MONASH IVF GROUP

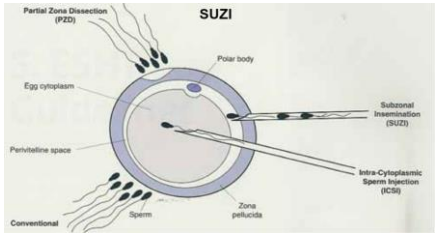
PIEZO ICSI: Better or Not?

ASPIRE Webinar Series

A/Prof Deirdre Zander-Fox- Regional Scientific Director Monash IVF Group Victoria and Malaysia

INTRODUCTION-ICSI

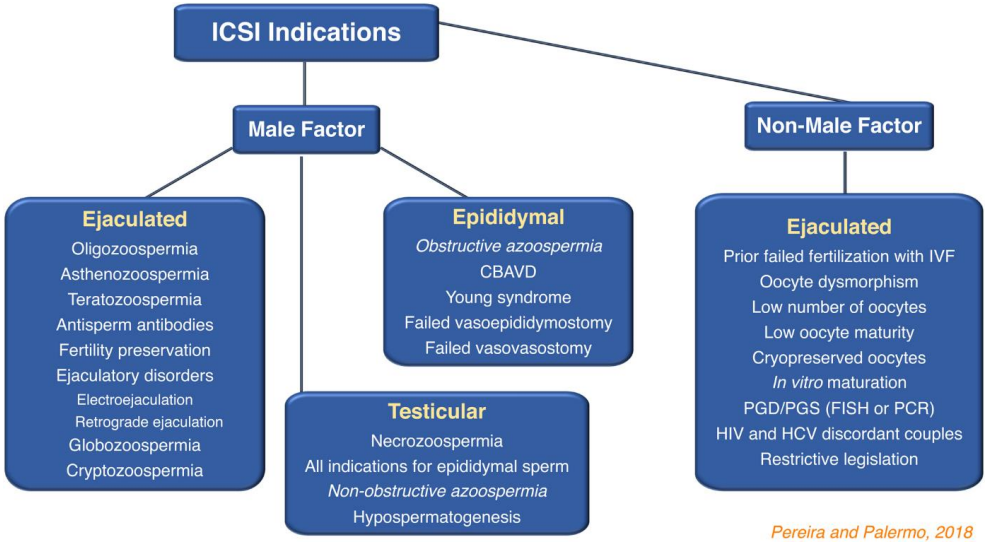
- Louise Brown born 1978
- Mother: Lesley Brown had blocked fallopian tubes
- IVF using standard insemination was tubal infertility was highly successful
  - Failed fertilization via standard insemination
  - Male factor infertility
- 1980's investigations into further optomising gamete interaction
  - Partial zona dissection
  - Creating a hole with acid tyrode
  - Inducing a hole/slit mechanically



- All failed to work or resulted in high rates of polyspermy
- Accidentally injected a sperm directly while attempting to induce interaction between oolemna and sperm
- Whenever this happened fertilization nearly always occurred
- First human pregnancy from ICSI was an accident (Palermo et al 1992)

