Mean	SE
Desi	Male	2	2.80-3.40	3.13	0.856	2.20-2.90	2.31	0.817	1.10-1.80	1.50	0.790	0.50-1.70	1.24	0.697
	Female	34	2.90-3.80	3.35	1.449	2.30-3.00	2.59	0.848	1.30-1.90	1.60	1.408	0.60-1.80	1.28	1.248
	Total	36	2.80-3.80	3.27	1.150	2.20-3.00	2.41	0.805	1.10-1.90	1.52	1.068	0.50-1.80	1.25	0.993
Large White Yorkshire	Male	6	3.10-3.40	3.35	0.849	2.10-2.50	2.31	1.173	1.10-1.70	1.40	0.801	0.50-1.70	1.01	1.530
	Female	30	3.20-3.50	3.36	0.827	2.20-2.60	2.40	1.248	1.20-1.80	1.52	1.321	0.60-1.80	1.25	1.468
	Total	36	3.10-3.50	3.36	0.833	2.10-2.60	2.36	1.203	1.10-1.80	1.46	1.061	0.50-1.80	1.31	1.103
Duroc x Large White Yorkshire	Male	4	2.90-3.30	3.14	0.731	2.20-2.60	2.31	1.813	1.10-1.60	1.50	0.078	0.50-1.60	1.20	0.060
	Female	32	3.00-3.40	3.36	0.860	2.30-2.80	2.48	1.801	1.20-1.70	1.58	0.081	0.60-1.70	1.26	0.063
	Total	36	2.90-3.40	3.34	0.795	2.20-2.80	2.38	1.807	1.10-1.70	1.49	0.079	0.50-1.70	1.24	0.061
Overall		108	2.80-3.80	3.32	0.928	2.10-3.00	2.39	1.279	1.10-1.90	1.51	0.743	0.50-1.80	1.21	0.835

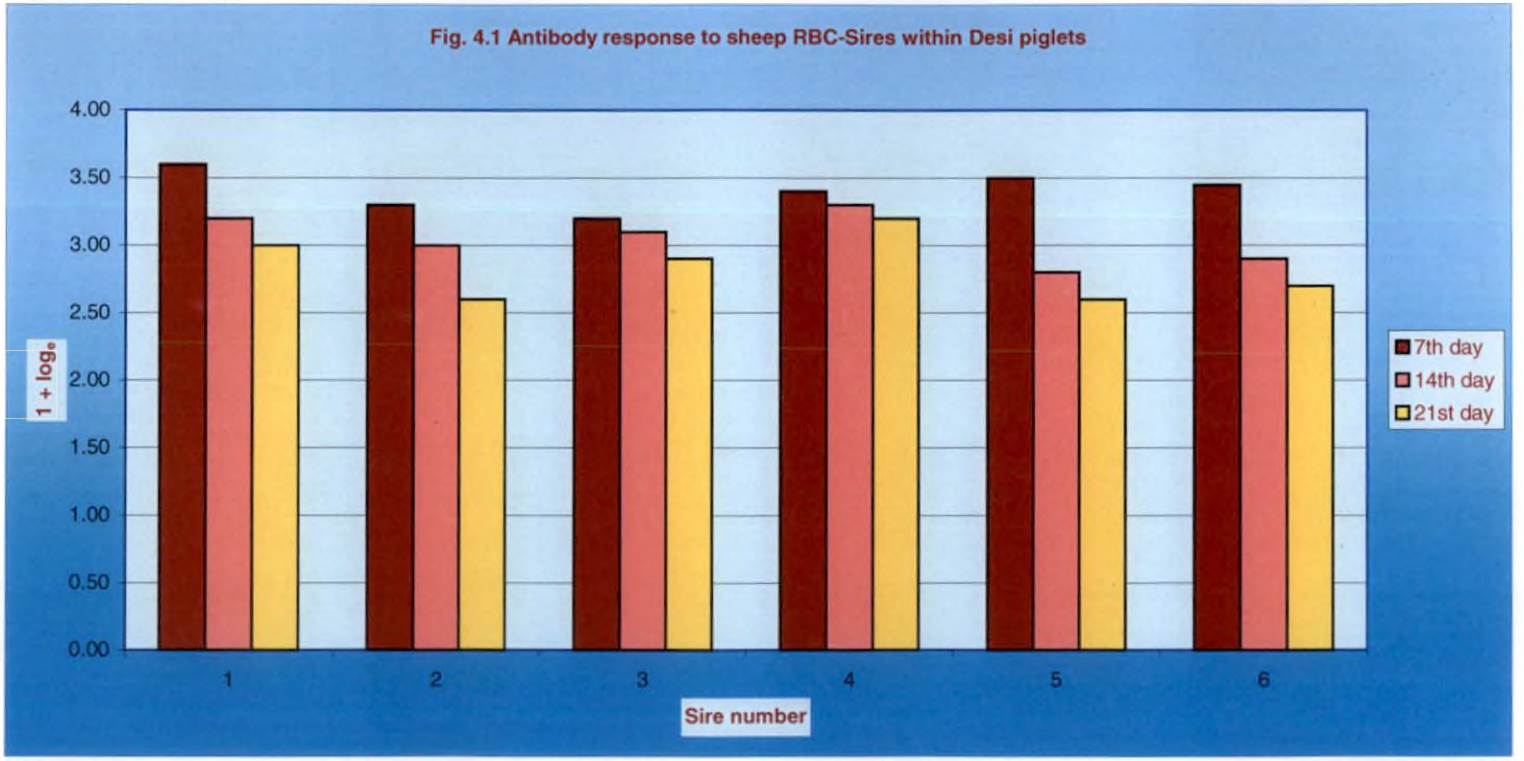


Fig. 4.1 Antibody response to sheep RBC-Sires within Desi piglets
4.00
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00
1 + log$_e$
1
2
3
4
5
6
Sire number
7th day
14th day
21st day

