

# Blackfly ecology and *O. volvulus* transmission in Uganda, Tanzania and Cameroon

ADAM HENDY (and many, many others)



# Introduction

## BLACKFLIES - LIFECYCLE

Egg



1-3 days

Larva



1 week –  
several months  
depending on  
species

Pupa



Several more  
days

Adult Male



Do not blood  
feed

Adult Female



Require blood  
meals every 3  
– 4 days for  
egg  
development

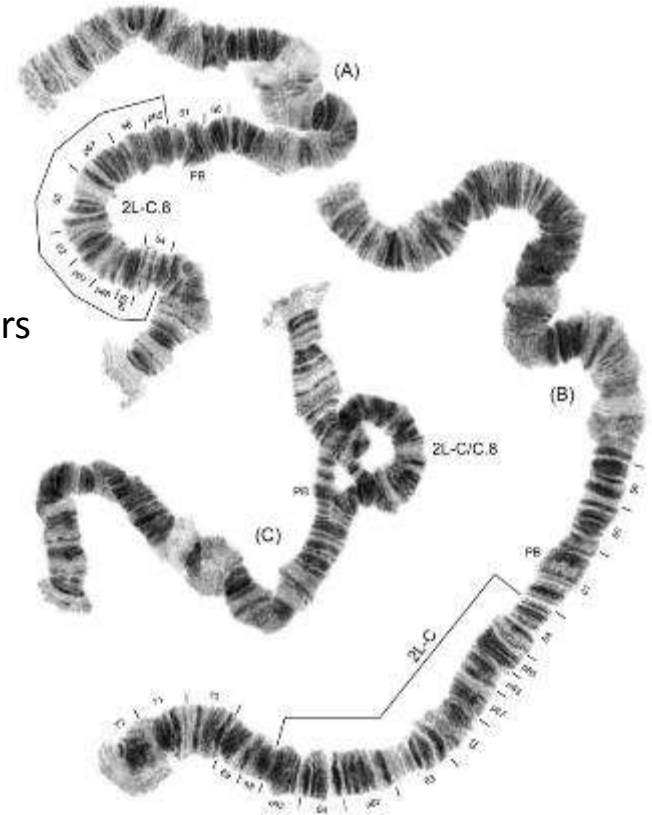
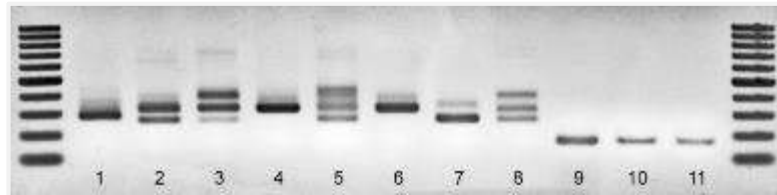
*Simulium damnosum* and *Simulium neavei*, two important vectors of *O. volvulus*.



# Background

## SPECIES COMPLEXES

- *Simulium damnosum* is a complex of species.
- All look remarkably similar.
- Differ in their biology, behaviour (some bite humans, others don't) and their ability to transmit parasites.
- Can be identified by banding patterns of giant polytene chromosomes of larvae.
- Can sometime be identified by PCR amplification of ITS1 gene.



# Background

## GENERAL METHODS

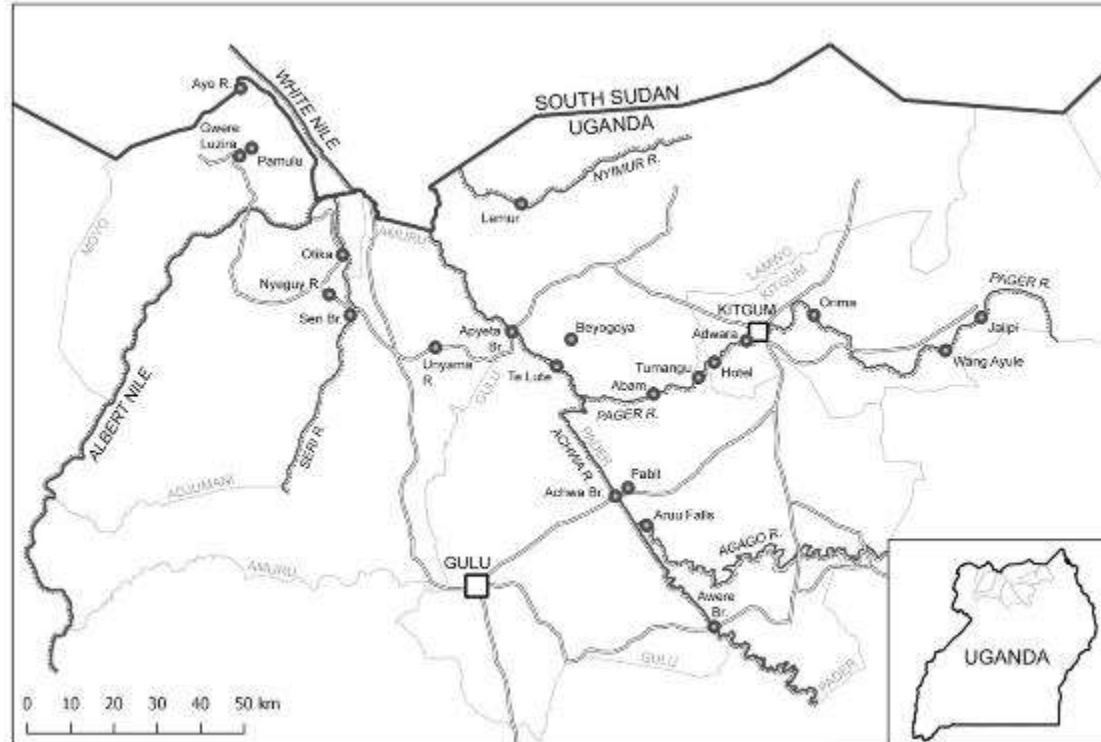
- Breeding site surveys: collecting larvae and pupae from rivers, streams etc.
- Identification by morphology, cytotaxonomy and
- Adult blackfly collections using human bait.
- Parasites identified by dissecting blackflies, or...
- Molecular identification of *Onchocerca* parasites conventional O-150 PCR or Real Time PCR/qPCR (Uni. Bonn).



# Uganda

## BACKGROUND

- Onchocerciasis was only discovered to be a problem in the Mid North focus post-war (ca. 2008).
- Control was initially through annual CDTI (2009), but switched to biannual CDTI + vector control in 2012.



# Uganda

## METHODS/RESULTS

- Breeding site surveys along major rivers and tributaries in 2012/13 (RJP) and 2014/15 (AJH).
- Adult blackfly collections using human bait in 2015/16.
- Human biting blackflies identified as: *S. damnosum* complex; *S. bovis*.
- Adult blackfly collections from cattle 2014/15.