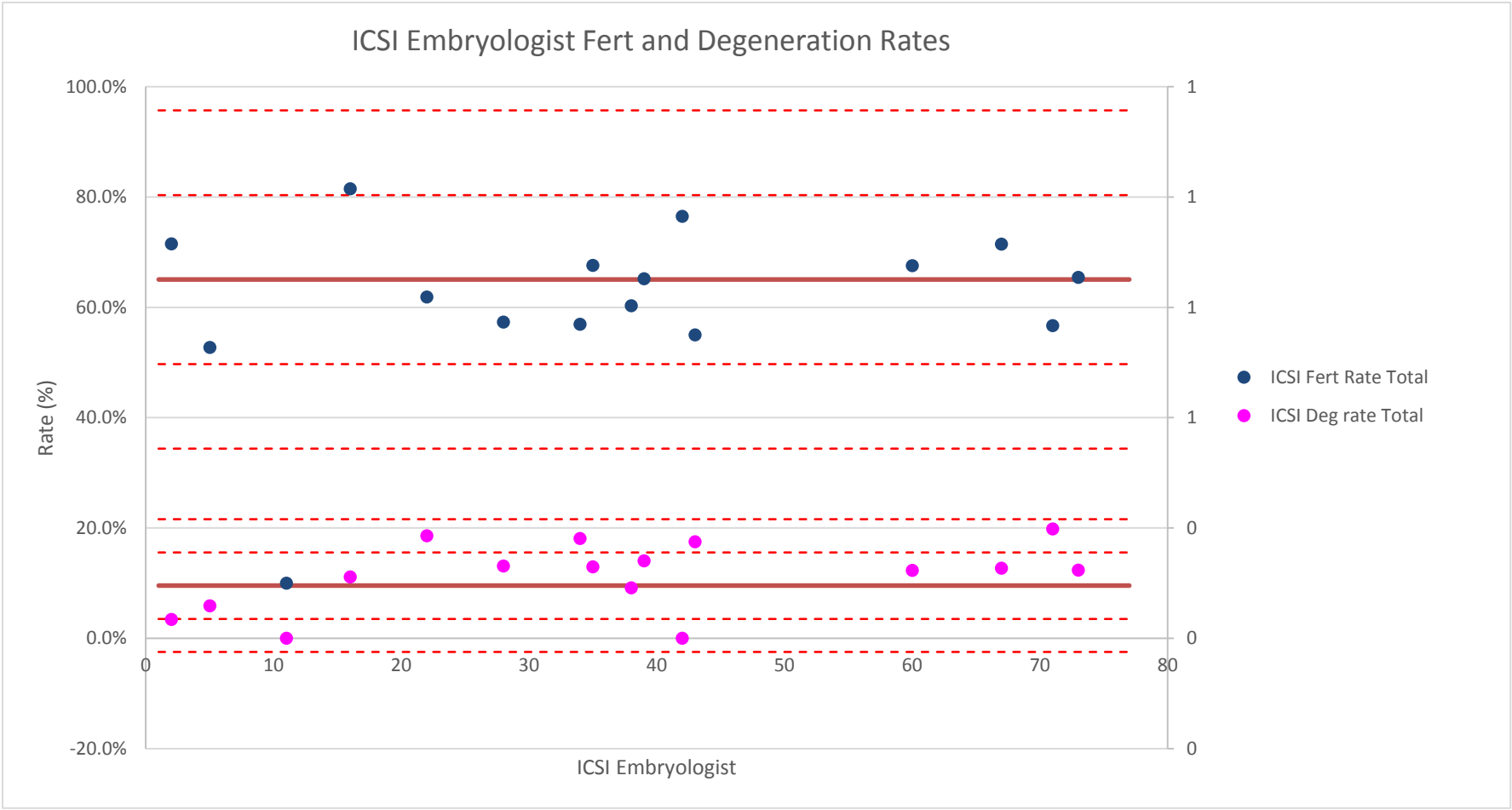
INTRODUCTION-ICSI

- Gained steady increase in popularity throughout the 1990's
  - Occasional motile in ejaculate
  - Testicular and epididymal sperm
  - Cryopreserved oocytes/IVM (zona hardening)
  - PGT-M
  - Sero-discordant couples
- Is invasive and can result in oocyte damage



Pereira and Palermo, 2018

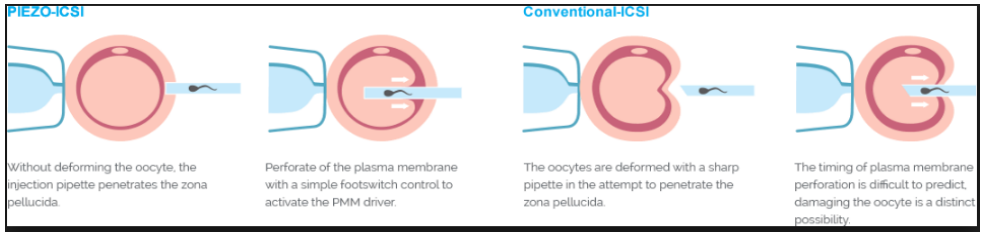
LABORATORY ICSI STATISTICS



MONASH IVF GROUP

INTRODUCTION-PIEZO

- PIEZO ICSI has been used primarily in research/animal industry due to the fragile nature of animal oocytes (such as mice)
- PIEZO ICSI resulted in improved fertilization and survival rates compared to conventional ICSI
- Used blunt ended micropipette
- Required operation fluid to be loaded into the pipette (mercury or fluorinert)
- A piezoelectric actuator is used to move the capillary in an ultra rapid fashion by producing ultra-fast submicron motion which propels the capillary forward.
- Actuator is made of material that when a voltage is applied it changes shape and thus extrudes a force which moves the capillary in a forward micro-jack hammer like motion