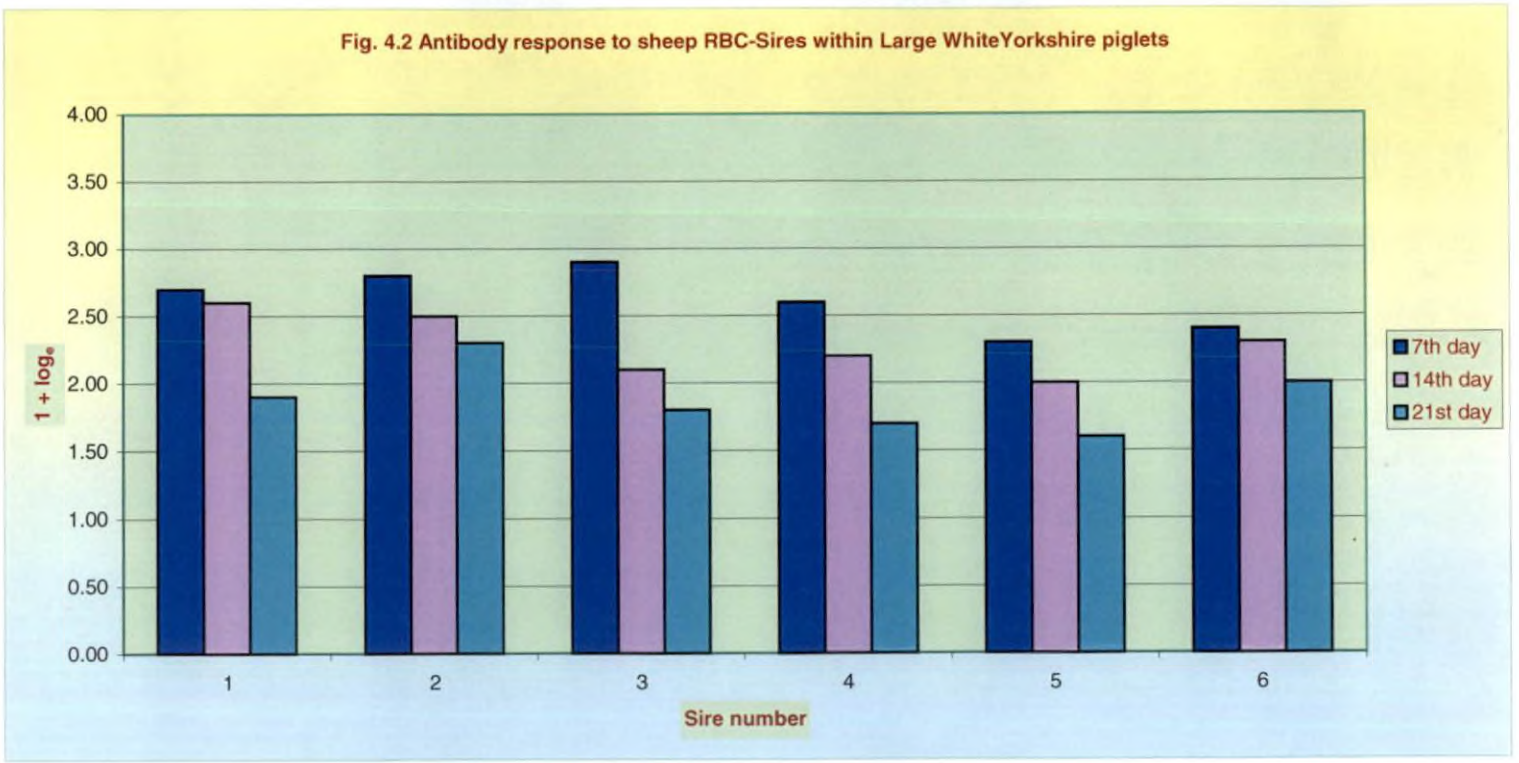


Fig. 4.2 Antibody response to sheep RBC-Sires within Large WhiteYorkshire piglets
4.00
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00
1 + log$_e$
1
2
3
4
5
6
Sire number
7th day
14th day
21st day

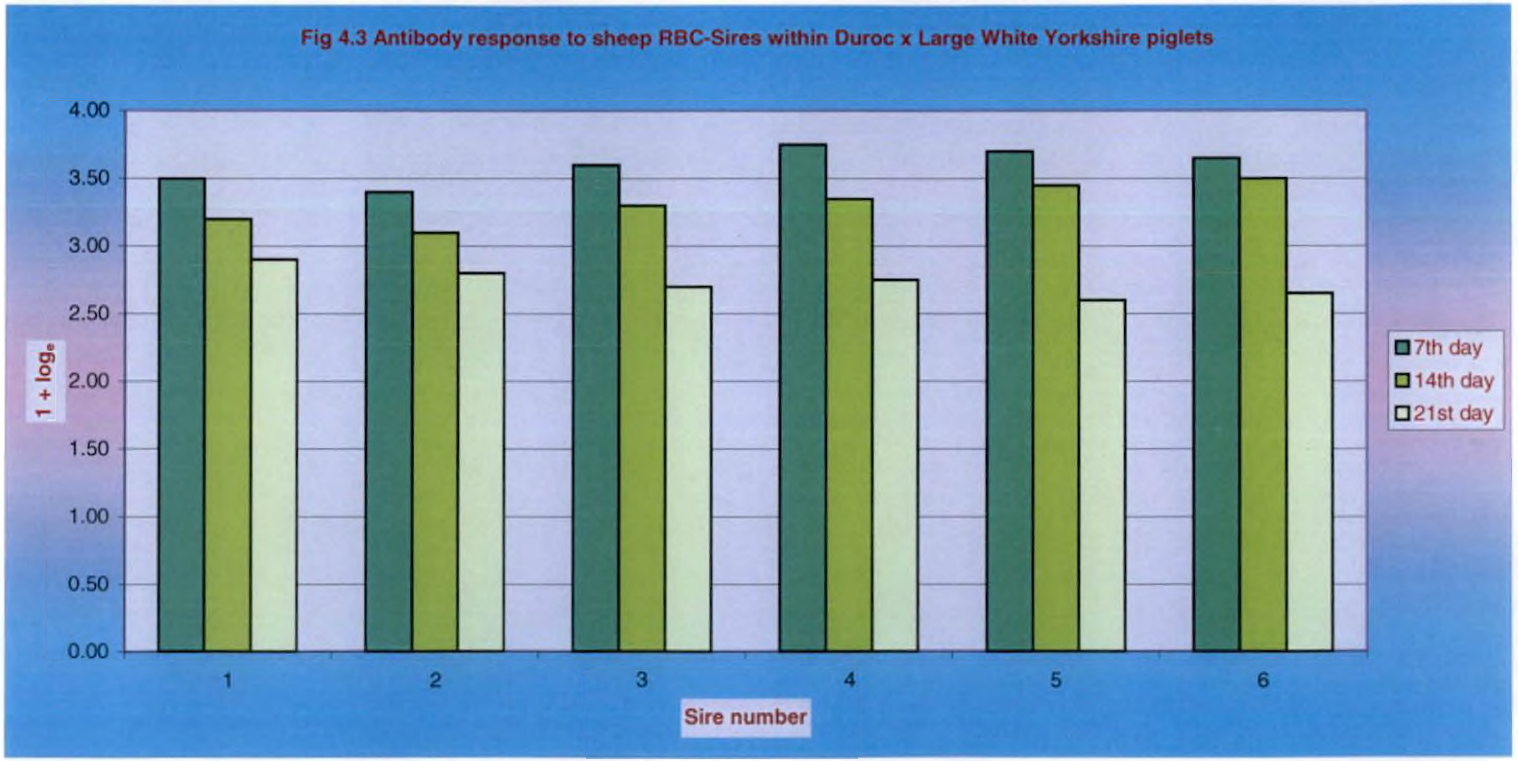


Fig 4.3 Antibody response to sheep RBC-Sires within Duroc x Large White Yorkshire piglets
4.00
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00
1 + log$_e$
1
2
3
4
5
6
Sire number
7th day
14th day
21st day

response to PHA-M had mean value of 2.359 ± 1.203 (2.10 – 2.60). The 48 hour response was 1.460 ± 1.061 (1.10-1.80) and 72 hour increase had least square mean of 1.31 ± 1.103 (0.50-1.80).

In Duroc x Large White Yorkshire crossbreds - the exhibited response values were in between that of Desi and Large White Yorkshire. The mean zero hour and 24 hour, 48 hour, 72 hour mean increase in skin thickness were 3.340 ± 0.795 (2.90-3.40), 2.381 ± 1.807 (2.20-2.80), 1.490 ± 0.079 (1.10-1.70) , 1.243 ± 0.061(0.50-1.70) respectively. The least square mean for different litter traits had no significance on cutaneous response to PHA-M.

4.3 HUMORAL IMMUNE RESPONSE

Antibody response to sheep RBC in Desi, Large White Yorkshire and Duroc x Large White Yorkshire pigs are shown in Table 4.3.

Naturally occurring antibodies could not be detected in three genetic groups. $1+\log_e$ transformed antibody titre on 7^{th} day in Desi piglets had mean of 3.625 ± 0.176 with a range of 2.39-4.47. The 14^{th} day titre value had least square mean of 3.290 ± 0.170 (2.39 – 4.47). The mean response on 21^{st} day was 3.213 ± 0.189 (2.39 - 4.47).

In Large White Yorkshire, the overall antibody titre on 7^{th} day was 2.718 ± 0.141(1.69-3.08). The antibody titre on 14^{th} day had least square mean of 2.732 ±0.410 (1.69-3.08). The mean value for antibody response to sheep RBC on 21^{st} day was 2.410 ± 0.409 (1.69 – 3.08).

