

Does vertical transmission play a role in generating the high prevalence and ubiquity of *Toxoplasma gondii* in natural populations of humans and animals?

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Overview

- Background to disease
- Transmission in mammalian hosts
- Sheep, rodents and humans

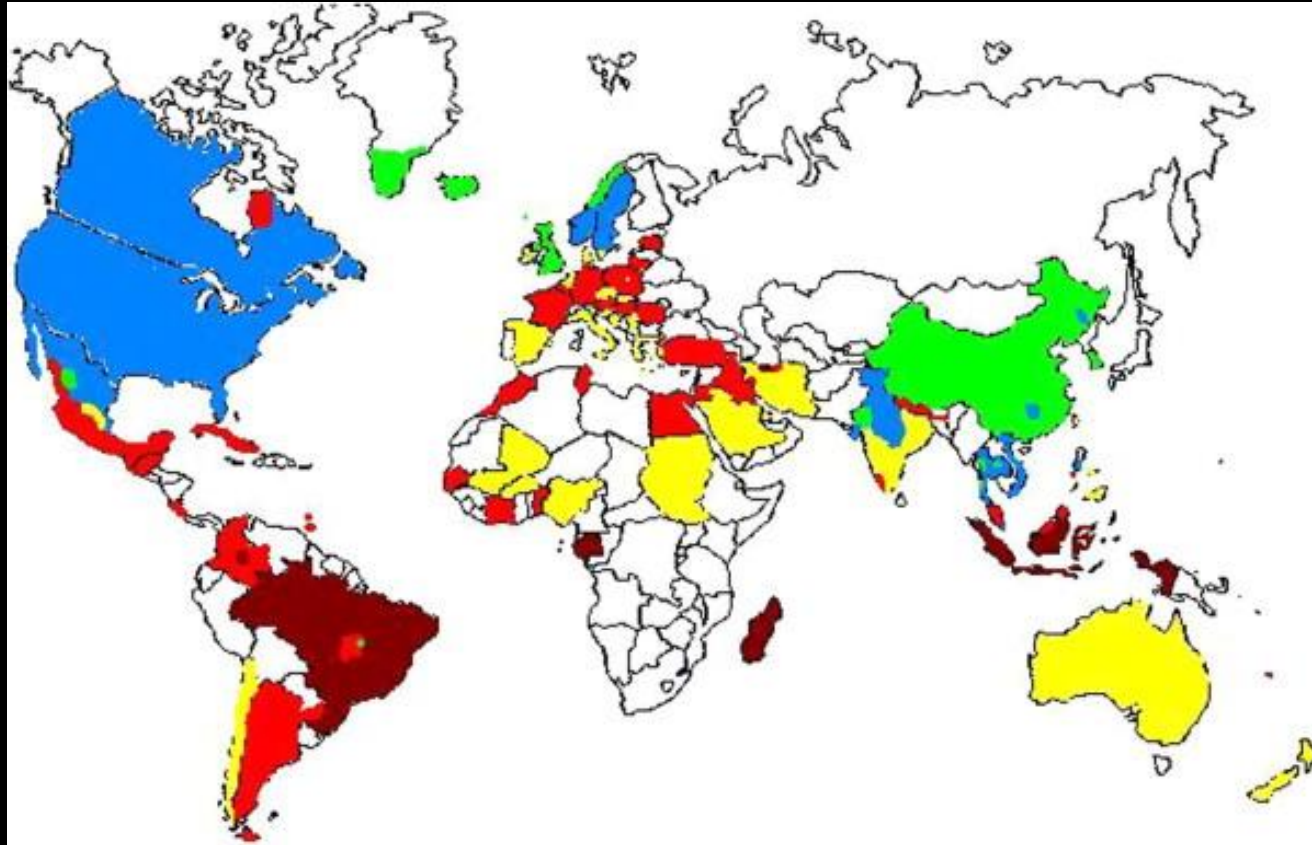
Toxoplasmosis

- *Toxoplasma gondii*
- Wide geographical range
- All warm blooded animals
- High prevalences
- Parasite of the cat

Disease

- Serious disease
- Abortion/miscarriage
- Ocular disease
- Domestic animals + humans

