Single versus double testing of meat-juice samples for Salmonella antibodies, in the Danish pig-herd surveillance programme

In Denmark, a national serological surveillance-and-control programme for Salmonella in pigs has been in operation since 1995. The programme is based on the Danish mix-ELISA and uses double testing (two ELISA-wells used per sample) of meat-juice samples taken in relation to slaughter. All herds are classified monthly into one of the three levels; the classification is based on the percentage of positive serological results in the previous 3 months. In connection with evaluation of the programme in 2001, we investigated whether single testing (testing in one well only) could be expected to be sufficiently precise compared to double testing. Data from the year 2000 were used, and mathematical modelling. Single testing was simulated by randomised selection of one of the two results in the double testing. A slight increase in the prevalence of Salmonella-positive samples (1.02-1.09 times more through the four quarters of the year 2000) was found in the simulated single testing, as compared to the double testing. Around 0.5% of the herds would be allocated to another herd level in single testing-almost equal numbers one level up and one level down. No herd being seronegative in double testing would be allocated to levels 2 or 3 (herds with >40 or >70%, respectively, serological reactors) in single testing. The prevalence of "false-positive" diagnoses (positive in single testing and negative in double testing) and inversely defined "false-negative" diagnoses varied from 4.2 to 8.7% and from 3.2 to 4.5%, respectively, through the four quarters of the year 2000. The probability of allocating a herd to a wrong level due to sampling error was on the average 6.2 (varying from 1.66 to over 100) times higher than the probability of allocating a herd to a wrong level due to the test inaccuracy introduced by going from double to single testing. This is, however, an average; a herd with a true prevalence close to one of the level border cut-offs (40 and 70% weighted seroprevalence, respectively) would have a higher risk of being allocated to a wrong level than a herd with a true prevalence far from the level border cut-offs. The results are based
on the current Danish sample sizes in the surveillance scheme, which implies that 60, 75 or 100 samples are taken annually in a herd, depending on its size. Other sample sizes would produce other results.

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The serologic response to Salmonella enteritidis and Salmonella typhimurium in experimentally infected chickens, followed by an indirect lipopolysaccharide enzyme-linked immunosorbent assay and bacteriologic examinations through a one-year period

Three groups of 100 individually marked salmonella-free chickens were followed for a period of 53 wk. The chickens were infected as day olds by crop instillation of 101 colony-forming units: one group with Salmonella enteritidis and a second group with Salmonella typhimurium. A third group was kept uninfected as controls. The groups were monitored bacteriologically by examination of cloacal swabs and organs and serologically by examination of serum and egg yolk by a lipopolysaccharide enzyme-linked immunosorbent assay throughout the period. Within the first week, 100% of birds in both infected groups were excreting salmonella bacteria in the feces. However, the number of fecal excretors declined rapidly with time, down to 6% in 16 wk for S. typhimurium and down to a similar level within the first 3 wk for S. enteritidis. For the latter, relapses with up to 40% positive birds were observed at the onset of egg production. For both S. typhimurium and S. enteritidis, positive bacteriologic cultures were obtained by sampling from internal organs at the end of the experiment, more than 1 yr from the time of infection. At the age of 6-7 wk, 50% of the chickens in the two infected groups showed a measurable serologic response in serum samples. The response persisted throughout the study in both serum and egg yolk, samples. The inclusion of serologic methods is a valuable additional tool in the detection of salmonella in poultry, but serology should be used in conjunction with bacteriologic methods in surveillance programs, in particular to detect flocks in early stages of infection before a measurable serologic response has been raised.

