Alien crayfish management in Mediterranean areas. The Italian experience.

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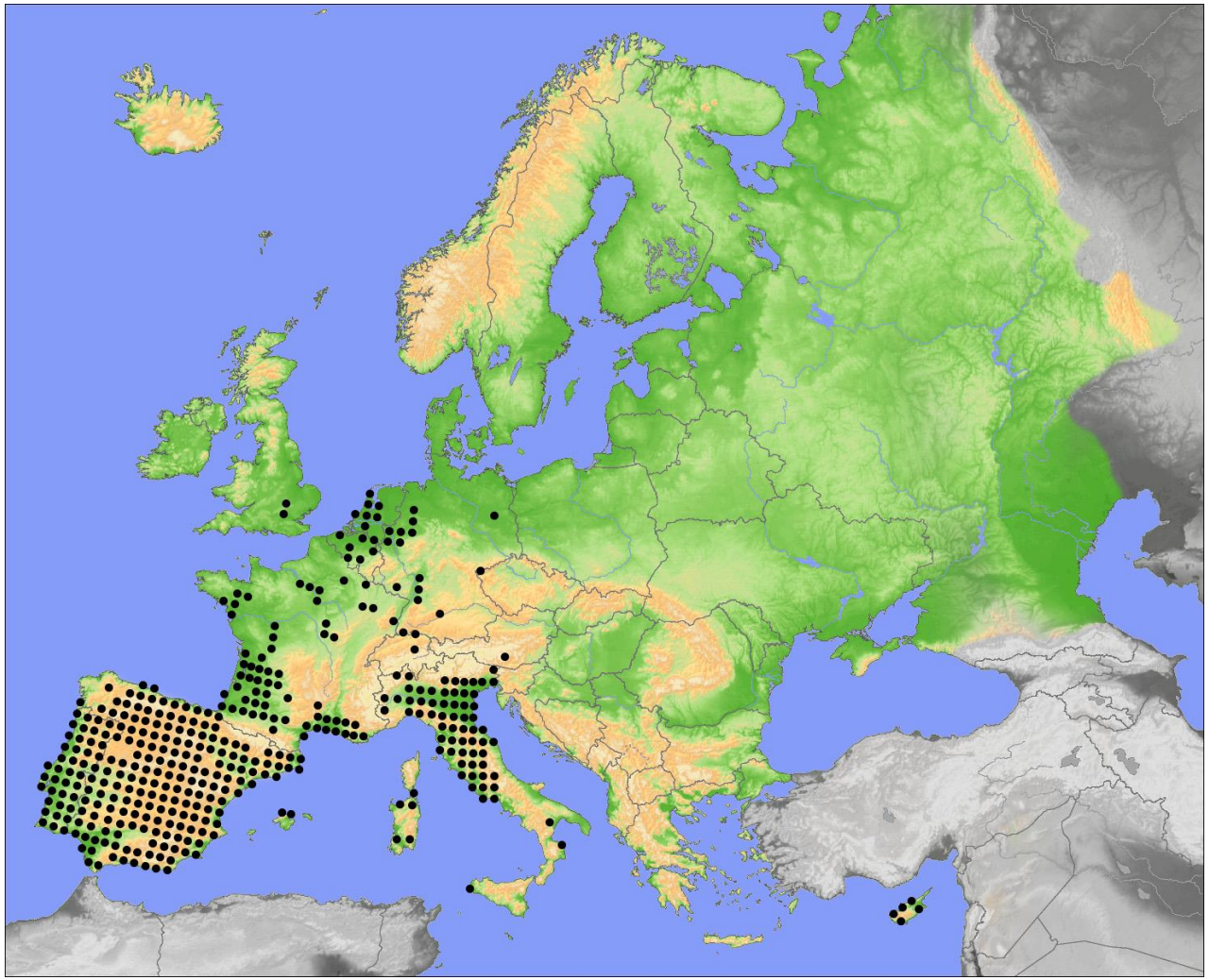
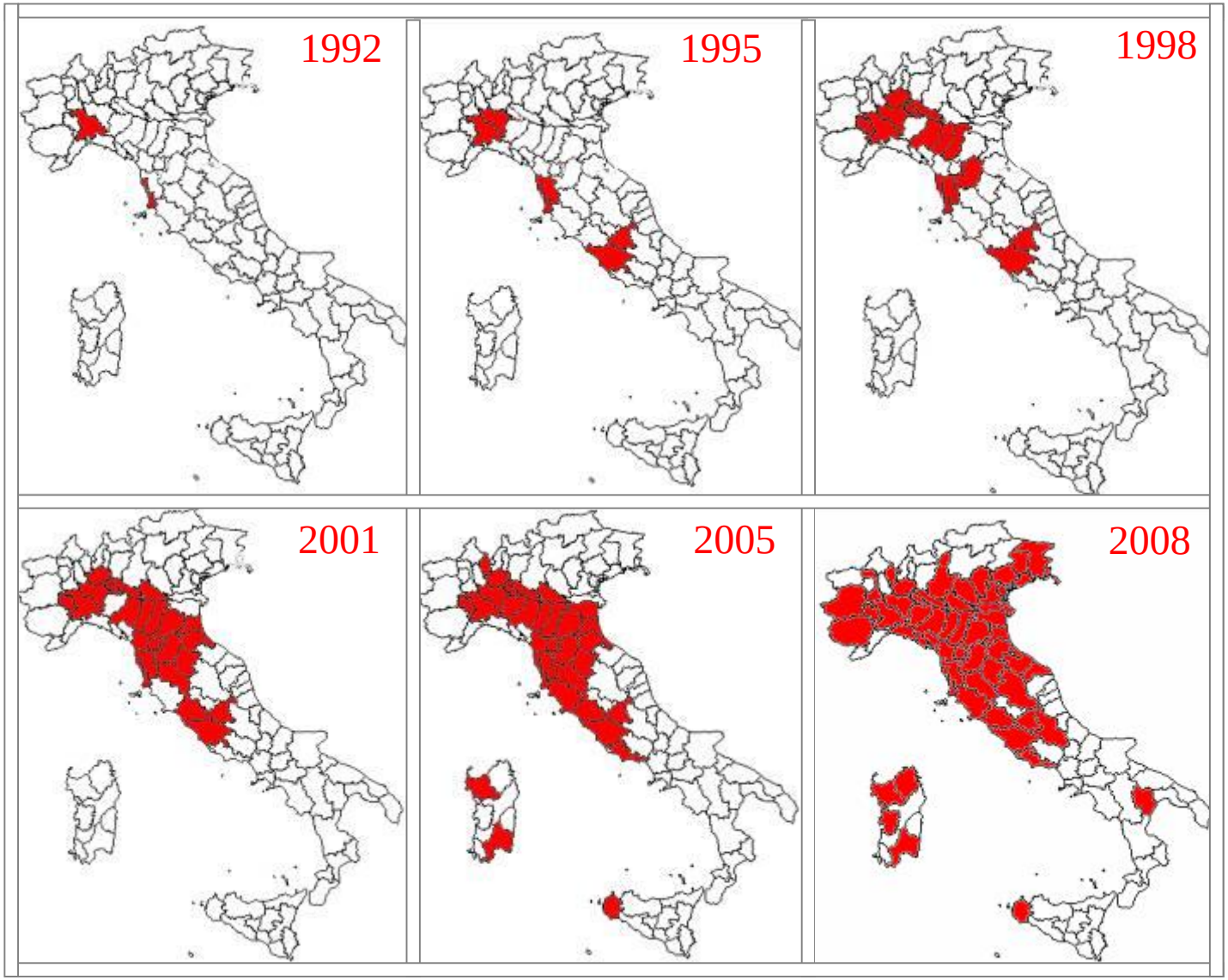
The red swamp crayfish, *Procambarus clarkii* (one of the 100 worst invasive species in Europe; Gherardi & Panov 2009).

