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ZOOPROFILATTICO
SPERIMENTALE
DELL'ABRUZZO
E DEL MOLISE
"G. CAPORALE"

European Vectors of Bluetongue: Insights from Italy



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dell' Abruzzo e del Molise

Phylum

ARTHROPODA

Class

INSECTA

Order

DIPTERA

Suborder

NEMATOCERA

Family

CULICINAE
CHIRONOMIDAE
CERATOPOGONIDAE
TIPULIDAE



Genus: **CULICOIDES**

Culicoides: morphological features



- Stout body
- Humped torax
- Wing pattern



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Culicoides' wings

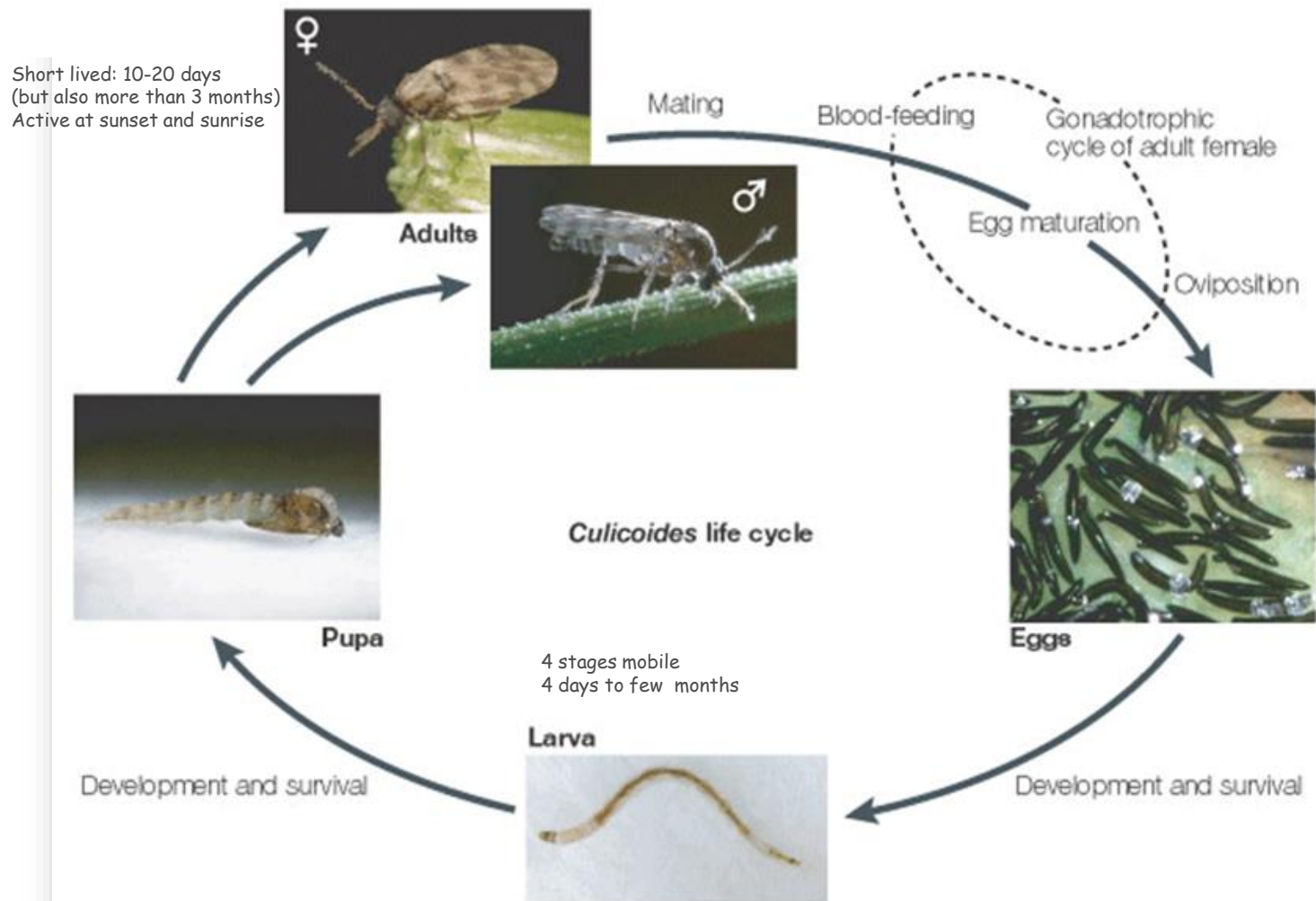


The whole process
is dependent on
temperature

Only adult females
do blood meal

Credits: Purse *et al.*, 2005

Culicoides life cycle



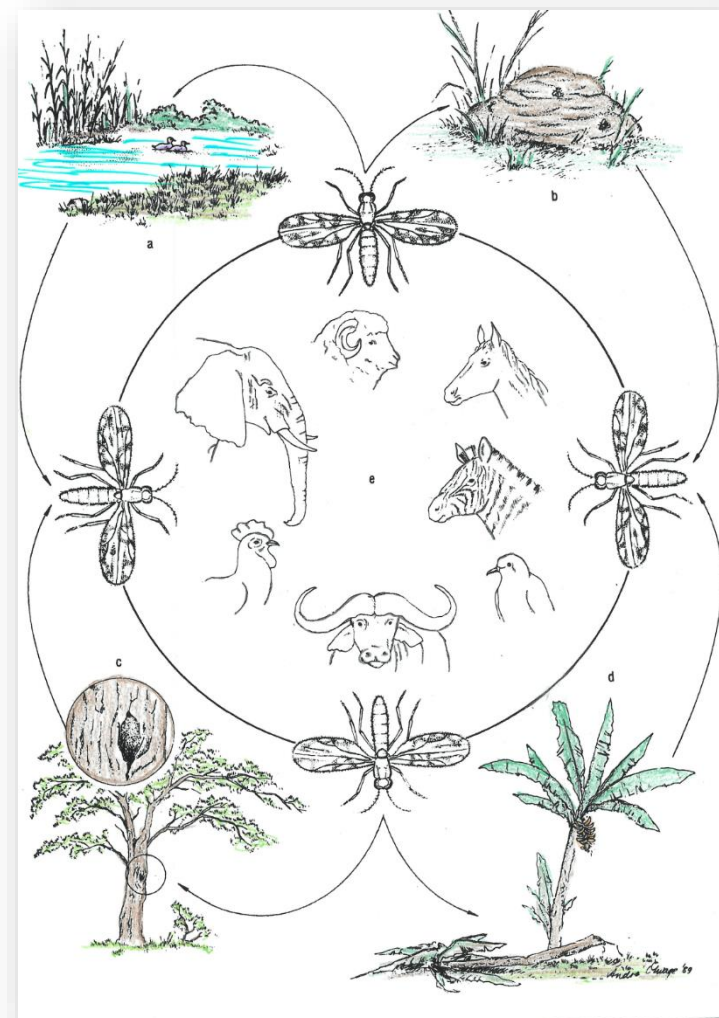


Organic matter and humidity



The distribution of a species depends on its breeding site

Culicoides breeding sites



Meiswinkel R. et al., 2004

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Culicoides: general features

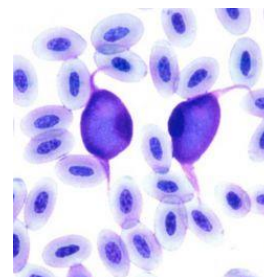


✓ 1 - 3 mm



✓ More than 1000 species

- ✓ Transmission of
 - allergic dermatitis (sweet itch)
 - protozoa
 - nematodes






> 75 ArBoviruses have been isolated
from Culicoides





Culicoides: general features

REOVIRIDAE - Sedoreoviridae

-  - Bluetongue virus
- African Horse sickness virus 
- Equine Encephalosis virus
-  - Epizootic Haemorrhagic Disease virus

