

# Pheromones and other semiochemicals of click beetles - a European perspective

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Wireworms, the larvae of click beetles (Coleoptera: Elateridae) are important soil-dwelling polyphagous pests all over the world.



Traditional forecast and monitoring involves labour-intensive soil sampling methods,



Photo L. Furlan



and to obtain wireworms from soil samples collected is time-consuming  
(several days or more).

Photo L. Furlan

Pheromone-baited traps are much easier and simpler to use.

However, the pheromone composition should be identified first!



Photo M. Tóth

On the picture: the YATLOR FUNNEL (or YF ) trap design specifically developed for pheromone trapping of click beetles (Furlan, Inform. Fitopat. 10:49, 2004)

# Pheromone structures - first identifications

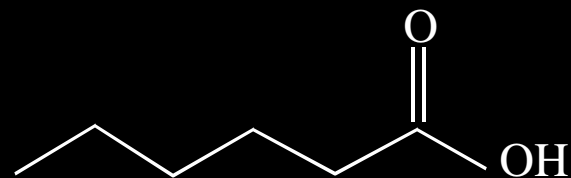
The very first chemical structures elucidated from click beetles (female-produced pheromone) came from North America and were organic acids



valeric acid (pentanoic acid)

*Limonius californicus*

(Jacobson, Science 159:208, 1968)



caproic acid (hexanoic acid)

*Limonius canus*

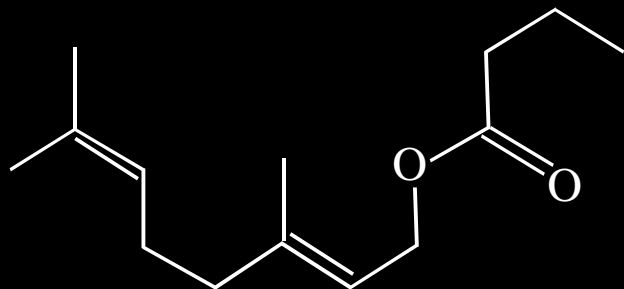
(Butler, Environ. Entomol. 4:229, 1975)

*Limonius californicus*  
www.bugguide.net

# Pheromone structures - first identifications

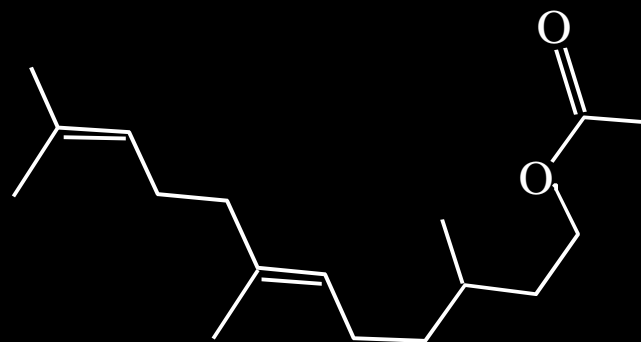
In Europe, starting from the eighties, a number of geranyl and farnesyl esters were identified mainly by scientists from the Soviet Union.

Example structures:



geranyl butyrate  
[(*E*)-3,7)-dimethyl-2,6-  
octadienyl butyrate]

i.e. *A. sputator*



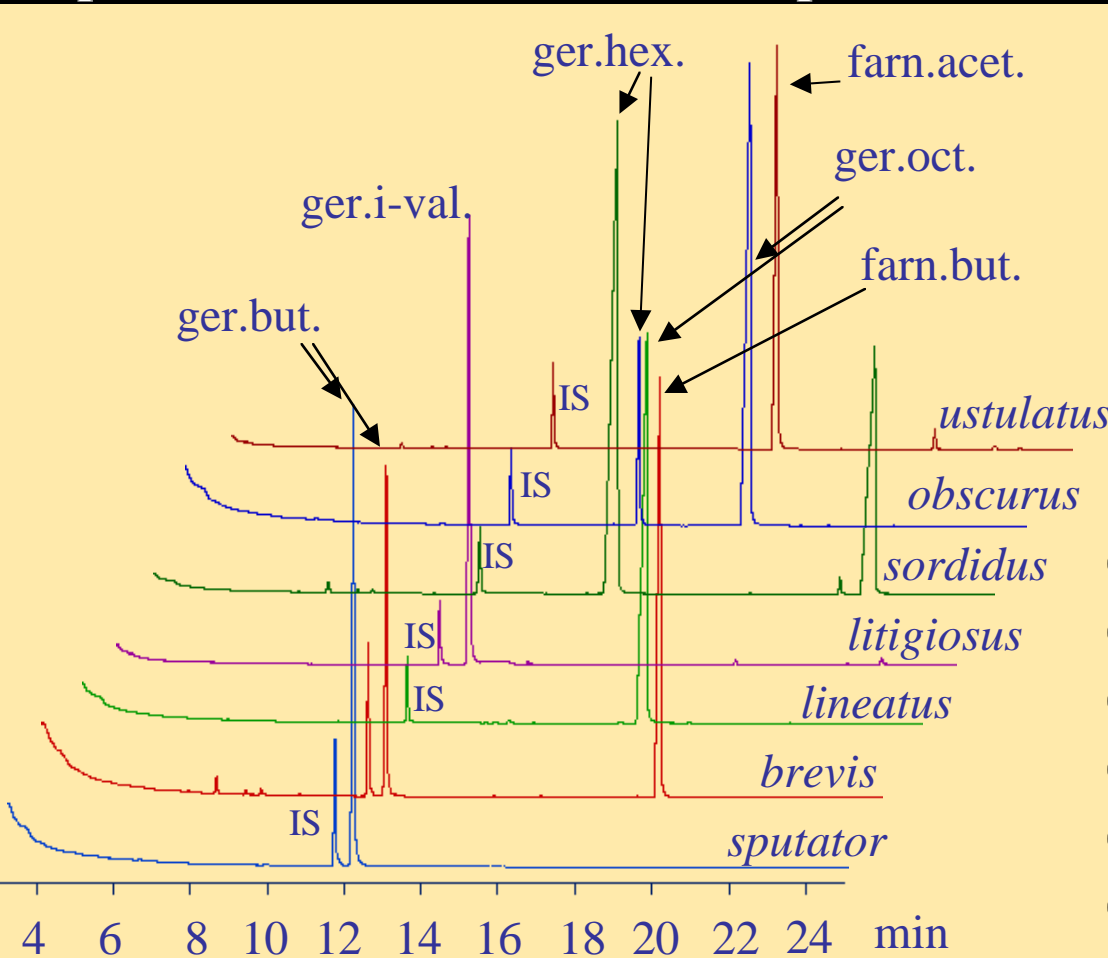
(*E,E*)-farnesyl acetate  
[(*E*)-3,7,11)-trimethyl-2,6,10-  
dodecatrienyl acetate]

i.e. *A. ustulatus*

First report on similar structures from: Oleschenko, 1979 (cited in Kamm, Coleopt. Bull. 37:16, 1983)

# Pheromone structures - my lab's contribution

In the 1990-s we identified new and revisited known (but inactive) pheromones of elaterids (important in Central and Western Europe)



- first identification
- improved published composition
- confirmed known composition

- *A. ustulatus*
- *A. obscurus*
- *A. sordidus*
- *A. litigiosus*
- *A. lineatus*
- *A. brevis*
- *A. sputator*
- *A. rufipalpis*
- *A. proximus*

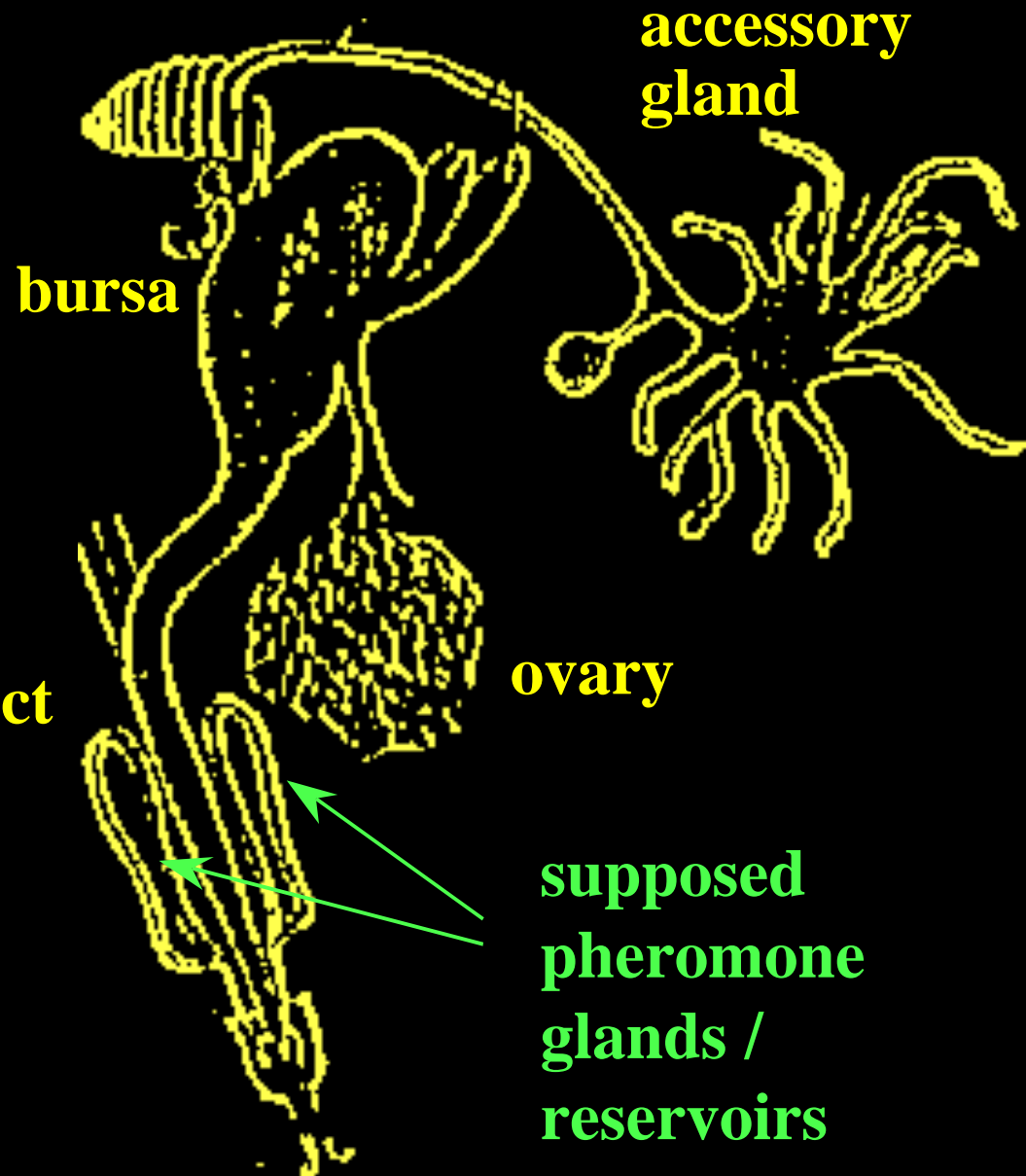
GC analysis of pheromone gland extracts of *Agriotes* spp.

(after Tóth, J. Chem. Ecol., 28:1641, 2002, and Pest Manag. Sci. 59:417, 2003)



# Pheromone structures - my lab's contribution

One reason for the relatively frequent necessity of revisiting published structures may be that most early identifications were based on direct gland extracts, which may not necessarily represent composition emitted into the air by the females.

