

Chestnut tree damage evolution due to *Dryocosmus kuriphilus* attacks

Authors: Eric Gehring^{1,2}, Bruno Bellosi¹, Nicola Reynaud¹, Marco Conedera¹

¹ Insubric Ecosystem Research Group, WSL Swiss Federal Research Institute, A Ramél 18, 6593 Cadenazzo, Switzerland

² Laboratory of Soil Biodiversity, University of Neuchâtel, Emile-Argand 11, 2000 Neuchâtel, Switzerland

Corresponding author: Eric Gehring, eric.gehring@wsl.ch, ORCID: 0000-0002-4338-540. Tel. 091 821 52 35

Published in: *Journal of Pest Science* <https://doi.org/10.1007/s10340-019-01146-0>

Abstract

Dryocosmus kuriphilus has become a major threat to *Castanea sativa* and chestnut cultivation in Europe since its introduction to Italy in 2002. Thankfully, the biological control agent *Torymus sinensis* has proven to effectively control and reduce the *D. kuriphilus* population. The reaction of chestnut trees to decreasing *D. kuriphilus*-pressure, however, has been sparsely investigated. In this study, we analyze the recovery patterns of chestnut trees since the pest epidemic peak in 15 sites in southern Switzerland that were differently affected by the dephased arrival of *D. kuriphilus* and *T. sinensis*. By using various tree damage and insect population indicators, we show how damage varies as a function of the time elapsed between pest arrival and biological control by *T. sinensis*. Specifically, trees in sites experiencing seven years of *D. kuriphilus* uncontrolled attacks show large dying crown portions and stress-induced reactions (e.g., suckers along the stem) compared to sites where the two insects arrived nearly simultaneously. Moreover, in warm and south-exposed sites, the *D. kuriphilus* population remains significant and damage on chestnut trees still persists suggesting a desynchronisation in the phenology of the antagonist with respect to the pest.

Keywords: *Castanea sativa*, *Torymus sinensis*, Tree damage, Recovery process, Damage recrudescence, Classical biological control.

Key message

- Knowledge regarding chestnut tree reactions due to decreasing *Dryocosmus kuriphilus* pressure is still lacking.
- We investigated the evolution of various tree damage and insect population indicators during the entire epidemic process.
- Damage level varies as a function of the time elapsed between pest arrival and biological control occurrence.
- We show the importance of promptly introducing the biological control agent in order to reduce persisting damage.

Introduction

The Asian Chestnut Gall Wasp (*Dryocosmus kuriphilus* Yasumatsu, Hymenoptera, Cynipidae) is an invasive gall maker that rapidly became epidemic in Europe since its arrival in Italy in 2002 (Brussino et al. 2002). The high invasion potential is characterized by its fast parthenogenetic (thelytokous) reproductive success (Stone et al. 2002; Aebi et al. 2011; Avtzis et al. 2019), its rapid spread, and its asymptomatic conditions, due to its minute size and specific biological cycle, which allow it to remain hidden in chestnut buds during nearly two thirds of its life (Panzavolta et al. 2012; Bernardo et al. 2013). In addition, its dispersion potential and invasive ability is increased by its optimal phenological synchrony with the main European host tree *Castanea sativa* (Miller) (Bernardo et al. 2013), the absence of efficient local natural enemies in the newly invaded areas (Aebi et al. 2006; Cooper and Rieske 2007; Matošević and Melika 2013; Quacchia et al. 2013), the continuity of chestnut groves in many European countries and the lack of efficient mechanical (Maltoni et al. 2012a) or chemical (Cooper and Rieske 2007) control options.

In the case of repeated and uncontrolled attacks by *D. kuriphilus*, overall tree fitness is reduced (Kato and Hijii 1997; Ugolini et al. 2014; Sartor et al. 2015; Gehring et al. 2018a) to the point where chestnut groves experience important reductions in fruit yield (up to 80% - Battisti et al. 2014; Sartor et al. 2015). When the *D. kuriphilus* infestation occur in conjunction with other stresses such as drought and/or weakness pathogens (e.g., the necrophitic *Cryphonectria parasitica* Murrill (Meyer et al. 2015; Rigling and Prospero 2018) and/or *Gnomoniopsis castaneae* G. Tamietti (Lione et al. 2016, 2019)), concerned trees may even die (Tarcali and Radocz 2009; Zhi-Yong 2009).

Due to the importance of the chestnut culture and related fruit production in many European countries and in Italy in particular (Bounous 2006), and inspired by the successful Japanese experience (Moriya et al. 2003), the University of Turin studied and trailed the use of the biocontrol agent *Torymus sinensis* Kamijo (Hymenoptera, Torymidae) which was then first released in 2005 in the province of Cuneo (Quacchia et al. 2008). Following the promising results of the Italian experience, many other release programs followed in France (Borowiec et al. 2014), Croatia, Hungary and Slovenia (Matošević et al. 2015), Turkey (Doğanlar and Savaş 2018), Spain (Pérez-Otero et al. 2017), Portugal and Slovakia (Personal communication by Ambra Quacchia), and Austria (LFI 2017).

Even though the effectiveness of *T. sinensis* in controlling the *D. kuriphilus* population has subsequently been demonstrated in Japan (Moriya et al. 2003), the United States of America (Rieske 2007), Croatia, Slovenia and Hungary (Matošević et al. 2017), and Italy (Ferracini et al. 2018a) information concerning chestnut recovery with respect to decreasing pest-pressure remains sparse. In the present study, we investigate the recovery patterns of chestnut trees in southern Switzerland in 15 sites differently affected by *D. kuriphilus* epidemics in terms of years prior to biological control by *T. sinensis*. Our specific aims are to:

- 1) investigate chestnut tree reactions to decreasing *D. kuriphilus* pressure as a consequence of its parasitism by *T. sinensis*.
- 2) define the threshold of *D. kuriphilus* parasitism by *T. sinensis* that induces the visible recovery of chestnut trees
- 3) describe the recovery patterns in different crown sectors (inner and outer crown area) and as function of the time-lag between *D. kuriphilus* and *T. sinensis* arrival.

Materials and methods

Study area

This study was carried out in the chestnut groves of canton Ticino (southern Switzerland). The climate is classified as moist-warm and temperate (annual mean precipitation from 1,318 to 2,134 mm, annual mean temperature ranging from 10.2 to 12.4 °C – MeteoSwiss climate normals for the period 1981-2010 and considering the main chestnut grove areas; <http://www.meteoswiss.admin.ch>) and soils as haplic podzol on crystalline bedrock (Blaser et al. 2008). The chestnut forests of canton Ticino cover an area of ca. 20,000 ha representing 15% of the entire cantonal forest. *C. sativa* is mainly found at elevations ranging from 200 m a.s.l. (lowest point at Lago Maggiore) up to 900-1,100 m a.s.l. (Krebs et al. 2012). As a result, the species forms a chestnut forest belt continuum across the whole study area with a mix of cultivated, abandoned and naturalized stands (Brändli 1998; Ceschi 2014) (Fig. 1).

D. kuriphilus entered Switzerland from the south most likely in 2007 (Forster et al. 2009) (arrow in Fig. 1). Its population grew rapidly and moved northwest mainly by natural dispersion and wind assisted flight (Oho and Shimura 1970; Graziosi and Rieske 2014), colonizing the entire chestnut area by 2013. The arrival of its specific antagonist, *T. sinensis*, was officially announced in Ticino in 2013, although its presence was recorded by the authors in southern Ticino from 2011 onwards. Because of the prohibition of an active release of the antagonist due to unresolved biosafety issues, its arrival in Switzerland is assumed to be the result of a natural migration process beginning with the Italian releases near the Swiss border combined with some sporadic cases of illegal introductions (Conedera et al. 2015). Similarly to *D. kuriphilus* but much quicker, *T. sinensis* colonized the entire chestnut area by 2014, arriving almost simultaneously with its prey in the northernmost sites. This resulted in very different time-lags between *D. kuriphilus* and *T. sinensis* arrival between southern and northern sites (Fig. 1).

