

# Effect of an additional GnRH 2 d after the initiation of the Resynch-25 program on pregnancy per artificial insemination in lactating dairy cows

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## Introduction

Timed-AI protocols for the reinsemination of nonpregnant cows significantly reduce the interval between inseminations (AI) in lactating dairy cows. One of the most used programs is the Resynch-25, which starts Ovsynch on d 25 after previous AI in all cows, and only nonpregnant cows continue with the program on d 32 after previous AI. Low ovulatory response to d 25 GnRH administration can compromise the proportion of cows synchronized to the program and pregnancy per AI (P/AI) after timed-AI. One of the factors that can affect ovulatory response to this d 25 GnRH is the stage of antral follicle development at the time of treatment. For instance, non-dominant follicles cannot ovulate due to a lack of LH receptors in the granulosa cells. Thus, we hypothesized that adding an extra GnRH 2 d after d25 GnRH would increase P/AI to the Resynch-25 program. Our objective was to determine the effect of an additional GnRH 2 d after initiating the Resynch-25 program on P/AI.

## Materials and methods

Lactating Holstein cows ( $n = 2,795$ ) from 2 commercial dairy farms were randomly assigned to receive GnRH only on d 25 (GnRH25) or on d 25 and 27 (GnRH25&27) between August 2020 and January 2021. Pregnancy diagnoses were performed on d 32 and 102 post-AI. Nonpregnant cows on d 32 were classified according to CL presence and estimated diameter: 1) CL larger than 20 mm, 2) CL smaller than 20 mm, and 3) no CL present. Pregnant cows did not receive any further treatment. Nonpregnant cows with a CL larger than 20 mm continued the Resynch program and received a PGF injection on d 32 and 33, followed by GnRH on d 34 and timed-AI on d 35 after previous AI. Nonpregnant cows with no CL present or a CL smaller than 20 mm received different strategies of synchronization protocols depending on parity and were not included in the analysis. Binary variables were analyzed by logistic regression using the GLIMMIX procedure of SAS. The statistical model included the fixed effects of treatment, parity, AI technician, service and the random effect of farm by cow id. Parity was categorized as primiparous or multiparous. Service number was classified as 2nd, 3rd, 4th or greater than 4th service. Cows inseminated with sexed semen ( $n = 25$ ) were excluded from the P/AI analysis.

## Results

Different than hypothesized, the proportion of nonpregnant cows that did not have a CL greater than 20 mm in diameter on d 32 did not differ ( $P = 0.40$ ) among treatments (GnRH25 = 32.2%, and GnRH25&27 = 34.5%). Additionally, treatment did not affect P/AI on d 32 ( $P = 0.70$ ) and d 102 ( $P = 0.73$ ) of the cows enrolled in the Resynch-25 program (P/AI d32: GnRH25 = 39.9% vs. GnRH25&27 = 38.4%; P/AI d102: GnRH25 = 30.8% vs. GnRH25&27 = 32.0%). There was an effect of parity ( $P = 0.01$ ) on d 32 and 102 P/AI (d32 P/AI: primiparous = 45.0% vs. multiparous = 33.2%; d102 P/AI: primiparous = 37.6% vs. multiparous = 28.2%). Pregnancy loss between d 32 and 102 also did not differ ( $P = 0.11$ ) between treatments (GnRH25 = 19.3% vs. GnRH25&27 = 11.6%, respectively).

## Significance

In conclusion, adding an extra GnRH 2 d after the initiation of the Resynch-25 program was not effective in increasing P/AI nor in increasing the proportion of cows with a CL larger than 20 mm in diameter on d 32 after previous AI.