Global *Toxoplasma* prevalence



< 10%



10 - 20%

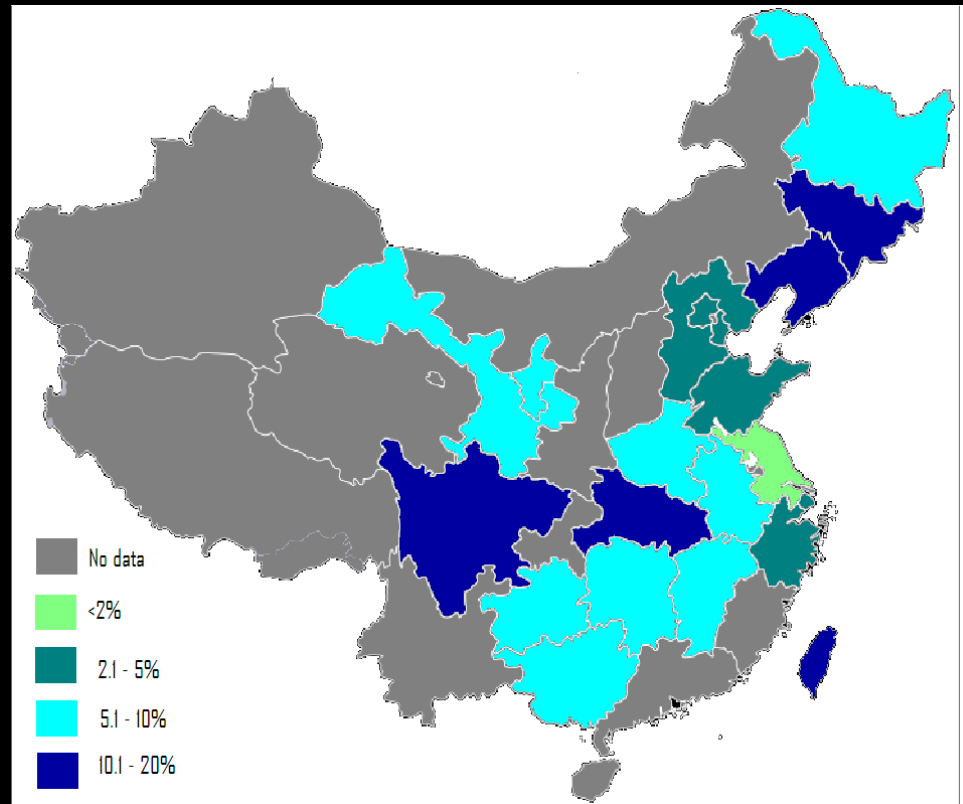


20 - 40%

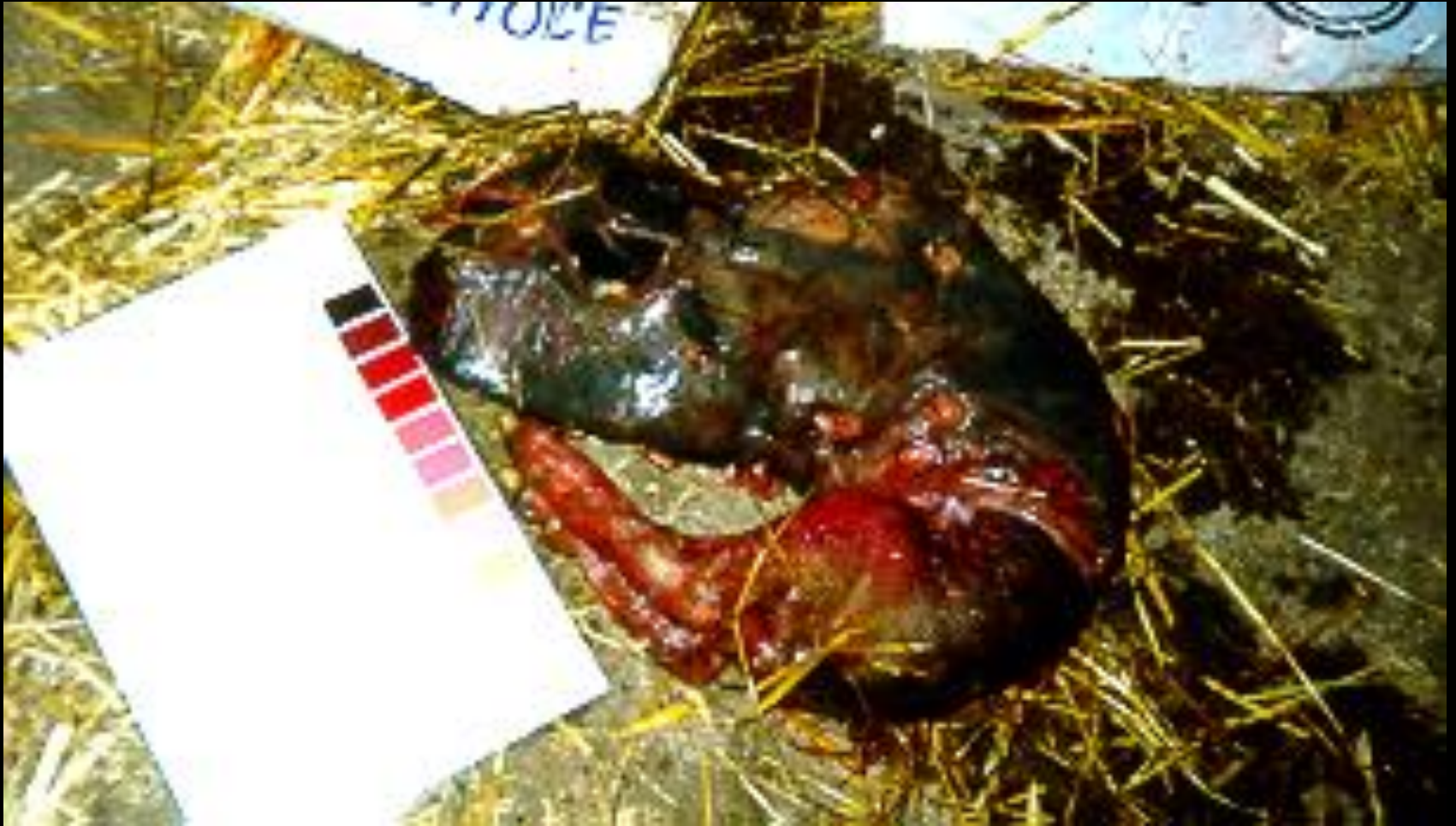


> 40%

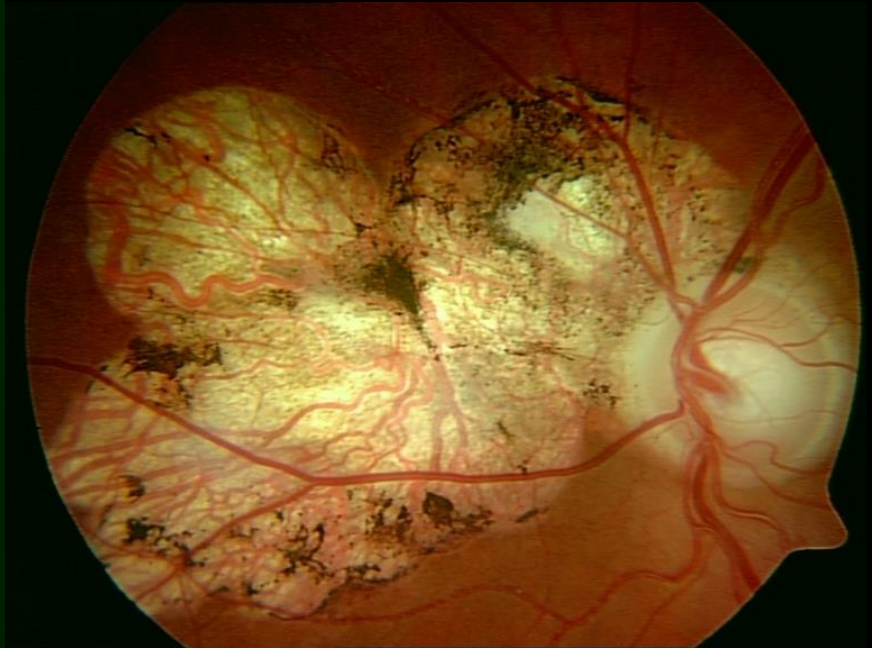
Toxoplasma prevalence in pregnant women in China



