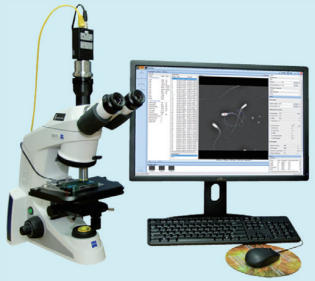
Assessment of Boar Sperm Motility by Computer-Assisted Sperm Analysis (CASA)

1



Herbie, Hsiu-Lien LIN

Breeding And Genetic Division
Livestock Research Institute
Tainan, Taiwan

Video Link: https://youtu.be/l6dqYppEz50

Outlines

2

- Boar sperm motility evaluation: predicting boar fertility
- How do I use CASA in my lab

- Quality control and assurance in CASA labs
- Conclusion

Predicting boar fertility

3

Boar sperm motility evaluation

- Microscopic evaluation
- Computer-assisted sperm analysis (CASA)
- CASA versus conventional microscope
- The CASA parameters and boar fertility

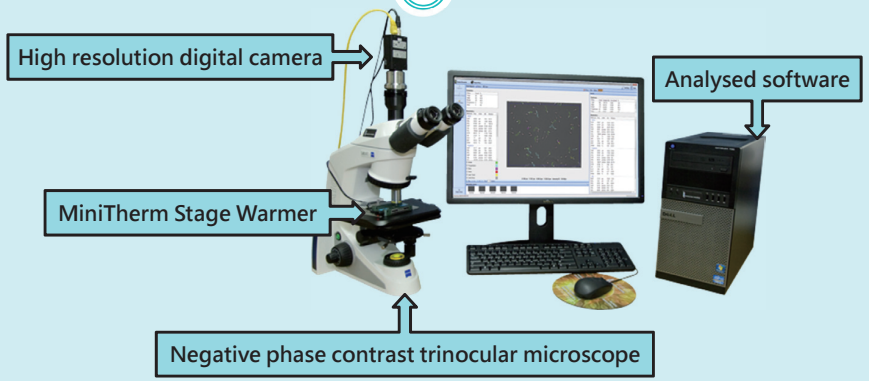
Microscopic evaluation

4

- Color
- Contamination
- Viscosity
- Concentration
- Motility is microscopically estimated by experience lab technicians

Computer-assisted sperm analysis (CASA)

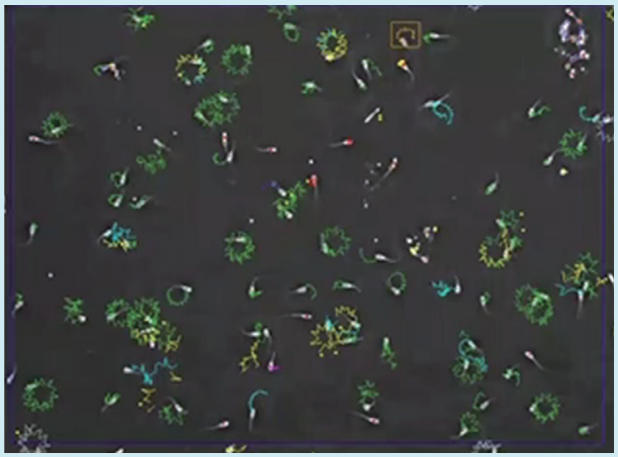
5



Place the sample slide on the stage → Focus the image → Capture images from selected fields → Begin analysis

