



A new insecticide based on *Clitoria* ternatea extract

ILARIA PERTOT

University of Trento, Center Agriculture Food Environment

LIVIA ZANOTELLI, OSCAR GIOVANNINI

Fondazione Edmund Mach, Research and Innovation Center



Clitoria ternatea L. - butterfly pea

- Fabaceae family
- Excellent forage legume (very good regrowth and yields)
- Cover crop
- Edible plant (young and tender parts of the plant, shoots, leaves, flowers and pods)
- Pharmacological properties

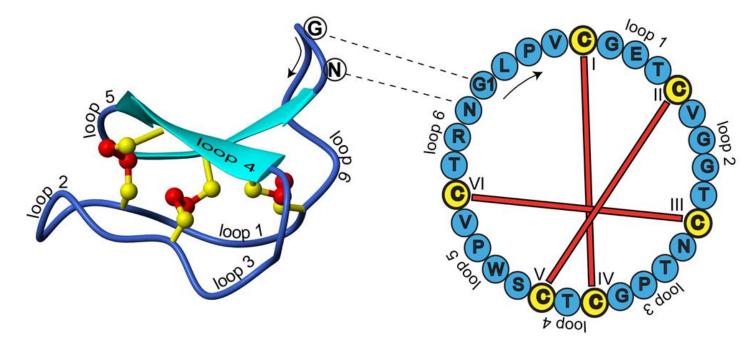




Insecticide properties

- Recent studies indicate that *C. ternatea* has insecticidal effects (cyclotides, flavonyl glycosides, proteins)
- Complex mechanism: ingestion, partially contact

Cyclotides: molecules composed of 28-37 amino acids in a head-to-tail cyclic backbone with three interlocking disulfide cystine bonds, mainly produced by plants as defence proteins





Insecticide properties

- **Cyclotides** are only found in four families (rubiaceae, violaceae, rabacae and cucurbitaceae), but they are widespread
 - extremely water soluble (around 1–10 g/L)
 - not readily degraded except in sunlight over weeks when the amino acid tryptophan is present
- Thousands of flavonoids present in plants: six are identified in C. ternatea extract
 - kaempferol, quercetin, myricetin, astragalin, kaempferol 3-neohesperiodoside and clitorin
 - low volatility, high water solubility and ready biodegradability
 - unlikely to persist or bioaccumulate



Toxicology

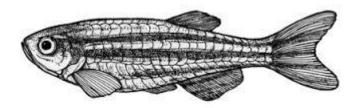
- Non toxic: low acute oral (LD50 > 2000 mg/kg bw) and dermal (LD50 > 2000 mg/kg bw) toxicity in rats
- Not a skin irritant in rabbits and the results of a skin sensitization study (local lymph node assay) did not provide evidence of a sensitisation potential
- The acute inhalational toxicity is likely to be low





Eco-Toxicology

- Non-toxic to mammals on an acute basis: no signs of toxicity when mammals were exposed to the highest level tested (oral and dermal routes)
- Fish, crustaceans, algae and aquatic plants: practically non-toxic an acute basis
- Bees: some sensitivity when tested at the highest concentrations
- Earthworms: some sensitivity for acute exposure at the highest concentrations in soils tested
- No phytotoxicity observed on tested crops





Possible uses of *C. ternatea* ethanolic extract against a wide range of phytophagous insects



Materials and methods

- Small scale trials under controlled conditions (lab, greenhouse)
- \bullet Concentration of the active ingredient in the formulated product: 400 g/l
- Dosage of the formulated product: 20 ml/l
- Untreated control (UTC): water
- Chemical standard reference
- Experiment carried out at least twice with 5 replicates/treatment
- Data of the experiments were pooled
- Statistics: ANOVA, Tukey's test ($\alpha = 0.05$)