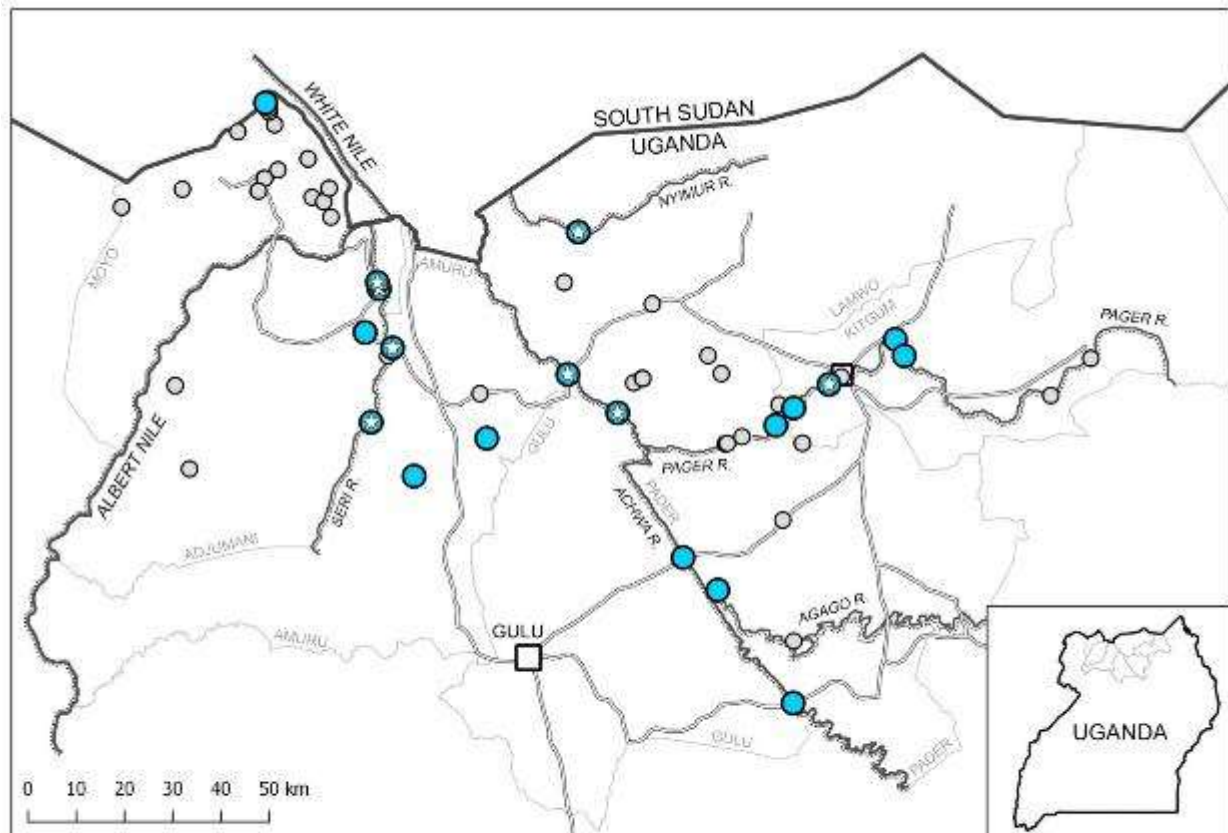
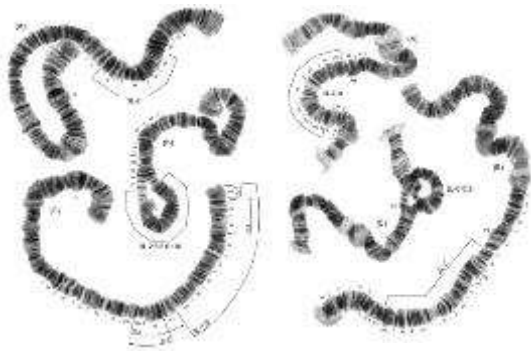


# Uganda

## RESULTS

### LARVAE & PUPAE

- *S. damnosum* complex breeding mainly in large rivers.
- Cytoforms: *S. damnosum s.str.* and *S. sirbanum*



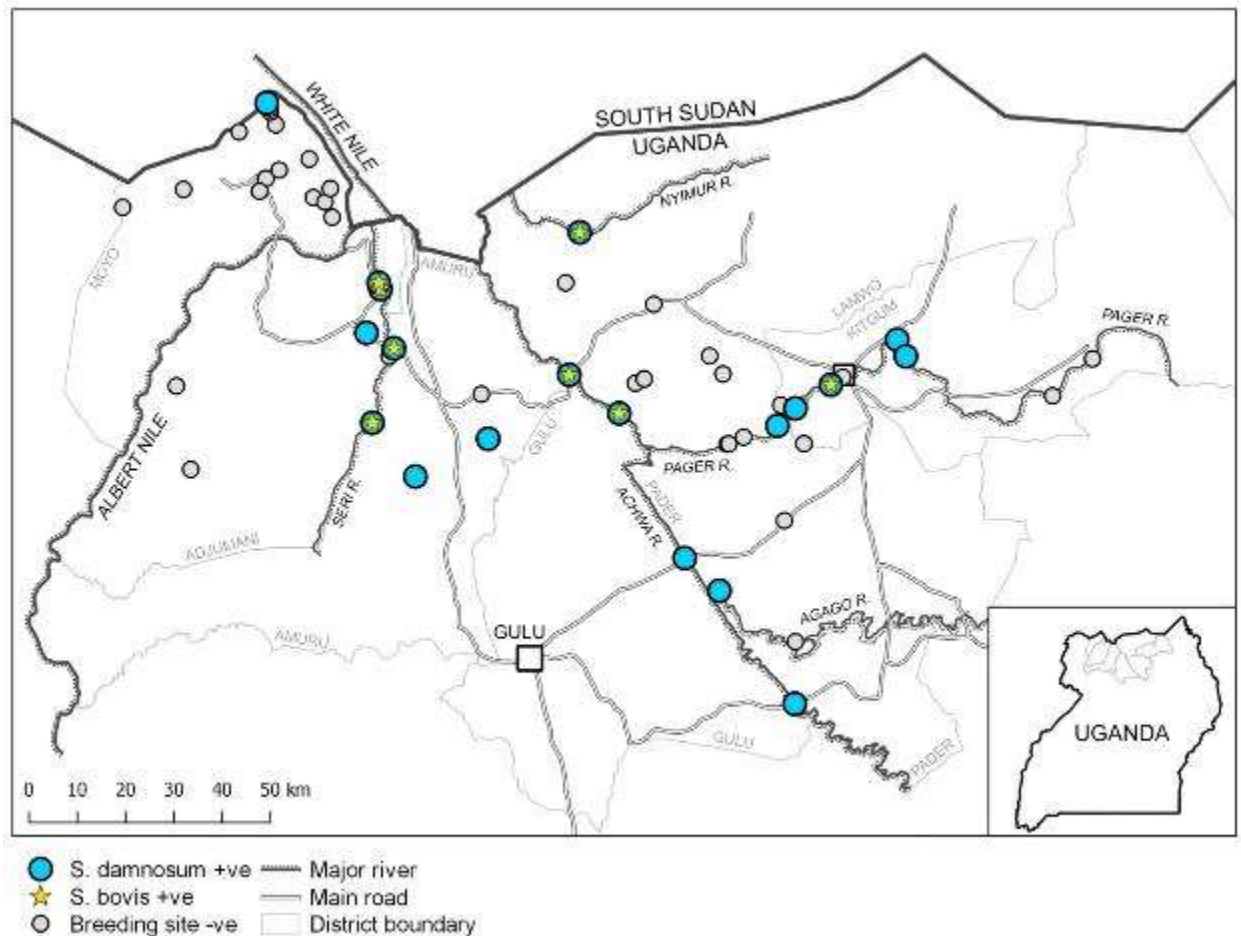
- *S. damnosum* +ve
- ☆ *S. bovis* +ve
- Breeding site -ve
- Major river
- Main road
- District boundary

# Uganda

## RESULTS

### LARVAE & PUPAE

- *S. bovis* occupying similar breeding sites along the Pager, Achwa, Nyimur and Seri rivers.