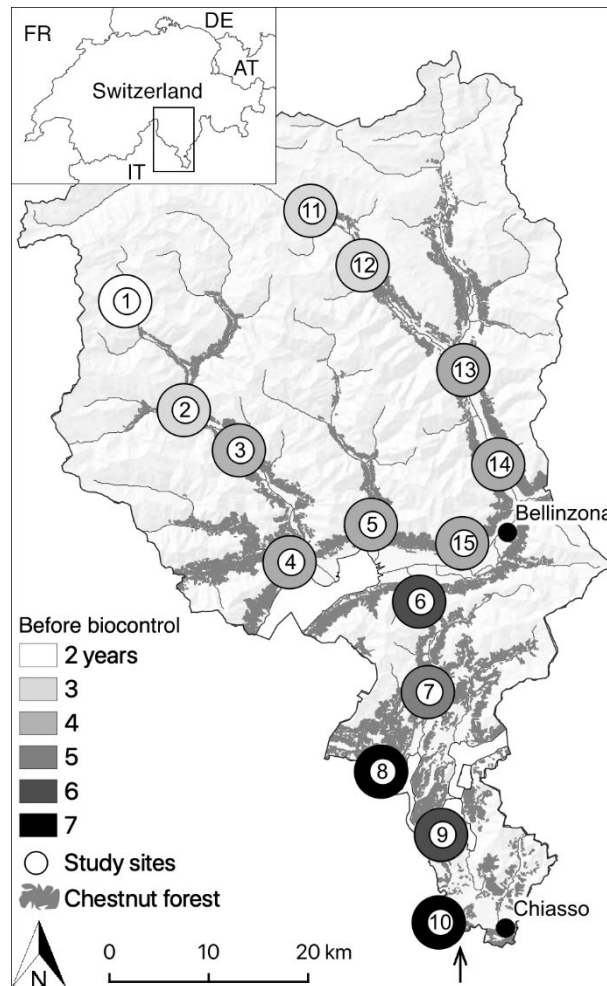


Figure 1 Location of the study sites. The top left subfigure shows the location of the study area (indicated by the small rectangle) within Switzerland. The numbered circles are colored according to the time-lag between *Dryocosmus kuriphilus* arrival and biological control by *Torymus sinensis*. The arrow in the lower part of the figure indicates the main *D. kuriphilus* and *T. sinensis* entry corridor

Study design and data collection

Study sites were selected in grids measuring 10 x 10 km so as to cover the entire area from north to south and to target heterogeneous situations in terms of *D. kuriphilus* and *T. sinensis* arrival years (Fig. 1, Table 1). General site characteristics such as geographic coordinates, altitude, slope, and aspect were recorded in the field (Table 1). From 2015 on, three temperature data loggers (Thermochron Logger DS1922L, iButton®) were placed in a shaded area in each site and the air temperature was measured at 2-hour intervals.

Within each site, ten mature trees were randomly selected in 2012 within an area of ca. 10 hectares and revisited during each field campaign (from 2013 to 2018). For every sampled tree, an overall visual crown assessment was first performed from four different perspectives for each tree and then averaged to a single value. Specifically, two different indices were recorded: the crown transparency of the living part (Crown transparency) using Müller and Stierlin's (1990) reference photography, and the proportion of the dead crown due to repeated *D. kuriphilus* attacks and *Cryphonectria parasitica* attacks (Dead crown).

Table 1 Sampling sites with detailed location and characteristics

Cluster		Site	Geographic coordinates (Swiss grid CH1903)		Altitude (m a.s.l.)	Slope (%)	Aspect (°)	Aspect transformed ^a	Temperature (°C) ^b	Arrival year		Epidemic peak	Biocontrol ^c
Group	ID		X	Y			360°	0-2	Mean [Min., Max]		Year		
1	3	Maggia	695588	125634	434	28	250	0.09	11.9 [-4.5, 31.3]	2011	2013	2013	2015
1	13	Biasca	718372	134801	382	42	200	0.09	11.0 [-5.2, 33.0]	2011	2013	2013	2015
1	15	Sementina	717928	115998	646	31	200	0.09	11.3 [-7.0, 33.7]	2011	2012	2013	2015
2	2	Cevio	689843	128927	475	41	155	0.66	10.7 [-5.0, 30.3]	2012	2013	2014	2015
2	4	Losone	700817	114163	318	4	40	2.00	10.9 [-5.3, 32.2]	2011	2012	2014	2015
2	5	Gordola	709094	117459	542	47	290	0.58	9.9 [-5.7, 31.1]	2011	2012	2013	2015
2	14	Claro	721970	124254	388	31	240	0.03	11.8 [-8.2, 34.3]	2011	2013	2013	2015
3	6	Rivera	713941	110347	552	15	50	2.00	11.5 [-6.8, 32.9]	2009	2012	2013	2015
3	7	Arosio	715290	100943	557	12	70	1.91	9.0 [-7.5, 28.6]	2009	2011	2013	2014
3	8	Pura	710578	92950	454	19	40	2.00	11.3 [-6.3, 30.2]	2008	2012	2013	2015
3	9	Melano	716143	85852	644	34	330	1.26	9.8 [-7.0, 28.6]	2008	2012	2013	2014
3	10	Stabio	715265	78329	422	17	110	1.42	11.6 [-6.7, 30.7]	2007	2011	2013	2014
4	1	Sonlerto	683916	139793	920	15	60	1.97	8.4 [-7.3, 28.7]	2013	2014	2014	2015
4	11	Faido	701571	149740	857	30	115	1.34	9.0 [-9.8, 28.9]	2012	2014	2014	2015
4	12	Lavorgo	708157	143244	748	16	135	1.00	9.0 [-7.3, 30.2]	2012	2013	2014	2015

^a Aspect transformed using Beers et al. (1966) equation $[\sin(\text{azimuth} + 45) + 1.0 = \text{southwest facing (xeric)}; 2 = \text{northeast facing (mesic)}]$.

^b Daily mean temperature from March 2015 to March 2018

^c Biocontrol threshold defined at 75% according to Quacchia et al. 2014

Table 2 Main indicators used for *Torymus sinensis*/*Dryocosmus kuriphilus* population and damage assessments

Indicators	Short name	Formula / Description	Values															
Population	<i>Dryocosmus kuriphilus</i> parasitism by <i>Torymus sinensis</i> (at gall level)	<i>D. kuriphilus</i> parasitism $D. kuriphilus \text{ parasitism} = \frac{T. sinensis \text{ alive}}{\text{chamber}} * 100$	From 0% (no <i>T. sinensis</i>) to 100% (chambers fully occupied). 75% is considered the biological control threshold level (Quacchia et al. 2014) when <i>D. kuriphilus</i> infestation significantly decreases.															
	Number of buds with <i>Dryocosmus kuriphilus</i> galls per available bud (at shoot level)	<i>D. kuriphilus</i> infestation $D. kuriphilus \text{ infestation} = \frac{\text{attacked buds}}{\text{available buds}} * 100$	<table border="1"> <tr> <td>0</td> <td>≤ 30</td> <td>> 30 ≤ 40</td> <td>> 40 ≤ 60</td> <td>> 60</td> </tr> <tr> <td>No</td> <td>Low</td> <td>Moderate</td> <td>High</td> <td>Very high</td> </tr> <tr> <td colspan="5">Damage level</td> </tr> </table>	0	≤ 30	> 30 ≤ 40	> 40 ≤ 60	> 60	No	Low	Moderate	High	Very high	Damage level				
	0	≤ 30	> 30 ≤ 40	> 40 ≤ 60	> 60													
No	Low	Moderate	High	Very high														
Damage level																		
Outer crown	Crown transparency assessment of the living part (at tree level)	Crown transparency Visual assessment of crown transparency based on the Sanasilva approach (Müller and Stierlin 1990).	30% is considered the damage threshold level (Gyoutoku and Uemura 1985, Murakami et al. 2009). <table border="1"> <tr> <td>0</td> <td>≤ 20</td> <td>> 20 ≤ 30</td> <td>> 30 ≤ 60</td> <td>> 60</td> </tr> <tr> <td>No</td> <td>Low</td> <td>Moderate</td> <td>High</td> <td>Very high</td> </tr> <tr> <td colspan="5">Damage level</td> </tr> </table>	0	≤ 20	> 20 ≤ 30	> 30 ≤ 60	> 60	No	Low	Moderate	High	Very high	Damage level				
0	≤ 20	> 20 ≤ 30	> 30 ≤ 60	> 60														
No	Low	Moderate	High	Very high														
Damage level																		
	Crown assessment of the dead part (at tree level)	Dead crown Visual assessment of dead branches from <i>D. kuriphilus</i> and <i>Cryphonectria parasitica</i> attack.	<table border="1"> <tr> <td>0</td> <td>≤ 20</td> <td>> 20 ≤ 30</td> <td>> 30 ≤ 60</td> <td>> 60</td> </tr> <tr> <td>No</td> <td>Low</td> <td>Moderate</td> <td>High</td> <td>Very high</td> </tr> <tr> <td colspan="5">Damage level</td> </tr> </table>	0	≤ 20	> 20 ≤ 30	> 30 ≤ 60	> 60	No	Low	Moderate	High	Very high	Damage level				
0	≤ 20	> 20 ≤ 30	> 30 ≤ 60	> 60														
No	Low	Moderate	High	Very high														
Damage level																		
Damage	Damage Composite Index (at branch level)	DCI $DCI = (\text{dead shoots} * 0.479 + \text{Reactivated dormant buds} * 0.525 + \text{galls on shoots} * 0.120) * 100$ Detailed assessment of branch architecture. For details, see Gehring et al. 2018a, b.	<table border="1"> <tr> <td>≤ 2.5</td> <td>> 2.5 ≤ 7.5</td> <td>> 7.5 ≤ 15</td> <td>> 15 ≤ 25</td> <td>> 25</td> </tr> <tr> <td>No</td> <td>Low</td> <td>Moderate</td> <td>High</td> <td>Very high</td> </tr> <tr> <td colspan="5">Damage level</td> </tr> </table>	≤ 2.5	> 2.5 ≤ 7.5	> 7.5 ≤ 15	> 15 ≤ 25	> 25	No	Low	Moderate	High	Very high	Damage level				
	≤ 2.5	> 2.5 ≤ 7.5	> 7.5 ≤ 15	> 15 ≤ 25	> 25													
No	Low	Moderate	High	Very high														
Damage level																		
Inner crown	Number of dormant buds (at shoot level)	Dormant buds Average number of buds remaining dormant during current vegetative season.																
Intense stress	Suckers per stem-linear- meter (at tree level)	Suckers Visual assessment of stem and branch suckers per linear meter in five categories.	<table border="1"> <tr> <td>0</td> <td>1-5</td> <td>6-10</td> <td>11-20</td> <td>> 20</td> </tr> <tr> <td>No</td> <td>Low</td> <td>Moderate</td> <td>High</td> <td>Very high</td> </tr> <tr> <td colspan="5">Stress level</td> </tr> </table>	0	1-5	6-10	11-20	> 20	No	Low	Moderate	High	Very high	Stress level				
0	1-5	6-10	11-20	> 20														
No	Low	Moderate	High	Very high														
Stress level																		