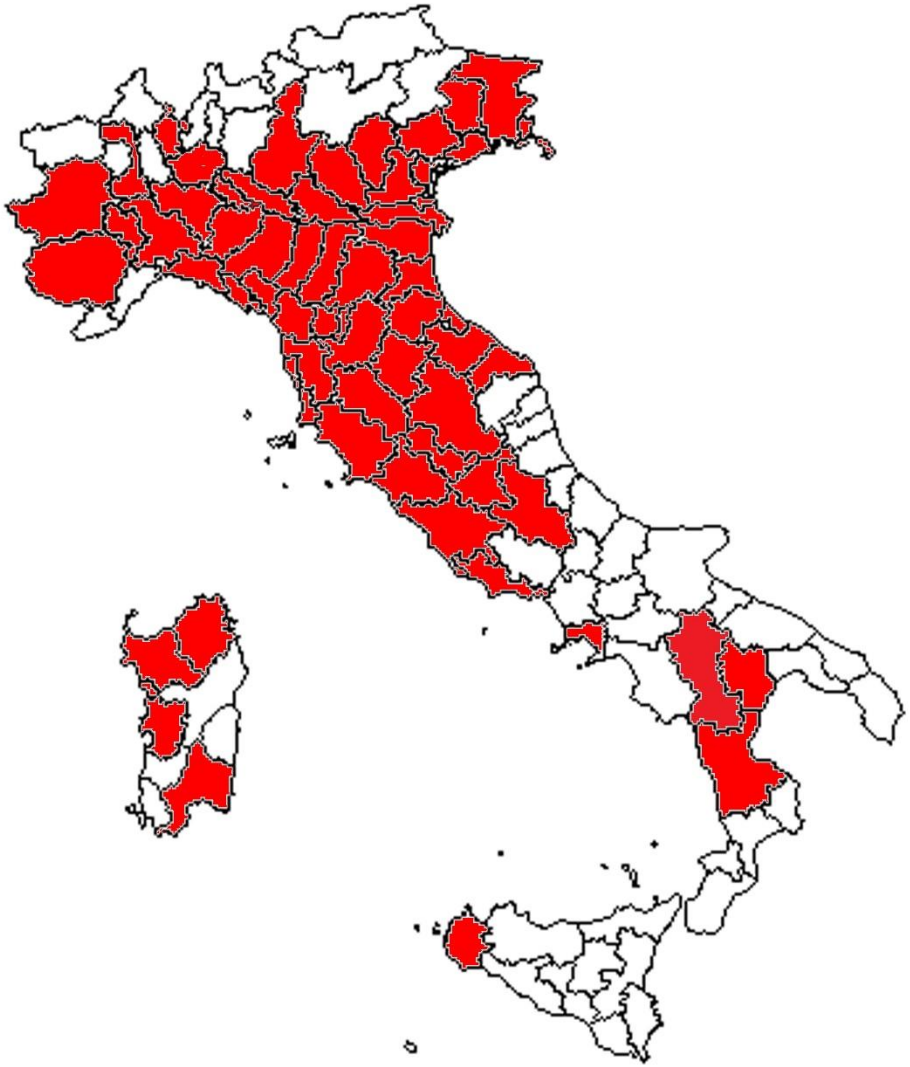
Europe



Kouba et al. 2014

Invasion in Italy

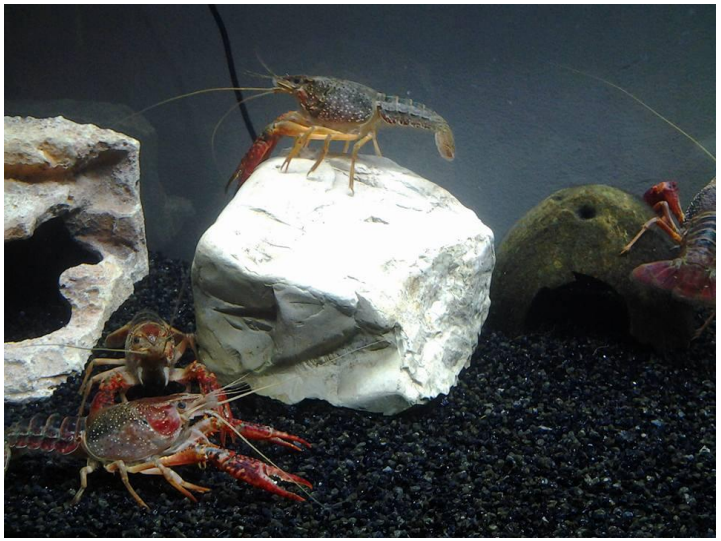




2015

And the invasion can continue…

The species is sold in the online aquarium trade (Mazza et al. 2015).



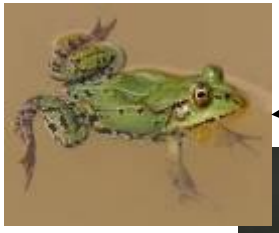
The perfect invader

Damages to human health

Economic impacts

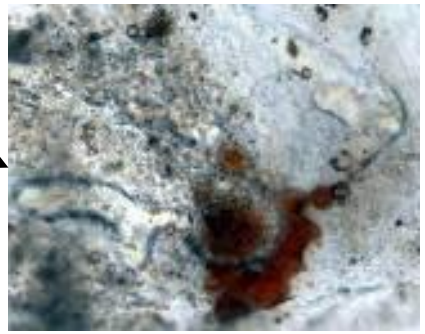
Competition

Predation



Habitat destruction

Transmission of pathogens



It is imperative to intervene!

- Mechanical methods (trapping)
- Biological methods (use of native predators, biocides, autocides).

- Protected areas in Tuscany
- Irrigation ditches in Emilia-Romagna
- Wetland areas in Friuli Venezia Giulia

Trapping

- Cat food as bait
- A high number for a long period
- Good for small populations/early detected populations
- Coupled with another method
- Used to monitor the population and the effect of the management actions (C.P.U.E.)

Pros: very simple and friendly use
Cons: cost of manpower, juveniles and ovigerous females are trap shy

Biological methods

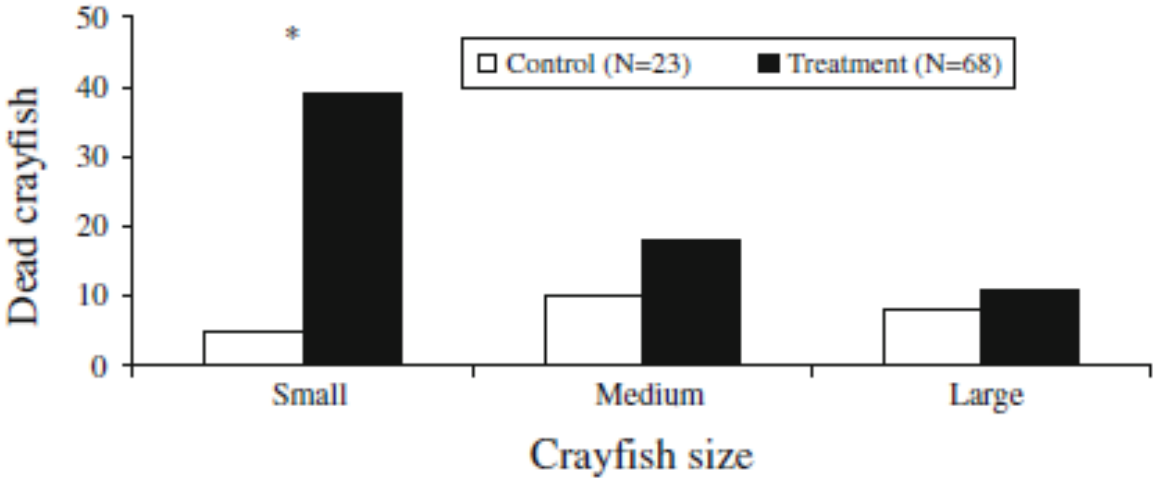
(a) The use of native predators

A good candidate for *P. clarkii*‘s biological control is the European eel.

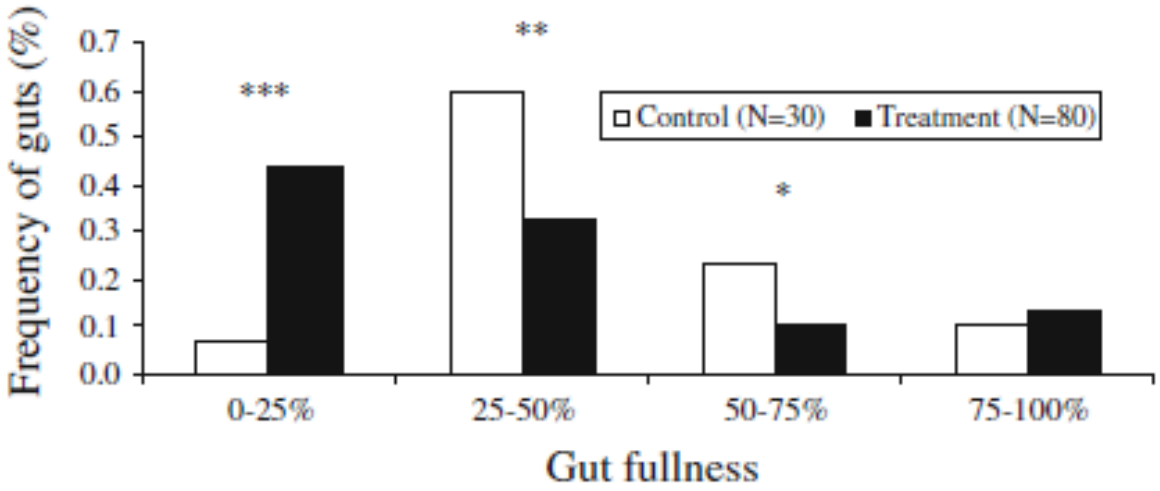
Pros:

1) it is an indigenous and
2) benthonic feeder; it
3) tolerates partially deoxygenated waters.