Abortion in sheep



Ocular disease – age 19 and 40



Life cycle of *Toxoplasma gondii*

Life cycle of *Toxoplasma gondii*

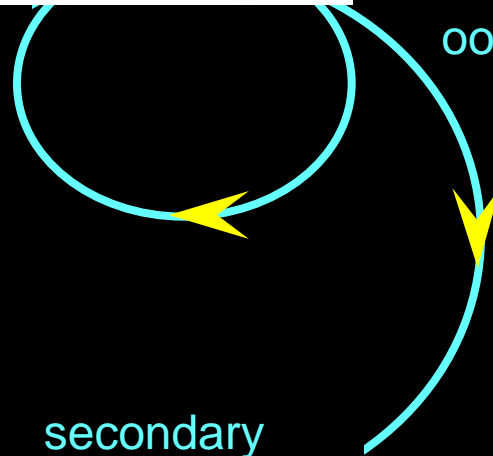


Cat/felids

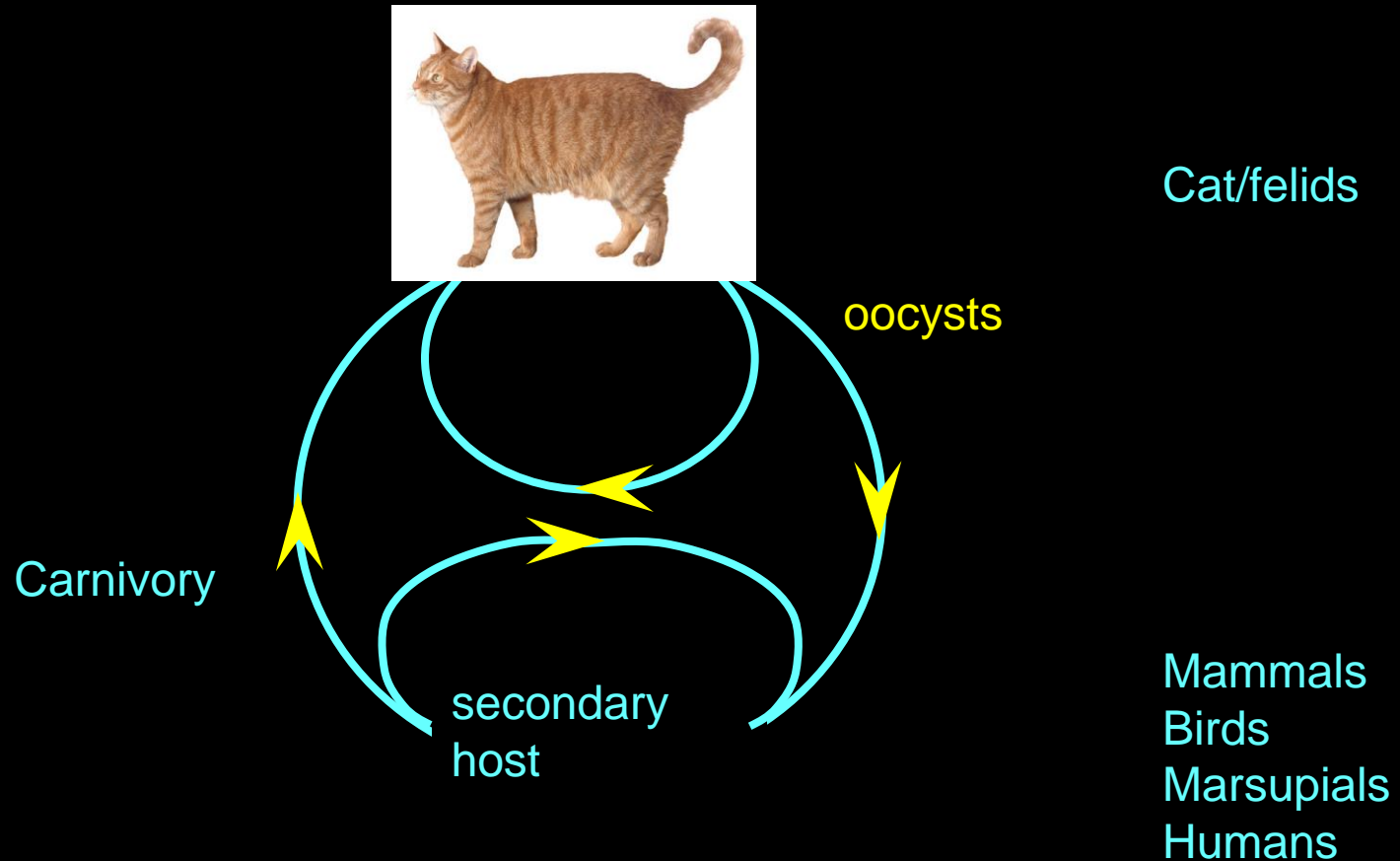
oocysts

secondary
host

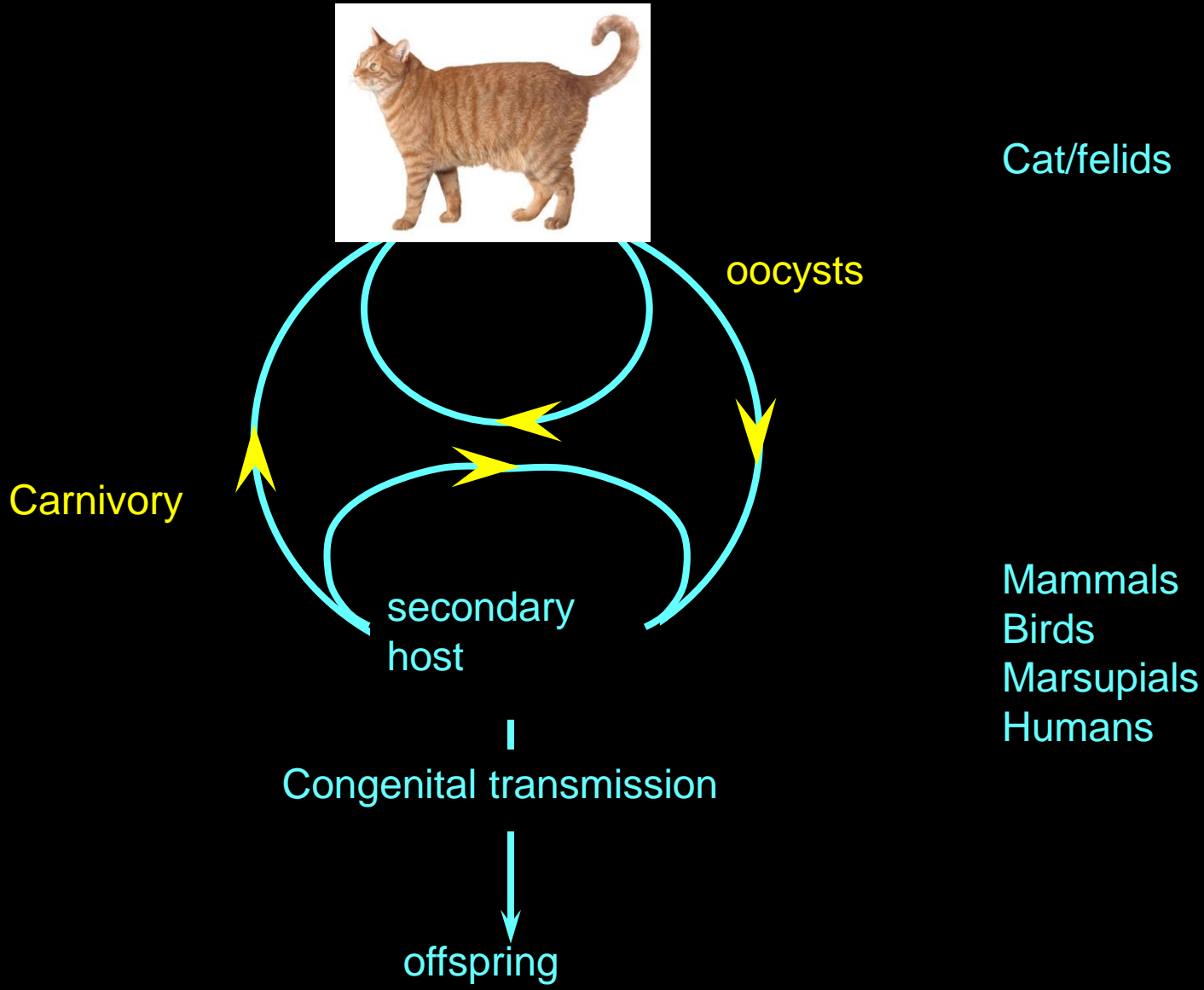
Mammals
Birds
Marsupials
Humans



Life cycle of *Toxoplasma gondii*



Life cycle of *Toxoplasma gondii*



Life cycle of *Toxoplasma gondii*



Cat/felids

oocysts

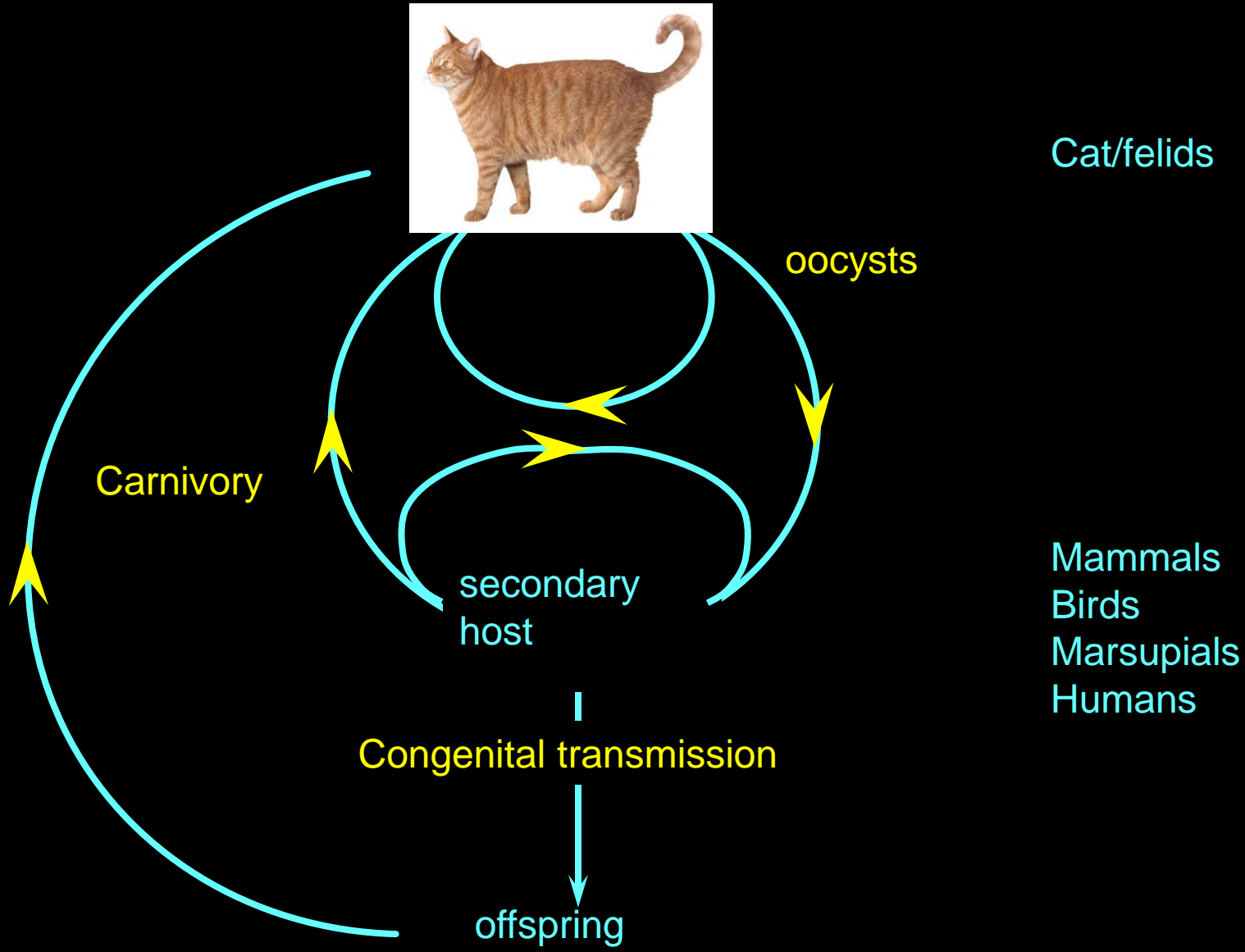
Carnivory

secondary host

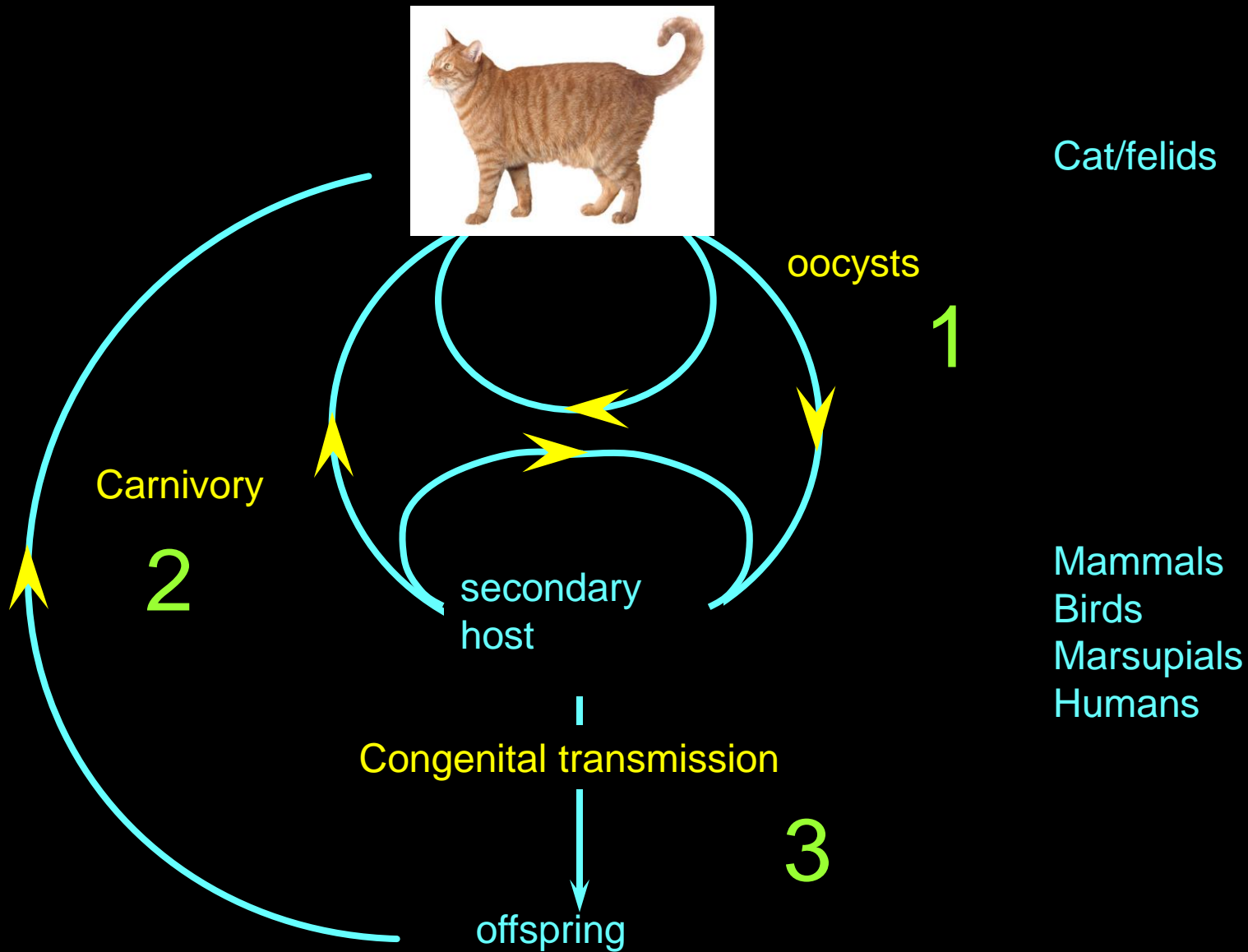
Mammals
Birds
Marsupials
Humans

Congenital transmission

offspring



Life cycle of *Toxoplasma gondii*



A dilemma...

- Cat is the only definitive host
- Parasite is ubiquitous and is often found at high prevalences

What is the importance of transmission cycles which bypass the cat?

Is congenital/vertical transmission important in natural populations of rodents?

Study 1

Wild population of mice:

Study and Objectives

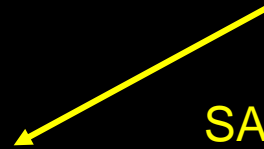
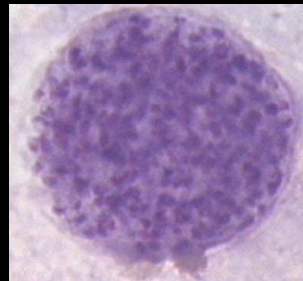
- 200 mice were trapped from an urban location
- Analysis of infection rates using PCR (SAG1 Gene)
- Analysis of transmission in pregnant mice



n = 200



n = 16



SAG1 - PCR

Results

- 59% of mice infected with Toxoplasma
- 16 mice pregnant
- 12 of the pregnant mothers were positive

Results - pregnant mice

Female status	Pregnant females	Number of foetuses	Infected foetuses %
Infected	12	47/63	74.6
Uninfected	4	0/15	0
Total	16	47/78	60.2%

Results - pregnant mice

Female status	Pregnant females	Number of foetuses	Infected foetuses %
Infected	12	47/63	74.6
Uninfected	4	0/15	0
Total	16	47/78	60.2%

Conclusions

- High frequency of congenital transmission (100% from infected mothers)
- 75% of pregnancies (overall) and 74.6% of foetuses
- Congenital transmission may be a general phenomenon

Study 2

Captive population of mice: Study and Objectives

