Transmission and life cycle patterns of *Toxoplasma*

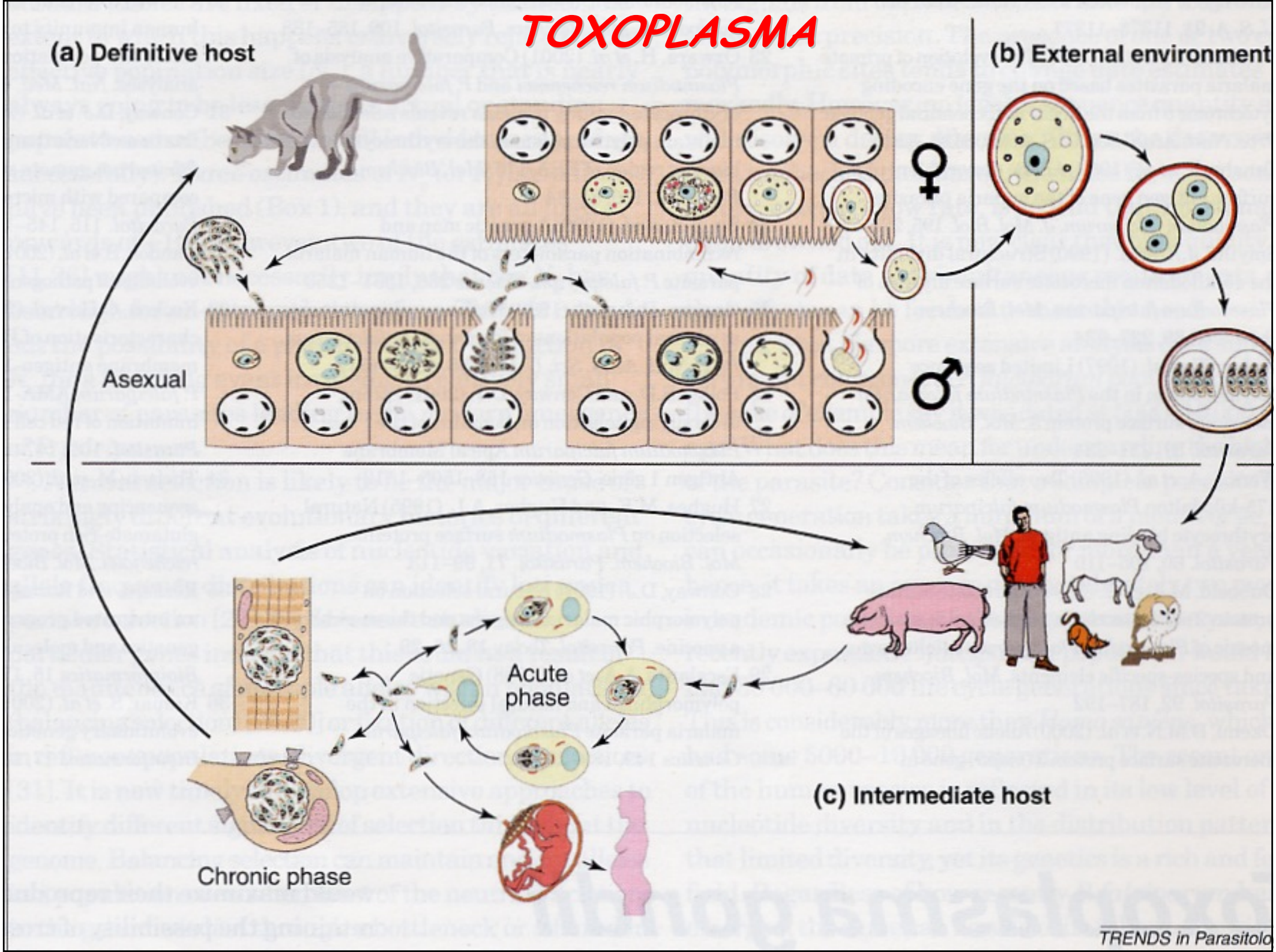
R. C. Andrew Thompson

World Health Organization for the Molecular Epidemiology of Parasitic Infections, School of Veterinary and Biomedical Sciences, Murdoch University, Western Australia

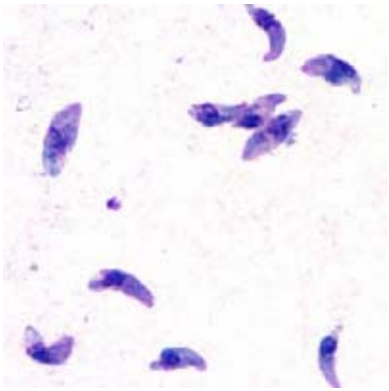
MURDOCH UNIVERSITY
PERTH, WESTERN AUSTRALIA

TOXOPLASMA IS A UBIQUITOUS PROTOZOAN PARASITE THAT INFECTS UP TO ONE THIRD OF THE WORLD'S HUMAN POPULATION AND A HUGE DIVERSITY OF VERTEBRATE FAUNA