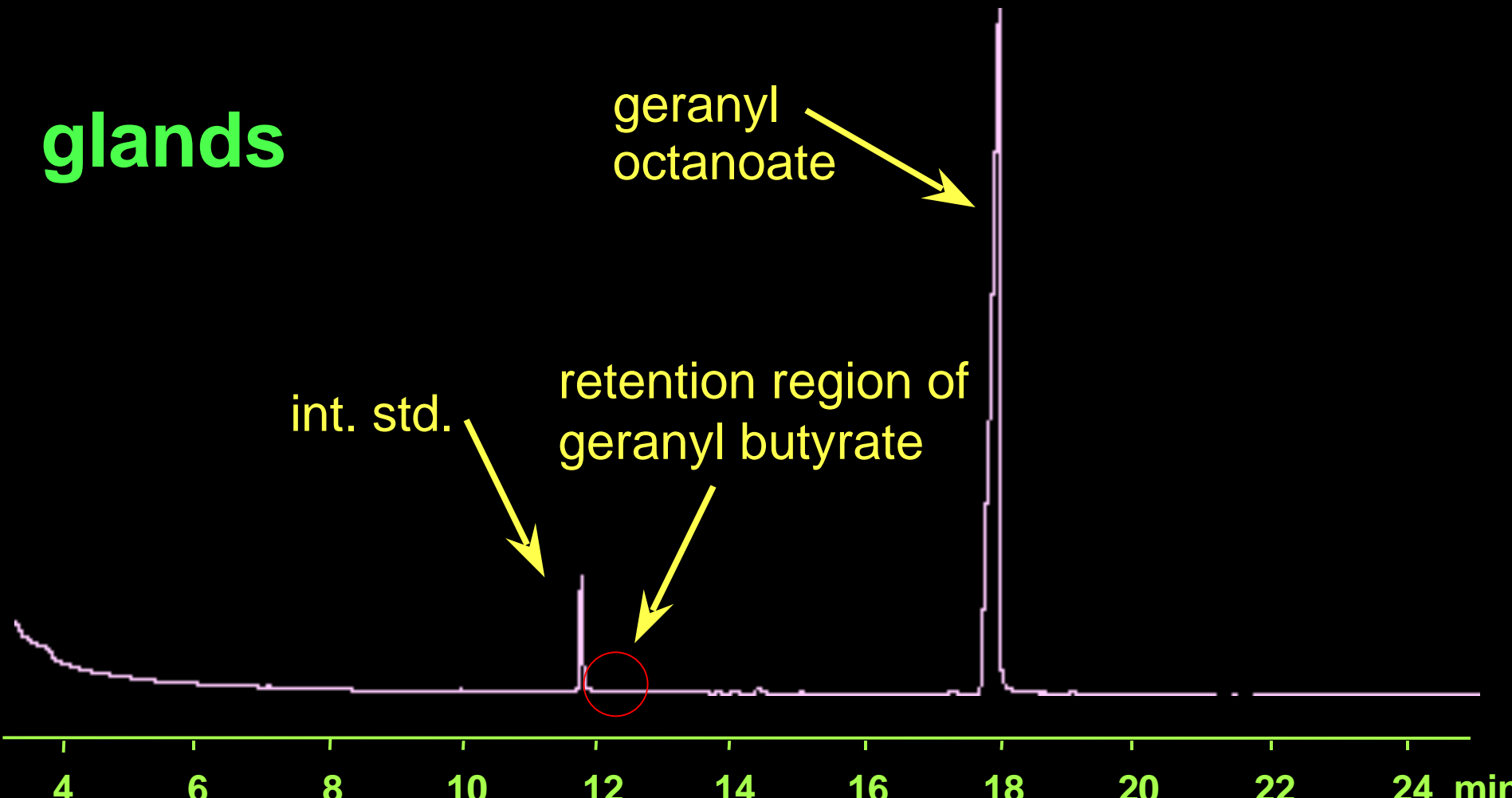


(After Ivaschenko, Zool. Zh. 59:225, 1980).

# Gland extract vs. volatile collection (example No. 1)

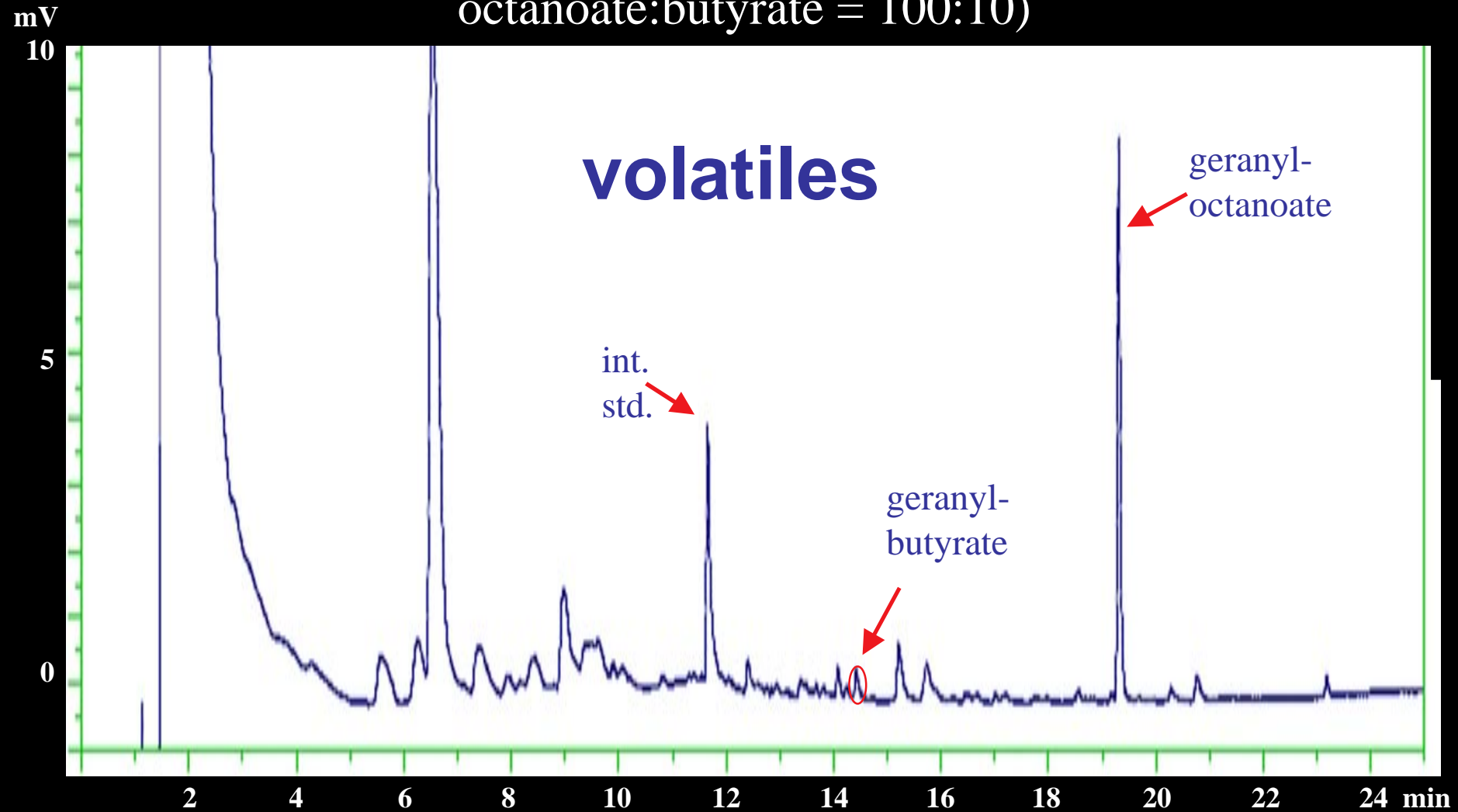
Gland extracts of *A. lineatus* were dominated by a single very large peak of geranyl octanoate, geranyl butyrate was hardly detectable (ratio of octanoate:butyrate = 100:<0.01)

**glands**



# Gland extract vs. volatile collection (example No. 1)

In volatile collections the butyrate was well visible (ratio of octanoate:butyrate = 100:10)



(After Vuts, Chemoecol. 22:23, 2012)

# Gland extract vs. volatile collection (example No. 1)

In the field the presence of the butyrate was absolutely necessary for attraction.

Total caught  
in test:  
**279**  
beetles

Rümlang,  
May 23 -  
August 14,  
1997

|  | Geranyl<br>octanoate | Geranyl<br>butanoate | E,E-farnesyl<br>acetate | Geraniol | Geranyl<br>hexanoate | Neryl<br>isovalerate | <i>A.lineatus</i> |
|--|----------------------|----------------------|-------------------------|----------|----------------------|----------------------|-------------------|
|  | 10                   | .                    | .                       | .        | .                    | .                    | 0                 |
|  | 30                   | .                    | .                       | .        | .                    | .                    | 2                 |
|  | 100                  | .                    | .                       | .        | .                    | .                    | 0                 |
|  | 10                   | <b>1</b>             | .                       | .        | .                    | .                    | <b>273</b>        |
|  | 10                   | .                    | <b>1</b>                | .        | .                    | .                    | 2                 |
|  | 10                   | .                    | .                       | <b>1</b> | .                    | .                    | 1                 |
|  | 10                   | .                    | .                       | .        | <b>1</b>             | <b>1</b>             | 0                 |
|  | 10                   | .                    | .                       | .        | .                    | <b>1</b>             | 1                 |

# Gland extract vs. volatile collection (example No. 1)

It appears that volatile collections reflect better the true pheromone composition of a given species than gland extracts do.

Total caught  
in test:  
279  
beetles

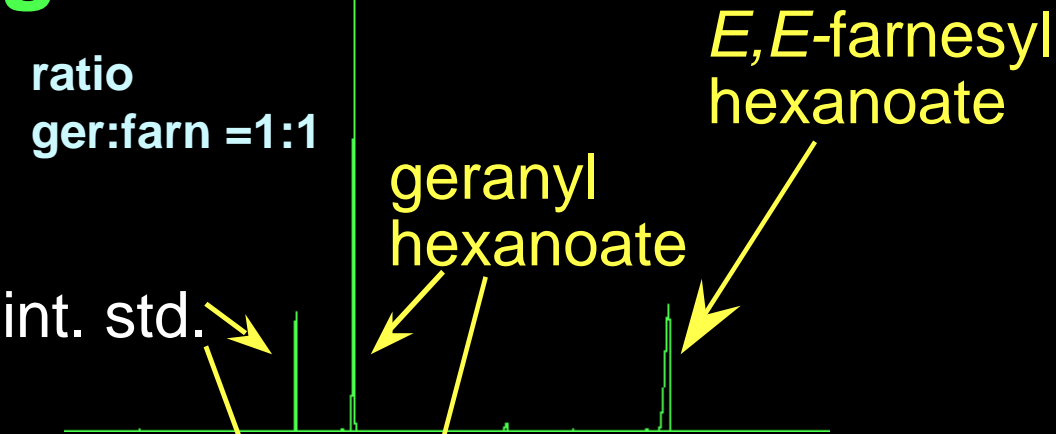
Rümlang,  
May 23 -  
August 14,  
1997

|  | Geranyl<br>octanoate | Geranyl<br>butanoate | E,E-farnesyl<br>acetate | Geraniol | Geranyl<br>hexanoate | Neryl<br>isovalerate | <i>A.lineatus</i> |
|--|----------------------|----------------------|-------------------------|----------|----------------------|----------------------|-------------------|
|  | 10                   | .                    | .                       | .        | .                    | .                    | 0                 |
|  | 30                   | .                    | .                       | .        | .                    | .                    | 2                 |
|  | 100                  | .                    | .                       | .        | .                    | .                    | 0                 |
|  | 10                   | 1                    | .                       | .        | .                    | .                    | 273               |
|  | 10                   | .                    | 1                       | .        | .                    | .                    | 2                 |
|  | 10                   | .                    | .                       | 1        | .                    | .                    | 1                 |
|  | 10                   | .                    | .                       | .        | 1                    | 1                    | 0                 |
|  | 10                   | .                    | .                       | .        | .                    | 1                    | 1                 |

# Gland extract vs. volatile collection (example No. 2)

In *A. sordidus*, in gland extracts large amounts of (*E,E*)-farnesyl hexanoate were present, with only traces in volatile collections.

