Degree-day based phenological forecasting model of saddle gall midge (*Haplodiplosis marginata*) (Diptera: Cecidomyiidae) emergence

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- 1 Degree-day based phenological forecasting model of saddle gall midge (Haplodiplosis marginata)
- 2 (Diptera: Cecidomyiidae) emergence
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- 10 Abstract

Outbreaks of saddle gall midge (*Haplodiplosis marginata*) affecting wheat and other cereals are difficult to anticipate and may not be identified until damage has occurred. Earlier work on this pest has shown that degree day models can be used to predict *H. marginata* emergence based on soil temperatures. Here, we show how the availability of regular long-term trapping data can be used to update and improve upon this earlier model by predicting the progression of emergence. The emergence of adult *H. marginata* at three sites in the UK was monitored over two flight seasons using pheromone traps. The data confirmed the presence of multiple peaks in emergence over several weeks. Rainfall events followed by an accumulation of 512DD (±9.11DD) above 0 °C could be used to predict peaks with greater accuracy than degree day accumulations alone. Cumulative percentage emergence as a function of degree day accumulations was best described by a probit model. The probit model predicted *H. marginata* emergence at other sites and years to within 4 days. Application of these models will enable growers to forecast peaks in emergence, make informed assessments of crop risk and time application of chemical controls appropriately and only where required.

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1. Introduction

Saddle gall midge, Haplodiplosis marginata, (von Roser) is an occasional pest of cereals across Europe. The larval stage of this insect is phytophagous, causing the formation of saddle-shaped depressions (galls) on the stems of host plants (Rowley et al., 2016). Crops most at risk are spring wheat and spring barley (Skuhravý et al., 1983; Skuhravý et al., 1993), but the insect will also damage winter wheat and barley (Pope and Ellis, 2013). Damaged plants can exhibit a loss in yield due to shrunken grains as a consequence of galls disrupting the flow of nutrients to the ear (Woodville, 1968; Golightly, 1979). Stem breakage and secondary attack from pathogens at the site of the gall can also occur (Nijveldt and Hulshoff, 1968; Golightly and Woodville, 1974; Gratwick, 1992; Skuhravý et al., 1993). Following a resurgence of *H. marginata* outbreaks in several European countries from 2010 onwards, attempts have been made to consolidate and extend current knowledge of this insect to better inform pest management options (Censier et al., 2015; Rowley et al., 2016). Such reviews have highlighted the lack of information concerning H. marginata development and life cycle events. Haplodiplosis marginata populations can fluctuate wildly on a yearly basis, making outbreaks difficult to anticipate (Woodville, 1973; Basedow, 1986). Larvae may remain in a period of extended diapause for at least six years (Nijveldt and Hulshoff, 1968) and have been observed to form cocoons in response to drought (Censier et al., 2014a). The exact causes of diapause termination in this species are currently unknown, however the importance of both temperature and moisture has previously been highlighted (Skuhravý et al., 1983; Gratwick, 1992). Adult midges generally begin to emerge between the end of April and early May (Censier et al., 2015; Rowley et al., 2016), however early stages of infestation are seldom recognised due to the inconspicuous nature of the midge (Harris and Foster, 1999). Once damage is evident, chemical control applications are often unsuccessful as the larvae are protected by the leaf sheath (Gratwick, 1992). The use of reliable monitoring and forecasting tools are therefore critical in effective management of this pest (Censier et al., 2015).

The sex pheromone of *H. marginata* has been identified as 2-nonyl butyrate (Censier et al., 2014b) which has led to the development of species specific pheromone traps (Censier et al. 2016, Rowley et al., 2017). This advance has made it possible to reliably monitor the emergence and flight activity, providing opportunity to easily study populations in the field (Censier et al., 2016). In pest management, pheromone monitoring can be used to time chemical controls appropriately (Witzgall et al., 2010). The traps, however only provide a limited amount of advanced warning of insect activity and cannot predict the peaks in emergence which have been observed previously in this species (Censier et al., 2016). In addition, traps can be difficult to maintain consistently over an entire flight season and give no indication as to the duration of emergence. Phenological forecasting is a tool used in pest management to predict insect emergence and activity by modelling the progression of a particular developmental stage in relation to environmental variables (Prasad and Prabhakar, 2012). Such models can be used to support pheromone monitoring, by predicting when to deploy traps and identifying periods of peak activity on a year-to-year basis. Successful forecasting models have so far been developed for other pest Cecidomyiidae such as orange wheat blossom midge Sitodiplosis mosellana (Géhin) (Elliot et al., 2009; Jacquemin et al., 2014); swede midge Contarinia nasturtii (Keiffer) (Hallett et al., 2007); sorghum midge Contarinia sorghicola (Coquillett) (Baxendale et al., 1984); blueberry gall midge Dasineura oxycoccana (Johnson) (Hahn and Isaacs, 2002); and pine needle gall midge Thecodiplosis japonensis (Uchida et Inouye) (Son et al. 2007). When combined with meteorological data, models can provide assessments of crop risk over a wide geographical area and prompt farmers to inspect crops or deploy monitoring traps (Prasad and Prabhakar, 2012). Outputs from models may also feed into more complex decision support systems to guide farmers on when to employ pest management strategies (Strand, 2000). An earlier degree day based model of the development of H. marginata in the soil stage successfully predicted onset of emergence of the insect to within 4 days at the sites tested (Rowley et al., 2016). Soil moisture has previously been identified as being important in H. marginata emergence (Skuhravý et al., 1983) and here, we attempt to expand upon the earlier model by identifying the role of rainfall