- Colony of mice set up from wild mice "founders"
- Colony existed for >5years and many generations
- Tested for *Toxoplasma* by SAG1-PCR

Results

- Tested 89 mice
- 68/89 (76.4%) infected
- Genotyped mice - highly inbred

Conclusions

- High prevalence of infection
- Closed colony, therefore cats not involved
- Congenital transmission may be a general phenomenon

Study 3

Wild population of woodmice living in an area relatively free of cats:
Study and Objectives

- *Apodemus sylvaticus* (Woodmice) collected from a cat free area
- 2 were pregnant
- Tested for *Toxoplasma* by SAG1-PCR

Results

- Tested 206 woodmice
- 84/206 (40.78%) infected
- 1 of 2 pregnant mice were infected and transmitted to the foetus (100% congenital)
- *Toxoplasma* transmission occurring at high frequency in the absence of cats

Results

- Comparison *Toxoplasma* prevalence in an area with and without cats
- With cats (>500 cats/km²) - 59%
- Without cats (<2.5 cats/km²) - 40.78%

Study 4

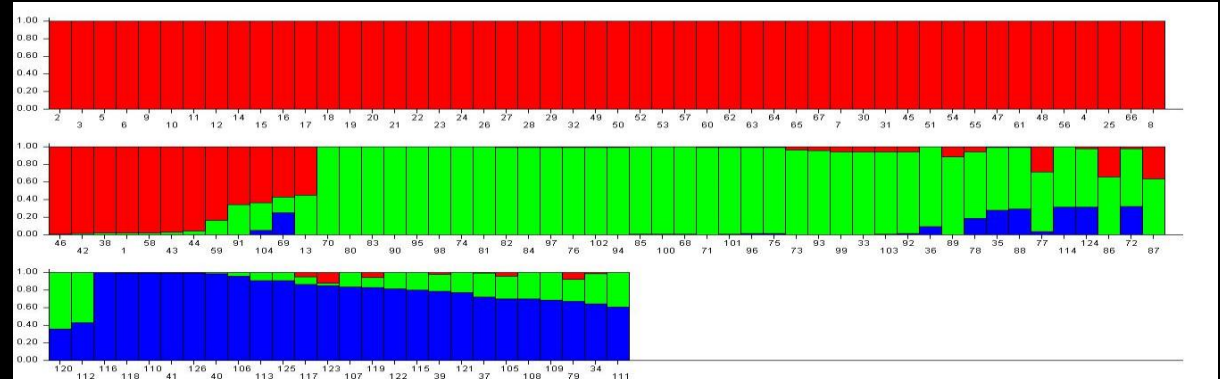
Spatial and genotypic distribution of infection in a natural population

Study and Objectives:

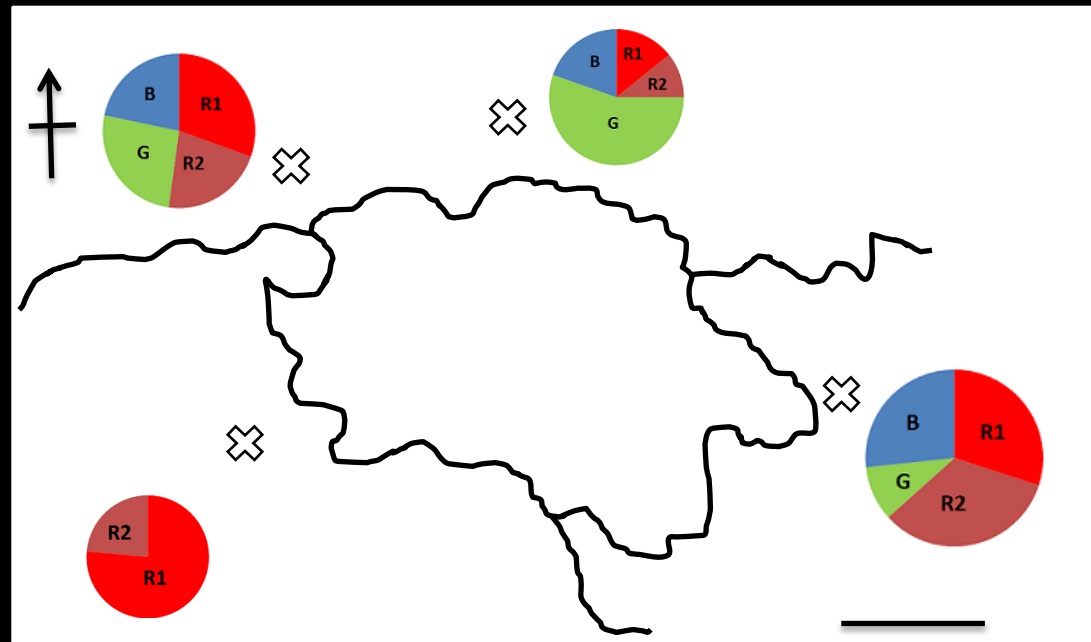
- *Apodemus sylvaticus* (Woodmice) collected and genotyped using microsatellite markers
- Tested for *Toxoplasma* by SAG1-PCR

Results

Populations
by micro -
satellite
DNA analysis



Geographical
locations



Results

Population	No. tested	No. Positive for <i>T. gondii</i>	Prevalence (%)
R1	37	5	13.5%
R2	25	8	32.0%
G	40	18	45.0%
B	24	13	54.2%

Conclusions

- A highly significant association between genetic group and prevalence ($P=0.004$)
- No significant association with location of mice ($P=0.125$)

Conclusions

- Evidence for high frequency of congenital transmission in both wild and captive populations of mice
- High prevalence even in the absence of cats

Is congenital/vertical transmission important in natural populations of sheep?