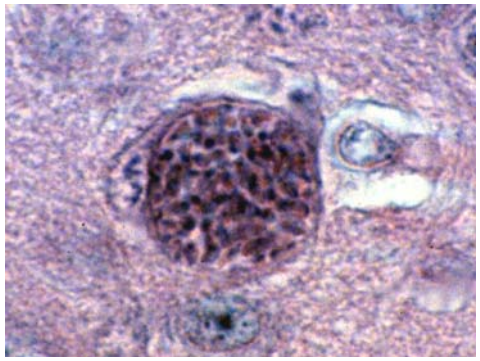
TOXOPLASMA


(a) Definitive host
(b) External environment
♀
♂
Asexual
Acute phase
Chronic phase
(c) Intermediate host
TRENDS in Parasitolo

Three infectious stages of *Toxoplasma*

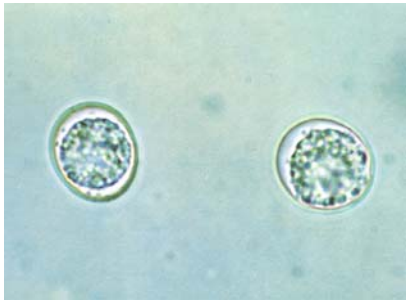


Tachyzoites



Bradyzoite
tissue cyst

Oocysts- environmental stage, in faeces of felids



Environmental
oocyst stage

GENETIC VARIATION

- Early studies concluded >95% of *T. gondii* isolates fall into three distinct genotypes
- These studies used isolates predominantly from humans and domestic animals in North America and Europe
- Ajzenberg et al (2004) proposed the genetic diversity of *T. gondii* in wildlife and geographically isolated areas is underestimated

TYPE X

TOXOPLASMA IN WILDLIFE

- Unusual genotype of *T. gondii* in California sea otters might represent an endemic *T. gondii* strain within this species = Type X (Miller *et al* 2004)

- 72% of all otter isolates are Type X (Conrad *et al* 2005)

- How is Type X maintained?