in the phenology of this insect. Additionally, intensive sampling of H. marginata populations using

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pheromone traps has enabled the development of a model to describe the cumulative percentage emergence of the insect over the flight season. These models not only improve upon the previously published version by predicting cumulative percentage emergence over the flight season, but can also be used to forecast periods of peak *H. marginata* emergence and provide a much more comprehensive understanding of the development of this insect in the soil stage.

2. Materials and Methods

2.1 Field data

Haplodiplosis marginata activity was monitored at three sites in the UK: Buckinghamshire (Bucks) and Oxfordshire (Oxon) in 2015 and 2016, and Wiltshire (Wilts) in 2016. Pheromone traps were placed in two fields at each site. All fields were in wheat with the exception of one field at Bucks in 2015 which was in field beans and one field at Oxon in 2016 which was in oilseed rape. Four pheromone traps per field were arranged in transects perpendicular to the field edge, at least 40 m into the crop with a distance of 20 m between traps. Pheromone traps consisted of a standard red delta trap with a removable sticky insert (Agralan Ltd, Ashton Keynes, UK) hung on a fibreglass cane. Pheromone lures comprised a polyethylene vial containing 0.5mg (R)-2-nonyl butyrate placed in the centre of the trap (Natural Resources Institute, University of Greenwich). The trapping period began approximately a week prior to the start of the flight season (mid-April to May) and sticky cards were changed every 3-4 days for 8 weeks, after which they were changed weekly until emergence ceased. The same pheromone lures were used throughout the field season. Numbers of *H. marginata* caught at each trapping interval were counted. Hourly soil temperatures and rainfall were obtained from the UK Meteorological Office MIDAS network based on readings from weather stations that were less than 20km from each site (Met Office, 2012).

2.2 Modelling peaks in emergence

Two degree day models were developed to attempt to describe *H. marginata* emergence patterns. Peaks in *H. marginata* activity were identified from catch numbers and the start and end dates were approximated as occurring midway between counts. The first model assumed a straightforward relationship between degree day accumulations from a single date of biofix to the start of each peak (Fig 1a). Here, different DD accumulations do not represent exact physiological requirements but are used to approximate the time to emergence for groups of insects experiencing different temperatures lower down the soil profile. The second model (Fig 1b) assumed equal DD accumulations between each rainfall event and the subsequent peak, as described by Jacquemin et al. (2014) from observations of *S. mosellana* emergence.

The same biofix was used as the start of DD accumulations for both models, defined as the date of first rainfall on or after 1st March. Here, biofix represents the time when conditions were suitable for pupation to occur post-diapause. The chosen biofix assumes the diapause requirements for *H. marginata* would have been met prior to 1st March, with little or no post-diapause development. It also assumes moisture is necessary for pupation to occur, as with previous models of *H. marginata* and *S. mosellana* development (Oakley et al., 1998; Elliot et al., 2009; Rowley et al., 2016). Degree day (DD) accumulations were calculated above 0°C from hourly temperature data as described in Rowley et al. (2016). Mean hourly temperatures above 0°C were summed for each day and then divided by 24 to give degree days (Cesaraccio *et al.*, 2001). Rainfall events were classified as daily rainfall over 1 mm following 3 days without precipitation. The threshold of 1 mm was used to account for inaccuracies in monitoring equipment. For both models, the coefficient of variation of DD accumulations was calculated for all sites and years.