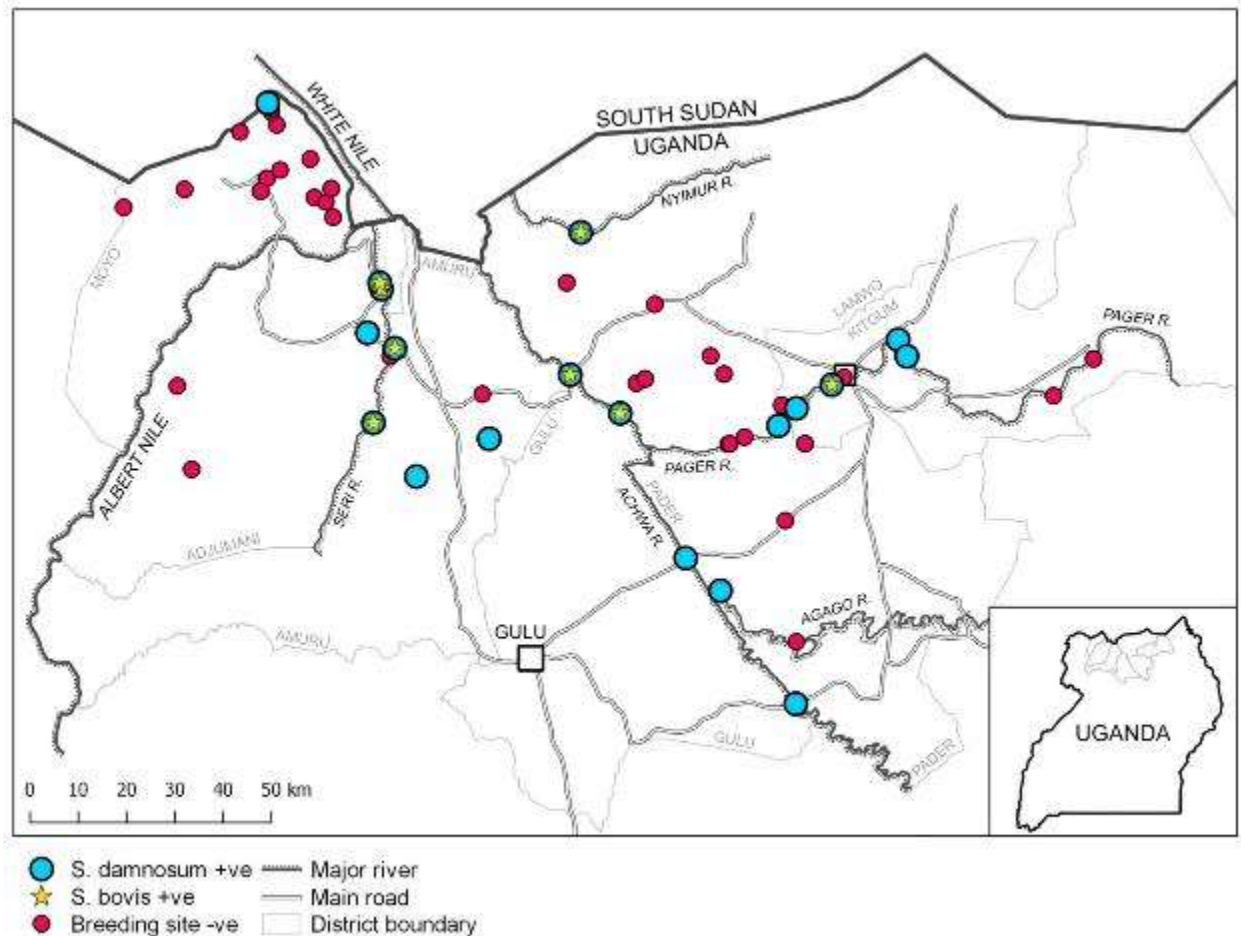


# Uganda

## RESULTS

### LARVAE & PUPAE

- Very little breeding (if any) in the smaller rivers/tributaries.

