LONGEVITY IN VITRO AND GLYCEROL TOXICITY OF EPIDIDYMAL SPERM RECOVERED FROM A WHITE RHINOCEROS (Ceratotherium simum)

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This study was designed as a preliminary step towards developing effective methods for the long-term storage of sperm from the white rhinoceros. Testes were removed from an adult bull within 1 h of death and transported on ice. By 3 h post-mortem, caudal epididymal sperm were suspended in TALP medium then aliquoted into 1 of 5 treatment groups: TALP medium, or an egg yolk-based diluent (Biladyl; Minitub, Germany) containing 0, 4, 8 or 12% glycerol (Gly). Aliquots of each treatment were incubated at refrigeration (4 to 10°C), room (22 to 25°C) or physiological (37 to 38°C) temperatures. At 4, 8 and 12 h post-collection (pc), all sperm treatments were evaluated for estimated motility and rate of forward progression (FP; 0=no movement to 5=fast linear movement) after incubation at 37°C for 10 min; percent live and acrosome intact (AcrInt) sperm were determined by counting 200 cells after vital staining (Aalseth & Saacke, Gam. Res. 15:73, 1986).

	Refrigeration					Room Temp					Physiol Temp				
		Biladyl %Gly					Biladyl %Gly					Biladyl %Gly			
At 4 h:	TALP	0	4	8	12	TALP	0	4	8	12	TALP	0	4	8	12
%Motile	85	65	75	50	50	80*	45*	35*	35*	30*	35	20	30	25	20
FP (0-5)	4	4	4	4	3	4	3	3	3	2	2	1	1	1	1
%Live	95	94	86	72	18	96	90	96	45	0	86	60	43	27	59
%AcrInt	100	84	0	0	9	95	48	0	0	0	82	60	0	1	0
At 8 h:															
%Motile	85	65	60	45	40	80*	35*	35*	20*	20*	20	20	5	5	0
FP (0-5)	4	4	3	3	3	4	3	2	2	2	2	2	1	1	0
%Live	81	95	90	74	3	92	87	73	30	34	11	31	18	4	1
%AcrInt	70	64	0	0	0	91	0	2	0	0	11	4	0	0	0
At 12 h:															
%Motile	85	60	60	40	35	80*	30*	25*	15*	15*	0	0	0	0	0
FP (0-5)	4	4	3	3	3	3	2	2	2	2	0	0	0	0	0
%Live	83	94	86	17	1	92	80	62	22	5	12	19	14	1	0
%AcrInt	66	5	0	0	0	11	0	0	0	0	4	7	0	0	0

*Marked head agglutination

At collection, the sperm were evaluated as 90% motile, FP=4, with 95% live and 100% AcrInt. There was no loss of motility in TALP medium at refrigeration for up to 12 h pc; however, there was a decrease ($p<0.05$; Chi^2) in the %AcrInt sperm by 8 h pc. At all levels and temperatures, glycerol apparently induced acrosome reactions by 4 h pc. Biladyl without glycerol at refrigeration and room temperature resulted in less than a 15% reduction in live sperm by 12 h pc; however, motility was reduced and, with increasing time and temperature, percentages of acrosome-reacted sperm increased. In summary, rhinoceros sperm can be stored for up to 12 h in TALP medium at refrigeration.