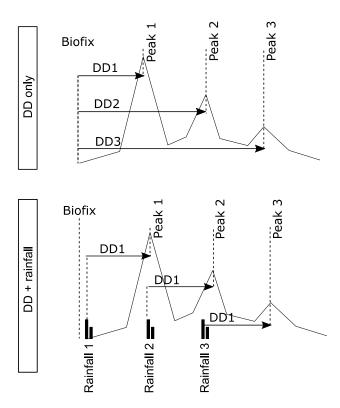


Figure 1. Representation of two different *Haplodiplosis marginata* emergence models. DD refers to degree day accumulations with numbers indicating unique DD values.

2.3 Percentage emergence model

Pooled field data for each site were used to calculate the cumulative percentage emergence for the entire flight period at each monitoring date. Monitoring dates were converted to degree days calculated from the predetermined date of biofix for each site and year, as described in section 2.2. Three regression models were tested to best describe the relationship between degree days and cumulative percentage emergence, all having previously been used successfully to describe insect development in response to time or temperature. A two-parameter Weibull function was fitted using nonlinear least squares regression:

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$$y = 100(1 - \exp(-(x/\alpha)^{\beta}))$$

Where y is the cumulative percentage emergence, x is cumulative degree days and α and β are model parameters. Two generalised linear models were also performed with binomial errors and logit or probit links (Forrest and Thomson, 2011). Model selection was done by comparing the adjusted r-squared and root mean square error (RMSE) values of models fitted to observed data (Damos et al.,

2010; Parker et al., 2011). The chosen model was validated against other sites and years for which the date of *H. marginata* emergence is known but which were not included in model development. All statistical analyses were done in R-3.2.2 (R Core Team, 2015).

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3. Results and Analysis

3.1 Modelling peaks in emergence

Emergence began no later than 2nd May (± 3 days) at all study sites and continued until as late as mid-July. Total site catches ranged from 1,755 to 20,384 individuals over the entire flight season. The catch data revealed apparent 'waves' of emergence of *H. marginata* over time, with a maximum catch rate of 200 individuals per trap per day. The first peak generally occurred soon after the initial emergence, with smaller subsequent peaks occurring at two- to three-week intervals. At the two sites for which data were obtained in both years, mean soil temperatures in April and May 2015 varied by only 2.33 °C whereas in 2016 the difference rose to 5.56 °C, reflecting a cooler April and warmer May than the previous year. Mean daily rainfall at the two sites was higher in 2016, averaging 2.28 mm compared with 1.31 mm in 2015. The maximum daily rainfall of 33.8 mm occurred in 2016 at the Oxon site. The DD-only model showed an average accumulation of 528.25DD (± 7.69DD) between the biofix and the onset of emergence. Accumulations between the biofix and subsequent peaks however were more variable, averaging 796.82DD (± 39.14DD) and 1083.68 (± 54.81DD) for peaks 2 and 3 respectively (Table 1). Across all sites and years, 14 out of 29 identified rainfall events could be linked to the beginning of peaks in emergence (Fig. 2). Subsequent rainfall events however, may have additionally contributed to increased emergence over the duration of the peak as represented by the shaded areas of Figure 2. The DD + rainfall model showed an average accumulation of 512.42DD (± 9.11DD) between a triggering rainfall event and a subsequent peak in emergence activity (Table 1, Fig.

Table 1. Degree day accumulations for the periods between biofix and emergence peaks (DD only model) and the periods between inductive rainfall events and emergence peaks (DD + rainfall model), calculated for each site and year. Mean, standard deviation (SD) and coefficient of variation (CV) values were calculated from the DD values for each peak separately for both models. Mean DD for all peaks was additionally calculated for the DD + rainfall model.

	DD only 1: Biofix - Peak			DD + rainfall : Rain - Peak		
	Peak 1 (DD1)	Peak 2 (DD2)	Peak 3 (DD3)	Peak 1 (DD1)	Peak 2 (DD1)	Peak 3 (DD1)
2015						
Bicester (Oxon)	537.24	764.91	1158.41	537.24	466.13	461.45
H. Wycombe (Bucks)	563.62	1054.46	1339.97	563.62	477.97	560.78
2016						
Bicester (Oxon)	484.68	746.86	-	484.68	551.44	-
H. Wycombe (Bucks)	534.13	694.30	926.43	534.13	486.59	516.16
Devizes (Wilts)	521.56	723.57	909.91	521.56	504.43	507.65
Mean	528.25	796.82	1083.68	528.25	497.31	511.51
SD	28.77	146.43	205.10	28.77	33.32	40.7
cv	0.054	0.184	0.189	0.054	0.067	0.079
				Mean (all peaks)		512.42
				(±\$	SEM)	(± 9.11)

