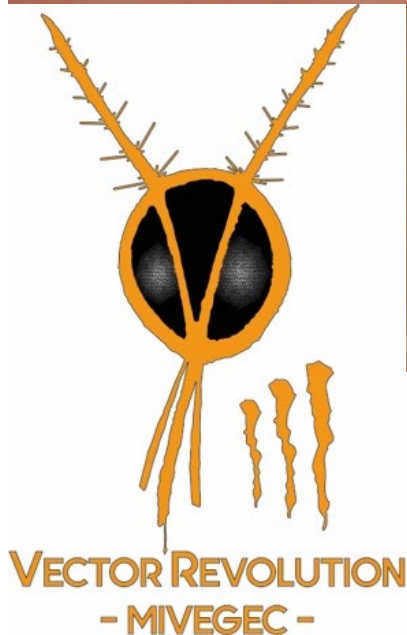


# Field evidence for manipulation of mosquito host choice by *Plasmodium falciparum*



Thierry Lefèvre,  
MIVEGEC lab,  
Univ. Montpellier, CNRS, IRD

# About 200 species of malaria parasites

A disease caused by a protozoan parasite from the genus *Plasmodium*

100 in Reptiles



40 in Birds



60 in Mammals



**Human malaria parasites:**

*Plasmodium falciparum*

*P. vivax*

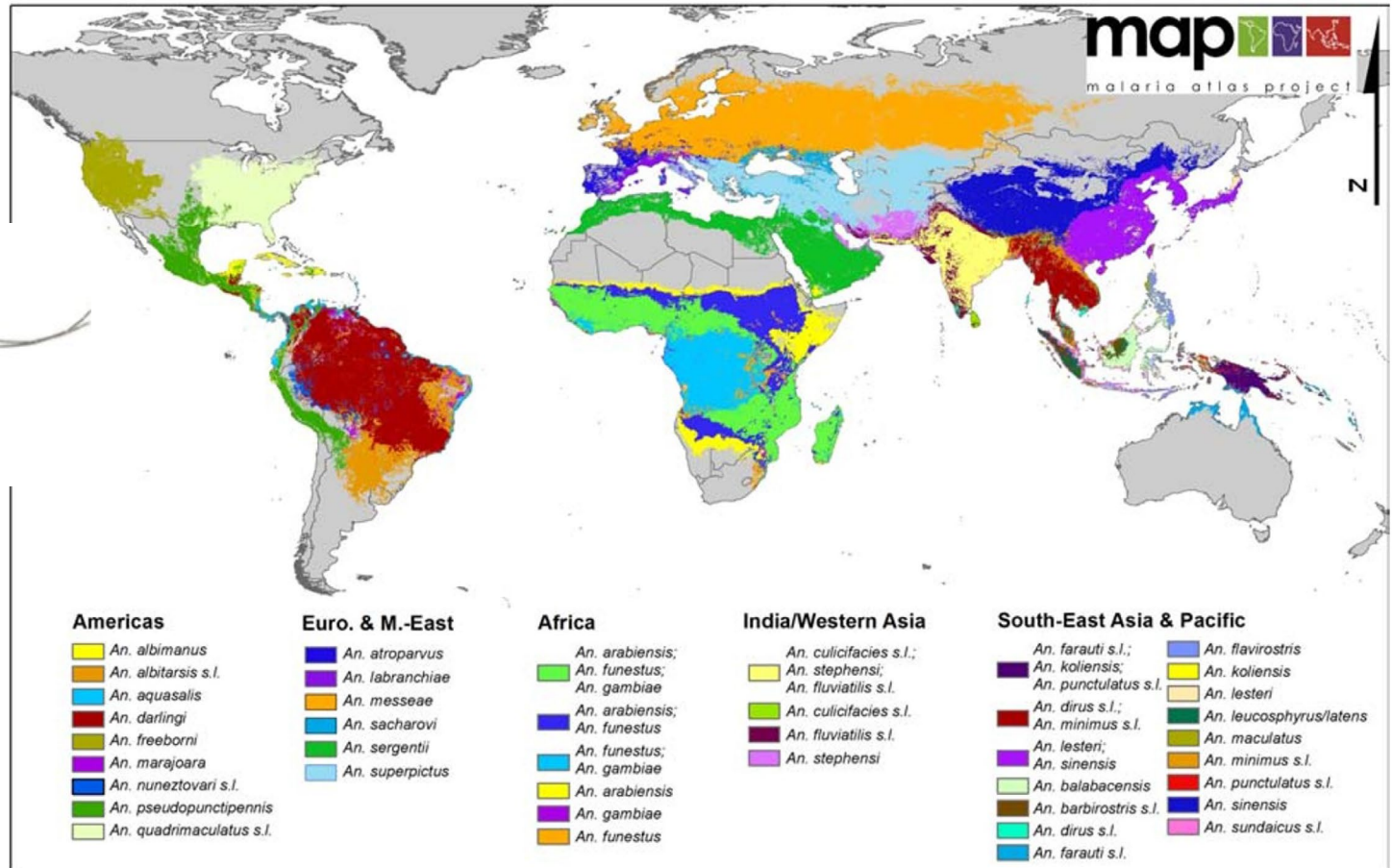
*P. ovale*

*P. malariae*

(*P. knowlesi*)



# Distribution of potential vectors of *P. falciparum*



~ 500 described species of *Anopheles* mosquitoes  
30-40 species possible vectors of *P. falciparum*



“To choose or not to choose, that is the question”



*Anopheles* mosquito-vertebrate host interactions

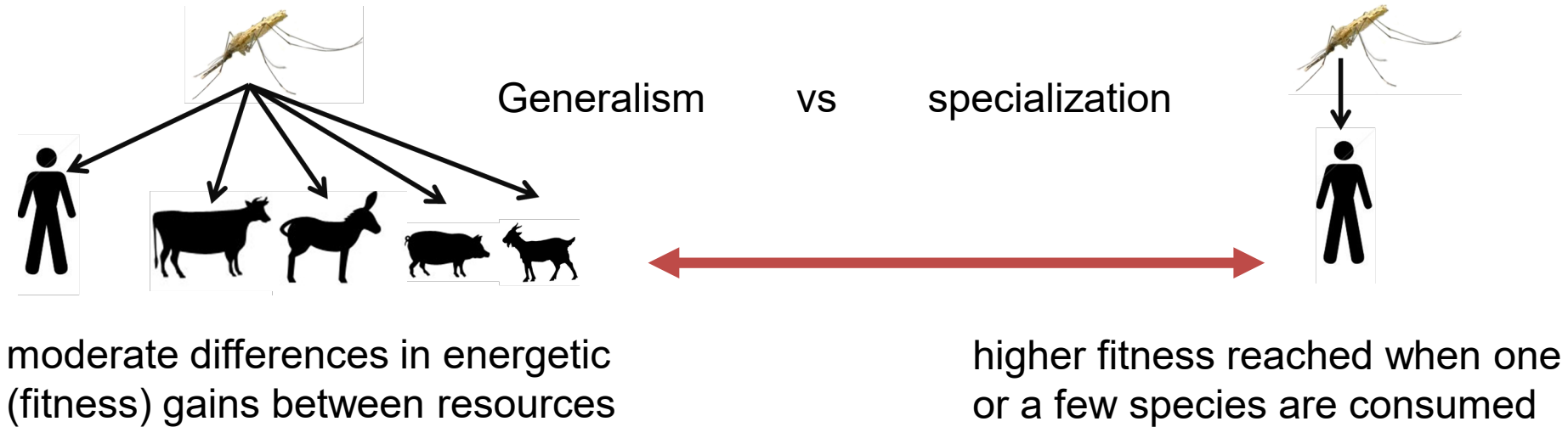
Ressource = blood meal

**“To choose or not to choose, that is the question”**

**Natural selection should favor mosquitoes that have a preference for resources which provide the highest fitness**

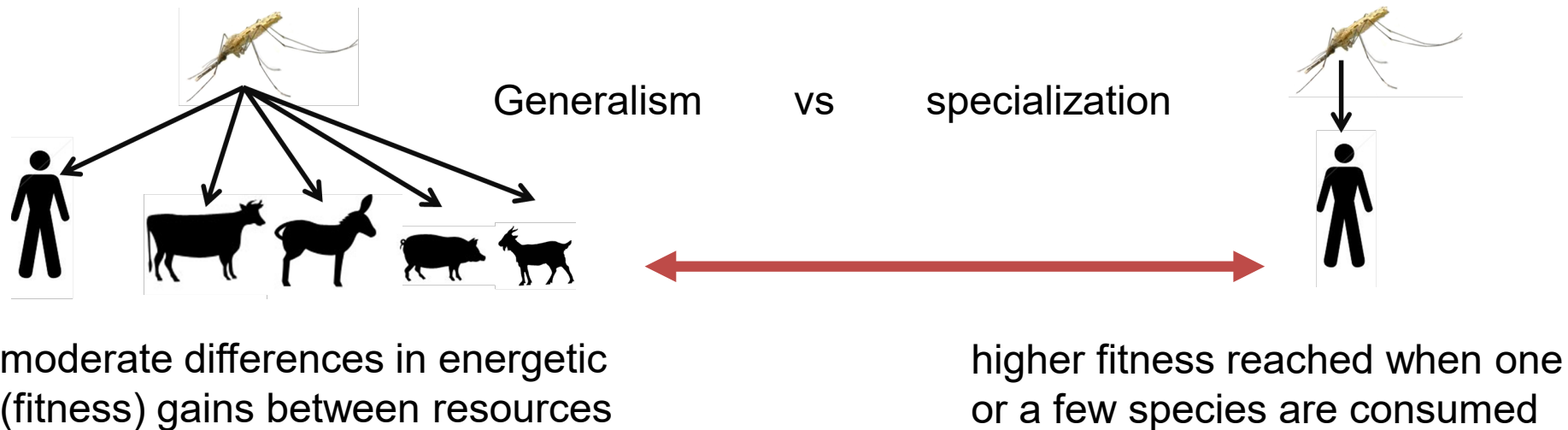
# “To choose or not to choose, that is the question”

**Natural selection should favor mosquitoes that have a preference for resources which provide the highest fitness**



# “To choose or not to choose, that is the question”

**Natural selection should favor organisms that have a preference for resources which provide the highest fitness**



**In contrast to phytophageous insects, there are very few studies on preference-performance relationships in *Anopheles* mosquitoes !!**

e.g. MC Singer et al. 1988 *Evolution*

**“To choose or not to choose, that is the question”**

What drive mosquito host choice ?



# “To choose or not to choose, that is the question”

What drive mosquito host choice ?

Certainly the interaction between:

- ❖ **Genetic preference** (related to performance/fitness?) Gillies, M.T. (1964) Selection for host preference in *Anopheles gambiae*. Nature



Divergent feeding preferences for humans or cows were generated within 5–6 generations of selection

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X

- ❖ Environmental factors (host diversity, accessibility, defensive behaviors, etc.)

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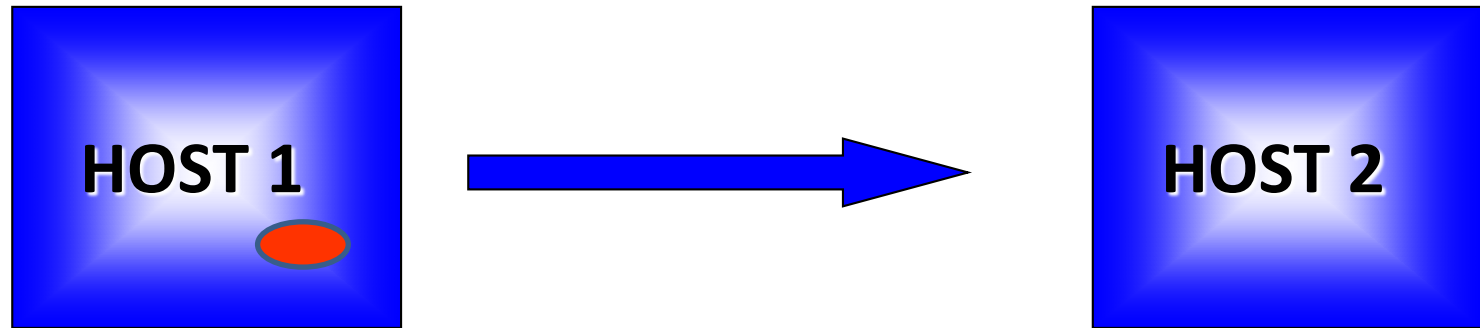
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X

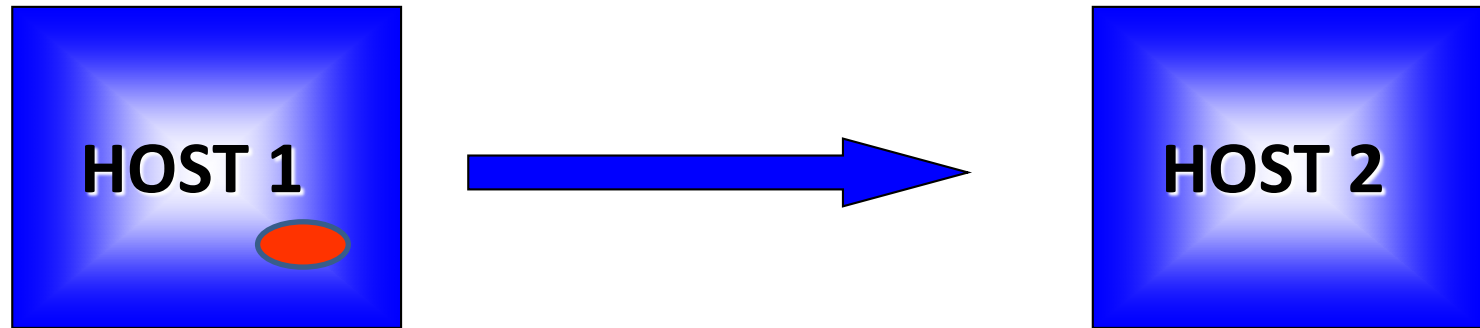
- ❖ Environmental factors (host diversity, accessibility, defensive behaviors, etc.)