Direct effect



Indirect effect

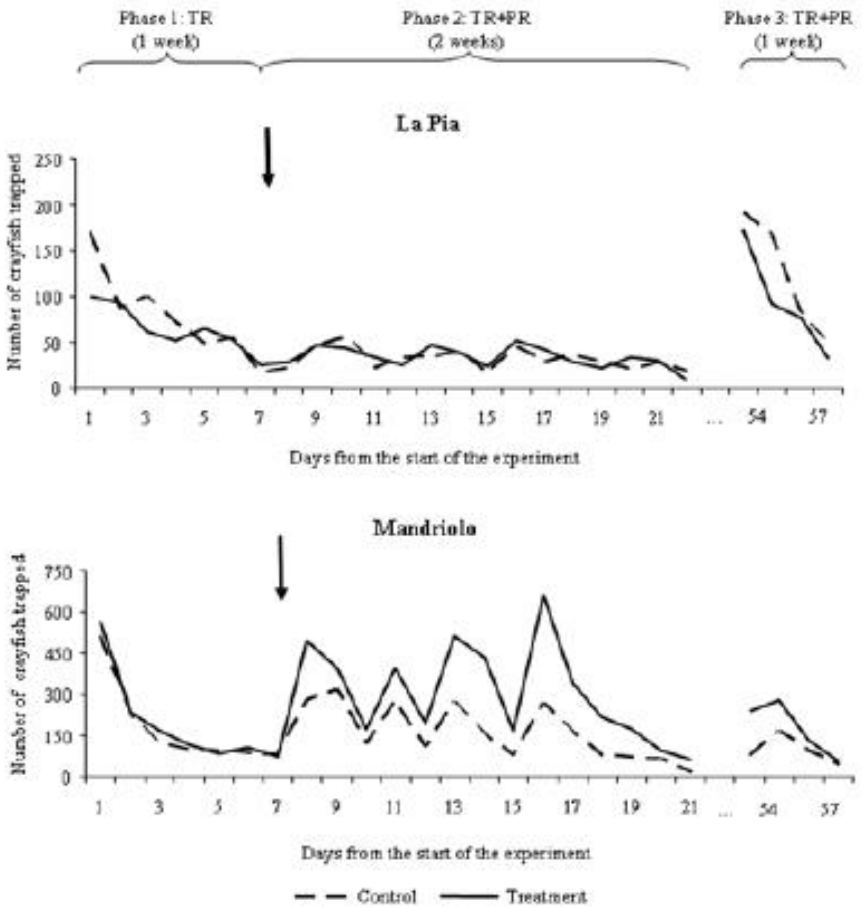


Righetti-La Monaca (Tuscany)

Aquiloni et al. 2010

Do the predators work?

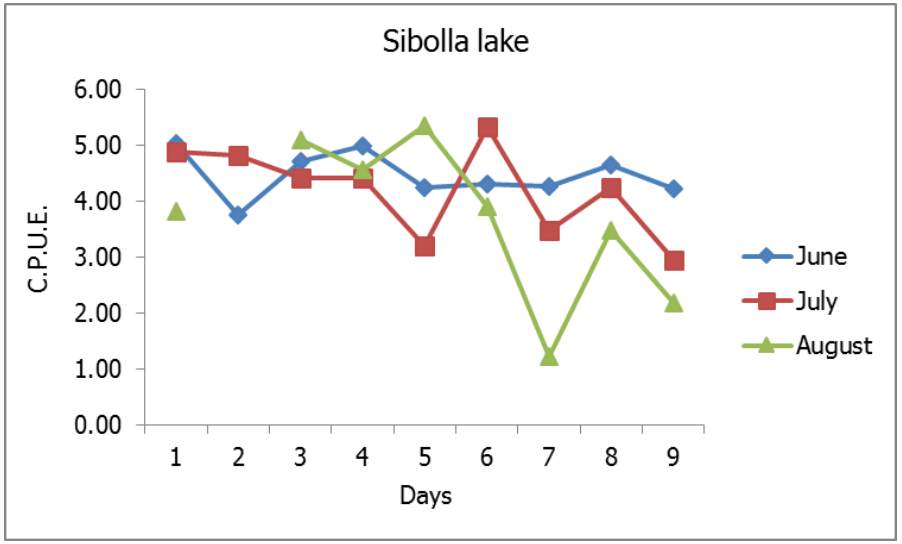
Irrigation ditches (Emilia-Romagna)



Aquiloni et al. 2010

Cons: restoring the habitats for the predator, long term effect, introduced more eels, traps detect large-medium crayfish.

LIFE SOS TUSCAN WETLANDS



Decrease of 55% in CPUE in 2014

Native predators are helping us!

Tricarico et al. 2015

Biological methods

(b) Biocides

Use of Pyblast for crayfish

Pros

Low toxicity for mammals and birds
No toxic for plants
Rapid decay with sunlight
No toxic residuals

Cons

Not selective for aquatic animals
More expensive compared to other pyretroids

Irrigation ditches (Emilia-Romagna)

To be used in restricted areas in order to maximize the number of dead individuals of the target species and to minimize the quantity of biocide in the habitat.

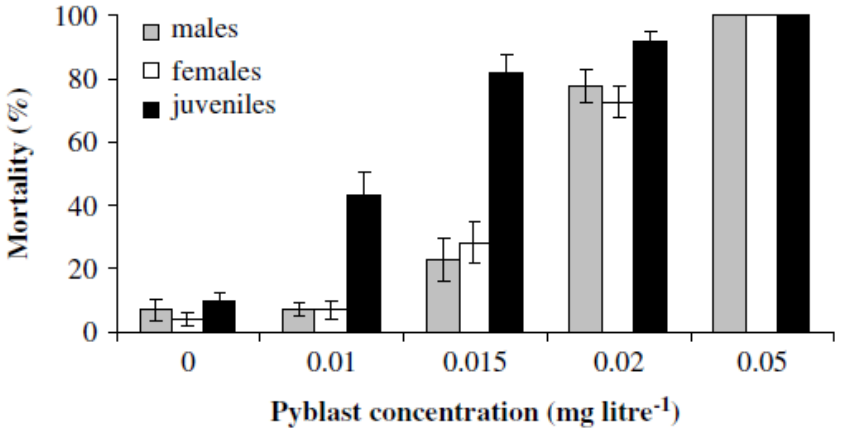


Figure 1. Crayfish mortality (in %) after 24 h, compared among five Pyblast concentrations and among groups (N = ten crayfish per group) during acute toxicity tests carried out in tanks (experiment 1).

0.05 mg/l is the optimal concentration (100% dead crayfish and 33% mortality of *Daphnia magna* used as bioindicator).

Table 3. Number of crayfish captured in the 24 h trapping sessions before and after experiment 4. In this experiment, Pyblast was applied in the water of an experimental section of a drainage channel

Trap number	Before	After
1	30	0
2	27	0
3	22	0
4	44	0
Total	123	0

Cecchinelli et al. 2010

Biological methods

(c) Autocides: pheromones

P. clarkii males respond to female sexual pheromones (as shown by their reduced aggressiveness) and do not require the sight of the female partner to be attracted to her (as shown by the more intense locomotion in the presence of the female odor than of her sight alone; Aquiloni et al. 2009).