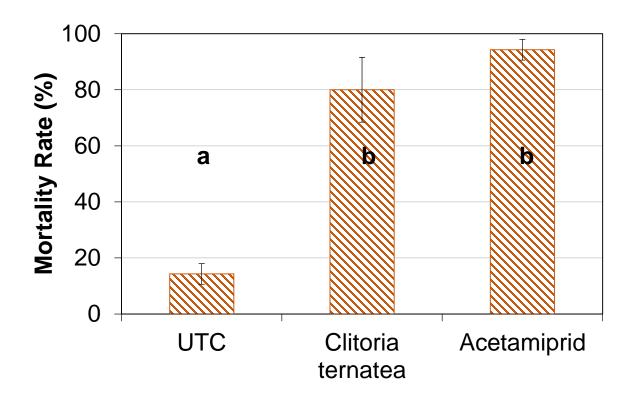


Target pests and chemical standard references and related dosages

Target	Reference Product 1		Reference Product 2		Reference Product 3	
	Active Ingredient	Dosage (ml or g/l)	Active Ingredient	Dosage (ml or g/l)	Active Ingredient	Dosage (ml or g/l)
Aphis gossypii	Flonicamid	0,14				
Antispila oinophylla	Acetamiprid	2,00				
Drosophila suzukii	Spinosad	0,20	Deltamethrin	0,70		
Frankliniella occidentalis	Abamectine	0,75				
Halyomorpha halys	Acetamiprid	2,00	Clorantraniliprole	0,18	Chlorpyrifos-Methyl	4,00
Lobesia botrana	Emamectine Benzoate	1,50				
Scaphoideus titanus	Flonicamid	0,14				
Trialeurodes vaporariorum	Abamectine	0,13				







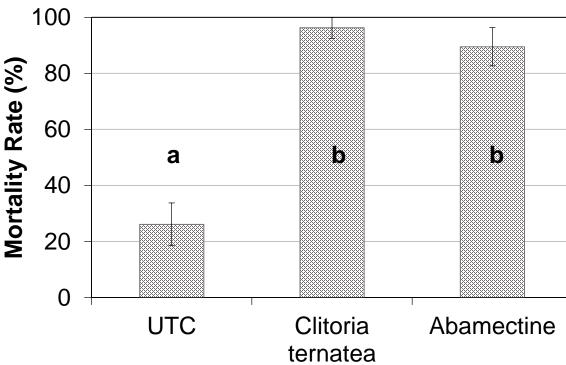
Antispila oinophylla

Mortality of *A. oinophylla* larvae, 40 hours after the treatment, was similar to chemical reference

Treatments (drops) were applied on leaves, above the mines containing active larvae inside, and incubated in Petri dishes; two experiments pooled





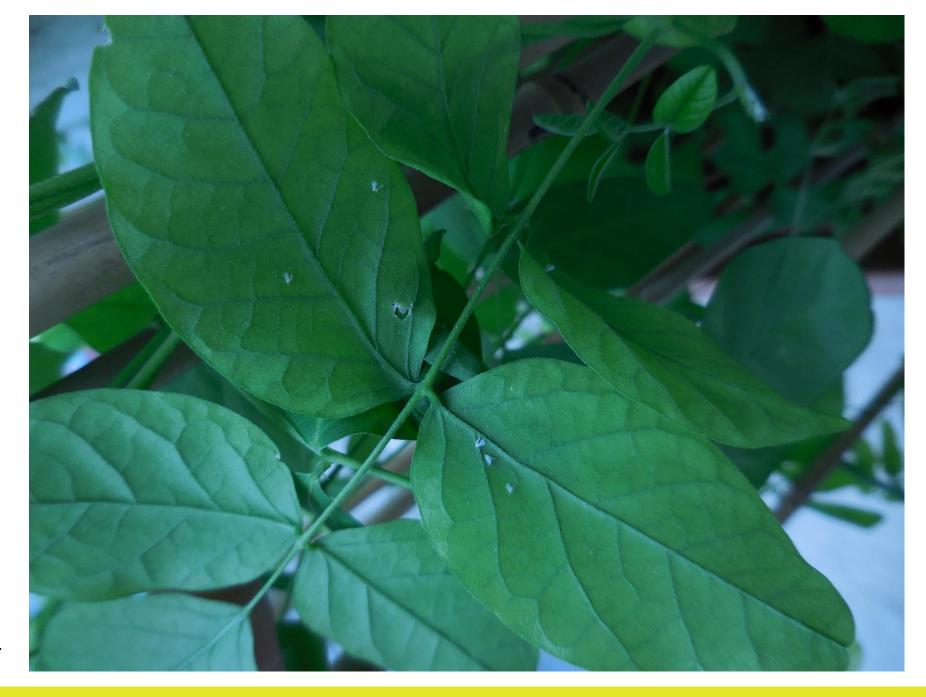


Trialeurodes vaporariorum

Mortality of nymphs, recorded as the number of specimens that did not emerge from the 'pupa' in the three weeks following the treatment