**glands**



**volatiles**

ratio  
ger:farn = 1:<0.1

?

Detailed description: This chromatogram shows the composition of volatile collections. The x-axis represents time in minutes, with major ticks at 15, 20, 25, 30, 35, 40, 45, and 50. The y-axis represents relative intensity. A very large peak is observed at approximately 26 minutes, labeled as geranyl hexanoate. A very small peak is visible at approximately 50 minutes, labeled as *E,E*-farnesyl hexanoate. The ratio of geranyl hexanoate to *E,E*-farnesyl hexanoate is noted as 1:<0.1. A question mark (?) is placed near the baseline at approximately 50 minutes, indicating a trace amount.



*A. sordidus*

Photo J. Vuts

# Gland extract vs. volatile collection (example No. 2)

In field tests on *A. sordidus*, no influence of the addition of the farnesyl compound could be observed.

**glands**

ratio  
ger:farn = 1:1

int. std.

*E,E*-farnesyl  
hexanoate

geranyl  
hexanoate

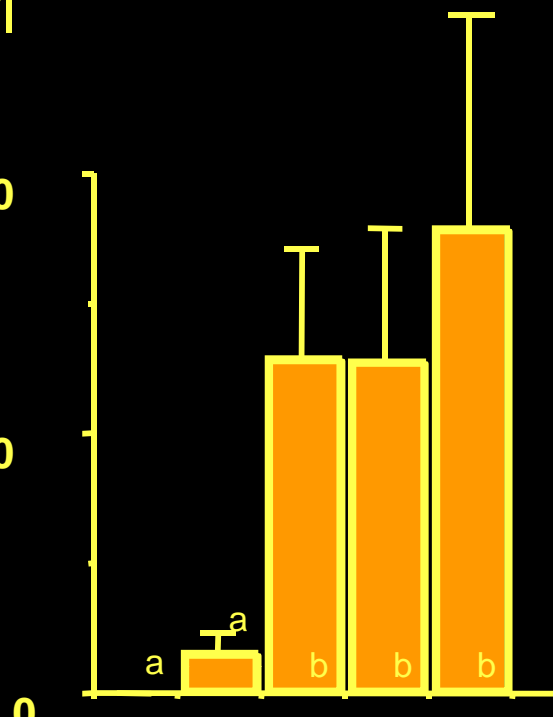
**volatiles**

ratio  
ger:farn = 1:<0.1

?

15 20 25 30 35 40 45 50 50 min

average catch/trap/inspection



Eraclea, Italy,  
1998

Total caught  
in test:  
535  
beetles

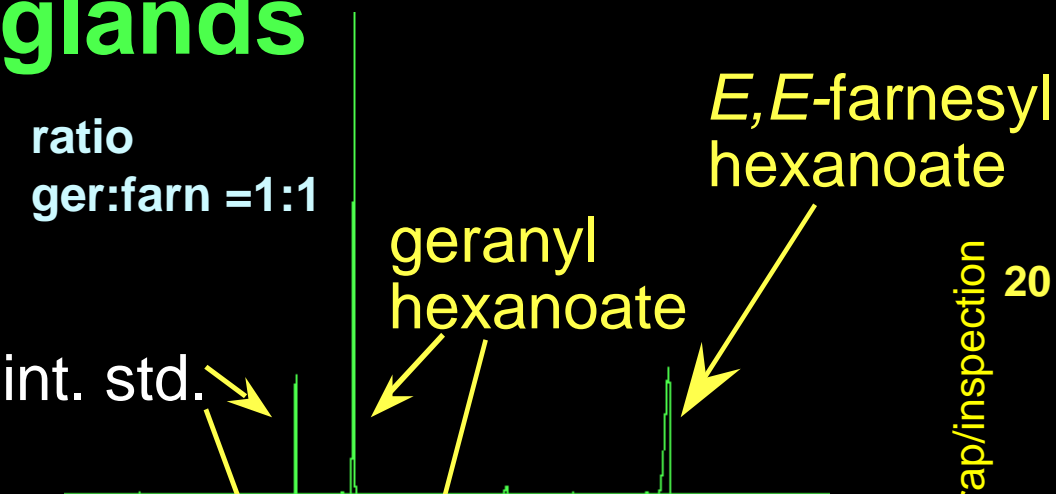
-- 3 30 30 30 geran. hex.  
30 30 30 3 -- *E,E*-farn. hex.  
(mg)

(after Tóth M, et al., unpublished)

# Gland extract vs. volatile collection (example No. 2)

Consequently, by definition, only geranyl hexanoate can be termed a pheromone component in this species.

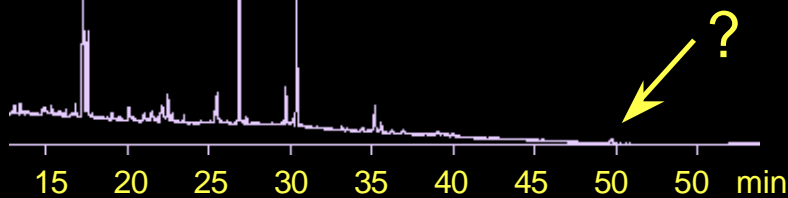
**glands**



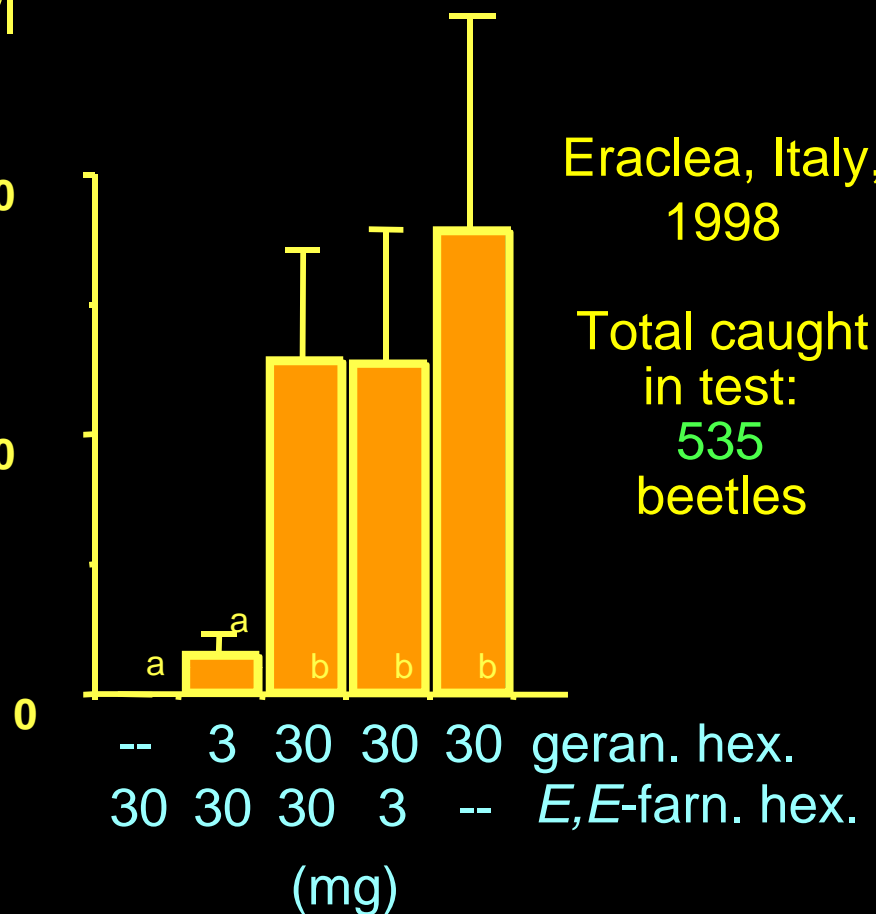
int. std.

**volatiles**

ratio  
ger:farn = 1:<0.1



average catch/trap/inspection



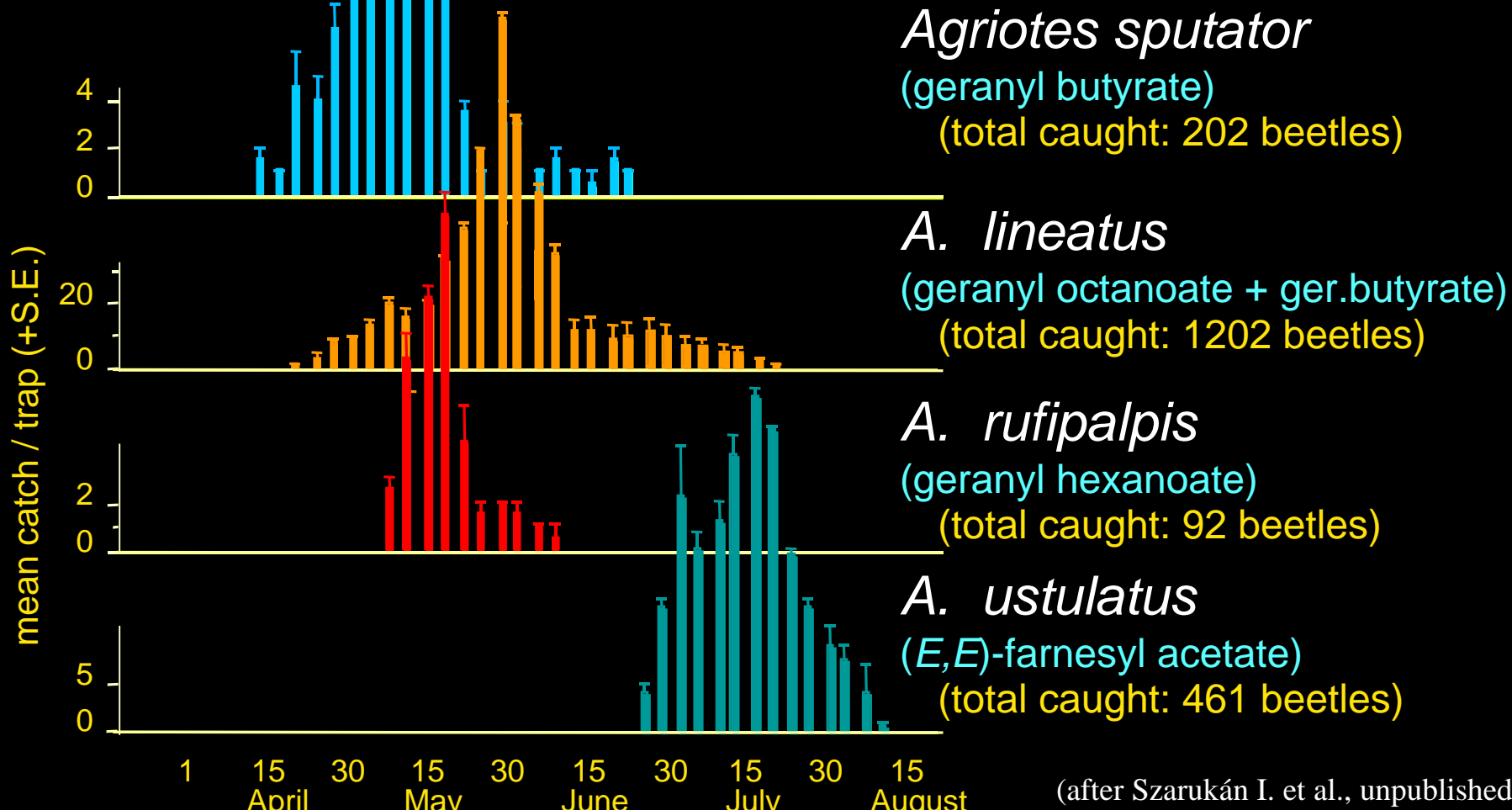
(after Tóth M, et al., unpublished)



# Pheromone traps in use in Europe

As a result of our studies pheromone traps are available for 9 *Agriotes* spp., (including all of the most important click beetle pests) in Europe.

