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The role of tabanids as potential African swine fever vectors in Estonian wild boar habitats: preliminary results

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BACKGROUND

Trace amounts of African swine fever virus (ASFV) DNA have been detected in insects collected in 2016 from an infected pig farm in Estonia (¹) and in 2019, in several farms in Lithuania (²). It has been experimentally demonstrated that stable flies (*Stomoxys calcitrans*) can transmit ASFV within a short period of time $(^3)$.

Tabanids are haematophagous flies and mechanical vectors for many infectious diseases, they should be considered as potential mechanical vectors for ASFV.

AIM OF THIS STUDY - to determine via DNA analysis which tabanid species feed on the wild boar (Sus scrofa)

MATERIAL AND METHODS

- Location: Wild boar baiting sites in Emmaste, Hiiumaa, Estonia
- 2 collection years: 2019 and 2020
- 4 collection periods in June, July and August
- 4 collection sites
- 4 trapping methods
- Infrared cameras to confirm wild boar in the area
- Storage: 75% ethanol; net caught tabanids stored dry in -20°C.
- All specimens morphologically identified to genus or species level.
- Homogenisation: individual or pooled (max no. 5 specimens) samples



Figure 2. Tabanid collection sites in Hiiumaa, Estonia, 2019 and 2020.

Figure 3. Trap placement in collection site 1. Nzi trap in the foreground, H-trap in the far right.



- **DNA samples analysed by real-time PCR** for the presence of suid-specific cytochrome b (cyt-b) gene
- Fwd. primer 5'-CCTCGCAGCCGTACATCTC-3'
- Rev. primer 5'-ACGTAGAATAGCGTAGGCGAATAA-3'

2019

One H-trap and one water tray per site

2020

- One H-trap + one Nzi-trap site.
- Entomological net catches by 2 persons, per 20 minutes once a day
- All traps urine-baited



Probe 5' FAM-TCCTGCACGAAACCGGATCCAACAAC-BHQ-1 3' (⁴)

Figure 4. Female Tabanus bromius specimens. Figure 5. DNA samples to be analysed by real-time PCR (RotorGene Q (Qiagen, Germany)).



Figure 6. Total number of tabanids per trapping method and year. H-traps and water trays were utilised in 2019; H-traps, Nzi-traps and net catches in 2020.

50 K2 K1 КЗ K4 Trapping site 2019 2020

No. of tabanids per trapping site and

year

Figure 7. Total number of tabanids by trapping site and year. In 2020, site K3 was switched to site 4 after one day of collection due to no evidence of wild boar in the area.

PRELIMINARY RESULTS AND CONCLUSIONS

753 individual tabanids were captured over the study period.

Table 1. Tabanid species captured from three locations near wild boar baiting

sites in Estonia, 2019 and 2020, and cytochrome b PCR analysis results.

		2019	2020	Total (% total)	No. tested for <i>cytb</i> (no. pos)
Genus	Species				
Atylotus	A. fulvus	1	-	1 (0.1%)	1 (0)
	A. plebeius	2	-	2 (0.3%)	2 (0)
Haematopota	H. crassicornis	-	4	4 (0.5%)	4 (0)
	H. pluvialis	166	553	719 (95.5%)	719 (0)
Heptatoma	H. pellucens	1	-	1 (0.1%)	1 (0)
Hybomitra	Hybomitra spp.	13	-	13 (1.7%)	13 (0)
Tabanus	T. bovinus	-	10	10 (1.3%)	10 (0)
	T. bromius	-	1	1 (0.1%)	1 (0)
	T. maculicornis	-	2	2 (0.3%)	2 (0)
		183	570	753	753 (0)

- 96% of all tabanids caught belonged to genus *Haematopota*.
- The most effective way of collection was entomological net, yielding • 496 tabanids in total (65.9% out of all tabanids caught in all periods) over 6 collection days.

350

300

None of the tabanids tested positive for wild boar contact, suggesting that tabanids don't play a significant role as wild boar pests and therefore as potential mechanical vectors for African swine fever.



The publication of this poster is supported by the European Union, European Regional Development Fund (Estonian University of Life Sciences ASTRA project "Value-chain based bioeconomy"

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A WORK IN PROGRESS: 2021

-- indicates no individuals of the species being caught

- Wild boar baiting sites: 6 sites in Tartumaa, Estonia; collection period 6 weeks.
- Commercial pig farms: 2 farms in Tartumaa, collection period 6 weeks: collection and identification of tabanids to determine their species diversity and relative abundance.
- All captured tabanids to be tested for wild boar/ pig contact by *cyt-b* PCR.