**Infection with PLASMODIUM ?**

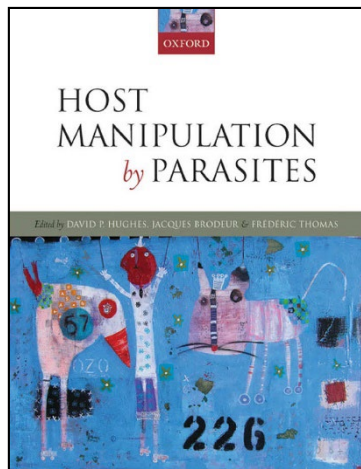
# Host behavioural manipulation by parasites



# Host behavioural manipulation by parasites

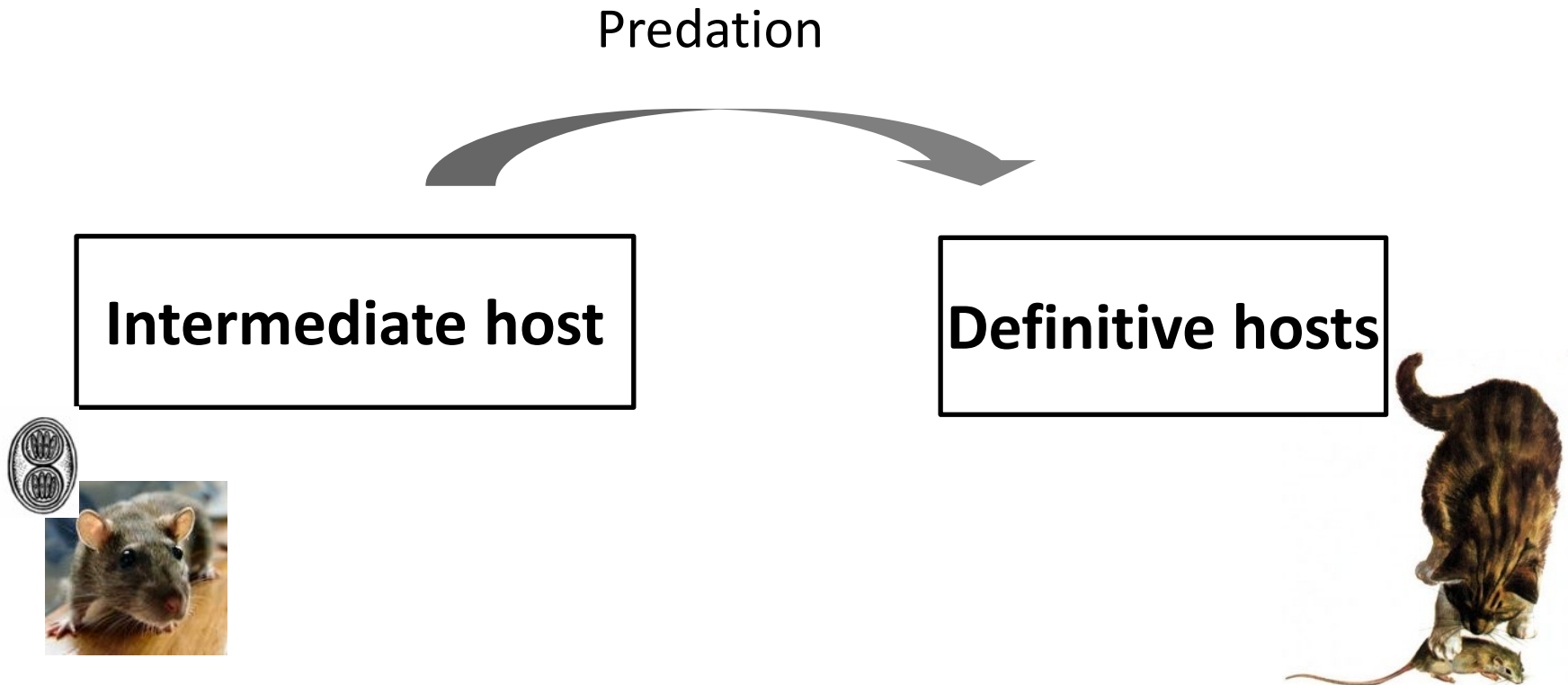


There are many examples supporting the existence of this parasite strategy of transmission



Hughes, Brodeur, Thomas, 2011 Oxford Univ. Press

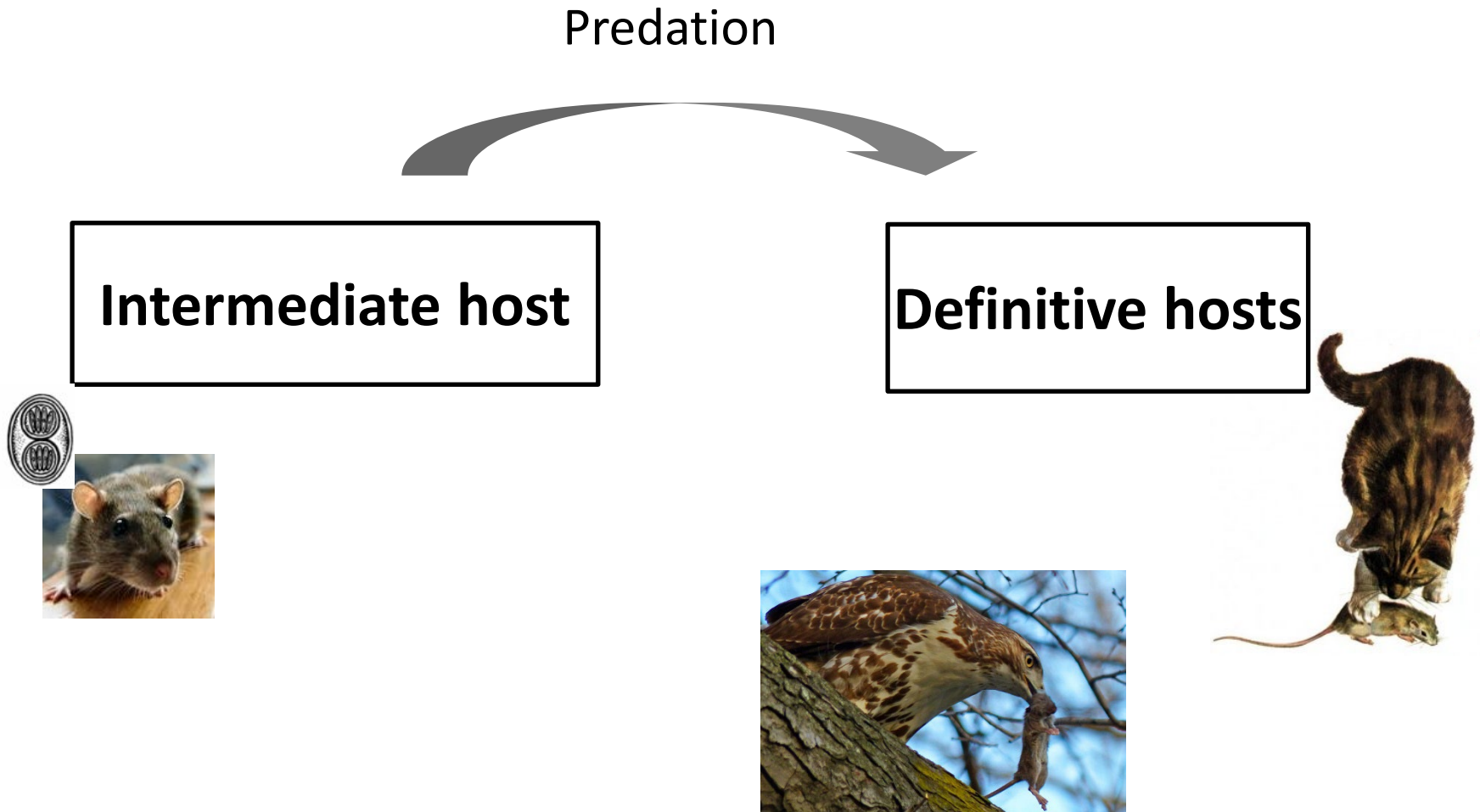
# Host behavioural manipulation by parasites



Trophically-transmitted parasites can alter the behaviour of their intermediate hosts in ways that increase predation rate by definitive hosts, hence favouring transmission

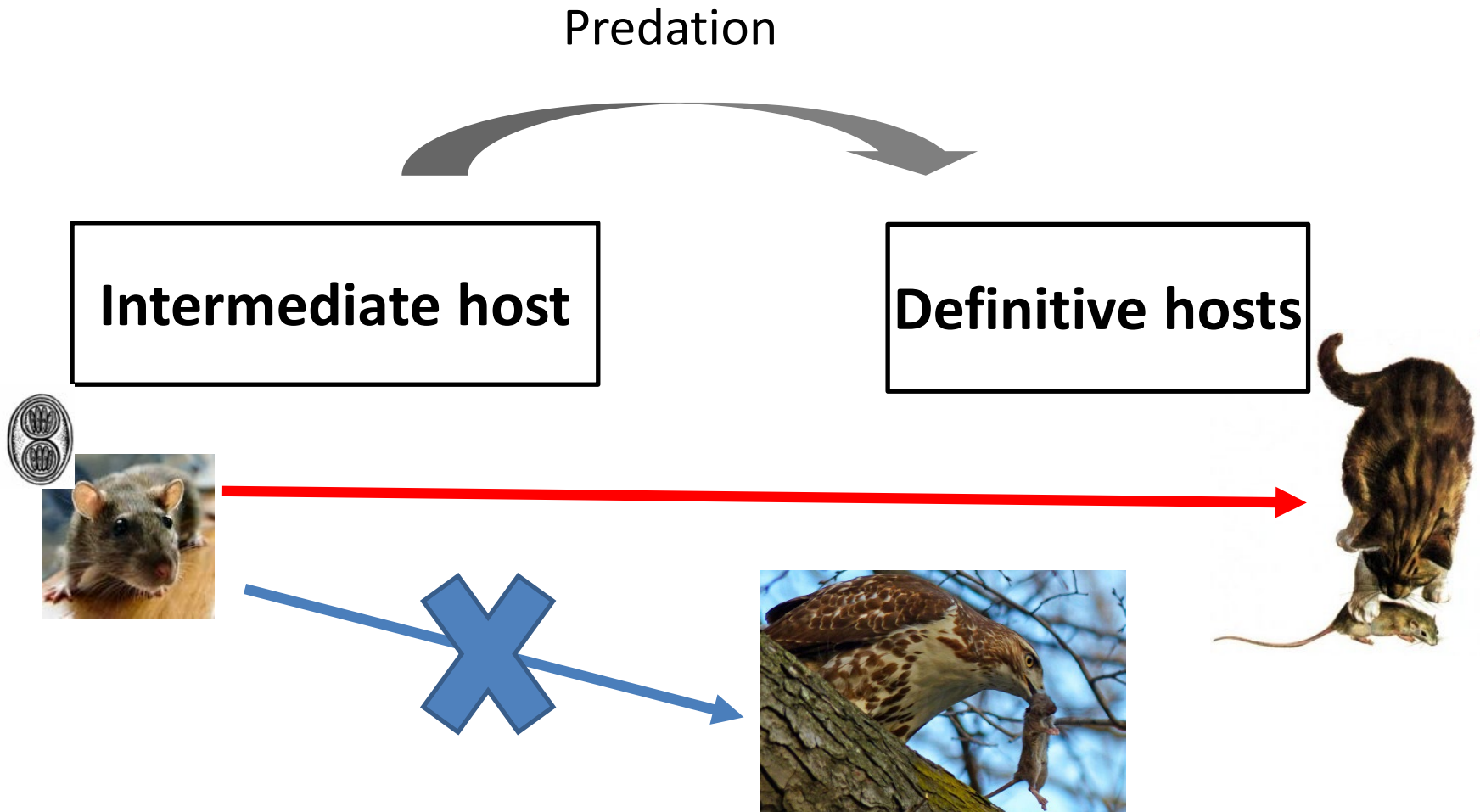


# Host behavioural manipulation by parasites



However, altering the behaviour of intermediate hosts can also increase predation rates by unsuitable hosts