Treatments were carried out on tobacco leaf disks infested by nymphs; two experiments pooled

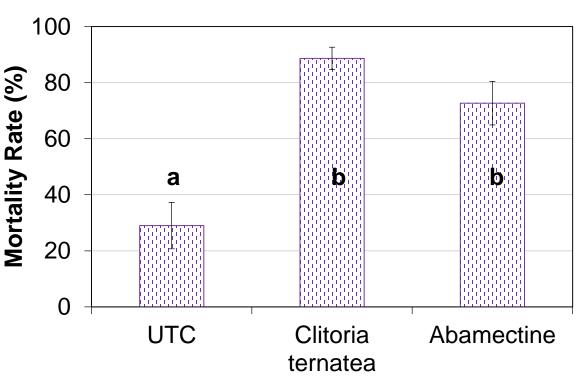




By Andrea Nesler





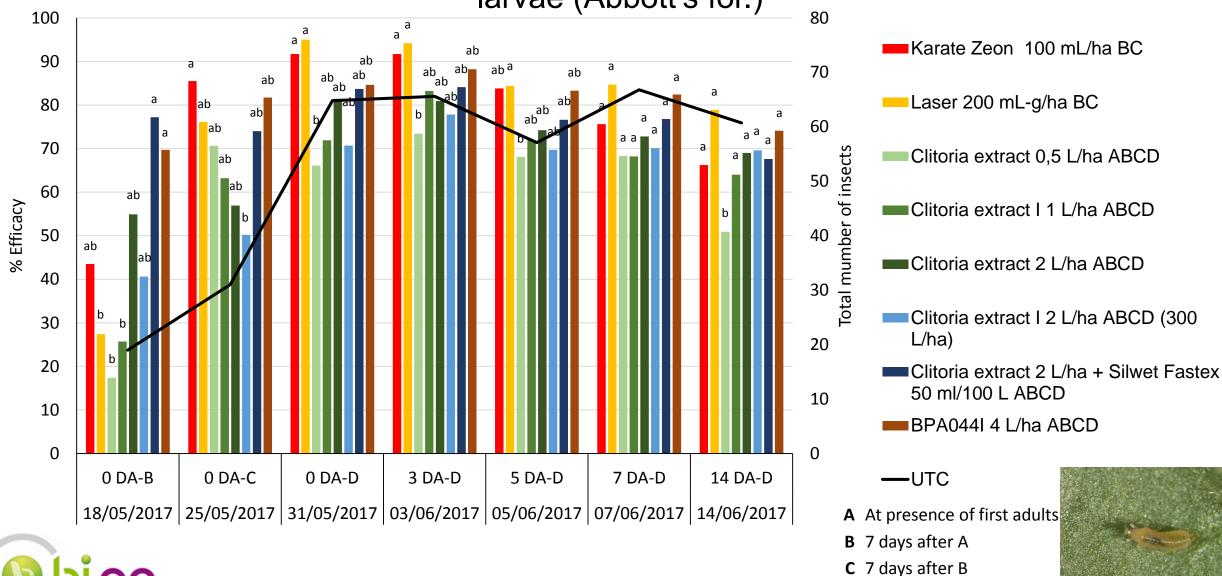


Frankliniella occidentalis

Mortality of nymphs, 48 hours after the treatment

Treatments were carried out on insects placed on bean leaf disk and then incubated in Petri dishes; two experiments pooled

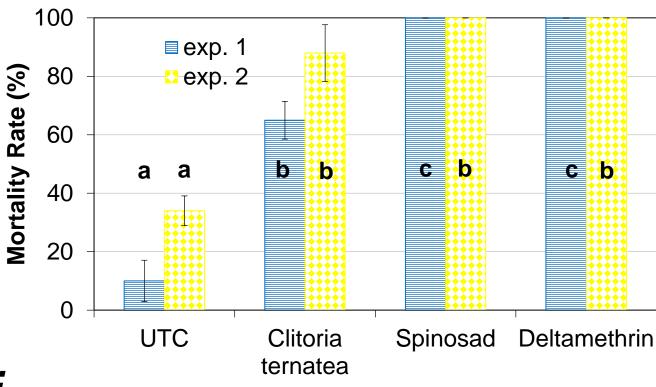
Insecticide efficacy calculated on total number of insects: adults and larvae (Abbott's for.)