Current views

- Sheep infected by oocyst
infected feed/bedding/water
- Immunity following infection -
safe to breed from infected
ewes
- Vertical transmission at low
levels

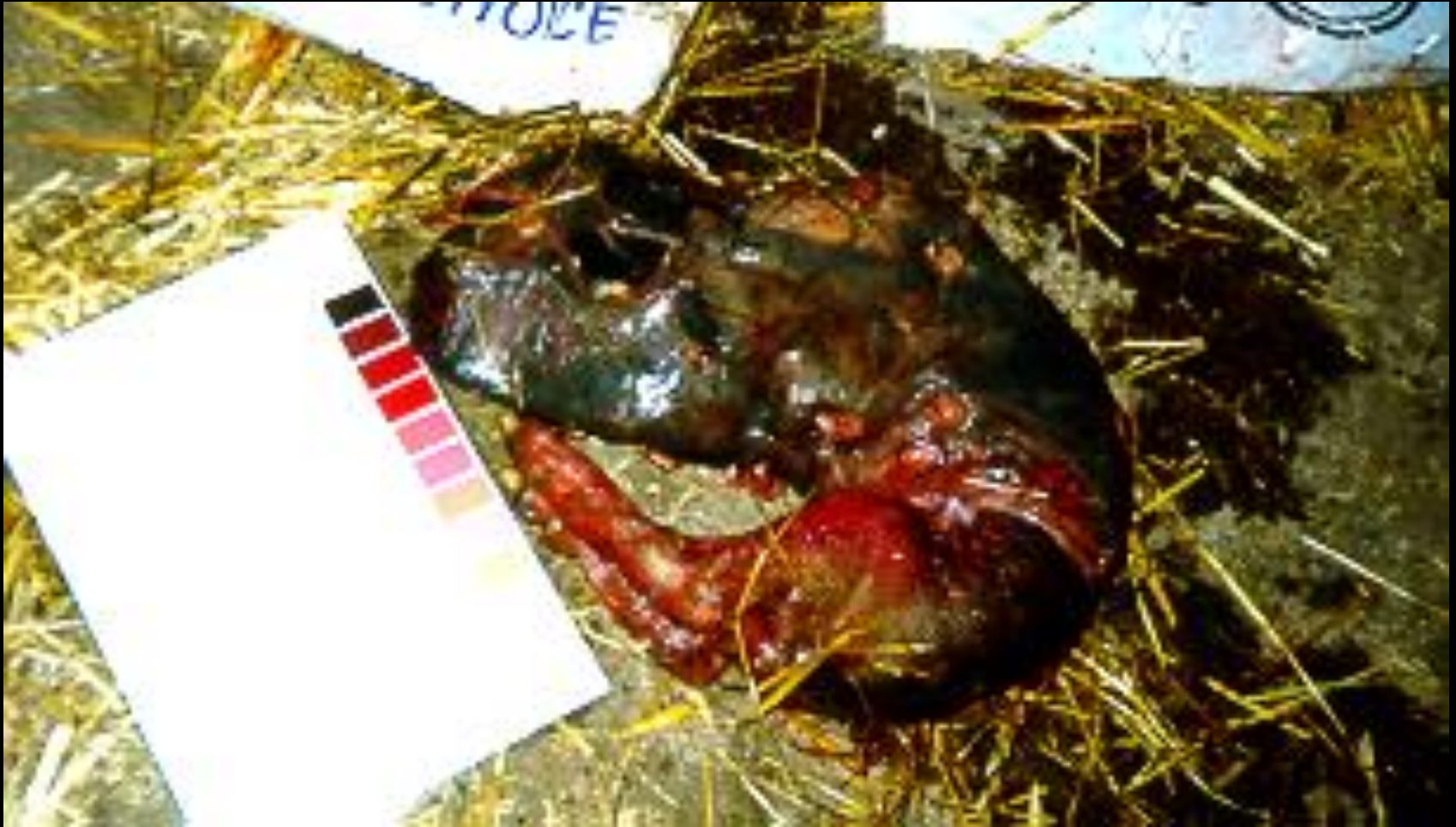
Study and Objectives

- Sheep are not carnivores
- Measure vertical transmission
- Use PCR to detect parasites from newborn lambs

Sampled umbilical cord tissue from newborn lambs



Sampled internal tissues from aborted lambs



Results - 5 lambings (3 years)

	Successful pregnancies	Unsuccessful pregnancies	Total pregnancies
Pregnancies	334	58	392
PCR Positive	218	53	271
Percentage	65%	91%	69%

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Conclusions

- High levels of congenital transmission
- Healthy infected lambs
- Abortion associated with infection
- Possible role for vertical transmission
- Wrong advice to farmers?

Do we detect differences
in *Toxoplasma* infection in
different families of
sheep?

Hypothesis 1

- Ingestion of oocysts from feed/bedding/water

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- Ingestion of oocysts from feed/bedding/water

Expectation 1

- Infection with *Toxoplasma* should be randomly distributed in families on the same farm

Hypothesis 2

- Vertical transmission: ewe to lamb

Hypothesis 2

- Vertical transmission: ewe to lamb

Expectation 2

- Different infection levels in different families of sheep

Study and Objectives

- Charollais pedigree flock
- Detailed lambing records going back 11 years
- Sampled lambs for *Toxoplasma* for 3 years
- Look for differences in infection levels in families

Family	Abortion %	Family	Abortion %
A901	48	R891	23
K891	40	D971	22
G921	40	S921	20
L921	38	I921	20
F891	36	T941	18
C921	33	U911	18
E921	33	V921	13
M981	30	W901	12
B921	29	X891	8
N971	27	Y901	8
H921	26	Z921	5
O921	26	J921	3
P881	24	AA891	0
Q911	23		

Family	Abortion %	Family	Abortion %
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B921	29	X891	8
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H921	26	Z921	5
O921	26	J921	3
P881	24	AA891	0
Q911	23		

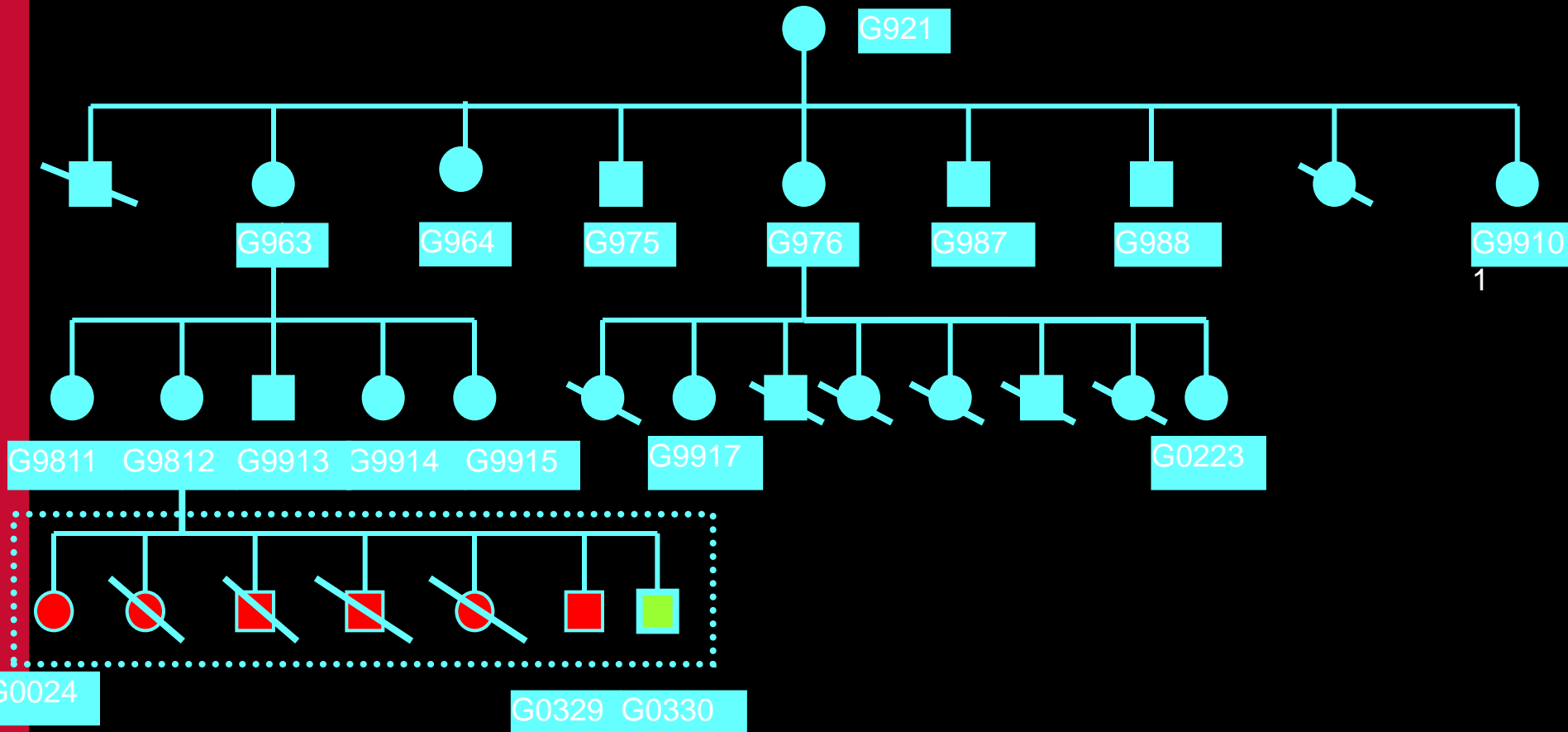
Family	Infection %	Family	Infection %
A901	100	R891	50
K891	100	D971	100
G921	86	S921	20
L921	100	I921	20
F891	83	T941	18
C921	83	U911	20
E921	100	V921	22
M981	75	W901	17
B921	75	X891	14
N971	100	Y901	0
H921	75	Z921	0
O921	73	J921	0
P881	67	AA891	0
Q911	60		