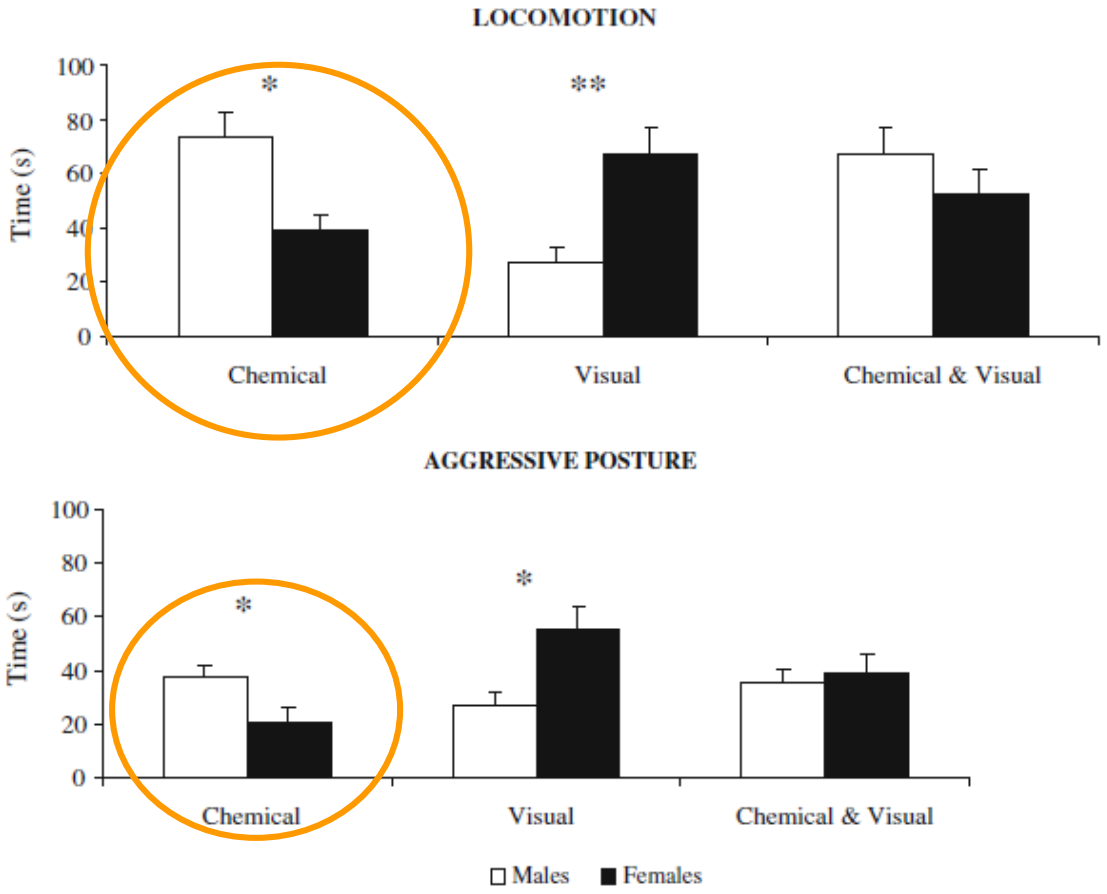


Fig. 3 Differences between the control and the test phases (mean± SE) in the time spent (in seconds) by *P. clarkii* males and females in locomotion and in the aggressive posture: comparisons between sexes per treatment. *One* and *two asterisks* denote significant differences at $p<0.01$ and $p<0.001$, respectively, after Mann–Whitney tests. $N=20$ per treatment

Do pheromones work in the wild?

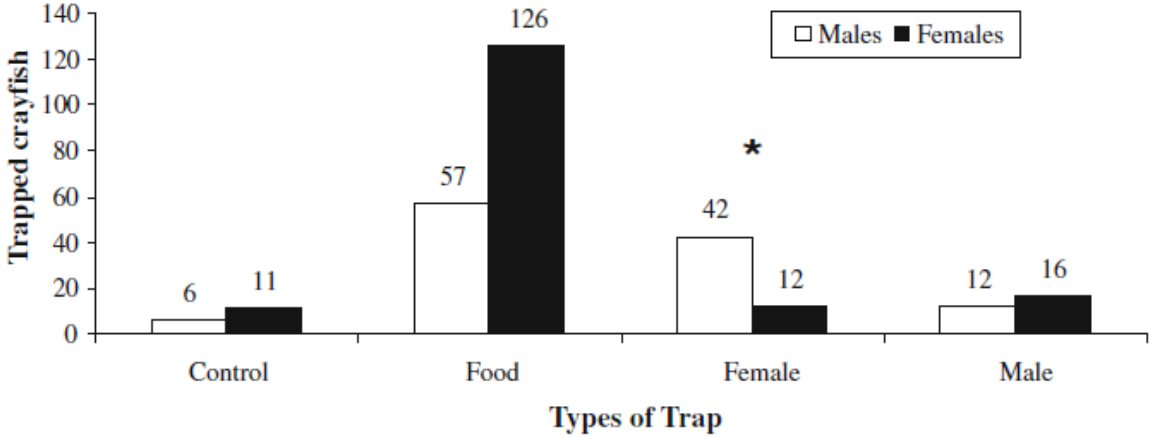
Control: empty trap (n=18)

Food (n=18)

Male: trap with sexually active male (n=18)

Female: trap with sexually active female (n=18)

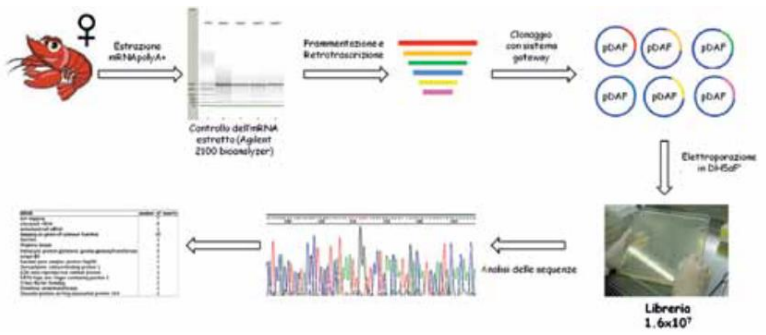
Righetti-La Monaca (Tuscany)

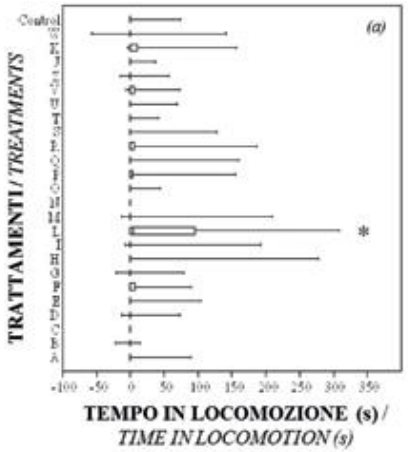


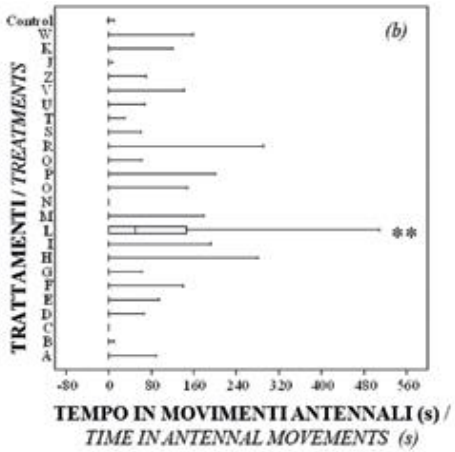
Aquiloni & Gherardi 2010

Pros: 1) species-specificity –the method can be used in the presence of indigenous crayfish– and 2) efficacy at low crayfish densities –it can be used as an early warning system.

Cons: sexual pheromones 1) are effective only during the reproductive season and 2) attract only males; 3) their chemical nature is unknown in crayfish –much money is needed for research.







Biological methods

(c) Autocides: SMRT

“Sterile male release technique”

Pro: encouraging results for the control of other freshwater organisms (the sea lamprey case).

But: the target species should be polygynous and its mating system should follow the Bateman’s principle –a few dominant males monopolize most copulations; sperm competition should not occur.

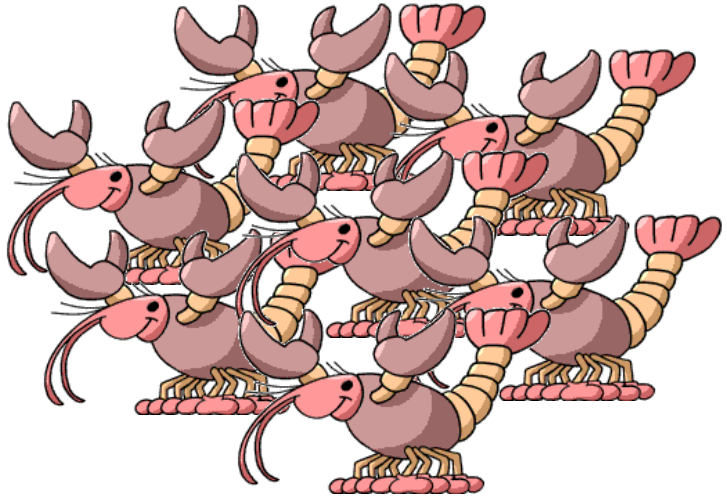
The first two requirements are met by *P. clarkii*.
From behavioral studies we know that large males 1) are dominant in intrasexual fights (Gherardi et al. 1999) and 2) are selected by females in mate choice (Aquiloni & Gherardi 2008).

