



### ■ THE PROBLEM ADDRESSED

## The main threat to citrus industry has no biological control agents in Europe

The citrus greening or “huanglongbing” (HLB) disease is one of the most devastating citrus diseases in the world (Bové, 2006). This disease is associated with three phloem  $\alpha$ -proteobacteria: ‘*Candidatus Liberibacter asiaticus*’ (CLas), ‘*Ca. L. americanus*’ (CLam) and ‘*Ca. L. africanus*’ (CLaf). HLB is transmitted by the African citrus psyllid *Trioza erytreae* (Del Guercio) (Hemiptera: Triozidae) and the Asian citrus psyllid *Diaphorina citri* Kuwayama (Hemiptera: Liviidae). None of these insect vectors was present in mainland Europe until 2014, when *T. erytreae* was detected in north-western Spain (Cocuzza *et al.*, 2017). Since then, the psyllid has spread towards the southwest of the Portuguese coast reaching Lisbon province (DGAV, 2019). Although the psyllid has not reached the main citrus-producing areas and HLB has not been detected (Siverio *et al.*, 2017), the spread of *T. erytreae* in mainland Europe has alarmed citrus growers from the Mediterranean countries. Recent studies carried out by TROPICSAFE and other research groups have demonstrated that the psyllid vector has no potential biological control agents that can control it in Europe.



- Left: citrus flush infested by *Trioza erytreae* with open gall-like structures produced by *T. erytreae* nymphs. Right: different stages of *T. erytreae*: eggs, nymphs and adults recently emerged.

### ■ THE PRACTICE/INNOVATION PROPOSED BY TROPICSAFE

## A parasitic wasp to prevent HLB in Europe

One of the main aims of TROPICSAFE is to develop advanced and novel pest management strategies that provide a reduction in the environmental impact of plant protection approaches. TROPICSAFE has initiated a classical biological control program to introduce the parasitoid *Tamarixia dryi* (= *Tetrastichus dryi*) (Waterston) (Hymenoptera: Eulophidae) from South Africa. This parasitoid is the most abundant and effective biological control agent of *T. erytreae* in Africa. The parasitoid was introduced in the Canary Islands, an archipelago in the Atlantic Ocean belonging to Spain. After rearing it and testing its specificity in collaboration with the Canarian Institute of Agricultural Research (ICIA) (Pérez-Rodríguez *et al.* 2019; Urbaneja-Bernat *et al.*, 2019), it was released in the spring of 2018 in the Canary Islands and in fall of 2019 in the northwest of Iberian Peninsula (Galicia, mainland Spain).



## ■ HOW IS TROPICSAFE IMPLEMENTING IT?

### Release of *Tamarixia dryi* in the field

Taking into consideration the success of *T. dryi* in other areas, and the results obtained under laboratory conditions, TROPICSAFE together with ICIA and the “Servicio de Sanidad Vegetal de Canarias” reared, released and monitored the parasitoid *T. dryi* in the field. The parasitoid was released in the spring of 2018 in a citrus orchard that belongs to ICIA and is located in La Laguna, northeast of Tenerife.

After its release, the spread of *T. dryi* was measured during the fall and winter of 2018 and 2019. For this, citrus trees from different localities throughout the Tenerife Island (60 × 40 km) were sampled to determine the presence of the parasitoid; the parasitism rates of *T. dryi* were also measured in two citrus orchards in 2019. For this, shoots infested with *T. dryi* were collected and transported to the laboratory. Once there, the number of alive and parasitized psyllid nymphs were counted under a binocular microscope to determine the potential of the parasitoid in field.

The same procedure was followed in mainland Spain (Pontevedra, Galicia) where *T. dryi* was released in three localities in the fall of 2019 and the spring of 2020. The spread of *T. dryi* was measured during the summer of 2020 and 2021 with the collaboration of the “Servicio de Sanidad Vegetal de Galicia” and the public companies TRAGSA and TRAGSATEC.



- The parasitoid *Tamarixia dryi* parasitizing the psyllid *Trioza erytreae* in the laboratory (left). A detail of the release of *T. dryi* in the field, the parasitoids are leaving the tube where they were transported (right).