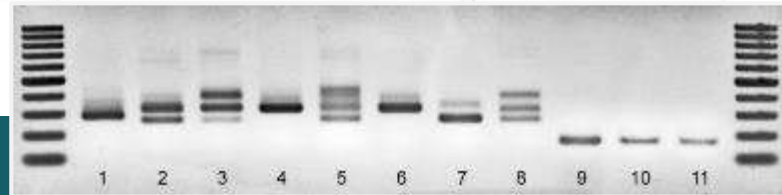
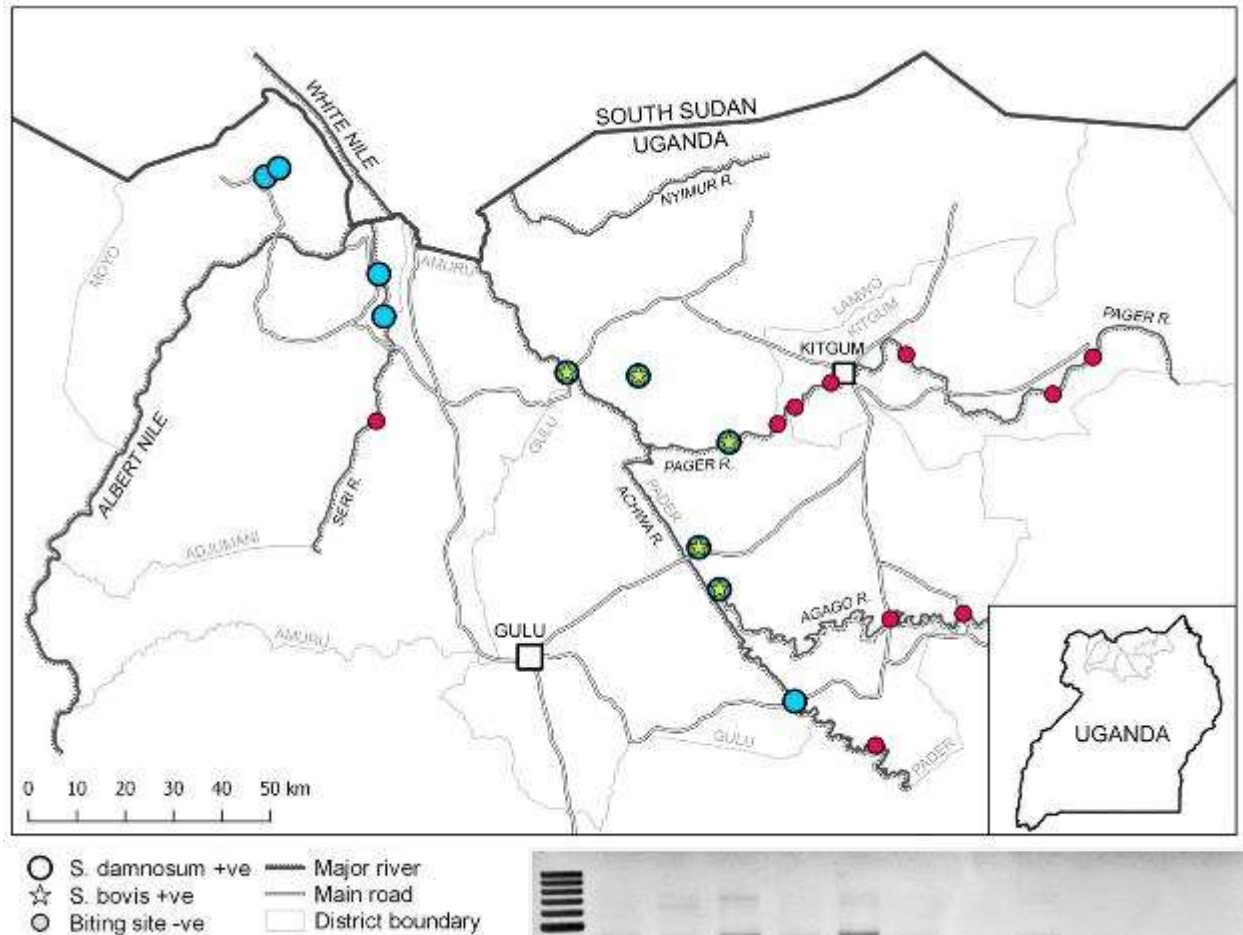


# Uganda

## RESULTS

### ADULT FLIES

- **4,807** *S. damnosum* s.l. collected in a combined 79 days.
- **4,674** (97.2%) in Moyo.
- **133** (2.8%) elsewhere.
- **772** *S. bovis* collected on human bait in Kitgum, Lamwo and Pader.
- Both species biting humans and cattle.

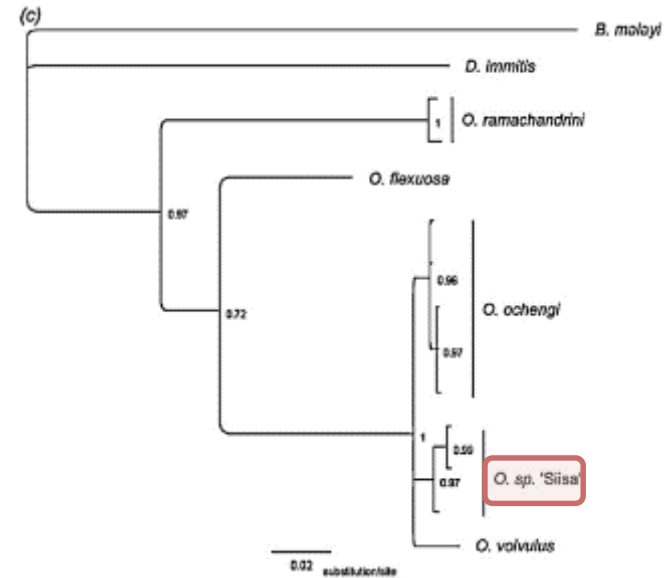


# Uganda

## RESULTS

### POOL SCREENING

- **No** detection of *O. volvulus* in *S. damnosum* s.l. from Kitgum, Lamwo and Pader.
- ***Onchocerca ochengi*** and ***Onchocerca* sp. 'Siisa'** (both bovine parasites) present in human biting *S. damnosum* and *S. bovis*.
- 'Siisa' is closely related to both *O. volvulus* and *O. ochengi*.
- Caused some problems w/ identification using conventional O-150 PCR (resolved through qPCR and sequencing).



Renz *et al.* 2013



# Tanzania

## BACKGROUND ...

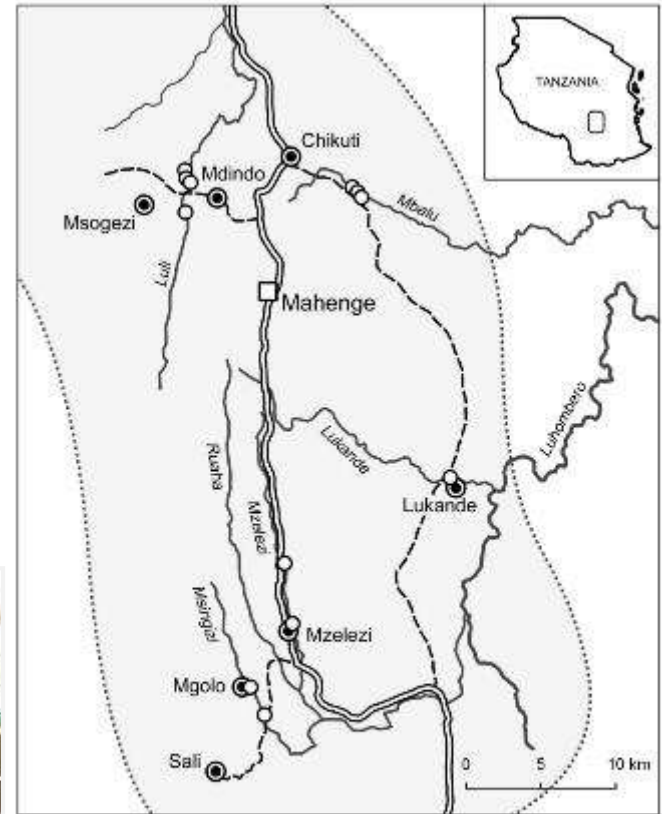
- Mahenge onchocerciasis focus in SE Tanzania has been little studied entomologically.
- Although W. Häusermann spent a year studying blackflies, *O. volvulus*, and onchocerciasis in the 1960's.
- His are the only pre-control (annual CDTI since 1997) entomological data documenting parasite transmission in the area.
- By dissecting >12,000 *S. damnosum* s.l., he found that 0.68% contained infective L3 stage parasites in their heads



# Tanzania

## METHODS

- Breeding site surveys in January 2015 and June 2016 .
- Adult blackfly collections made at seven sites throughout June 2016 (time of peak transmission).



