

Invasion of *Ceratitis capitata* W. (Diptera, Tephritidae) from coastal to inland areas of Dalmatia region of Croatia: E-traps as an improved detection tool

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Abstract: The Mediterranean fruit fly, *Ceratitis capitata* (Wied.), is a pest of great economic importance worldwide and in the coastal areas of Croatia. After the introduction and establishment of this pest in the coastal areas, there was no evidence of its spread to the less favourable neighbouring areas north of the coast. In order to investigate the spread of *C. capitata* to inland areas, a grid of conventional and electronic traps was established in 25 sites, at the edges of the pests' current distribution, where populations expand and contract their range seasonally. For each site, data was collected on host species, habitat description, elevation and geographic location using a GPS. Two types of conventional traps were used to monitor *C. capitata* population: Tephri traps baited with 3C lure (tri-methyl amine, putrescine, and ammonium acetate) and Jackson traps baited with trimedlure. A new improved detection tool, e-traps, was developed under the EU Horizon 2020 project “Fruit flies In Silico Pest Management (FF-IPM)”. This improved method of fruit fly detection and monitoring was incorporated into a conventional network of traps. The e-traps were set using locally purchased SIM cards and batteries. A total of 12 e-traps were successfully set at six sites (two e-traps per site, one with 3C bait and one with trimedlure) in trapping stations adjacent to existing Tephri traps and Jackson traps. Traps were inspected at weekly intervals and the number of males and females of *C. capitata* and other non-target insects were recorded. Surveys conducted in 2020 and 2021 showed that *C. capitata* has spread and invaded large parts of the sub-Mediterranean inland area of Split – Dalmatia and Šibenik – Knin counties. The e-traps provided a good early warning of the presence of the *C. capitata* in coastal and invaded areas.

Key words: Mediterranean fruit fly, Dalmatia, spreading, e-traps; Tephri traps, Jackson traps

Introduction

The Mediterranean fruit fly, medfly, *Ceratitis capitata* (Wiedemann 1824; Diptera, Tephritidae) is considered one of the most significant tephritid fruit fly species worldwide. Medfly is an extremely dangerous polyphagous pest of fruits and vegetables reported from five different continents in temperate, subtropical and tropical climates (Gilstrap et al., 1987). Originated from tropical Sub-Saharan Africa (De Meyer et al., 2002), *C. capitata* spread to many Mediterranean countries after introduction through the Iberian Peninsula in 1842, to Italy in 1863 and to France 1885 (Fimiani, 1989) and furthermore along temperate areas of Mediterranean (Deschepper et al., 2021). *C. capitata* is the most important invasive tephritid

fruit fly species in fruit production worldwide affecting over 260 different plant species. In temperate areas of Mediterranean, like northern Greece and coastal Croatia, due to its high biotic potential and adaptability to different ecological conditions, medfly complete four to five generations per year (Papadopoulos et al., 2001; Bjeliš et al., 2014). In the Mediterranean area, the distribution of the medfly has traditionally been considered limited to the 41st parallel north (Robinson and Hooper 1989; Israely et al., 2004). However, several publications have reported the ability of medfly to overwinter as larvae in cold winter conditions and survive at temperatures below freezing (Papadopoulos et al., 1996; 2001). Recent experiments conducted in Central Dalmatia in Croatia confirm the possibility that the *C. capitata* can successfully overwinter as a pupa (females emerged of overwintering pupae remained alive until the beginning of August) and as an adult (some adults from the basement in an urban area remained alive until the end of spring) at the basement of the houses and continue its biological cycle as soon as seasonal weather conditions are suitable for the development of the species (Bjeliš et al., 2020).