Debrecen, Hungary, 1999



(after Szarukán I. et al., unpublished)

# Pheromone traps in use in Europe

One application of the traps in recent years was to determine the occurrence and abundance of the click beetle spp. in Europe.

## *A. sordidus* / *rufipalpis* bait

Geranyl hexanoate

1998 - 2006

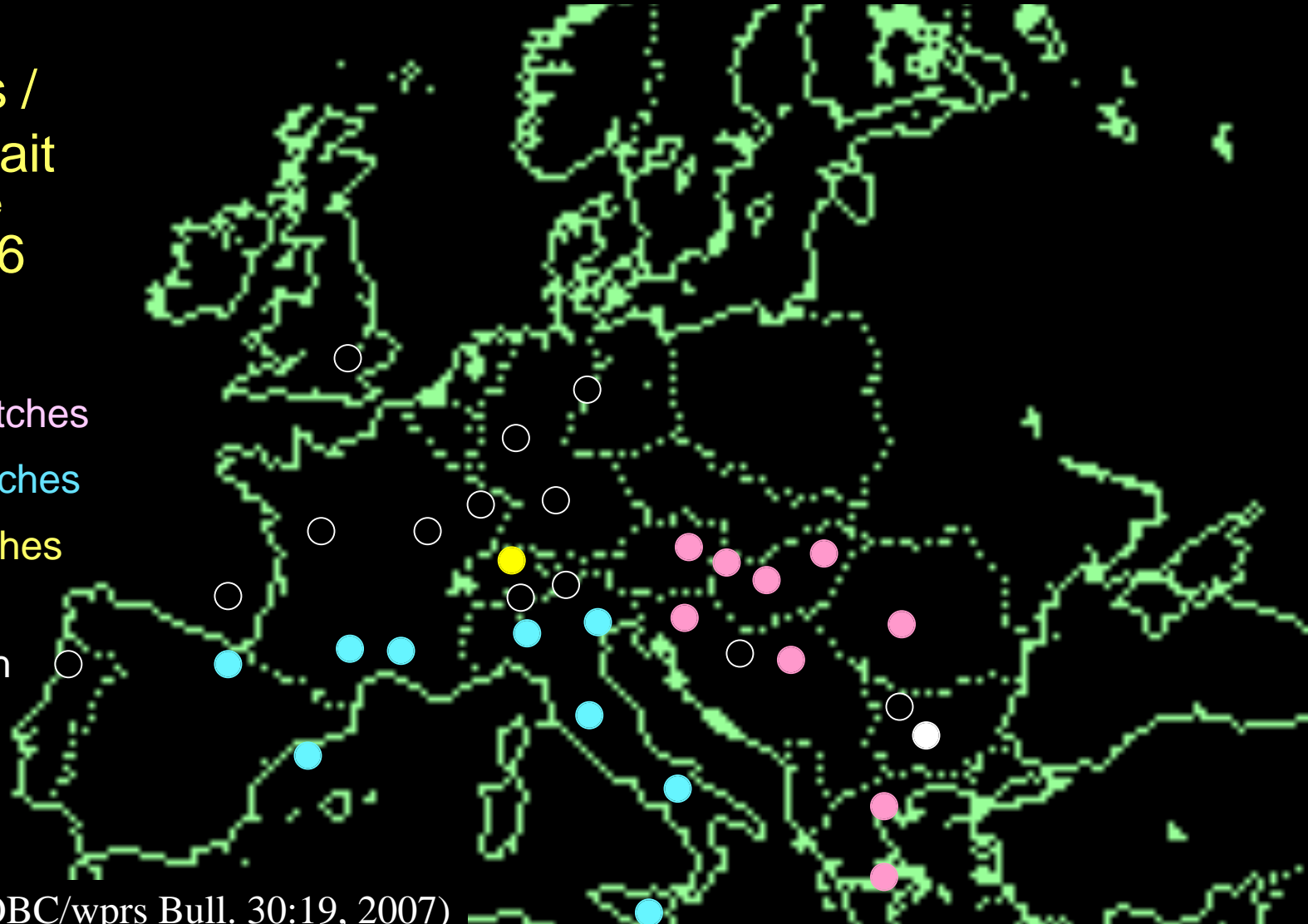
● *rufipalpis* catches

● *sordidus* catches

● *gallicus* catches

○ *Cidnopus*  
*pilosus* catch

○ no catch



(after Furlan, IOBC/wprs Bull. 30:19, 2007)

# Pheromone traps in use in Europe

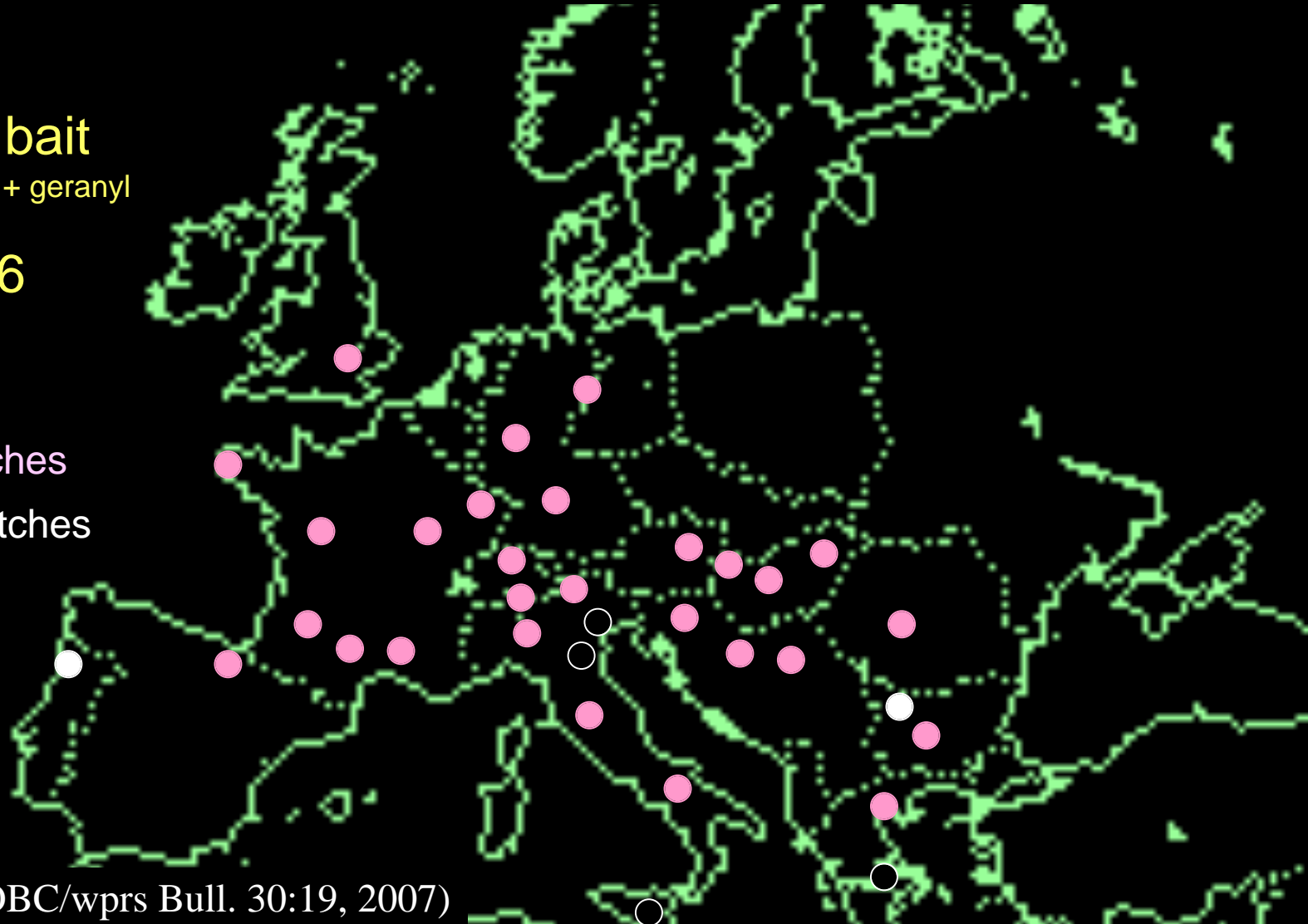
One application of the traps in recent years was to determine the occurrence and abundance of the click beetle spp. in Europe.

## *A. lineatus* bait

Geranyl octanoate + geranyl butyrate

1998 - 2006

- *lineatus* catches
- *proximus* catches
- no catch



(after Furlan, IOBC/wprs Bull. 30:19, 2007)

# Females responding to the pheromone

Click beetle pheromone traps capture large numbers of males. In some species, however, users reported on the presence of females in the catch!

The phenomenon most frequently was reported  
in:

*A. sordidus* and *A. brevis* –  
Lorenzo Furlan, Italy

*A. ustulatus* – István Szarukán,  
Hungary

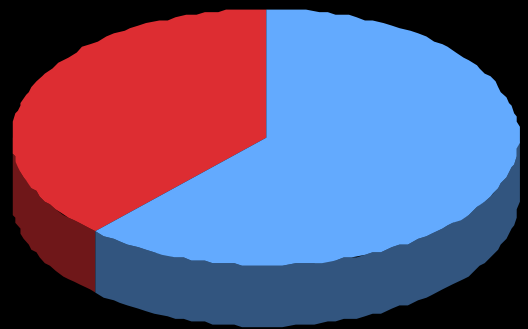


# Females responding to the pheromone

On closer scrutiny, beetle catches in pheromone baited traps indeed contained a sizeable proportion of female beetles in these species!

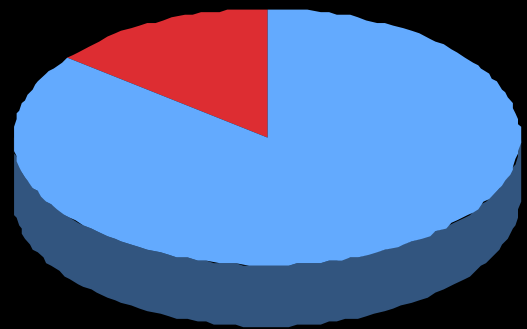
## *A. brevis*

pheromone components:  
(*E,E*)-farnesyl butyrate +  
geranyl butyrate  
Veneto, Italy, 2001  
Total caught: 1173 beetles



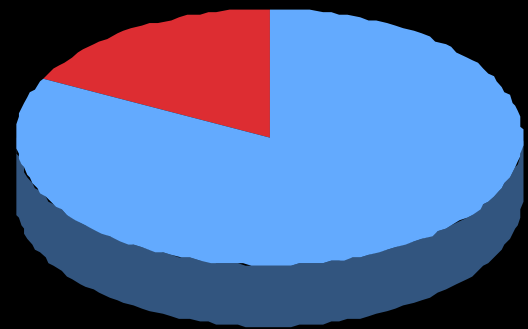
## *A. sordidus*

pheromone component:  
geranyl hexanoate  
Berton-Greggio, Italy, 2001  
Total caught: 367 beetles



## *A. ustulatus*

pheromone component:  
(*E,E*)-farnesyl acetate  
Eraclea, Italy, 2000  
Total caught: 114 beetles



# Females responding to the pheromone

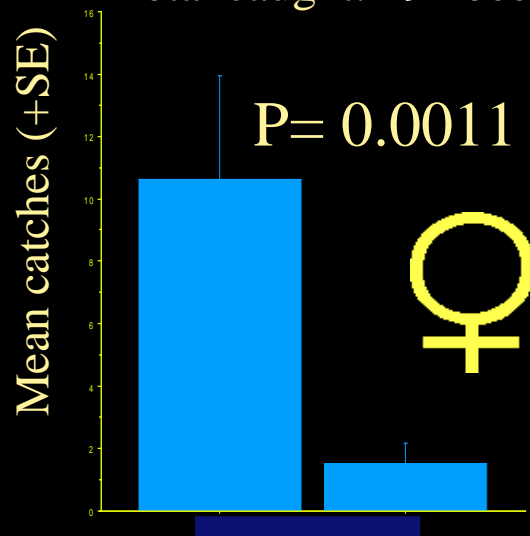
Female catches in pheromone baited traps were significantly higher than in unbaited traps.