For the latter, very old dead branches and snags are excluded from the assessment. Recent dieback (ca. ten years) can be identified by the presence of small terminal twigs and/or lower order branches (Table 2). Moreover, on each tree, one to three branches, depending on the heterogeneity of the damage, were subsequently sampled every year rotating among different heights and orientations in order to preserve the tree crown. These branches were assessed for *D. kuriphilus* damage and tree reactions using the Damage composite index (DCI - for details see Gehring et al. 2018a, b) which includes parameters such as the number of dead shoots, reactivated dormant buds, developed buds, and the number and position of *D. kuriphilus* galls. In addition, the average number of dormant buds per shoot was recorded (Table 2).

In this manuscript we define the outer crown as the part issued from all the buds that should normally develop and grow (not dormant bud) during a vegetative season and that can be easily assessed with the crown transparency approach and the *D. kuriphilus* infestation indicator. The inner part refers on contrary to the branch-architectural parts of the crown involving dormant buds, reactivated dormant buds and shoots (dead or alive; please refer to Gehring et al. 2018a for the precise definitions). The inner crown are best assessed by the DCI and the dormant buds indicator (Table 2).

D. kuriphilus infestation was computed with the method proposed by Kotobuki et al. (1985) which consists of counting the proportion of attacked previous-year buds (number of attacked buds per available buds) at shoot level on the collected branches (Table 2).

In every site, at least 100 galls were randomly analyzed from the collected branches, additional trees, and crown parts. The galls were then dissected and the presence of *T. sinensis* in the cells recorded in order to calculate the level of *D. kuriphilus* parasitism by *T. sinensis* (hereafter called *D. kuriphilus* parasitism; Table 2). *T. sinensis* larvae were identified morphologically by their characteristic red-brownish ventral stripes, which are a quite reliable identification trait for the species at this development stage. In fact, potential confusion with native *Torymus* species exists for *T. cyaneus*, *T. notatus* and *T. affinis* only (Gómez et al. 2008), which have however to our knowledge never been reared from *D. kuriphilus* galls so far (Aebi et al. 2006, 2007; Bigler et al. 2006; Matošević and Melika 2013; Quacchia et al. 2013; Palmeri et al. 2014; Francati et al. 2015; Kos et al. 2015; Bernardinelli et al. 2016; Colombari and Battisti 2016a; Panzavolta et al. 2018; Ferracini et al. 2018b; Bonsignore et al. 2019), what drastically decreases the chance of misidentification.

During the final field campaign in 2018, additional 50 trees were randomly selected in every site and both the proportion of dead crown and the number of suckers per stem-linear-meter were estimated in order to have a current picture of the still detectable general stress level resulting from the epidemic (Table 2).

Data processing and analysis

The various tree damage and insect population indicators were initially calculated at each sample collection level (gall, shoot, branch) as described in Table 2 and then averaged at tree and site levels. Site aspect was

transformed in order to be analyzed using the equation proposed by Beers et al. (1966) which assigns the value of 0 to southwest facing sites (assumed to be more xeric), 2 to the northeast facing sites (more mesic), and an intermediate values for site aspects in between.

A hierarchical cluster analysis (using Euclidean distance with the Ward.d2 clustering method; see Murtagh and Legendre 2014) was performed to group sites based on their geo-physiographic characteristics (see columns “arrival year”, “altitude”, “slope”, “aspect transformed”, and “temperature” in Table 1). The resulting groups were then selected and used as references to analyze possible trends and patterns in the evolution of the *D. kuriphilus* infestation since 2012, the *D. kuriphilus* parasitism, as well as the tree damage indicators.

Daily mean temperatures were first calculated at site level and then summed over three different periods: T1 (yearly site temperature) = yearly (from March 1st to February 28th) average of the daily mean temperature; T2 (*T. sinensis* development period) = average of daily mean temperatures from October 1st (average *T. sinensis* pupation time in our study sites, data not shown) to March 15th (average *T. sinensis* emergence time in our study sites, data not shown); T3 (degree days) = average of the daily mean temperature from January 1st (that is the conventional starting time for calculating degree days for most insect species; Herms 2004) to March 15th.

Descriptive statistics combined with univariate comparative analysis (using Mann-Whitney *U* tests with significant differences at $p < 0.05$) were used in order to quantitatively describe and summarize the evolution of the indicators over the years. Linear regression was used to model the relationship between the 2018 tree damage and insect population indicators and site characteristics (Table 2). Specifically, *D. kuriphilus* infestation, DCI, and crown transparency were separately used as response variables in three different models with all site characteristics as explanatory variables.

All statistical analyses were performed using the R statistical package version 3.5.1 (R Core Team 2018). Figure 1 was generated using the open source geographic information system QGIS (QGIS Development Team 2018). Figure 2 was made using the ggplot2 data visualization package for R (Wickham 2016).

Results

Chestnut trees recovery

The cluster analysis based on site characteristics revealed four major groups (Table 1). Group 1 includes the most sun-exposed sites with high average temperatures at altitudes ranging between 382 and 646 m. a.s.l. At the other extreme, group 4 includes cooler sites at the highest altitudes (857-920 m. a.s.l.) where the time-lag between *D. kuriphilus* arrival and the biocontrol by *T. sinensis* did not exceed 3 years. In between, group 2 includes sites that are similar to those in group 1, but are slightly less sun-exposed and cooler,

whereas group 3 includes sites at average altitudes, not particularly sun-exposed and where the time-lag between *D. kuriphilus* arrival and the biocontrol by *T. sinensis* exceeded 4 years (Fig. 1 and Table 1).

D. kuriphilus parasitism by *T. sinensis* reached the biocontrol threshold of 75% in all sites in 2015. In groups 3 and 4, it remained constant until 2018, whereas it decreased to ca. 60% in groups 1 and 2 (Fig. 2). Simultaneously, *D. kuriphilus* infestation drastically decreased after 2014 in all groups and stabilized below the damage threshold level of 30% in groups 2 (~15/20%), 3 and 4 (~5%), whereas it significantly increased again from 2016 onwards in group 1 where it reached an average value of 32% in 2018. Damage indices follow a similar trend with group 1 experiencing the most severe damage followed by groups 2, 3, and 4, respectively. Specifically, crown transparency decreased from 2015 in all sites and significantly increased again since 2017 in groups 3 and 4. The DCI showed a somewhat similar trend but with much more fluctuation and a significant increase from 2016 on in groups 1 and 2 (Fig. 2). Finally, the later arrival of *D. kuriphilus* in group 4 (Table 1) is clearly visible in Figure 2 as expressed by the delayed damage index increase with respect to the other groups.