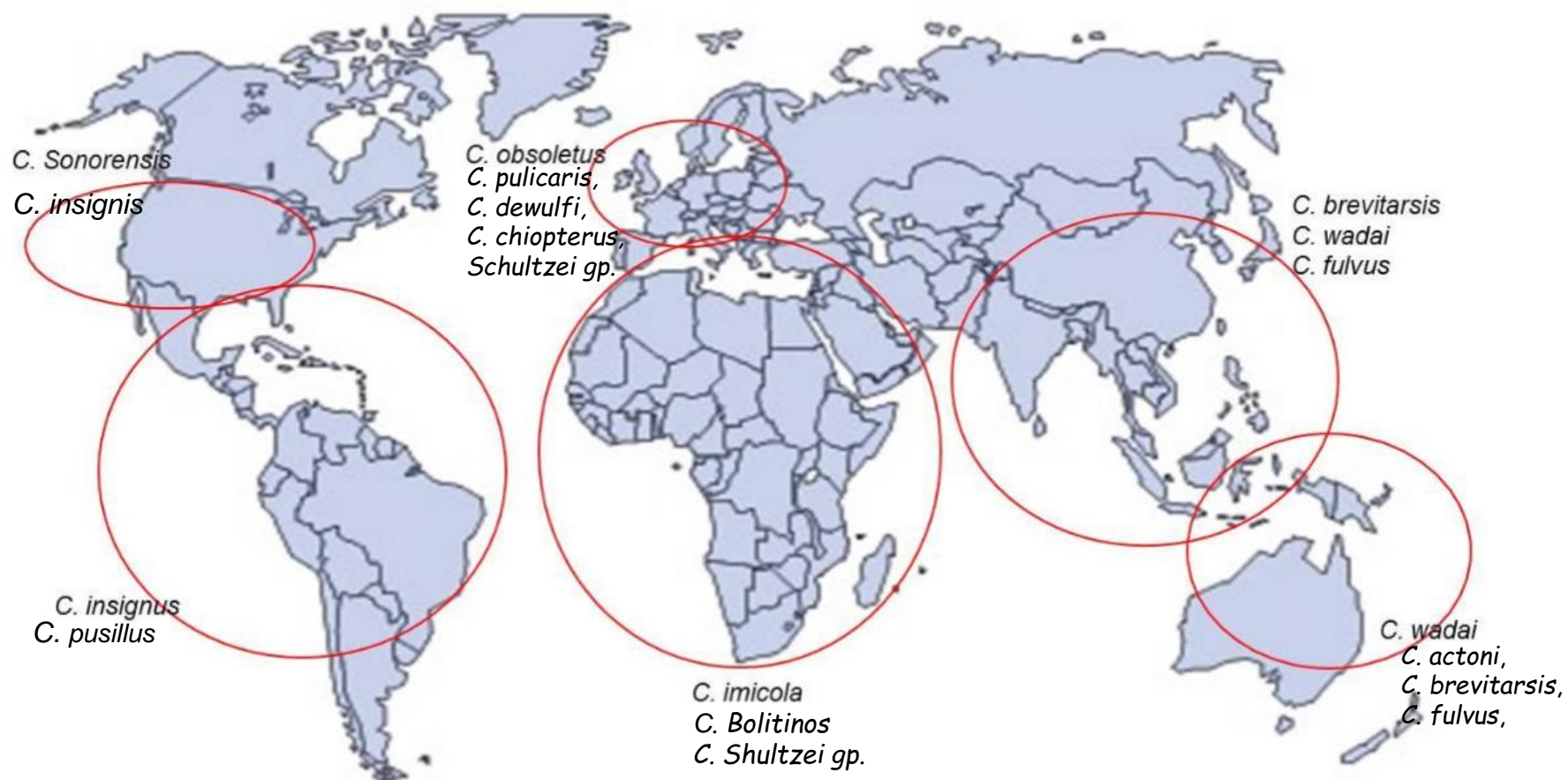
BUNYAVIRIDAE

- Akabane virus
-  - Schmallenberg virus
-  - Oropouche virus

RABDOVIRIDAE

- Bovine Ephemeral Fever virus

Worldwide distribution of the major *Culicoides* vectors.

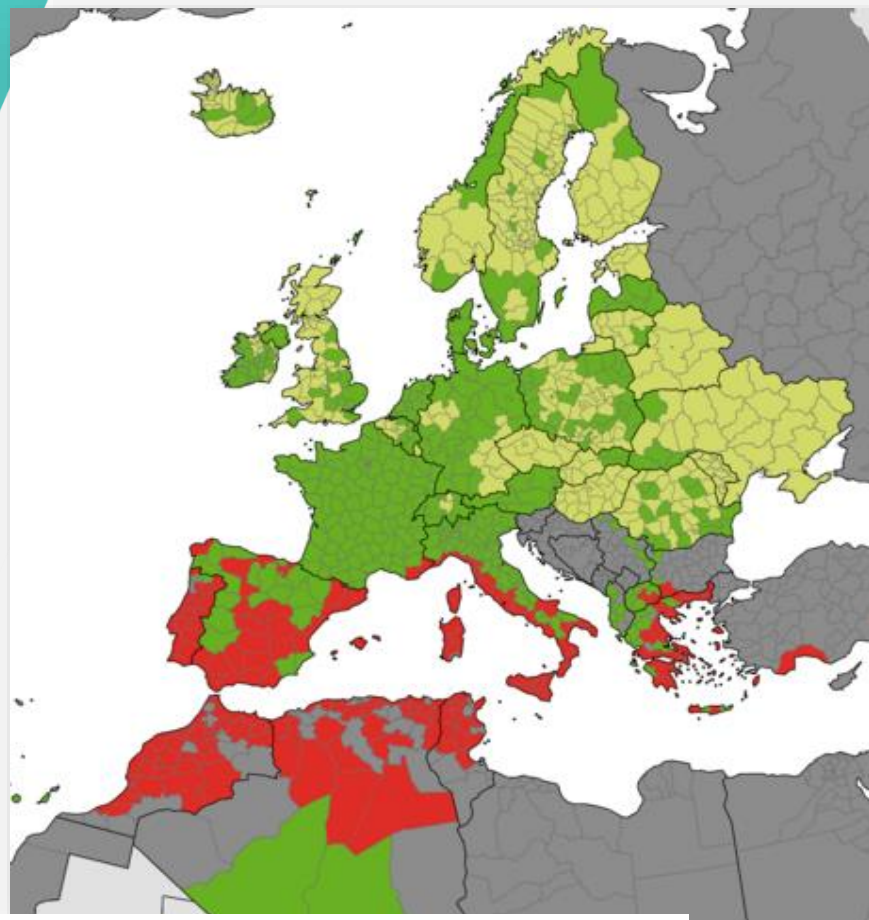


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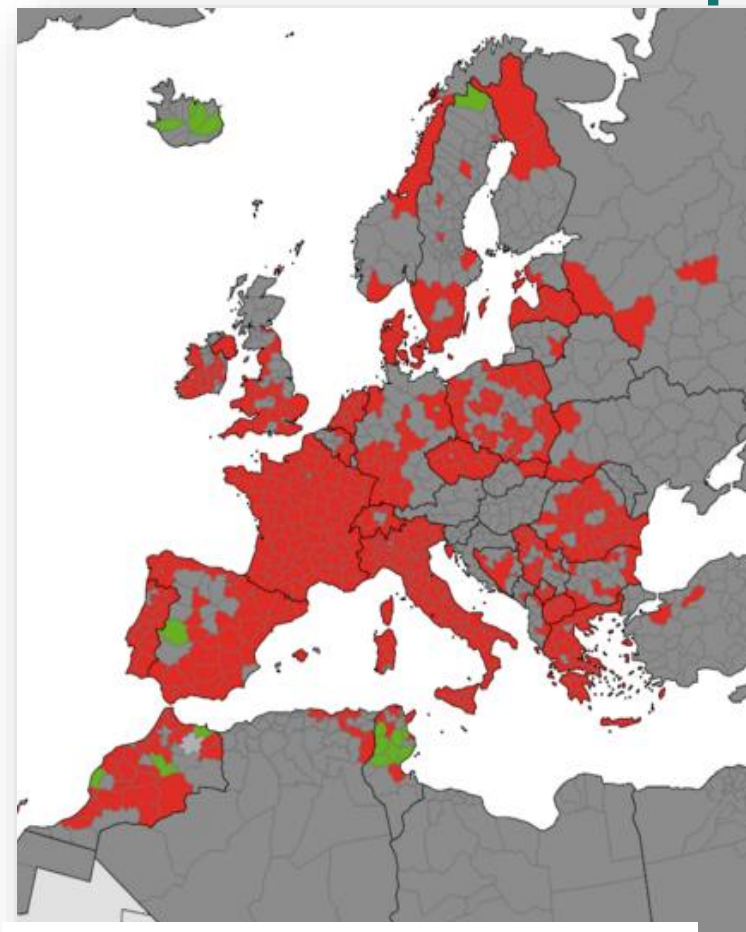
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Culicoides imicola

In Europe...



