



Fig. 1: Schematic drawing attract-and-kill.

MATERIALS & METHODS

The **y-olfactometer** assay is an in-house development to investigate the level of attraction of *Ixodes ricinus* nymphs (Fig.2).

Test conditions: 3 min test time, flow of 47.8 ml/min, 77-80% RH; 20-24°C, 520-650 ppm room CO_2 level.

<u>Behaviours:</u> left / right decision after 10 cm distance, walk out, not moving.



Fig. 2: Y-olfactometer with *I. ricinus* nymphs.

REFERENCE

Humbert, P., Vemmer, M., Giampà, M., Bednarz, H., Niehaus, K. & Patel, A. V. (2017). Co-encapsulation of amyloglucosidase with starch and Saccharomyces *cerevisiae* as basis for a long-lasting CO₂ release. World Journal of Microbiology and Biotechnology, 33(4), 71.



In Fig. 3 we show a significantly attractive effect of CO_2 on *I. ricinus*. 24-28% of nymphs decide did not between test and control side. Side preferences were excluded.

The screening of 39 compounds of different classes in regards to their level of attraction of *I. ricinus* nymphs to date demonstrates only significantly repellent effects on *I. ricinus*.

[%] cisio 50%

* P ≤ 0.05 15-41 ticks

DEVELOPMENT OF A BIOLOGICAL TICK TRAP BASED ON ATTRACT-AND-KILL STRATEGY

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RESULTS & DISCUSSION

ATTRACTIVE COMPOUNDS



Fig. 3: Choice of *I. ricinus* in y-olfactometer assays tested against CO_2 .



AGGREGATION COMPOUNDS

Beside the attraction also the arrest of the ticks on the capsules is important. Therefore known aggregation compounds were screened.



*** P ≤ 0.001 ** P ≤ 0.01 * P ≤ 0.05 ± SD



Fig. 5: Choice of *I. ricinus* nymphs in a static four-chamber olfactometer.

For the first time we could demonstrate aggregation pheromones of the classes of purines and other substances for *I. ricinus* nymphs (Fig. 5).

Combinations tested so far did not show any synergistic effect. Compounds will be further evaluated to show if they are suitable for microporation into the capsules and if they increase contact time between tick and capsules.



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MATERIALS & METHODS

refined four-chamber static olfactometer was used for sreening of potential aggregation compounds (Fig. 6).

Test conditions: One test container included 4 stripes filter paper of 3.75 cm² (1 treated with test compound, 3 control), 3 h observation time, 20 ticks per test run, 20°C, 90% RH.



Control filter paper

Test filter paper Gauze

Walking arena

Fig. 6: Static four-chamber olfactometer with *I. ricinus* nymphs.

CONCLUSIONS

We go further with the screening of attractive compounds for *I. ricinus*. Arresting and attracting substances combined will be coupled with the entomopathogenic fungus as kill component. Such a trap could serve protection against ticks in areas frequently used by humans.

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