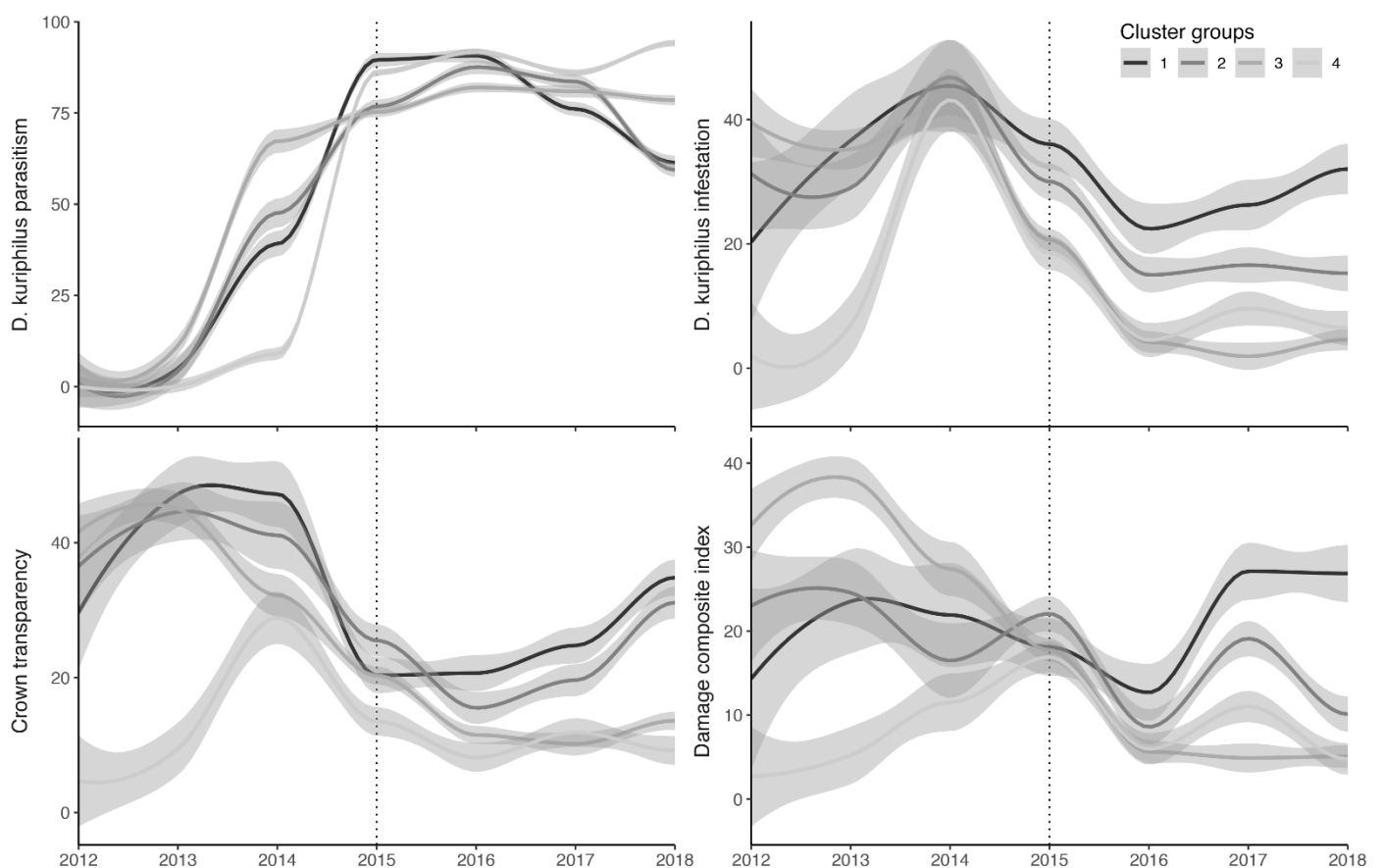


Figure 2 Evolution of selected epidemic and damage indices from 2012 to 2018 by cluster group (see Table 1). The vertical dashed line indicates the average date (between 2014 and 2015) when biocontrol by *Torymus sinensis* was reached in the study area. Grey bands show 95% confidence intervals

Using 2018 data only, the best linear models with DCI and *D. kuriphilus* infestation as the response variables, respectively, included the transformed site aspect (negatively related) and the sum of temperatures from October 2016 to March 2017 (T2; positively related) as the explanatory variables. The two models had adjusted R-squared values of 0.73 and 0.69 (Table 3). The model with crown transparency as the response variable also retained site aspect and a temperature as the best explanatory variables but in this case the yearly sum of temperature (from March 2016 to March 2017) had greater explanatory power. None of the models were affected by multicollinearity problems ($VIF < 2$) nor did diagnostic plots reveal any violation of model assumptions.

Table 3. Best linear models considering various damage index as response variables and only 2018 data

Model variables		Estimate	Confidence Interval		t value	Var. imp. ^a %	Adj. R ²	R ²
Response ^b	Explanatory		2.50%	97.50%				
Crown transparency	Intercept	-22.229	-89.187	44.73	-0.723			
	Aspect t. ^d	-7.348	-14.923	0.227	-2.114	48.1	0.62	0.68
	T1 ^c	0.016	0.001	0.031	2.281	51.9		
<i>D. kuriphilus</i> infestation	Intercept	5.303	-22.411	33.016	0.417			
	Aspect t.	-10.392	-16.12	-4.663	-3.953	68.9	0.69	0.74
	T2 ^c	0.025	-0.006	0.055	1.78	31.1		
DCI ^b	Intercept	-7.496	-27.468	12.476	-0.818			
	Aspect .t	-6.053	-10.181	-1.924	-3.195	50.2	0.73	0.77
	T2 ^c	0.032	0.01	0.054	3.166	49.8		

^aVar.imp. refers to variable importance as calculated by the function varImp() contained in the R-package caret (Kuhn 2008). ^bSee Table 2 for details for the definitions of the response variables. ^cT = sum of daily mean temperature exceeding 0°C; T1 = from 15.03.16 to 14.13.17; T2 = from 15.10.16 to 15.03.17. ^dAspect t. = Aspect transformed according to the equation proposed by Beers et al. 1966.

D. kuriphilus parasitism thresholds inducing tree recovery

The first signs of significant tree recovery with respect to all indicators (DCI, number of dormant buds, crown transparency, and *D. kuriphilus* infestation) are registered when *D. kuriphilus* parasitism reaches mean values of 79% (95%CI [74, 84]). Recovery from *D. kuriphilus* infestation is already visible at lower parasitism levels (68% [48, 88]), whereas the number of dormant buds reacts at higher parasitism levels only (86% [83, 89]) (Table 4). The situation is similar, although at much lower *D. kuriphilus* parasitism values, when considering the previous year (Table 4).

Role of the time-lag between pest and antagonist arrivals

The time-lag between *T. sinensis* arrival and the first sign of tree recovery is longer in sites in groups 1, 2, and 3, compared to that in group 4 (Table 5). For example, the first signs of recovery after the arrival of *T. sinensis* regarding the crown transparency index in group 1 are visible 1.3 years later (on average 3.3 years; 95% CI [2.6, 4.0]) than in group 4 (on average 2.0 years; 95% CI [0.9, 3.1]). This trend is similar when considering the other indices. It is also worth noting that the recovery process began in the outer crown area nearly one year sooner than the inner part (Table 5).

Table 4 *Dryocosmus kuriphilus* parasitism thresholds for first signs of recovery.

Crown area	Index	<i>D. kuriphilus</i> parasitism rate (%) ^a	
		Recovery year	Previous year
Outer	Dk infestation	68 [48, 88]	37 [18, 55]
	Crown transparency	75 [59, 91]	40 [20, 60]
Inner	DCI ^b	81 [73, 88]	55 [34, 77]
	Dormant buds	86 [83, 89]	57 [38, 76]
	All ^b	79 [74, 84]	47 [39, 54]

^a Values represent means with 95% lower and upper confidence intervals.