- |   |                                          |       |                 |
|---|------------------------------------------|-------|-----------------|
| ● | <i>S. damnosum</i> adult collection site | - - - | District road   |
| ○ | <i>S. damnosum</i> breeding site         | —     | River           |
| — | Main road                                | ⋯     | Extent of Focus |

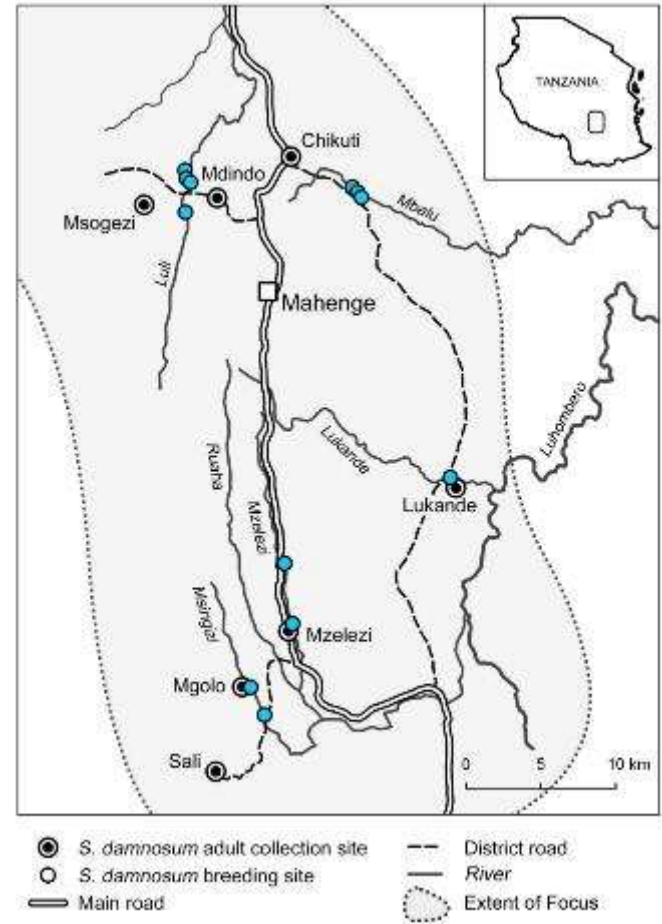
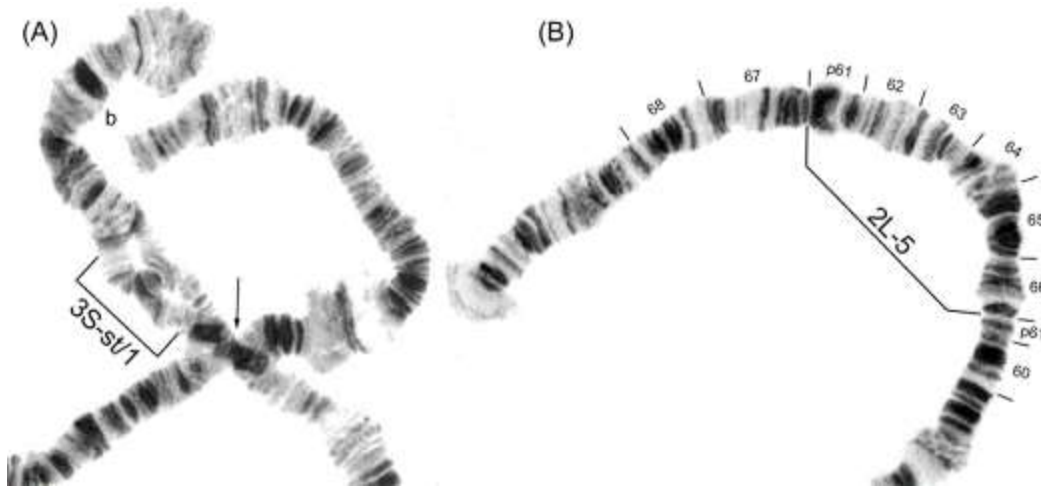


# Tanzania

## RESULTS

### LARVAE & PUPAE

- *S. damnosum* s.l. collected breeding in Luli, Mbalu, Lukande, Mzelezi and Msingizi rivers.
- *S. damnosum* complex: 'Nkusi J', *S. kilibanum* and 'Turiani' breeding sympatrically.



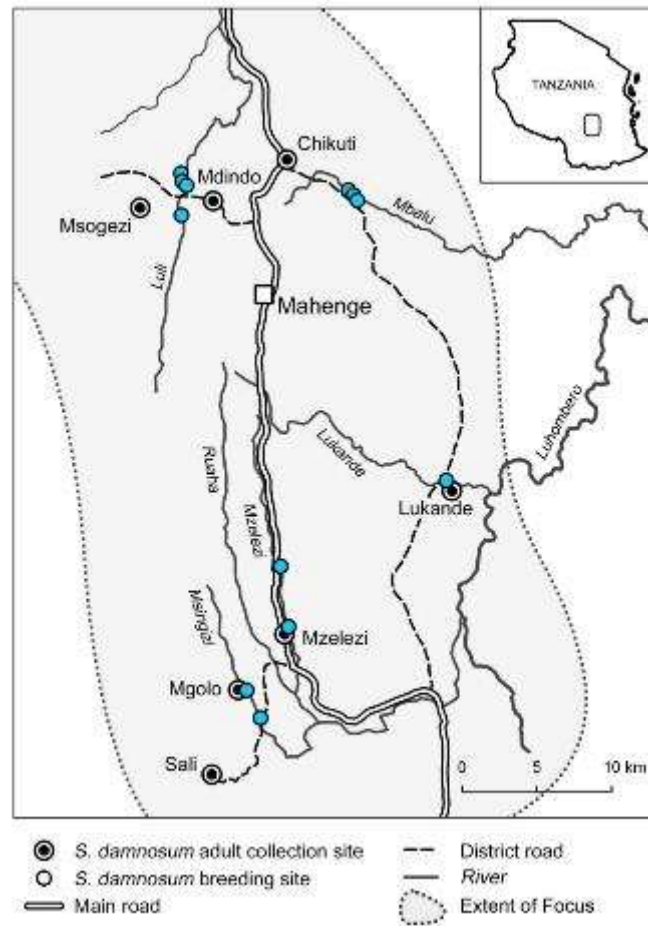
# Tanzania

## RESULTS

### ADULT FLIES & POOL SCREENING

- 16,943 adult blackflies collected.
- 99.8% of human biting blackflies were *S. damnosum* s.l., 0.2% were *S. nyalalandicum*.