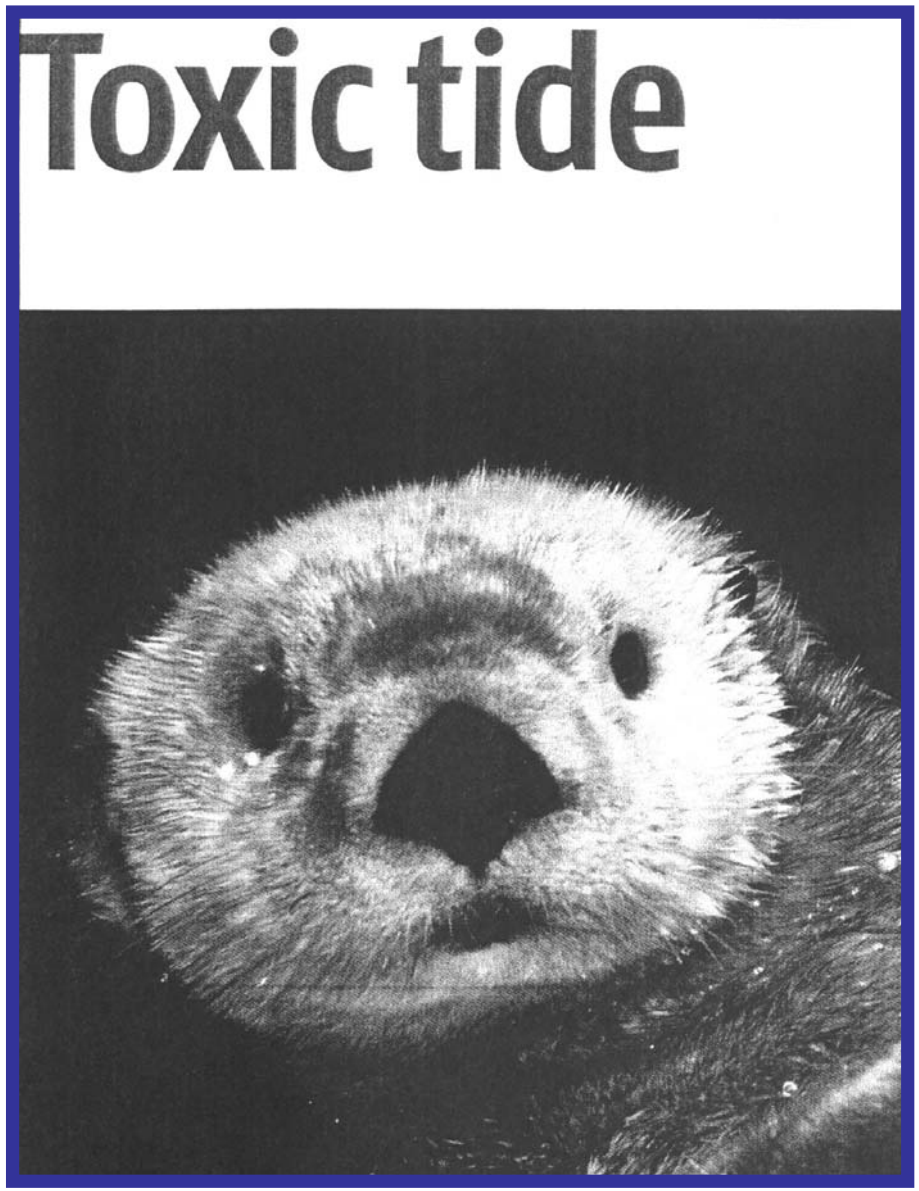
SEVIER

TRENDS in Parasitology

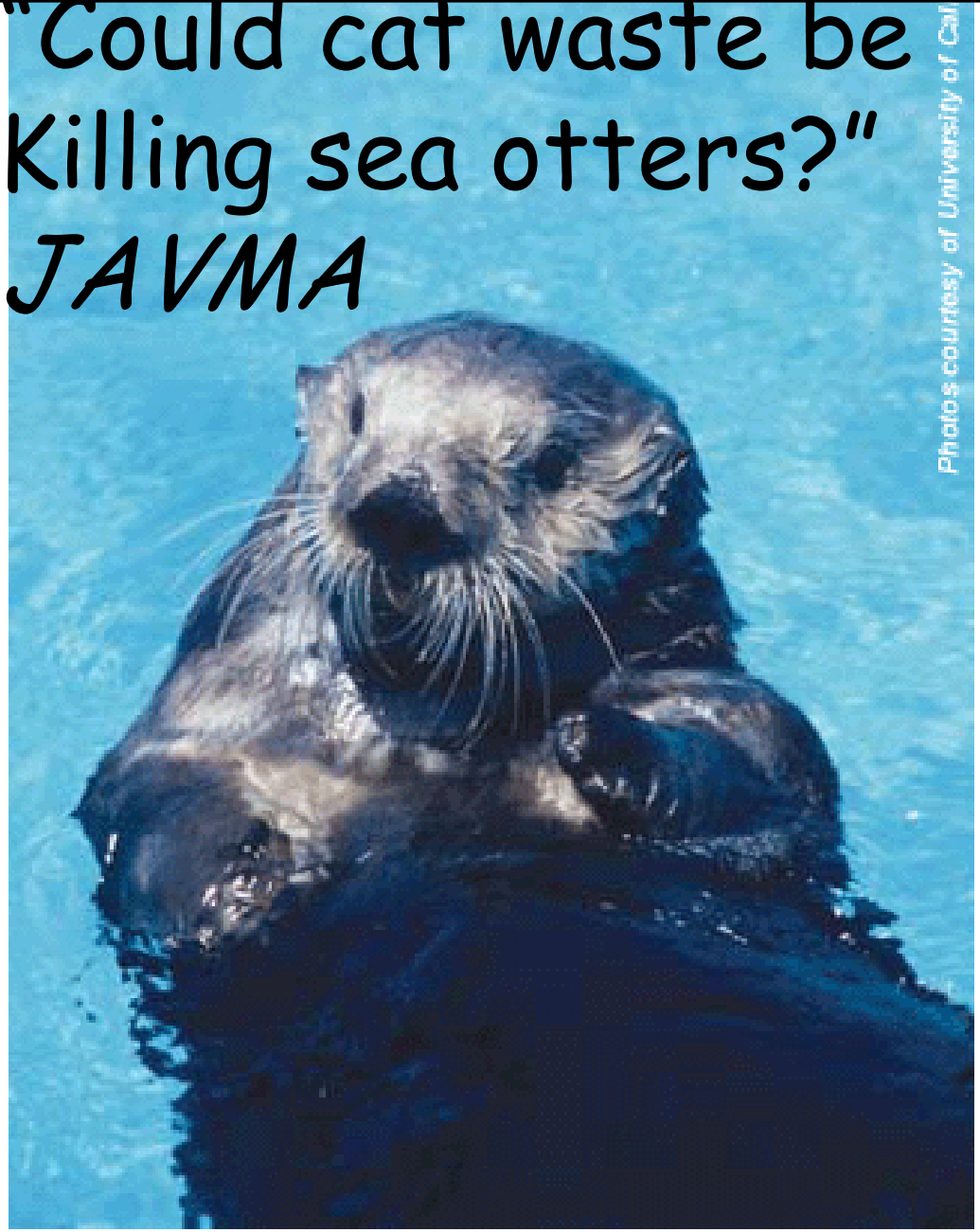
FORMERLY PARASITOLOGY TODAY

Zoonotic protozoa: from land to sea

"TOXIC TIDE"
New Scientist



"Could cat waste be Killing sea otters?"
JAVMA



A California study finds *Toxoplasma gondii* infections in southern sea otters exposed to freshwater runoff

HOW DID THE SEA OTTERS BECOME INFECTED WITH *TOXOPLASMA* ?