## *A. brevis*

pheromone components:  
(*E,E*)-farnesyl butyrate +  
geranyl butyrate

Eraclea, Italy, 2000

Total caught: 194 beetles



pheromone  
bait

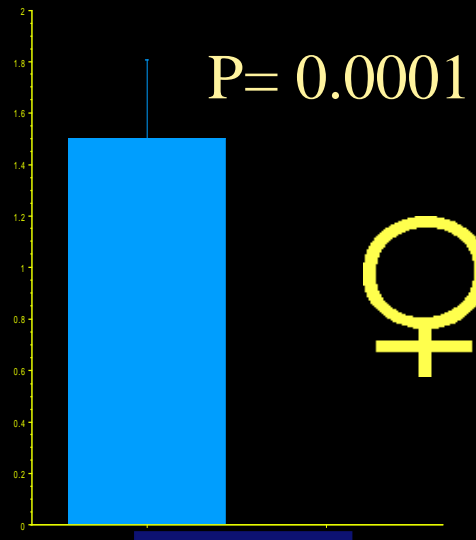


## *A. sordidus*

pheromone component:  
geranyl hexanoate

Berton-Greggio, Italy, 2001

Total caught: 42 beetles

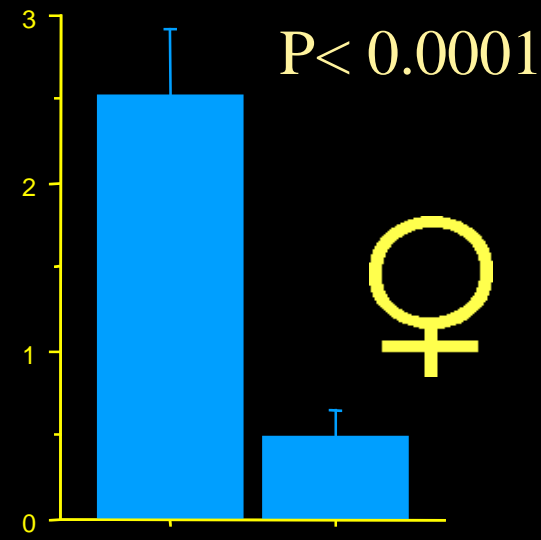


## *A. ustulatus*

pheromone component:  
(*E,E*)-farnesyl acetate

Debrecen, Hungary, 2003

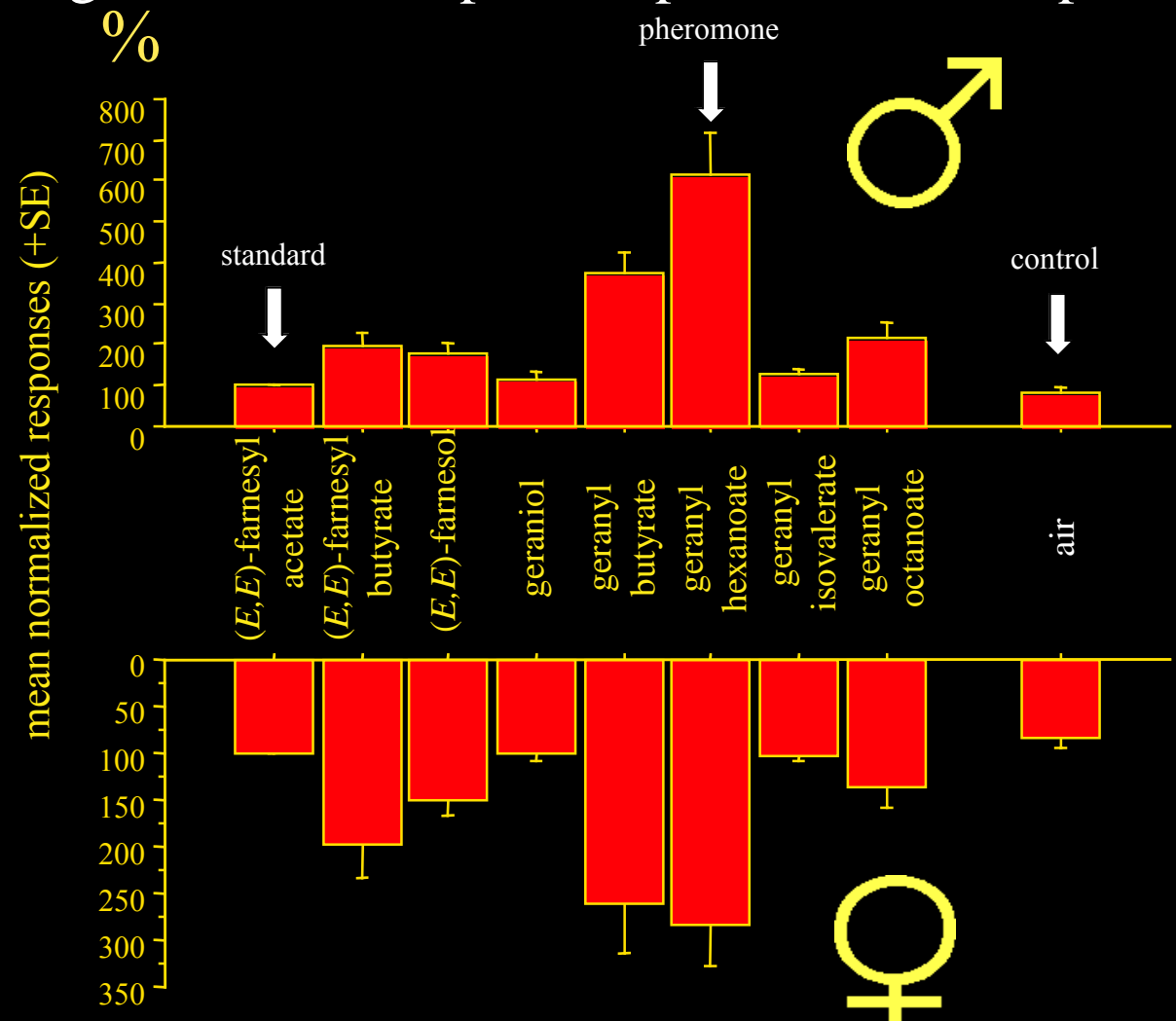
Total caught: 116 beetles



# Females responding to the pheromone

In EAG screenings of synthetic pheromone compounds of click beetles, female antennae showed similar response spectra as male antennae, responding best to the respective pheromone component.

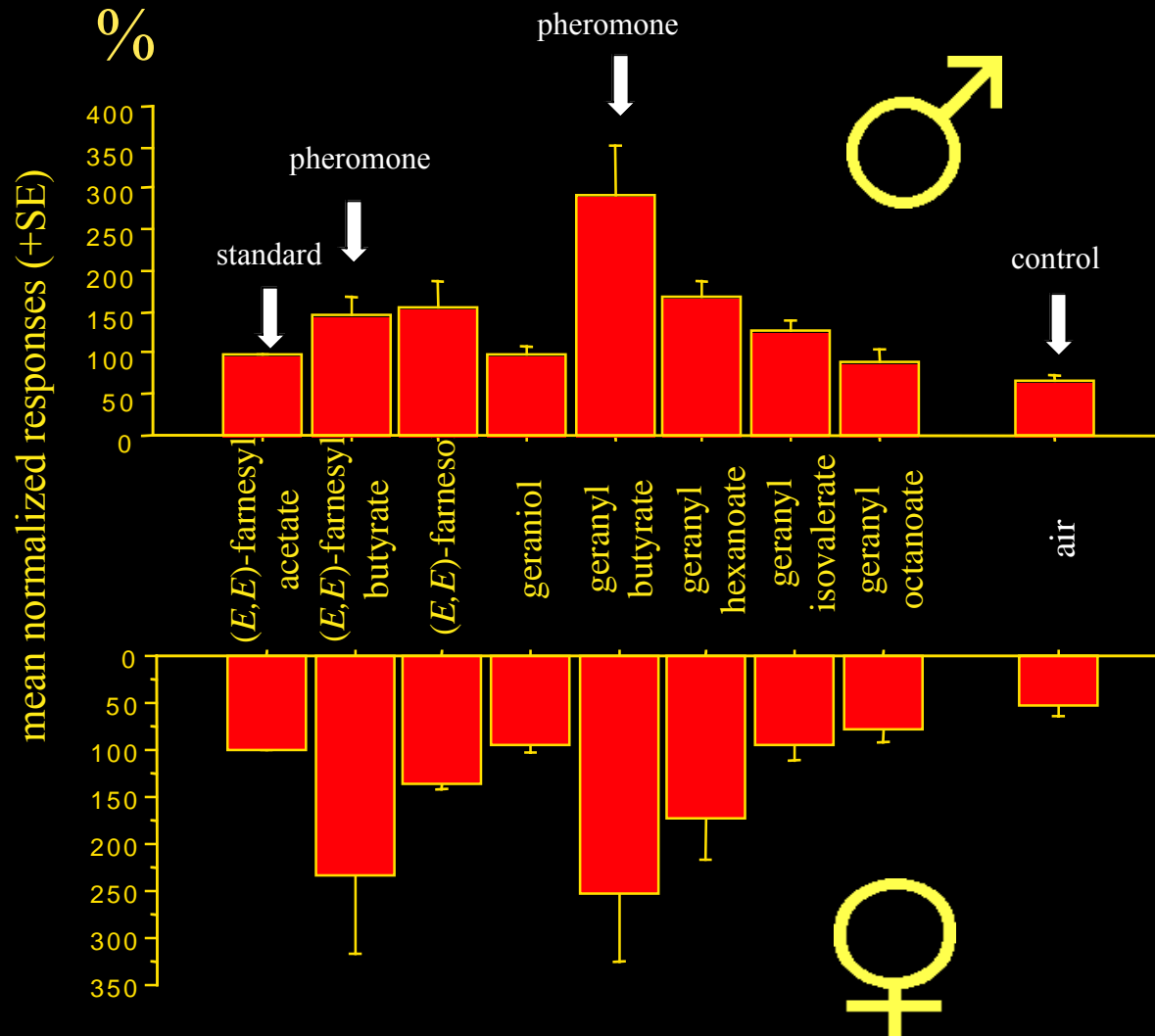
*Agriotes sordidus*



# Females responding to the pheromone

This suggested that females were capable of detecting their respective pheromone components.