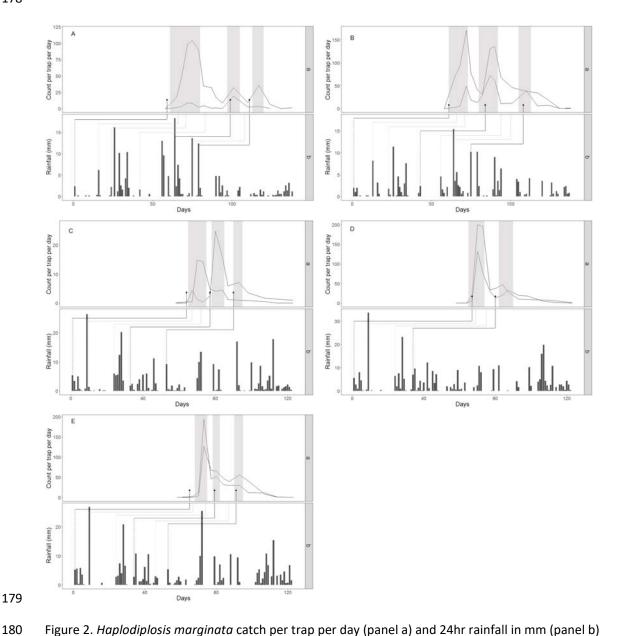


Figure 2. *Haplodiplosis marginata* catch per trap per day (panel a) and 24hr rainfall in mm (panel b) for each day of the trapping period. Black arrows represent inductive rainfall events, grey lines indicate non-inductive rainfall events. Horizontal lines represent degree day accumulations of 512 DD. Shading represents identified periods of peak emergence with the start and end dates approximated to the midpoint of trapping intervals. A) Bucks 2015, B) Oxon 2015, C) Bucks 2016, D) Oxon 2016, E) Wilts 2016

3.2 Percentage emergence model

The Weibull model was the best fitting model based on the adjusted r-squared value, accounting for 91% of the variation in the data. The GLM with probit link had the lowest RSME values (Table 2). Generation of the models showed that the Weibull model had a poor fit at the lower end of the data however, suggesting that it would be a poor predictor of the start of H. marginata emergence (Fig. 3). The probit model was therefore selected for validation and predicted that 10% emergence of *H. marginata* would occur at 550DD post-biofix. When validated against other sites and years, the probit model agreed with the observed date of emergence to within a maximum of 4 days (± 4 days), with the error reflecting uncertainty in the true emergence date as a result of the sampling interval (Table 3).

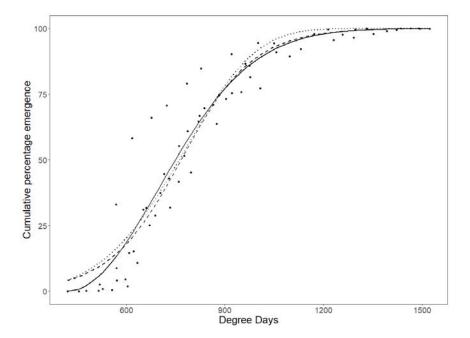


Figure 3. Percentage cumulative emergence of *Haplodiplosis marginata* as a function of accumulated degree days from biofix for all sites and years studied. Predicted emergence based on the probit model (solid line), weibull model (dotted line) and binomial GLM (dashed line) shown.

Table 2. Parameter estimates and standard error (SE) for all models. RMSE and adjusted R² values shown. Predicted DD accumulations required for 10%, 50% and 90% based on the selected model.

Model	Parameter	Est. value	SE	P-value	RMSE	Adj. R²	Model p	redictions
Binomial GLM	α	7.126	0.01616	<0.001	10.03	0.89		
	β	-47.16	0.10725	< 0.001				
Weibull	α	822.061	10.6095	<0.001	10.67	0.91		
	β	4.696	0.3915	<0.001				
Probit model	α	4.124	0.00845	<0.001	9.98	0.89	10%	550.04
	β	-27.308	0.05616	<0.001			50%	750.47
							90%	1023.93

Table 3. Observed and predicted 10% emergence dates for probit model for other sites and years, data from which were not used in model development. Differences in days between observed and predicted dates shown. Error in brackets represents uncertainty in emergence dates which are given as a midpoint between sampling dates.