^b All = average value of all indices

Table 5 Time-lag in years (with 95% confidence intervals) between *Torymus sinensis* arrival and the first signs of tree recovery using various indices and for different cluster groups

Crown area	Index	Cluster groups				All
		1	2	3	4	
Outer	Dk infestation	3.0 [1.9, 4.1]	2.8 [1.3, 4.3]	3.0 [2.1, 3.9]	1.7 [1.0, 2.4]	2.7 [2.1, 3.3]
	Crown transparency	3.3 [2.6, 4.0]	3.2 [2.3, 4.1]	3.8 [3.1, 4.5]	2.0 [0.9, 3.1]	3.2 [2.7, 3.7]
Inner	DCI	4.3 [2.6, 6.0]	3.8 [3.3, 4.3]	3.8 [3.4, 4.2]	2.3 [1.6, 3]	3.6 [3.1, 4.1]
	Dormant buds	4.3 [2.6, 4.9]	4.0 [3.2, 4.8]	4.4 [3.9, 4.9]	2.7 [1.4, 4]	3.7 [3.2, 4.2]

The average damage severity for sites in group 3 experiencing a longer time-lag between the arrival of *D. kuriphilus* and the occurrence of *T. sinensis* biocontrol (~5 - 7 years) is significantly greater in terms of crown transparency (Fig. 3 top) and malformation of the branch architecture (Fig. 3 bottom) compared to those in group 4 where the two insects arrived nearly simultaneously (*T. sinensis* biocontrol within 2-3 years of *D. kuriphilus* arrival). Furthermore, considering 2018 data only (i.e., the present situation where, for every site, at least four years have passed since biocontrol occurrence), 30% of the crown area is still dead in sites that experienced four or more years of uncontrolled *D. kuriphilus* attack. The proportion of dead crown progressively and significantly decreases to 5% in sites with a shorter time-lag (group 4). This is especially pronounced in the site "Sonlerto" (Fig. 4 bottom). Consequently, the stress level endured by trees as

indicated by the number of epicormic resprouts (“Suckers” in Fig. 4) follows a similar trend with a significant increase as a function of the time-lag.

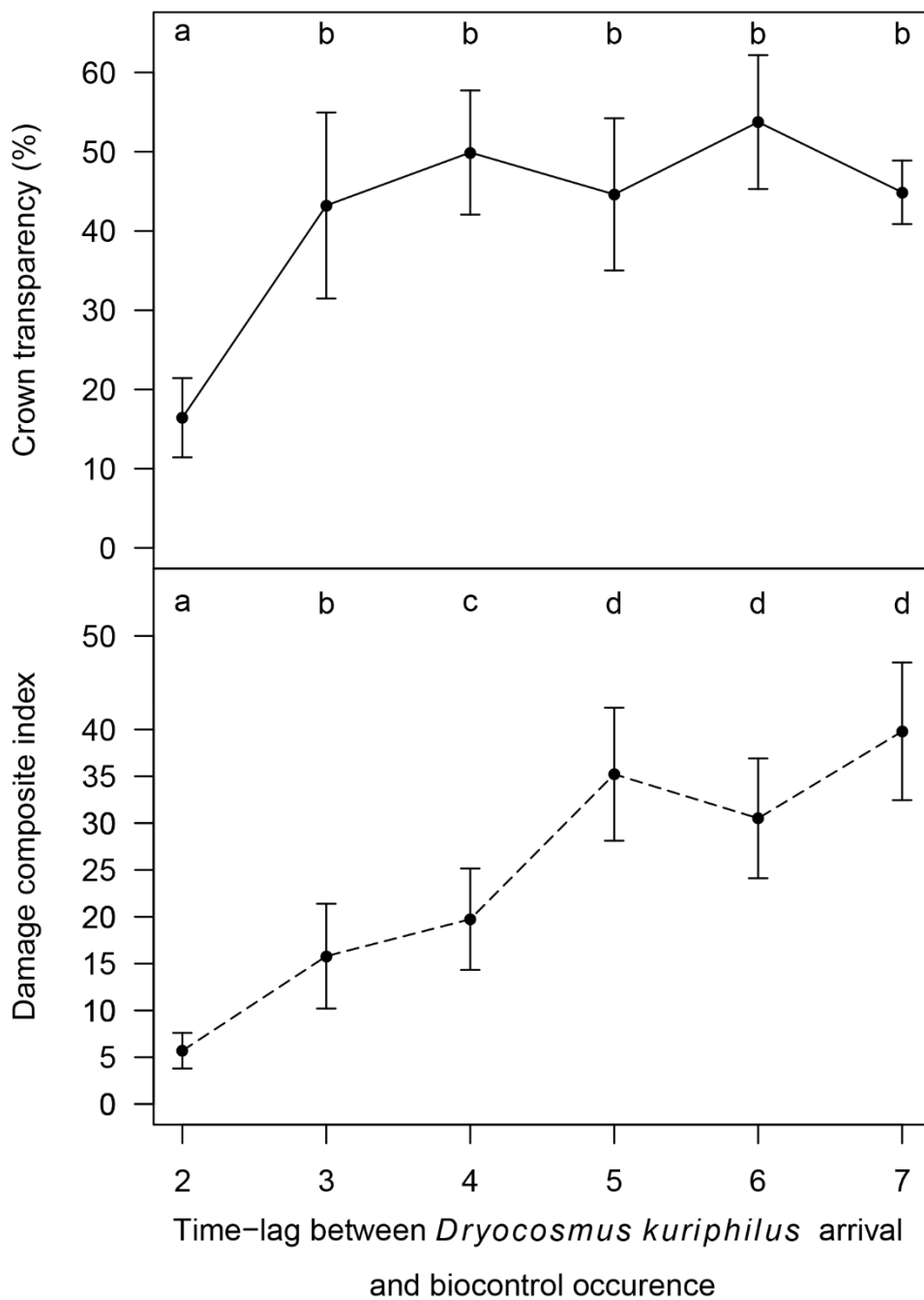


Fig 3 Damage severity during the epidemic peak by time-lag between the *Dryocosmus kuriphilus* arrival and the first biological control year by *Torymus sinensis*. Only data from the epidemic peak to the year before biocontrol occurrence in the sites in question was used to generate this figure (see Table 1). Error bars show 95% confidence intervals. Letters indicate significant differences with $p < 0.05$ according to the non-parametric Mann-Whitney U test with Holm adjustment

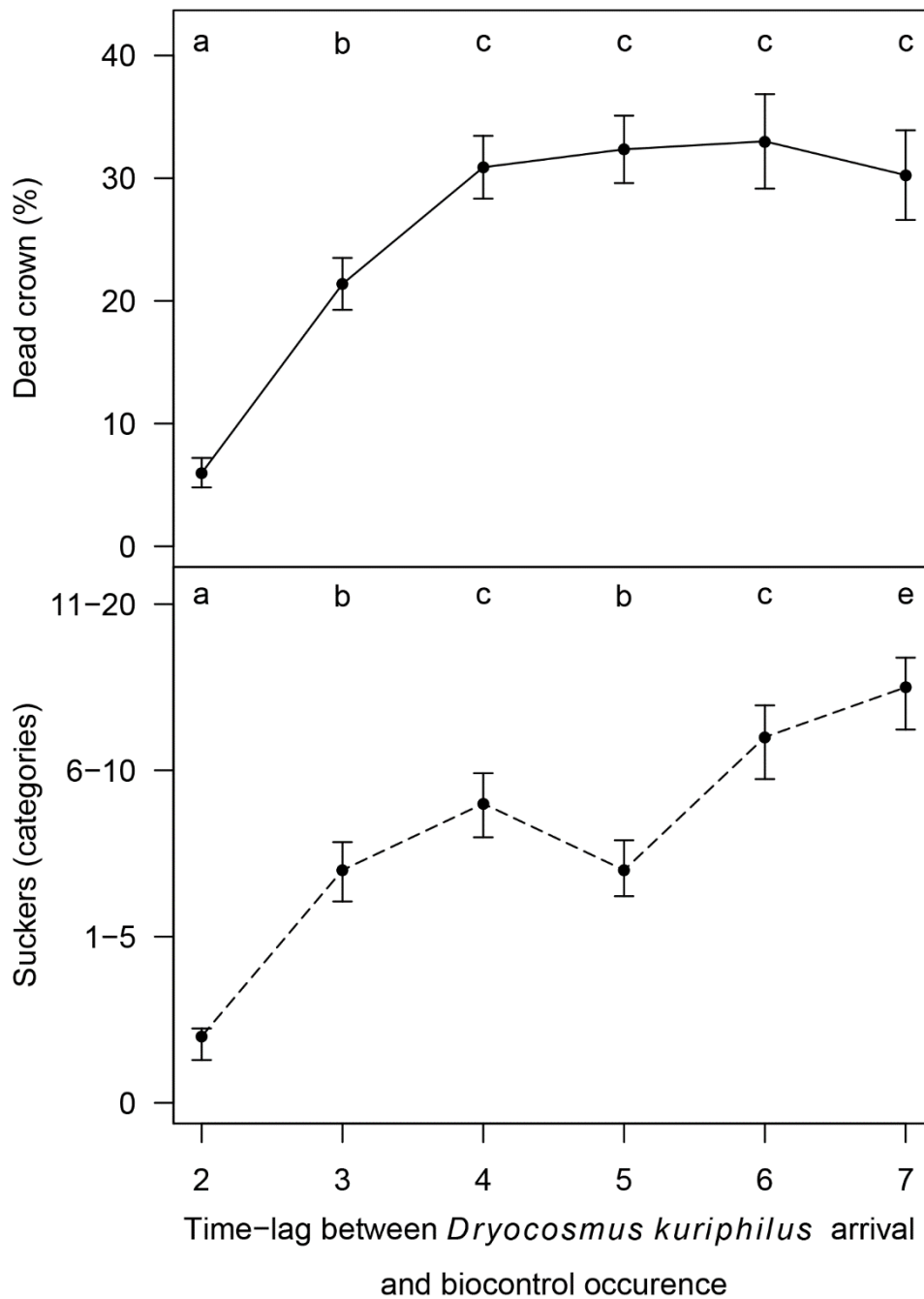


Figure 4 Damage severity and tree reaction in 2018 according to the time-lag between *Dryocosmus kuriphilus* arrival and the first year of biological control by *Torymus sinensis*. Error bars show 95% confidence interval. Letters indicate significant differences with $p < 0.05$ according to the non-parametric Mann-Whitney U test with Holm adjustment