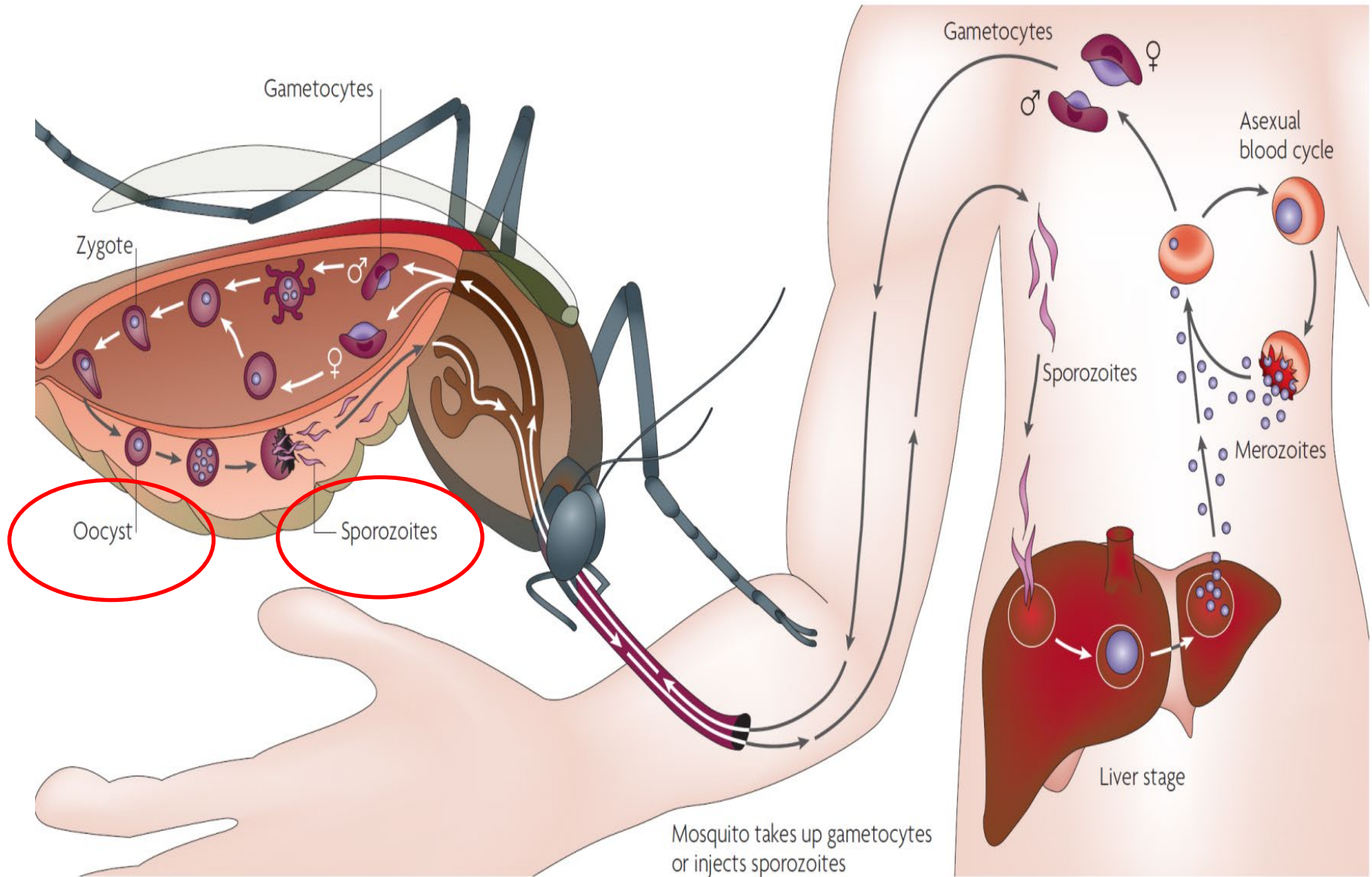
# Host behavioural manipulation by parasites



In response, some parasites have evolved specific manipulation, i.e. the ability to enhance transmission toward appropriate hosts and/or reduce predation by unsuitable hosts

# *Plasmodium* ability to manipulate their hosts?

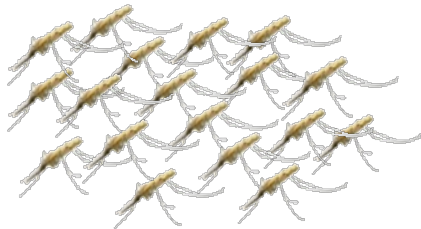
# The lifecycle of *P. falciparum*



# *Plasmodium* ability to manipulate their hosts?

# *Plasmodium* ability to manipulate their hosts?

1/ infected vertebrate hosts are more attractive to mosquito vectors



Uninfected



Infected

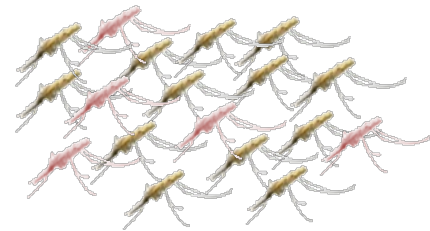
Lacroix et al. 2005, Cornet et al. 2012, De Moraes et al. 2014, Batista et al. 2014, Kelly et al. 2015, Busula et al. 2015, Emami et al. 2017, Robinson et al. 2018, etc.



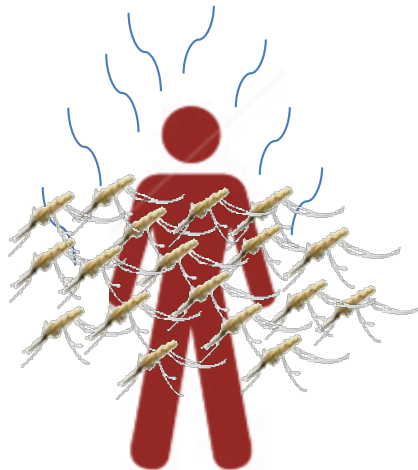
# *Plasmodium* ability to manipulate their hosts?

1/ infected vertebrate hosts are more attractive to mosquito vectors

2/ Infected mosquitoes display increased biting / feeding rate



Uninfected



Infected



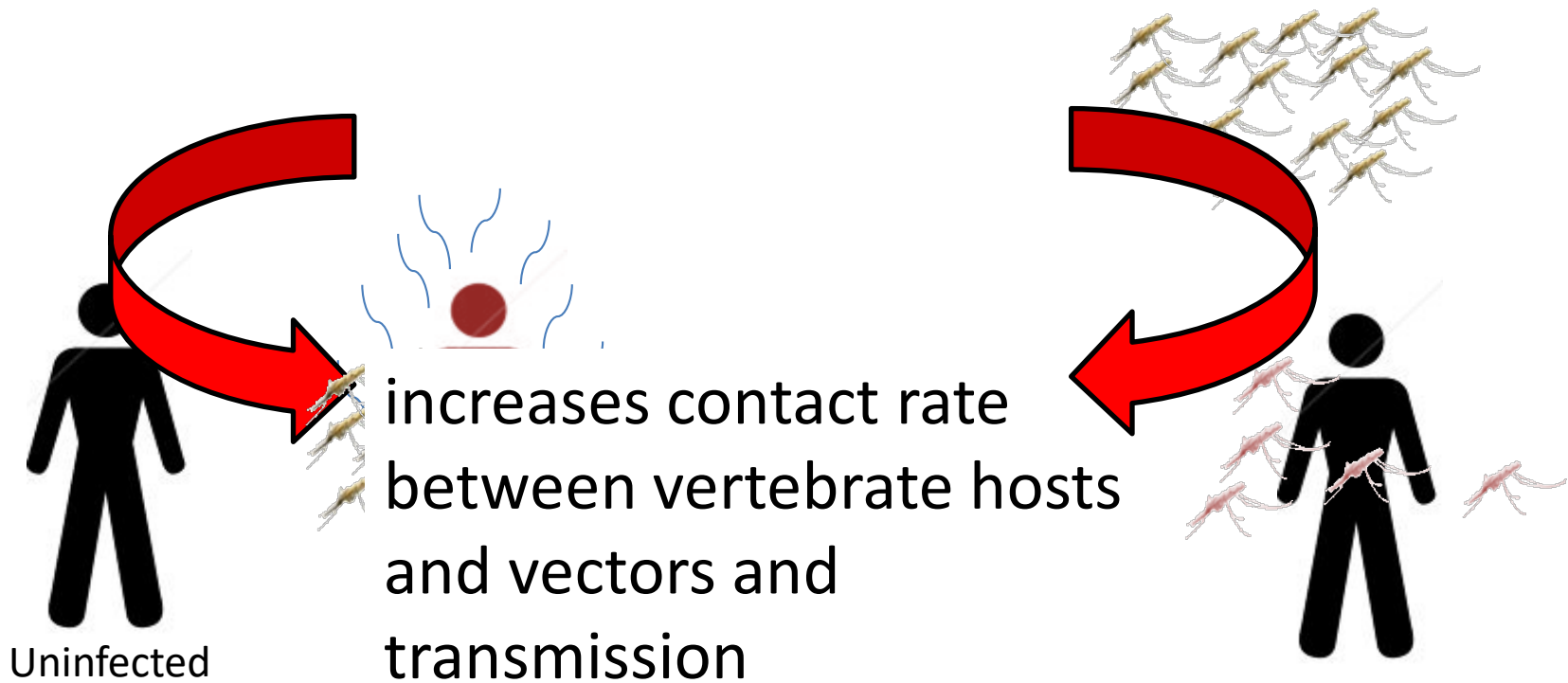
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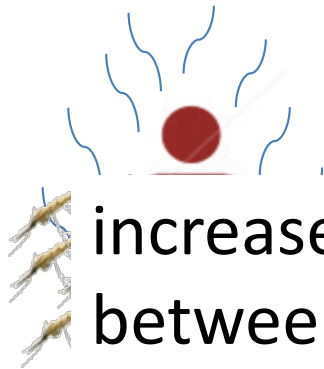


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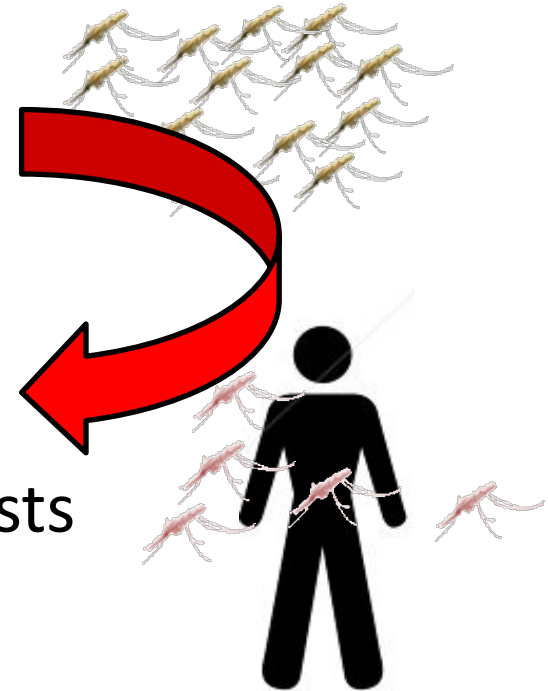
# *Plasmodium* ability to manipulate their hosts?

1/ infected vertebrate hosts are more attractive to mosquito vectors



increases contact rate  
between vertebrate hosts  
and vectors and  
transmission

2/ Infected mosquitoes display increased biting / feeding rate



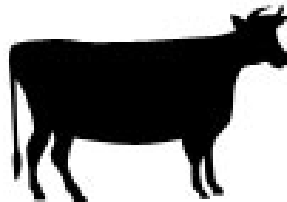
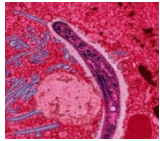
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# *Plasmodium* ability to manipulate their hosts?