CASA objective parameters

6



Tracking
- Motile %
- Progressive%

Kinematic
- Velocity
- Linearity
- Head movement

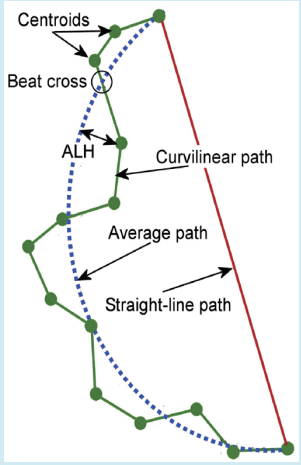
Morphology
- Tail
- Droplets

Concentration

CASA kinematic parameters definition

7

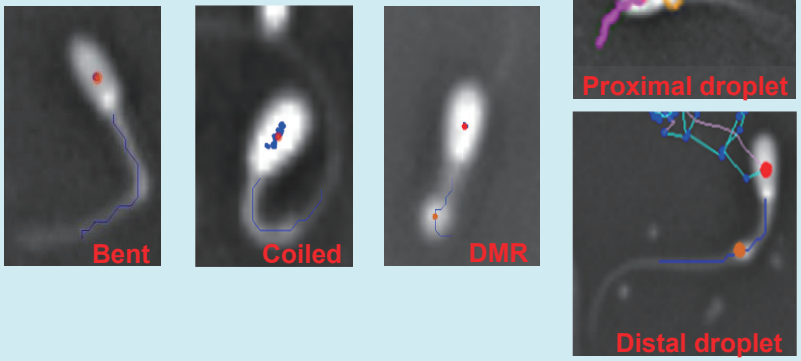
CASA parameter	CASA definition
ALH	Amplitude of lateral head displacement (μm)
BCF	Beat cross frequency (Hz)
DAP	Distance average path (microns)
DCL	Distance sperm travels in a curved line (μm)
DSL	Distance sperm travels in a straight line (μm)
LIN	Linearity (VSL divided by VCL)
MOT	Motility percentage of sperm >2.5 μm DSL
FPM	Progressive forward motility (% sperm >4.5 μm DSL)
STR	Straightness (VSL divided by VAP)
VAP	Velocity average path (μm/s)
VCL	Velocity curved line (μm/s)
VSL	Velocity straight line (μm/s)
WOB	Wobble (VAP divided by VCL)



[Adapted from Didion, 2008] & [Adapted from Rupert et al., 2014]

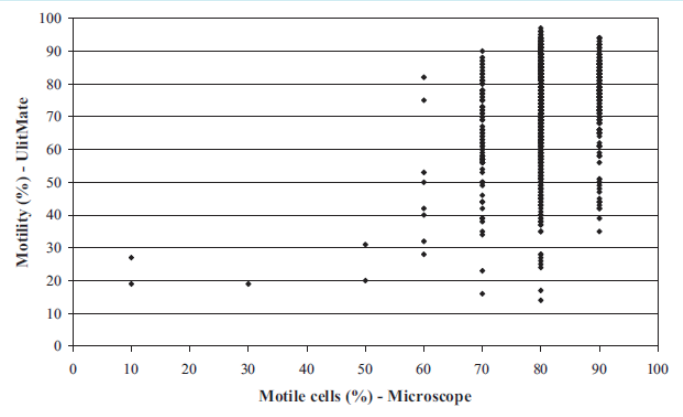
CASA sperm morphology

8



CASA versus conventional microscope

9

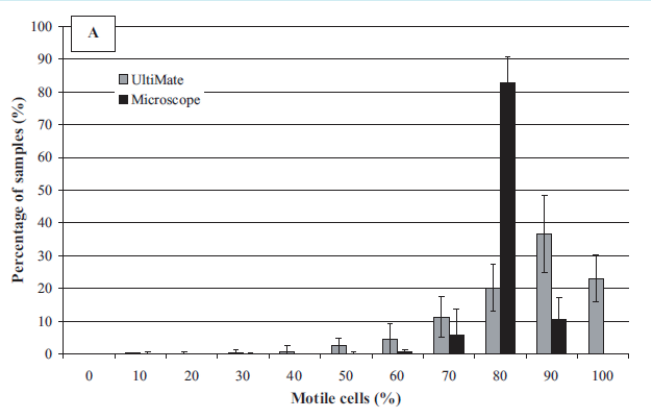


[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA versus conventional microscope

10

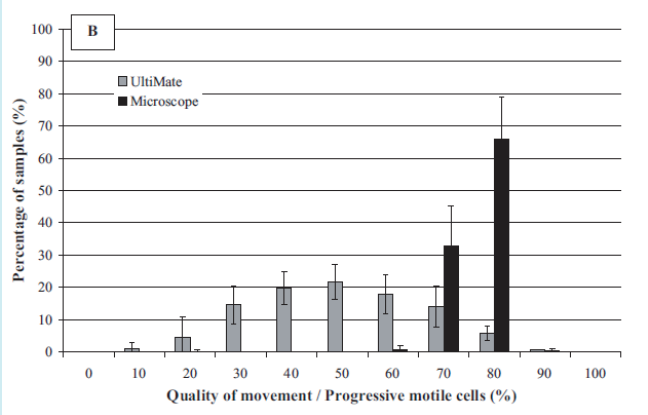


[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA versus conventional microscope