Discussion

In this study, we assess the evolution of the *D. kuriphilus* epidemic from its arrival to biological control by *T. sinensis*, as well as the resulting damage evolution and recovery patterns, in 15 sites located in southern Switzerland. Generally, damage peaked between 2013 and 2014 and started decreasing as soon as *T. sinensis* reached notable levels of parasitism on *D. kuriphilus*, proving its effectiveness as a biocontrol agent as already shown by other studies (Moriya et al. 2003; Borowiec et al. 2018; Ferracini et al. 2018a; Avtzis et al. 2019). As a general rule, the first signs of chestnut tree recovery are visible as soon as *D. kuriphilus* parasitism reaches values of 80% on average, a parasitism rate that could be considered as the recovery and biocontrol threshold for the chestnut tree. This is in line with other studies that have confirmed a decrease in infestation in terms of gall numbers when the *D. kuriphilus* parasitism reaches the 75% level (Quacchia et al. 2014). By considering previous year parasitism levels, one could also predict recovery to occur one year after *D. kuriphilus* parasitism reaches values of about 50%.

This threshold does not depend on the time-lag between the first *D. kuriphilus* attack and the arrival of *T. sinensis*. In fact, external crown parts react immediately to the achieved biocontrol. This is likely due to the permanent healthy condition of the apical buds which are usually formed after the *D. kuriphilus* oviposition period (Maltoni et al. 2012b). When biocontrol is achieved, the lateral buds of the terminal shoot will also be attacked less such that the external part of the crown will soon consist of normal-shaped branches. This initial recovery process, when the gall-free external crown coat reinvigorates the leaf area, is well reflected in the crown transparency index and the *D. kuriphilus* infestation. The inner crown area, in contrast, experiences a delayed recovery of, on average, one year and varies as a function of the time elapsed between *D. kuriphilus* arrival and the occurrence of *T. sinensis* biocontrol. This is reflected by specific indicators such as DCI and the number of dormant buds. In fact, in sites of severe and repeated *D. kuriphilus* attacks, branch corruption includes the consumption and depletion of tree reserves (dormant buds, Gehring et al. 2018a), the failure of bud production from galled shoots (Kato and Hijii 1997), and the dieback of infested branches (Maltoni et al. 2012b; Gehring et al. 2018a), which all require more time and energy to be restored and renewed.

The speed and completeness of inner crown recovery and eventually of the whole tree depend strongly on the time-lag between the first *D. kuriphilus* attack and the arrival of *T. sinensis*. In sites where the two insects arrived nearly simultaneously (cluster group 4. *D. kuriphilus* and occurrence of *T. sinensis* biocontrol ≤ 3 years), severe damage never occurred and the recovery process was quasi-imminent keeping both the branch and the crown architecture of the affected trees intact. In contrast, sites that experienced more than four years of uncontrolled *D. kuriphilus* attacks suffered overall from more severe crown corruption and tree reserve exhaustion to the point that, even after four years of *T. sinensis* biocontrol (present situation; cluster

group 3. *D. kuriphilus* and occurrence of *T. sinensis* biocontrol ≥ 6 years), long-lasting damage is still visible in the form of dead crown parts. This crown deterioration could also be the result of an indirect interaction between *D. kuriphilus* and the chestnut blight (*Cryphonectria parasitica*) that uses the flickering holes of abandoned galls as entry points (Meyer et al. 2015). Moreover, in the locality that suffered from long-lasting uncontrolled attacks, the majority of trees are rebuilding the damaged crown by producing an elevated number of suckers along the trunk and the main branches in reaction to past severe stress. It is therefore reasonable to argue that a proportion of these trees may never fully recover and will be seriously threatened in the immediate future by secondary agents such as hail-induced chestnut blight and drought. Last but not least, possible recrudescence of the *D. kuriphilus* infestation may periodically increase again the pressure on the chestnut trees as it has been already reported for Japan in 2000 and 2007 (Toyama et al. 2009) and as generally predicted by the model developed by Paparella et al. (2016).

In this study we were able to detect different epidemic trajectories based on the geo-physiographic site characteristics and the time-lag between the arrival of the pest and its antagonist. Of particular interest from an epidemiological point of view is the evolution of damage and the *D. kuriphilus* parasitism in cluster 1, where the *Dryocosmus* infestation never reached low levels, and damage increased again in 2017 and 2018 to ca. 35/40% as a consequence of a decrease in *D. kuriphilus* parasitism. One possible explanation for such a fluctuation is the south facing aspect and the related generally high temperatures at these sites. As shown by our model analysis, the damage level in 2018 is positively correlated with the sum of temperature from October 2016 to March 2017 and negatively correlated with aspect. Based on the fact that the time period in question roughly corresponds to development stages of *T. sinensis* from pupation to time of emergence (personal observation of our study sites), we hypothesize a possible earlier emergence of the majority of the antagonist population in such warm sites that causes a desynchronization with gall appearance. This, necessarily follows the leaf unfold phenology of the chestnut tree. In addition, high temperatures combined with arid conditions (southern exposure sites) may also influence the longevity of *T. sinensis* adults as demonstrated by laboratory trials that highlighted a significant decrease in lifespan comparing temperatures of 15 and 25 °C (Quacchia et al 2008) and in the case of food deprivation (Picciau et al. 2019). This may cause a partial failure to control the *D. kuriphilus* population and lead to consequent increased damage the year after.

In optimal condition (forest stand continuum and wind) *T. sinensis* may recolonize the unfavourable sites by wind-aided flight starting from the suitable areas (Colombari and Battisti 2016b). However, in our study area, favourable (e.g., north-facing) and unsuited (south-facing) sites are often at least three kilometres away and not always connected by chestnut forests. Its immediate recolonization success will thus highly depend on the dominant direction of winds and the *T. sinensis* phenology. Late emerging parasitoid may in fact arrive at the unfavourable site when galls are too big and lignified to be parasitized.

Finally, all these possible factors (synchronization, longevity problems and recolonization) are weather-dependent and thus subjected to strong year-to-year variations. We should thus expect important yearly fluctuations of *D. kuriphilus* parasitism at medium or high damage levels without a decrease to undetectable levels of the *D. kuriphilus* population, especially in less favourable sites. If this will reveal to be true, forecasting the future will become very difficult in such a complex geomorphological area with various exposures.

In conclusion *T. sinensis* has shown to be a very effective biological control agent of *D. kuriphilus* on many species of the genus *Castanea* worldwide. The control of the *D. kuriphilus* population by *T. sinensis* displays however also very dynamic and unexpected aspects, especially where or when environmental conditions cause a possible desynchronisation of the phenology of the two insects and a failure of its parasitic action. The history of the *D. kuriphilus* epidemic in southern Switzerland reported here teaches us that the best way to avoid long-lasting gallwasp-induced damage on chestnut trees, resulting in heavily weakened trees with disrupted crowns, is to release the specific antagonist *T. sinensis* as soon as the pest is detected in the area. We are thus convinced that the downside related to the introduction of *T. sinensis* such as the possible hybridization with *T. beneficus* (Yara et al. 2010), the loss of native parasitoid richness (Ferracini et al. 2018b) and the rare host-range expansion (Ferracini et al. 2017) are and will be by far more acceptable than possible consequences on chestnut forests without its introduction or arrival.

Author contributions

MC and EG conceptualized the study and developed the research design. BB with EG designed the field protocols. BB, EG, and NR conducted the fieldwork. BB and NR inserted data in the database. EG analyzed the data, wrote the manuscript draft and submitted the paper. MC revised the manuscript. MC procured the research funding and coordinated the project. All authors read and approved the manuscript.

Funding: This study was partially funded by the Forest Service of Canton Ticino and the Federal Office for the Environment FOEN.