Location	Alt.	No. Days	Total Catch	No. Pooled	No. Pools*	<i>O. volvulus</i> +ve		Infection Rate (Heads)			
						Bodies	Heads	L3**	95% CI -/+		
Msogezi	603m	17	4273	2056	16	11	6	0.37%	0.13%	0.83%	
Mdindo	548m	17	4157	3210	25	25	15	0.72%	0.38%	1.26%	
Chikuti	459m	17	3001	2681	27	27	8	0.36%	0.14%	0.72%	
Mgolo	465m	17	2589	2164	15	15	7	0.43%	0.16%	0.92%	
Mzelezi	333m	17	1812	1423	11	11	6	0.62%	0.21%	1.43%	
Sali	876m	12	672	614	6	6	5	1.65%	0.46%	4.23%	
Lukande	355m	12	407	304	4	2	4	-	-	-	
			Total	16943	12452	104	97	51	0.57%	0.43%	0.74%



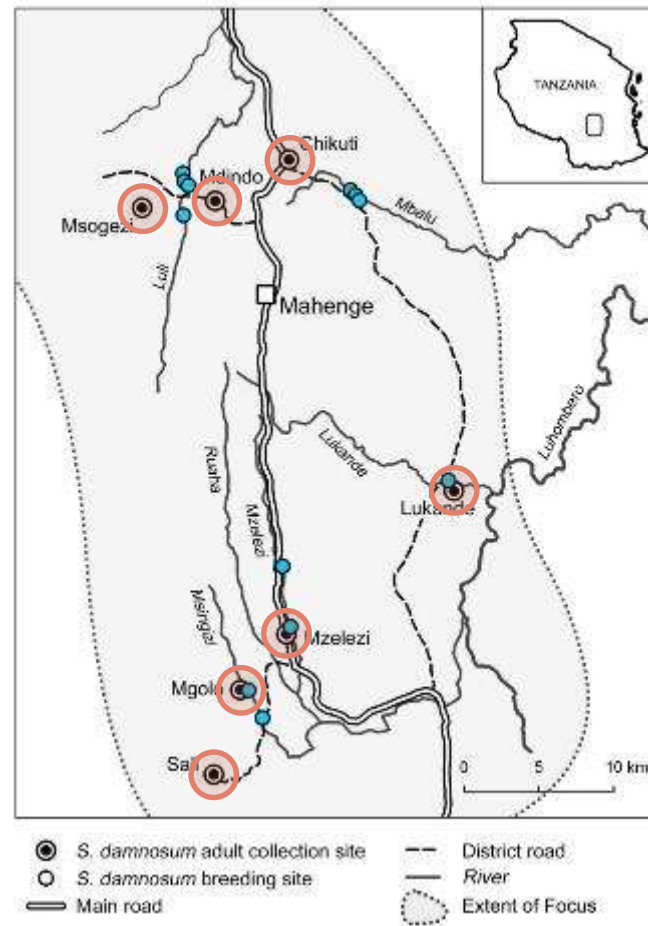
# Tanzania

## RESULTS

### ADULT FLIES & POOL SCREENING

- *O. volvulus* present in *S. damnosum* s.l. at all adult sites.
- 97/104 pools of bodies and 51/104 pools of heads were PCR positive.

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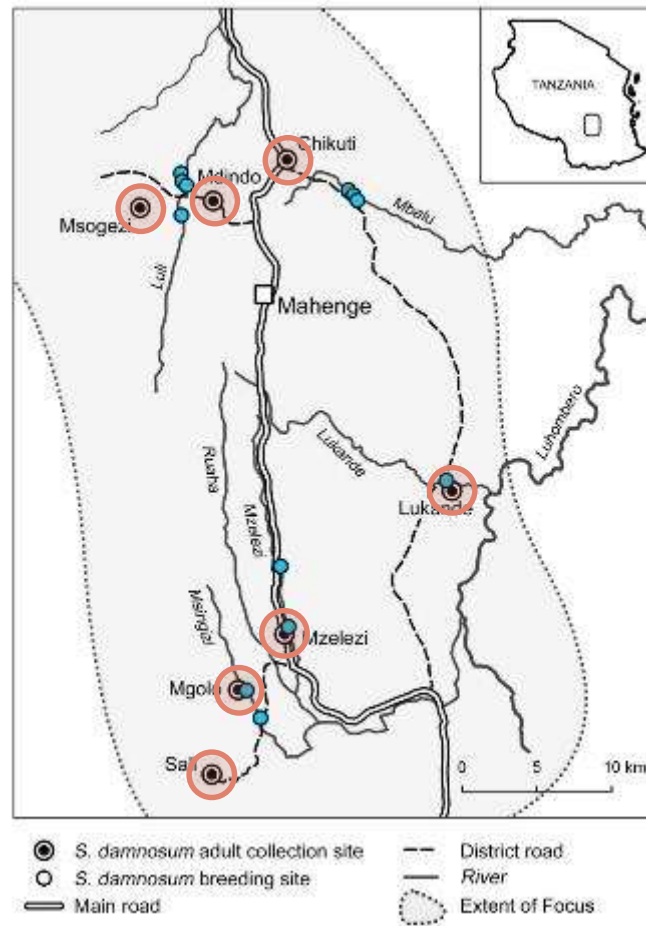
# Tanzania

## RESULTS

### ADULT FLIES & POOL SCREENING

- An estimated **0.57%** (95%CI: 0.43% - 0.74%) of *S. damnosum* s.l. possessed infective L3 stage parasites in their heads.

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			Total	16943	12452	104	97	<b>0.57%</b>	<b>0.43%</b>	<b>0.74%</b>



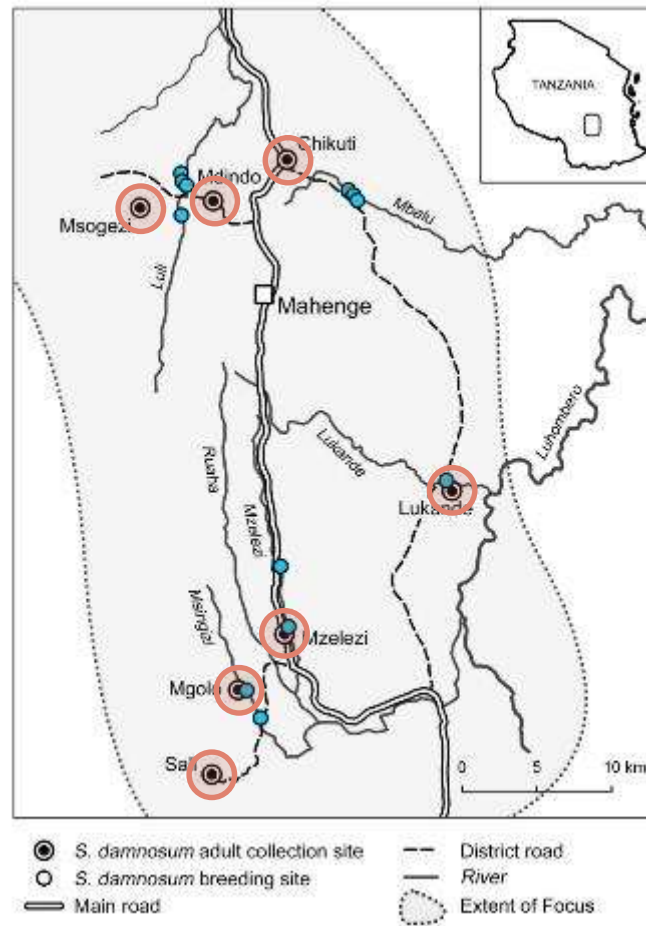
# Tanzania

## RESULTS

### ADULT FLIES & POOL SCREENING

- Similar to Häusermann's infection rate of **0.68%**.
- Transmission is ongoing.