Family	Infection %	Family	Infection %
A901	100	R891	50
K891	100	D971	100
G921	86	S921	20
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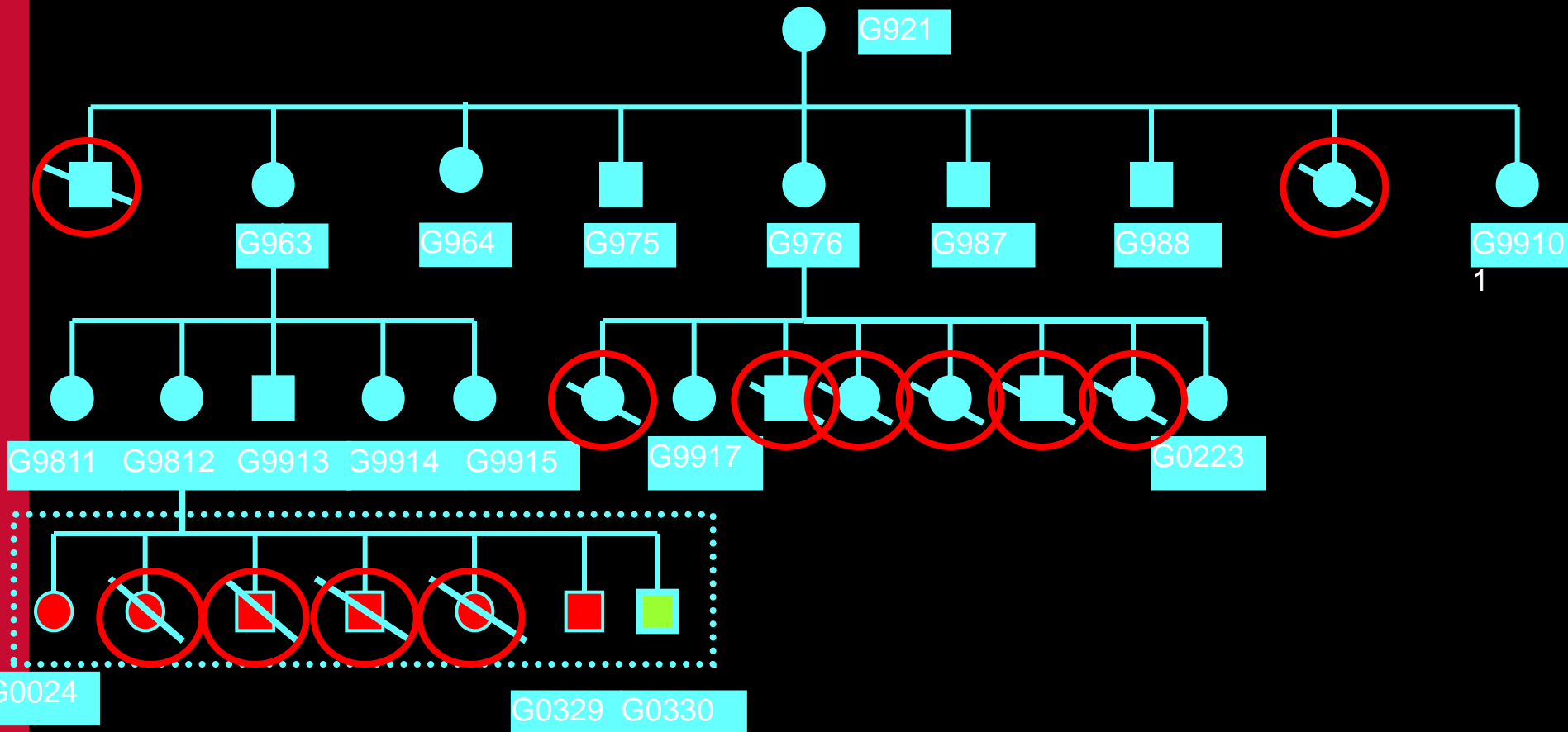
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M981	30	75	W901	12	17
B921	29	75	X891	8	14
N971	27	100	Y901	8	0
H921	26	75	Z921	5	0
O921	26	73	J921	3	0
P881	24	67	AA891	0	0
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O921	26	73	J921	3	0
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Q911	23	60			

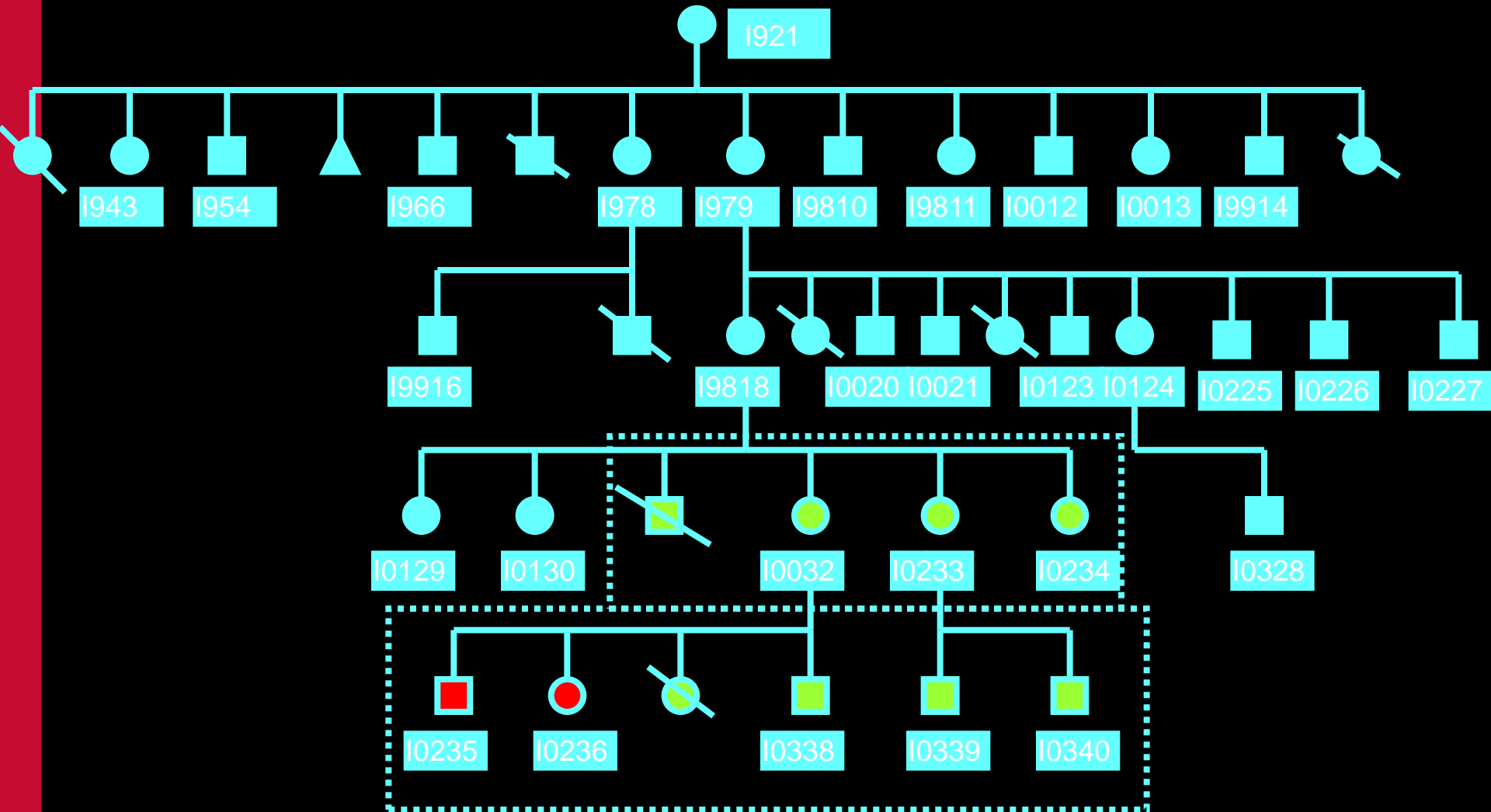
Family with high frequency of abortion and infection