Compliance with ethical standards

Conflict of interest: the authors declare that they have no conflict of interest.

References

- Aebi A, Schoenenberger N, Bigler F (2011) Evaluating the use of *Torymus sinensis* against the chestnut gall wasp *Dryocosmus kuriphilus* in the Canton Ticino, Switzerland. *Agroscope Reckenholz-Tänikon Rep 72*
- Aebi A, Schönrogge K, Melika G, et al (2007) Native and introduced parasitoids attacking the invasive chestnut gall wasp *Dryocosmus kuriphilus*. *EPPO Bull* 37:166–171. doi: 10.1111/j.1365-2338.2007.01099.x
- Aebi A, Schönrogge K, Melika G, et al (2006) Parasitoid recruitment to the globally invasive chestnut gall wasp *Dryocosmus kuriphilus*. *Gall. Arthropods Their Assoc. Ecol. Evol.* 103–121
- Avtzis DN, Melika G, Matošević D, Coyle DR (2019) The Asian chestnut gall wasp *Dryocosmus kuriphilus*: a global invader and a successful case of classical biological control. *J Pest Sci* (2004) 92:107–115. doi: 10.1007/s10340-018-1046-1
- Battisti A, Benvegna I, Colombari F, Haack RA (2014) Invasion by the chestnut gall wasp in Italy causes significant yield loss in *Castanea sativa* nut production. *Agric For Entomol* 16:75–79. doi: 10.1111/afe.12036
- Beers TW, Dress PE, Wensel LC (1966) Notes and observations: aspect transformation in site productivity research. *J For* 64:691–692
- Bernardinelli I, Bessega D, Zanolli P, et al (2016) Survey of indigenous parasitoids affecting the invasive chestnut gall wasp *Dryocosmus kuriphilus* in the Friuli Venezia Giulia region (North-East Italy). *EPPO Bull* 46:286–289. doi: 10.1111/epp.12296
- Bernardo U, Iodice L, Sasso R, et al (2013) Biology and monitoring of *Dryocosmus kuriphilus* on *Castanea sativa* in Southern Italy. *Agric For Entomol* 15:65–76. doi: 10.1111/j.1461-9563.2012.00588.x

- Bigler F, Babendreier D, Kuhlmann U (2006) Environmental impact of invertebrates for biological control of arthropods : methods and risk assessment. CABI Pub
- Blaser P, Kernebeek P, Tebbens L, et al (2008) Cryptopodzolic Soils in Switzerland. *Eur J Soil Sci* 48:411–423. doi: 10.1111/j.1365-2389.1997.tb00207.x
- Bonsignore CP, Vono G, Bernardo U (2019) Environmental thermal levels affect the phenological relationships between the chestnut gall wasp and its parasitoids. *Physiol Entomol*. doi: 10.1111/phen.12280
- Borowiec N, Thaon M, Brancaccio L, et al (2014) Classical biological control against the chestnut gall wasp *Dryocosmus kuriphilus* (Hymenoptera, Cynipidae) in France. *Plant Prot Q* 29:9–12
- Borowiec N, Thaon M, Brancaccio L, et al (2018) Early population dynamics in classical biological control: establishment of the exotic parasitoid *Torymus sinensis* and control of its target pest, the chestnut gall wasp *Dryocosmus kuriphilus*, in France. *Entomol Exp Appl* 166:367–379. doi: 10.1111/eea.12660
- Bounous G (2006) Revival of chestnut culture in Mediterranean countries: factors to improve the quality of productions. *Adv Hortic Sci* 20:7–15
- Brändli U-B (1998) Die häufigsten Waldbäume der Schweiz. Ergebnisse aus dem Landesforstinventar 1983-85: Verbreitung, Standort und Häufigkeit von 30 Baumarten. *Ber Eidgenöss Forsch.anst Wald Schnee Landsch* 342:1–279
- Brussino G, Bosio G, Baudino M, et al (2002) Pericoloso insetto esotico per il castagno europeo. *Inf Agrar* 58:59–61
- Ceschi I (2014) Il bosco del Canton Ticino, 2a ed. agg. Dipartimento del territorio, Divisione dell’ambiente, Sezione forestale, Locarno
- Colombari F, Battisti A (2016a) Native and introduced parasitoids in the biocontrol of *Dryocosmus kuriphilus* in Veneto (Italy). *EPPO Bull* 46:275–285. doi: 10.1111/epp.12297
- Colombari F, Battisti A (2016b) Spread of the introduced biocontrol agent *Torymus sinensis* in north-eastern Italy: dispersal through active flight or assisted by wind? *BioControl* 61:127–139. doi: 10.1007/s10526-015-9712-1
- Conedera M, Gehring E, Quacchia A (2015) Danni da cinipide: il peggio è passato? L’ape. *Riv. Svizz. di Apic.* 18–19
- Cooper WR, Rieske LK (2007) Community Associates of an Exotic Gallmaker, *Dryocosmus kuriphilus* (Hymenoptera: Cynipidae), in Eastern North America. *Ann Entomol Soc Am* 100:9. doi: 10.1603/0013-8746(2007)100[236:CAOAE]2.0.CO;2
- Doğanlar M, Savaş SK (2018) Studies on release methods of *Torymus sinensis* (Hymenoptera : Torymidae), parasitoid of *Dryocosmus kuriphilus* Yasumatsu), into Turkey from Italy. *Acta Hortic* 1220:109–112. doi:

10.17660/actahortic.2018.1220.16

Ferracini C, Bertolino S, Bernardo U, et al (2018a) Do *Torymus sinensis* (Hymenoptera: Torymidae) and agroforestry system affect native parasitoids associated with the Asian chestnut gall wasp? Biol Control 121:36–43. doi: 10.1016/j.biocontrol.2018.01.009

Ferracini C, Ferrari E, Pontini M, et al (2018b) Effectiveness of *Torymus sinensis*: a successful long-term control of the Asian chestnut gall wasp in Italy. J Pest Sci (2004) 1–7. doi: 10.1007/s10340-018-0989-6

Ferracini C, Ferrari E, Pontini M, et al (2017) Post-release evaluation of non-target effects of *Torymus sinensis*, the biological control agent of *Dryocosmus kuriphilus* in Italy. BioControl. doi: 10.1007/s10526-017-9803-2

Forster B, Castellazzi T, Colombi L, et al (2009) Die Edelkastaniengallwespe *Dryocosmus kuriphilus* (Yasumatsu) (Hymenoptera, Cynipidae) tritt erstmals in der Südschweiz auf. Mitteilungen der Schweizerischen Entomol Gesellschaft 82:271–279

Francati S, Alma A, Ferracini C, et al (2015) Indigenous parasitoids associated with *Dryocosmus kuriphilus* in a chestnut production area of Emilia Romagna (Italy). Bull Insectology 68:127–134

Gehring E, Bellosi B, Quacchia A, Conedera M (2018a) Assessing the impact of *Dryocosmus kuriphilus* on the chestnut tree: branch architecture matters. J Pest Sci (2004) 91:189–202. doi: 10.1007/s10340-017-0857-9

Gehring E, Bellosi B, Quacchia A, Conedera M (2018b) Evaluating *Dryocosmus Kuriphilus*-induced Damage on *Castanea Sativa*. J Vis Exp 1–9. doi: 10.3791/57564

Gómez JF, Nieves-Aldrey JL, Hernández Nieves M (2008) Comparative morphology, biology and phylogeny of terminal-instar larvae of the European species of Toryminae (Hym., Chalcidoidea, Torymidae) parasitoids of gall wasps (Hym. Cynipidae)

Graziosi I, Rieske LK (2014) Local spread of an exotic invader: using remote sensing and spatial analysis to document proliferation of the invasive asian chestnut gall wasp. Acta Hort 1019:113–118. doi: 10.17660/ActaHortic.2014.1019.17

Herms DA (2004) Using Degree-Days and plant phenology to predict pest activity. In: Krischick V, Davidson J (eds) IPM (Integrated Pest Management) of Midwest landscapes: Tactics and tools for IPM. University of Minnesota, Minnesota Agriculture Experiment Station, St. Paul, pp 49–59

Kato K, Hijii N (1997) Effects of gall formation by *Dryocosmus kuriphilus* Yasumatsu (Hym., Cynipidae) on the growth of chestnut trees. J Appl Entomol 121:9–15. doi: 10.1111/j.1439-0418.1997.tb01363.x

Kos K, Kriston E, Melika G (2015) Invasive chestnut gall wasp *Dryocosmus kuriphilus* (Hymenoptera: Cynipidae), its native parasitoid community and association with oak gall wasps in Slovenia. Eur J Entomol 112:698–704. doi: 10.14411/eje.2015.091

