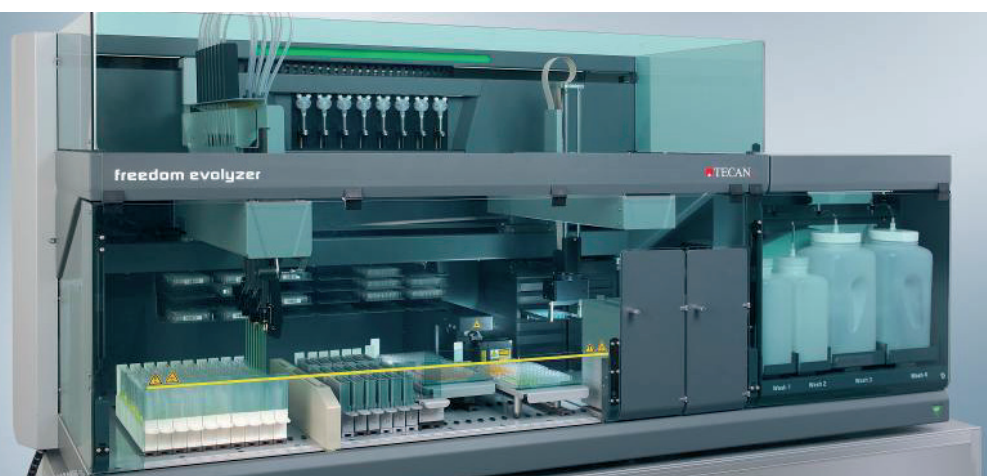


Screening of animal diseases using ELISA

Process automation helps to control PRRS and other diseases in swine herds



Introduction

First described in the US in 1987, Porcine Reproductive and Respiratory Syndrome (PRRS) is a viral swine disease that causes a range of reproductive and respiratory disorders. The major component of the PRRS is reproductive failure, causing premature births, late-term abortions and stillbirths, weak piglets, decreased farrowing rates and delayed return to estrus, with the consequence of significantly reduced production rates. Respiratory problems often lead to secondary infections, and cause increased mortality in young piglets.

Several studies have estimated financial losses of US\$113-236 per sow per year (see Ref. 1, 2) and annual losses of US\$560 million per year in the US alone (see Ref. 3). Since November 1990, when the first case of PRRS was detected in Germany, it has spread throughout Europe causing major production losses. The vaccines that are currently available for PRRS do not offer protection against different strains of the virus, and are therefore not a fully effective method of prevention.

Objectives of the SVA

The Swedish National Veterinary Institute (SVA) in Uppsala is a governmental institute in veterinary medicine with a staff of 400 people. One of SVA's primary objectives is to be well prepared to react immediately to serious contagious and infectious diseases of animals, by rapid and reliable diagnosis.

The EU membership in 1995 brought about a strengthening of the monitoring of swine and poultry for diseases that have not been detected in the country. The urgency of a larger scale testing for the PRRS virus in Sweden has recently increased following the first reports of PRRS outbreaks in eight pig farms in July 2007.

Freedom EVOlyzer[®] configuration

With the increased importance of an efficient monitoring and control procedure for PRRS and other diseases the SVA required a flexible and reliable instrument that would allow a reduction in manual pipetting. The SVA needed an automated platform solution capable of both the application of a wide range of assays with a low number of samples, and

performing a limited range of standard assays on a large number of samples.

In 2005 the threat of an avian influenza pandemic demonstrated the need for large scale testing. The following year, the SVA decided to increase its processing capacity by using a Freedom EVOlyzer® platform with the following configuration: 150 cm worktable, four fixed pipetting tips, 24-plate microplate storage, one ambient temperature incubator, one heated incubator and eight-channel washer.

The SVA now uses the Freedom EVOlyzer workstation for the monitoring of PRRS. The validation of classical swine fever and Aujeszky's disease monitoring have also been completed. All three validated tests are now being performed in the same run. In the framework of control programs, 3000-5000 samples are currently being analyzed yearly.

Procedure description

The IDEXX HerdChek* PRRS 2XR Antibody Test Kit from IDEXX B.V., Netherlands allows detection of antibodies against both European and North American strains of PRRS virus in swine serum by ELISA to detect seroconversion, ie. both infected and vaccinated (immunized) animals. The kit can be used to monitor a suspicious herd or to check whether animals are vaccinated.

Based on the Instructions for Use the serum (diluted in 1:40 using dilution buffer) and the positive and negative controls (undiluted) are pipetted in duplicate into a 96 well format strip plate with 12 strips of eight wells, coated with PRRS antigen and normal cells in alternate rows.

The plate is incubated for 30 minutes at room temperature and washed 3-5 times with wash buffer, after which 100 µl of anti-swine HRP conjugate is added and incubated for 30 min at room temperature. The plate is washed again and 100 µl TMB substrate is added. After 15 min of room temperature incubation the reaction is stopped by adding 100 µl stop solution. The results are obtained by measurement of optical density at 650 nm.



Jane Borg explains to her colleague Gunnel Svedlund the schedule for running ELISA tests on the Freedom EVOlyzer platform.

Summary

Tecan's Freedom EVOlyzer workstation automates ELISAs from start to finish and increases the productivity and efficiency of today's modern clinical, veterinary, pharmaceutical and research laboratories.

The laboratory in the SVA is able to adapt very quickly to daily changes in sample load which is particularly important during disease outbreaks, when the laboratory must quickly deal with changing demands upon throughput and the required range of tests. It is particularly invaluable when the number of samples exceeds what can be processed manually in laboratories like the SVA, where both manual and automated processing are being used. The flexibility of the Freedom EVOlyzer platform is ideal for running a large number of assays on a limited number of samples, or large number of samples on a limited number of assays.

Acknowledgments

We would like to thank Dr Lena Renström from the SVA in Uppsala, Sweden for sharing with us her experience of using the Freedom EVOlyzer workstation in her daily work.

References

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The Freedom EVOlyzer is compliant according to the IVD directive 98/79/EC and is an Annex III-compliant open system for ELISA processing.

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