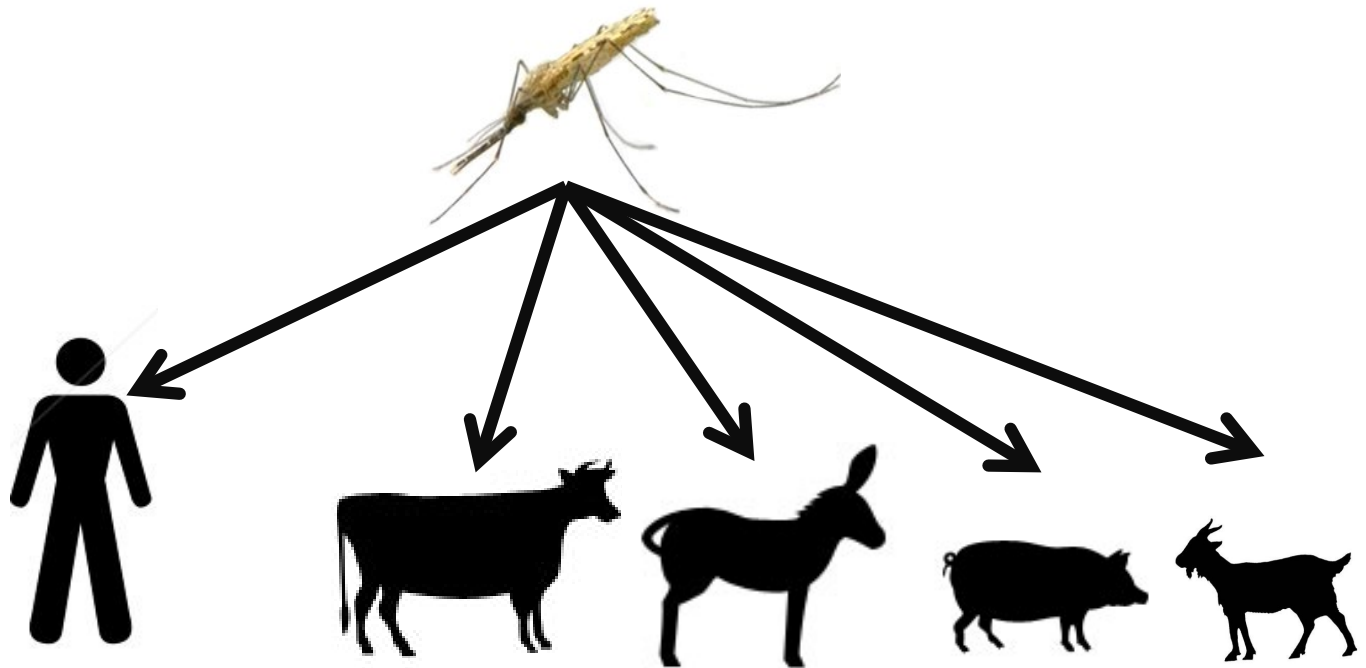
- not all vertebrate blood sources are suitable hosts for the parasite

*Plasmodium  
falciparum*



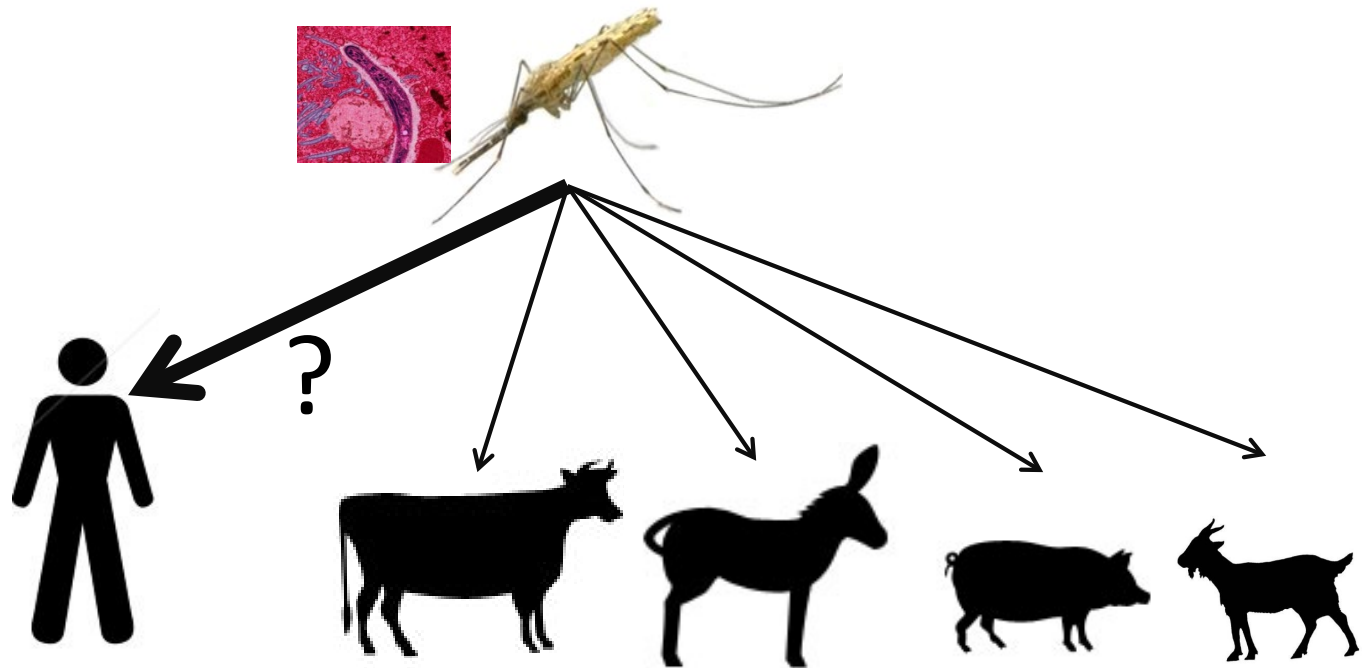
# *Plasmodium* ability to manipulate their hosts?

- not all vertebrate blood sources are suitable hosts for the parasite
- Malaria vectors can feed on a wide range of host species



# *Plasmodium* ability to manipulate their hosts?

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- Malaria vectors can feed on a wide range of host species



**HYPOTHESIS: Do *P. falciparum* manipulate mosquito host choice in ways that enhance parasite transmission toward human?**



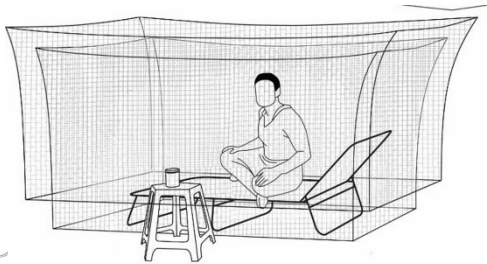
# Study sites

## 3 villages of South-Western Burkina Faso:

- Samandeni
- Soumouosso
- Klesso



# Experiment 1. Mosquito host preference



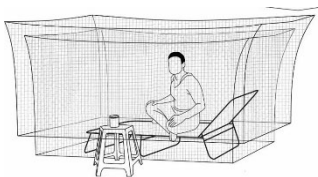
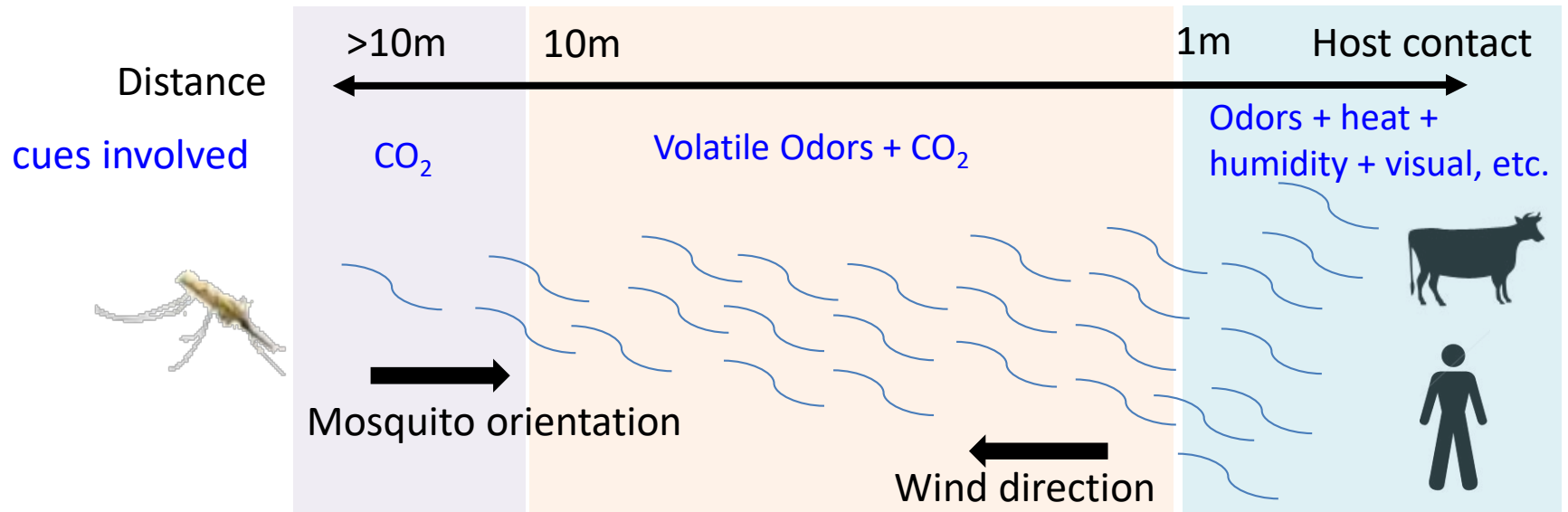
Odour-Baited Double Nets Traps (OBDNTs)



Odour-Baited Entry Traps (OBETs)

For each type of assay, two traps, set side by side in dual choice tests and releasing either human or calf odors were used to determine mosquito preference

# Experiment 1. Mosquito host preference



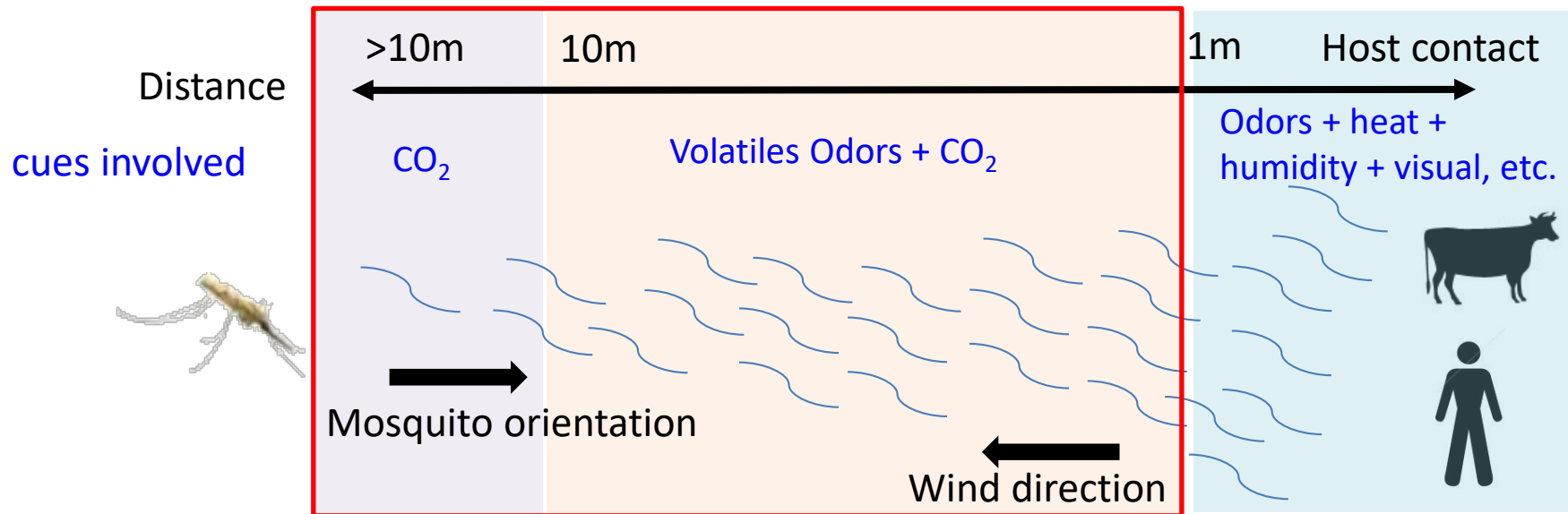
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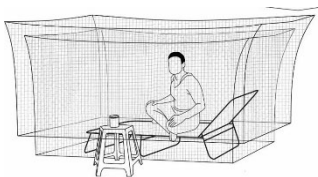
Odour-Baited Entry Traps (OBETs)



# Experiment 1. Mosquito host preference



long-range odour-mediated host preference, but...  
do not inform on the final realized choice of the mosquitoes

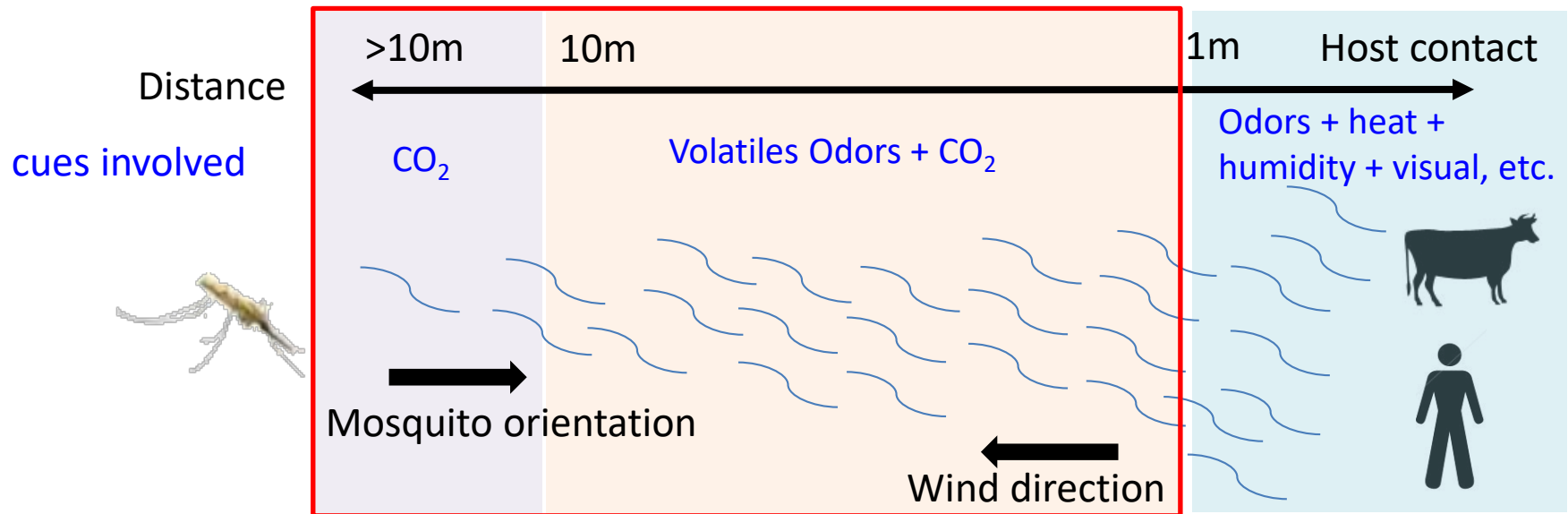


Odour-Baited Double Nets Traps (OBDNTs)