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Monitoring porcine reproductive and respiratory syndrome virus infection status in swine herds based on analysis of antibodies in meat juice samples

An indirect ELISA test was developed as a novel tool aimed at monitoring the herd infection status of swine herds. Meat juice samples from pig carcasses were analysed for the presence of antibodies against porcine reproductive and respiratory syndrome virus (PRRSV). A study of samples from herds with known PRRS status was undertaken. The PRRS status of the herds was evaluated based on the analysis of blood samples by another serological test (blocking ELISA) capable of differentiating between infection with PRRSV of the American type and European type. The specificity of the indirect ELISA test on meat juice samples was 0.98. The sensitivity of the test depended on the type of the PRRS strain involved. The apparent prevalence in herds infected with the American type of PRRSV was 0.44. The apparent prevalence in herds infected with the European type of PRRSV was 0.64. Herd level sampling and herd level criteria for assessing the PRRS status of herds by the new test were developed. Herds were classified as PRRS negative or PRRS seropositive based on 10 meat juice samples collected randomly at slaughter throughout a 3-month-period. Herd PRRS status classification by the indirect ELISA was validated in 47 herds by collection of blood samples from the herds. Eighteen herds were classified as PRRS negative by both test systems. Twenty-nine herds were classified as PRRS seropositive by both test systems. Acceptable herd classification was achieved using this test.

Serologic reactions against Salmonella in samples from broiler parent stock with and without preceding colibacillosis: A case-control study

In the Danish Salmonella Control Program, eggs from broiler parent flocks are surveyed by serologic analysis every 4 wk for antibodies against Salmonella lipopolysaccharide O-antigens 1, 4, 5, 9, and 12 (Mix-enzyme-linked immunosorbent assay [ELISA]) and 6 and 7 (Infantis-ELISA). The antibody response is measured in percentage optical density (OD%) of a strong positive reaction, and the cutoff value has been determined to be 40 OD%. Two or more reactors above 40 OD% will place the parent flock under suspicion. There has been concern about possible cross-reactions between Salmonella spp. and other Enterobacteriaceae, e.g., Escherichia coli, because a high specificity of a Salmonella antibody test is desirable. Moreover, false-positive Salmonella results have economic consequences and impede planning the production. A case-control study based on cases of clinical E. coli infections (colibacillosis) from two Danish hatcheries, supplying about 62% of the Danish broiler production, was described. In order to eliminate a possible bias from age and season, the controls were matched on age of the birds and on time of submitting the samples. This study shows that flocks with preceding colibacillosis did not have higher salmonella reactions than matched flocks without a preceding colibacillosis. This observation was confirmed in longitudinal studies.
Evaluation of a serological Salmonella Mix-ELISA for poultry used in a national surveillance programme

A Mix-ELISA using lipopolysaccharide antigens from Salmonella enterica serotype Enteritidis and Typhimurium? was evaluated using samples collected over time in the Danish salmonella surveillance programme for poultry. Serological samples (n = 42813) taken from broiler-breeder flocks after a year of bacteriological monitoring with negative results were used for calculating the flock and individual test specificities, which were 0.997 and 0.999, respectively. Layer flocks from the table egg sector were used for calculation of positive predictive values. In the survey, flocks were examined for salmonella by Mix-ELISA and by faecal culture, and in case of a positive result in either of these a repeated, serological testing was performed, and 60 animals were organ-cultured. If one of these samplings was positive, the flock was declared salmonella infected. In a period of 3 months, 35 flocks were found to be positive in the routine samples. Of these, 32 were serologically positive, 2 both serologically and faecally positive and 1 flock only faecally positive. For flocks serologically positive in the surveillance programme, a positive-predictive value of 0.62 for organ culture positivity was found, and while considering serological follow-up samples, the value was 0.95.

General information

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Evaluation of a polyclonal blocking ELISA and a complement fixation test detecting antibodies to Actinobacillus pleuropneumoniae serotype 2 in pig serum

General information

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Mycoplasma hyopneumoniae infection in pigs; duration of the disease and evaluation of four diagnostic assays

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Nation-wide Salmonella enterica surveillance and control in Danish slaughter swine herds

