Study of *Toxoplasma gondii* and *Neospora caninum* in the Spanish Ibex (*Capra pyrenaica*) population of Sierra Nevada National Park (Andalucía, S.E. España)

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- **INTRODUCTION**
- MATERIAL AND METHODS
- RESULTS
- DISCUSSION
- CONCLUSIONS

(Billins, 2013; Hudson *et al.,* 2002; Karesh *et al.,* 2012)

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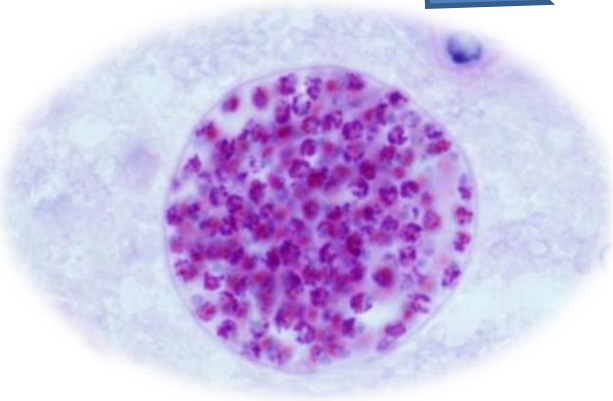
Epidemiological interactions

(Frenkel *et al.*, 1975; Afonso *et al.*, 2006; Morand, 2015)

Hosts

(Kutz *et al.*, 2014; Penczykowski *et al.*, 2015)