Materials and methods

Area description

The experimental area is located between 43.2753 to 44.3135 north and 16.1244 to 16.4227 east geographic coordinates, covering over 2600 km² of highland areas. According to Köppen's classification (Köppen, 1936), the study area is characterized by a sub-Mediterranean climate, characterized by hot, dry summers, and cool wet winters (Cfa). This type of climate differs from the Mediterranean climate mainly in the slightly higher amounts of precipitation and their more even distribution throughout the year, although the summers are also quite dry. Main medfly host plants in the target area are apple – *Malus domestica* L., pear – *Pyrus communis* L., peach – *Prunus persicae* and plum – *Prunus domestica* L. and much rarely represented are quince – *Cydonia oblonga* L. and fig – *Ficus carica* L.

Trapping methodology

Three different trapping systems were used in the research. Two conventional types of traps were used to detect and to monitor the medfly population: Tephri traps baited with 3C lure (tri-methyl amine, putrescine, and ammonium acetate) and Jackson traps baited with trimedlure. Conventional traps were active during the summer-fall (trapping season) of 2020-21. A new improved detection tool, e-traps, was used to evaluate its performance and ability to provide constant daily information and early warning alerts (not yet automatized) of the *C. capitata*. The e-traps were set using locally purchased SIM cards and batteries and were active during the trapping season of 2021. The e-traps were incorporated into a conventional network of traps in order to contrast their trapping capabilities. A total of 12 e-traps were successfully set at six sites (2 e-traps per site, one with 3C lure and one with trimedlure) in trapping stations adjacent to existing Tephri traps and Jackson traps. Conventional traps were inspected at weekly intervals and the number of males and females of *C. capitata* was recorded. E-traps uploaded images were inspected on a daily basis.

Results and discussion

Medfly detection results show that the pest was detected at two sites in Šibensko – kninska County and at six sites in Splitsko – dalmatinska County (Figure 1, Table 1). Before 2000, the

medfly was found only in coastal regions (Figure 1). The geographic locations (coordinates and elevations above sea level) where medfly was caught in 2020-21 are summarized in Table 1. Geographic locations (coordinates and elevations above sea level) where *C. capitata* were captured in 2020-21 are summarized in Table 1.

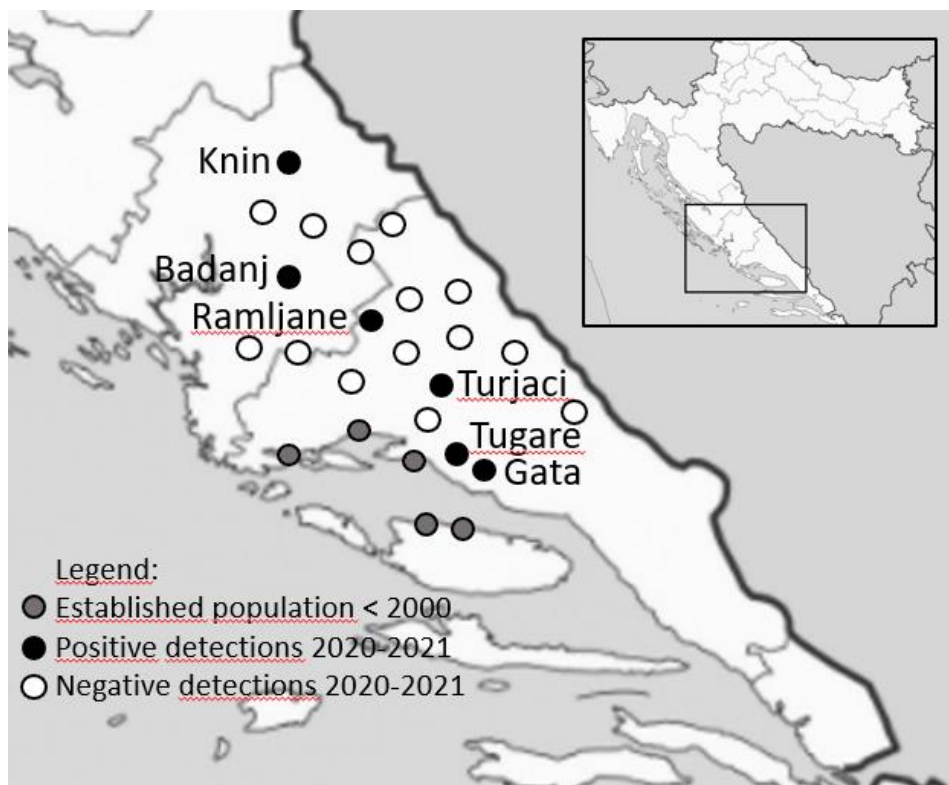


Figure 1. Trapping network in Splitsko – dalmatinska and Šibensko – kninska counties and *Ceratitis capitata* detections during 2020-2021.

