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Introduction

- Wood bison (*bison bison athabasca*) and plains bison (*bison bison bison*) are threatened subspecies native to North America [1]
- Collection of oocytes, semen and embryos are required to connect valuable and inaccessible genetics from geographically distant herds in a bio-secure manner [2]
- There is a need for minimum-handling protocols to allow for oocyte and embryo collections that are feasible in the field

Objectives

- Compare oocyte collection procedures on sedated, recumbent bison vs bison restrained in a hydraulic chute (Experiment 1)
- Compare the superstimulatory responses of a single vs multiple-dose treatment protocol and modes of administration methods (manual injection [restrained] vs field darting [unrestrained] bison; Experiment 2)
- Compare embryo development from oocyte collections at random stages of the ovarian follicular status vs superstimulated (Experiment 2)

Materials and Methods

Experiment 1:

- Mature wood bison (n=16) were used for transvaginal ultrasound-guided oocyte collection during the anovulatory season
- Oocyte collections occurred at random stages of ovarian follicular status
- Half of the bison were collected in a hydraulic chute without sedation (n=8) and the other half were sedated

Experiment 2:

- 2 x 2 design to assess 2 superstimulation protocols (see below) and 2 methods of drug administration (manual injection and field darting; n = 4 per group)

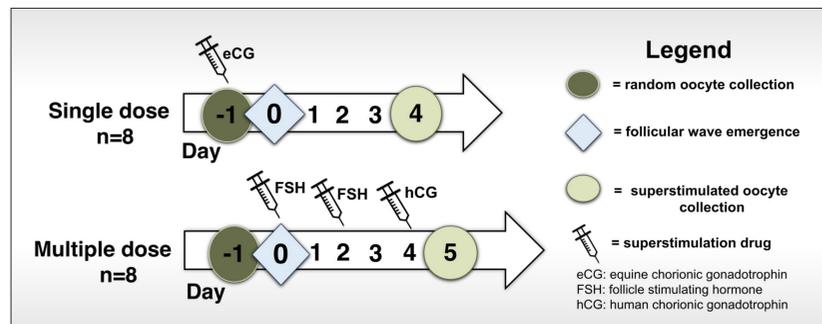


Figure 1. Oocyte collection procedures for experiment 2

- Compact oocytes were *in vitro* matured, fertilized and cultured
- Cleavage development was assessed on Day 3 (Day 0 = day of fertilization)
- Embryo development was assessed on Days 7-10
- Data were compared by ANOVA and GLIMMIX

Results

Experiment 1:

Table 1. Collection statistics		
	Chute (n=8)	Sedated (n=8)
Collection time (mins)	6.9 ± 1.0	8.9 ± 1.0
Follicles available	11.5 ± 1.9	9.3 ± 1.8
Oocyte recovery rate ¹	58/92 (63%)	44/69 (64%)

No differences were detected between chute-restrained vs sedated groups for any endpoint
¹ Number of COC recovered out of the number of follicles aspirated

Experiment 2:

- No effect of superstimulation protocol or method of drug administration for any endpoint; hence, data were combined to compare the effect of COC collection at random (non-superstimulated) vs superstimulated

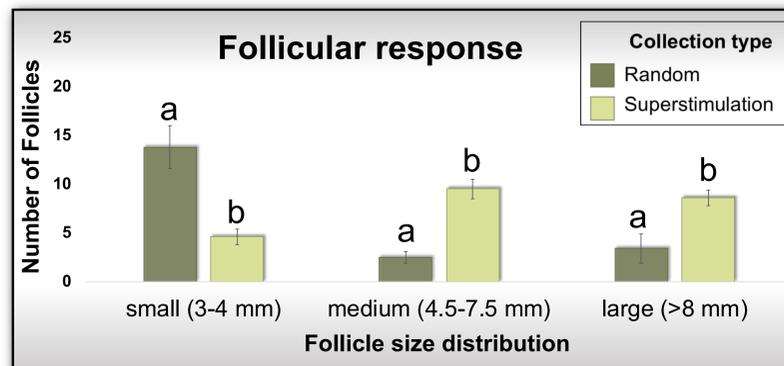


Figure 2. Follicle size distribution (mean ± SEM) for wood bison. (n=16 per group)
^{a,b}within follicular size categories, values with no common superscripts are different (P<0.05).

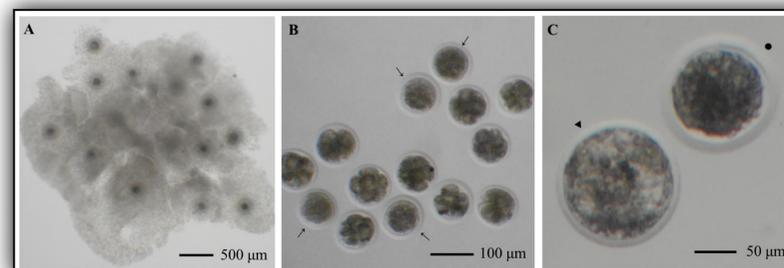


Figure 3. Wood bison cumulus-oocyte complexes (COC) after 25-28 hours of *in vitro* maturation (A). *In vitro* produced wood bison embryos on day 3 (B) and Day 9 (C). (→, unfertilized oocytes; ▶, blastocyst stage grade 1; •, early blastocyst stage grade 1; Day 0 = day of *in vitro* fertilization).

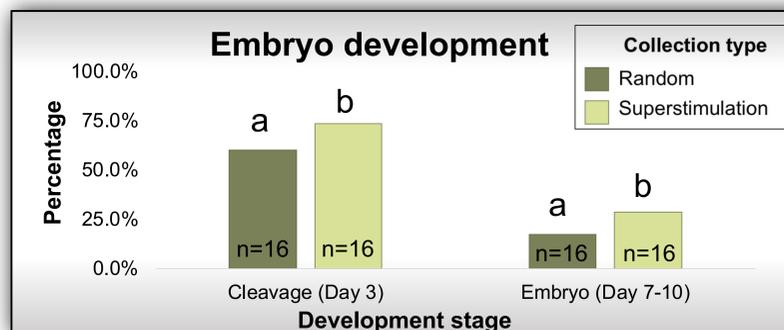


Figure 4. Embryo development distribution (%) for wood bison, Day 0 = day of fertilization
^{a,b}within embryo development categories, values with no common superscripts are different (P<0.05).

Conclusions

- No difference was detected in the duration of procedure time or oocyte recovery rates on sedated bison vs chute collections
- Administration of ovarian superstimulation treatments through field darting is an effective method
- Ovarian superstimulation through a single-dose protocol is as effective as a multiple-dose protocol
- Use of ovarian superstimulation increased the number of medium and large follicles available for collection and improved embryo development



We conclude that the minimum-handling protocols tested are feasible for use in free roaming bison herds

Literature Cited

- [1] COSEWIC. 2013. COSEWIC assessment and status report on the Plains Bison *Bison bison bison* and the Wood Bison *Bison bison athabasca* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. xv + 109 pp. (www.registrelep-sararegistry.gc.ca/default_e.cfm).
- [2] Adams GP, Zwiefelhofer ML, Palomino JM, Cervantes M, Yang S, Anzar M, McCorkell RB, Mastromonaco GF (2020) Development and use of a genome biobank to restore the genetic diversity of North American bison. Special Report for Parks Canada. ISBN 978-0-660-35863-5, CAT. NO. R62-566/2020E-PDF, Pp. 1-97.

Acknowledgments



This study was approved by the University of Saskatchewan's Animal Research Ethics Board and adhered to the Canadian Council on Animal Care guidelines for humane animal use