Pathogens



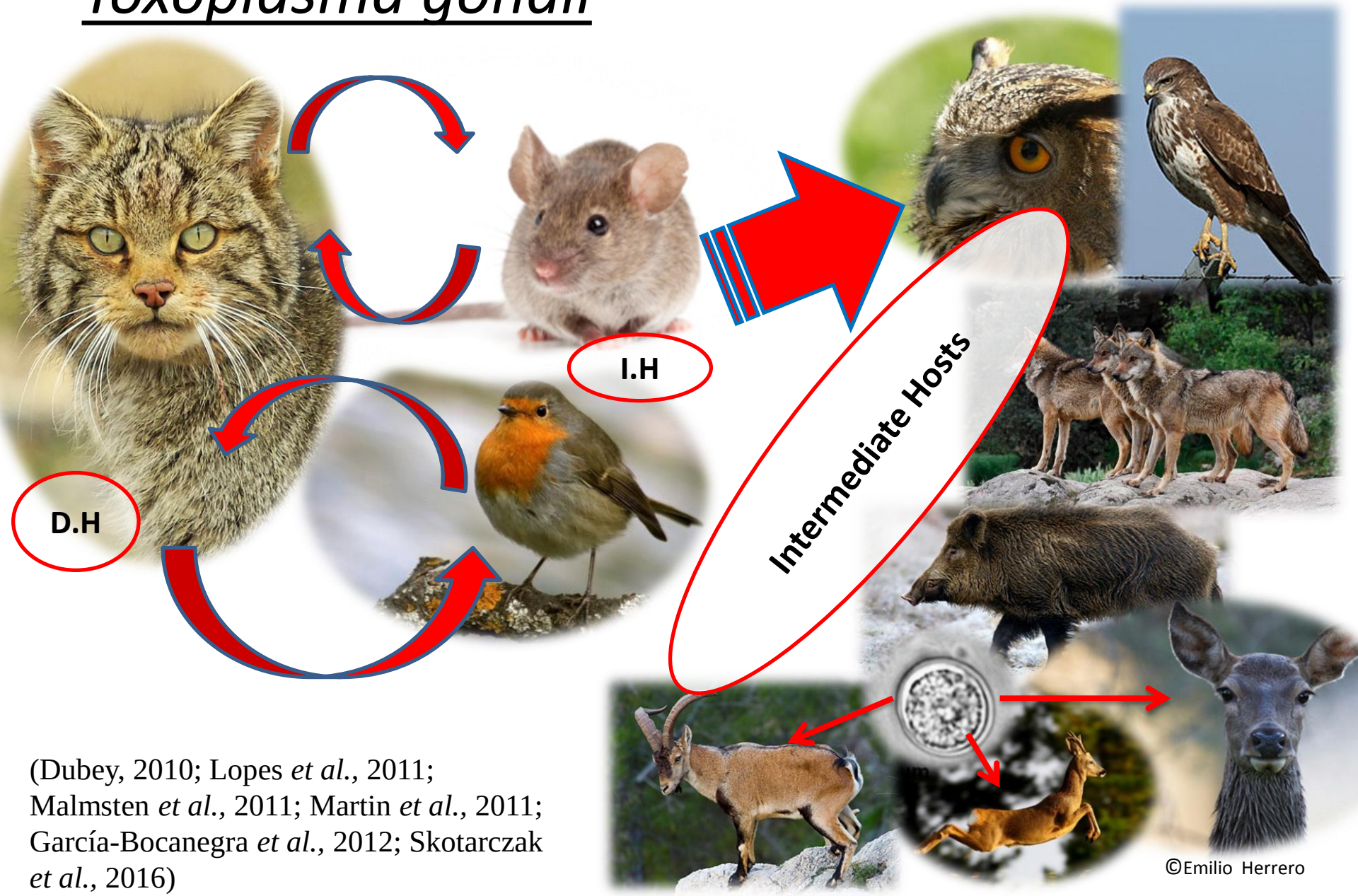
ILLNESS

Environment

SIERRA NEVADA GLOBAL WARMING OBSERVATORY

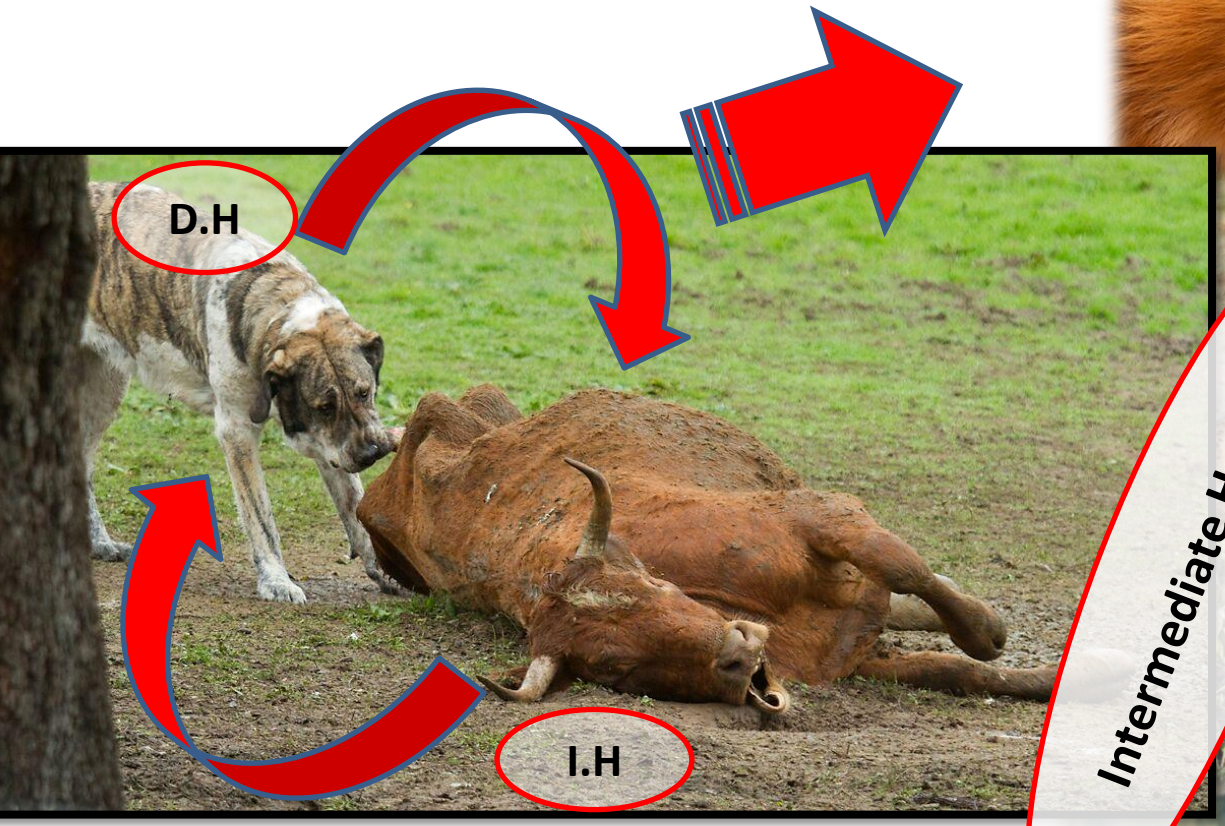
(Granados y Cano-Manuel, 2015)

Toxoplasma gondii



(Dubey, 2010; Lopes *et al.*, 2011; Malmsten *et al.*, 2011; Martin *et al.*, 2011; García-Bocanegra *et al.*, 2012; Skotarczak *et al.*, 2016)

Neospora caninum



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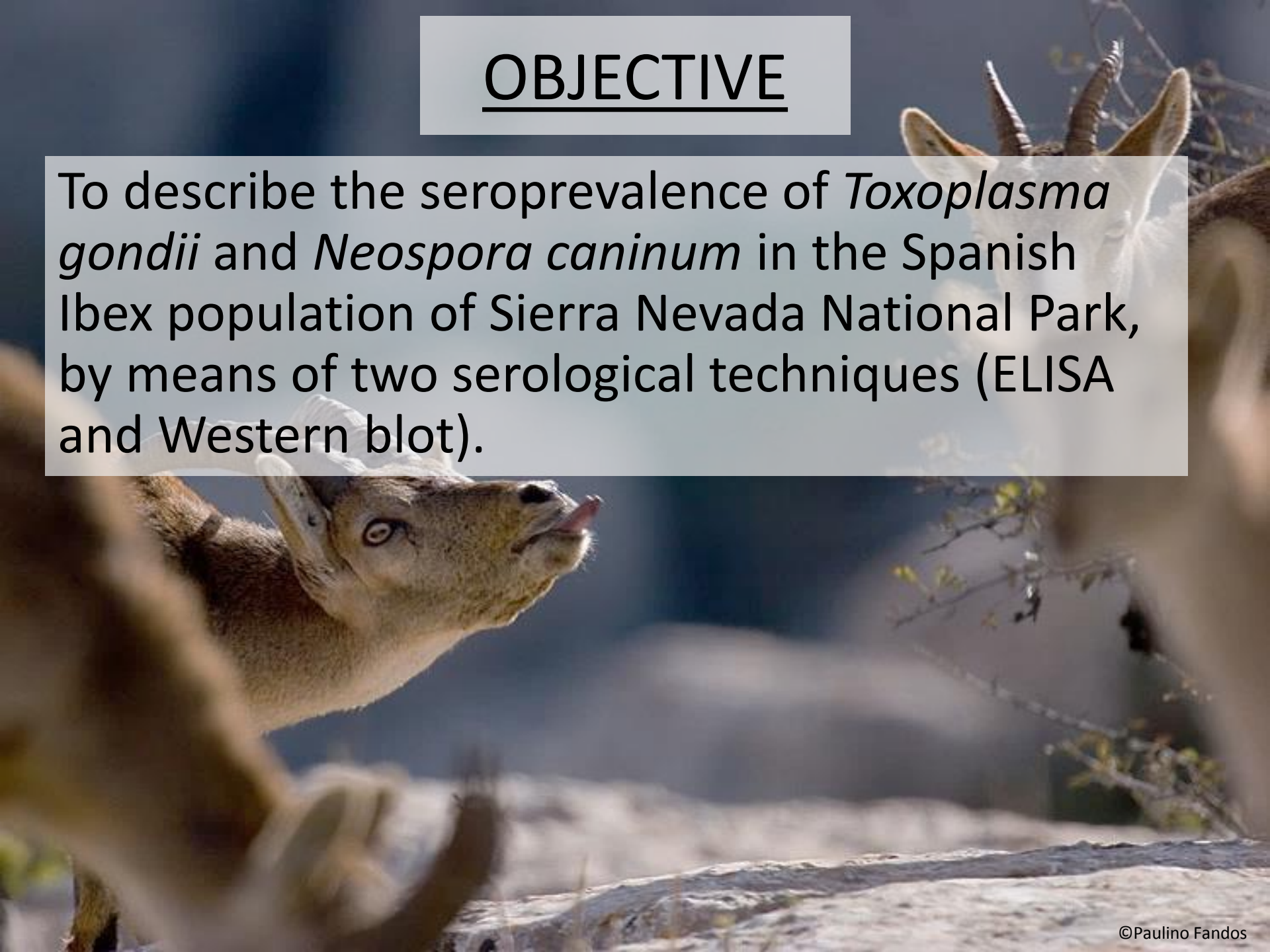
Intermediate Hosts

(Dubey y Lindsay, 1996; McAllister *et al.*, 1998; Dubey, 2003; Gondim *et al.*, 2004; Dubey *et al.*, 2007; García-Bocanegra *et al.*, 2012)

(Almería *et al.*, 2013)

OBJECTIVE

To describe the seroprevalence of *Toxoplasma gondii* and *Neospora caninum* in the Spanish Ibex population of Sierra Nevada National Park, by means of two serological techniques (ELISA and Western blot).