Table 1. List of the sites with *Ceratitis capitata* detections during 2020-2021.

Location	Northing	Easting	Altitude [m]	Number of captures in 2020	Number of captures in 2021
Šibensko – kninska County					
Badanj	43°51'59.6016	16°11'5.6292	319	6	0
Knin	44°3'13.5504	16°12'44.5824	241	1	0
Splitsko – dalmatinska County					
Ramljane	43°44'50.4456	16°21'35.5896	408	1	0
Gata	43°27'53.2512	16°42'27.7056	341	28	1
Turjaci	43°39'37.278	16°39'33.93	311	28	3
Tugare	43°28'25.5396	16°38'12.174	255	2	0

The first pest detection (1 female) in 2020 occurred in Knin (June 30, 2020) in a small orchard of apples (*Malus domestica* L.) and pears (*Pyrus communis* L.). After this discovery, the capture of *C. capitata* was recorded at intervals of a few days at Tugare (one female) on a peach tree (*P. persica* L.) and Split (two females and three males) on a fig tree (*F. carica* L.). In 2021, *C. capitata* was detected for the first time at the Podstrana site, in the Tephri 3C traps on 10/08 and in the E trap on 14/08. Detections of *C. capitata* at the Badanj, Knin, Ramljane and Tugara sites from 2020 were not observed in 2021. Similarly, captures at the Gata site in 2020 in conventional traps were not observed in Tephri 3C traps in 2021, but one capture (one male) was recorded in the e-trap (28/09/2021) set on a single apple tree (*M. domestica*) at this site in 2021.

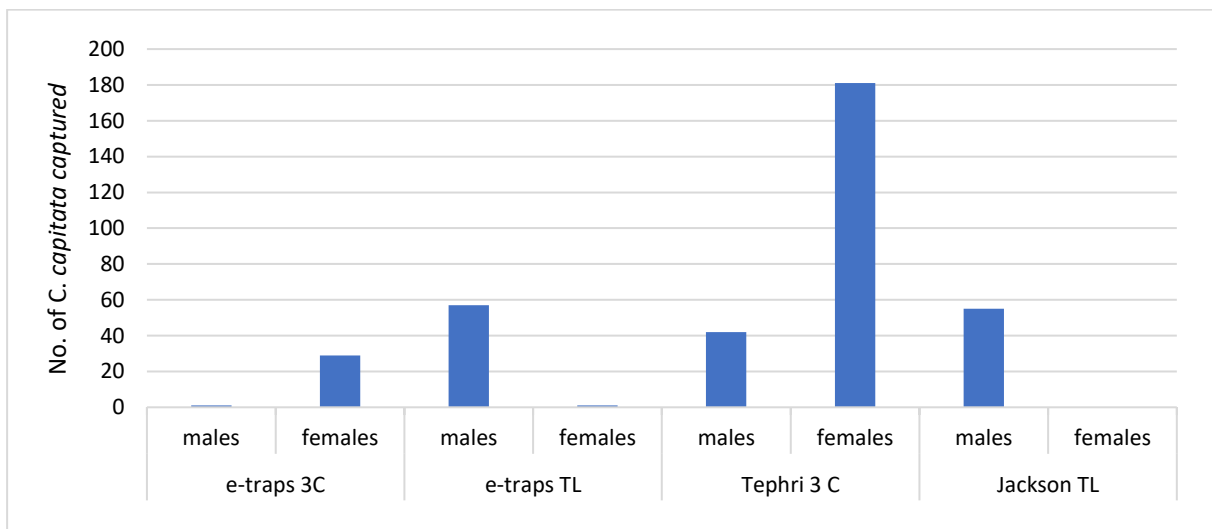


Figure 2. Total captures of *Ceratitidis capitata* on different combinations of traps (e-trap, Tephri and Jackson traps) and lures (3C; TL-trimedlure) at Podstrana site during 2021.