		Probit Model		
Site	Observed emergence date	Predicted emergence date	Days difference (Obs – Pred	
2014				
Royston (Herts)	30 th April (± 0 days)	27 th April	3	
Bicester (Oxon)	3 rd May (± 3.5 days)	30 th April	3	
H. Wycombe (Bucks)	3 rd May (± 3.5 days)	30 th April	3	
Aylesbury (Bucks)	3 rd May (± 3.5 days)	2 nd May	1	
2015				
Royston (Herts)	2 nd May (± 2 days)	1 st May	1	
Aylesbury (Bucks)	9 th May (± 4 days)	5 th May	4	
Glemsford (Suffolk)	3 rd May (± 3 days)	2 nd May	1	
Thirsk (N. Yorks)	9 th May (± 2 days)	9 th May	0	
Devizes (Wiltshire)	3 rd May (± 3 days)	30 th April	3	
2016				
Glemsford (Suffolk)	5 th May (± 3 days)	6 th May	-1	
Max. diff	4 (± 4days)			
SD (Obs	1.93			

4. Discussion

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The work presented here clearly demonstrates how rainfall and soil temperature can be used to develop forecasts of H. marginata emergence. The association between soil temperature and moisture and the onset of emergence in this species has been made in earlier work (Rowley et al., 2016). With the availability of more detailed catch data, we have improved upon this earlier model to develop a cumulative emergence forecast for the entire flight season. Furthermore, we have confirmed the role of rainfall events in triggering H. marginata emergence, showing that moisture as well as temperature is crucial to the post-diapause development of this insect. Moisture is an important but often underappreciated aspect of insect phenology and development (Tauber and Tauber, 1976; Tauber et al, 1998). Rainfall is reported to be important in the emergence of a number of Cecidomyiidae species such as Hessian fly Mayetiola destructor (Say) (Woli et al., 2014), swede midge (Chen et al., 2011) and apple leaf curling midge Dasineura mali (Kieffer) (Cross et al., 2009) and in the case of the latter acts to increase the number of suitable oviposition sites by increasing the rate of host plant growth (Cross et al., 2009). The incorporation of soil moisture or rainfall into a degree day-based phenological model can lead to improved precision in some species (e.g. Baxendale and Teetes, 1983; Baxendale et al., 1984; Tauber et al., 1994). Early attempts to model the emergence pattern of orange wheat blossom midge, S. mosellana, recognised the importance of soil moisture in pupation and therefore emergence (Basedow, 1977; Basedow and Gillich, 1982; Oakley et al., 1998; Elliot et al., 2009). It was not until this insect was studied under controlled conditions and with more frequent field sampling that rainfall events could be linked to the inducement of this final stage of development (Jacquemin et al., 2014). Here, we show a similar effect of rainfall on *H. marginata* emergence based on field data. The onset of a precipitation period followed by the accumulation of 512DD above 0 °C predicted an increase in H. marginata emergence to within 3 days of the midpoint of the observed peaks. Just under 50% of all rainfall events were found to be inductive. The model proposed here and that proposed by Jacquemin et al., (2014) agree with theories of insect development which state that post-diapause, insects can remain in a state of

'readiness' until an environmental cue triggers the onset of pupation (Tauber and Tauber, 1976; Hodek, 1996; Koštál, 2006). Such a mechanism ensures that development typically resumes when conditions are favourable regardless of when diapause is terminated. If no cue is received, the insect recommences diapause for another year (Tauber and Tauber, 1976). This would account for the proportion of S. mosellana and H. marginata larvae that undergo extended diapause (Nijveldt and Hulshoff, 1968; Basedow, 1977), which is thought to be up to twelve years in the case of S. mosellana (Barnes, 1952). Not all rainfall events were directly linked to increased catch rates but smaller peaks in emergence may have been overlooked at the trapping interval used or due to weather conditions affecting the catch rate on particular days. Additionally, many rainfall events while not initiating peaks in emergence may still have contributed to the high numbers of insects being caught during the trapping interval (Fig. 2). Soil conditions in the preceding days might also determine the impact of a particular rainfall event. Factors such as landscape, soil type and structure will affect the permeability and moisture retention of soil, and thus the impact of any one rainfall event. Soil type however, was not found to be of significance in the emergence of swede midge, when investigated alongside soil moisture (Chen and Shelton, 2007). In their studies on sorghum midge, Baxendale et al. (1983, 1984) found that rainfall delayed emergence and drier years correlated with lower heat requirements. No such pattern was observed here, the site with the earliest emergence (Oxon 2016) also experienced 43% more rain than the next wettest site in the four weeks prior to emergence. This site also had a greater initial rate of emergence, resulting in only two clear emergence peaks rather than three (Fig. 2). This may have been due to the 33.8mm of rainfall which the site received on the 9th March, resulting in a greater proportion of larvae encountering favourable pupation conditions at once. Due to the trapping intervals used in this study and other factors affecting insect numbers over the flight season, the relationship between rainfall amount and trap catch could not be determined. It may be possible to examine this relationship under controlled conditions however, which could add additional predictive value to the model described here.