Culicoides obsoletus/scoticus

European Centre for Disease Prevention and Control and European Food Safety Authority. Biting-midge maps [internet]. Stockholm: ECDC; 2023. Available from: <https://ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/biting-midge-maps>

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Subgenus Avaritia ***Culicoides imicola***

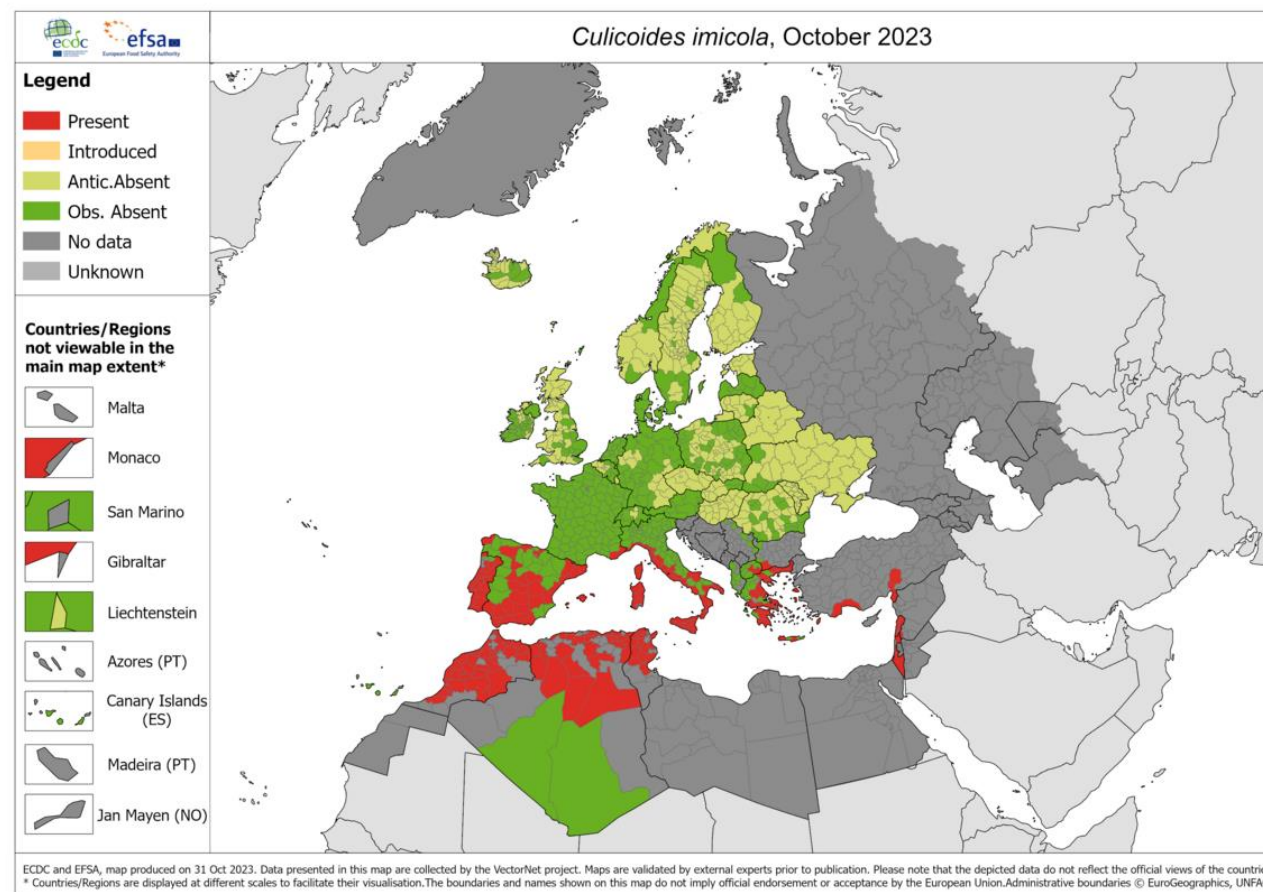


**Breeding sites: muddy environments
rich in organic matter, even artificial
man-made, sunny, with low vegetation**





Subgenus *Avaritia* *Culicoides imicola*



European Centre for Disease Prevention and Control and European Food Safety Authority. Biting-midge maps [internet]. Stockholm: ECDC; 2023. Available from: <https://ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/biting-midge-maps>



- They can live for more than three months.
- Active also during the day.
- Active even inside farms.
- Adults are active even in winter, with low temperatures.

Subgenus *Avaritia* *Obsoletus complex*

- ✓ *Cryptic species: Culicoides obsoletus s.s.*
Culicoides scoticus
Culicoides montanus

PCR



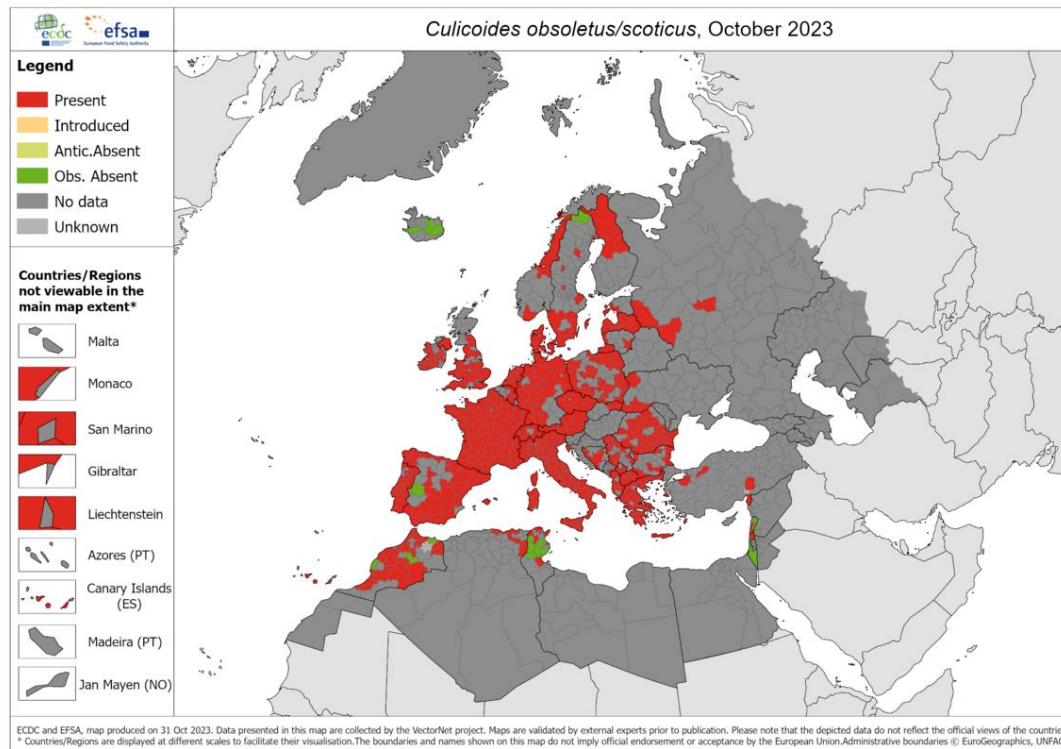
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Subgenus *Avaritia* *Obsoletus complex*