In Duroc x Large White Yorkshire crossbreds – titre values were with least square mean of 3.510 ± 0.149 (2.39-4.47), 3.207 ± 0.169 (2.39 - 4.47) and 2.918 ± 0.174 (2.39-4.47) on 7^{th} , 14^{th} and 21^{st} day respectively. The least square

Table 4.3 Antibody response to sheep RBC in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets

Breed	Sex	N	Pre - inoculation titre			1+ $\log_e$ of titre								
						7th day			14th day			21st day		
			Range	Mean	SE	Range	Mean	SE	Range	Mean	SE	Range	Mean	SE
Desi	Male	2	0-0	0	0	2.386-4.465	3.455	0.163	2.386-4.465	3.269	0.172	2.386-4.465	3.201	0.182
	Female	34	0-0	0	0	3.079-4.465	3.772	0.187	3.079-4.465	3.356	0.169	2.386-4.465	3.216	0.191
	Total	36	0-0	0	0	2.386-4.465	3.625	0.176	2.386-4.465	3.290	0.170	2.386-4.465	3.213	0.189
Large White Yorkshire	Male	6	0-0	0	0	1.693-2.386	2.081	0.136	1.693-2.386	2.716	0.413	1.693-2.386	2.312	0.401
	Female	30	0-0	0	0	2.386-3.079	2.916	0.148	2.386-3.079	2.766	0.312	2.386-3.079	2.413	0.412
	Total	36	0-0	0	0	1.693-3.079	2.718	0.141	1.693-3.079	2.732	0.410	1.693-3.079	2.410	0.409
Duroc x Large White Yorkshire	Male	4	0-0	0	0	2.386-3.079	3.250	0.142	2.386-3.079	3.201	0.181	2.386-3.079	3.001	0.171
	Female	32	0-0	0	0	2.386-4.465	3.651	0.153	2.386-4.465	3.281	0.171	2.386-4.465	3.013	0.183
	Total	36	0-0	0	0	2.386-4.465	3.510	0.149	2.386-4.465	3.207	0.169	2.386-4.465	2.918	0.174
Overall		108	0-0	0	0	1.693-4.465	3.220	0.153	1.693-4.465	3.091	0.239	1.693-4.465	2.842	0.258

Table 4.4 Least square mean for cutaneous response to PHA-M in pre-weaning mortality (among littermates) and enteritis classes

Independent variable		N	24 hour		48 hour		72 hour	
			Mean	SE	Mean	SE	Mean	SE
			P=0.9990 NS		P=0.9933 NS		P=0.9528 NS	
	Yes	26	2.725	0.2261	2.23	0.2725	1.730	0.2098
	No	82	2.767	0.2921	2.486	0.332	1.596	0.2990
			P=0.7840 NS		P=0.7179 NS		P=0.9831 NS	
	Yes	24	2.617	0.3540	2.260	0.2893	1.051	0.3091
	No	84	2.725	0.3205	2.121	0.3631	1.692	0.2880

NS – Non-Significant

Table 4.5 Least square mean for antibody response to sheep RBC in pre-weaning mortality (among littermates) and enteritis classes

Independent variable		N	7^{th} day		15^{th} day		21^{st} day	
			Mean	SE	Mean	SE	Mean	SE
			P=0.3349 NS		P=0.2360 NS		P=0.3156 NS	
	Yes	26	3.8366	0.5087	3.4724	0.5111	3.0365	0.2921
	No	82	3.4179	0.2164	3.243	0.8586	2.0840	0.1274
			P=0.2006 NS		P=0.4031 NS		P=0.1101 NS	
	Yes	24	3.3722	0.7674	3.1985	0.6344	2.5353	0.6139
	No	84	3.477	0.6334	3.202	0.6139	2.7084	0.5255

NS – Non-Significant

Table 4.6 Correlations among immune response traits

	1	2	3	4	5	6
Cutaneous response to phytoheamagglutinin at 24 hour (1)	1.000					
Cutaneous response to phytoheamagglutinin at 48 hour (2)	0.738**	1.000				
Cutaneous response to phytoheamagglutinin at 72 hour (3)	0.745**	0.711**	1.000			
Antibody response to Sheep RBC on 7th day (4)	-0.219*	-0.209*	-0.133 NS	1.000		
Antibody response to Sheep RBC on 14th day (5)	-0.089 NS	-0.073 NS	0.069 NS	0.794**	1.000	
Antibody response to Sheep RBC on 21st day (6)	0.086 NS	0.111 NS	0.083 NS	0.816**	0.893**	1.000

** - Significant at 1 % level *-Siginificant at 5 % level NS-Non- siginificant

Table 4.7 Correlations of immune response traits with weight at birth, weight at weaning and pre-weaning mortality among littermates

	Weight at birth	Weight at weaning	Pre-weaning mortality
Cutaneous response to PHA-M at 24 hour post-injection	0.065 NS	-0.098 NS	0.109 NS
Cutaneous response to PHA-M at 48 hour post-injection	-0.254**	-0.053 NS	0.160 NS
Cutaneous response to PHA-M at s72 hour post-injection	0.321**	-0.123 NS	0.032 NS
Antibody response to Sheep RBC on 7^{th} day $(1 + \log_e)$	0.343**	0.105 NS	-0.028 NS
Antibody response to Sheep RBC on 14^{th} day $(1 + \log_e)$	0.363**	-0.176 NS	-0.074 NS
Antibody response to Sheep RBC on 21^{st} day $(1 + \log_e)$	0.783**	-0.036 NS	-0.099 NS

** -Significant at 1 % level NS-Non- siginificant

mean for different litter traits had no significance on antibody response to sheep RBC.

4.4 SIRE EFFECTS

Sire effects on antibody response to sheep RBC were evaluated (Fig.4.1, 4.2 and 4.3).

The effects of sires within Desi, Large White Yorkshire and Duroc x Large White Yorkshire were found to be highly significant ($P<0.01$) on antibody response to sheep RBC on 7^{th}, 14^{th} and 21^{st} day.

4.5 ENTERITIS AND PRE- WEANING MORTALITY

Table 4.4 presents least square mean for cutaneous response to PHA-M in pre- weaning mortality (among littermates) and enteritis classes.

The association of cutaneous response to phytohaemagglutinin with pre-weaning mortality (among littermates) and enteritis were found to be non-significant in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets.

Least square mean for antibody response to sheep RBC in pre - weaning mortality (among littermates) and enteritis classes is given in Table 4.5.

The association of antibody response to sheep RBC with pre- weaning mortality (among littermates) and enteritis were also found to be non - significant in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets.

4.6 CORRELATION ANALYSIS

Table 4.6 details the correlation among immune response traits.

Correlation between cutaneous response to PHA-M at 24 hour and 48 hour (0.738), 24 hour and 72 hour (0.745), 48 hour and 72 hour (0.711) were highly significant (P<0.01). Correlation of antibody response to sheep RBC on 7^{th} day with 14^{th} day (0.794), 7^{th} day with 21^{st} day (0.816), 14^{th} day with 21^{st} day (0.893) were also found to be highly significant (P<0.01).

Antibody response to sheep RBC on 7^{th}, 14^{th}, 21^{st} day with cutaneous response to PHA-M at 72 hour associates non-significantly. Correlation of antibody response to sheep RBC on 14^{th} day with cutaneous response to phytohaemagglutinin at 24 hour and 48 hour were found to be non-significant. Association between antibody response to sheep RBC on 7^{th} day and cutaneous response to PHA-M at 24 hour (-0.219) 48 hour (-0.209) were negative and significant (P < 0.05). Antibody response to sheep RBC on 21^{st} day correlates non-significantly with cutaneous response to PHA-M at 24 hour and 48 hour.

Table 4.7 shows correlations of immune response traits with weight at birth, weight at weaning and pre- weaning mortality.