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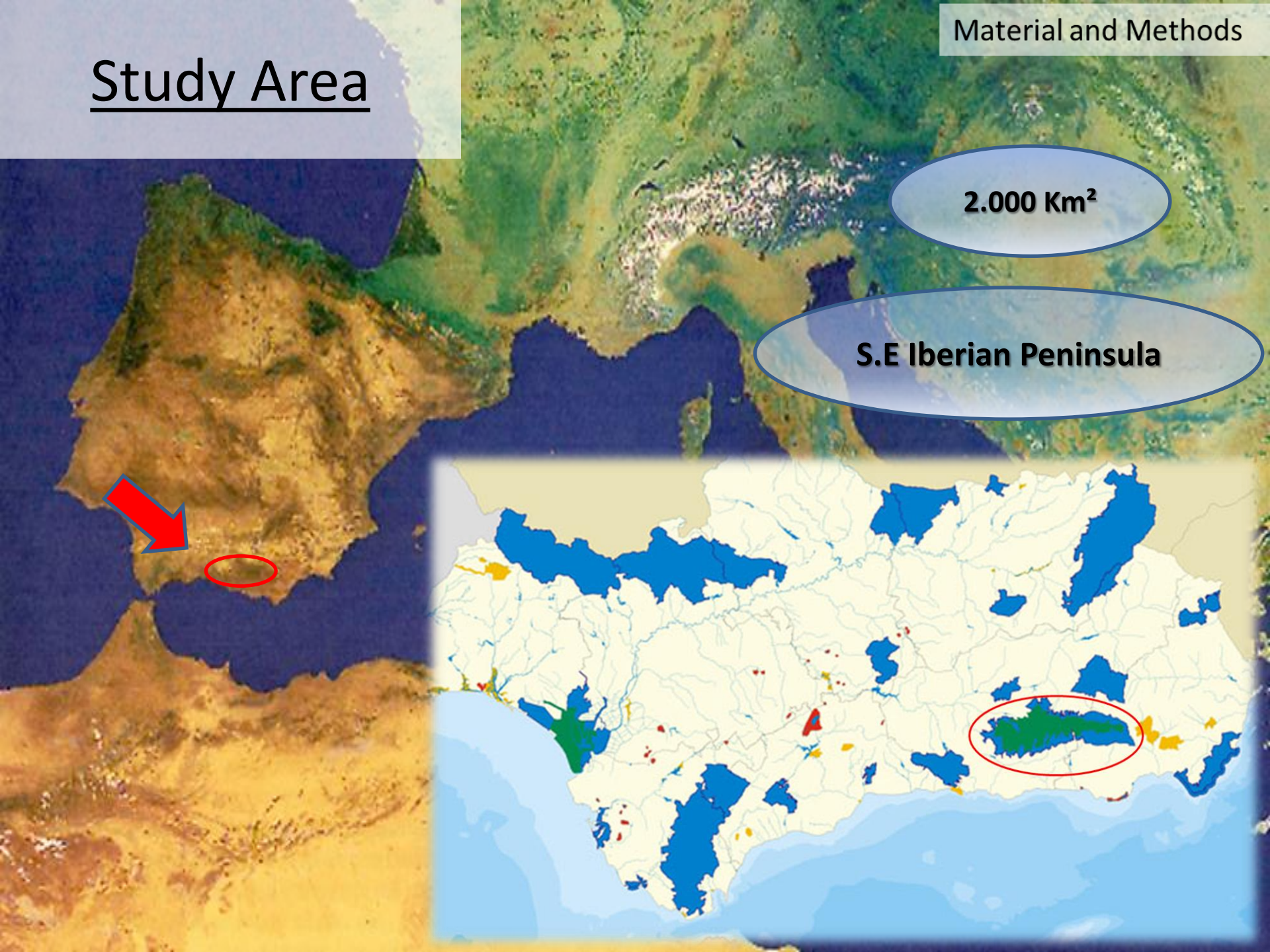
Study Area

2.000 Km²

S.E Iberian Peninsula



Sierra Nevada National Park

The largest endemism-area in the Iberian Peninsula

15.000 individuals (*Capra pyrenaica*)

(Pérez *et al.*, 2002; Aspizua *et al.*, 2010)

Studied Animals

(Casas *et al.*, 2011)

CAPTURED BETWEEN 2010-2015

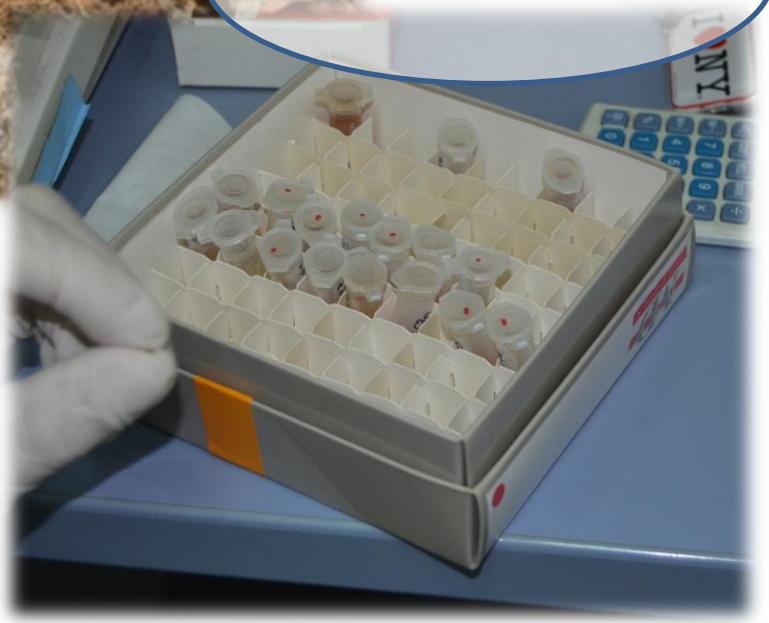
FREE-RANGING ANIMALS

ANESTHESIA

Studied Animals

BLOOD SAMPLING

LABORATORY



JUGULAR VENEPUNCTURE

CENTRIFUGED
COAGULATION (LABORATORY TEMPERATURE)

Studied Animals

n = 147

(Pérez, 2002)