Location	Alt.	No. Days	Total Catch	No. Pooled	No. Pools*	<i>O. volvulus</i> +ve		Infection Rate (Heads)			
						Bodies	Heads	L3**	95% CI -/+		
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			Total	16943	12452	104	97	51	<b>0.57%</b>	<b>0.43%</b>	<b>0.74%</b>



# Cameroon

## BACKGROUND & OBJECTIVES

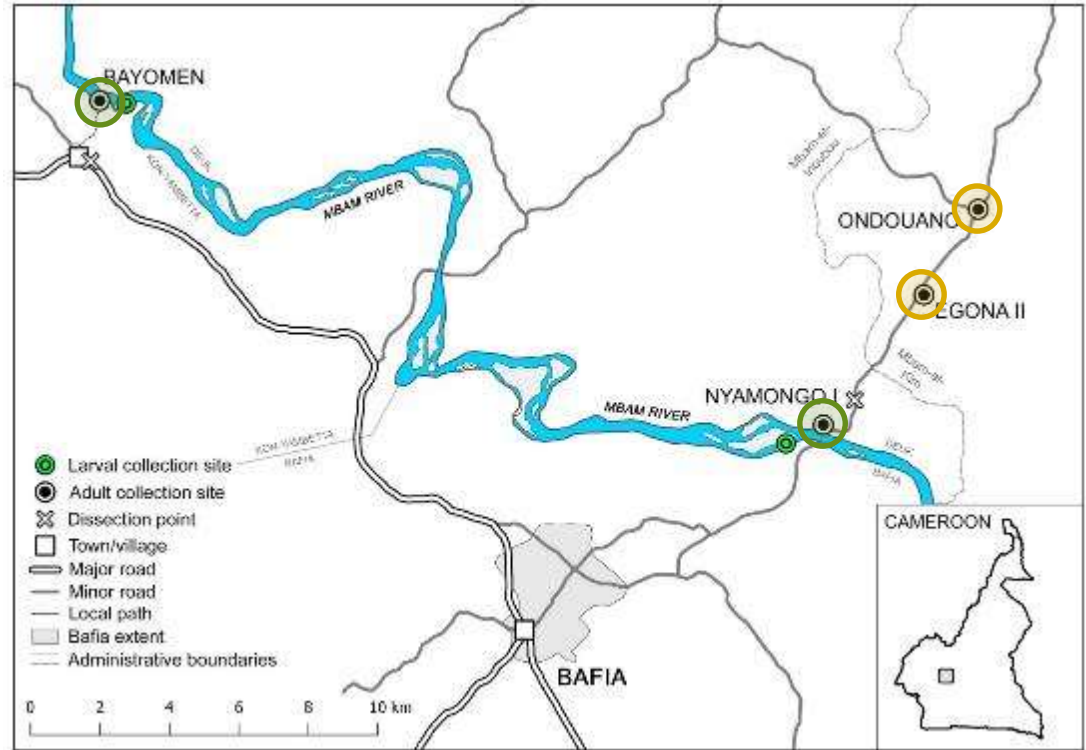
- Onchocerciasis in villages along the Mbam River near Bafia historically associated with severe pathologies.
- Annual CDTI has been ongoing since 2000.
- A 12-month entomological and study of parasite transmission was conducted in 1993/94, prior to MDA with ivermectin (Barbazan *et al.* 1998).
- We **repeated** a similar study in 2016/17.



# Cameroon

## METHODS

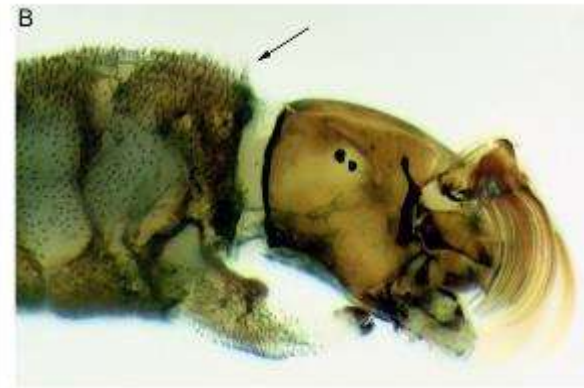
- Four collection sites, two riverside, two further away.
- Collections made for 3 consecutive days per month, for 12-months between July 2016 – June 2017.
- Dissected a proportion of catch to determine parity rates and *O. volvulus* infection rates.
- Preserved the remaining material for pool screen analysis (ongoing).



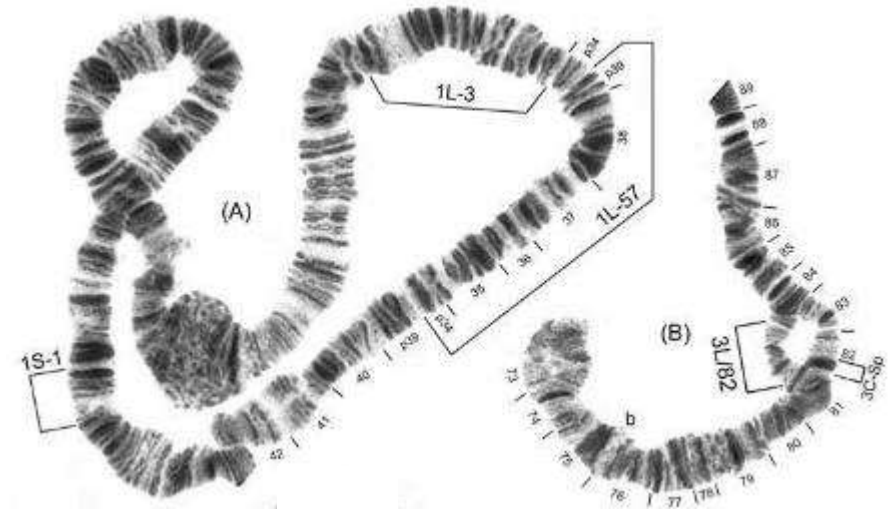
# Cameroon

## RESULTS

### LARVAE & PUPAE



- *S. damnosum* complex: *S. mengense* & a chromosomal variant of *S. squamosum* E (not usually found in Cameroon)
- *S. mengense* appears not to be biting, so presumably *S. squamosum* E2 is (but needs confirming – collections were very limited)