Association of cutaneous response to PHA-M at 24, 48 and 72 hour with weaning body weight, pre-weaning mortality were found to be non-significant. Correlations between antibody response to sheep RBC on 7^{th}, 14^{th}, 21^{st} day and birth weight were positive and highly significant (P<0.01). Antibody response to sheep RBC on 7^{th}, 14^{th} and 21^{st} day associates non-significantly with weaning body weight and pre-weaning mortality. Even though correlations were non-significant, they revealed a negative trend. The association between cutaneous response to PHA-M at 24 hour and birth weight was non-significant. Correlation

Table 4.8 Heritability estimates of immune response traits

Traits	Heritability	Standard Error
Pre-injection skin thickness	0.41	0.30
Cutaneous response to phytohaemagglutinin at 24 hour	0.72	0.35
Cutaneous response to phytohaemagglutinin at 48 hour	0.64	0.25
Cutaneous response to phytohaemagglutinin at 72 hour	0.51	0.20
Antibody response to Sheep RBC on 7^{th} day post-inoculation	0.84	0.28
Antibody response to Sheep RBC on 14^{th} day post-inoculation	0.87	0.39
Antibody response to Sheep RBC on 21^{st} day post-inoculation	0.81	0.33

of cutaneous response to PHA-M at 72 hour with birth weight (0.0321) was positive and highly significant ($P<0.01$). Correlation between cutaneous response to phytohaemagglutinin at 48 hour and birth weight (-0.254) was negative and highly significant ($P<0.01$).

4.7 HERITABILITY ESTIMATION

Table 4.8 documents the heritability estimates of immune response traits.

Heritabilities of 0.41 ± 0.30, 0.72 ± 0.35, 0.64 ± 0.25, 0.51 ± 0.20, 0.84 ± 0.28, 0.87 ± 0.39, 0.81 ± 0.33 were estimated for pre-injection skin thickness, cutaneous response to PHA-M at 24, 48, 72 hours post-injection and antibody response to sheep RBC on seventh, 14^{th}, 21^{st} day post - inoculation.

5. DISCUSSION

Genetic resistance to disease in livestock is often due to many genes (polygenes) and not a single pair of genes. In such cases the development of a strain of animals resistant to disease would be long, slow process because the animals produced would have to be exposed to the disease to determine if they were resistant before selections could be made. Probably the most important problem encountered in developing strains of genetically resistant animals is that genetic resistance to disease appears to be specific and not general. Selection for superior performance in livestock includes a certain amount of automatic natural selection for genetic resistance to disease because those that perform best must be healthy and free from infections. To date, man has done very little deliberate selection for genetic resistance to disease, although nature has always selected in this direction. However, the development of strains of animals resistant to certain diseases has been successful.

5.1 LITTER TRAITS

The highest birth weight, body weight at weaning, litter size at birth and weaning were recorded in Duroc x Large White Yorkshire, medium in Large White Yorkshire and least in Desi. The litter traits were highly significantly different in different .genetic groups ($P< 0.01$). This result is in agreement with reports of Lessard *et al* (2002) and contradiction with the findings of Morrow-Tesch *et al* (1994).

The highest birth weight, weaning body weight, litter size at birth and weaning were recorded in Duroc x Large White Yorkshire indicating the prolificacy and faster growth rate of crossbreds. A lower body weight, weight at weaning, litter size at birth and weaning were observed in Desi. An attributable reason could be genotype- environment interaction. Desi has been developed

originally in a scavenging environment where occasional feeding is allowed, a better feed conversion efficiency and litter performance were noticed. In a better management condition where computed feeding is practiced, leads to more fat deposition and it may affect reproductive and other litter performance. In filthy environment, a better performance could be expected.

5.2 CELL-MEDIATED IMMUNE RESPONSE

The 24 hour post-injection increase in skin thickness had a mean value of 2.405 ± 0.805 with a range of 2.20-3.00 (mm). The mean value for cutaneous response to PHA-M at 48 hour was 1.520 ± 1.068 (1.10-1.90). The increase in skin thickness 72 hour post- injection had least square mean of 1.251 ± 0.993 (0.50-1.80).

In Large White Yorkshire, 24 hour increase in skin thickness had mean of 2.359 ± 1.203 (2.10 – 2.60). The 48 hour post- injection increase in skin thickness to PHA-M had least square mean of 1.460 ± 1.061(1.10 - 1.80) and 72 hour increase had mean value of 1.310 ± 1.103(0.50-1.80).

In Duroc x Large White Yorkshire cross breds, cutaneous response values were in between that of Desi and Large White Yorkshire.The 24,48,72 hour increase in skin thickness in response to phytohaemagglutinin were 2.381 ± 1.807 (2.20-2.80), 1.490± 0.079(1.10-1.70), 1.243 ± 0.061 (0.50-1.70) respectively.

Significant difference in PHA response in cow, pig and sheep was noticed by Eguchi *et al.* (1988). Raszyk and Pillich (1990) observed states of allergy in response to phytohaemagglutinin. CMI assessed by skin sensitivity testing revealed immunosuppressive effects of specific diseases in broiler chicks (Singh *et al.*, 1990).

A highly effective CMI was observed in Desi suggesting the most resistant nature of native pigs. In Duroc x Large White Yorkshire cross breeds, a good CMI response was noticed. The theoretical basis may be the buffering effect of heterosis in F_1 individuals which provides the ability to withstand adverse environmental conditions. Low immune level was recorded in Large White Yorkshire pointing the poor adaptability and susceptibility to diseases in pure exotic breeds in tropical climate.

The least square mean for various litter traits had no significance on cutaneous response to phytohaemagglutinin on 24, 48 and 72 hour post- injection. Future investigations in more animals and across a wider age range are necessary.

The results identify immune functioning in swine and importance to establish methodologies that might be needed to develop new strains of pigs resistant to diseases. Scientists should identify breeding stock that is healthier, by virtue of its innate disease resistance properties, which will improve the economy of production program.

5.3 HUMORAL IMMUNE RESPONSE

The seventh day post -inoculation antibody titre ($1 + \log_e$) in Desi had least square mean of 3.625 ± 0.176 with a range of 2.39 – 4.47 and 14^{th} day response had mean of 3.290 ± 0.170 (2.39-4.47) . The mean response on twenty first day was 3.213 ± 0.189 (2.39-4.47).

In Large White Yorkshire, the overall antibody titre on 7^{th} day had least square mean of 2.718 ± 0.141 (1.69-3.08). Antibody titre value had a mean of 2.732 ± 0.410 (1.69-3.08) on 14^{th} day. The mean response on 21^{st} day was 2.410 ± 0.409 (1.69-3.08).

In Duroc x Large White Yorkshire cross breds, antibody titre values had mean of 3.510 ± 0.419 (2.39-4.47), 3.207 ± 0.169 (2.39-4.47), 2.918 ± 0.174 (2.39-4.47) on 7^{th}, 14^{th} and 21^{st} day respectively. This is in concordance with the findings of Rajan (2002).

Meeker *et al.* (1987) found that the relationship between humoral immune response and growth traits were negatively associated. Generation and maintenance of immune response to one antigen (keyhole limpet hemocyanin) in swine was observed by Binns *et al.* (1988). A moderate antibody response to specific monomorphic antigen expressed on peripheral blood lymphocytes (PBL) and red blood cells (RBC) was noticed in sera of ewes and does by Mac Laren *et al.* (1992). van Heugten *et al.* (1994) reported a high level humoral immune response after injecting sheep RBC in weanling pigs. Rajan (2002) reported that different litter sizes at birth and weaning had no significant effect on seventh, fourteenth and twenty first day post-inoculation response.

Antibody response to sheep RBC on 7^{th}, 14^{th} and 21^{st} day associated negatively with litter traits.

It has been found that litters which were having a better birth weight showed a better antibody response. It could be because of the reason that better body weight at birth is a sign of better health.