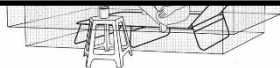
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# Experiment 1. Mosquito host preference

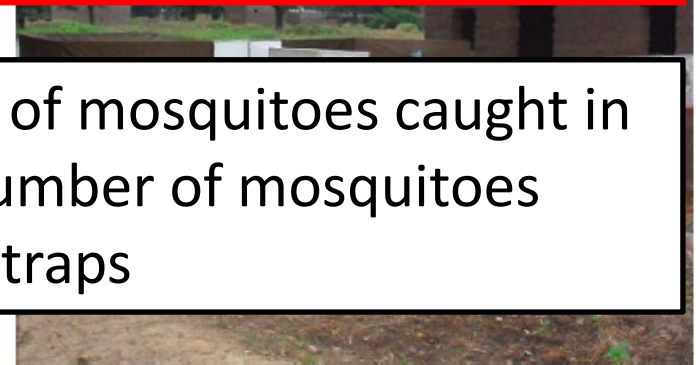


long-range odour-mediated host preference, but...  
do not inform on the final realized choice of the mosquitoes

Anthropophilic Index (AI) = the number of mosquitoes caught in the human-baited trap over the total number of mosquitoes caught in both human- and calf- baited traps



Odour-Baited Double Nets Traps (OBDNTs)



Odour-Baited Entry Traps (OBETs)

## Experiment 2: Mosquito blood-feeding pattern

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Indoor collection of freshly blood-fed mosquitoes in:  
human dwellings, unoccupied houses, and animal shades



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Indoor collection of freshly blood-fed mosquitoes in:  
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Focuses on the mosquito final decision by  
identifying bloodmeal origin (PCR or ELISA tests)  
retrospectively





## Experiment 2: Mosquito blood-feeding pattern

Indoor collection of freshly blood-fed mosquitoes in:  
human dwellings, unoccupied houses, and animal shades



Focuses on the mosquito final decision by  
identifying bloodmeal origin (PCR or ELISA tests)  
retrospectively

The Human Blood Index (HBI): the proportion of meals taken off  
humans

# Laboratory processing of field-collected mosquitoes

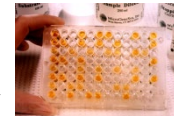


• Head / thorax

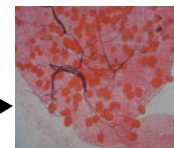


(1) Sporozoite qPCR detection

• Midgut



(3) ELISA or PCR blood-meal determination human, cattle, goat/sheep, pig, donkey/horse, dog and chicken



(4) Microscopical observation of oocysts and PCR checking

• Ovaries



(5) Microscopical observation of ovaries to determine the parity rate

→ Three infection status: uninfected individuals, oocyst-infected and sporozoite-infected mosquitoes

# Results

## ➤ Experiment 1. Mosquito host preference



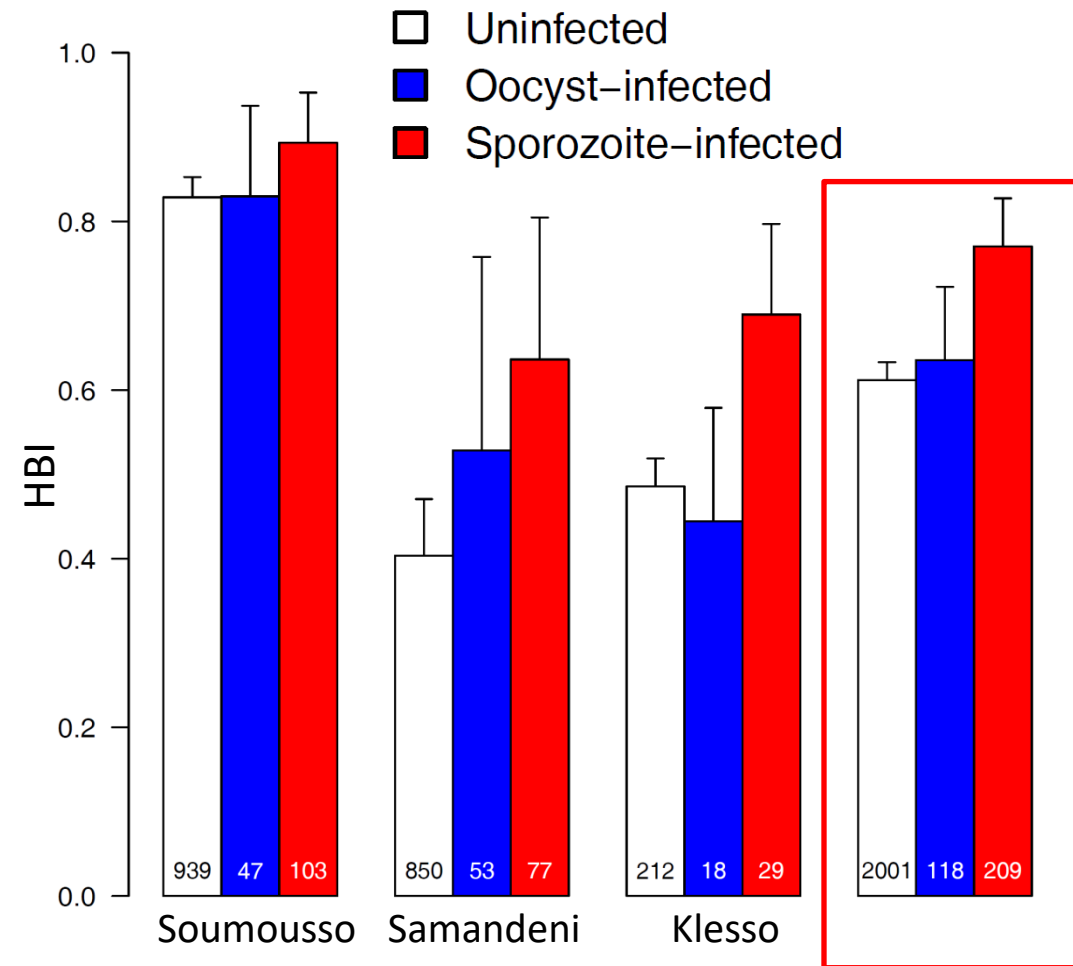
- The infection status (uninfected vs oocyst-infected vs sporozoite-infected) was successfully determined in 584 mosquitoes collected in the odour-baited traps
- no effect of infection on AI. Uninfected, oocyst-infected, and sporozoite-infected mosquitoes displayed similar host preferences ( AI~ 60%). *Nguyen et al. 2017 using a dual port olfactometer and experimentally infected mosquitoes in the lab*

## ➤ Experiment 2. Blood Feeding Pattern



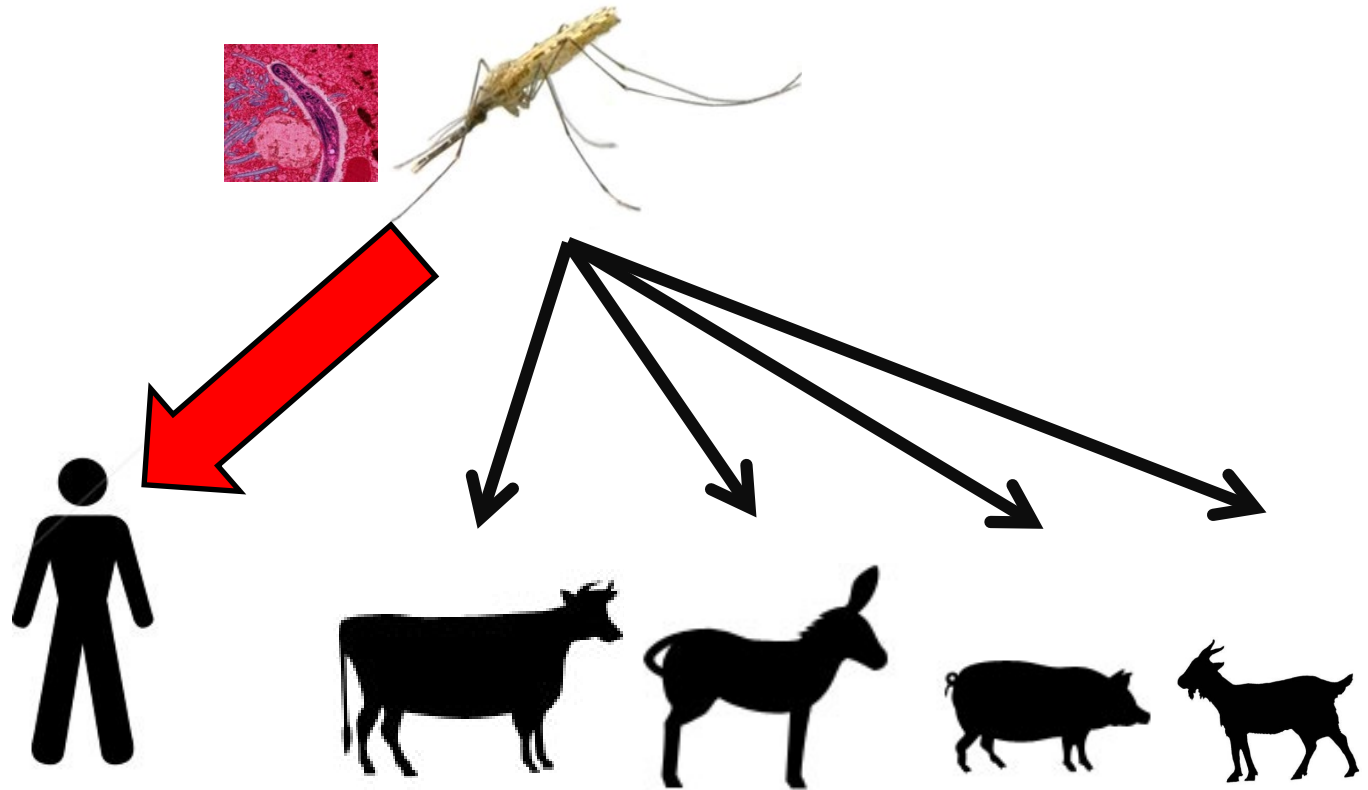
- The Blood-meal origin and infection status (oocyst-infected vs. sporozoite-infected vs. uninfected) of 2328 *Anopheles gambiae* sl were successfully determined.

# The effect of infection on HBI



$HBI_{\text{sporozoite-infected individuals}} = 77 \pm 5.7\%$   
 $>$   
 $HBI_{\text{oocyst-infected individuals}} = 63.6 \pm 5.7\%$   
 $=$   
 $HBI_{\text{uninfected individuals}} = 61.1 \pm 2.1\%$   
  
 $(LRT \chi^2_2 = 13.007; P = 0.0015)$

# Discussion and conclusions



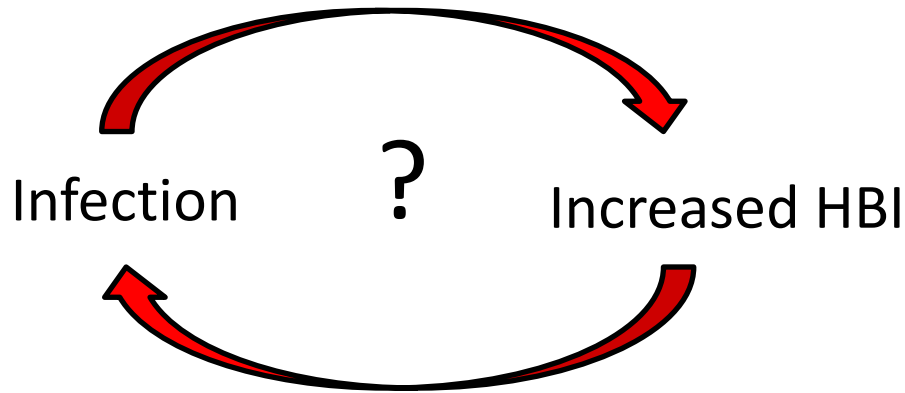
Sporozoites of *P. falciparum* enhance bloodfeeding on human, the suitable host for the parasite.

Is the parasite responsible for these changes?

