

Table S1. Embryo development after ICSI with and without heat inactivated sperm.

Method of activation	No. of surviving oocytes (exp.)	No. of oocytes activated (PN %)	No. of embryos developing to (%)	
			2-cell	Blastocyst
ICSI with fresh sperm	43 (3)	43 100%	36 83.7%	22 51.2% ^{a,b,c,d,e,f}
ICSI with frozen sperm	45 (3)	41 91.1%	38 84.4%	12 26.7% ^a
ICSI + HT 2 min 30 sec	27 (2)	23 85.2%	18 66.7%	6 22.2% ^b
ICSI + HT 5 minutes	47 (3)	38 80.9%	28 61.7%	8 17% ^c
ICSI + HT 10 minutes	49 (3)	38 77.6%	33 67.3%	3 6.1% ^d
ICSI + HT 20 minutes	48 (3)	13 27%	11 22.9%	2 4.1% ^e
ICSI + HT 30 minutes	47 (3)	8 17%	1 2%	0 ^f

HT = Heat treatment at 56°C

a, non-significant difference $P= 0.268$
b,c,d,e,f, represent a significant difference. ($P= 0.0197, 0.007, 0.0038, 0.0023, 0.000$) respectively.
Statistical analysis was performed by student's T test

Table S2. Embryo development after ICSI with various activation protocols

ICSI method	No. Of surviving oocytes (exp.)	No. Of oocytes activated (PN)	No. Of embryos developing to (%)	
			2-cell	Blastocyst
ICSI with fresh sperm	43 (3)	43 100%	36 83.7%	22 51.2% ^{abc}
ICSI with frozen sperm	45 (3)	41 91.1%	38 84.4%	12 26.7%
Inactive ICSI* & 5µM Ionomycin	45 (3)	36 80%	26 57.8%	6 13.3% ^{ade}
Inactive ICSI* & 5mM Strontium	44(3)	40 90.9%	40 90.9%	15 34.1% ^e
Inactive ICSI & Nus-A hPLCζ injection	80 (3)	77 96.3%	68 85%	29 36.3% ^{cd}
ICSI fresh sperm & Nus-A hPLCζ injection	37 (3)	36 97.3%	28 78.4%	17 45.9% ^b
ICSI frozen sperm & Nus-A hPLCζ injection	38 (3)	35 92%	29 76.3%	17 44.7%

*Inactive ICSI= Sperm with heat treatment at 56°C for 30mins

a significantly different ($P= 0.009$)

b no significant difference ($P = 0.068$)

c no significant difference ($P = 0.079$)

d significantly different ($P = 0.008$)

e significantly different ($P =0.059$)

Statistical analysis was performed by student's T test