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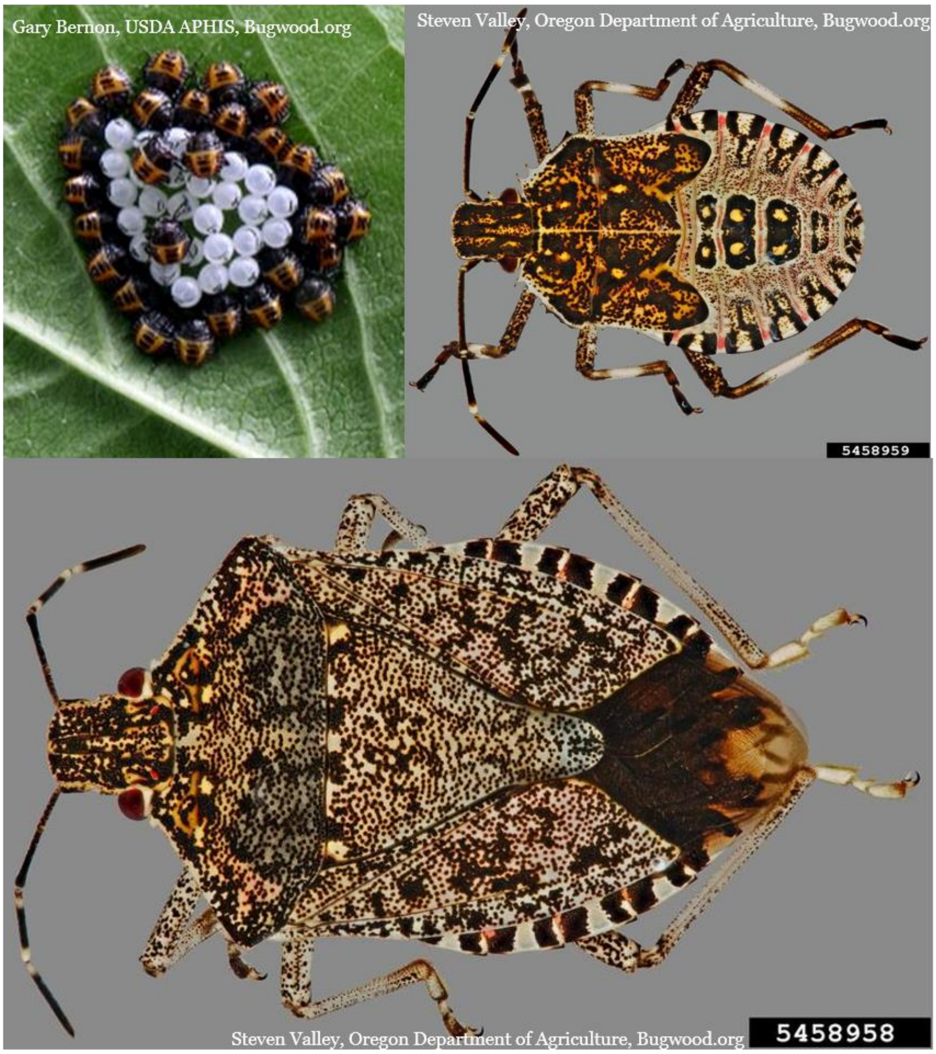
Monitoring for the Brown Marmorated Stink Bug (BMSB) *Halyomorpha halys* in Scotland

PHC2019/01 - Project Final Report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1 Executive Summary

1.1 Background

The increase in global trade brings with it the risk of spread of new pests and diseases into Scotland. *Halyomorpha halys*, Brown Marmorated Stinkbug (BMSB) is an invasive pest that has already become established in North America and several European countries. The insect aggregates inside houses over winter and can cause problems as an urban nuisance pest in homes as well as being a pest of agriculture. Preventing introduction of this species is particularly difficult as overwintering adults aggregate in sheltered spots and buildings and may be moved from infested areas with numerous commodities. BMSB has been intercepted entering the UK on several occasions in recent years, located on passenger luggage or imported goods. However, establishment of populations has not been reported. Detection is difficult until damage is noticed. The BMSB attacks a wide range of hosts including *Rubus idaeus* (raspberry) and *Prunus avium* (sweet cherry). The potential for economic impact in Scotland is not yet fully understood, however conditions in Scotland may be suitable for establishment. In order to meet trade requirements (Scottish soft fruit industry requires pest freedom status in order to maintain trade with countries setting out strict biosecurity measures for BMSB) and to allow early detection and control of this pest, a pilot Scottish BMSB monitoring exercise was carried out. Modelling of climatic suitability for BMSB establishment in Scotland was also undertaken.

1.2 Key Research question

The aim of this project was to gain a better understanding of the future threat of *Halyomorpha halys* the Brown Marmorated Stink Bug (BMSB) and to determine the likely outcomes for any potential outbreak in Scotland. We investigated the possibility that BMSB presents a significant threat to Scotland's soft and tree fruit industries and the likelihood of the pest becoming established in Scotland under current and future climate conditions and growing practices.

1.3 Research undertaken

Co-ordinated monitoring for the presence of BMSB was undertaken by teams at SASA and the James Hutton Institute. To determine the presence and abundance of BMSB in Scotland, 10 trapping sites were selected based on their proximity to locations considered to be 'risk points' for introduction and 'at risk' for potential damage to industry. Commercial traps were set up and they remained in position for approximately 12 weeks between July and October 2019. The morphology of any potential specimens trapped in the commercial traps was examined under a binocular microscope. Individuals of interest were removed and tentatively identified using classical taxonomy. A reference collection of voucher specimens of common UK stinkbug species was established. DNA barcoding was conducted via amplification and sequencing of the CO1 subunit (Cytochrome Oxidase) and comparison of the resulting sequences with public DNA databases. A process-oriented climate-based niche model software package CLIMEX (Kriticos *et al.*, 2017) was used by a team at SRUC to determine the areas in Scotland that are suitable for the establishment of BMSB under current and future climates. Scottish climate data was collected, and the model was used to assess the risk of BMSB becoming established both outdoors and within protected cropping environments.