→ Need to rule out two other possibilities

## Discussion and conclusions

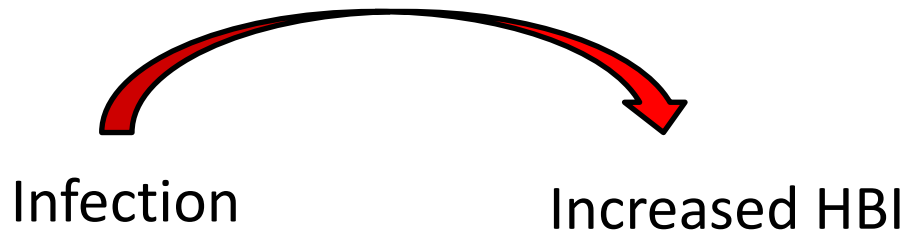
**rule out the potential confounding effect of a mere intrinsic mosquito characteristic.**



Infected mosquitoes may show increased HBI not because of being infected but just because of an innate propensity to feed on human, thus making these mosquito individuals more likely to become infected

## Discussion and conclusions

**rule out the potential confounding effect of a mere intrinsic mosquito characteristic.**

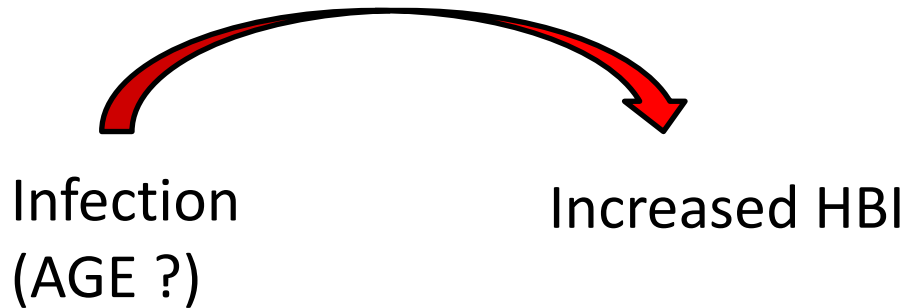


Infected mosquitoes may show increased HBI not because of being infected but just because of an innate propensity to feed on human, thus making these mosquito individuals more likely to become infected

NO! here the HBI of oocyst-infected individuals was similar to that of uninfected individuals and lower than that of sporozoite-infected individuals

# Discussion and conclusions

**rule out the potential confounding effect of mosquito age**



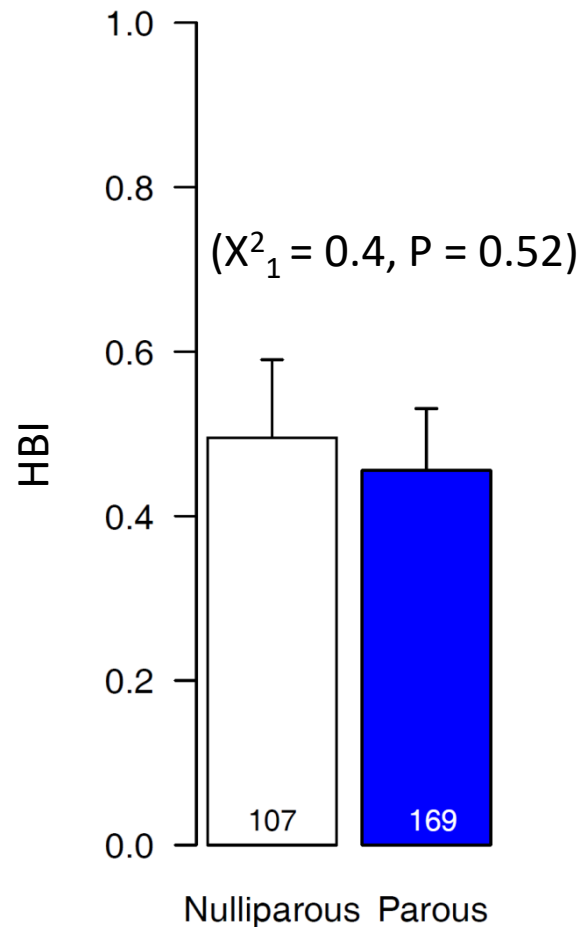
Sporozoite-infected mosquitoes may display increased HBI not because they carry sporozoites but because they are older ?



# Results

## effect of parity rate on HBI

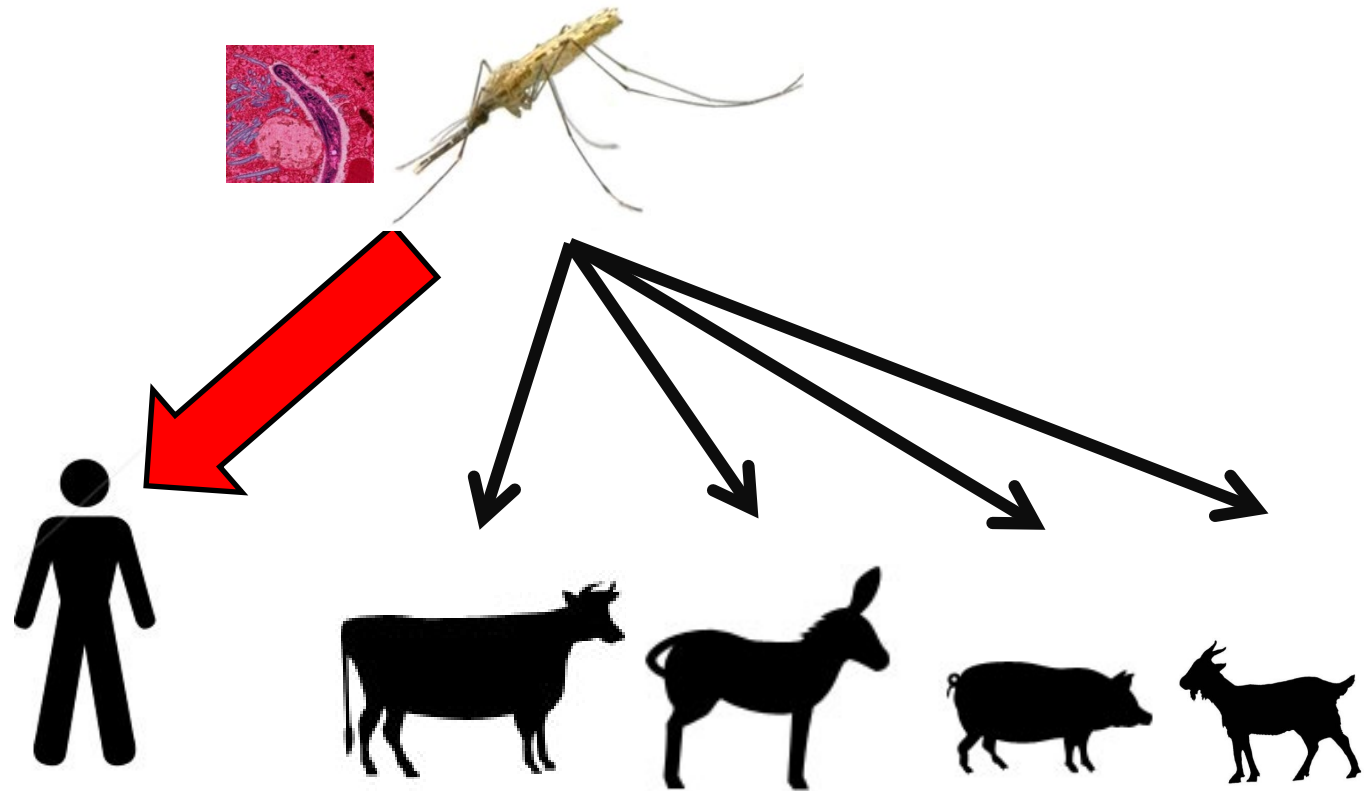
To determine whether mosquito age could influence HBI, we dissected a subset of mosquito ovaries to determine their parity rate.



Age effect? A priori no because HBI parous = HBI non-parous.

Parity rate is only a rough proxy for mosquito age and further studies are required using more precise age determination

# Perspectives

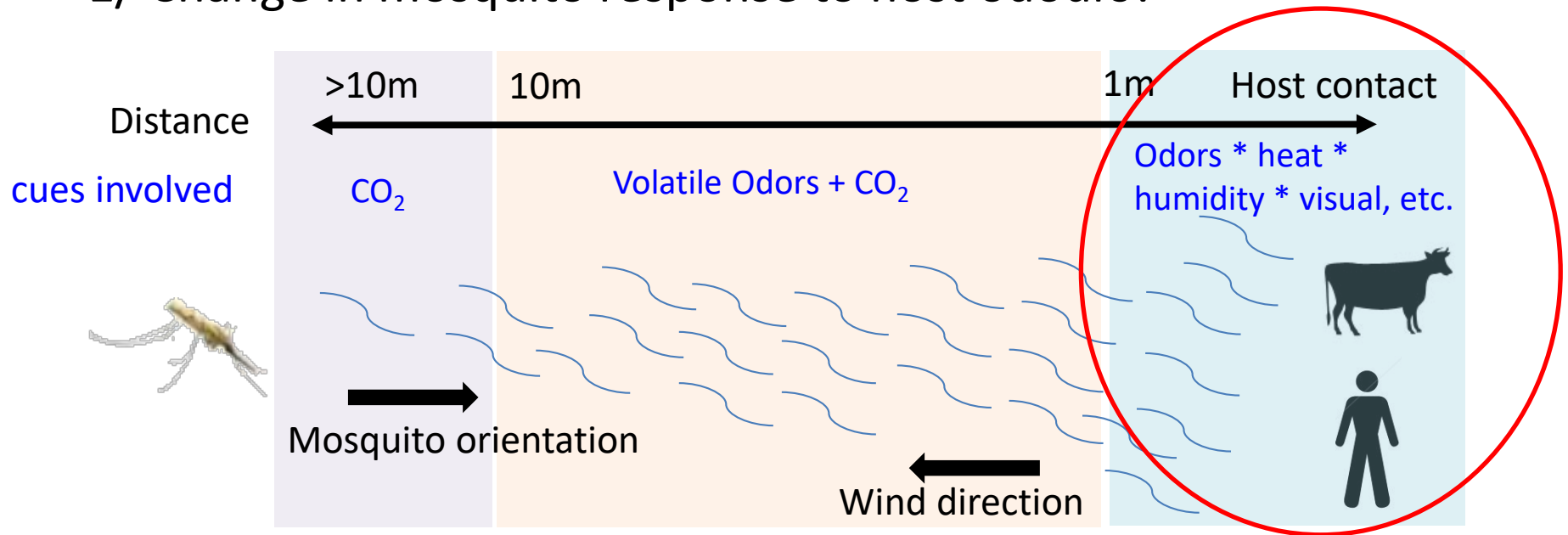


Our results suggest that *P. falciparum* alters mosquito host choice in ways that enhance parasite transmission toward suitable hosts and/or reduce mosquito attraction to unsuitable hosts i.e. specific manipulation.

- Underlying proximate mechanisms?

# Perspectives

## 1/ Change in mosquito response to host odours?



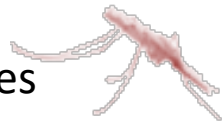
Parasites might manipulate mosquito short-range behaviours only.

The combinations of short-range stimuli (odors, heat, humidity, visual) are host specific and may inform of suitability for parasite development before the mosquito engages in selection and feeding

# Perspectives

2/ Spatial “rendez-vous” with the human host: Change in mosquito resting behaviour (endophily/phagy)?

Infectious  
mosquitoes



infectious mosquitoes may exhibit an enhanced tendency to enter (or a decreased tendency to exit) house interstices regardless of emitted odors.

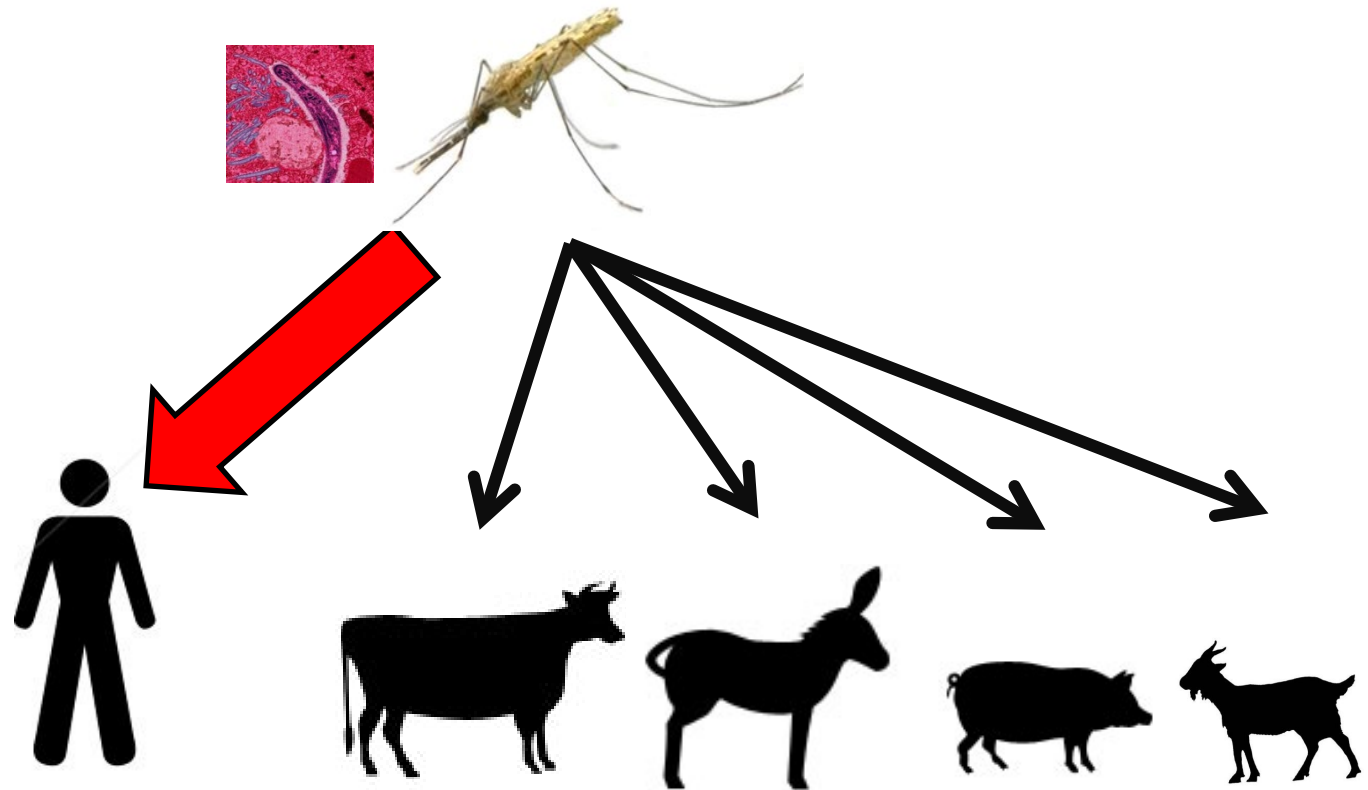
# Perspectives

3/ “temporal rendez-vous” with the human host: Change in mosquito temporal activity



