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JOURNAL ARTICLE

# Hyaluronic acid enhances the zona pellucida-induced acrosome reaction of macaque sperm

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Hyaluronic acid (HA) not only surrounds the zona pellucida as part of the cumulus matrix but also is present throughout the zona pellucida and the perivitelline space of many mammalian oocytes. However, most in vitro techniques to study sperm-oocyte interaction eliminate HA from the oocyte through enzymatic digestion and/or do not expose sperm to HA prior to zona pellucida binding. This study explores the effect

of preincubation of sperm or oocytes with HA on sperm-zona pellucida binding and subsequent acrosome reaction of bound sperm. Cynomolgus macaque semen was washed. Incubated, chemically capacitated with dibutryl cyclic adenosine monophosphate (dbcAMP) and caffeine, and sperm-zona pellucida binding assays were performed. In one experiment, sperm were pretreated with HA (100 micrograms/ml) during the last 10 minutes of the 30-minute period for chemical capacitation. In another experiment, only the oocytes were preincubated in the media containing HA. In the third experiment, gametes were exposed to HA for 10 minutes after sperm had been allowed to bind to zonae pellucida. The preincubation of either sperm or zonae pellucida with HA enhanced the percentage of bound sperm that were acrosome reacted. However, HA did not affect the number of sperm bound to zonae pellucida. When sperm already bound to the zonae pellucida were exposed to HA, there was no increase in the percentage of bound acrosome-reacted sperm. The HA enhancement of the acrosome reaction of sperm bound to the zona pellucida shown in this study required less than 1 minute of sperm exposure to HAtreated zonae during the zona binding process. However, this enhancement was observed only if the HA exposure preceded sperm-zona binding. This result suggests that HA is interacting with the sperm surface, possibly via a receptor, at the time of initiation of the acrosome reaction. It is unlikely that the effects noted in the current experiments were the result of motility retention or improvement because the full enhancement of the acrosome reaction was observed when only the oocytes were pretreated with HA.

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