The study demonstrated that indicators of genetic resistance like elicitation of humoral immune response when exposed to specific antigen would make selection for resistance more rapid and successful. The variation in quantity and persistency of response in three genetic groups may be due to influence by multiple genes underlying immune traits. Results suggest selection for high or low antibody response in young piglets in early or late antibody production. To maximize the efficiency of selection for early immune response, breeder must determine the optimum age, timing of antibody evaluation in given population

and these values must be revalidated and updated as selection proceeds. Much more work needs to be done in this area.

5.4 SIRE EFFECTS

The effects of sires within Desi, Large White Yorkshire and Duroc x Large White Yorkshire were found to be highly significant ($P<0.01$) on antibody response to sheep RBC on 7^{th}, 14^{th} and 21^{st} day. Similar findings were reported by Saxena *et al.* (1997).

Mortensen *et al.* (2004) were able to detect significant sire effects on antibody response to *Mycobacterium paratuberculosis* in Danish Holstein cows.

Variation in immune response with respect to sire point out the high degree of heritability associated with immune response traits. This measure of paternal effects or variation in genotype can be assigned to genetic influence on immune traits.

5.5 ENTERITIS AND PRE-WEANING MORTALITY

The association of cutaneous response to PHA-M ,antibody response to sheep RBC with pre-weaning mortality among littermates and enteritis were found to be non significant in all the three genetic groups. This is in conformation with the observation of Lillehoj (1991) and Lamont (1998).

Genetic and environmental variables influence animal's resistance to infectious disease. Understanding the mechanisms underlying disease incidence is likely to lead in long term to therapeutically useful intervention for infectious and autoimmune diseases. There is great potential for genetic studies of disease

resistance to reveal novel defence mechanisms and can lead to new biotherapeutic approaches and cost effective control mechanisms.

5.6 CORRELATION ANALYSIS

Correlation analysis between immune response traits and weaning body weight, pre-weaning mortality revealed a non-significant association. Correlation among immune response traits were significant. This result is in accordance with the studies made by Mazumdar *et al.* (2005) and contradictory to the observation of Brown (2005).

The significant correlation between immune response traits underline the analogy with respect to CMI and humoral immunity and sensitization of T-cells and B-cells by particular type antigen. Unlike immunogens that activate only the lymphocyte clones bearing the appropriate antigen receptor, polyclonal activators stimulate both B-cell and T-cell clones regardless of antigenic specificity. It is apparent that two immune systems interact intimately. The cytokine messengers triggered by certain elements of antigen are common both in cell-mediated and humoral immune responses.

Genetic correlations are determined by statistical procedures to estimate the probability that many of the same genes affect two traits. The other is to conduct experiments where selection is practiced for only one trait and then determine the correlated response of other traits as progress is made in selection. Genetic correlations among traits are an indication of how two traits will respond together when selection for either one is practiced. If the genetic correlation between two traits is positive, then favourable response to selection by one trait will be echoed by the other. If the genetic correlation is negative, then when one improves because of selection, the other will decline. A negative or positive genetic correlation between two traits doesn't necessarily mean that association

of the two traits is desirable or undesirable. Sometimes, even though the genetic correlation is negative it is considered as favourable.

5.7 HERITABILITY ESTIMATION

Heritability estimates of 0.41 ± 0.30, 0.72 ± 0.35, 0.64 ± 0.25, 0.51 ± 0.20, 0.84 ± 0.28, 0.87 ± 0.39, 0.81 ± 0.33 were noted for pre-injection skin thickness, cutaneous response to phytohaemagglutinin at 24, 48, 72 hour post-injection and antibody response to sheep RBC on seventh, 14^{th}, 21^{st} day post-inoculation. This study provides supportive evidence for the findings of Rajan (2002).

The effects of various genetic and nongenetic factors on immune response to sheep RBCs in guinea fowl were estimated by Saxena *et al.* (1997). The estimate of heritability for immune response to sheep RBCs in guinea fowl was 0.35 ± 0.17. The heritability of responsiveness to diphtheria and tetanus, as estimated by parent-offspring regression, was 0.21 ± 0.51 and 1.21 ± 0.40 (Raberg *et al.*, 2003). There was little evidence that natural variation in antibody responsiveness to these antigens reflected nutritional conditions during early life, responsiveness to at least one of the antigens (tetanus) had a strong genetic component.

Mortensen *et al.* (2004) estimated the genetic variation and heritability of the ability to establish an immune response by producing antibodies to *Mycobacterium paratuberculosis*. The bivariate model with daily milk yield and optical density as dependent variables showed a significant heritability of the ability to produce *Mycobacterium paratuberculosis* antibodies (genetic variance = 0.054) and a non-significant genetic correlation of –0.037 between daily milk yield and optical density. When a sire model was used the estimated heritability was 0.091.

High heritabilities were estimated for immune response traits in the present study. In quite heritable traits, differences in breeding values of animals have large effect on performance and differences in environments are less important. High heritability estimates indicate the additive gene action underlying immune traits, high correlation between phenotype and genotype, mating best to best can make significant genetic change. Due to highly heritable nature of immune response traits, in conjunction with production traits selection for immune traits can be practiced in swine production enterprises.

Summary

6. SUMMARY

Resistance to disease in general is known to be a genetically viable characteristic of domestic animals. Recognition of this fact makes it possible to think in terms of breeding selectively for enhanced resistance. Knowledge of the mechanisms through which genetic variation is expressed makes it possible to think in terms of manipulating immune responsiveness selectively in order to improve overall breed resistance.

Magnitude of cell mediated and humoral immune response to specific antigens were assessed in the present study. Relation of immune response traits with economic traits of swine were analyzed and the immune responsiveness in three genetic groups namely Desi, Large White Yorkshire and Duroc x Large White Yorkshire were compared.

1. Litter size at birth, litter size at weaning, birth weight, weaning body weight (kilograms) measured were 9.316 ± 0.531, 6.313 ± 0.238, 0.806 ± 0.312, 8.316 ± 0.136 for Desi 12.306 ± 0.312, 10.112 ± 0.112, 1.631 ± 0.221, 11.106 ± 0.126 for Large White Yorkshire and 14.132 ± 0.432, 10.136 ± 0.216, 1.890 ± 0.126, 14.312 ± 0.136 for Duroc x Large White Yorkshire respectively.

2. The least square mean for birth weight, weaning body weight, litter size at birth, litter size at weaning had probability values of $P = 0.0042$, $P = 0.0093$, $P= 0.0081$, $P = 0.0068$ respectively. The litter traits were highly significantly different in different genetic groups ($P< 0.01$).

3. The least square mean for different litter traits had no significance on cutaneous response to PHA-M and antibody response to sheep RBC.

4. The increase in skin thickness at 24, 48 and 72 hour post- injection was highest in Desi, medium in Duroc x Large White Yorkshire and least in Large White Yorkshire.

5. Cutaneous response to PHA-M and occurrence of enteritis, pre- weaning mortality associated non-significantly.

6. Naturally occurring antibodies could not be detected in three genetic groups as evidenced by zero pre-inoculation titre.

7. The maximum titre giving the reaction in microhaemolytic assay on 7^{th} , 14^{th} and 21^{st} day serum samples was in Desi. Serum samples from Duroc x Large White Yorkshire had medium mean antibody titre and that of large White Yorkshire had the least titre.

8. Peak antibody response to SRBC was obtained on seventh day post-inoculation in all the three genetic groups.

9. Sire effect was highly significant ($P<0.01$) with antibody response on 7^{th}, 14^{th} and 21^{st} day in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets.

10. Correlations of antibody response on seventh, 14^{th} and 21^{st} day were non - significant with occurrence of enteritis and pre- weaning mortality.