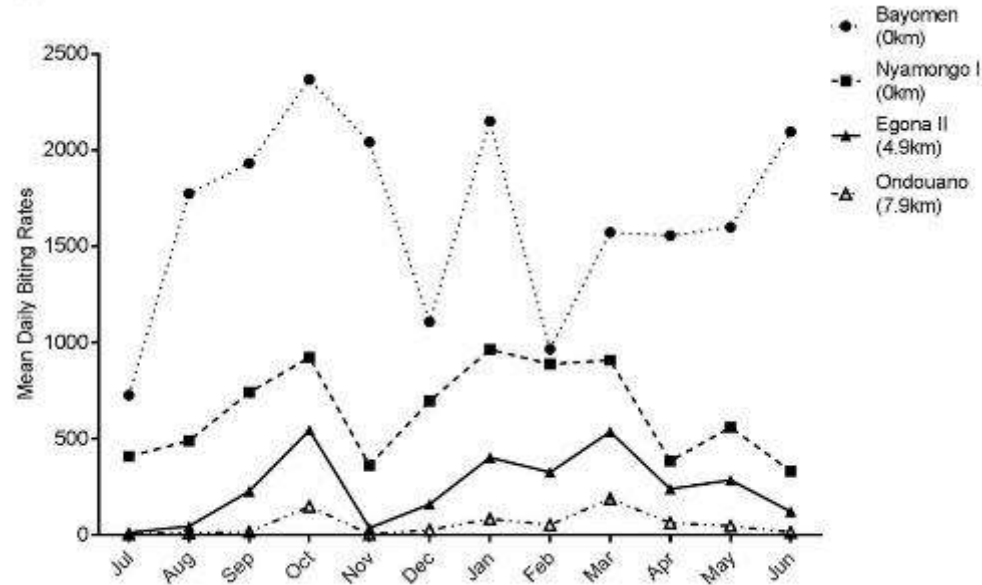


# Cameroon

## RESULTS

### ADULT FLIES – BITING RATES

- Biting rates highest at the two riverside sites.
- Biting rates are more or less consistently high at Bayomen (ABR >600,000).
- A shorter biting peak around October at Nyamongo I – Ondouano.
- A longer peak in biting between January and March.
- Majority of biting is towards the end of the long dry season.

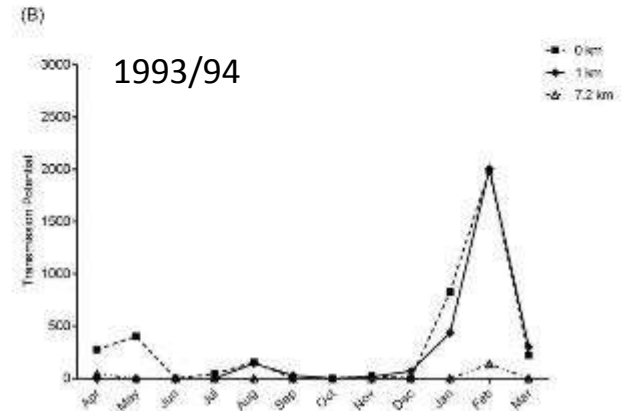
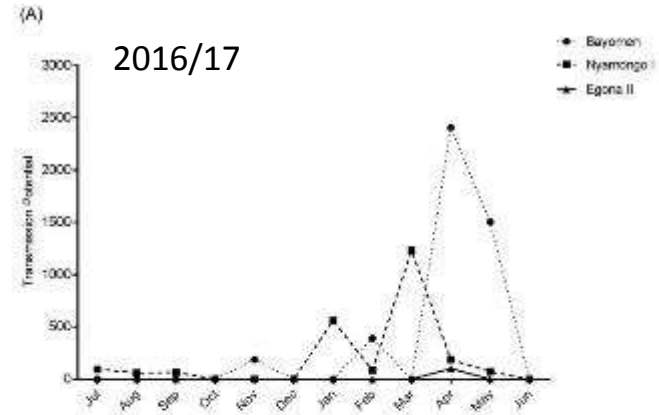


# Cameroon

## RESULTS

### ADULT FLIES – *O. VOLVULUS* TRANSMISSION

- 93,563 flies collected.
- 9,281 dissected.
- Remaining flies being pool screened.
- Preliminary transmission data based on dissections.
- Highest annual transmission potentials at riverside: Bayomen (ATP **4,488**), Nyamongo I (ATP **2,360**).
- Low transmission at Egona 2 (ATP **104**) and Ondouano (ATP **0**)
- Infection rates are relatively low (Bayomen: L3H = 0.13%, Nyamongo I = 0.36%), but very high biting rates!!!







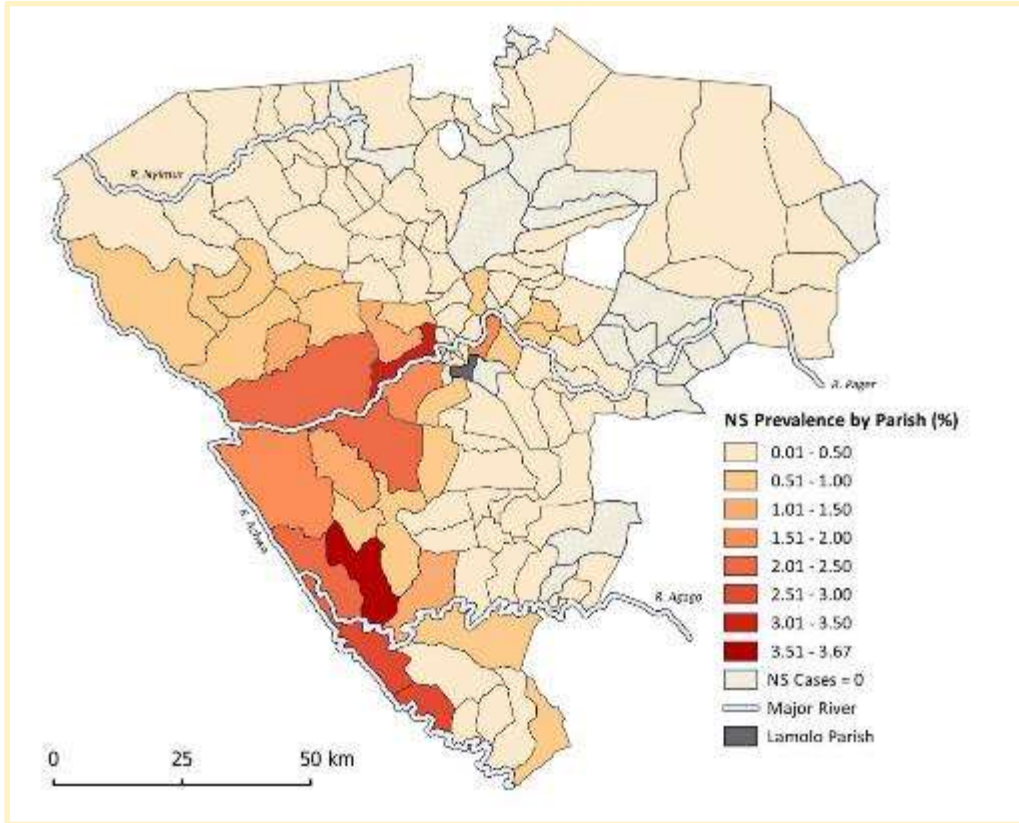


**INSTITUTE  
OF TROPICAL  
MEDICINE  
ANTWERP**

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- **NS Case** data from MoH (2012)
- **Denominator:** UBOS population projections based on 2002 census.
- Populations have been displaced/resettled in that time. How **reliable** are the data?
- Currently the *best available* data (more recent data now available).
- **Lamolo parish** an outlier. Pop. = 1500 (UBOS), NS cases = 207 (13.8% prevalence).