## ■ HOW IS IT WORKING?

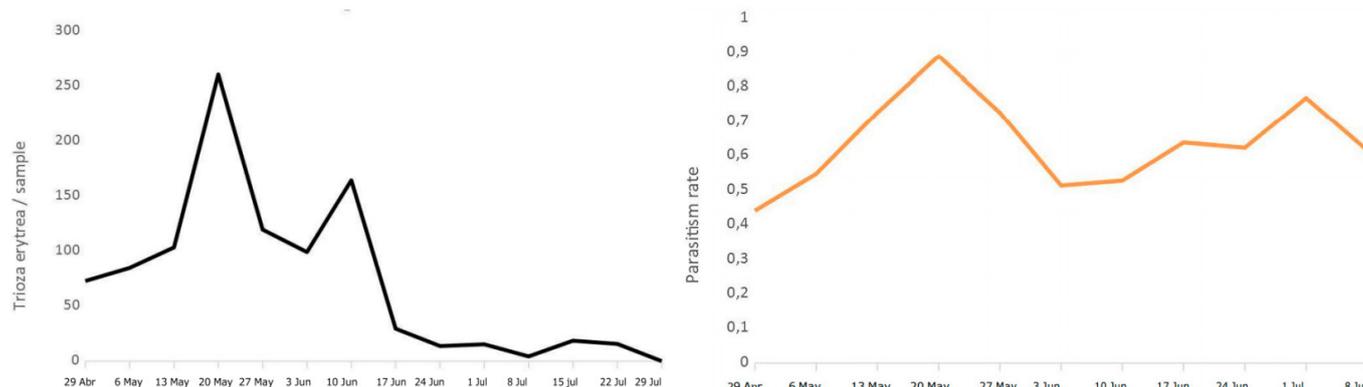
### *Tamarixia dryi* controls *Trioza erytreae* in the Canary Islands

The field data demonstrates that *T. dryi* spread rapidly throughout the Tenerife Island. After its release in the northeast of the island in spring of 2018, the parasitoid was detected in the west and south of Tenerife (50 km from the release point) in the fall of 2018. Moreover, the parasitoid was recovered in 80% of the 83 locations sampled in the fall and winter of 2018 and 2019. Taking into consideration the different climates of the north (humid) and south (dry) parts of the island, its rapid spread suggests that *T. dryi* will be able to establish and spread once it is released in the Iberian Peninsula.

The dynamics of *T. erytreae* and the parasitism rates by *T. dryi* were measured in two citrus orchards that had high levels of the psyllid in previous years. The levels of *T. erytreae* were extremely low in 2019, likely by the introduction of the parasitoid *T. dryi*. In the two citrus orchards sampled in 2019, parasitism rates reached almost 90% and the



populations of the psyllid did not recover after summer, even when new flush was available. These results demonstrate the efficacy of *T. dryi* as biological control agent of *T. erytreae* in the field in Tenerife.



- Seasonal trend of *Trioza erytreae* and parasitism rate by its parasitoid *Tamarixia dryi* in the Canary Islands in 2019.

## *Tamarixia dryi* controls *Trioza erytreae* in mainland Spain

The field data demonstrates that *T. dryi* spread throughout Pontevedra between the summer of 2020 and 2021. After its release in three localities from Pontevedra, the parasitoid spread up to 2 km from the releasing point in 2020 and more than 40 km in the summer of 2021. Therefore, *T. dryi* has been also able to establish and spread in the Iberian Peninsula.

In the summer of 2020, the percentage of parasitism in the citrus shoots from spring was  $38.9 \pm 3.2\%$  in Romai,  $25.7 \pm 3$  in O Grove and  $7.6 \pm 2.3\%$  in O Rosal. In O Grove, it was also possible to calculate the level of parasitism in the citrus shoots from summer. The percentage of parasitism reached  $75.2 \pm 3.6\%$ . All the parasitized nymphs of *T. erytreae* were parasitized by *T. dryi* and hyperparasitoids were not recovered.

In the summer of 2021, the density of *T. erytreae* decreased dramatically, likely due to the presence of the *T. dryi*, and it was not possible to calculate the percentage of parasitism. After this success, the parasitoid has been released in all the areas infested by *T. erytreae* in mainland Spain and Portugal.



- A colony of nymphs of *Trioza erytreae* parasitized by the parasitoid *Tamarixia dryi* in Galicia in summer 2020.

**KEY WORDS**

Citrus, integrated pest management (IPM), classical biological control, *Tamarixia dryi*, *Trioza erytreae*

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