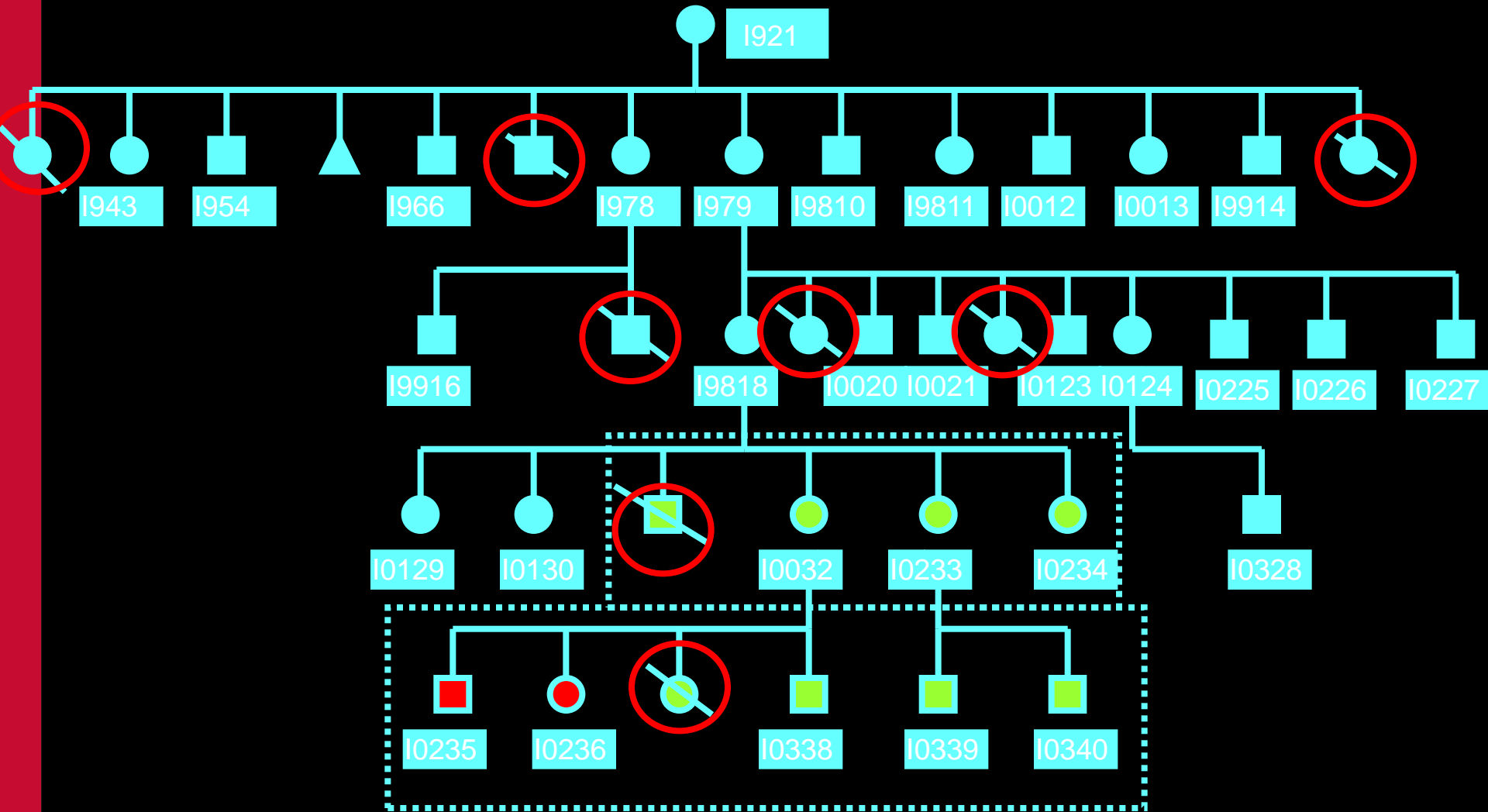
Family with high frequency of abortion and infection



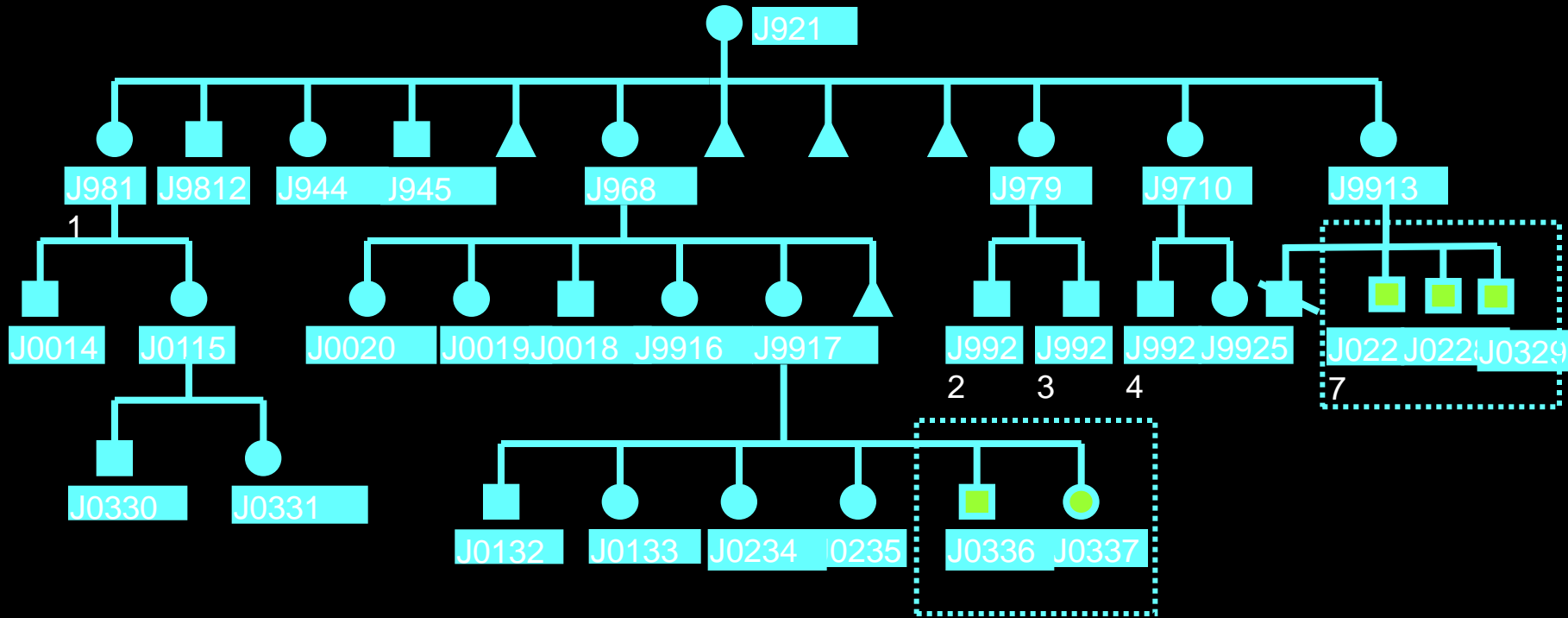
Sheep family with intermediate frequencies of abortion and infection