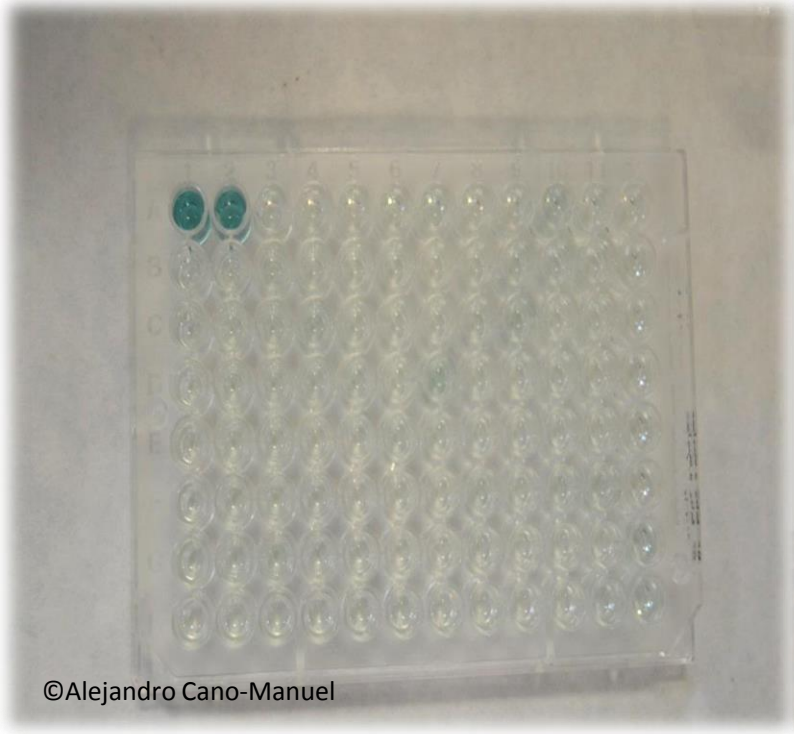
n: 147	CATEGORY	SAMPLE NUMBER	%
AGE	KIDS	5	3,4
	JUVENILES	18	12,2
	SUB-ADULTS	16	10,9
	ADULTS	93	63,3
	OLD	15	10,2
SEX	MALES	113	76,9
	FEMALES	34	23,1
CAPTURE ZONE	ALPUJARRA	27	18,3
	PONIENTE	118	80,3
	MARQUESADO	2	1,4

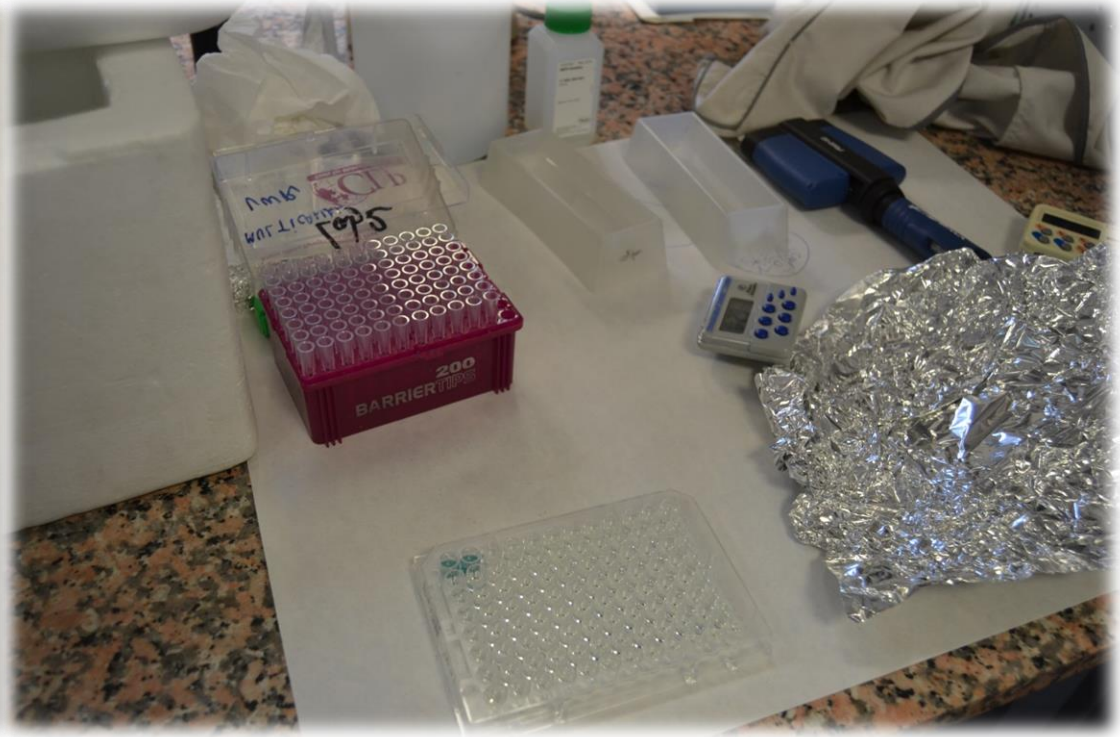
ELISA *in house*

Sonicated tachyzoites

- *Tg*-ME49 for *Toxoplasma gondii*.
- *Nc*-1 for *Neospora caninum*.

(Álvarez-García *et al.,* 2002; Chávez-Velásquez *et al.,* 2005; Gutiérrez-Expósito *et al.,* 2013).