D 7 days after C







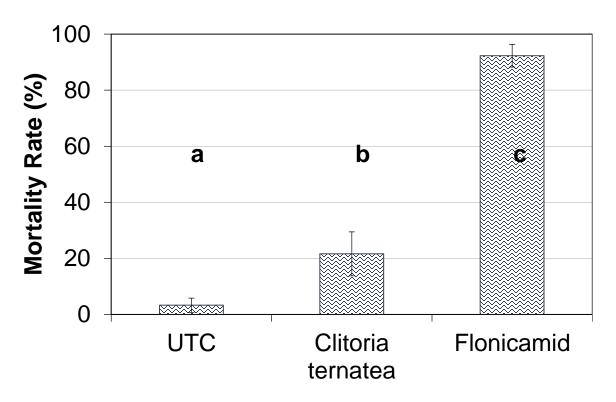
Drosophila suzukii

Mortality of adults 48 hours after the treatment

Treatments were sprayed directly on insects, which were then incubated in Petri dishes; some variability in the efficacy between trials (exp1 and 2) was noticed







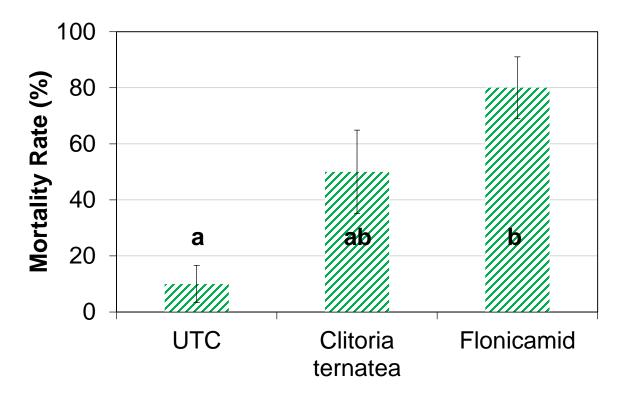
Aphis gossypii

Mortality of nymphs, 4 days after the treatment, was lower than chemical reference, but higher than UTC

Treatments were carried out on insects placed on zucchini leaf disk in Petri dishes; two experiments pooled







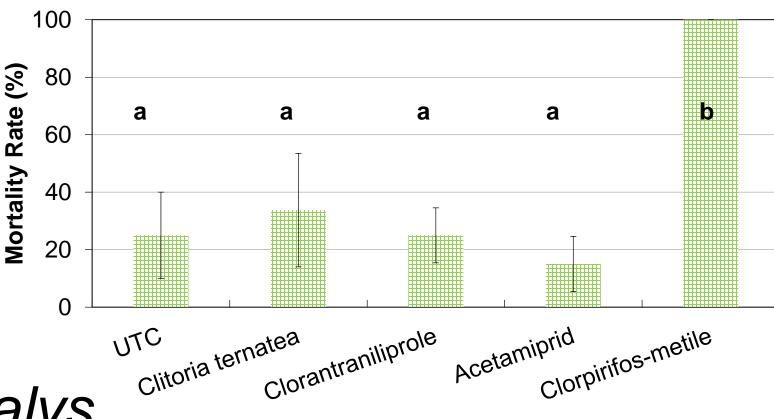
Scaphoideus titanus

Mortality of nymphs, 7 days after the treatment

Treatments were carried out on grapevine leaf disks, insects were then placed on the leaf disks and incubated in Petri dishes







Halyomorpha halys

Mortality of adults up to 14 days after the treatment

Treatments were carried out on crabapples, insects were then placed on the apples and incubated in boxes; two experiments pooled



Conclusions

- No toxicity for mammals
- Almost no toxicity for the environment
- Good persistency and good efficacy against several target insect species (but not all)
- Selectivity (expected)
- No phytotoxicity







Thank you for your attention!





Andrea Nesler Sandro Frati