*P. falciparum* could manipulate mosquito rhythms in a way that increases bites on unprotected people

# Perspectives



Our results suggest that *P. falciparum* alters mosquito host choice in ways that enhance parasite transmission toward suitable hosts and/or reduce mosquito attraction to unsuitable hosts i.e. specific manipulation

- Underlying proximate mechanisms?
- Confirm this phenotype in controlled laboratory conditions

# Peer Community Journal

Section: Evolutionary Biology



## RESEARCH ARTICLE

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Correspondence  
amelie.vantaux@gmail.com

## Field evidence for manipulation of mosquito host selection by the human malaria parasite, *Plasmodium falciparum*

Amélie Vantaux<sup>1,2</sup>, Franck Yao<sup>1</sup>, Domonbabele FdS Hien<sup>1</sup>, Edwige Guissou<sup>1</sup>, Bienvenue K. Yameogo<sup>1</sup>, Louis-Clément Gouagna<sup>2</sup>, Didier Fontenille<sup>2</sup>, François Renaud<sup>2</sup>, Frédéric Simard<sup>2</sup>, Carlo Constantini<sup>2</sup>, Frédéric Thomas<sup>2</sup>, Karine Mouline<sup>1,3,2</sup>, Benjamin Roche<sup>2,4</sup>, Anna Cohuet<sup>2</sup>, Kounbobr R Dabiré<sup>1,3</sup>, and Thierry Lefèvre<sup>1,3,2</sup>

Volume 1 (2021), article e13

<https://doi.org/10.24072/pcjournal.13>



Amélie Vantaux

# THANK YOU



# Results

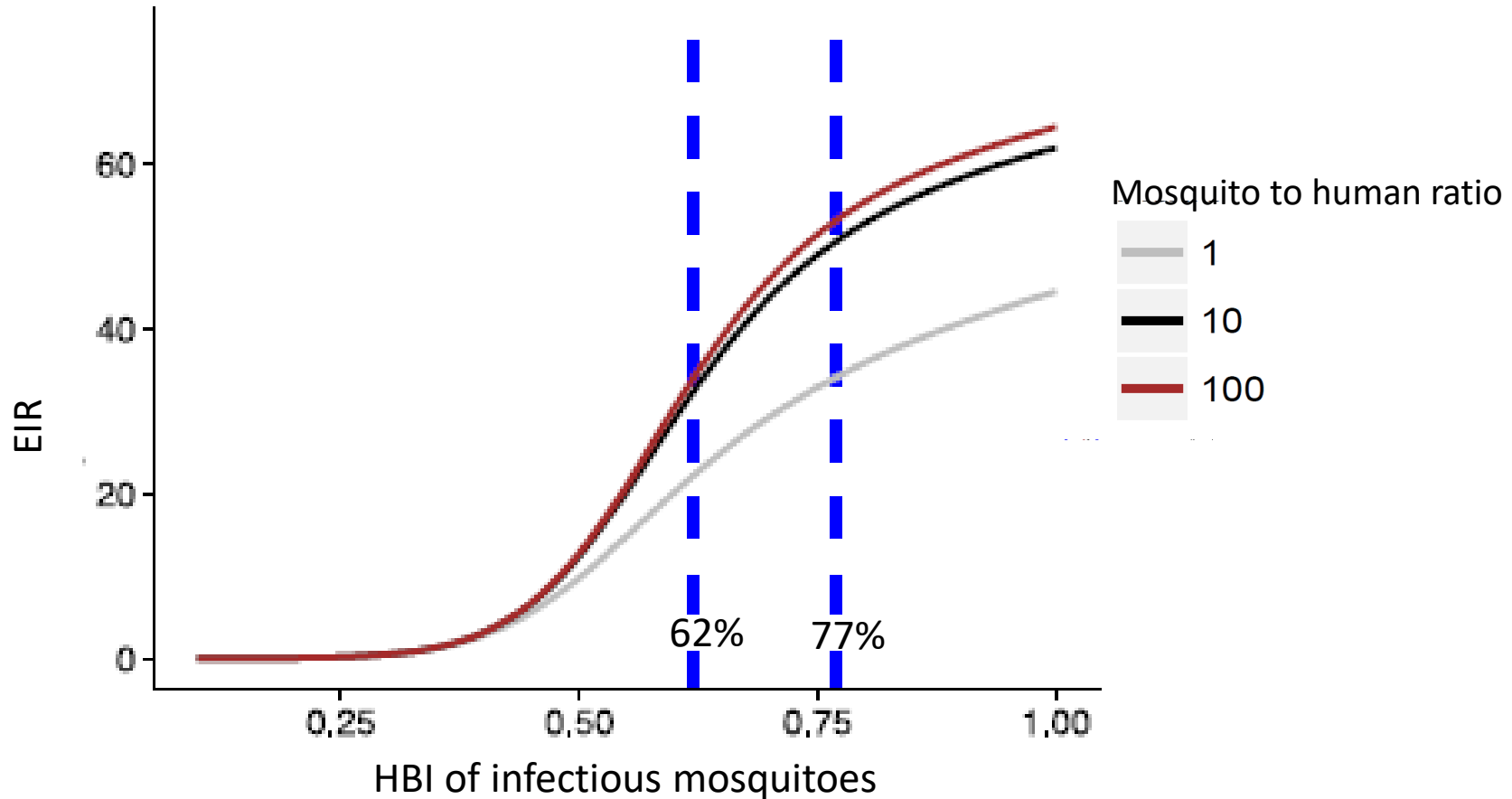
Consequences on transmission potential?



# Results

## Consequences on transmission potential?

Entomological Inoculation Rate (number of infectious bites received by a person over one year)



The observed increased anthropophagy (from 62% to 77%) in infectious females has important epidemiological consequences with up to **250% increase in parasite transmission**

# Implications for management and control?

- ❑ Deciphering the underlying mechanisms (which cues involved?) should help designing traps specifically targeting infectious mosquito females (which cues? Where and when?)



Blood feeding

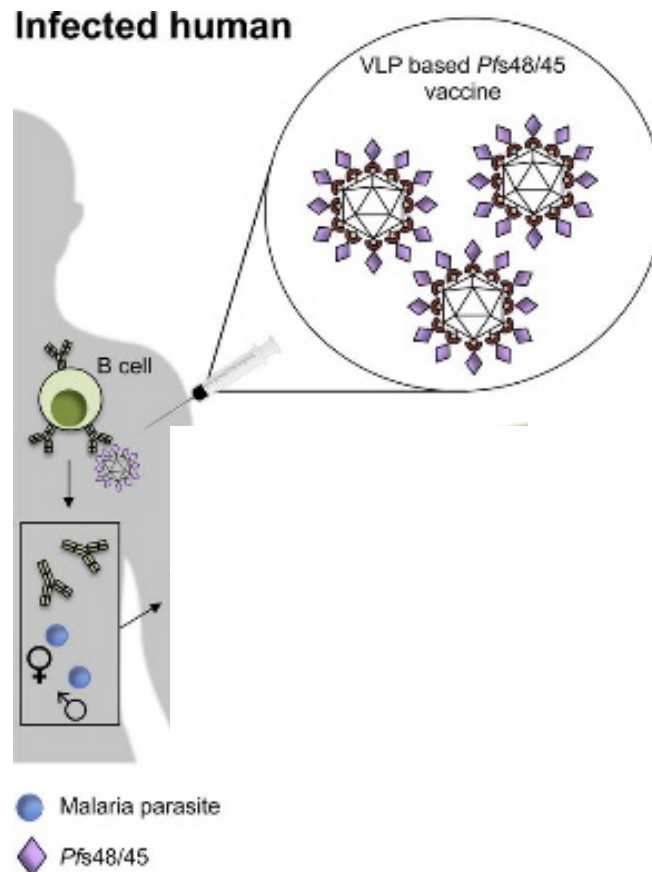


# Implications for management and control?

- ❑ The development of transmission-blocking strategies: curing the vector to stop transmission

## 1/ “Altruistic” Vaccine

First clinical trial of a transmission-blocking vaccine in the field (2019-2024). Adrian Hill Lab

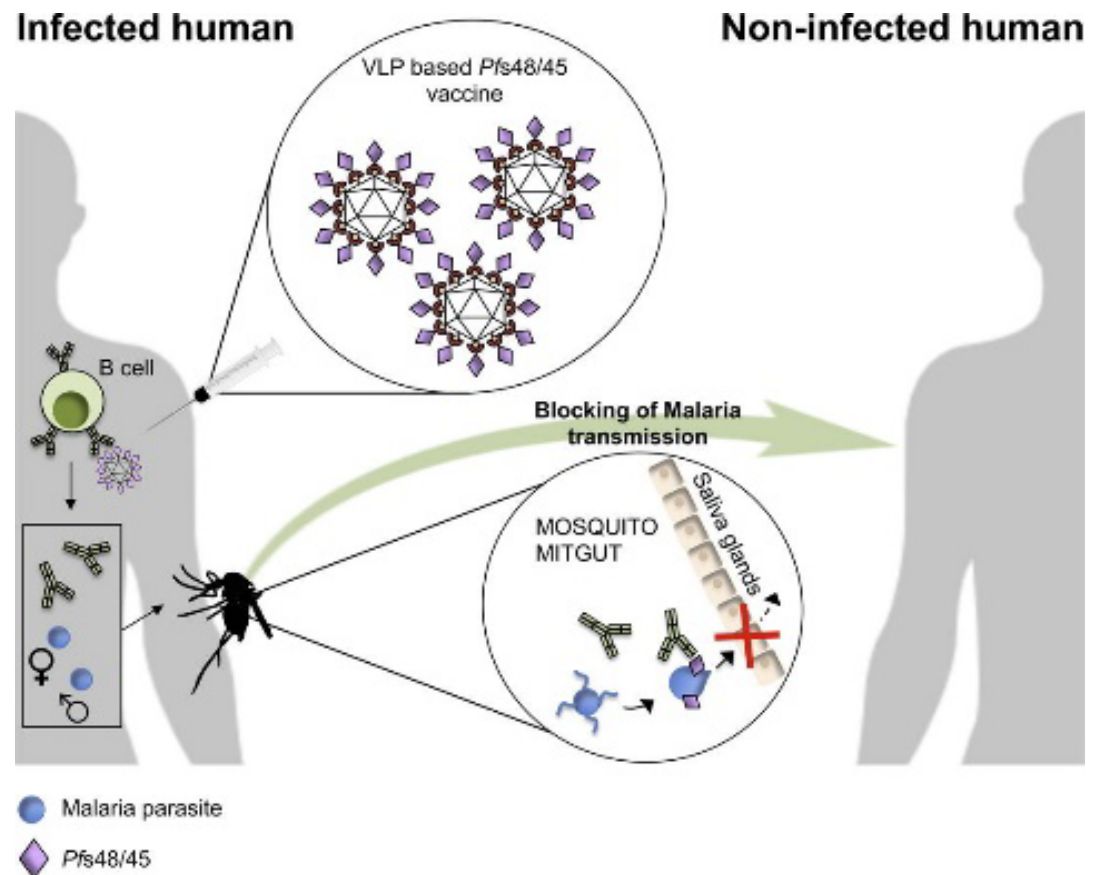


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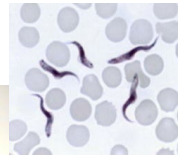


# Perspectives: Beyond malaria

## Bite more or bite better?

- Other Vector-borne parasites of medical importance ?
  - Generalist/opportunistic vectors with broad range of hosts
  - Specialist parasites with narrow host range

*Glossina* spp. –  
*Trypanosoma* spp.



?

# Perspectives: Beyond malaria

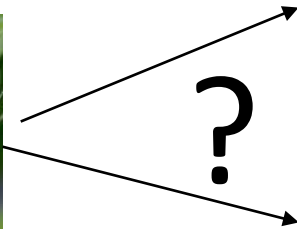
## Bite more or bite better?