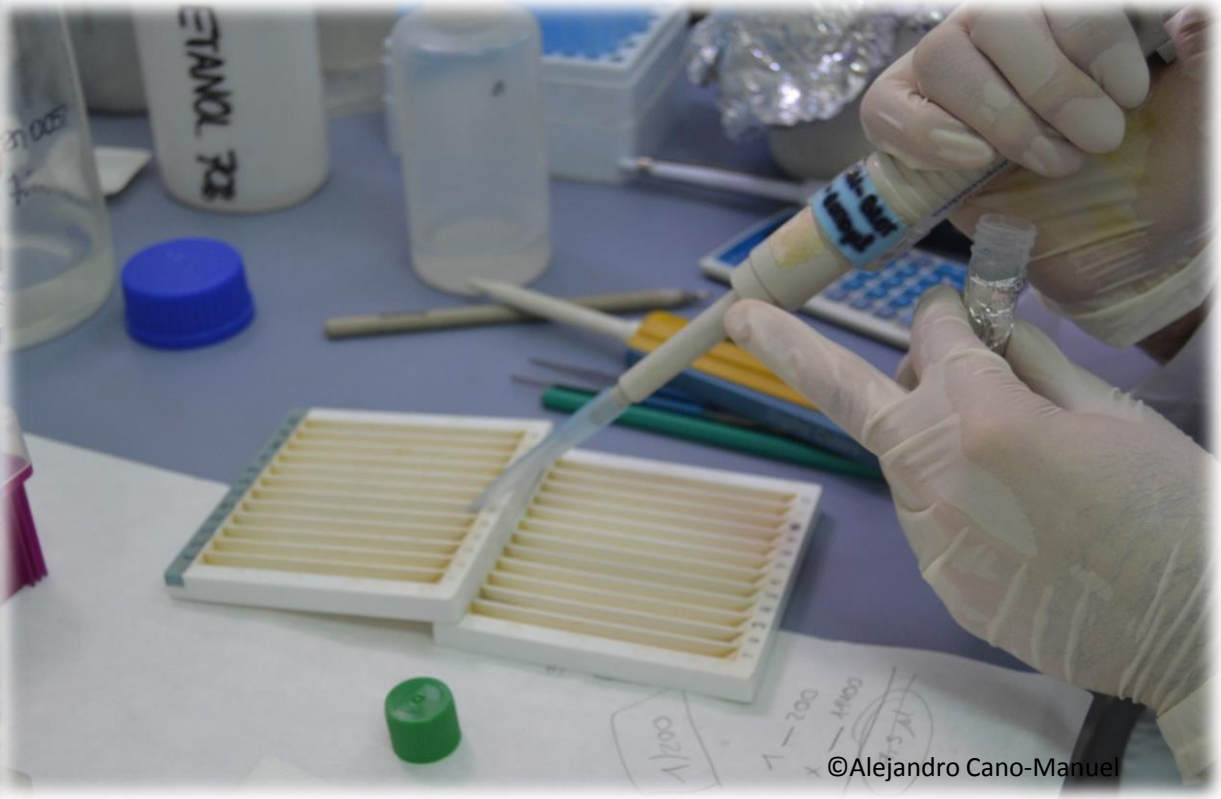


Western blot

Positive and/or doubtful samples

Control Samples	*T. gondii*	*N. caninum*
Domestic Goat	+ and - Control	+ and - Control
Sheep	+ and - Control	+ and - Control
Cow	NO	+ and - Control

(Álvarez-García *et al.*, 2002; Chávez-Velásquez *et al.*, 2005; Fernández-García *et al.*, 2009; Gutiérrez-Expósito *et al.*, 2013)

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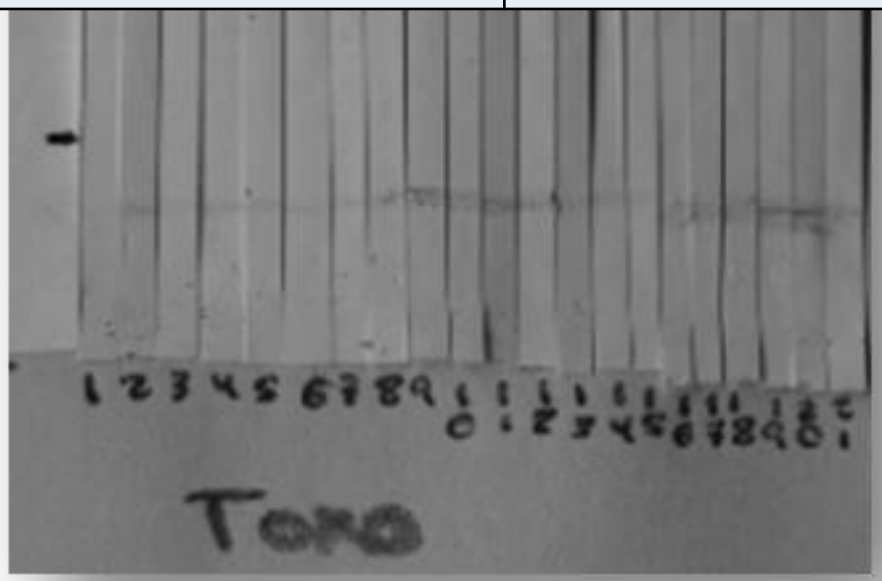
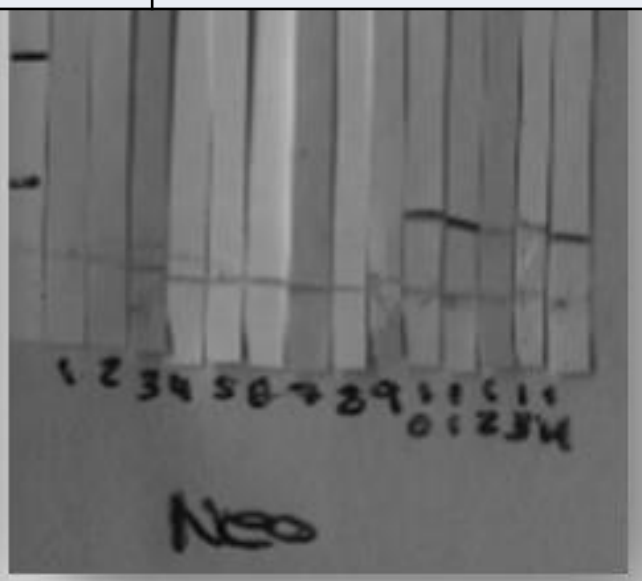
Results