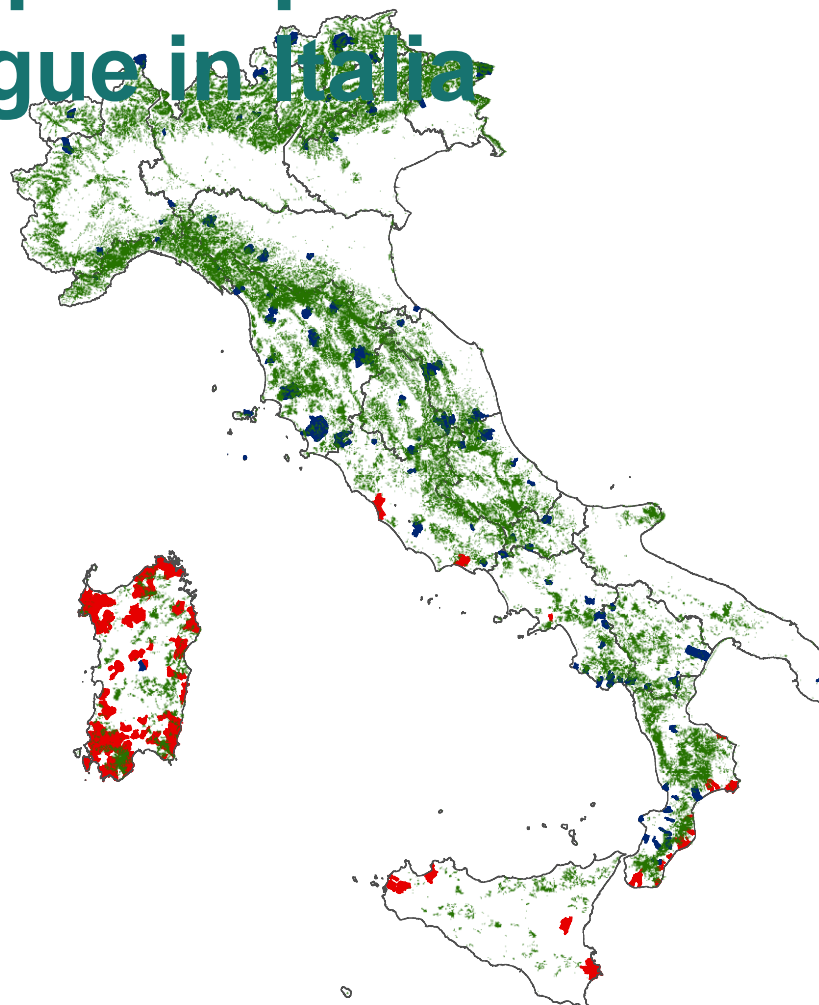


European Centre for Disease Prevention and Control and European Food Safety Authority. Biting-midge maps [internet]. Stockholm: ECDC; 2023. Available from: <https://ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/biting-midge-maps>

Obsoletus complex e *C. imicola* sono i vettori più importanti della Bluetongue in Italia

■ *C. imicola*
100 largest collections

■ Obsoletus Complex
100 largest collections

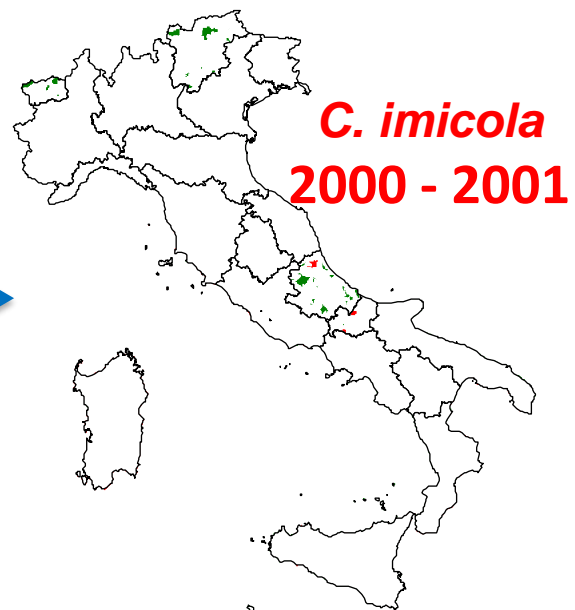
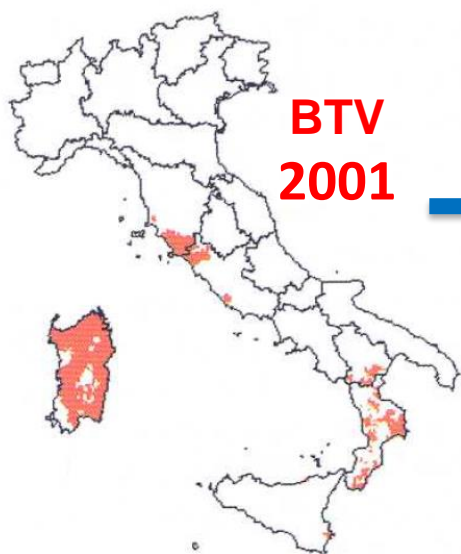
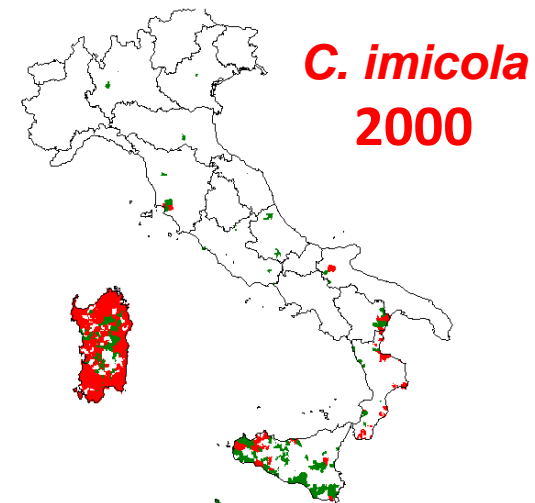
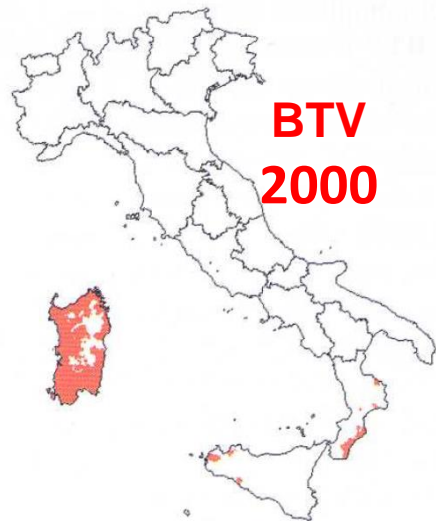


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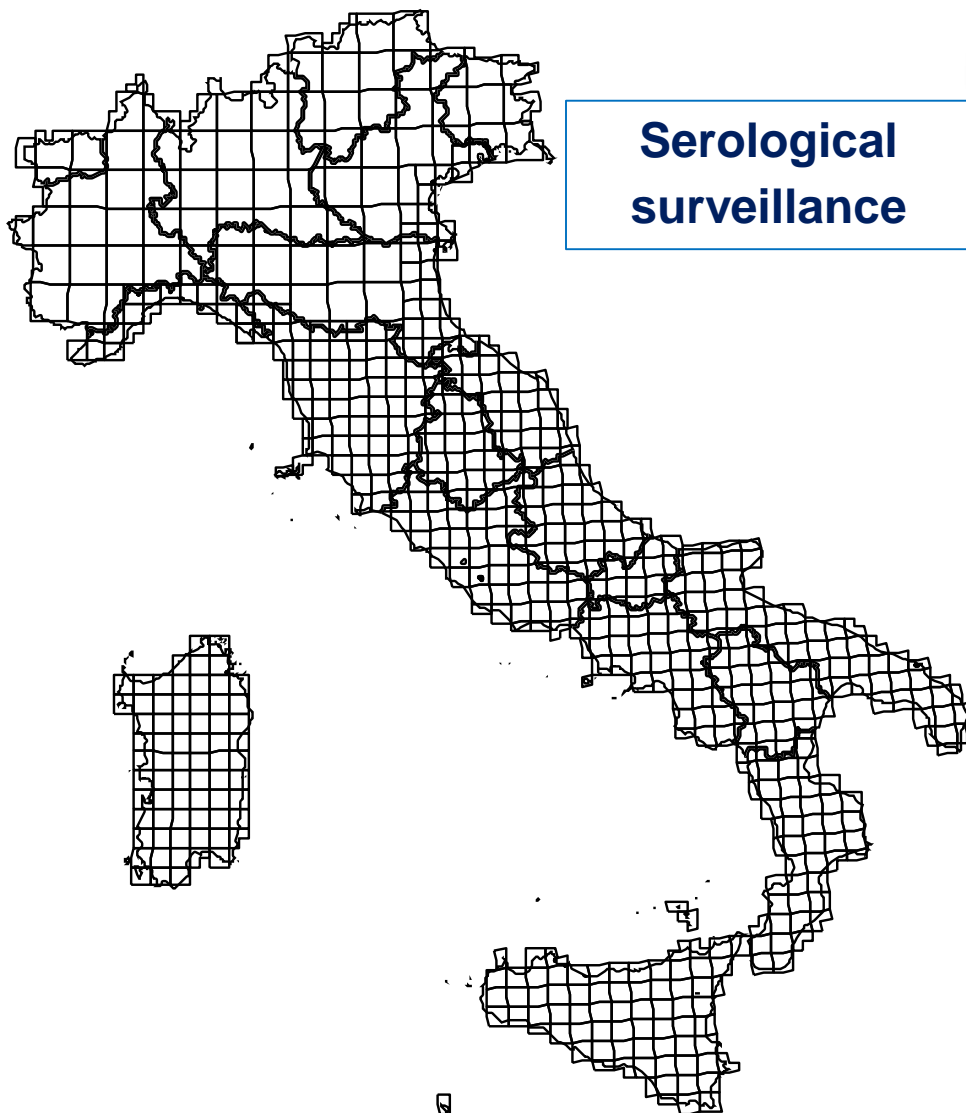
SNAPSHOT



5,000 collections

1,247 municipalities

According to *C. imicola* distribution, high and low risk areas were defined in 2001



Serological
surveillance



Entomological
surveillance

Permanent traps:

1 per 1600km²

Weekly collections

Mobile traps:

In outbreaks,
seroconversions, etc...