A nation-wide Salmonella enterica surveillance and control programme was initiated in Danish finishing herds over the first quarter of 1995. In Denmark, all swine for slaughter are identifiable by a unique herd code. For each herd code, and depending on the herd's annual kill, random samples ranging from four to more than 60 swine are obtained quarterly at the abattoir. A meat sample from each pig is frozen, and meat juice (harvested after thawing) is examined for specific antibodies against S. enterica using an indirect enzyme-linked immunosorbent assay (ELISA). The ELISA combines several S. enterica O-antigens, and allows detection of antibody response after a variety of different S. enterica serovar infections. Results are transferred to a central database, which each month (based on meat-juice tests obtained in the previous 13 weeks) assigns all herds into three S. enterica infection levels: Level 1, in which the S. enterica prevalence is deemed low and acceptable; Level 2, where there is a moderate prevalence of S. enterica seroreactors (from > 50% in the smallest to > 10% in the largest herds); Level 3, in which S. enterica seroreactor prevalence is clearly unsatisfactory (> 50% for most herd sizes). Irrespective of Salmonella level, all herds receive a monthly update on the current results of the S. enterica test results. If a herd is categorized in Level 2 or 3, it must receive an advisory visit by a practising veterinarian and a local swine extension specialist, and certain management hygiene precautions must be taken. If a herd is categorized in Level 3, the finishers from the herd must additionally be slaughtered under special hygiene precautions. This is supervised by the veterinary authorities. During 1995, 604 000 samples were tested for S. enterica, corresponding to 3.0% of the total kill. In December 1995, 15 522 herds (representing > 90% of the national production) were categorized into one of the three levels: 14 551 herds (93.7%) in Level 1; 610 herds (3.9%) in Level 2; 361 herds (2.3%) in Level 3. The proportion of serologically positive meat-juice samples collected during 1995 ranged from a mean of 2.9% in smaller herds (101-200 swine slaughtered per year) to 6.1% in relatively large herds (more than 5000 swine slaughtered per year).

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Evaluation of a Polyclonal Blocking ELISA Detecting Antibodies to Actinobacillus pleuropneumoniae Serotype 2 in pig serum

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Contributors: Sørensen, V., Barfod, K., Nielsen, J. P., Feld, N. C.
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Comparison of four different methods for demonstration of Mycoplasma hyopneumoniae in lungs of experimentally inoculated pigs

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Demonstration of Mycoplasma hyopneumoniae by a monoclonal sandwich ELISA

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Contributors: Friis, N. F., Pedersen, M. W., Ahrens, P., Feenstra, A. A., Feld, N. C., Sørensen, V., Barfod, K.
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Evaluation of a monoclonal blocking ELISA detecting antibodies to Mycoplasma hyopneumoniae on a single-pig level

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Contributors: Barfod, K., Sørensen, V., Feld, N. C.
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Evaluation of a Polyclonal Blocking ELISA Detecting Antibodies to Actinobacillus pleuropneumoniae Serotype 2

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Contributors: Sørensen, V., Barfod, K., Nielsen, R., Feld, N. C., Nielsen, J. P., Christensen, J.
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Evaluation of four laboratory methods for demonstration of Mycoplasma hyopneumoniae in the lungs of pigs

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Sammenligning af fire forskellige metoder til påvisning af M. hyopneumoniae

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The humoral immune response to experimental Mycoplasma hyopneumoniae infection in pigs in relation to clinical signs and pathological lesions

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Contributors: Sørensen, V., Barfod, K., Feenstra, A. A., Feld, N. C.
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Application of enzyme-linked immunosorbent assay for the surveillance of Mycoplasma hyopneumoniae infections in pigs

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ELISAs for detection of Salmonella infections in poultry, pigs and cattle

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Contributors: Nielsen, B., Feld, N. C., Hoorfar, J., Lind, P.
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Calculation of Herd Sensitivity and Herd Specificity for a Monoclonal Blocking ELISA Detecting Antibodies to Mycoplasma hyopneumoniae in Pig Serum and Colostrum

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Publication date: 1992

Host publication information
Title of host publication: Proceedings of the International Organization for Mycoplasmology
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Evaluation of a monoclonal blocking ELISA and IHA for antibodies to Mycoplasma hyopneumoniae in SPF-pig herds

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Contributors: Sørensen, V., Barfod, K., Feld, N. C.
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Evaluation of a monoclonal blocking enzyme-linked immunosorbent assay detecting antibodies to Mycoplasma hyopneumoniae in pig serum and colostrum

General information
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