Table 2. Prevalence of *Toxoplasma gondii* antibodies in marine mammals

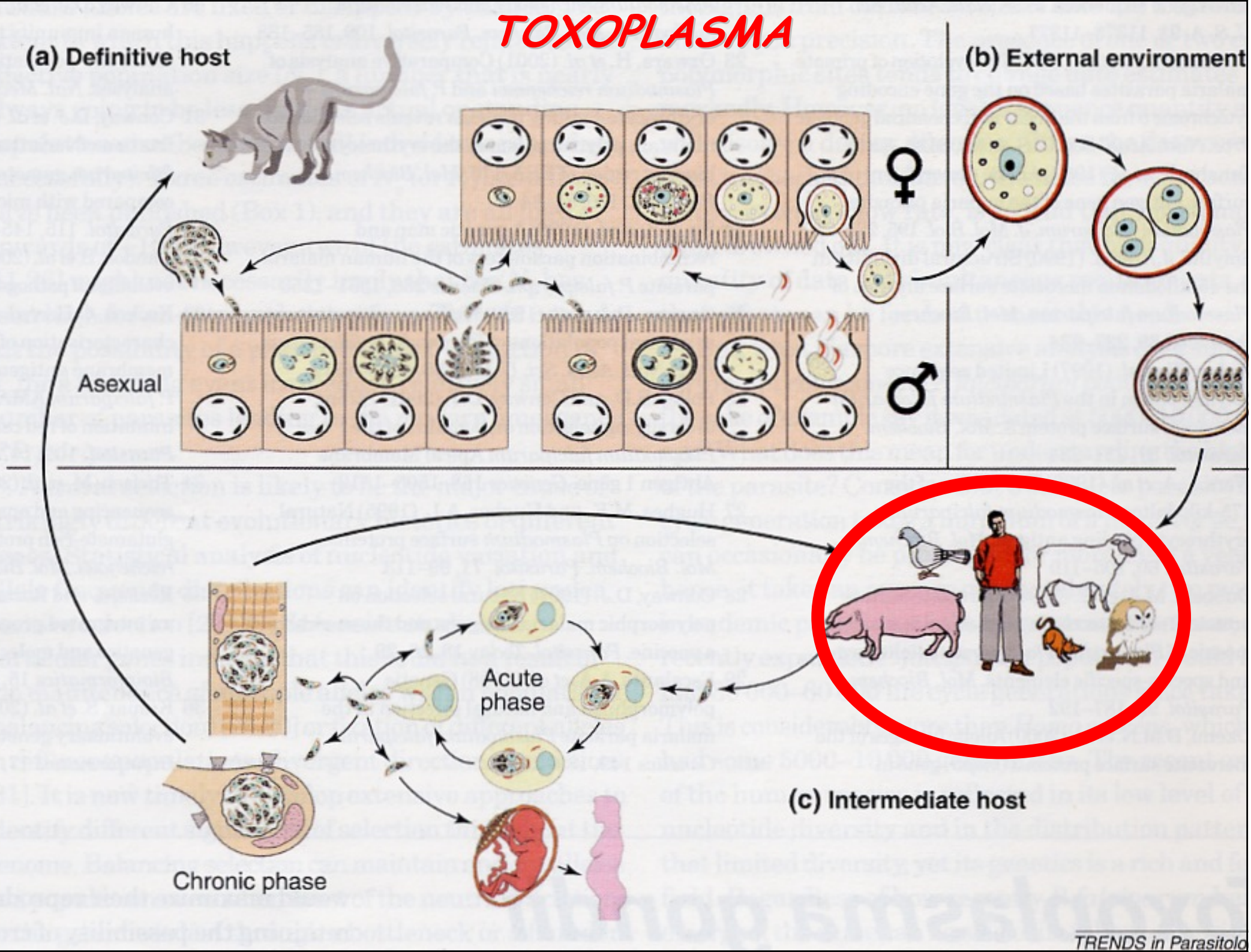
Species (common name)	Source	No. tested	No. positive	% positive (MAT ≥ 1:25
Enhydra lutris **(sea otter)**				
Dead	CA, USA	100	82	82.0
	CA, USA	107	66	62.0[b]
	WA, USA	15	7	47.0
Live	AK, USA	65	0	0[b]
	CA, USA	116	49	42.0[b]
	CA, USA	80	29	36.0[b]
	WA, USA	30	18	60.0
	WA, USA	21	8	38.0[b]
Odobenus rosamarus **(Pacific walrus)**	AK, USA	53	3	5.6
Zalophus californianus **(Californian sea lion)**	CA, USA	27	8	29.6
Phoca vitulina **(Harbor seal)**	AK, USA	311	51	16.4
	Canada	34	3	8.8
	WA, USA	380	29	7.6
Cystophora cristata **(Hooded seal)**	Canada	60	1	1.7
Phoca hispida **(Ringed seal)**	AK, USA	32	5	15.6
	Canada	34	3	8.8
Halichoerus grypus **(Grey seal)**	Canada	122	11	9.0
Erignathus barbatus **(Bearded seal)**	AK, USA	8	4	50.0
Phoca groenlandica **(Harp seal)**	Canada	112	0	0.0
Phoca largha **(Spotted seal)**	AK, USA	9	1	11.1
Phoca fasciata **(Ribbon seal)**	AK, USA	14	0	0
Tursiops truncatus **(Bottlenose dolphin)**	Spain	7	4	57.1
	CA, USA	94	91	96.8
	FL, USA	47	47	100.0
Stenella coeruleoalba **(Striped dolphin)**	Spain	36	4	11.1
Delphinus delphis **(Common dolphin)**	Spain	4	2	50.0

[a]Abbreviation: MAT, modified agglutination test.
[b]Indirect fluorescent antibody test.