2001: surveillance was extended to all the Italian regions

AIMS OF SURVEILLANCE:



- To continuously refine the distribution and abundance maps of the main vector *C. imicola* in Italy.
- To evaluate the role of other vector species
- To detect BT virus in *Culicoides* midges during outbreaks/virus circulation.
- To identify seasonally free zones.

These aims can be achieved only with data that are both comparable and sensitive.

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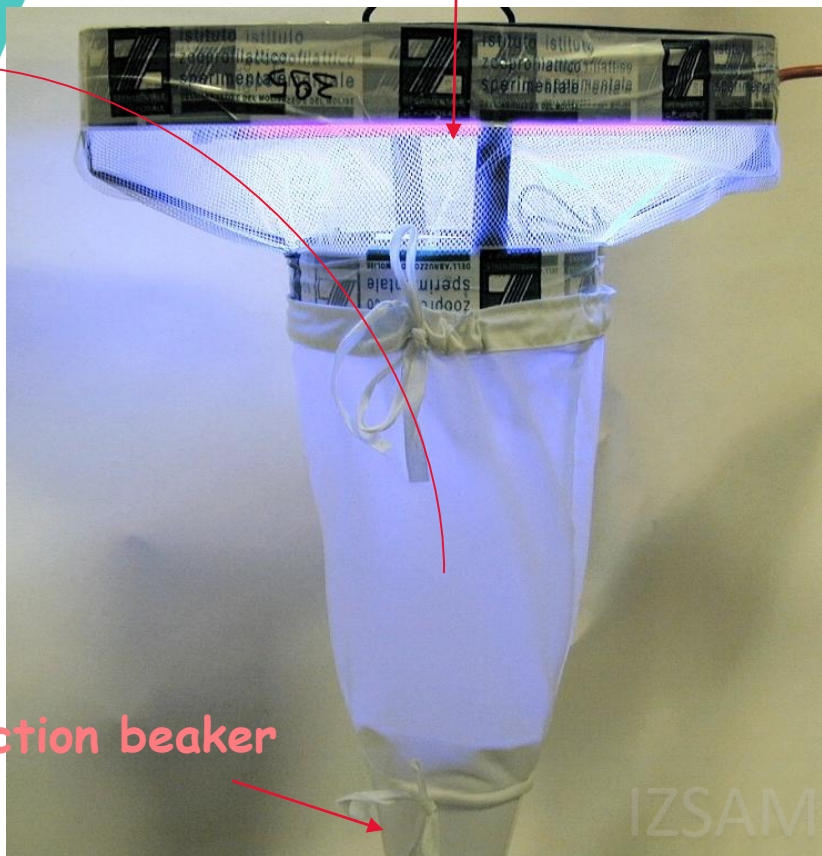
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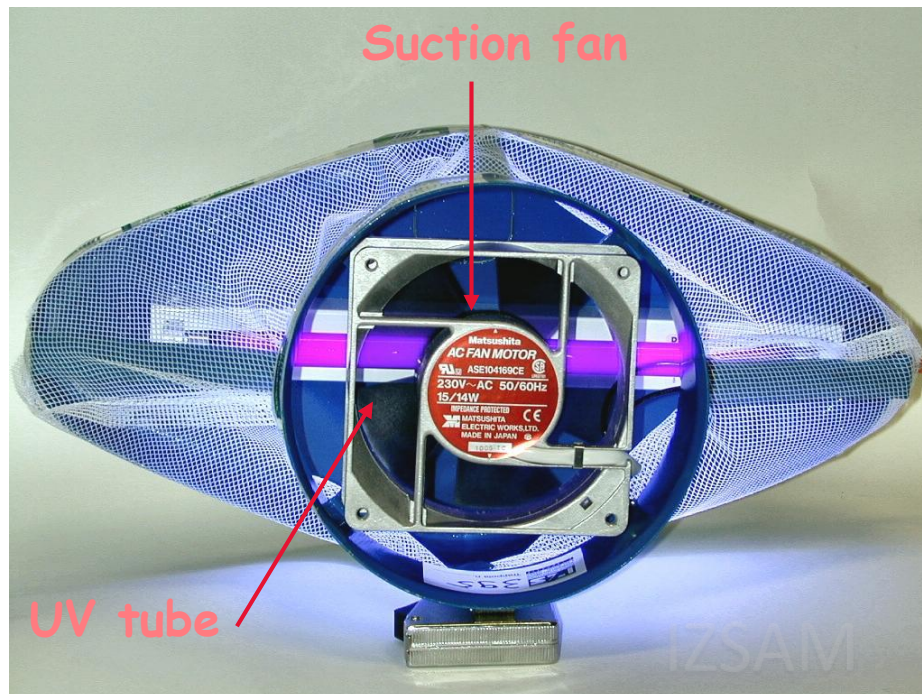
Collection beaker

Net



BLACK LIGHT TRAPS

Suction fan



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CHOICE OF TRAPPING SITES



- Cattle, sheep, goats
- An higher number of animals
- Shelters not completely closed



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CHOICE OF TRAPPING SITES



- Proximity to animals where they rest at dusk and during the night
- Available electricity

CATCHING PROCEDURE



160 cm from the ground

Solution in collection beaker:

- Water + liquid soap
- PBS + antibiotics + soap

At least 1 hour before sunset...

The next early morning...

Filter the collection and dip the gauze, with insects inside, in one container with:

- 70% ethanol and store at room temperature but in a cool place, avoiding direct sunlight
- PBS with antibiotics (if the aim is virus isolation), store at +4°C and analyse within few days



Btv vectors...2000

- *C. imicola*
- *C. obsoletus/C. scoticus* ??
- *C. pulicaris* ??



C. pulicaris



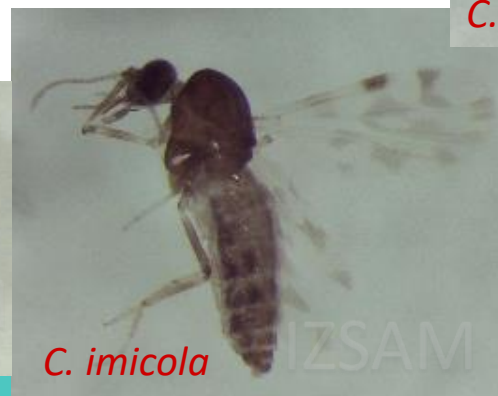
C. obsoletus/C. scoticus



C. imicola

BTV potential vector species...2025

- *C. imicola* (PCR + isolamento)
- **Obsoletus complex** (PCR + isolamento)
 - *C. obsoletus* (PCR)
 - *C. scoticus* (PCR)
 - *C. montanus* (PCR)
- *C. pulicaris* (PCR + isolamento)
- *C. lupicaris* (PCR)
- *C. newsteadi* (PCR)
- *C. punctatus* (PCR)
- *C. dewulfi* (PCR)
- *C. chiopterus* (PCR)



Vector species of Culicoides midges implicated in the 2012-2014 Bluetongue epidemics in Italy

Maria Goffredo*, Monica Catalani, Valentina Federici, Ottavio Portanti, Valeria Marini, Giuseppe Mancini, Michela Quaglia, Adriana Santilli, Liana Teodori & Giovanni Savini

Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise 'G. Caporale', Campo Boario, 64100 Teramo, Italy.

* Corresponding author at: Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise 'G. Caporale', Campo Boario, 64100 Teramo, Italy.