ELISA	Positive	Prevalence (%)
Toxoplasma gondii	17	11,5
Neospora caninum	7	4,7





Results

T. gondii

Category	Nº
MALES	10
FEMALES	7
KIDS	1
JUVENILES	1
SUB-ADULTS	1
ADULTS	12
OLD	2
PONIENTE	9
ALPUJARRA	8
MARQUESADO	0

N. caninum

Category	Nº
MALES	3
FEMALES	4
KIDS	1
JUVENILES	1
SUB-ADULTS	0
ADULTS	5
OLD	0
PONIENTE	2
ALPUJARRA	5
MARQUESADO	0

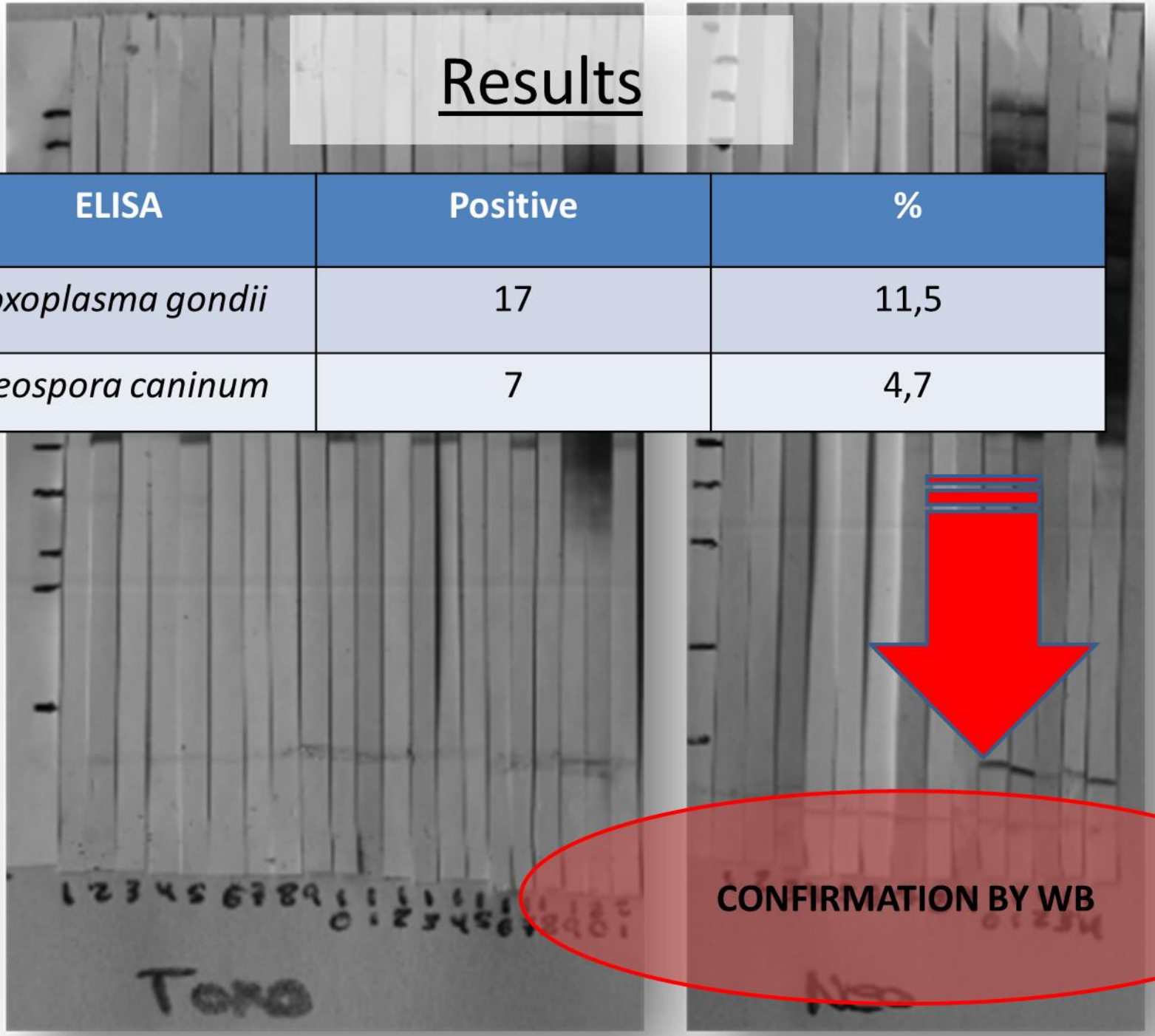
SEX

AGE

CAPTURE ZONE

Results

ELISA	Positive	%
Toxoplasma gondii	17	11,5
Neospora caninum	7	4,7



Results

ELISA	Positive	%
Toxoplasma gondii	17	11,5
Neospora caninum	7	4,7

WB	Positive	Prevalence (%)
Toxoplasma gondii	**0**	**0**
Neospora caninum	**0**	**0**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Hospedador	n	Técnica	Prevalencia %	Área	Autores
Capreolus capreolus	278	2	39,20	Península Ibérica	Gamarra *et al.* 2008
Odocoileus hemionus columbianus	42	2,3	0,00	Whasington State(USA)	Dubey *et al.* 2008
Odocoileus hemionus hemionus	43	2,3	32,55	Washington State (USA)	Dubey *et al.* 2008
Sus scrofa	148	2, 4	0,45	Champagne-Ardenne y Córcega (Francia)	Richomme *et al.* 2009
Capreolus capreolus	160	1,5	13,70	Galicia (España)	Panadero *et al.* 2010
Ammotragus lervia	91	2	1,49 (salvajes), 24,00 (cautivos)	Parque Regional Sierra Espuña (SERP) y Estación Experimental de Zonas Áridas (CSIC) (España)	Candela *et al.* 2009
Capreolus capreolus	20	1	52,00	Wallonia, Flandes, Bruselas (Bélgica)	De Craeye *et al.* 2010
Cervus elaphus	13	1	3,00	Wallonia, Flandes, Bruselas (Bélgica)	De Craeye *et al.* 2010
Capreolus capreolus	60	2	40,00	Francia	Aubert *et al.* 2010
Ovis gmelini	31	2	16,00	Francia	Aubert *et al.* 2010
Cervus elaphus	24	2	4,00	Francia	Aubert *et al.* 2010
Alces alces	417	5	20,00	Suecia	Malmsten *et al.* 2011
Capreolus capreolus	199	5	34,00	Suecia	Malmsten *et al.* 2011
Odocoileus virginianus	62 (M), 170 (I)	3, 4, 6, 7	32,25 (M), 53,52 (I)	Iowa y Minnesota (USA)	Dubey *et al.* 2009
Capra pyrenaica hispanica	531	2	27'50	Andalucía (España)	García-Bocanegra *et al.* 2012

TÉCNICA	ELISA	MAT	NAT	PCR	DAT	IFAT	Wb
Nº	1	2	3	4	5	6	7

Toxoplasma gondii Prevalence

Discussion

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< 40% in the Iberian Peninsula