11

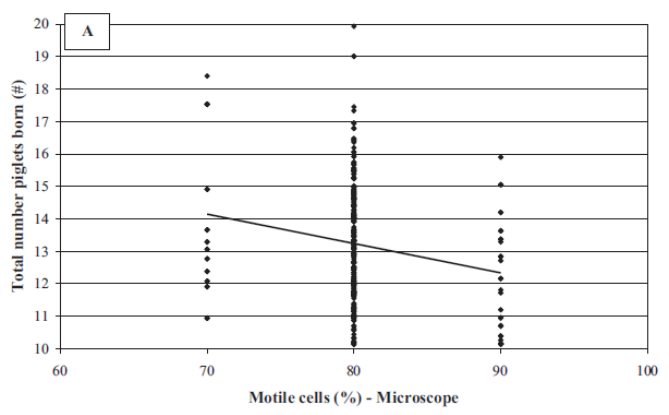


[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA versus conventional microscope

12

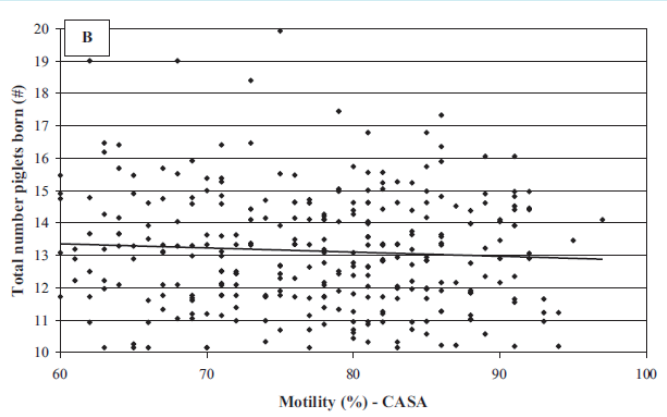


[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA versus conventional microscope

13



[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA parameters and boar fertility

14

Boar or semen characteristic	Number of records, or mean ± SD
Number of boars	2,367
Number of ejaculates	45,532
Number of genetic boar lines	15
Number of AI laboratories	7
Number of AI production locations	9
Age of boars, mo	24 ± 11
Number of days between ejaculation	4.34 ± 2.51
Number of sperm cells per ejaculate	$84 \times 10^9 \pm 11 \times 10^9$
Number of sperm cells in a dose (80 mL)	$1.87 \times 10^9 \pm 0.42 \times 10^9$
Number of doses produced per ejaculate	37 ± 16
General	
Motility, %	87.4 ± 6.4
Progressive motility, %	78.2 ± 8.6
Direction and movement	
Velocity average path, μm/s	95.1 ± 20.5
Velocity straight line, μm/s	68.5 ± 18.4
Velocity curvilinear, μm/s	175.2 ± 37.3
Amplitude of lateral head displacement, μm	7.3 ± 1.3
Beat cross frequency, Hz	39.3 ± 2.8

[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA parameters and boar fertility

15

Effect of +1 SD on	FR, %	TNB
Motility	NS[2]	0.128
Progressive motility	1.058	NS
Velocity average path	NS	0.246
Velocity straight line	NS	−0.092
Velocity curvilinear	−0.373	NS
Amplitude of lateral head displacement	NS	−0.027
Beat cross frequency	−0.728	NS

[1]Effects are expressed as change in FR or TNB by +1 SD value of the CASA variable. The CASA variable had no significant effect on FR or TNB.

[2]NS = not significant.