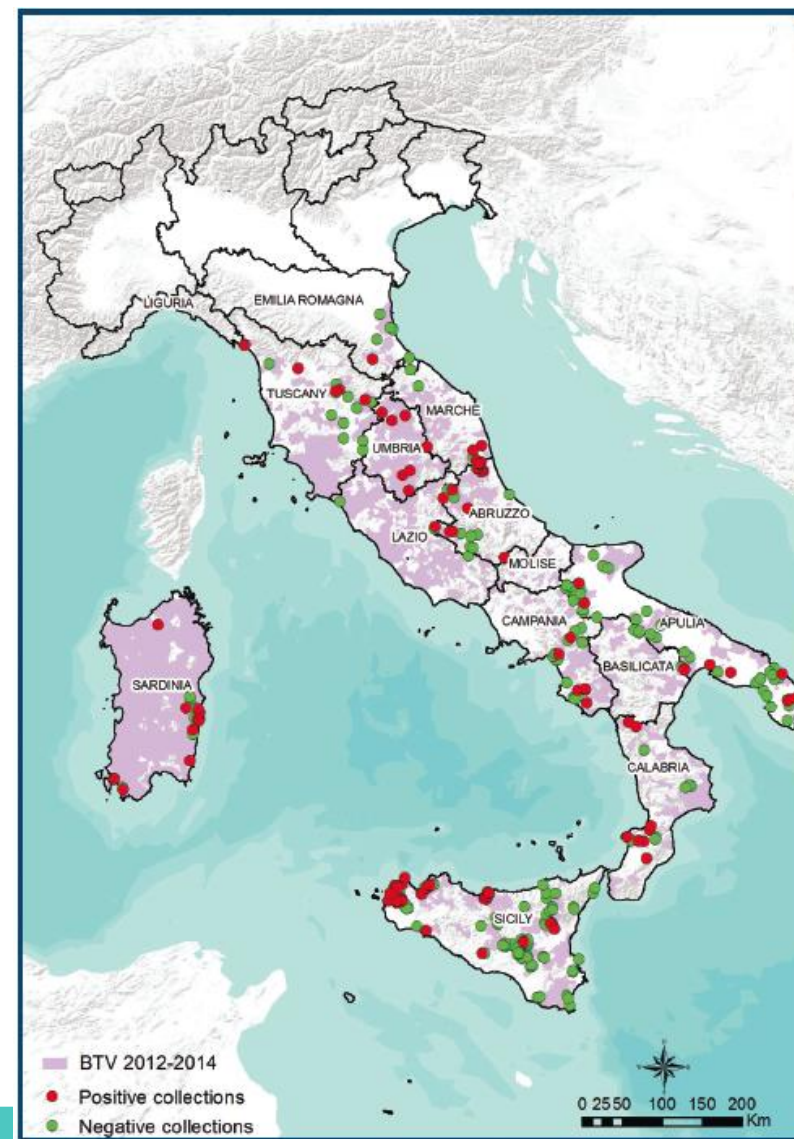
Tel.: + 39 0861 332416, Fax: +39 0861 332251, e-mail: m.goffredo@izs.it.

Veterinaria Italiana 2015, **51** (2), 131-138. doi: 10.12834/VetIt.771.3854.1

Accepted: 06.06.2015 | Available on line: 30.06.2015

BTV affected farms: 339
Total of collections: 651

BTV IN ITALY 2012-2014



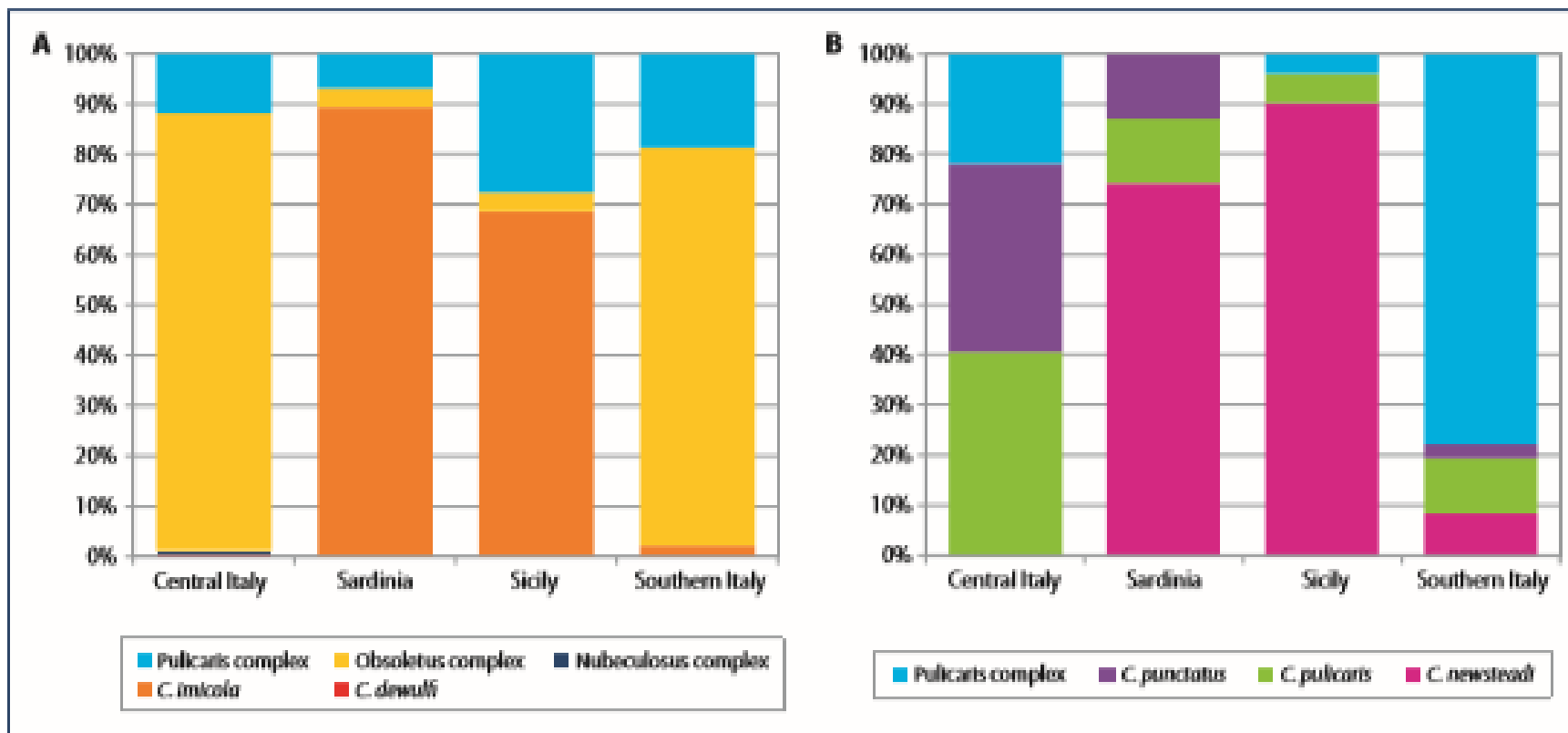
POOLS

Figure 3. Species composition of the BTV tested (n = 2,925; A and B) and positive (n = 1,107; C and D) pools in Sardinia, Sicily, Southern (Calabria, Apulia, Campania regions) and Central Italy (Abruzzo, Marche, Umbria, Tuscany, southern parts of Emilia Romagna and Liguria regions). The species of the *Pulicaris* complex are shown as complex (A and C) and in detail (B and D).



83,102 *Culicoides*
2,925 tested pools
1,107 positive pools

Species composition of BTV positive pools





Ministero della Salute

DIPARTIMENTO DELLA SALUTE UMANA, DELLA SALUTE
ANIMALE E DELL'ECOSISTEMA (ONE HEALTH), E DEI RAPPORTI
INTERNAZIONALI
EX DIREZIONE GENERALE DELLA SANITA' ANIMALE E
DEI FARMACI VETERINARI

Ufficio 3

Sanità animale e gestione operativa del Centro nazionale di lotta ed
emergenza contro le malattie animali e unità centrale di crisi

Registro - Classif. P-I.1.a.e/2024/1

Regioni e P.A.
Assessorati sanità
Servizi veterinari

II.ZZ.SS.

e, p.c.:
Rep. di San Marino – UOS San. Vet. e I.A.
servizio.veterinario@iss.sm
antonio.putti@iss.sm

CESME c/o IZSAM

Comando Carabinieri per la Tutela della Salute

Associazioni di categoria
bovini, bufalini, ovini e caprini

Organizzazioni dei veterinari

Uff. 8 Ex DGSAF
Uff. 3 Ex SEGGEN

Oggetto: Febbre catarrale degli ovini (Bluetongue) – orientamenti sulle misure di controllo e di gestione sul territorio nazionale e sulle attività di sorveglianza sierologica ed entomologica.