Discussion

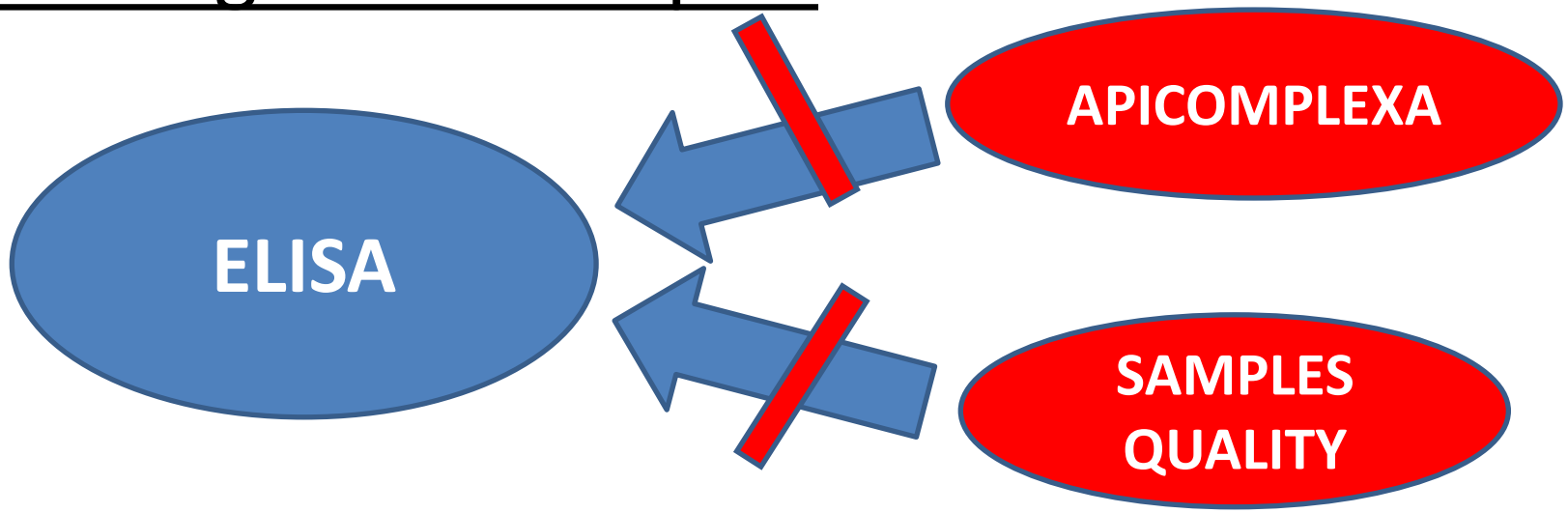
Hospedador	n	Técnica	Prevalencia %	Área	Autor
Odocoileus hemionus columbianus	42	2, 3	16,00	Whasington State (USA)	Dubey *et al.* 2008
Odocoileus hemionus hemionus	43	2, 3	18,60	Whasington State (USA)	Dubey *et al.* 2008
Capreolus capreolus	160	1, 5	6,80	Galicia (España)	Panadero *et al.* 2010
Capreolus capreolus	20	1	2,70	Wallonia, Flandes, Bruselas (Bélgica)	De Craeye *et al.* 2010
Alces alces	417	1	11,56	Suecia	Malmsten et al. 2011
Capreolus capreolus	199	1	1,00	Suecia	Malmsten *et al.* 2011
Odocoileus virginianus	62 (M), 170 (I)	3, 4, 6, 7	70,96 (M), 88,23 (I)	Iowa y Minnesota (USA)	Dubey *et al.* 2009
Capra pyrenaica hispanica	531	1, 6	5,10	Andalucía (España)	García-Bocanegra *et al.* 2012
Cervus elaphus	237	1, 6	11,80	Cataluña, Andalucía, Castilla-La Mancha (España)	Almería *et al.* 2007
Ammotragus lervia	13	1, 6	7,70	Sur y Centro de la Península Ibérica (España)	Almería *et al.* 2007
Capreolus capreolus	33	1, 6	6,10	Sur y Centro de la Península Ibérica (España)	Almería *et al.* 2007
Sus scrofa	298	1,6	0,30	Asturias, Andalucía, Castilla y León, Castilla-La Mancha, Cataluña (España)	Almería *et al.* 2007
Capreolus capreolus	66	1	7,60	Trentino, Alpes (Italia)	Bregoli *et al.* 2006
Cervus elaphus	125	1	3,20	Trentino, Alpes (Italia)	Bregoli *et al.* 2006
Rupicapra rupicapra	503	1	1,40	Trentino, Alpes (Italia)	Bregoli *et al.* 2006

TÉCNICA	ELISA	MAT	NAT	PCR	DAT	IFAT	Wb
Nº	1	2	3	4	5	6	7

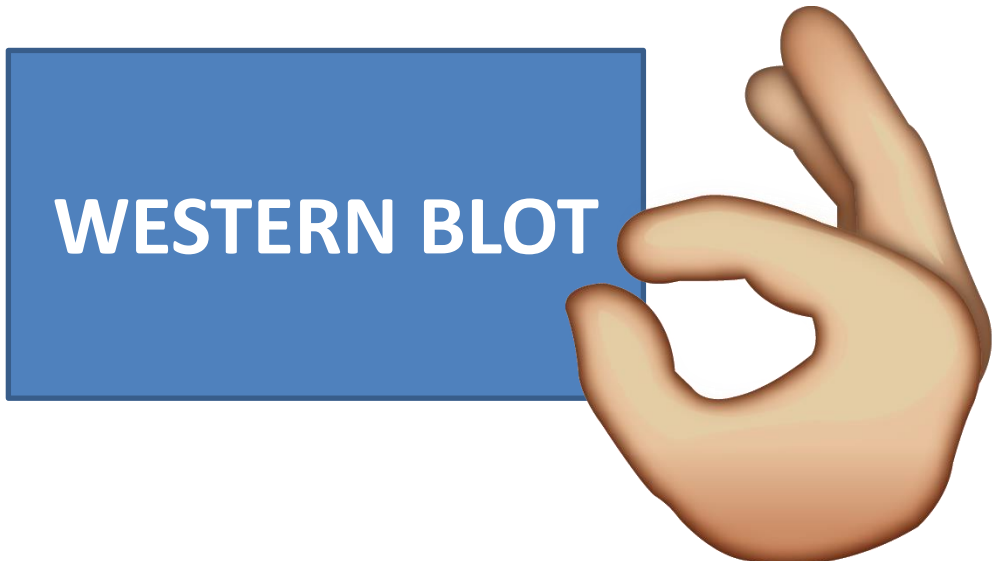
Neospora caninum Prevalence

< 12% in the Iberian Peninsula

Serological techniques



TÉCNICA	ELISA	MAT	NAT	PCR	DAT	IFAT	Wb
Nº	1	2	3	4	5	6	7



(Söndgen *et al.*, 2001; Chávez-Velásquez *et al.*, 2004; Chávez-Velásquez *et al.*, 2005; Gutiérrez-Expósito *et al.*, 2012; Nasir et al., 2012; Gutiérrez-Expósito *et al.*, 2013; Gazzonis *et al.*, 2014)

Carnivores in Sierra Nevada

- Fox (*Vulpes vulpes*)
- Feral dogs (*Canis lupus familiaris*)