*Agriotes brevis*

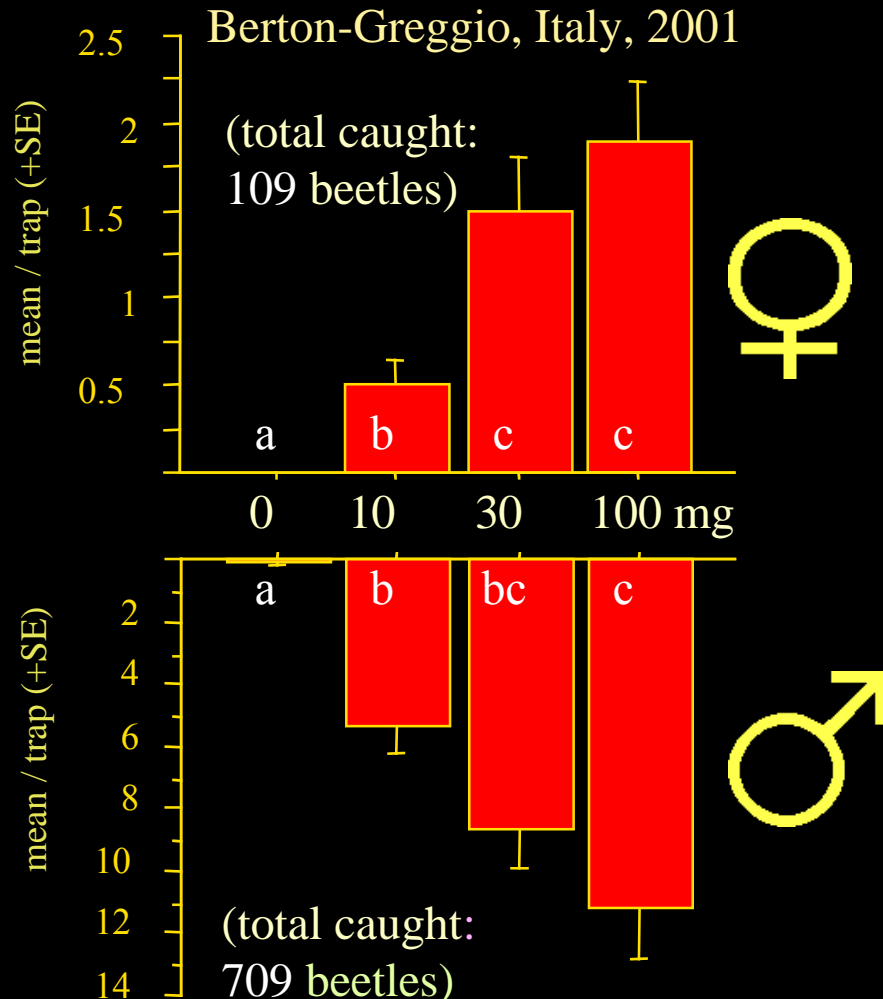




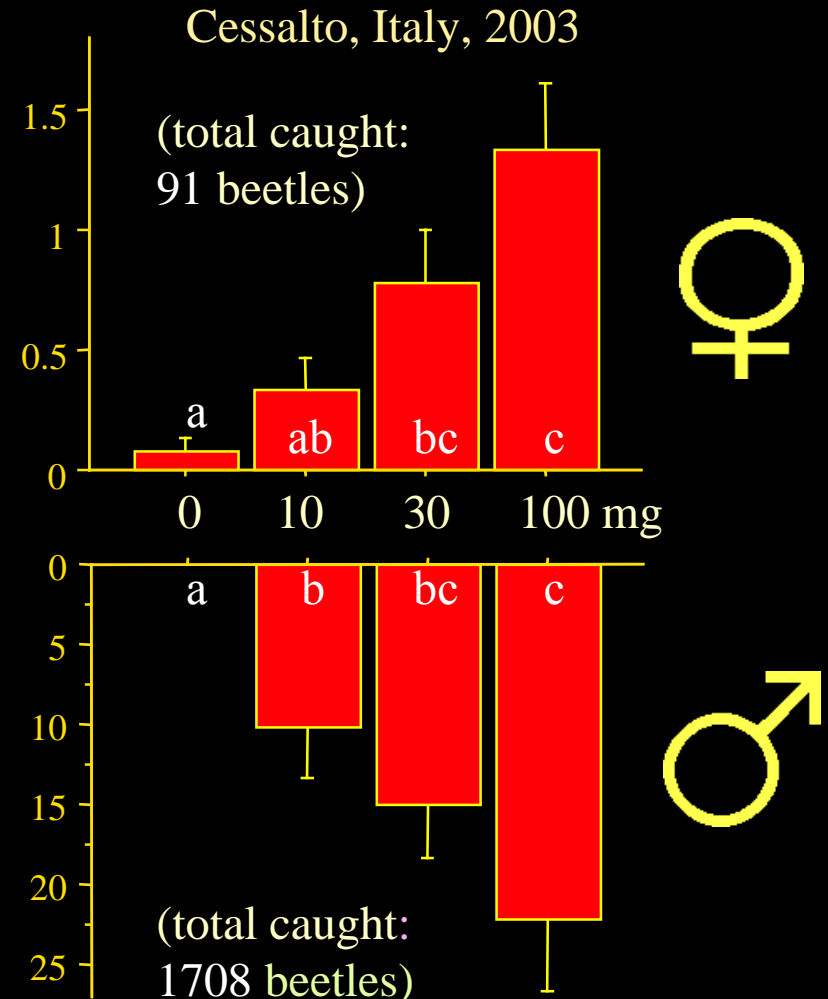
# Females responding to the pheromone

Female catches in pheromone baited traps showed dose dependence, similar to catches of male beetles.

## *A. sordidus*



## *A. ustulatus*

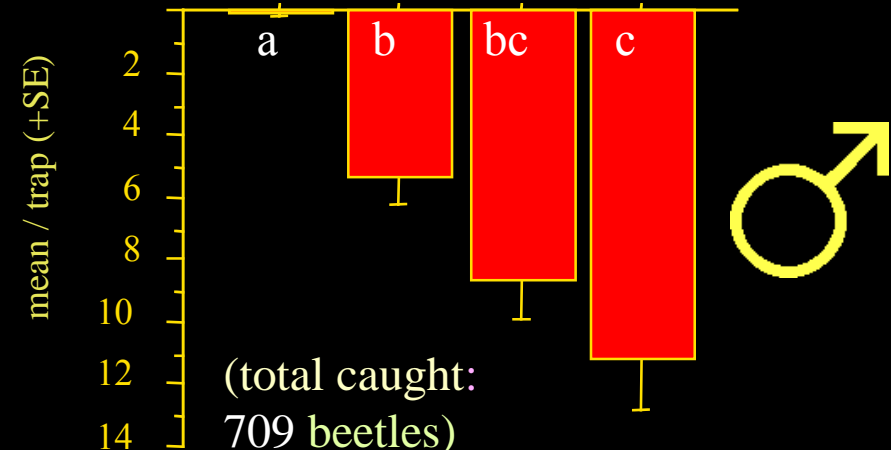
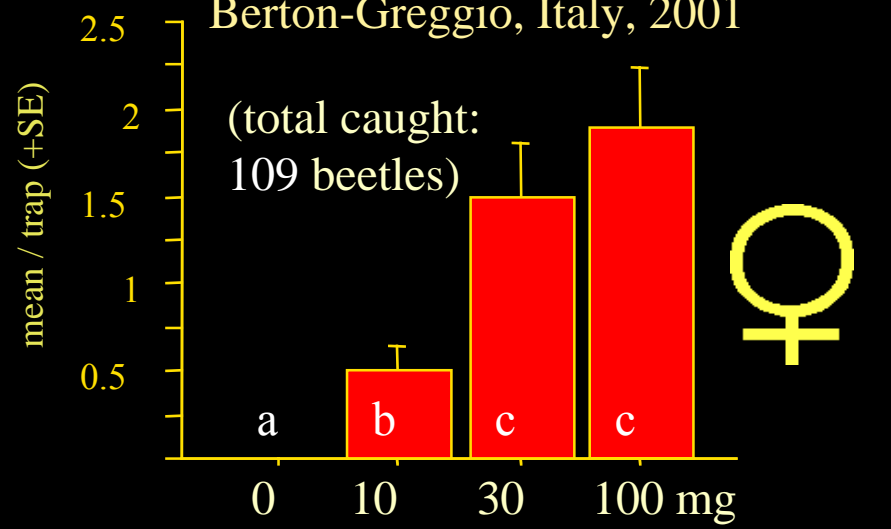


# Females responding to the pheromone

The demonstrated female attraction to the resp. pheromone brings up the possibility of developing a trap catching females also.

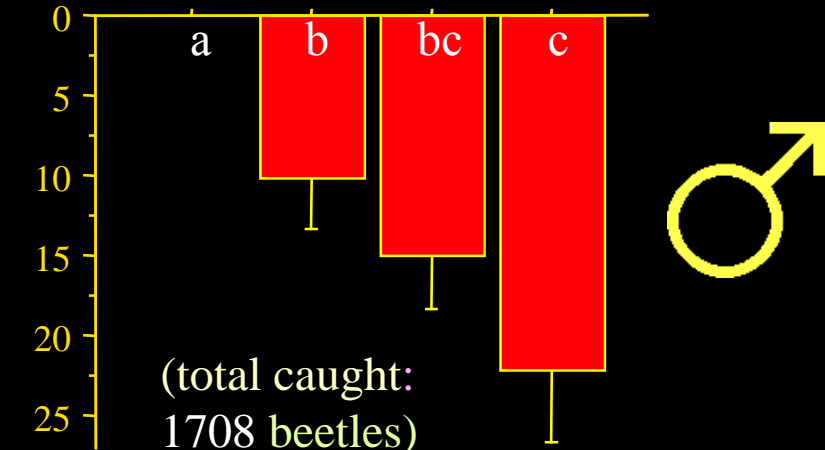
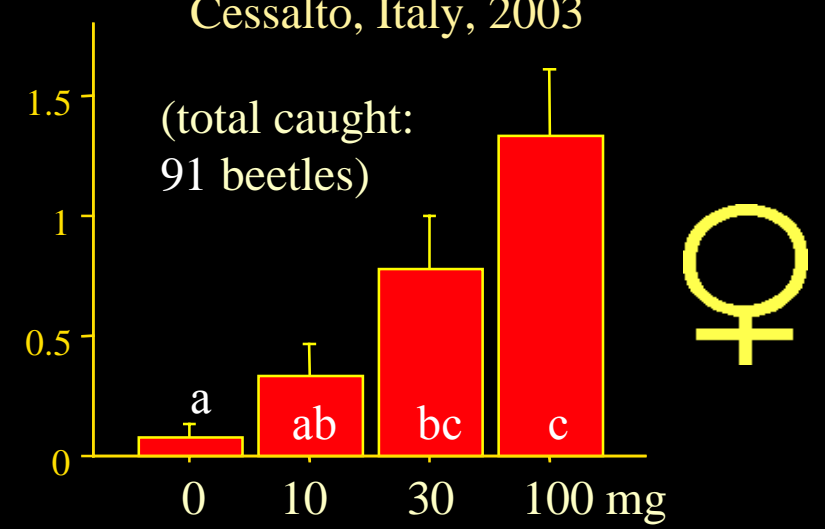
## *A. sordidus*