11. High heritability estimates of 0.41 ± 0.30, 0.72 ± 0.35, 0.64 ± 0.25, 0.51 ± 0.20, 0.84 ± 0.28, 0.87 ± 0.39, 0.81 ± 0.33 were noted for pre-injection skin thickness, cutaneous response to PHA-M at 24, 48, 72 hour post-inoculation and antibody response to sheep RBC on 7^{th}, 14^{th}, 21^{st} day post-inoculation.

12. Antibody response to sheep RBC on 7^{th}, 14^{th}, 21^{st} day with cutaneous response to PHA-M at 72 hour associated non-significantly. Correlation between antibody response to sheep RBC on 7^{th} day and cutaneous response to PHA-M at 24 hour (-0.219), 48 hour (-0.209) were negative and significant ($P<0.05$).

13. Correlations of cutaneous response to PHA-M at 24, 48 and 72 hour with weaning body weight, pre-weaning mortality were found to be non-significant. Correlations between antibody response to sheep RBC on 7^{th}, 14^{th}, 21^{st} day and birth weight were positive and highly significant ($P<0.01$).

14. Correlation of cutaneous response to PHA-M at 72 hour with birth weight (0.321) was positive and highly significant ($P<0.01$). Correlation between cutaneous response to phytohaemagglutinin at 48 hour and birth weight (-0.254) was negative and highly significant ($P<0.01$).

References

REFERENCES

Asahi, H., Kawabata, M., Sendo, F., Naiki, M., Hosaka, Y. and Kobayakawa, T. 1984. The presence of anti-sheep red blood cell heterophile antibodies and their characteristics in murine *Schistosomiasis japonia*. *Microbiol. Immunol.* 28: 1241-1256

Biberthaler, P., Bogner, V., Baker, H.V., Lopez, M.C., Neth, P., Kanz, K.G., Mutschler, W., Jochum, M. and Moldawer, L.L. 2005. Genome-wide monocytic mRNA expression in polytrauma patients for identification of clinical outcome. *Shock* 24: 11-19

Binns, R.M. and Licence, S.T. 1981. A major subpopulation of Fc receptor bearing lymphocytes revealed by rosette formation in dextran media: studies with pig, sheep and rat lymphocytes. *J. Immunol. Methods.* 43: 153-162

Binns, R.M., Licence, S.T., Gurner, B.W. and Coombs, R.R. 1983. The demonstration of sIg, MHC and T cell antigens and Fc receptors on the lymphocyte surface by antiglobulin rosetting reactions: some technical considerations. *J. Immunol. Methods* 63: 69-80

Binns, R.M., Pabst, R. and Licence, S.T. 1988. Subpopulations of T lymphocytes emigrating in venous blood draining pig thymus labeled *in vivo* with flurochrome. *Immunology* 63: 261-267

Brown, J.M. 2005. Exogenous administration of immunomodulatory therapies in hematopoietic cell transplantation: an infectious disease perspective. *Curr. Opin. Infect. Dis.* 18: 352-358

Buhler, L., Friedman, T., Iacomini, J. and Cooper, D.K. 1999. Xenotransplantation-State of the art - update 1999. *Front. Biosci.* 14: 16-32

Campbell, K.L. and George, J.W. 1984. Application of the enzyme-linked immunosorbent assay to detect canine erythrocyte antibodies. *Am. J. Vet. Res.* 45: 747-750

Carruthers, V.R. and Morris, C.A. 1988. Quantity and composition of digesta in the reticulo-rumen of cows offered two diets differing in K:Na ratio. *Newzealand J. Agri. Res.* 31: 121-127

Cerrone, M.C. and Kuhn, R.E. 1991. Description of a urease-based micro ELISA for the analysis of limiting dilution microcultures. *J. Immunol. Methods* 138: 65-75

Chow, C.C., Clermont, G., Kumar, R., Lagoa, C., Tawadrous, Z., Gallo, D., Betten, B., Bartels, J., Constantine, G., Fink, M.P., Billiar, T.R. and Vodovotz, Y. 2005. The acute inflammatory response in diverse shock states. *Shock* 24: 74-84

Cox, K.O. and Cox, J. 1981. Specificity of autoantibodies of autoantibody-specific suppressor cells elicited by RBC from various strains of rats. *Aust. J. Exp. Biol. Med. Sci.* 59: 55-61

Cox, K.O. and McAuliffe, A. 1983. Induction and suppression of red blood cell auto-antibodies in mice injected with rat bromelain treated red blood cells. *Cell Immunol.* 81: 22-27

Eguchi, M., Kuniyasu, C. and Kishima, M. 1988. Passive hemagglutination test for detection of antibodies against *Taylorella equigenitalis* in sera of mares. *Vet. Microbiol.* 18: 155-161

Ekkel, E.D., Kuypers, A.H., Counotte, G.H. and Tielen, M.J. 1995. The phytohaemagglutination (PHA) skin test as an indicator of stress-induced changes in immune reactivity in pigs. *Vet. Q.* 17: 143-146

Evans, A.S. and Niederman, J.C. 1982. EBV-IgA and new heterophile antibody tests in diagnosis of infectious mononucleosis. *Am. J. Clin. Pathol.* 77: 555-560

Ficinska, J., Van Minnebruggen, G., Nauwynck, H.J., Bienkowska-Szewczyk, K. and Favoreel, H.W. 2005. Pseudorabies virus glycoprotein gD contains a functional endocytosis motif that acts in concert with endocytosis motif in gB to drive internalization of antibody-antigen complexes from the surface of infected monocytes. *J. Virol.* 79: 7248-7254

Gombart, A.F., Borregaard, N. and Koeffler, H.P. 2005. Human cathelicidin antimicrobial peptide (CAMP) gene is a direct target of the vitamin D receptor and is strongly up-regulated in myeloid cells by 1,25-dihydroxy vitamin D3. *Faseb J.* 19: 1067-1077

Hagberg, M., Wattrang, E., Niskanen, R., Traven, M., Hoglund, J. and Lunden, A. 2005. Mononuclear cell subsets in bronchoalveolar lavage fluid during *Dictyocaulus viviparus* infection of calves: a potential role for gamma/delta TCR-expressing cells in airway immune responses? *Parasite Immunol.* 27: 151-161

Hammam, H., Refai, M., Bisping, W. and Kirpal, G. 1989. Studies on the efficiency of absorbed bovine PPD in tuberculin and serological tests. *Zentralbl. Veterinarmed.* 36: 175-179

Harvey, W.R.1985.Least squares analysis of data with unequal subclass number,USDA,ARS.I-4

Hoesel, L.M., Neff, T.A., Neff, S.B., Younger, J.G., Olle, E.W., Gao, H., Pianko, M.J., Bernacki, K.D., Sarma, J.V. and Ward, P.A. 2005. Harmful and protective roles of neutrophils in sepsis. *Shock* 24: 40-47

Huang, C.L., Yang, Y.H., Wang, L.C., Lin, Y.T., Tsai, Y.Y. and Chiang, B.L. 2005. Humoral and cellular immune response after measles vaccination in Taiwan. *J. Microbiol. Immunol. Infect.* 38: 169-175

Inger, E.L., Eva, W., Lena, M., Maria, M., Lena, A.E., Leif, A. and Caroline, F. 1998. Mapping quantitative trait loci for immune capacity in the pig. *J. Immunol.* 161: 829-835

Ishizawa, S. and Daniels, J.C. 1980. Cellular and humoral hypersensitivity to adrenal antigen in experimental adrenalitis. *Immunol. Commun.* 9: 437-451

Jensen, P.T. and Christensen, K. 1981. Genetic studies on the *in vitro* PHA - transformation of porcine blood lymphocytes. *Vet. Immunol. Immunopathol.* 2: 133-143

Jutel, M., Akadis, M., Blaser, K. and Akdis, C.A. 2005. Are regulatory T cells the target of venom immunotherapy? *Curr. Opin. Allergy Clin. Immunol.* 5: 365-369

Kasukawa, R. 2001. Advances in immuno-serology tests in clinical medicine: auto - antibodies, immune complexes and DNA analysis. *Rinsho Byori.* 49: 219-224