Figure 2. shows the total catch in Podstrana in all traps during the 2021 season. We can note that the largest catch of female *C. capitata* was made in Tephri traps baited with the 3 components (3C). A total of 181 females were collected in this trap throughout the season, in contrast to only 29 female medflies caught in the E trap baited with 3C during the same period. Tephri traps baited with 3C also captured 42 males; few males were captured in the E-trap baited with 3C. There was less difference in catches in the traps baited with Trimedlure compared to the conventional Jackson traps and the e-trap. The Jackson trap captured a total of 55 males throughout the season, while the e-traps had a slightly smaller lead with a total of 57 males captured during the monitoring period.

Figure 3 describes the capture of *C. capitata* in Podstrana during the different months of 2021. In early December, large numbers of adult *C. capitata* were confirmed in Podstrana. Podstrana is located at sea level, so autumn temperatures in December have not yet affected adult medfly populations (unlike higher elevation sites where medflies were not caught at this late date). The earliest captures of females in this region were observed in August in both the E trap and the Tephri trap baited with Trimedlure. The early detection of one female in the E-trap (Trimedlure) on a single tree of *P. persica* L. (peach) in August is interesting because of the nature of the bait and the fact that the E-trap was able to provide early warning of medfly presence. The detection of medfly in the E-trap was preceded by the detection of female medfly

in the Tephri trap that was baited with Trimedlure. Following the only two captures recorded in this region in August, additional traps were set in September in the immediate vicinity of figs (*F. carica* L.) and mandarins (*C. reticulata* L.). In September, we recorded a total of 129 adult *C. capitata* in all traps. In the same month, the Tephri trap, which was baited with 3 C, captured the most adults (68 females and 12 males) with a total of 80 (Fig. 3). The higher numbers of captures from the Tephri 3 C trap continued from October to December: 50 (42 females and 8 males) adult *C. capitata* in October, 44 in November (34 females and 10 males), and 48 adults (36 females and 12 males) in December. Recent results of *C. capitata* detection were also observed at the Podstrana site in both conventional and E-traps. The higher catch rate ("effectiveness") of the Tephri 3 C trap compared to the other traps is especially highlighted in September and October.