Berton-Greggio, Italy, 2001



## *A. ustulatus*

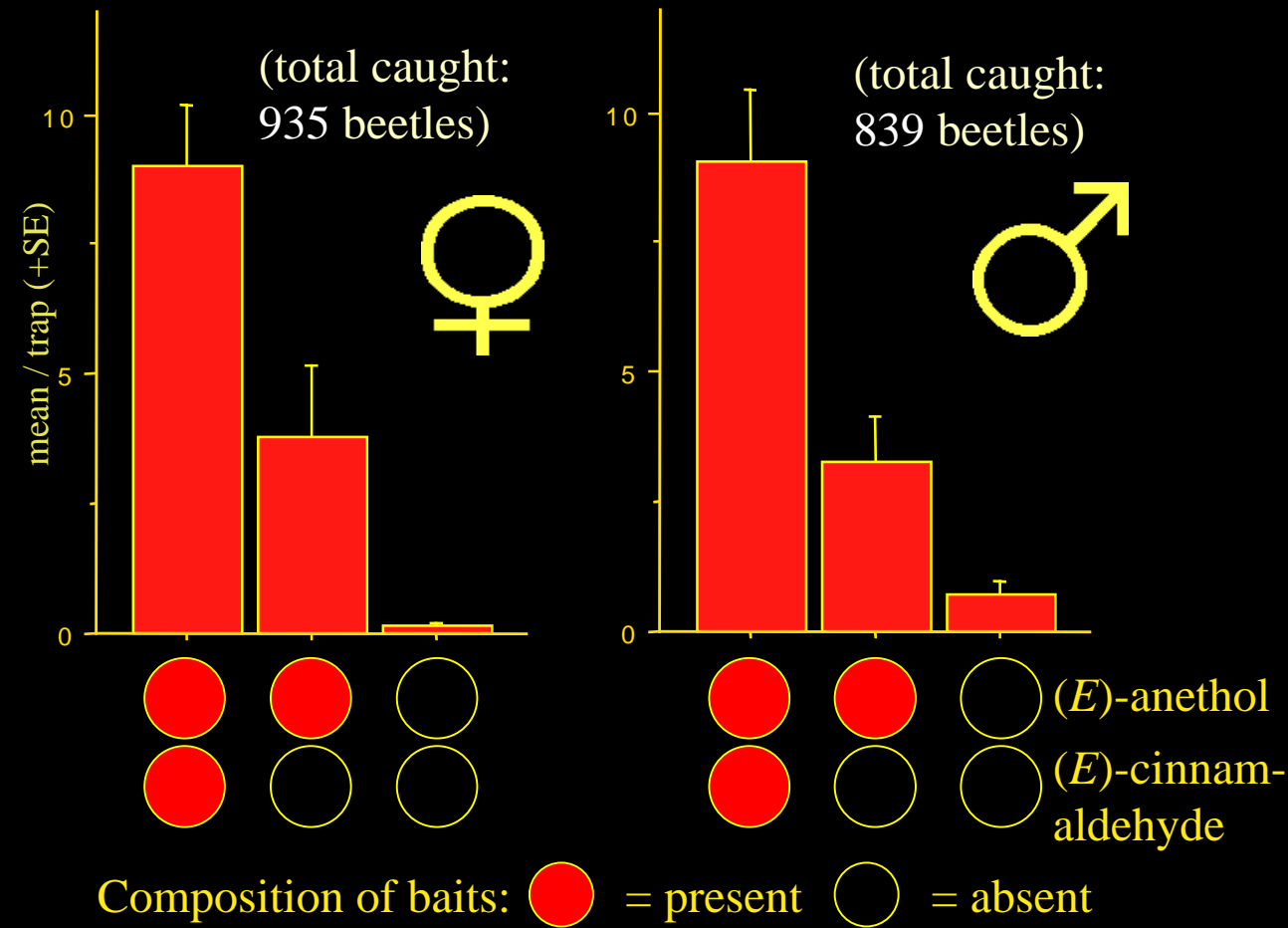
Cessalto, Italy, 2003



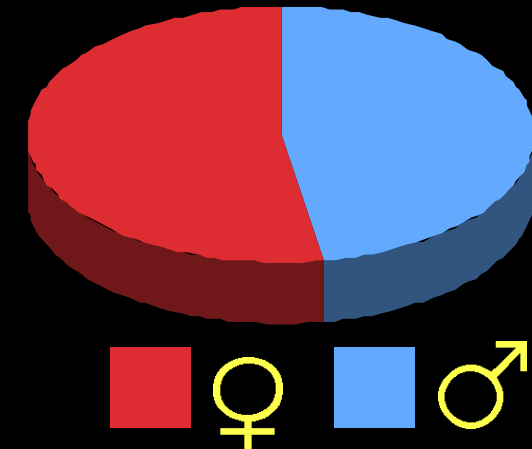
# Efficient click beetle traps for both sexes?

In the flower visitor *A. ustulatus*, a floral lure has been discovered, which attracts both females and males.

Debrecen, Hungary, 2006



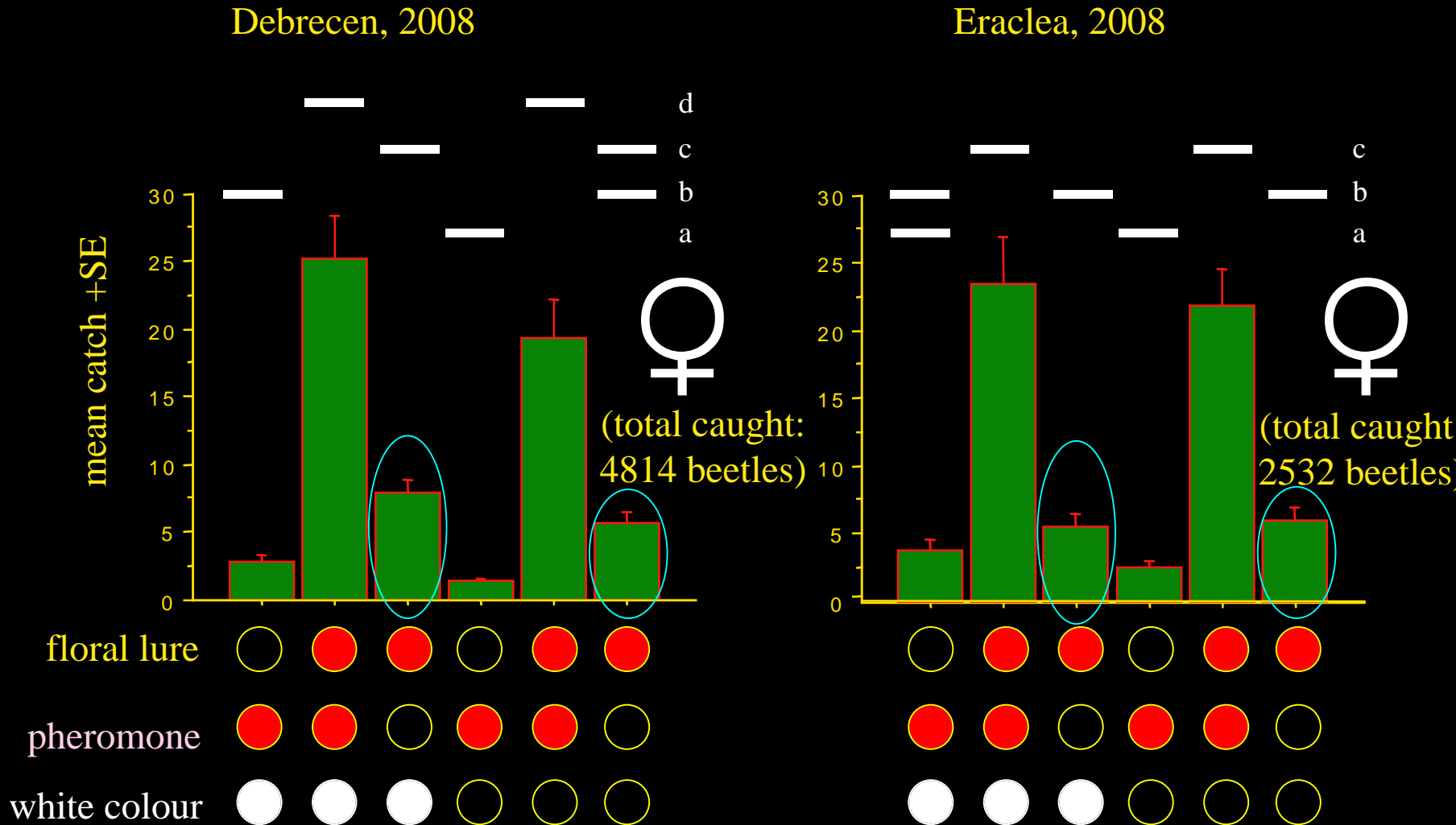
Sex ratio in catch of the 2 component floral lure (total caught 1232 beetles)



(After Tóth, Acta Phytopath. Hung. 46:235, 2011)

# Efficient click beetle traps for both sexes?

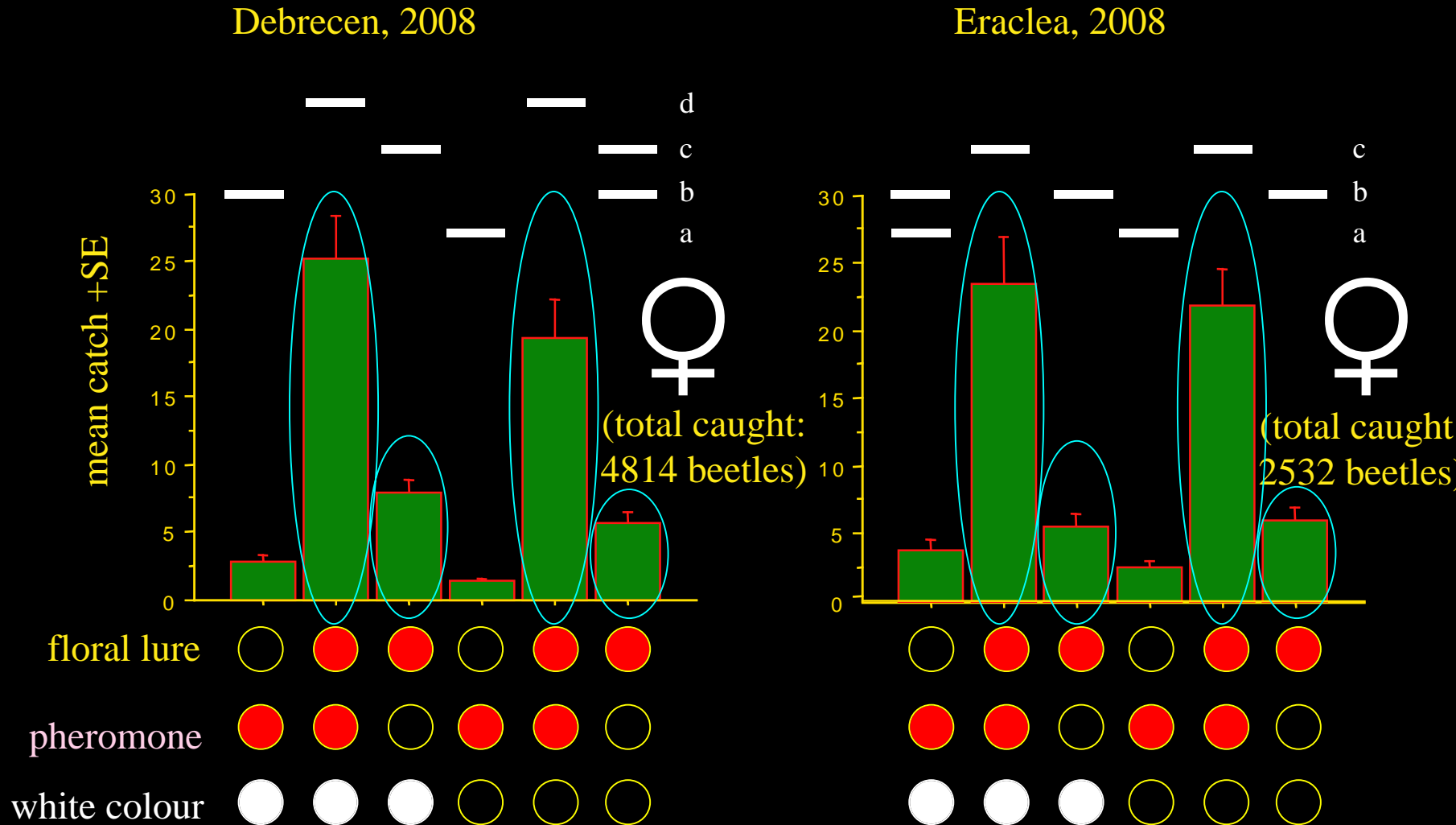
White colour (a known attractive visual cue for *A. ustulatus*) showed negligible influence on traps baited with floral or pheromonal lures



(After Tóth M. et al., in preparation)

