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SHORT COMMUNICATION

Measurement of Leptin and Insulin-like Growth Factor-I in Seminal Plasma from Different Species

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Summary

The multi-functional proteins, insulin-like growth factor-I (IGF-I) and leptin were present in seminal plasma from different species. Concentrations of IGF-I in equine and porcine semen were 20 and 17.5 ng/ml, respectively. Seminal plasma concentrations of leptin were 1 ng/ml in human and 11 ng/ml in porcine samples.

Key words

Leptin • Semen • Reproduction • Insulin-like growth factor

Seminal plasma hormones, cytokines, and growth factors affect sperm motility and fertilization (Aitken 1994). IGF-I has been measured in human and bovine semen and is correlated with semen quality (Glander *et al.* 1996, Henricks *et al.* 1998). IGF-I has been associated with onset of puberty, fertilization and embryonic development (Spiteri-Grech and Nieschlag 1992). Studies of knockout mice indicate that leptin is also involved in regulating these physiological events (Kiess *et al.* 1998). Unlike IGF-I, the primary structure of leptin is different among equine, porcine and humans. Equine leptin concentrations could not be validated. Because IGF-I has been measured in human semen previously it is not reported here. Therefore, our objectives were to identify IGF-I in porcine and equine, and leptin in porcine and human seminal plasma and promote further research on the roles these hormones have in male reproductive physiology.

Cryopreserved semen samples from three anonymous human donors (ages 20 to 40), a 15-year-old quarterhorse and two Duroc and Yorkshire boars (ages 2 to 4 years) were assayed for IGF-I and/or leptin. The IGF-I extraction and radioimmunoassay procedure are described in the work of Henricks *et al.* (1998). The level of quantitation of the assay was 0.1 ng/ml. Intra- and inter-assay coefficients of variation were 6.0 and 2.0 %, respectively. Seminal plasma leptin concentrations were assayed using a multi-species leptin RIA kit; the anti-human leptin antibody has 80 % cross-reactivity with porcine leptin (Linco, St. Louis, MO, Sinha *et al.* 1996). Inter- and intra-assay coefficients of variation were 12.0 and 11.0 %, respectively. The level of quantitation was 0.1 ng/ml.

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IGF-I concentrations in equine and porcine seminal plasma ranged from 15-25 and 5-25 ng/ml, respectively (Table 1). Seminal plasma leptin concentrations ranged from 9.2 - 13.38 and 0.55 - 1.15 ng/ml for porcine and human samples, respectively. The recovery rate of IGF-I in equine and porcine seminal plasma was 87 %. Recovery rates for leptin were 93 % in porcine and 103 % in human seminal plasma samples. Parallelism and additivity experiments (Fig. 1) performed to ensure assay optimization revealed that equine samples did not meet validation criteria for leptin.

Table 1. IGF-I and leptin in seminal plasma from different species

Species	**Sample**	**Replicates**	**IGF-I Concentration (ng/ml ± S.E.M.)**
Equine	E	8*	20.00 ± 2.00
Porcine	A	6	20.00 ± 1.15
	B	6	25.00 ± 2.80
	C	6	5.00 ± 0.54
	D	6	20.00 ± 4.07
			Leptin Concentration (ng/ml ± S.E.M.)
Porcine	A	6	13.38 ± 0.30
	B	6	10.70 ± 0.56
	C	6	10.25 ± 1.13
	D	6	9.20 ± 1.35
Human	F	8*	0.81 ± 0.01
	G	8*	0.67 ± 0.07
	H	8*	1.10 ± 0.03

** Samples from four different days were assayed in duplicate. Data in the Summary were obtained by calculating the means of concentrations presented in Table 1.*

The concentration of IGF-I in equine and porcine semen was similar to that in human and approximately four times lower than that found in bovine semen (Henricks *et al.* 1998). Differences in ejaculate volume, growth hormone, steroids, and nutritional factors may contribute to the variation in semen IGF-I concentrations. The concentration of leptin in porcine semen was approximately ten-fold higher than in human semen; body composition differences and hormonal and nutritional factors may have contributed to the difference. Future research should focus as to whether IGF-I in porcine and equine seminal plasma and leptin in porcine and human semen are indicators of semen quality or whether they are involved in regulating sperm motility as in other species (Glander *et al.* 1996, Henricks *et al.* 1998).

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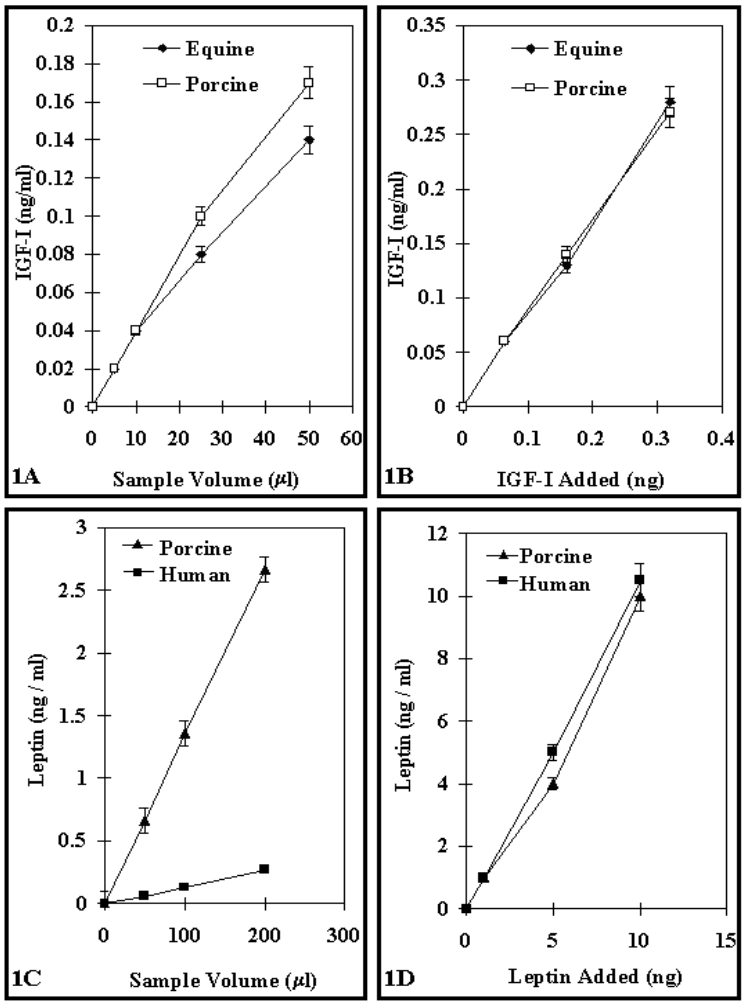

Fig. 1. *Validation experiments for IGF-I and leptin. The graphs depict typical dose-response kinetics to increasing sample volume (A & C) and addition of exogenous hormone (B & D).*

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Reprint requests

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