PIEZO VIDEO

PIEZO-HUMAN

- First published in human in 1996 with comparable results to conventional ICSI (Huang et al 1996)
  - Did not use operation fluid
  - Used hydraulic fluid to open and etch the PIEZO pipette
- Second study demonstrated: (Yanagida et al 1998)
  - used mercury as operation fluid
  - improved ICSI survival rates (88.1% vs 81.4% $p<0.001$)
  - increased fertilization rates (79.4% vs 66.4% $p<0.001$)
  - pregnancy rates were also increased (23.1% vs 14.9%, $p<0.05$)

PIEZO-HUMAN

- Study assessed effect of PIEZO compared to conventional ICSI in patients with male factor infertility and failed insemination via IVF (Takeuchi et al 2001)
- Demonstrated significant improvements in patient outcomes
- Used mercury for operation fluid

	Conventional ICSI	PIEZO ICSI
Cycles	122	204
Oocytes Retrieved	781 (6.4±0.5)	1681 (8.3±0.4)
Oocytes Injected	655	1344
Fertilisation rate	83.1%	90.3%*
Embryos cleaved	84.6%	88.1%*
Implantation Rate	10.4%	11.7%

PIEZO-HUMAN

- 14 years later another study was published for PIEZO ICSI in human
- Used Fluorinert as operation liquid (Hiraoka et al 2015)
- Trialed two different diameter pipettes (thin and tick wall diameter)

	Conventional ICSI	Conventional PIEZO	Thin PIEZO
Oocytes retrieved	624	717	679
Survival post injection	90%[a]	95%[b]	99%[c]
Fertilisation	68%[a]	75%[b]	89%[c]
Good quality D3	37%[a]	43%[b]	55%[c]
Clinical Pregnancy	19%[a]	21%[a]	31%[b]
Live birth	15%[a]	16%[a]	25%[b]

MIVFG CLINICAL TRIAL

Hypothesis: Use of piezo ICSI technology will increase fertilisation rates and decrease oocyte degeneration rates compared to conventional ICSI

Site: Repromed

Partnership: Vitrolife and Primetech

Training: Kameda Ladies Clinic (Dr Kenichiro Hiraoka) and Primetech

Operation liquid: MEA tested, ultra-purified medical grade perfluoro-n-octane, Vitrolife

- N=69 patients in each arm
- Prospectively case matched for comparison
- Clinician recommends ICSI as insemination method
- ≥6 mature oocytes
- Exclusion
  - Surgical sperm
  - <30% fertilization on previous ICSI cycle

CLINICAL TRIAL RESULTS

	Conventional ICSI	PIEZO ICSI	P Value
N value	69	69	
Maternal Age	34.4 (3.9)	35.2 (3.4)	NS
Maternal BMI	24.8 (5.8)	27.3 (5.9)	0.044
Paternal Age	36.4 (4.4)	35.6 (4.7)	NS
Paternal BMI	26.5 (3.6)	29.5 (6.4)	0.013
Previous Cycles	0.5 (0.9)	0.6 (0.9)	NS
AMH	26.3 (17.9)	22.1 (16.7)	NS
Sperm Conc	67.1 (58.1)	66.3 (53.4)	NS
Sperm motility	41.8 (19.3)	46.2 (16.8)	NS
Sperm Morphology	7.1 (4.9)	6.7 (4.9)	NS
Oocytes Retrieved	12.7 (4.5)	13.1 (5.1)	NS
Oocytes Injected	10.9 (4.1)	10.7 (4.2)	NS