[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA parameters and boar fertility

16

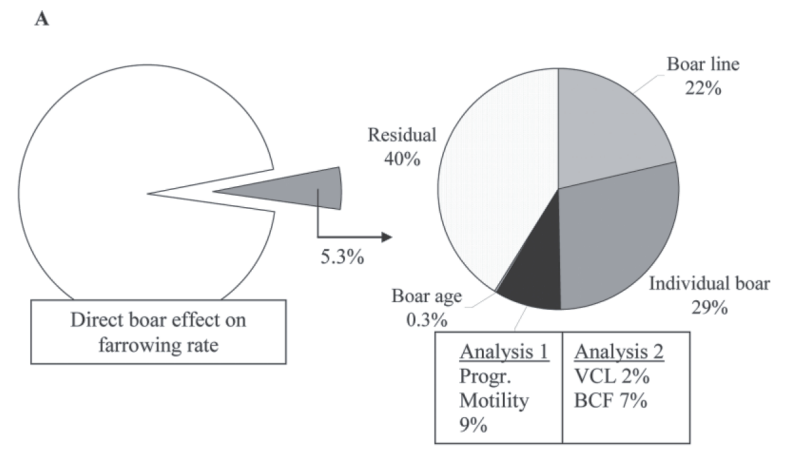
	Motility,[1]* %								
Item	65	70	75	80	85	90	95	100	
n	16	120	754	2,554	8,006	17,450	15,586	1,046	
Total number born	12.58	13.36	13.9	14.06	13.96	14.03	14.12	14.48	
	Progressive motility,[1]* %								
	55	60	65	70	75	80	85	90	95
n	52	395	1,672	3,113	6,166	10,788	13,191	9,240	902
Farrowing rate, %	84.59	83.17	86.34	84.01	85.64	86.61	86.67	87.56	87.04
	Velocity average path,[2]* μm/s								
	50	60	70	80	90	100	110	120	130	140
n	838	1,784	2,758	3,844	5,536	7,671	9,559	8,473	3,983	982
Total number born	13.72	13.83	13.86	13.85	14.02	14.08	14.13	14.11	14.24	14.03
	Velocity curvilinear,[3]* μm/s								
	100	125	150	175	200	225	250	275	300
n	982	3,430	6,352	9,486	12,022	8,752	3,498	802	160
Farrowing rate, %	88.23	86.26	86.4	85.46	87.13	86.50	86.63	86.77	80.38

[Adapted from Broekhuijse et al., 2012]

2017/11/8

CASA parameters and boar fertility

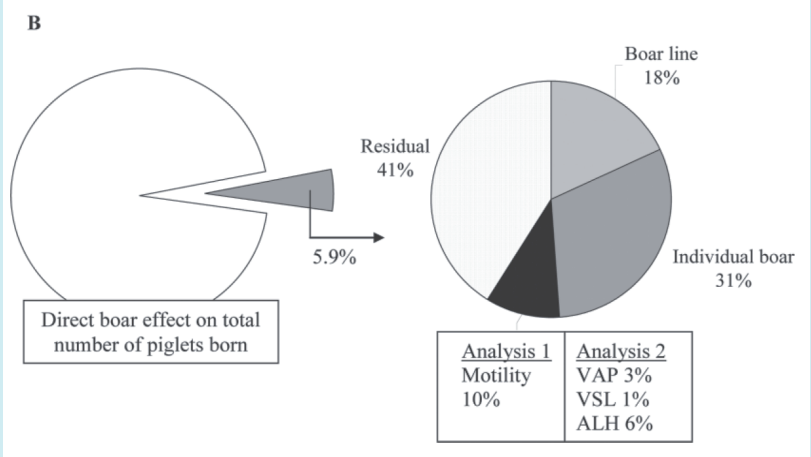
17



[Adapted from Broekhuijse et al., 2012] 2017/11/8

CASA parameters and boar fertility

18



[Adapted from Broekhuijse et al., 2012] 2017/11/8

Lab technician training

19

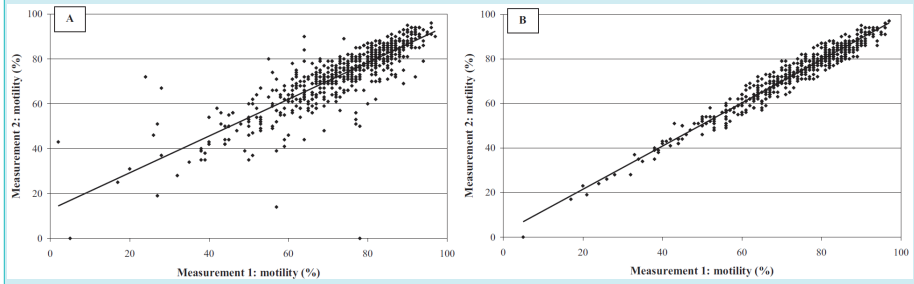
	Before training	During training	Current situation
Number of ejaculates	600	614	635
Number of cells per analysis	413 ± 35	456 ± 43	387 ± 61
Repeatability	71%	85%	96%
Coefficient of variation in motility scores	4.7%	3.3%	1.9%

Results represent mean ± SD.