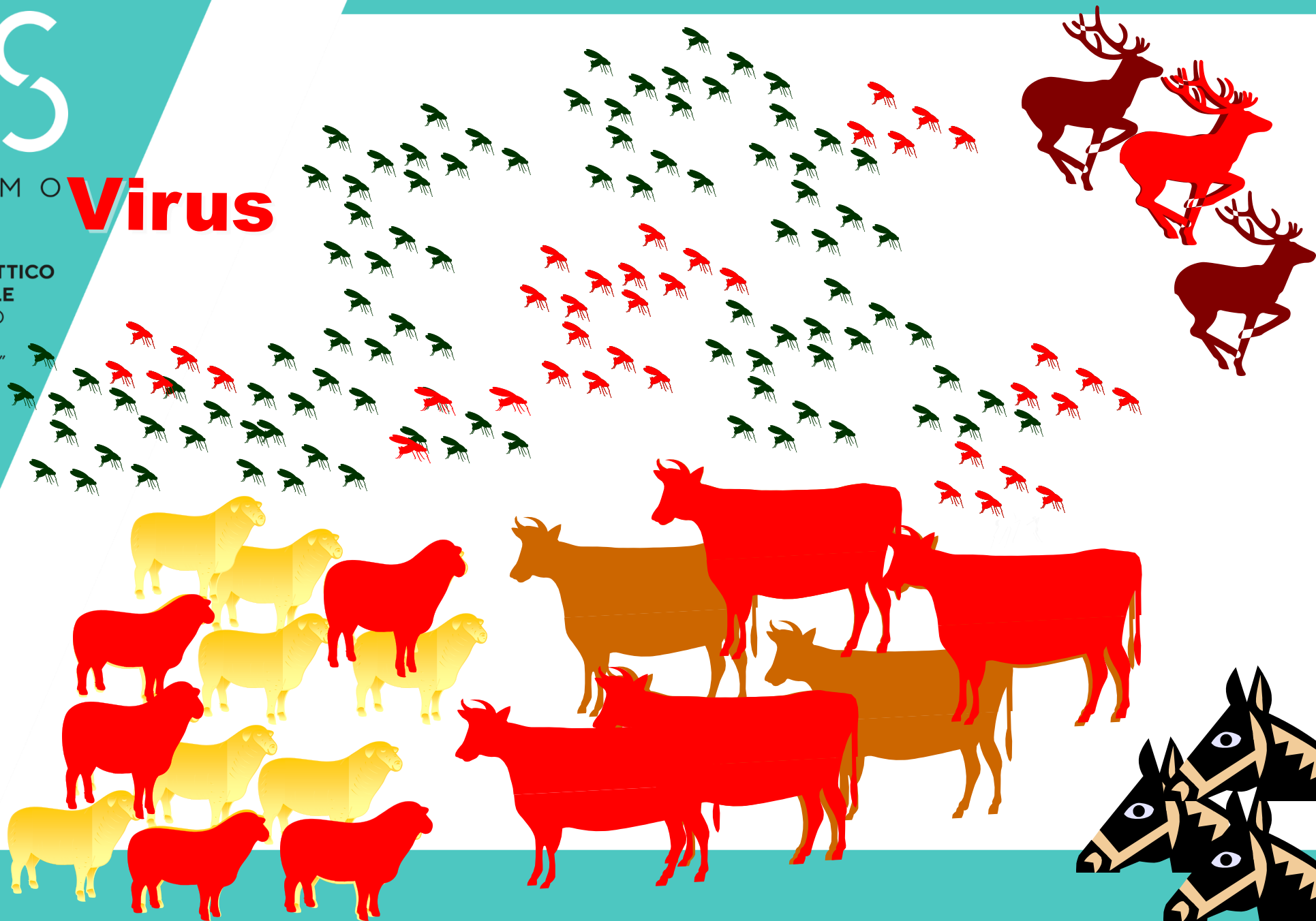
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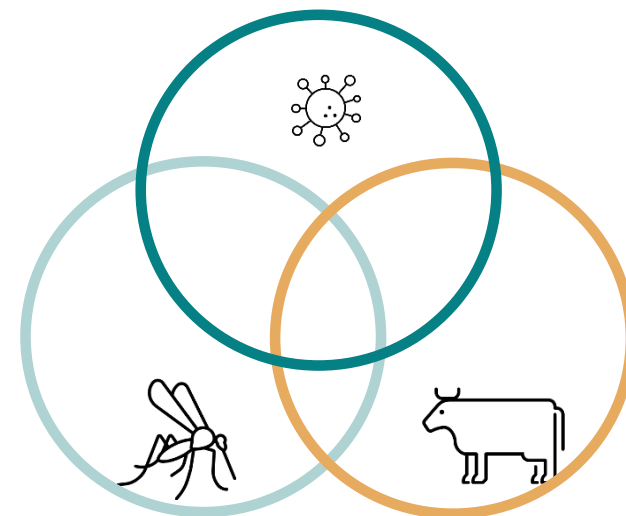
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Virus



The successful transmission of an arboviruses depends on a complex relationship between the virus, the vectors and the vertebrate hosts

- Vector competence
- Duration of the viraemia
- Duration of the incubation period(in the vector and in the vertebrate host)
- Behavioral patterns of the vector (eg host preference, day or night activity, frequency of meals, etc..)of the vertebrates (eg birds and mammals migrations)
- Factors of population abundance of vector species
- Availability of susceptible hosts
- Environmental factors must be favorable to virus, vectors and vertebrates



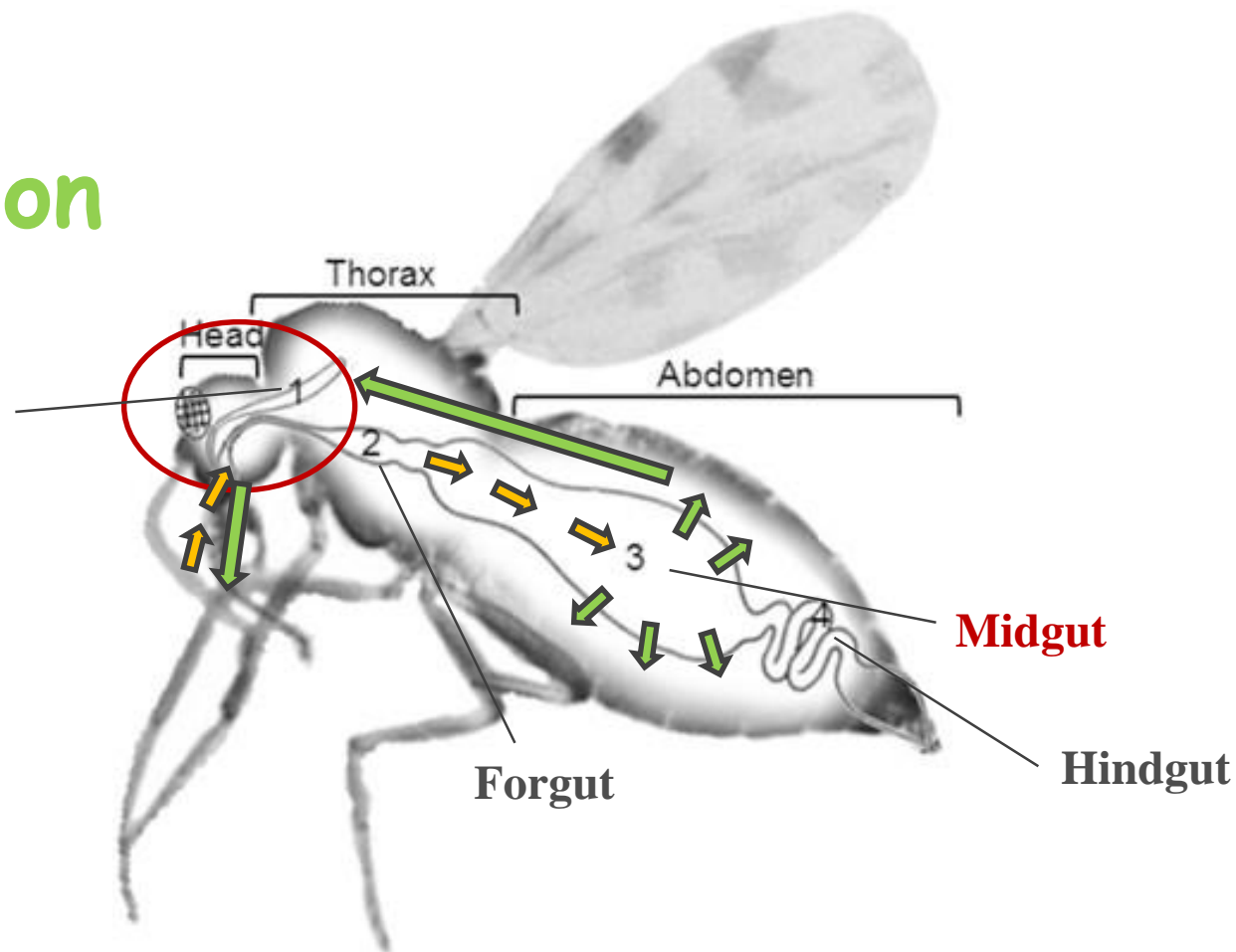
VECTOR COMPETENCE:

Ability of an arthropod to become infected from viremic hosts and then to transmit virus to susceptible hosts

AFTER AN INFECTED BLOOD MEAL...

Infection

Ghiandole salivari



Extrinsic incubation period: 1-2 weeks

Trasmissione

Main criteria to determine the VECTOR COMPETENCE

1. Field evidence that the distribution and seasonality of the virus is associated to those of the suspected vector
2. Recovery of the virus from wild-collected arthropods whose abdomens are free from visible fresh blood
3. Ability of the arthropod to become infected by feeding on a viremic host or an artificial meal
4. Ability to transmit the infection biologically by bite

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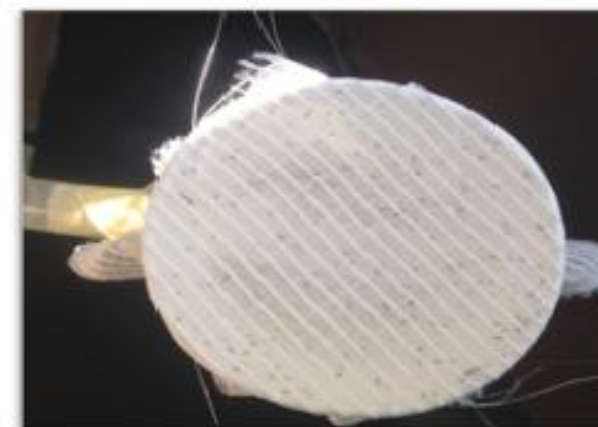
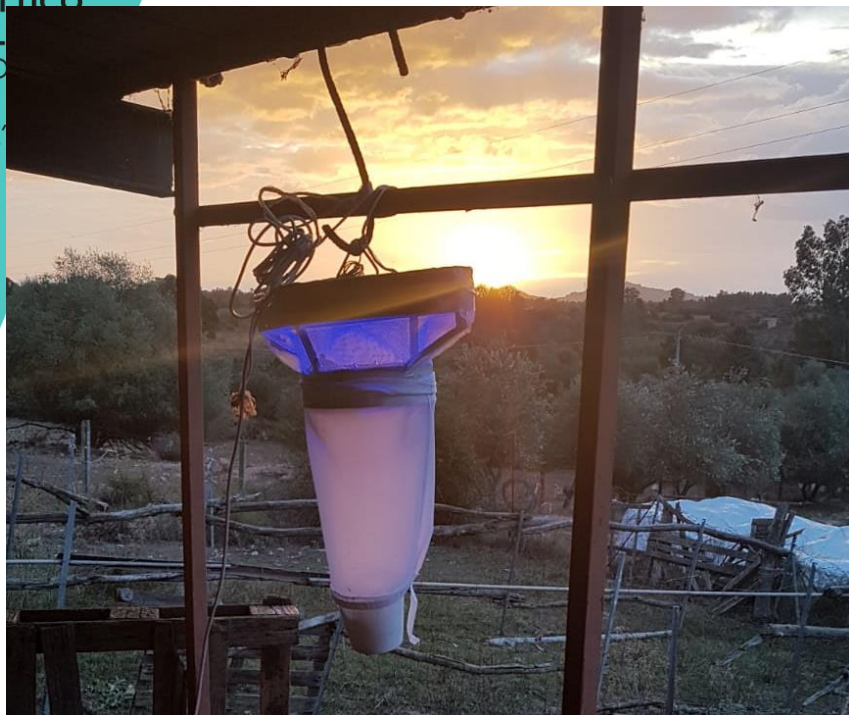
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Vector competence studies

field activities

On each farm a variable number (4-7) of UV blacklight suction traps operated on 2 or 3 consecutive days



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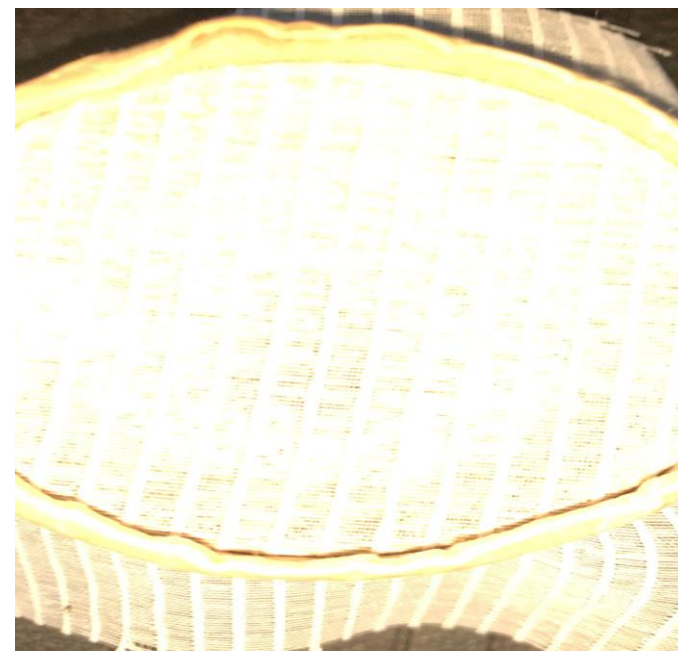
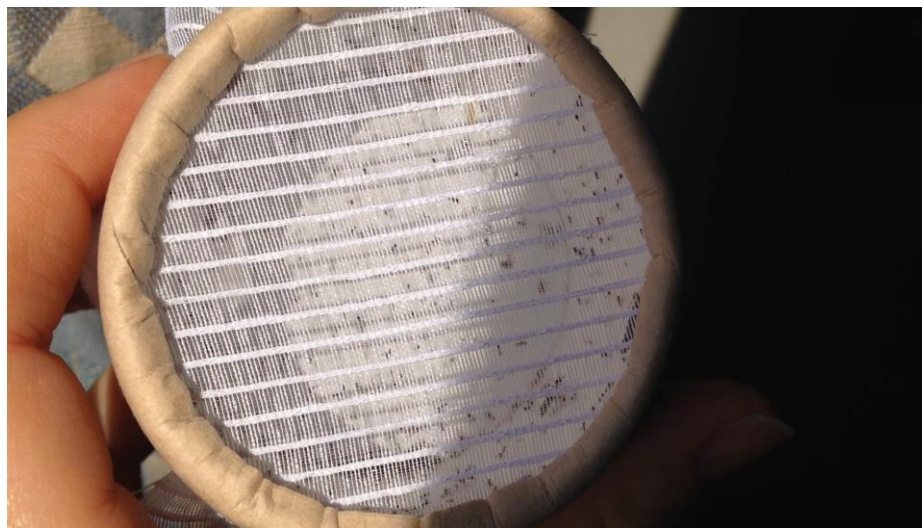
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Vector competence studies

field activities



After collection, alive ***Culicoides*** were transferred to the **lab** and were kept for **2 days of acclimation** with sucrose solution 10% and **1 day of starvation**, at 25°C and 70%>HR>40%.

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Vector competence studies

field activities



Vector competence studies

lab activities

Live caught *Culicoides* were fed using the **Hemotek** blood feeding system (blood at 37°C) with Parafilm membrane and/or **cotton pledget**, for 45-60 minutes.