(Dubey *et al.*, 2007; Marco *et al.*, 2008; Barea-Azcón, 2012)

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N. caninum

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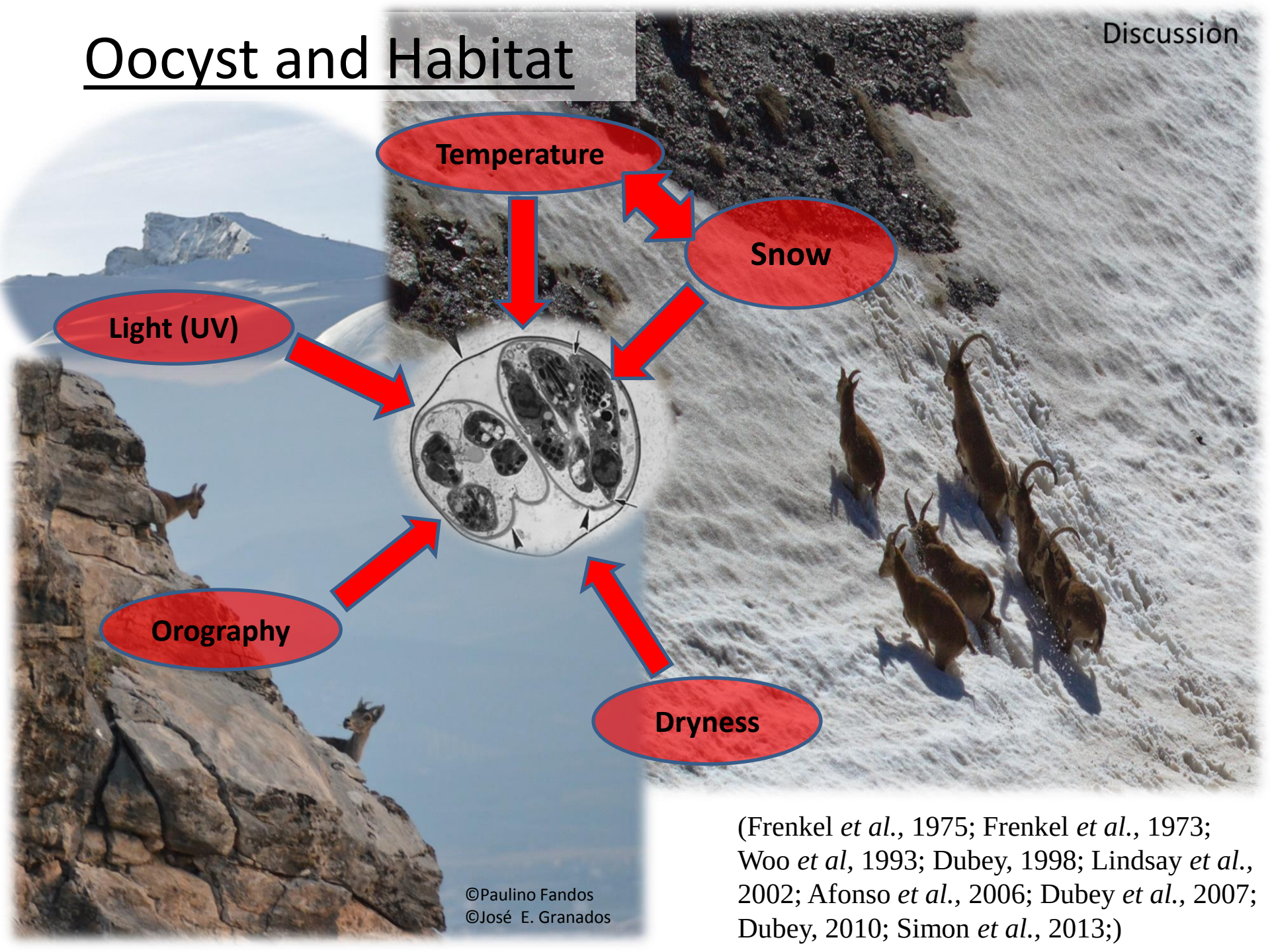
T. gondii

- Wildcat (*Felis silvestris silvestris*)
- Domestic cat (*Felis catus*)

(Moleón y Gil-Sánchez, 2003; Barea-Azcón, 2012; Gil-Sánchez *et al.*, 2015)

Oocyst and Habitat



(Frenkel *et al.*, 1975; Frenkel *et al.*, 1973; Woo *et al*, 1993; Dubey, 1998; Lindsay *et al.*, 2002; Afonso *et al.*, 2006; Dubey *et al.*, 2007; Dubey, 2010; Simon *et al.*, 2013;)

“Hypoendemic island”

Areas with a reduced presence of infectious pathogens due to the bioclimatic characteristics.

(Vaumourin *et al.*, 2015)

T. gondii oocyst

N. caninum
muscular cyst

(Méthot, 2012; Vander Wal *et al.*, 2014)

COINFECTION

Sarcoptes scabiei

(Pérez *et al.*, 1997; Alasaad *et al.*, 2008; Sarasa *et al.*, 2011; Carvalho *et al.*, 2015)

(Vander Wal *et al.*, 2014; Penczykowski *et al.*, 2015)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First

The seroepidemiological study of *Toxoplasma gondii* and *Neospora caninum* in the Spanish Ibex (*Capra pyrenaica*) based on the results obtained by non-validated techniques, requires the Western blot technique to confirmthe positive samples, in order to ensure that the epidemiological conclusions are properly supported.

El estudio epidemiológico de *Toxoplasma gondii* y *Neospora caninum* en la cabra montés (*Capra pyrenaica*) basado en los resultados mediante técnicas serológicas no validadas, requiere la confirmación de casos dudosos y positivos mediante WB, con el fin de que las conclusiones epidemiológicas estén debidamente sustentadas.

Second

The absence of seropositive individuals indicates that Sierra Nevada Park is an hypoendemic area for these protozoan parasites. The habitat of the Spanish Ibex is probably a determining factor that makes difficult the natural nesting of *T. gondii* and *N. caninum*, favoring that these mountainous areas are "hypoendemic islands" for both parasites from an epidemiological point of view.

La ausencia de individuos seropositivos indica que el Espacio Natural de Sierra Nevada es una zona hipoendémica para estos parásitos. El hábitat que ocupa la cabra montés posiblemente sea determinante en la dificultad para el anidamiento natural de *T. gondii* y *N. caninum*, propiciando que estas áreas montañosas sean “islas hipoendémicas” desde un punto de vista epidemiológico para ambos parásitos.


THANK YOU
GRACIAS
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