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Waves of emergence in *H. marginata* have been observed before from data collected using non-specific traps (Censier et al., 2016). Such waves may arise because not all larvae encounter the conditions necessary for pupation all at once, for example due to their depth in the soil. From an ecological perspective, this strategy increases the chance of coincidence between newly emerged adults and a suitable growth stage of the host plant. It is particularly relevant for *H. marginata*, which has a short adult lifespan of only 1 - 7 days (Nijveldt and Hulshoff, 1968; Popov *et al.*, 1998). The pheromone traps used here only captured adult males of *H. marginata*. As males tend to emerge 3 – 5 days before females (Skuhravý *et al.* 1983), the models described here based on male emergence would give sufficient warning of female emergence for farmers to be able to check crops for evidence of egg laying. The sex ratio of females to males has previously been reported by Skuhravý *et al.* (1983) as 59:41 and 54:46 based on emergence trap and Möricke trap catches, respectively. The numbers of males caught in these traps were therefore likely to be slightly lower than, or comparable with, the number of females emerging.

The importance of the start date in calculating DD accumulations is widely recognised (Pruess, 1983) and model accuracy can be improved if there is a biological basis for the date selected (e.g. Riedl et al., 1976). This date is commonly referred to as the 'biofix'. Previous work on the development of *H. marginata* proposed a point of biofix as the first rainfall event once mean daily soil temperatures rose above 6 °C after the 1st March (Rowley et al., 2016). Now, with more comprehensive emergence data, we propose simplifying the model to remove the temperature threshold. Laboratory observations of *H. marginata* development suggest that pupation will proceed at 10 °C but not at 5 °C, therefore the lower developmental threshold is likely to fall within this range (Baier, 1963; Nijveldt and Hulshoff, 1968). The 1st March is often selected as a start date for DD accumulations where there is a no lower developmental threshold data to draw upon (Pruess, 1983), such as models of *S. mosellana* emergence (Wise and Lamb, 2004; Elliot, 2009). The same date was used here based on a lack of observed *H. marginata* development in the field prior to 1st March previously (Pope and Ellis, 2013), and the assumption that earlier soil temperatures had little effect on *H. marginata* development as they were

at the lower end of the developmental threshold range. Photoperiodism can also play a major role in the termination of insect diapause, which further validates removing the temperature threshold from the biofix estimate (Tauber and Tauber, 1976; Saunders, 2014).

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The DD-based cumulative emergence model proposed here for H. marginata predicted 10% emergence would occur at 550 DD post-biofix and was accurate to within 4 days (± 4 days) when validated against observations from other sites and years. The value for 10% emergence was deemed to be an appropriate proxy for the start of emergence given the error involved in trapping insects at very low densities; it is unlikely that the earliest onset of emergence will have been recorded particularly in 2014 when pheromone traps were not available. The predictive capacity of the model to within 4 days is on a par with the previously published DD emergence model (Rowley et al., 2016). The advantage of the new model is the ability to predict cumulative emergence over the entire flight season, rather than just the start date. This will enable the midpoint and conclusion of flight periods to be estimated and aid in the assessment of the need for chemical controls or the effectiveness of insecticides applied earlier in the emergence period. It may mean that pest management options can be used more judiciously, so that chemical controls are only applied if the crop is at a vulnerable growth stage prior to the mid-point of emergence. The model produced here relies on data collected by a national network of weather stations. This means that forecasts can be made for different parts of the country, providing estimates based on local weather conditions. Multiple pest forecast models could be used to identify different periods of activity from the same meteorological data, for example the VIPS automatic forecasting system developed in Norway (NIBIO, 2017) and the CIPRA model in Quebec (Bourgeois et al., 2005). Ideally, such a forecasting system would be used alongside crop growth forecasts to provide an assessment of crop risk throughout the flight period. Continued application and evaluation of predictive models for H. marginata and other pest species will further improve the reliability of such forecasts in the future.

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