Goal: change the reproductive output, but not the behaviour!

A sample of 40 sexually mature males of large size was subject to 20 Gy x-ray irradiation (5 minutes).

Clinical linear accelerator Philips S175

20 Gray



Histological analyses (18C+18T)

Sexual behaviour in 80 pairs (40 C+ 40T)

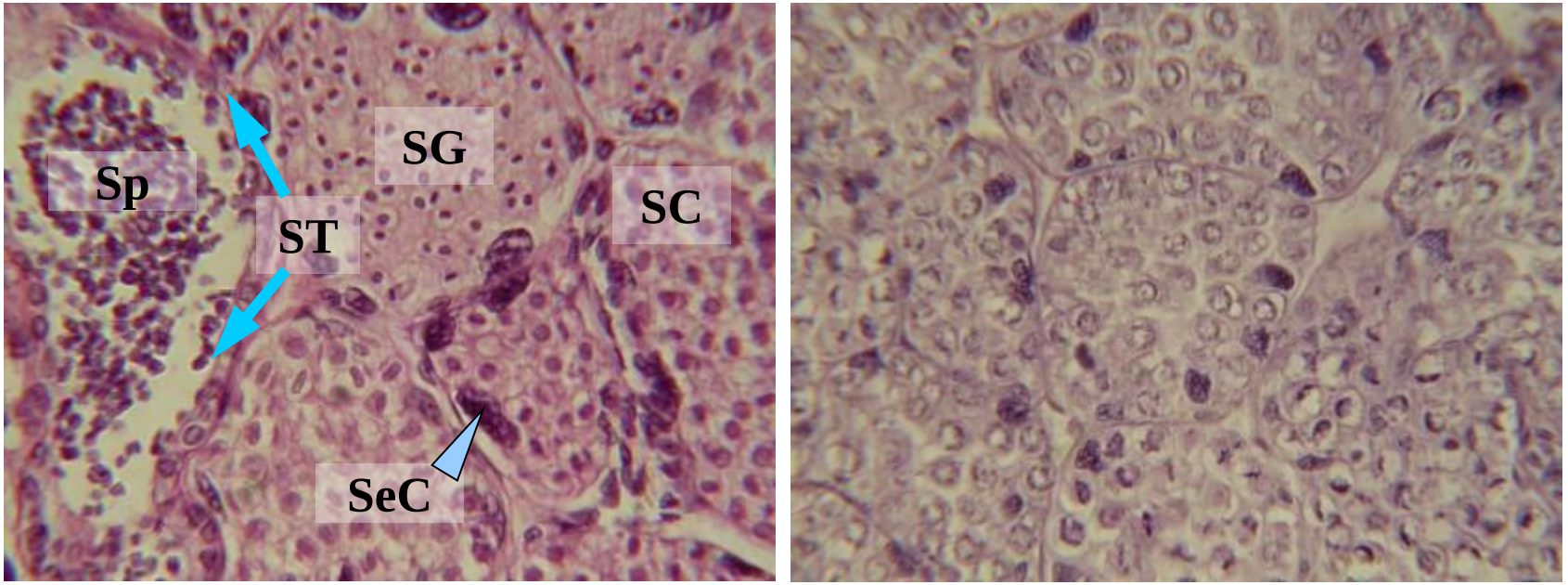
Reproductive output (eggs, offspring)

Histological analyses

X-ray irradiation affected male gonads by: 1) reducing the number of spermatogonia, 2) lowering the nuclear activity of seminal cells, and 3) causing necrosis of spermatocytes and edema in seminiferous tubules.

Control male

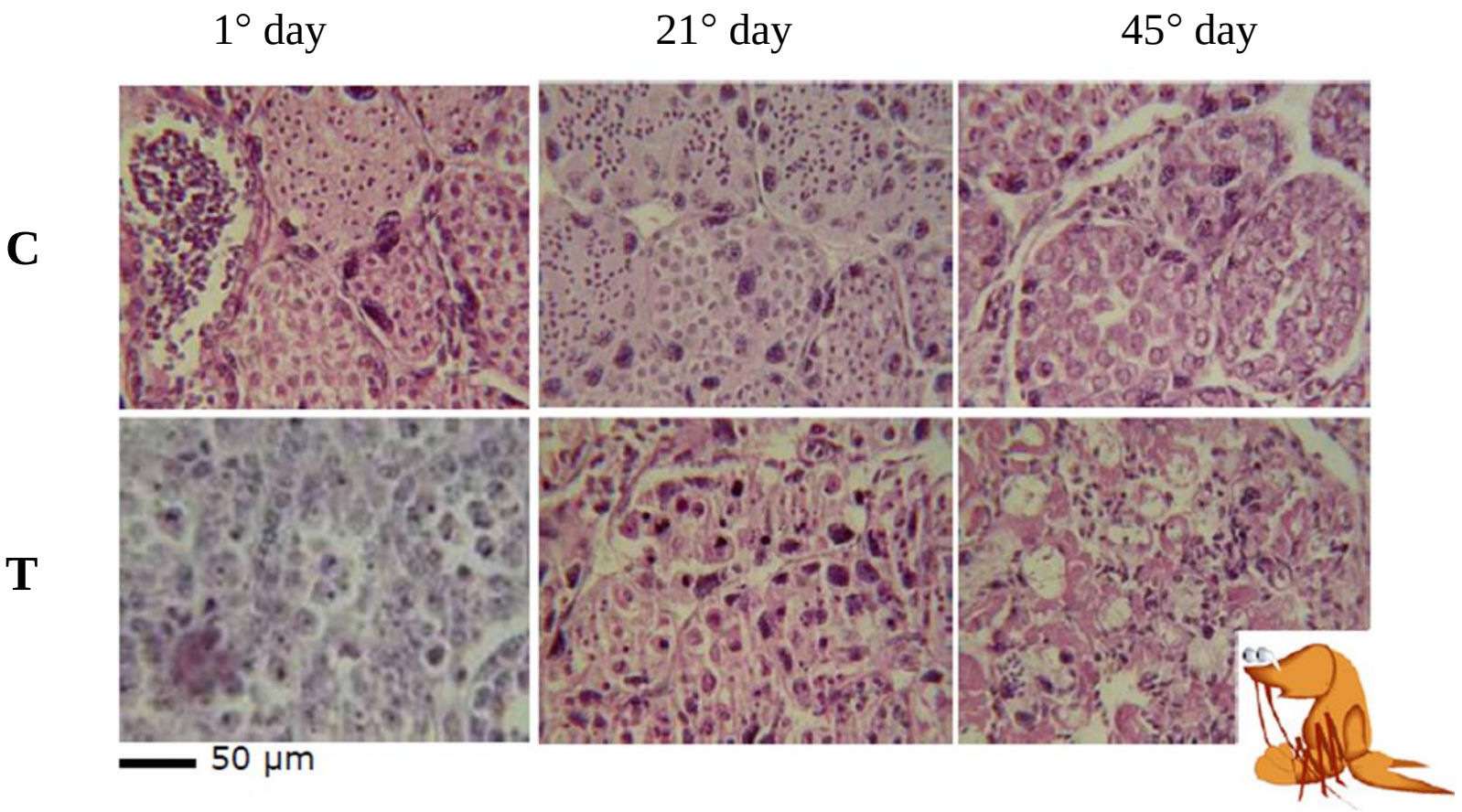
Irradiated male (1 day after)



ST=seminiferous tubules (arrows); SeC=Sertoli cells (arrowheads); SG= spermatogonia; SC= spermatocytes; SP=spermatids

