

## The feasibility of a swine semen storage system produced with bacteriostatic molecules to control bacterial proliferation

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### Abstract

The development of a sustainable swine industry is driven by current social and environmental concerns. Utilizing antibiotics in semen extenders is one of the issues associated with pig breeding. It causes the emergence of multidrug-resistant bacteria due to its widespread application. In addition, the antibiotic may have an adverse effect on the viability of sperm and the vaginal and cervical microbiota of sows. Consequently, animal reproduction biotechnologies have sought out products with technologies capable of minimizing antibiotic use. In this study, we assessed the efficacy of the Bactibag swine semen storage bag, which features a bacteriostatic mechanism to restrict bacterial growth. Six boars' ejaculates were collected using the "gloved-hand" technique. To produce insemination doses, the ejaculate was assessed for volume, sperm motility, and concentration. The semen was diluted with and without antibiotics (0.25 g/L of gentamicin sulfate) at 37°C at in-house made Beltsville Thawing Solution (BTS). Semen doses were produced with a total motility sperm concentration of  $2.5 \times 10^8$  sperm in a volume of 90 mL, filled into two different plastic semen storage: Bactibag blisters (Bactibag, IMV Technologies, France) and conventional blisters (GTB Bag, IMV Technologies, France), resulting in four experimental groups: semen diluted in BTS with antibiotics and added to the Bactibag blisters (BBA group) and conventional blisters (GTA group) and semen diluted in BTS without antibiotics and added to the Bactibag blisters (BBN group) and conventional blisters (GTN group). The doses of semen were then kept at 16°C for up to 120 hours (5 days). On a plate, bacterial growth was assessed after 72 hours and 120 hours of storage. We found a relationship between the storage blisters, the use of gentamicin, and time. The use of gentamicin in both type of blisters effectively inhibited bacterial growth from storage day 3 to day 5 ( $P < 0.05$ ). After 72 hours of storage, the results of the BBN group were comparable to the GTA group. There were no differences in bacterial growth between 72 and 120 hours of storage in BBA group. At 120 hours of storage, the BBN group was as effective as the GTA group at inhibiting bacterial growth. Upon examining the bacterial growth over the assessed period (from 72h to 120h), we found that Bactibag blisters alone, without the addition of gentamicin, was able to restrict the bacterial growth. Our results demonstrate the feasibility of the Bactibag blisters as an essential tool for preventing bacterial growth at porcine insemination doses stored at 16°C for up to 5 days.