



**Figure S1.** D4Z4 methylation variation in oocytes from the same donors. Different oocytes from a given donor (numbers 1-34) are indicated by differently colored (blue, red, green, yellow and mauve) dots on a vertical line.

**Table S1.** Semen parameters.

	<b>Mean ± standard deviation</b>	<b>Range</b>
<b>Donor Age (years)</b>	38.2±5.7	26.3-54.0
<b>Donor BMI (kg/m<sup>2</sup>)</b>	26.1±4.2	17.5-51.7
<b>Sperm volume (ml)</b>	3.2±1.3	0.2-8.0
<b>Sperm concentration (x 10<sup>6</sup>/ml)</b>	68.4±46.7	5.3-250
<b>Morphology (% normal)</b>	8.4±3.6	3.0-18.0
<b>Motility (% motile)</b>	53.7±11.8	35.0-85.0

**Table S2.** Clinical parameters of oocyte donors.

	<b>Mean ± standard deviation</b>	<b>Range</b>
<b>Oocyte donors (N)</b>	53	
<b>Donor age (years)</b>	34.3±3.8	28-42.7
<b>Donor BMI (kg/m<sup>2</sup>)</b>	26.6±5.8	18.9-42.5
<b>Previous number of OPUs</b>	2.2±2.0	0-12
<b>OPUs for this study (N)</b>	56	
<b>Oocytes per OPU</b>	9.6±4.4	2-21
<b>GV oocytes per OPU</b>	1.9±1.0	1-5
<b>Day of embryo transfer</b>	3.9±1.0	3-6
<b>Number of embryos transferred</b>	1.7±0.5	1-3
<b>Clinical pregnancy (N)</b>	14	
<b>% Clinical pregnancy / OPU</b>	25	95% CI 13.7-36.3
<b>Group 1: OPU with at least one GV oocyte &lt; 2.5% D4Z4 methylation (N)</b>	17	
<b>Clinical pregnancy (N)</b>	6	
<b>% Clinical pregnancy / OPU</b>	35.3	95% CI 12.6-58.1
<b>Group 2: OPU with no GV oocyte &lt; 2.5% D4Z4 methylation (N)</b>	39	
<b>Clinical pregnancy (N)</b>	8	
<b>% Clinical pregnancy / OPU</b>	20.5	95% CI 7.8-33.1

OPU = Oocyte pick-up; BMI = Body mass index; GV = Germinal vesicle;  
95%CI = 95% confidence interval

**Table S3.** Primers for multiplex (outer) and singleplex (inner) PCR and pyrosequencing.

Target region	Primer	Sequence (5'-3') <sup>a</sup>	Number of CpGs	Annealing temperature
<i>DUX4</i>	Forward outer	AGGAAGGTAGGGAGGAAAA		59°C
	Reverse outer	ACAAAACCTCAACCTAAAAATATAC		
	Forward inner	GGGTTGAGGGTTGGGTTTATA		58°C
	Reverse inner	*ACAAAACCTCAACCTAAAAATATAC		
	Sequencing	GGGTTGGGTTTATAGT	9	
<i>GTL2</i> (paternally imprinted)	Forward outer	GAATTATAGGGAATGATGGTGTA		59°C
	Reverse outer	CCAAAATACTAACTACTCCTTAAACA		
	Forward inner	AGGGTTAGGAAGTTTAGTAGGTTA		60°C
	Reverse inner	*ACTACTCCTTAAACAAAAAACACATAAT		
	Sequencing	GTAGTAAATTAAAGTGTATTAGAGA	2	
<i>PEG3</i> (maternally imprinted)	Forward outer	GGTTGTTGATTGGTTAGTATAG		59°C
	Reverse outer	CACTCACCTCACCTCAATAC		
	Forward inner	GGTGTAGAAGTTTGGGTAGTTG		57°C
	Reverse inner	*CTCACCTCACCTCAATACTAC		
	Sequencing	TGTTTATTTGGGTTGGT	4	

<sup>a</sup> Primers indicated by a star are biotinylated at the 5' end.

**Table S4.** Primers for deep bisulfite sequencing.

Assay	Primer	Sequence (5'-3') <sup>a</sup>	Amplicon length	Number of CpGs	Annealing temperature
<i>DUX4</i>	Forward	GAAGGTAGGGAGGAAAAG	259 bp	30	58°C
	Reverse	ACAAAACCTCAACCTAAAAATATA			