Aquiloni et al. 2009

Histological analyses



Histological analyses

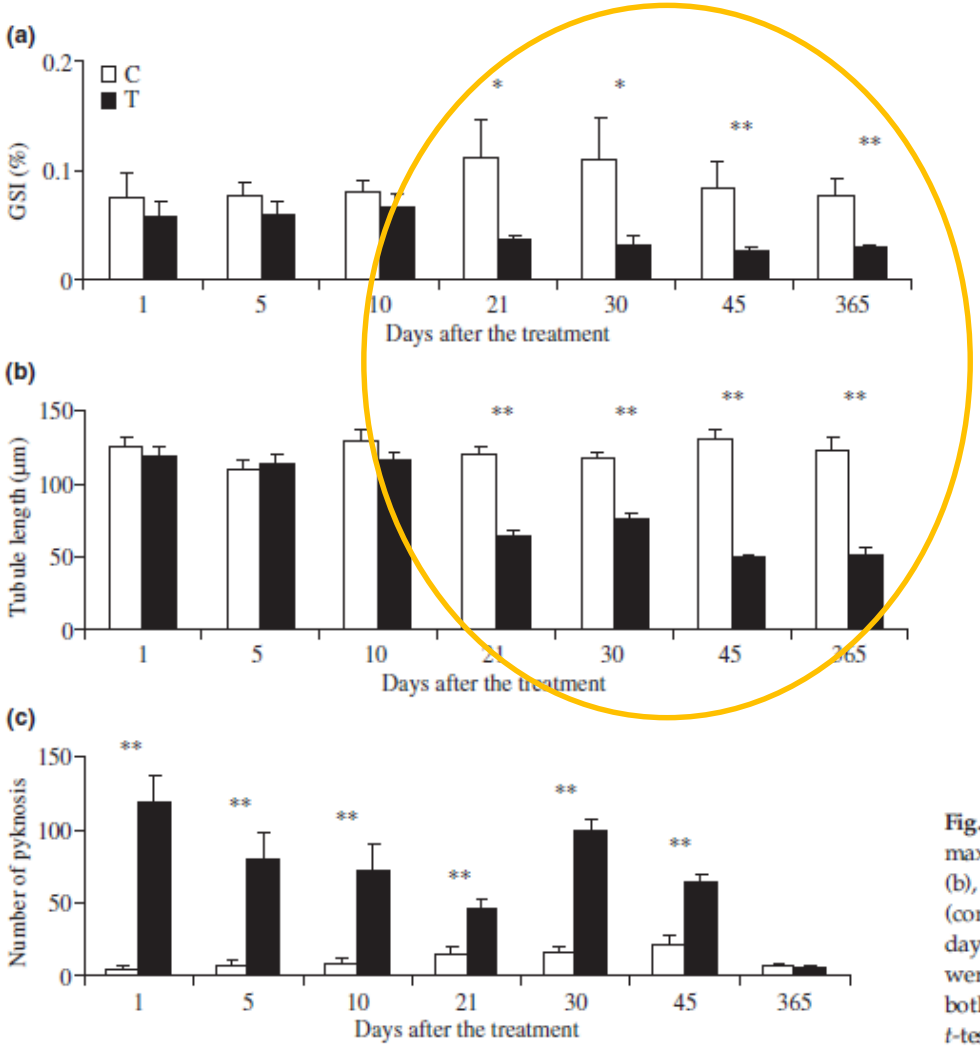
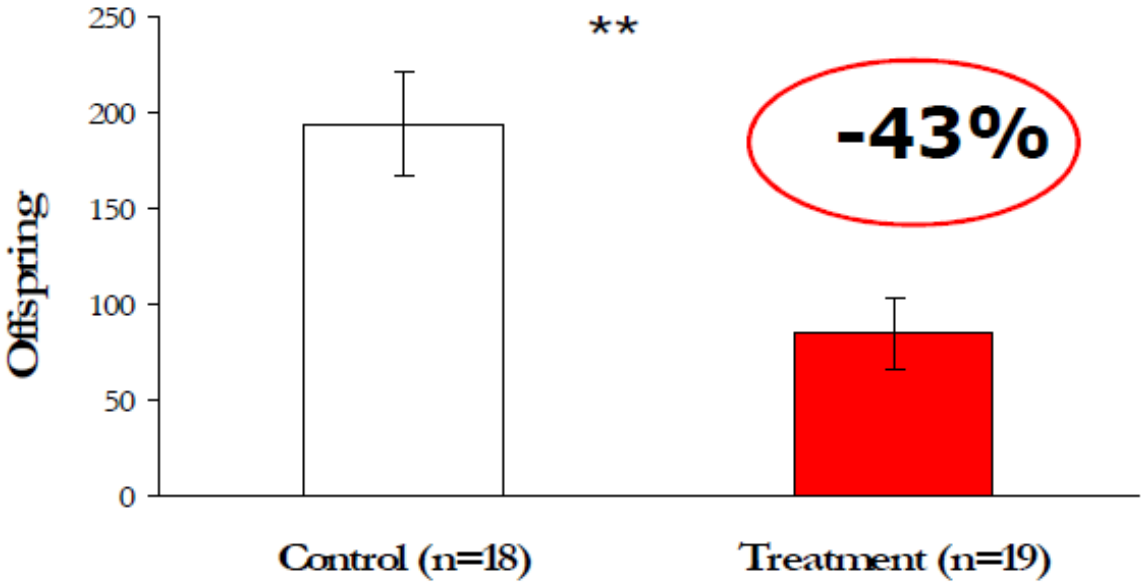


Fig. 2 Gonadosomatic index (GSI) (a), maximum length of seminiferous tubules (b), and number of pyknosis (c) between C (control) and T (irradiated) males after days from the treatment. Sample sizes were 21 in (a), 25 in (b) and 10 in (c) for both C and T males. One-tailed Student's *t*-tests; $*P < 0.05$, $**P < 0.001$

Aquiloni et al. 2009

Reproductive output



$t_{35}=3.035$, P=0.01

No difference for sexual behaviour.
A nearly significant difference for aborted eggs (> in T).

Aquiloni et al. 2009

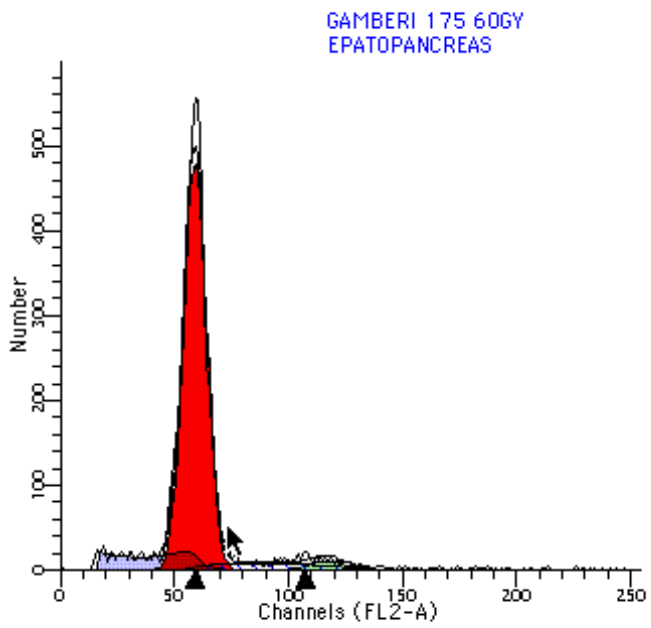
SMRT 2!

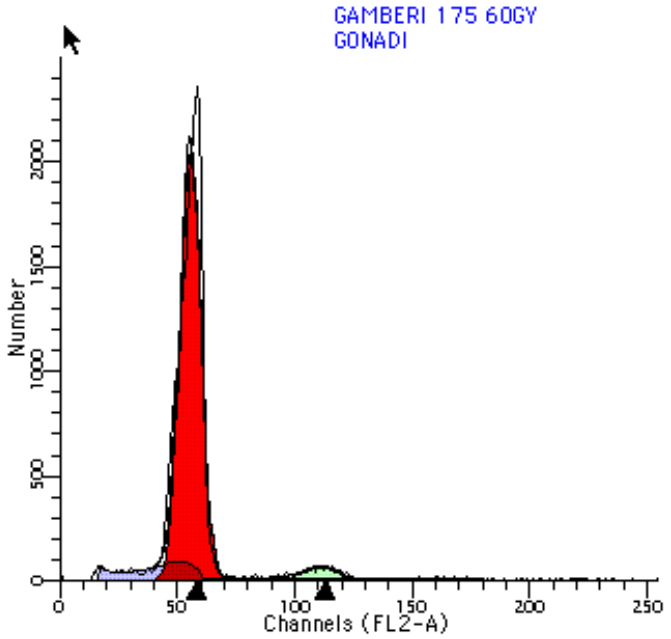
- Three different doses (20-40-60 Gy).
- Histological analyses, sexual behaviour, reproductive output.

- Application in the wild.

