

# Quality control protocols to avoid reproductive toxicity from plastics

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Magapor, in close collaboration with the University of Zaragoza, has tested and demonstrated the repro-toxic effect of chemical substances contained in plastic materials of semen containers. These substances known as 'Non Intentionally Added Substances' (NIAS), migrate to the content making its effect by direct contact.

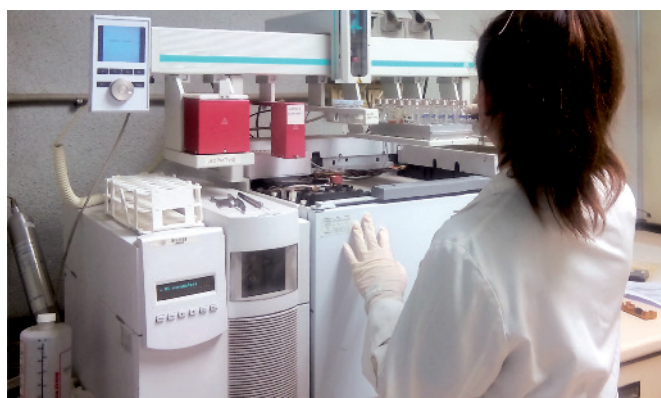
It is not the first case of reproductivity in the livestock industry, but it is the first to be recorded and published for public knowledge. This discovery may serve as a warning for the food and livestock industries (or others) of the need to implement efficient control measures because these toxics can be found in any type of plastic.

The case research involved three years of work in close cooperation between the Technical Service of Magapor and the Department of Analytical Chemistry of the Faculty of Engineering of the University of Zaragoza, ending with the publication of an article in Scientifics reports section of Nature in May 2014: "Compounds from multilayer plastic bags cause reproductive failures in artificial insemination".

## Total traceability

This article explains how, thanks to the total traceability of the products, it was able to detect a problem which affected some batches of semen packs for artificial swine insemination, which were quickly retired from market in a preventing way and communicating simultaneously to all the clients the possible existence of an incident that could affect farm productivity. In this way, the problem was limited to some clients and a period of time equal to three usage weeks.

In intensive pig production systems, 100% of fecundation is performed using artificial insemination. Semen from boars is collected, diluted with an appropriate aqueous solution and finally placed into an



impermeable to gas exchange plastic container (high barrier) to preserve it.

Spermatozoa quality is always checked during processing, and parameters such as motility, morphology, vitality and acrosome integrity are measured before insemination. Diluted semen is only accepted for artificial insemination if these parameters achieve the required thresholds.

Reproductive efficiency in swine is high (target > 90% farrowing rate), but some failures (5-7%) are acceptable because the origin can be related to many reasons such as parity, season, stress, poor semen quality, diseases or others.

The main failures in reproductive performance are related to non-infectious reproductive problems, such as anoestrus, as well as problems related to ovulation, oocyte production, fertilisation, and implantation leading to foetal death or mummification and stillbirth.

From February to April 2010 a dramatic increase in the regular return rate in sow farms was detected. The problem started suddenly in a large swine production system in the South of Spain, with a variable increase in the return rate from 10 to approximately 100%.

Cyclical repetition was observed after 17-22 days of insemination with seminal doses from different insemination centres distributed in plastic packs. In some cases, a reduction in litter size was reported.

All internal and external sperm

quality controls, such as tests for membrane functionality, abnormal forms, concentration, sperm motility and acrosome status, had produced satisfactory results when tests were performed during semen processing, transport and farm storage.

No changes in health status, feeding strategy or body condition of the sows were observed. Neither disease, season, parity, stress, poor semen nor insemination technique could be established as the cause of this problem.

The analysis of the traceability system established a possible relation between the plastic containers and the reproductive failure. This hypothesis was reinforced when suspicious bags were removed and replaced by the previous validated batches, and the farms recovered the normal reproduction rates.

During the chemical analysis of the semen containers unexpected compounds could be identified such as BADGE, a cyclic lactone and an unknown phthalate until then, that leached into the semen at concentrations of 0.2-2.5mg/L.

Repetition grade varied depending on the toxic quantity detected on the analysis, obtaining sow repetition percentages which were from <15% to 80-100%.

At laboratory level, these toxics did not produce any abnormality in routine analysed parameters (motility, agglutination, test of endosmosis, osmotic resistance test, acrosome integrity). Spermatic cells preserved in these containers passed all of the

routine quality control tests, and no differences were observed between the control and the suspicious bags (p. 0.05). In vitro fecundation tests and endocrine profiler panel analysis (EPP) did not show any alterations, whereas the in vivo tests confirmed the described failure.

Despite the strict internal quality controls established by Magapor to validate its materials, this fact caused a leap in protocols, affecting the reproductive results.

Until then no one protocol worldwide had been able to detect these previously unknown toxics.

This is the first time that the connection between toxic compounds migrating to plastic substances and the reproductive toxicity by contact has been proved.

## Conclusion

Detecting any compound or pathogen involves knowing its existence, action mechanism and the effects it produces. Once identified, there should exist the necessary diagnosis methods. It should be pointed out that any compound in enough quantity has toxic effects, having to evaluate the effect of any substances on biological functions considering dosage and the combined effect of the substances.

Later research has demonstrated that not only multilayer containers present these type of toxics, but all plastic materials in general (heat-sealed tubes included), so it is essential that good internal quality control of plastic containers (regardless if they are multilayer or monolayer) is able to detect the minimum change in composition or functioning.

Since this case occurred Magapor has implemented in a complementary way to its quality control protocol the RTC system (Reproductive Toxicity Control), which is the chemical plastic level systematic analysis to be able to identify the presence of unknown substances in its materials. This chemical analysis is highly valued because until that moment these compounds had not been taken into account as they were unknown. ■