- Kotobuki K, Mori K, Sato Y (1985) 2 methods to estimate the tree damage by chestnut gall wasp *Dryocosmus kuriphilus*. Bull fruit tree Res Stn A 2:29–36
- Krebs P, Koutsias N, Conedera M (2012) Modelling the eco-cultural niche of giant chestnut trees: new insights into land use history in southern Switzerland through distribution analysis of a living heritage. J Hist Geogr 38:372–386. doi: 10.1016/j.jhg.2012.01.018
- Ländliches Fortbildungs Institut LFI (2017) Edelkastanien als Solitärbaum Kastanien-Kulturen
- Lione G, Danti R, Fernandez-Conradi P, et al (2019) The emerging pathogen of chestnut *Gnomoniopsis castaneae*: the challenge posed by a versatile fungus. Eur J Plant Pathol 153:671–685. doi: 10.1007/s10658-018-1597-2
- Lione G, Giordano L, Ferracini C, et al (2016) Testing ecological interactions between *Gnomoniopsis castaneae* and *Dryocosmus kuriphilus*. Acta Oecologica 77:10–17. doi: 10.1016/j.actao.2016.08.008
- Maltoni A, Mariotti B, Jacobs DF, Tani A (2012a) Pruning methods to restore *Castanea sativa* stands attacked by *Dryocosmus kuriphilus*. New For 43:869–885. doi: 10.1007/s11056-012-9323-y
- Maltoni A, Mariotti B, Tani A (2012b) Case study of a new method for the classification and analysis of *Dryocosmus kuriphilus* Yasumatsu damage to young chestnut sprouts. IForest 5:50–59. doi: 10.3832/ifor0598-008
- Matošević D, Lacković N, Kos K, et al (2017) Success of classical biocontrol agent *Torymus sinensis* within its expanding range in Europe. J Appl Entomol 141:758–767. doi: 10.1111/jen.12388
- Matošević D, Lacković N, Melika G, et al (2015) Biological control of invasive *Dryocosmus kuriphilus* with introduced parasitoid *Torymus sinensis* in Croatia, Slovenia and Hungary. Period Biol 117:471–477. doi: 10.18054/pb.2015.117.4.3445
- Matošević D, Melika G (2013) Recruitment of native parasitoids to a new invasive host: first results of *Dryocosmus kuriphilus* parasitoid assemblage in Croatia. Bull Insectology 66:231–238
- Meyer JB, Gallien L, Prospero S (2015) Interaction between two invasive organisms on the European chestnut: Does the chestnut blight fungus benefit from the presence of the gall wasp? FEMS Microbiol Ecol 91:1–10. doi: 10.1093/femsec/fiv122
- Moriya S, Shiga M, Adachi I (2003) Classical biological control of the chestnut gall wasp in Japan. In: 1st International Symposium on Biological Control of Arthropods. USDA-Forestry Service,., Honolulu Hawaii, pp 407–415
- Müller E, Stierlin HR (1990) Sanasilva Kronenbilder : mit Nadel- und Blattverlustprozenten. Eidgenössische Forschungsanstalt für Wald, Schnee und Landschaft, Birmensdorf
- Murtagh F, Legendre P (2014) Ward's Hierarchical Agglomerative Clustering Method: Which Algorithms

Implement Ward's Criterion? J Classif 274–295. doi: 10.1007/s00357-014-9161-z

Oho N, Shimura I (1970) Research process on the chestnut gall wasp and some recent problems about its damage. Shokubutsu Boeki (Plant Prot 24:421–427

Palmeri V, Cascone P, Campolo O, et al (2014) Hymenoptera wasps associated with the Asian gall wasp of chestnut (*Dryocosmus kuriphilus*) in Calabria, Italy. Phytoparasitica 42:699–702. doi: 10.1007/s12600-014-0411-8

Panzavolta T, Bracalini M, Croci F, et al (2012) Asian chestnut gall wasp in Tuscany: Gall characteristics, egg distribution and chestnut cultivar susceptibility. Agric For Entomol 14:139–145. doi: 10.1111/j.1461-9563.2011.00551.x

Panzavolta T, Croci F, Bracalini M, et al (2018) Population Dynamics of Native Parasitoids Associated with the Asian Chestnut Gall Wasp (*Dryocosmus kuriphilus*) in Italy. Psyche (New York) 2018:. doi: 10.1155/2018/8078049

Paparella F, Ferracini C, Portaluri A, et al (2016) Biological control of the chestnut gall wasp with *Torymus sinensis*: A mathematical model. Ecol Modell 338:17–36. doi: 10.1016/j.ecolmodel.2016.07.023

Pérez-Otero R, Crespo D, Mansilla JP (2017) *Dryocosmus kuriphilus* Yasumatsu, 1951 (Hymenoptera: Cynipidae) in Galicia (NW Spain): pest dispersion, associated parasitoids and first biological control attempts. Arq Entomolóxicos 17:439–448

Picciau L, Alma A, Ferracini C (2019) Effect of different feeding sources on lifespan and fecundity in the biocontrol agent *Torymus sinensis*. Biol Control 134:45–52. doi: 10.1016/J.BIOCONTROL.2019.04.002

QGIS Development Team (2018) QGIS Geographic Information System. Open Source Geospatial Foundation Project.

Quacchia A, Ferracini C, Nicholls JA, et al (2013) Chalcid parasitoid community associated with the invading pest *Dryocosmus kuriphilus* in north-western Italy. Insect Conserv Divers 6:114–123. doi: 10.1111/j.1752-4598.2012.00192.x

Quacchia A, Moriya S, Bosio G (2014) Effectiveness of *Torymus sinensis* in the biological control of *Dryocosmus kuriphilus* in Italy. Acta Hortic 1043:199–204

Quacchia A, Moriya S, Bosio G, et al (2008) Rearing, release and settlement prospect in Italy of *Torymus sinensis*, the biological control agent of the chestnut gall wasp *Dryocosmus kuriphilus*. Biocontrol 53:829–839. doi: 10.1007/s10526-007-9139-4

R Core Team (2018) R: A language and environment for statistical computing. R Foundation for Statistical Computing

Rieske LK (2007) Success of an exotic gallmaker, *Dryocosmus kuriphilus*, on chestnut in the USA: A historical

account. EPPO Bull 37:172–174. doi: 10.1111/j.1365-2338.2007.01100.x

Rigling D, Prospero S (2018) *Cryphonectria parasitica*, the causal agent of chestnut blight: Invasion history, population biology and disease control. Mol Plant Pathol 19:7–20. doi: 10.1111/mpp.12542

Sartor C, Dini F, Torello Marinoni D, et al (2015) Impact of the Asian wasp *Dryocosmus kuriphilus* (Yasumatsu) on cultivated chestnut: Yield loss and cultivar susceptibility. Sci Hortic (Amsterdam) 197:454–460. doi: 10.1016/j.scienta.2015.10.004

Stone GN, Schönrogge K, Rachel J, et al (2002) The population biology of oak gall wasps (Hymenoptera: Cynipidae). Annu Rev Entomol 633–68

Tarcali G, Radocz L (2009) Experiences of a Study Trip in China on the Research of Chestnut Blight and Gall wasp. Analele Univ din Oradea, Fasc Prot Mediu XIV:410–419

Toyama M, Higaki M, Mishiro K, et al (2009) Population fluctuations of the chestnut gall wasp, *Dryocosmus kuriphilus* Yasumatsu, and its natural enemies in the vicinity of Tsukuba over the past fifteen years. In: Moriya S (ed) “A Global Serious Pest of Chestnut Trees, *Dryocosmus kuriphilus*: Yesterday, Today and Tomorrow”, Proceedings of the Japan-Italy joint international symposium. Tsukuba, Japan, pp 18–20

Ugolini F, Massetti L, Pedrazzoli F, et al (2014) Ecophysiological responses and vulnerability to other pathologies in European chestnut coppices, heavily infested by the Asian chestnut gall wasp. For Ecol Manage 314:38–49. doi: 10.1016/j.foreco.2013.11.031

Wickham H (2016) ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag, New York

Yara K, Sasawaki T, Kunimi Y (2010) Hybridization between introduced *Torymus sinensis* (Hymenoptera: Torymidae) and indigenous *T. beneficus* (late-spring strain), parasitoids of the Asian chestnut gall wasp *Dryocosmus kuriphilus* (Hymenoptera: Cynipidae). Biol Control 54:14–18. doi: 10.1016/j.biocontrol.2010.03.006

Zhi-Yong Z (2009) Study approaches on the chestnut gall wasp, *Dryocosmus kuriphilus* Yasumatsu in China. Acta Hortic 844:425–432. doi: 10.17660/ActaHortic.2009.844.59