[Adapted from Broekhuijse et al., 2011] 2017/11/8

Lab technician training

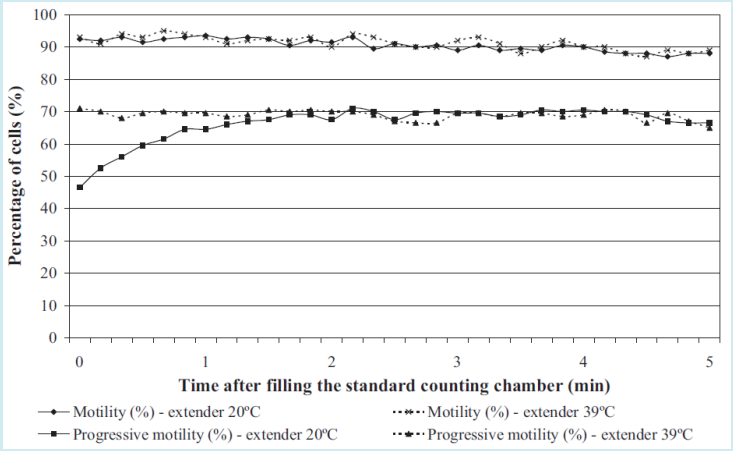
20



[Adapted from Broekhuijse et al., 2011] 2017/11/8

Sample temperature

21

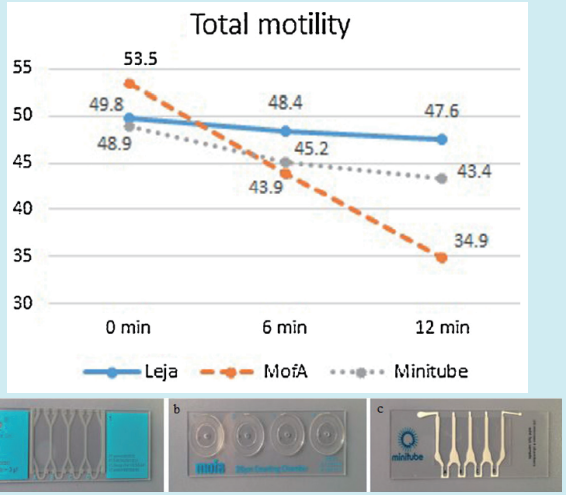


[Adapted from Broekhuijse et al., 2011]

2017/11/8

Viewing chamber

22

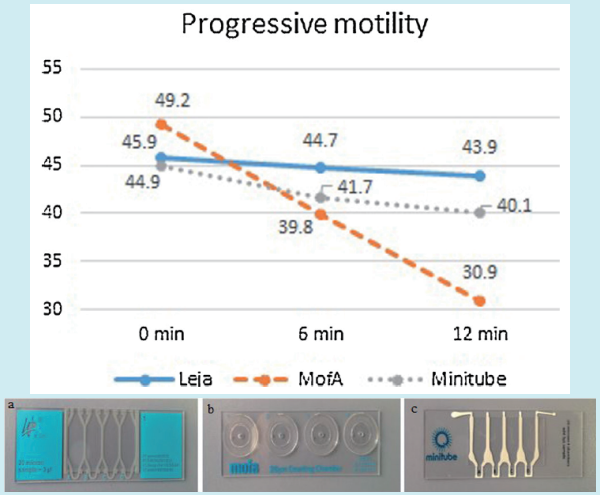


[Adapted from Ib˘anescu et al., 2016/Bull sperm]

2017/11/8

Viewing chamber

23



[Adapted from Ib˘anescu et al., 2016/Bull sperm]

2017/11/8

Different CASA systems

24