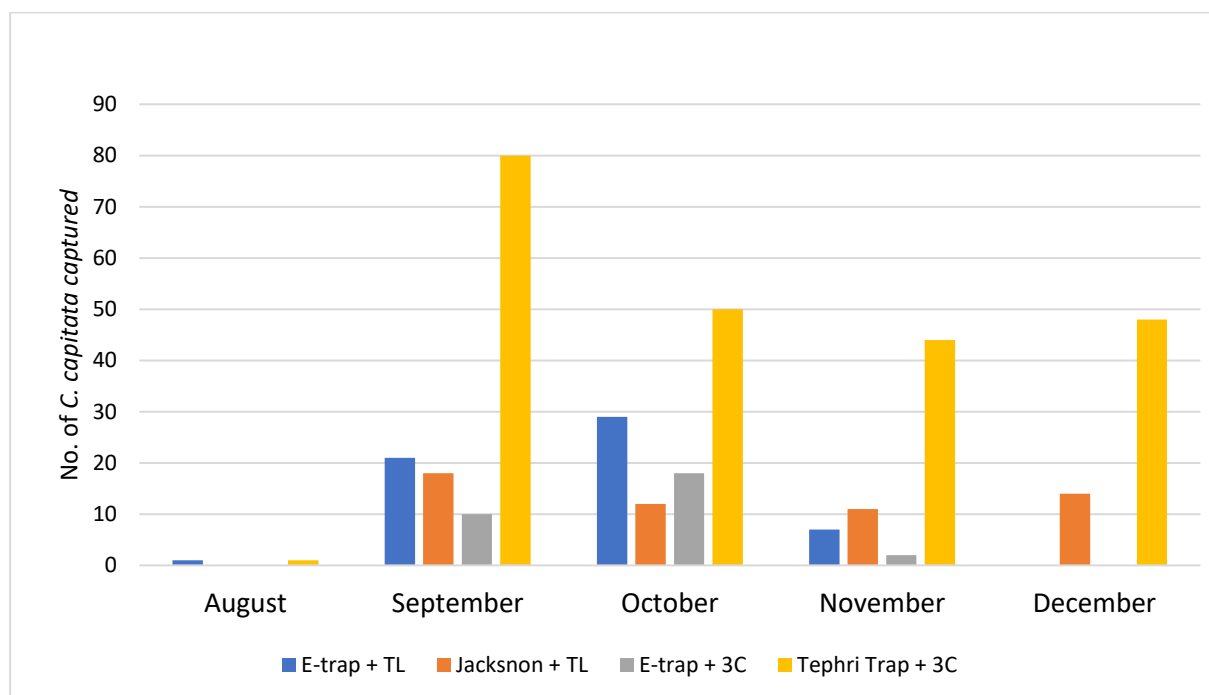


Figure 3. Total captures of *Ceratitits capitata* on different trapping system on the Podstrana site during 2021.

The occurrence of *C. capitata* was detected in 2020-2021 in two areas of Šibensko – kninska County and in seven areas of Splitsko – dalmatinska County (Table 1). In Šibensko – kninska County, *C. capitata* was found in the localities of Knin and Badanj. In Knin, only one adult female was caught on an apple tree (*M. domestica*) at the end of June (2020). In Badanj, we detected six adult female *C. capitata* on the same host (*M. domestica*) in early October (2020.). Surprisingly, this detection was not observed the second year of monitoring in Šibensko – kninska County. In this area, e-traps were set at the Maovice and Badanj sites, but they did not yield any detection even in 2021.

Medfly catches in Ramljane and Tugare were low in 2020. In Tugare locality, two catches were recorded in a tephri trap curled with 3 C, one in late June and the other in early August. They were caught on the peach (*P. persica*), in close proximity to the fig tree (*F. carica*). In Ramljane, only one male was caught on apples (*M. domestica*) in early October (2020) in a

tephri trap, attracted with 3 *C.* Notwithstanding the poorer condition of individual apple trees in Ramljane, these detections were not observed at the same sites in 2021.

The detection patterns found in 2020-21, particularly in the interior areas, are interesting and indicate the possible invasion pathways of *C. capitata* in this region. *C. capitata* catches were observed at elevations up to 400 m above sea level (e. g., Ramljane, Figure 1). Similarly, catches have been observed in an entire corridor of ravines extending from the sea coast to Knin (Figure 1). While it is not yet clear whether *C. capitata* has become established in this area or whether it is reintroduced from the lowlands yearly (probably through human activities and the fruit trade), the presence of *C. capitata* inland suggests a process of expansion of the pest. It is very possible that the main environmental factor driving this *C. capitata* expansion process is climate change, exacerbated by human trade and fruit transport. This process is expected to intensify in the future, leading to the spread and establishment of *C. capitata* and other pests in areas where they were not present in the past. For example, *C. capitata* has been detected in temperate areas of Europe, such as Austria, where the pest was not present in the past (Egartner et al., 2020). Thus, the current study and observed patterns suggest a mechanism of the spread of *C. capitata* and other pests in Europe that needs to be controlled and prevented. The study also provides some good evidence for the effectiveness of the McPhail e-trap as a tool to control the spread process of *C. capitata* by providing early warnings and remote sensing of the spread process.

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