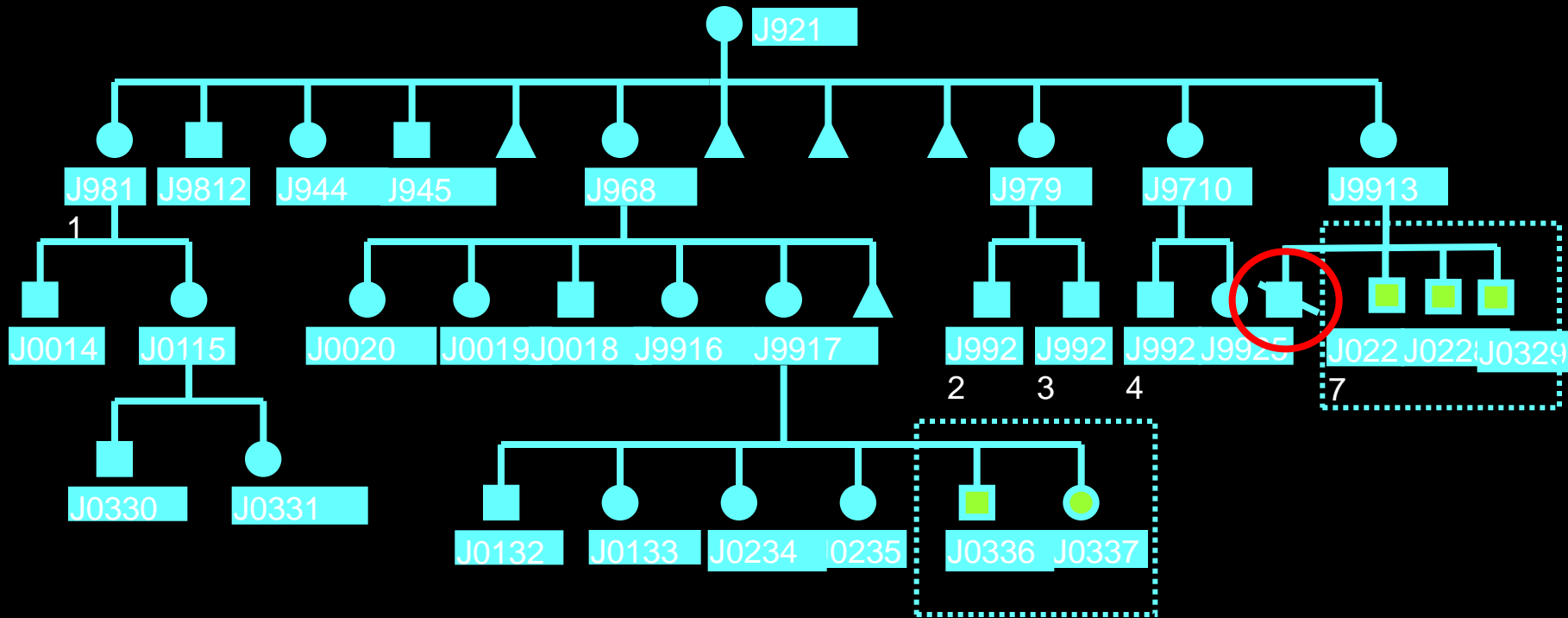
Sheep family with intermediate frequencies of abortion and infection



Sheep family with low frequency of abortion and infection



Sheep family with low frequency of abortion and infection



Results

- Abortion rates: highly significant difference from randomness ($P < 0.01$)
- Infection rates: highly significant difference from randomness ($P < 0.01$)
- Highly significant correlation between frequency of abortion and frequency of infection ($r = 0.89$, $n = 27$, $P < 0.01$)

Significance

- Strong evidence for the importance of vertical transmission
- Breeding from infected families may **increase** infection and abortion rates

What is the risk of increasing infection by breeding from infected ewes?

- High risk of subsequent abortion 55%
- High risk of subsequent infection 69%

Evidence that primary infection of Charollais sheep with *Toxoplasma gondii* may not prevent foetal infection and abortion in subsequent lambings

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Is congenital/vertical
transmission important in
humans?

Current views

- Congenital transmission occurs at very low levels (1-2/1000).
- Can result in significant disease - but rare
- Associated with infection during pregnancy
- Serological detection systems commonly used

Study and Objectives - 2 studies

- Miserata Central Hospital, Libya
- 276 umbilical cord samples from 272 pregnancies
- Stepping Hill Hospital, Manchester, UK
- 94 umbilical cord samples

Results - human samples Libya

	Successful pregnancies	Unsuccessful pregnancies	Total pregnancies
Pregnancies	267	5	272
PCR Positive	27	0	27
Percentage	10.1%	0%	9.93%

Results - human samples Libya

	Successful pregnancies	Unsuccessful pregnancies	Total pregnancies
Pregnancies	267	5	272
PCR Positive	27	0	27
Percentage	10.1%	0%	9.93%

Results - human samples Manchester

	Successful pregnancies	Unsuccessful pregnancies	Total pregnancies
Pregnancies	99	0	99
PCR Positive	33	0	33
Percentage	33.3%	0	33.3%

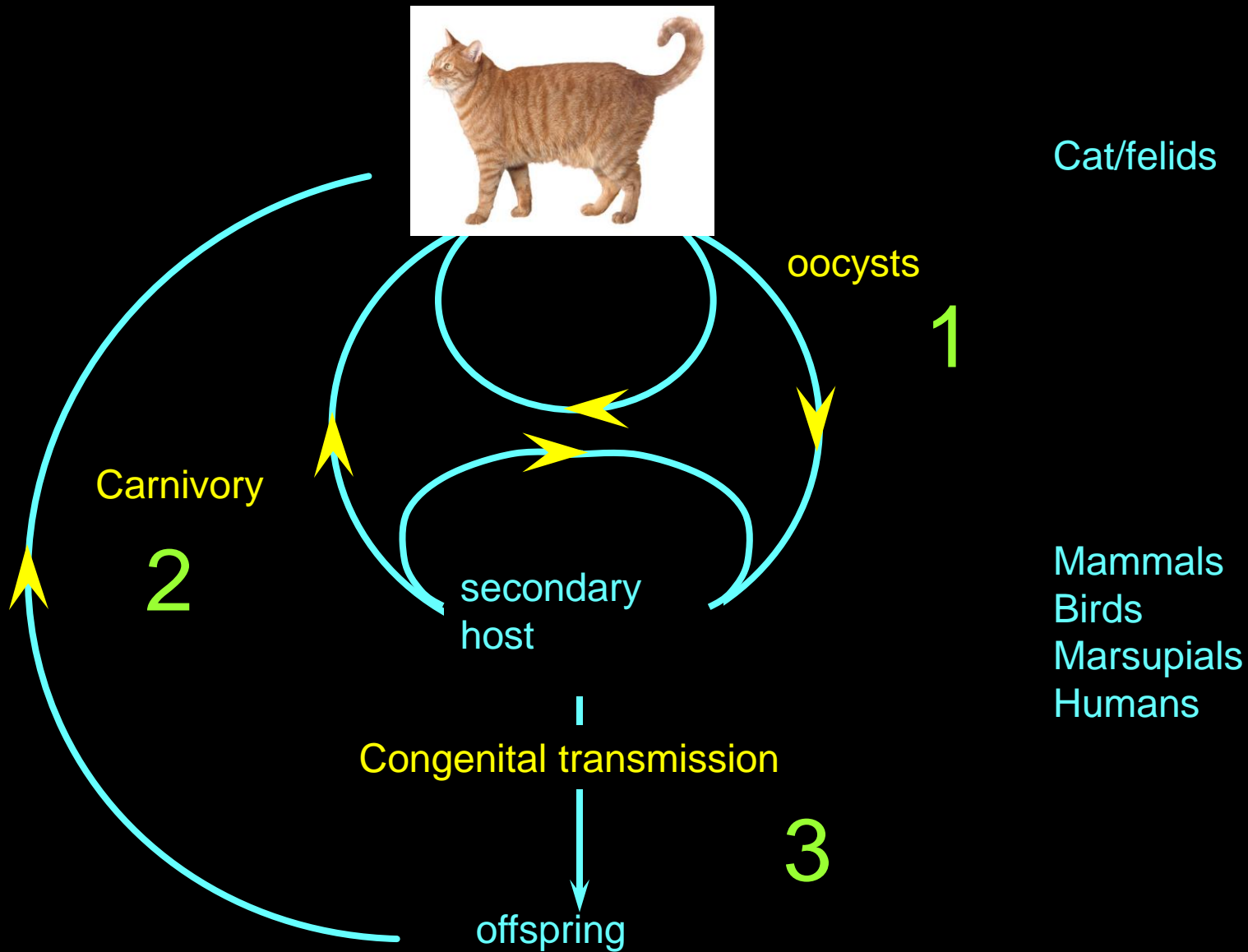
Results - human samples Manchester

	Successful pregnancies	Unsuccessful pregnancies	Total pregnancies
Pregnancies	99	0	99
PCR Positive	33	0	33
Percentage	33.3%	0	33.3%

Conclusions

- High frequency of congenital transmission
- Healthy infected babies born
- Possible evidence for vertical transmission

Life cycle of *Toxoplasma gondii*



Overall Conclusions

- Our data are consistent with transmission cycles which bypass the cat
- Our data suggest that vertical transmission may be a possible explanation of the wide prevalence and ubiquity of this parasite

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