Cellular proliferation (Lab test)

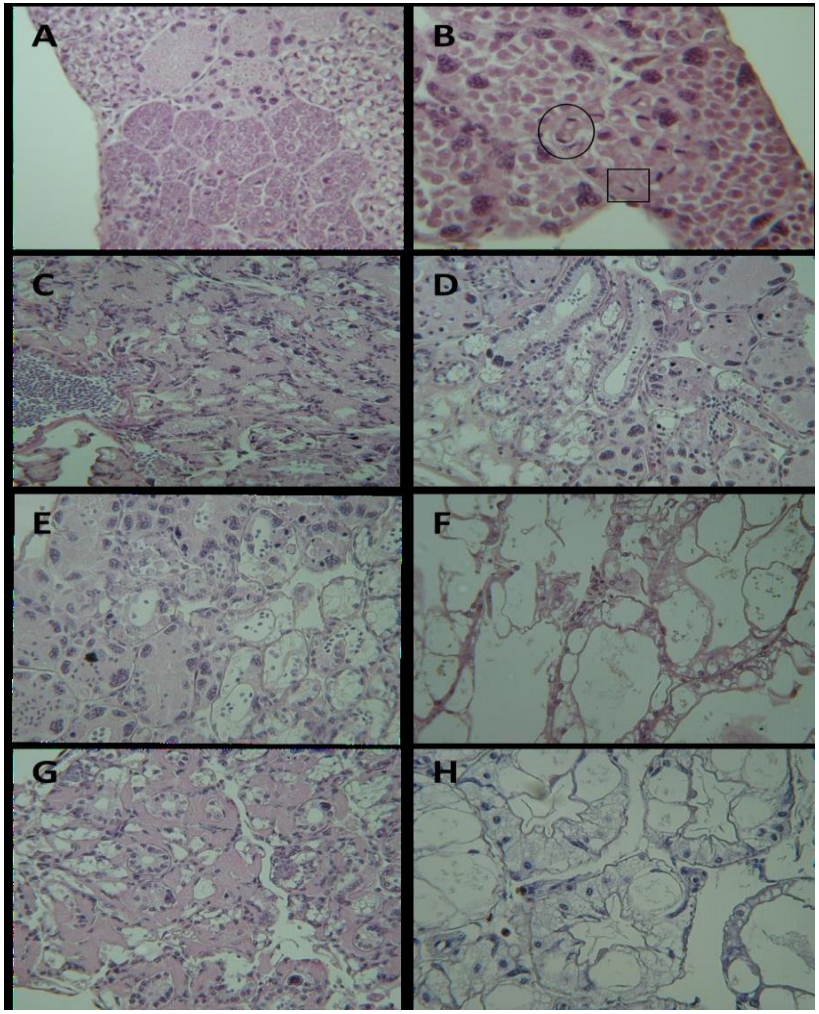
Flow cytometry

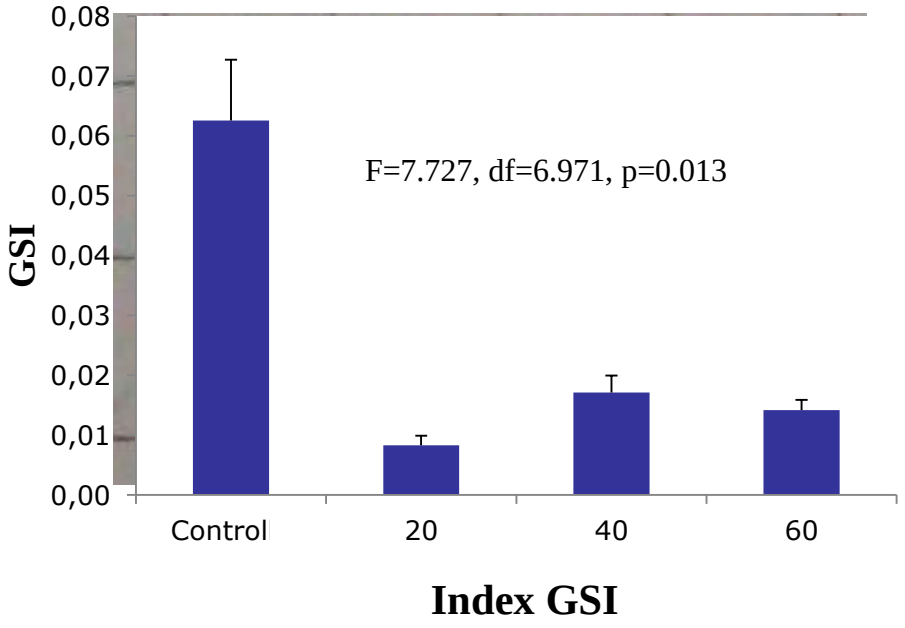




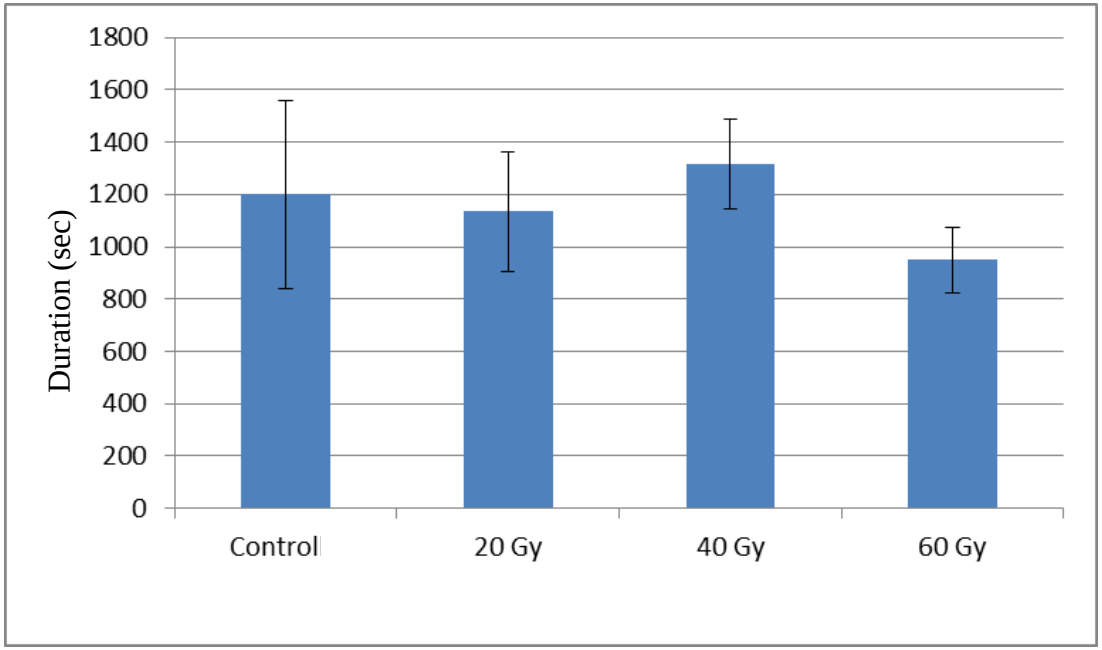
The spectrum of gonads belonging to treated animals (at each dose) shows no spermiogenesis and it is similar to a tissue without cellular proliferation (used as control).

Histological analyses (Lab test)



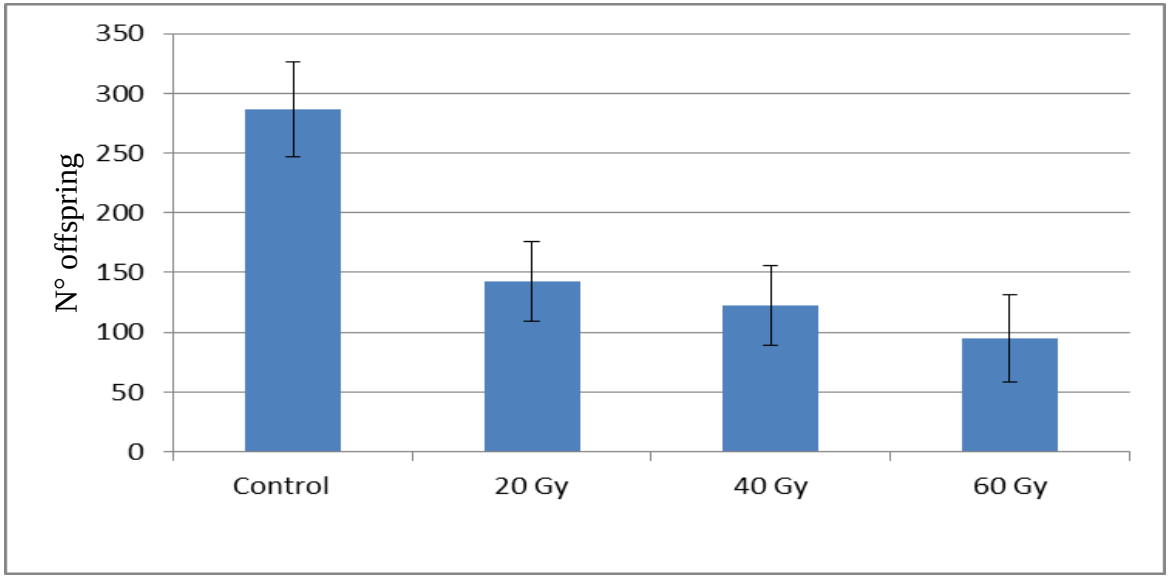


Sexual behaviour (Lab test)



The treated males did not differ from the control ones. No difference for duration and number (more than 80% for all males) of matings (a little decrease in duration for males treated at 60 Gy).