Data in parenthesis indicates standard deviation

Zander-Fox et al- manuscript in preparation

CLINICAL TRIAL RESULTS

	ALL		Maternal age <38 years		Maternal age >38 years	
	Est [95% CI]	P-Value	Est [95% CI]	P-Value	Est [95% CI]	P-Value
Unadjusted (PIEZO vs ICSI)						
% oocytes fertilised	14.33 [12.05, 16.61]	<0.0001	11.82 [5.60, 18.05]	<0.0001	21.92 [10.41, 33.43]	0.000
% oocytes degenerate	-5.02 [-6.21, -3.82]	<0.0001	-5.57 [-8.95, -2.20]	0.001	-3.28 [-9.15, 2.59]	0.273
% oocytes abnormally fertilised	-4.55 [-5.56, -3.54]	<0.0001	-3.09 [-5.67, -0.51]	0.019	-9.17 [-15.14, -3.20]	0.003
Adjusted (PIEZO vs ICSI)						
% oocytes fertilised	**14.05 [11.68, 16.41]**	**<0.0001**	**11.83 [5.55, 18.12]**	**<0.0001**	**28.02 [8.19, 47.8]**	**0.006**
% oocytes degenerate	**-3.79 [-5.12, -2.46]**	**<0.0001**	**-4.56 [-8.03, -1.10]**	**0.01**	-4.33 [-14.90, 6.30]	0.684
% oocytes abnormally fertilised	**-4.75 [-5.91, -3.59]**	**<0.0001**	**-2.71 [-5.60, 0.18]**	**0.066**	-9.52 [-20.2, 1.11]	0.079

For women >35 years of age, the fertilization (*P* = .008) and blastocyst development (*P* = .016) rates with Piezo-ICSI were significantly higher than ICSI (Furuhashi *et al* 2019).

Zander-Fox et al- manuscript in preparation

CLINICAL TRIAL RESULTS

	ALL		Maternal age <38 years		Maternal age >38 years	
	Est [95% CI]	P-Value	Est [95% CI]	P-Value	Est [95% CI]	P-Value
Unadjusted (PIEZO vs ICSI)						
% embryo cryopreserved	8.70 [2.20, 15.20]	0.009	6.38 [-0.82, 13.57]	0.082	15.94 [1.63, 30.27]	0.029
% embryo utilisation	8.81 [1.20, 15.62]	0.011	5.80 [-1.74, 13.34]	0.132	17.98 [3.48, 32.49]	0.015
Adjusted (PIEZO vs ICSI)						
% embryo cryopreserved	**10.01 [2.80, 17.23]**	**0.006**	7.52 [-0.55, 15.58]	0.068	**29.05 [8.04, 50.06]**	**0.007**
% embryo utilisation	**9.79 [2.57, 17.01]**	**0.008**	7.13 [-1.09, 15.36]	0.089	**35.9 [14.26, 57.6]**	**0.001**

Patients having PIEZO had on average an extra embryo vitrified compared to conventional ICSI

Zander-Fox et al- manuscript in preparation

CLINICAL TRIAL RESULTS

	Conventional ICSI	PIEZO-ICSI	P-Value
% Positive HCG	68.0% (17/25)	71.4% (15/21)	0.801
% Clinical Pregnancy	60.0% (15/25)	57.1% (12/21)	0.845
% Viable Pregnancy	52.0% (13/25)	52.4% (11/21)	0.979
% Live Delivery	48.0% (12/25)	47.6% (10/21)	0.979

- No baby abnormalities noted in ether arm of this trial

Zander-Fox et al- manuscript in preparation

TO CONSIDER

- Requires added equipment for the ICSI rig
- Requires loading of an operation fluid into the injection pipette
- Increases time to undertake the microinjection process
- Learning curve for laboratories to master the technology

CONCLUSIONS

- Less invasive (no cytoplasmic aspiration)
- Increased fertilization and increased embryo numbers available per cycle
- Greatest benefit in poorer prognosis patients (low ovarian reserve or advanced maternal age)
- Currently undertaking a phase 2 clinical trial- multi center with sibling split as a strengthened trial design
- Further studies warranted to investigate whether PIEZO results in increased cumulative pregnancy and live birth rates

THANKYOU

Kevin Lam
Dr Leanne Pacella-Ince
Cathy Tully
Professor Kelton Tremellen
Dr Kenichiro Hiraoka