Fayer *et al* 2004

How is a 'novel' strain of *Toxoplasma* maintained in aquatic mammals?

TOXOPLASMA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TRENDS in Parasitolo

RAISES
QUESTIONS ABOUT
TRANSMISSION
AND STRAIN TYPE

If a natural life cycle for *Neospora caninum* is by repeated vertical transmission, could this also be a natural life cycle for *T. gondii* ?

The role of vertical transmission in the epidemiology of toxoplasmosis needs to be re-evaluated

Johnson, 1997

Toxoplasma - Congenital transmission

Sheep - 61% of pregnancies of which 70% are successful

Duncanson *et al* 2001

Wild mice - 75% of pregnancies

Marshall *et al* 2004

Congenital transmission may be sufficient to explain the maintenance of *Toxoplasma* in natural populations of sheep without requiring new infections from oocysts excreted by cats

Duncanson et al. (2001)

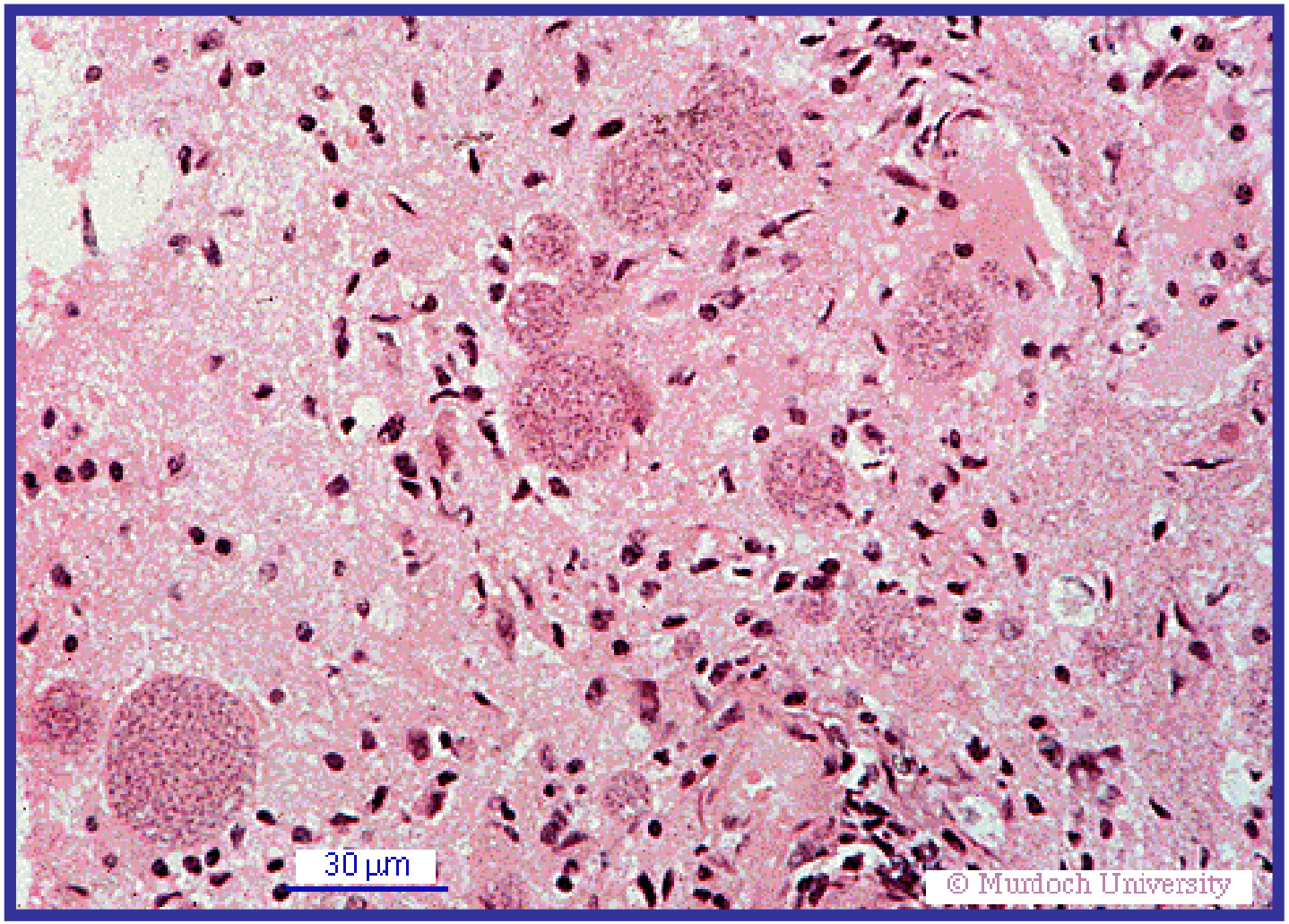
In Australia, there is no correlation between *Toxoplasma* in native mammals and the distribution of feral cats

We need to reconsider the role of vertical transmission in the maintenance of *Toxoplasma* in wildlife populations

TOXOPLASMOSIS in AUSTRALIAN WILDLIFE

Suggested that *Toxoplasma* may have been the cause of decline of some populations of native fauna in Australia *BUT* this has never been investigated or quantified.