Reproductive output (Lab test)



Treatment induced a decrease in offspring of **50%, 57% and 67%** at the different X-ray dose.

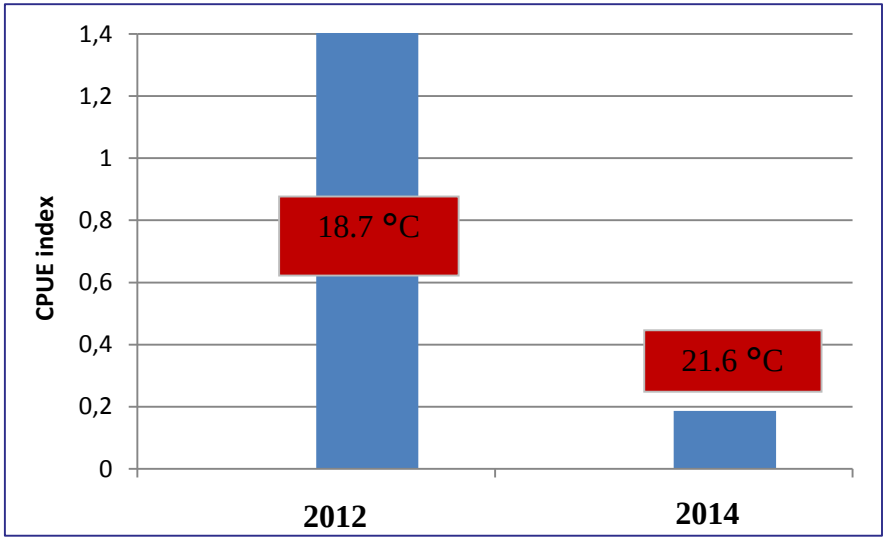
Application in the wild: Lake Casette (Pordenone, Friuli Venezia Giulia) (7 ha)

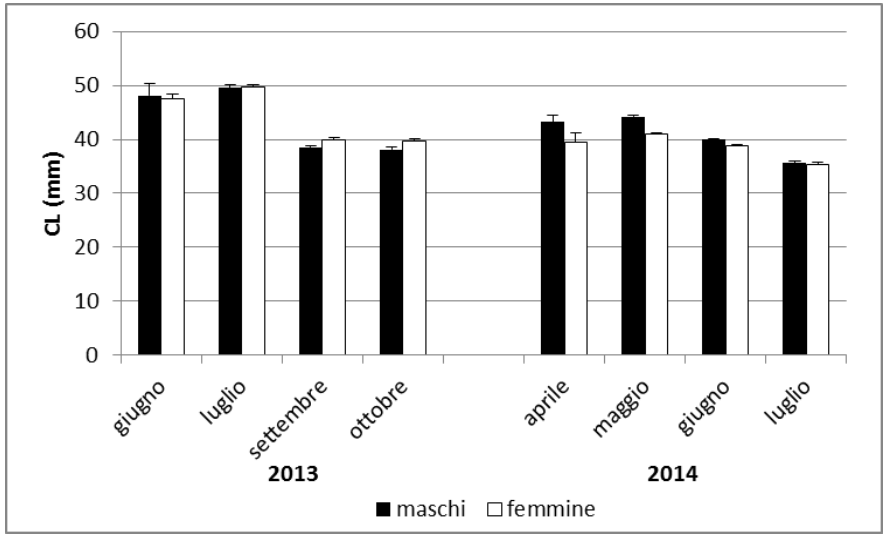
ESTIMATED POPULATION (CPUE): 10.419 individuals

Combination of trapping and SMRT

Individuals removed by traps: 4670

Released sterile males:
566 (at 20 Gy) in 2013 and
250 (at 40 Gy) in 2014





A population decrease of **87% in only two years of activity**

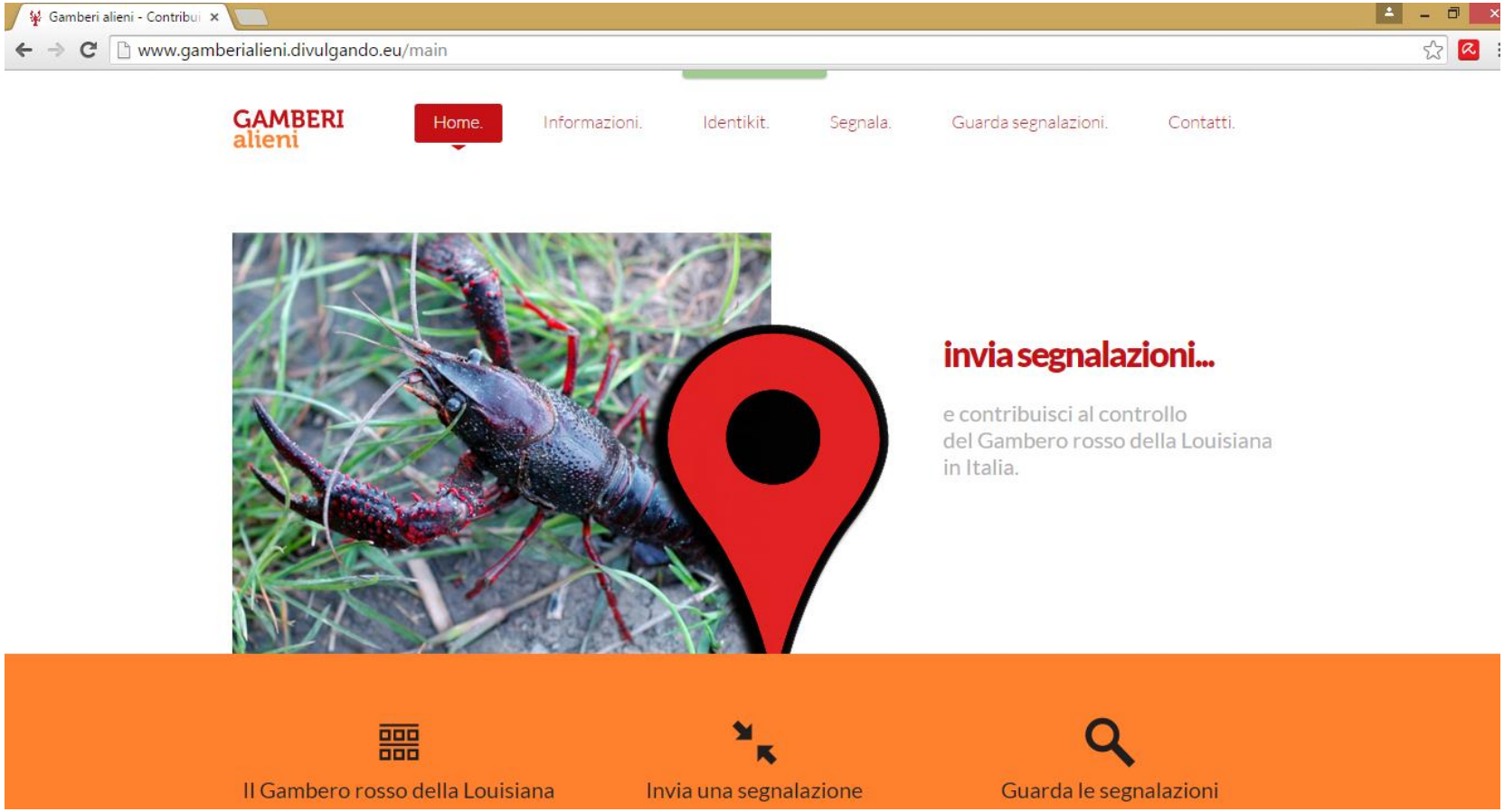
Reduction in size of caught individuals

Pro: low cost, non-invasive method, no harm for the habitat, good for restricted areas.

To sum up

- Improvements were achieved for some techniques (particularly SMRT).
- An unique and efficacious method for all the habitat types seems not to exist.
- The Integrated Pest Management approach, using a range of control and containment techniques to suit specific sites, is recommended to yield the best results.

Increase awareness!



THANK YOU!
GRACIES!
GRACIAS!