Kawano, S., Kohmura, H., Ohta, S. and Takahashi, N. 1990. Immunological properties of BMY - 28100 and Cefepime. *Jpn. J. Antibiol.* 43: 1275-1288

Kawano, S., Kohmura, H., Ohta, S. and Takahashi, N. 1990. Antigenicity study of buspirone hydrochloride in guinea pigs and mice. *J. Toxicol. Sci.* 1: 15-30

Kean, R.P., Cahancer, A., Freeman, A.E. and Lamont, S.J. 1994. Direct and correlated responses to multi trait divergent selection for immunocompetence. *Poult. Sci.* 73: 18-32

Kouchi, Y., Imamura, K., Izumi, K. and Yoshida, R. 1989. Antigenicity tests on propiverine hydrochloride in guinea pigs and mice. *J. Toxicol. Sci.* 14: 143-151

Lamont, S.J. 1998. Impact of genetics on disease resistance. *Poult. Sci.* 77: 1111-1118

Lessard, M., Taylor, A.A., Braithwaite, L. and Weary, D.M. 2002. Humoral and cellular immune responses of piglets after castration at different ages. *Can. J. Anim. Sci.* 82: 519-526

Li, T.Y. and Yeh, M.L. 2005. The application of qi-gong therapy to health care. *Hu Li Za Zhi.* 52: 65-70

Lillehoj, H.S. 1991. Lymphocytes involved in cell mediated immune responses and methods to assess cell immunity. *Poult. Sci.* 70: 1154-1164

Mac Laren, L.A., Anderson, G.B., Bon Durant, R.H., Edmondson, A.J. and Bernoco, D. 1992. Maternal serum reactivity to species specific antigens in sheep-goat interspecific pregnancy. *Biol. Reprod.* 46: 1-9

Mazumdar, T., Anam, K. and Ali, N. 2005. Influence of phospholipid composition on the adjuvanticity and protective efficacy of liposome encapsulated *Leishmania donovani* antigens.*J. Parasitol.* 91: 269-274

McIntosh, J.T., Marris, C.A., Cockrem, F.R.M., McLaren, R.D. and Gravett, I.M. 1988. Genetics of susceptibility to bloat in cattle. IV. Girth measurements, bloat scores, saliva proteins, blood components, milk production, live weight, and intake data. *Newzealand. J. Agri. Res.* 31: 133-144

Meeker, D.L., Rothschild, M.F., Christian, L.L., Warner, C.M. and Hill, H.T. 1987. Genetic control of immune response to pseudorabies and atrophic rhinitis vaccines. 1. Heterosis, general combining ability and relationship to growth and back fat. *J. Anim. Sci.* 64:407-413

Meier, S. and Drake, C. 1982. Development of a latex - conjugated immuno-cytological marker for scanning election microscopic analysis of quail-chick chimeras. *J. Exp. Zool.* 224: 25-37

Miller, F.R. and Blazkovec, A.A. 1979. Guinea pig cell - mediated tumor immunity: the chromium release assay detects both cytolysis and serum blocking for synergenic chemically - induced tumors. *Immunol. Commun.* 8: 31-47

Mohmmed, A., Kishore, S., Patra, K.P., Dasaradhi, P.V., Malhotra, P. and Chauhan, V.S. 2005. Identification of Karyopherin beta as an immunogenic antigen of the malaria parasite using immune mice and human sera. *Parasite Immunol.* 27: 197-203

Molina, P.E. 2005. Neurobiology of the stress response: contribution of the sympathetic nervous system to the neuroimmuno axis in traumatic injury. *Shock* 24: 3-10

Morrow-Tesch, J.L., McGlone, J.J. and Salak - Johnson, J.L. 1994. Heat and social stress effects on pig immune measures. *J. Anim. Sci.* 72: 2599-2609

Mortensen, H., Nielsen, S.S. and Berg, P. 2004. Genetic variation and heritability of antibody response to *Mycobacterium paratuberculosis* in Danish Holstein cows. *J.Dairy.Sci.* 87:2108-2113

Nakano, Y., Kohayashi, Y., Tsujino, M. and Hagano, K. 1987. Antigenicity study of rokitamycin. *Jpn. J. Antibiot.* 40: 608-612

Nesterenko, V.G., Novikova, T.K., Fontalin, L.N., Rabakova, E.I., Grunev, S., Wechnik, E. and Sidorova, E.V. 1983. ACA - 1, a new differentiation antigen of activated lymphoid cells. *Cell Immunol.* 79: 253-264

Nestor, K.E., Anderson, J.W. and Patterson, R.A. 2000. Changes in genetic parameters over thirty generations of selection for increased body weight. *Poult. Sci.* 79: 445-452

Nguven, N.T., Luketich, J.D., Schatz, S., Tran, Q., Ho, H.S. and Schaver, P.R. 1999. Effect of open and laparoscopic surgery on cellular immunity in a swine model. *Surg. Laparosc. Endosc. Percutan Tech.* 9: 176-180

Ochiya, T., Baba, T., Onozaki, K., Yaoita, H., Vyeno, K. and Hashimoto, T. 1983. Suppression of cutaneous delayed type hypersensitivity by macrophage disappearance reaction. *Cell Immunol.* 81: 134-143

Ponelics, N., Hirsch, T., Krehmeier, U., Denz, C., Patel, M.B. and Majetschak, M. 2005. Cytosolic ubiquitin and ubiquitylation rates in human peripheral blood mononuclear cells during sepsis. *Shock* 24: 20-25

Poutrel, B. and Caffin, J.P. 1983. A sensitive microassay for the determination of hemolytic complement activity in bovine milk. *Vet. Immunol. Immunopathol.* 5: 177-184

Raberg, L., Stjernman, M. and Hasselquist, D. 2003. Immune responsiveness in adult blue tits: heritability and effects of nutritional status during ontogeny. *Oecologia* 136: 360-364

Rajan, M.R. 2002. Porcine immune response as marker trait for selective breeding. Ph.D. thesis, Kerala Agricultural University, Thrissur. p. 143

Rao, T.D., Maghazachi, A.A., Fari, A.M., Basch, R.S. and Philips - Quagliata, J.M. 1992. T 560: an (H-2b x H-2a) F1 hybrid, phosphoryl choline (PC) -

binding, murine B cell lymphoma that bears receptors for Ig A and Ig G, presents antigen and secretes IL - 4. *Int. Immunol.* 4: 107-118

Raszyk, J. and Pillich, J. 1990. A simple immunologic skin test in piglets. *Vet. Med.* 35: 553-560

Rice, T., An, P., Gagnon, J., Lean, A.S., Skinner, J.S., Wilmore, J.H., Bouchard, C. and Rao, D.C. 2002. Heritability of HR and BP response to exercise training in the heritage family study. *Med. Sci. Sports Excerc.* 34: 972-979

Richters, C.D., Hoekstra, M.J., du Pont, J.S., Kreis, R.W. and Kamperdijk, E.W. 2005. Immunology of skin transplantation. *Clin. Dermatol.* 23: 338-342

Rimler, R.B., Rebers, P.A. and Phillips, M. 1984. Lipopolysaccharides of the Heddleston serotypes of *Pasteurella multocida*. *Am. J. Vet. Res.* 45: 759-763

Samarineau, M.P., Pascale, F. and Enache, M. 2000. Immunoenzymate detection of gamma interferon in swine lymphocyte culture stimulated with phytohaemagglutinin (PHA). *Stud. Res. Vet. Med.* 7: 67-70

Sawata, A., Kume, K. and Nakase, Y. 1982. Hemagglutinin of *Haemophilus paragallinarum* serotype 2 organisms: occurrence and immunologic properties of hemagglutinin. *Am. J. Vet. Res.* 43: 1311-1314