MOST CLINICAL CASES OF TOXOPLASMOSIS IN NATIVE MARSUPIALS ARE IN CAPTIVE ANIMALS



TOXOPLASMA
INFECTION COMMON
IN WILD
POPULATIONS OF
NATIVE MARSUPIALS
WITH NO DISEASE

Prevalence of *Toxoplasma* antibodies in Australian Marsupials

- **3.3%** in Bennett's wallabies (n=151)[Johnson et al., 1988]
- **17.7%** in Tasmanian pademelons (n=85)[Johnson et al., 1988]
- **6.7%** in Eastern Barred Bandicoots (n=150) [Obendorf et al., 1996]
- **6.3%** in the Common Brushtail Possum (n=142) [Eymann et al., 2006]
- **21.1%** in Western Grey Kangaroos in Perth (n=95) [Parameswaran et al., 2008]

Brisbane mum's anguish

KANGAROO MEAT BLINDED MY BABY

A seemingly innocent snack at a cocktail party left her child at risk for life

WHEN pregnant Rhonda Ganko went into a Brisbane restaurant in good health, she never thought that she'd leave with an infection that threatened the life of her unborn baby and blinded her child.

Rhonda is so angry about what happened, she decided to talk to *Woman's Day* to warn other expectant mothers to be on their guard against eating undercooked kangaroo meat.

The only positive aspect of the Gankos's tragedy is that it has alerted the medical world to the potential

party I'd been to when I was pregnan with Isabella. I had eaten a sma amount of meat on a thin slice of brea The meat was covered with garnish an it wasn't until I bit into it that I realise it was very rare. I was upset at the tim because it was something I would neve have touched had I been able to se what it was."

Before leaving the party, Rhond discovered that it was kangaroo mea which is almost always served rare. Sh didn't think about it again until Isabell was diagnosed with toxoplasmosi

VERTICAL TRANSMISSION OF *TOXOPLASMA* IN AUSTRALIANMARSUPIALS

	DAMS	POUCH YOUNG
KANGAROOS:		
	10 Seropositive*	7 Seropositive
	10 PCR positive**	2 PCR positive
WOYLIES:		
	1 Seropositive	1 Seropositive
	1 PCR positive	1 PCR positive

* ELISA & MAT
** ITS1 & B1

VERTICAL TRANSMISSION OF *TOXOPLASMA* IN AUSTRALIANMARSUPIALS

- *T. gondii* detected in the pouch young of 2 chronically infected western grey kangaroos (*Macropus fuliginosus*) and 1 woylie (*Bettongia penicillata*).

- Vertical transmission of *T. gondii* in Australian marsupials is not uncommon.

- Maybe important in the maintenance of *T. gondii* infection in Australian marsupial populations.