➤ In vector-borne plant parasites:

- Polyphageous vectors
- Parasites with narrow host range

- ❑ The cassava mosaic virus disease complex. polyphageous vector (*Bemisia tabaci*) and specific to cassava ?

CMV-infected  
vector



cassava



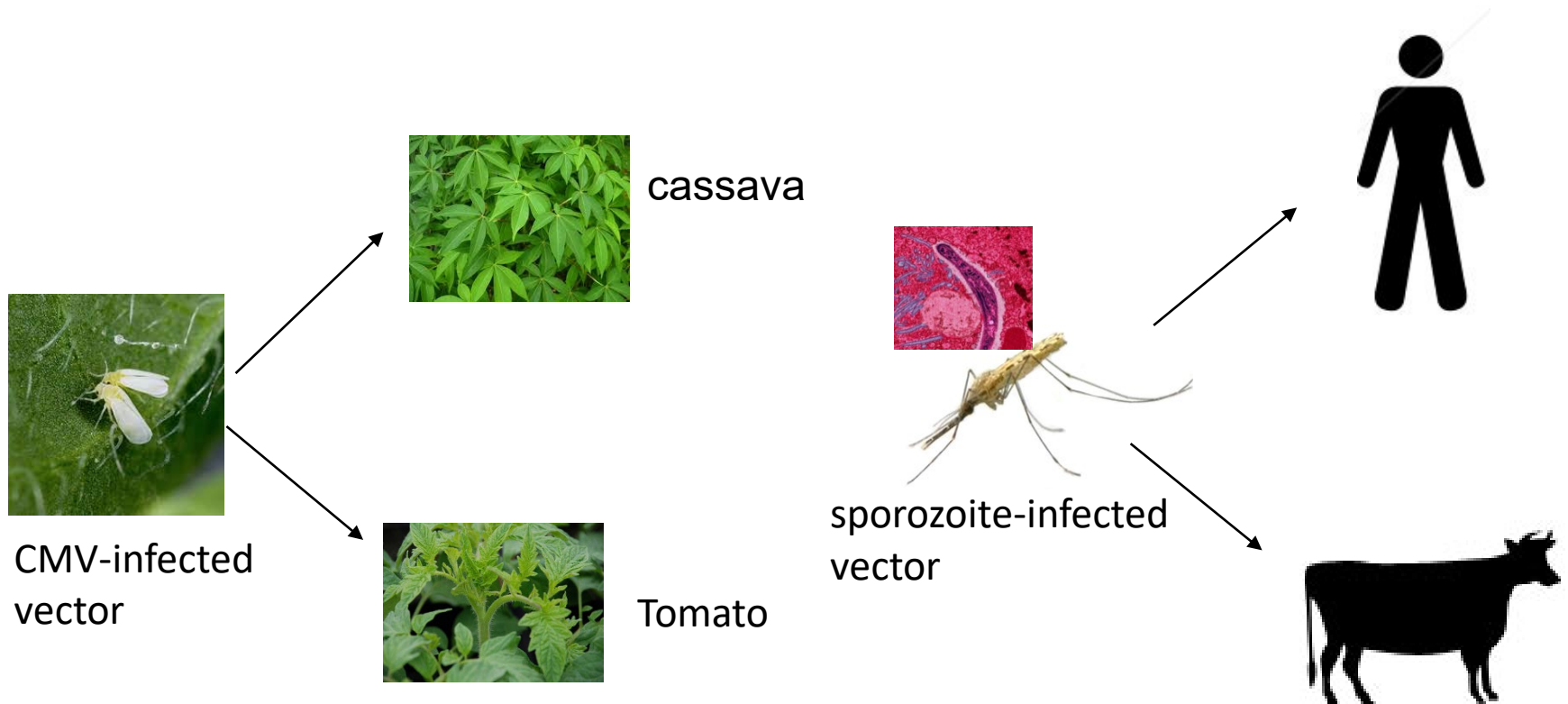
Tomato

- ❑ The sweet potato viruses transmitted by various polyphagous vectors (aphids like *Myzus persicae* and *Aphis gossypii*), but the host range seems to be restricted to *Convolvulaceae* and mostly the genus *Ipomoea* ?

# Perspectives: Beyond malaria

Bite more or bite better?

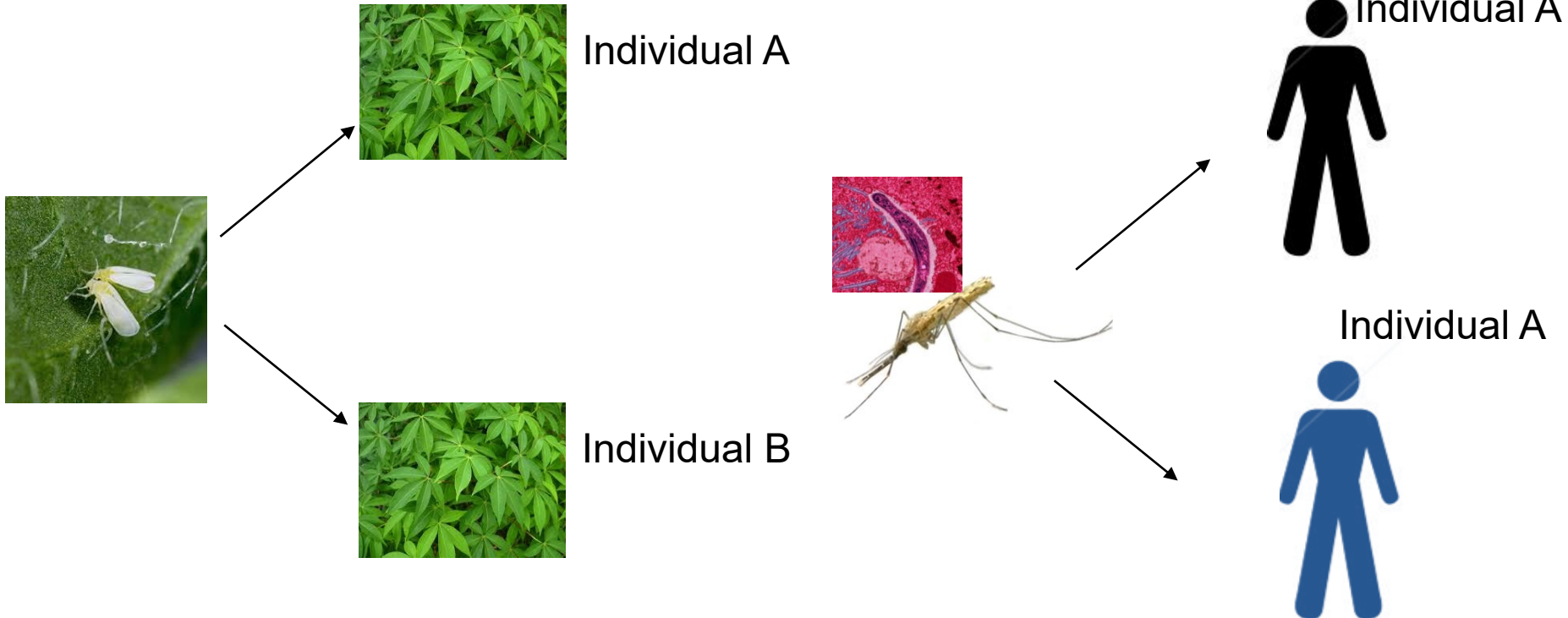
At the inter-specific level



# Perspectives: Beyond malaria

Bite more or bite better?

At the intra-specific level





# Perspectives: beyond manipulation of feeding behaviour

Contact rate is one determinant of transmission intensity among others

Vectorial capacity

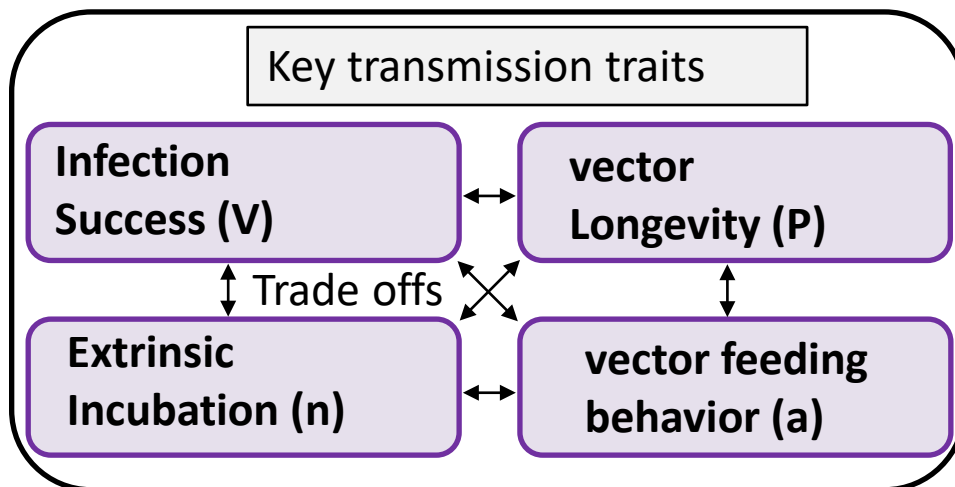
$$C = \frac{ma^2 V p^n}{-\ln(p)}$$

# Perspectives: beyond manipulation of feeding behaviour

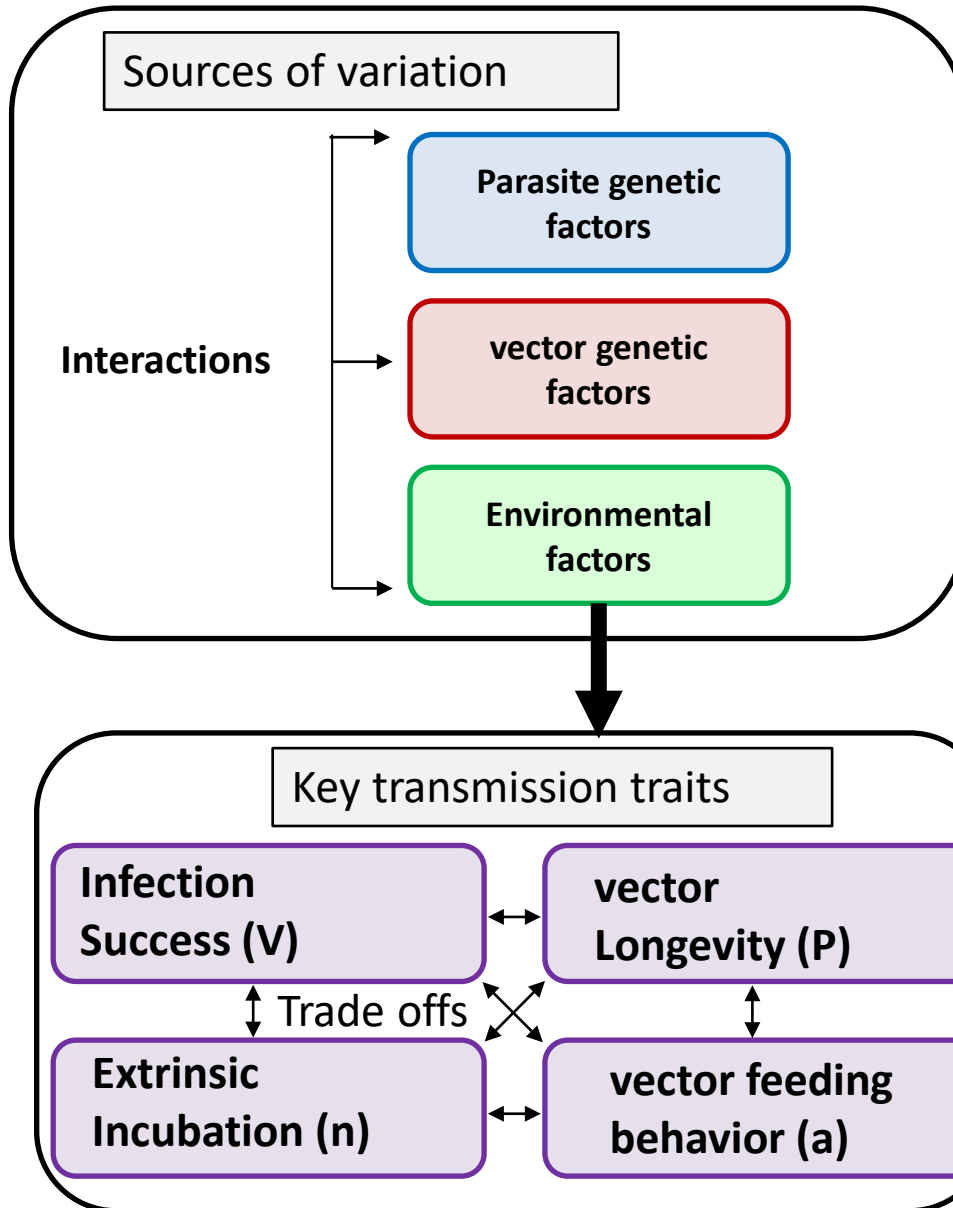
Contact rate is one determinant of the intensity of transmission intensity among others...

Vectorial capacity

$$C = \frac{ma^2Vp^n}{-\ln(p)}$$



# Perspectives: beyond manipulation of feeding behaviour



transmission intensity among

Vectorial capacity

$$C = \frac{ma^2Vp^n}{-\ln(p)}$$