Saxena, V.K., Singh, H., Pal, S.K. and Kumar, S. 1997. Genetic studies on primary antibody response to sheep erythrocytes in guinea fowl. *Br.Poult.Sci.* 38:156-158

Scheepens, C.J., Hessing, M.J., Hensen, E.J. and Henricks, P.A. 1994. Effect of climatic stress on the immunological reactivity of weaned pigs. *Vet. Q.* 16: 137-143

Shibata, K., Matsuda, K., Wada, H., Aruga, F. and Yamada, Y. 1991. *Antigenicity studies on catena - (s) - [mu - (N alpha - (3 - amino propionyl) histidinato (2-) - N1, N2, O:N tau) - Zinc]. Arzneimittel forschung.* 41: 1048-1052

Singh, G.S., Chauhan, H.V., Jha, G.J. and Singh, K.K. 1990. Immunosuppression due to chronic ochratoxicosis in broiler chicks. *J. Comp. Pathol.* 103: 399-410

Sivaraman, G.K., Kumar, S., Saxena, V.K., Singh, N.S. and Shivakumar, B.M. 2005. Genetics of immunocompetent traits in a synthetic broiler dam. *Br. Poult. Sci. 46: 169-174*

Splichalova, A., Splichal, I., Ivebichavsky, I. and Hojna, H. 2004. Expression of inflammatory markers in pig amnion after intra amniotic infection with nonpathogenic or enteropathogenic *Escherichia coli. Folia Microbiol.* 49:751-756

Takahashi, K. and Avai, Y. 1983. Antigenicity of suloctidil. *J. Toxicol. Sci.* 8: 231-242

Takemoto, M., Mat Suo, K., Katoh, S. and Yoshida, R. 1988. Antigenicity test of cefodizime sodium. *J. Toxicol. Sci.* 1: 231-243

Takemoto, M., Matsuo, K., Oguro, M. and Kouchi, Y. 1994. Antigenicity tests of tazobactam/piperacillin, tazobactam and piperacillin. *J. Toxicol. Sci.* 2: 249-262

Tuchscherer, M., Kanitz, E., Otten, W. and Tuchscherer, 2002. Effect of pre-natal stress on cellular and humoral immune responses in neonatal pigs. *Vet. Immunol. Immuno Pathol.* 86: 195-203

van der Meijden, A.P., de Jong, W.H., de Boer, E.C., Steerenberg, P.A., Debruyne, F.M. and Ruitenberg, E.J. 1989. Immunological aspects of intravesical administration of Bacillus Calmette Guerin (BCG) in the guinea pig. *Urol. Res.* 17: 47-55

van der waaiji, E.H., Bijma, P., Bishop, S.C. and Vanarendonk, J.A. 2000. Modeling selection for production traits under constant infection pressure. *J. Anim. Sci.* 78: 2809-2820.

van Heugten, E., Spears, J.W., Coffey, M.T., Kegley, E.B. and Qureshi, M.A. 1994. The effect of methionine and aflatoxin on immune function in weanling pigs. *J. Anim. Sci.* 72: 658-664

Visscher, A.H., Janes, L.L.G., Niewold, T.A. and Greef, K.H. 2002. Disease incidence and immunological traits for the selection of healthy pigs. A review. *Vet. Q.* 24: 29-34

Waylan, A.T., Kayser, J.P., Gnad, D.P., Higgins, J.J., Starkey, J.D., Sissom, E.K., Woodworth, J.C. and Johnson, B.J. 2005. Effects of L-carnitine on fetal growth and the IGF system in pigs. *J. Anim. Sci.* 83:1824-1831

Wilson, A.B. and Coombs, R.R. 1985. Fc gamma - receptor - bearing, non-B lymphocytes in human peripheral blood: cytophilic immunoglobulin binds almost exclusively to large granular lymphocytes. *Cell Immunol.* 90: 196-207

Yamamoto, R., Uenishi, H., Hatsuse, H., Sato, E., Awata, T., Yasue, H. and Takagaki, Y. 2005. TRAV gene usage in pig T-cell receptor alpha c DNA. *Immunogenetics*. 57: 219-225

Yang, H., Parkhouse, R.M. and Wileman, T. 2005. Monoclonal antibodies that identify the CD3 molecules expressed specifically at the surface of porcine gammadelta - T cells. *Immunology.* 115: 189-196

Yen, J.T. and Pond, W.G. 1987. Effect of dietary supplementation with vitamin C or carbadox on weanling pigs subjected to crowding stress. *J. Anim. Sci.* 64: 1672-1681

Yi, S., Feng, X., Wang, Y., Kay, T.W., Wong, Y. and Connell, P.J. 1999. $CD4^+$ cells play a major role in the Fas/Fas ligand lytic pathway. *Transplantation* 67: 435-443

EVALUATION OF PORCINE IMMUNE RESPONSES AMONG DIFFERENT GENETIC GROUPS

JEEBA K. GEORGE

Abstract of the thesis submitted in partial fulfilment of the requirement for the degree of

Master of Veterinary Science

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University, Thrissur

2005

Department of Animal Breeding and Genetics
COLLEGE OF VETERINARY AND ANIMAL SCIENCES
MANNUTHY, THRISSUR-680651
KERALA, INDIA

ABSTRACT

Porcine immune responses were evaluated using PHA skin test and microhaemolytic assay in this study. Investigation was undertaken in three genetic groups namely Desi, Large White Yorkshire and Duroc x Large White Yorkshire. The economic traits studied were birth weight, litter size at birth, weaning litter size and weaning body weight. The cell-mediated immune response was assessed by noting cutaneous response to intradermal injection of phytohaemagglutinin. Humoral immune response was assessed by noting antibody response to sheep red blood cells. Correlation of immune response with growth, disease occurrence and mortality among the littermates were also evaluated.

The highest birth weight, body weight at weaning, litter size at birth and weaning were recorded in Duroc x Large White Yorkshire, medium in Large White Yorkshire and least in Desi. The increase in skin thickness at 24, 48 and 72 hour post- injection of PHA-M was highest in Desi, medium in Duroc x Large White Yorkshire and least in Large White Yorkshire. The correlations of cutaneous response to phytohaemagglutinin with pre- weaning mortality among littermates and enteritis were found to be non-significant in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets.

Among three genetic groups, serum samples from Desi piglets had a higher mean antibody titre on 7^{th}, 14^{th}, 21^{st} day than the other two. Medium titre was noted in Duroc x Large White Yorkshire and least in Large White Yorkshire. Sire effect was highly significant with antibody response on seventh, fourteenth and 21^{st} day post inoculation. The correlations of antibody response to sheep RBC with pre- weaning mortality among littermates and enteritis were also found to be non-significant in Desi, Large White Yorkshire and Duroc x Large White Yorkshire piglets. The effects of sires within Desi, Large White Yorkshire and

Duroc x Large White Yorkshire were found to be highly significant ($P<0.01$) on antibody response to sheep RBC on 7^{th}, 14^{th} and 21^{st} day. Different litter traits had no significant effect on cutaneous response to PHA-M and antibody response to sheep RBC.

High heritabilities were estimated for pre-injection skin thickness, cutaneous response to PHA-M at 24, 48, 72 hour post-injection and antibody response to sheep red blood cells on 7^{th}, 14^{th}, 21^{st} day post- inoculation.

Correlations of antibody response to sheep RBC on 14^{th} day with cutaneous response to phytohaemagglutinin at 24 hour and 48 hour were found to be non-significant. Antibody response to sheep RBC on 21^{st} day correlated non-significantly with cutaneous response to PHA-M at 24 hour and 48 hour.

Antibody response to sheep RBC on 7^{th}, 14^{th} and 21^{st} day associated non-significantly with weaning body weight and pre-weaning mortality. Even though correlations were non-significant, they revealed a negative trend. The association between cutaneous response to PHA-M at 24 hour and birth weight was non-significant.