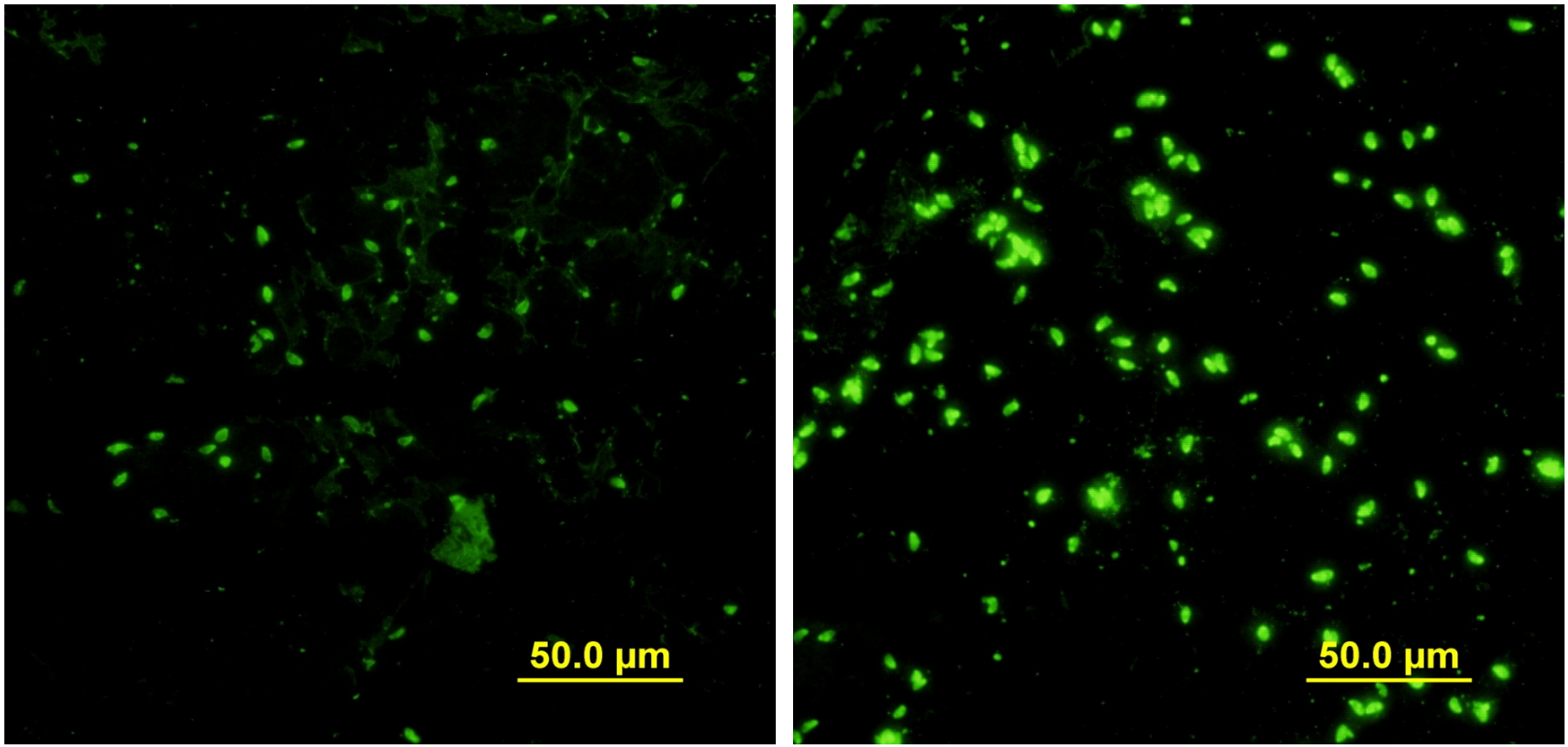
A Pilot Study of *T.gondii* in Free-range Chickens in Western Australia

Kamlang Chumpolbanchorn *et al*

Serology

- 20 sera from 5-week-old free-range chickens
- The sera were collected in August 2008 from a processing plant