# Efficient click beetle traps for both sexes?

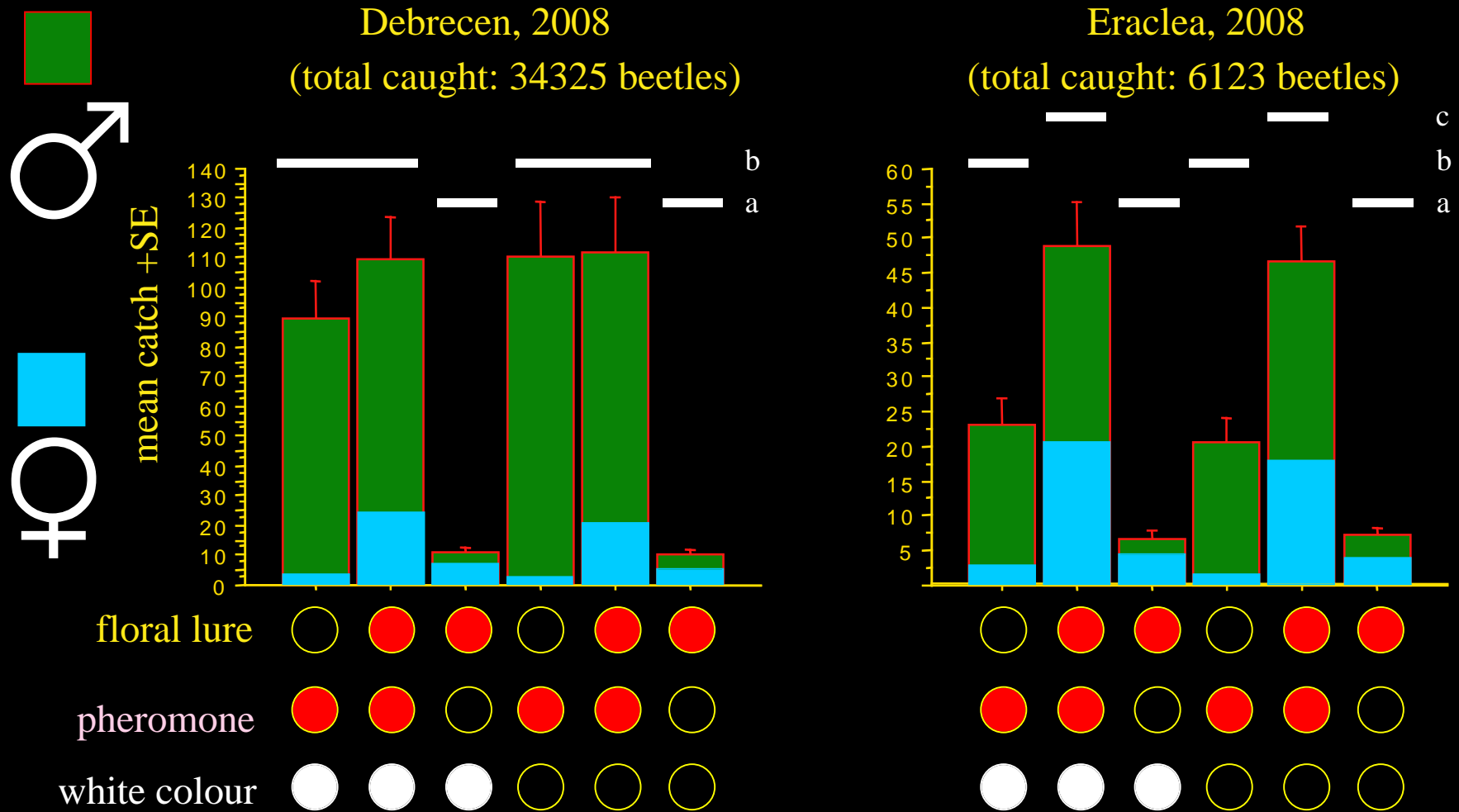
The addition of the pheromone to the floral lure in the same trap increased catches of females!



(After Tóth M. et al., in preparation)

# Efficient click beetle traps for both sexes?

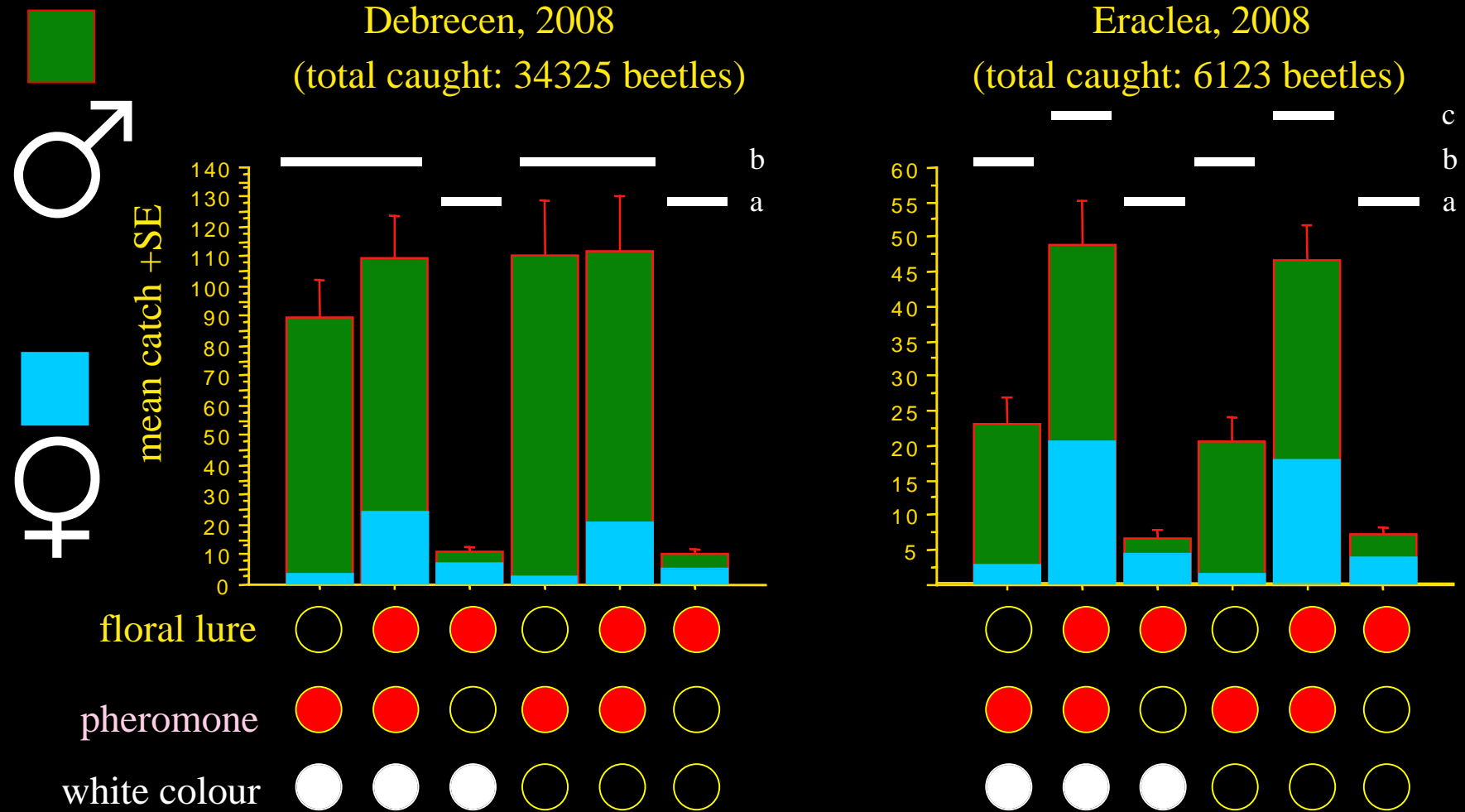
Most female specimens were caught in traps with both floral AND pheromonal baits



(After Tóth M. et al., in preparation)

# Efficient click beetle traps for both sexes?

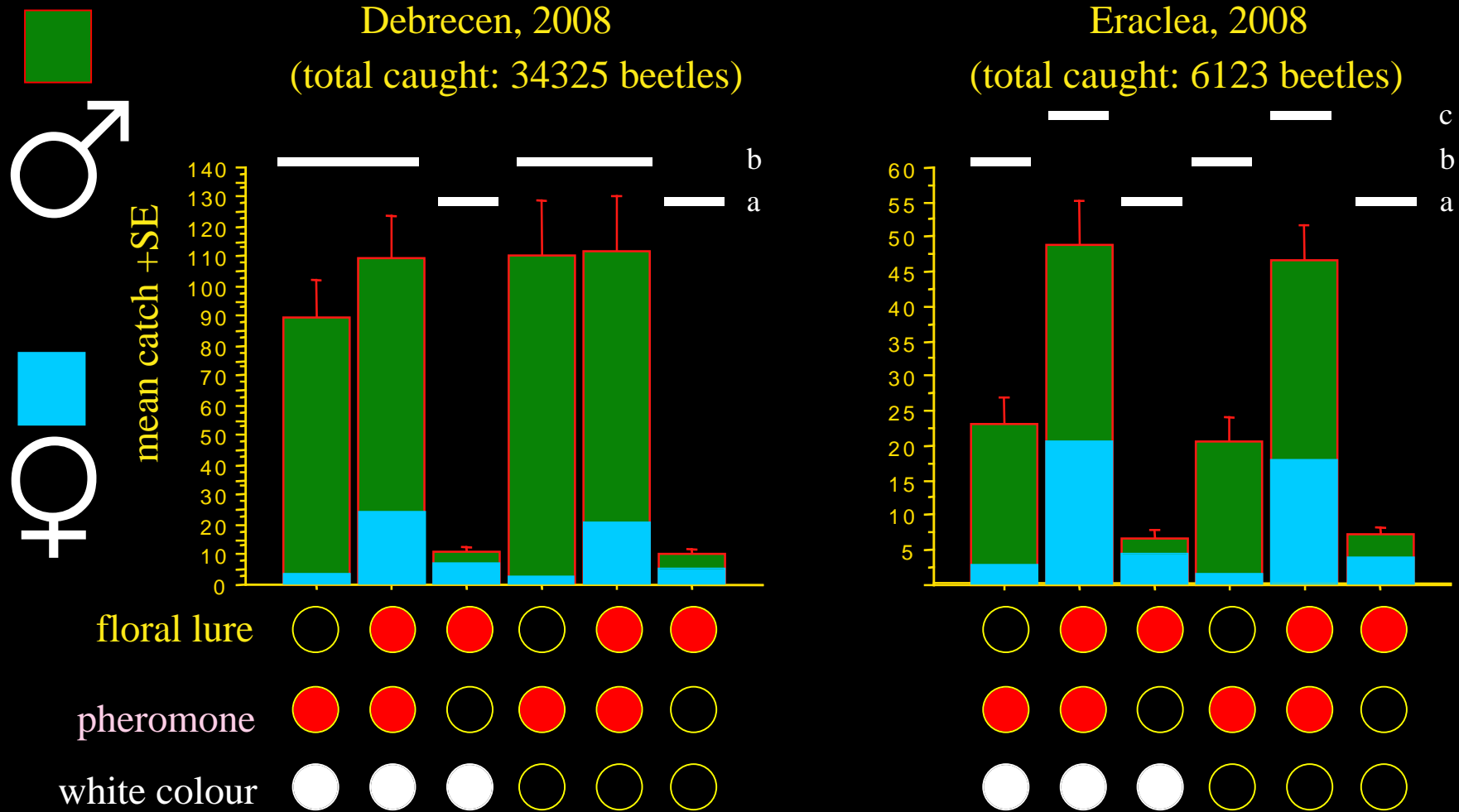
A similar phenomenon (that the presence of the pheromone increases the effect of floral bait on females) is unusual with sex pheromones



(After Tóth M. et al., in preparation)

# Efficient click beetle traps for both sexes?

The phenomenon is frequently reported with aggregation pheromones, so elaterid pheromones may have to be reclassified.



(After Tóth M. et al., in preparation)



# Acknowledgements



Scientists cooperating in  
unpublished results shown:

Bálintné Csonka, É. (Hungary)

Francke, W. (Germany)

Furlan, L. (Italy)

Subchev, M. (Bulgaria)

Szarukán, I. (Hungary)

Tolasch, T. (Germany)

Toshova, T. (Bulgaria)

Vuts, J. (Hungary)

Yatsynin, V.G. (Russia)