Results
Positive sera 18/20

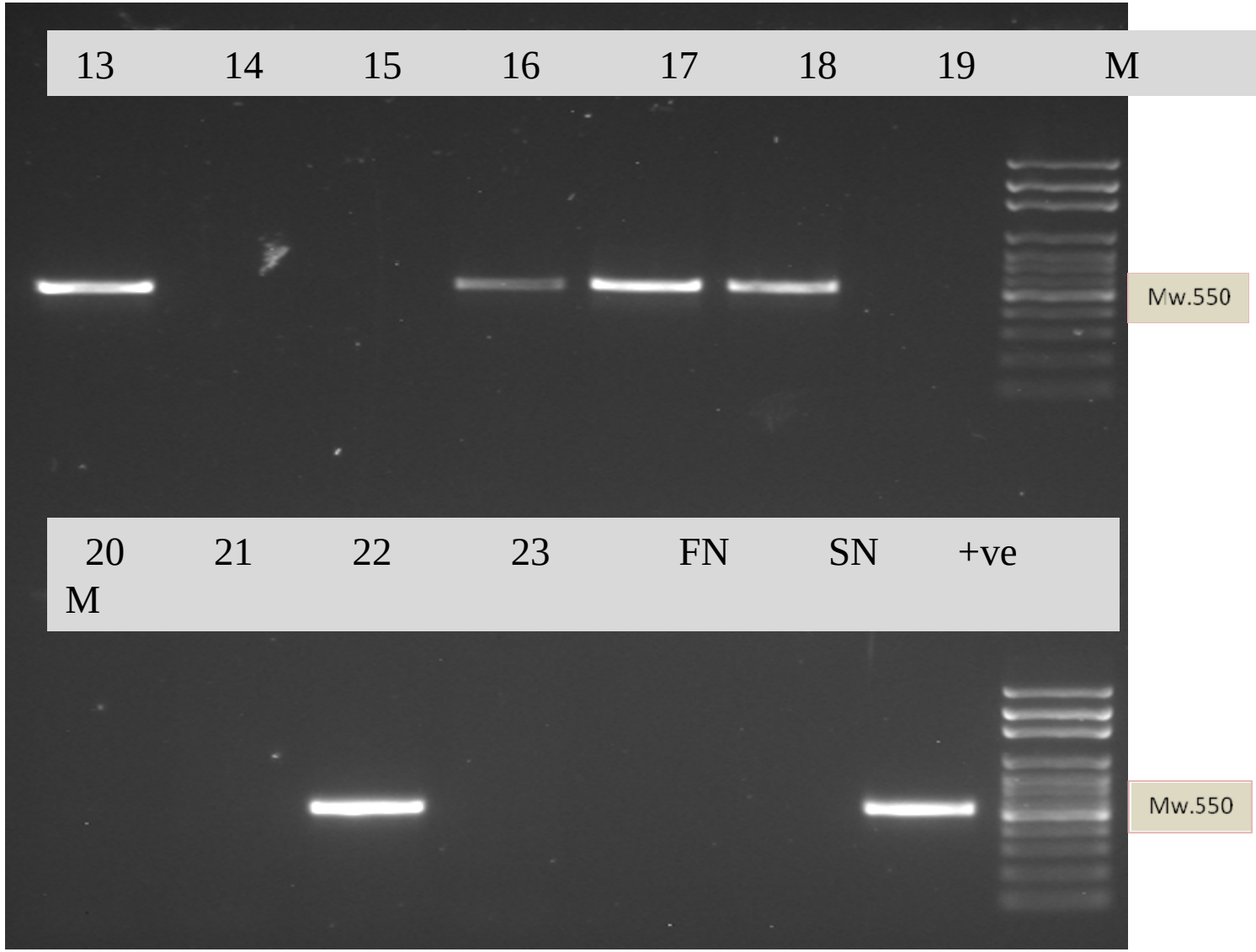


Molecular Biology

- 27 brains and 23 spleens from the same flock were collected in the same day
- Nested PCR with B1 gene

Results

Positive brains & spleens 22/50



Genetic diversity among Australian isolates of *Toxoplasma gondii*

Sampling locations

- Kimberley Archipelago
- Boodie and Montebello Islands
- Barrow Island
- Lorna Glen
- Faure Island
- Julimar
- Mt Caroline
- Karakamia
- Dryandra
- Upper warren
- Lake Magenta

Study Species

Kangaroo (*Macropus fuliginosus*)

Brush tailed possum (*Trichosurus vulpecula)*

Chuditch (*Dasyurus geoffroi*)

Woylie *(Bettongia penicillata)*

Shark Bay mouse (*Pseudomys praeconis*),

Western barred bandicoot (*Perameles bougainville*),

Banded hare-wallaby (*Lagostrophus fasciatus*),

Rock wallaby (*Petrogale* sp.), **Boodie** (*Bettongia lesueur*),

Bilby (*Macrotis lagotis*), **Quenda** (*Isoodon obesulus*),

Golden bandicoot (*Isoodon auratus*)

Objectives

- Obtain tissue samples from naturally infected animals in Australia, particularly those from wildlife
- Isolate *T. gondii* DNA from tissue samples using PCR for multiple genetic loci
- Sequence PCR products and identify single nucleotide polymorphisms (SNPs) present
- Compare Australian *T. gondii* isolates to types I, II and III


100
PACK


Please switch on bench sucker if you have open formalin, glycol or alcohol samples.

INITIAL STUDY

- 13 samples tested positive for *T. gondii* DNA using PCR for the B1 gene
- 6 of these had sequences consistent with the type I strain of *T. gondii*
- 7 samples had sequences inconsistent with strains I, II, III and X

Origin	Sample	317	360	366	378	504	533	Remarks
	Type I	G	C	T	G	G	A	(Grigg and Boothroyd, 2001)
	Type II/III	G	C	C/T	G	C/G	A	(Grigg and Boothroyd, 2001)
	Type X	G	C/G	T	G	C	A	(Miller et al, 2004)
Wild kangaroo	C14B	G	C	C/T	G	G	A/C	Atypical genotype
Wild kangaroo	C9B	G	C	T	A/G	C/G	A	Atypical genotype
Kangaroo meat retail	K2.8	G	C	T	A/G	C/G	A	Atypical genotype
Wild kangaroo	R7B	G	C	T	A	G	A	Atypical genotype
Wild kangaroo	J10T	G	C	T	G	C	A	Atypical genotype
Captive meerkat	A13	G	C	T	A	C/G	A	Atypical genotype
Wild kangaroo	J6B	T	G	T	G	C	A	Atypical genotype
Wild kangaroo	Q1T	G	C	T	G	G	A	Consistent with type I
Wild kangaroo PY	PYR19H	G	C	T	G	G	A	Consistent with type I
Wild woylie	A1b	G	C	T	G	G	A	Consistent with type I
Wild woylie PY	A1Ya	G	C	T	G	G	A	Consistent with type I
Horse meat	A7	G	C	T	G	G	A	Consistent with type I
Captive mouse	A8	G	C	T	G	G	A	Consistent with type I

ONGOING MOLECULAR CHARACTERISATION

Positives	Strain type*
8/13 Kangaroo	2 Type I, 6 Novel
12/17 Woylie	6 Type I, 2 Type II, 3 Novel
1/2 Possum	Novel
1 Bandicoot	Novel
12/19 Chuditch	TBS
1 Bilby	TBS
1 Quokka	TBS
5/8 Crow	4 Type 1, 1 Novel

*B1, SAG 3, SAG 4

TOXOPLASMOSIS in AUSTRALIAN WILDLIFE

It is presumed that *Toxoplasma* was introduced into Australia with domestic cats.

BUT???

THANKYOU