1.4 Main findings

Traps collected a substantial number of arthropods but no BMSB were found.

Two traps recorded one native shield bug each (*Acanthosoma haemorrhoidale* [Hawthorn shield bug] and *Elasmostethus interstinctus* [Birch shield bug] but all other individuals collected were from alternative arthropod families. This suggests that the pheromone lures did not selectively attract native shield bug species since these did not appear over-represented in the catch. Traps in more sheltered locations collected fewer individuals than those in more exposed situations, suggesting that the majority of individuals caught were via accidental impact rather than lured.

A process-oriented climate-based niche model software package (CLIMEX) used to analyse Scottish temperature climatic data and output suggests that, in Scotland, there would be a limited annual reproductive success under current climatic conditions in an outdoor situation. Climate change projections show that in outdoor environments in Scotland, BMSB populations would be unlikely to become established until the end of the century. However, temperature data from protected environments suggests that there is the potential for 2 or even 3 generations per year, depending on when BMSB is introduced. The tendency of BMSB to congregate does make it more likely that Scotland could end up with small 'clusters' of populations, particularly if sheltering indoors over the winter months. There is potential for individuals who have successfully made it through reproduction at a lower average temperature to accelerate genetic drift if enough variation exists in the introduced population.

1.5 Recommendations and next steps

Monitoring for the presence of BMSB has contributed to the improved surveillance and detection of plant health threats to Scotland and has demonstrated that the needs of stakeholders are being facilitated.

Modelling of the potential distribution of the pest has identified that protected cropping may provide key areas for establishment and should be the focus for future surveillance and a key focal point for future policy decisions and outbreak management. Opportunities to develop links with pest practitioners should be encouraged to aid the development of risk analysis and control strategies.

Since the completion of this project, BMSB has been intercepted several times in the south of the UK by researchers at NIAB EMR. The most recent detection was reported widely in a general press release on the 22nd August 2020 when a flying adult was caught on a pheromone trap at an RSPB nature reserve in Rainham Marshes. Glen Powell (NIAB EMR) stated that 'this suggests that adults may be actively dispersing in search of mates and food sources.' When the cumulative degree days for the Rainham Marshes site are assessed, BMSB could potentially have completed 1.9 generations in 2018, 1.5 generations in 2019, and 1.1. generations to date (end August) in 2020.

There is a risk that the chosen sites for monitoring in Scotland have not been able to detect the pest, even though it may have become established elsewhere in Scotland. Therefore, one recommendation would be to encourage growers to undertake pheromone trapping themselves, particularly if they grow crops under protected environments and their growing sites are within areas indicated by the modelling results as being potentially able to support the establishment of BMSB.

The development of a voucher specimen collection, and a UK stinkbug DNA barcode database, has strengthened the emergency response plan and increased the capacity for rapid identification. Industry resilience has been improved and we have developed greater capacity to respond to potential outbreaks.

Stakeholders, inspectors and members of the public should be encouraged to report potential BMSB sightings and seek accurate species identification via teams at the James Hutton Institute, SRUC and SASA. A central reporting point, potentially through the PHC website, could facilitate reports from members of the public and increase the possibility of early detection. Novel findings and control advice should continue to be reported to stakeholders at appropriate KE events.

2 Introduction

The increase in global trade brings with it the risk of spread of new pests and diseases into Scotland. Transportation of non-native species by human action can allow species into novel habitats beyond their normal geographic range. The Brown Marmorated Stink Bug (BMSB), *Halyomorpha halys*, (Hemiptera: Pentatomidae) is one such species which has already established and spread to every Northern Hemisphere continent including North America and mainland Europe. BMSB is an invasive polyphagous pest species that can cause significant economic loses to agriculture (Maistrello *et al.,* 2017, Valentin *et al.,* 2017). It attacks a wide range of hosts including a huge variety of fruit, vegetable and non-crop plants (Leskey *et al.,* 2012, Lee *et al.,* 2013). It feeds by piercing and sucking on plant tissues, and on fruits it can cause deformities that make the crop unmarketable. The damage can also aid the transmission of secondary infections from other pathogens. In addition to its status as an agricultural pest, BMSB also has nuisance status as large aggregations of adults can be found overwintering in man-made structures. When disturbed the stink bugs emit a foul-smelling scent (Watanabe *et al.,* 1978).

Preventing the introduction of this species is particularly difficult as aggregations of adults can be moved from infested areas with numerous commodities and there have often been interceptions associated with trade and postal goods. It can be difficult to determine distribution records based on detection alone, as many reports represent populations or individuals associated with human-mediated transport, which do not necessarily indicate that the pest is established. Detection can be difficult until damage is noticed, by which time populations may have reached levels that result in economic impacts (Valentin *et al.,* 2017). Identification requires a level of expertise, as this species is similar to native Pentatomidae. Early detection and better understanding of the source and likelihood of introductions is key to limiting the impact of this pest. Therefore, monitoring and modelling studies can be useful to estimate the current and potential distribution of BMSB.

BMSB is similar in appearance to several native British shield bug species, such as *Dolycoris baccarum (see Bugwwod for images)* and *Pentatoma rufipes (see Bugwwod for images)*. Adult BMSB can be distinguished from most native species by their black and white antennae, mottled legs and underside, and the absence of hairs on their body (see Figure 1).

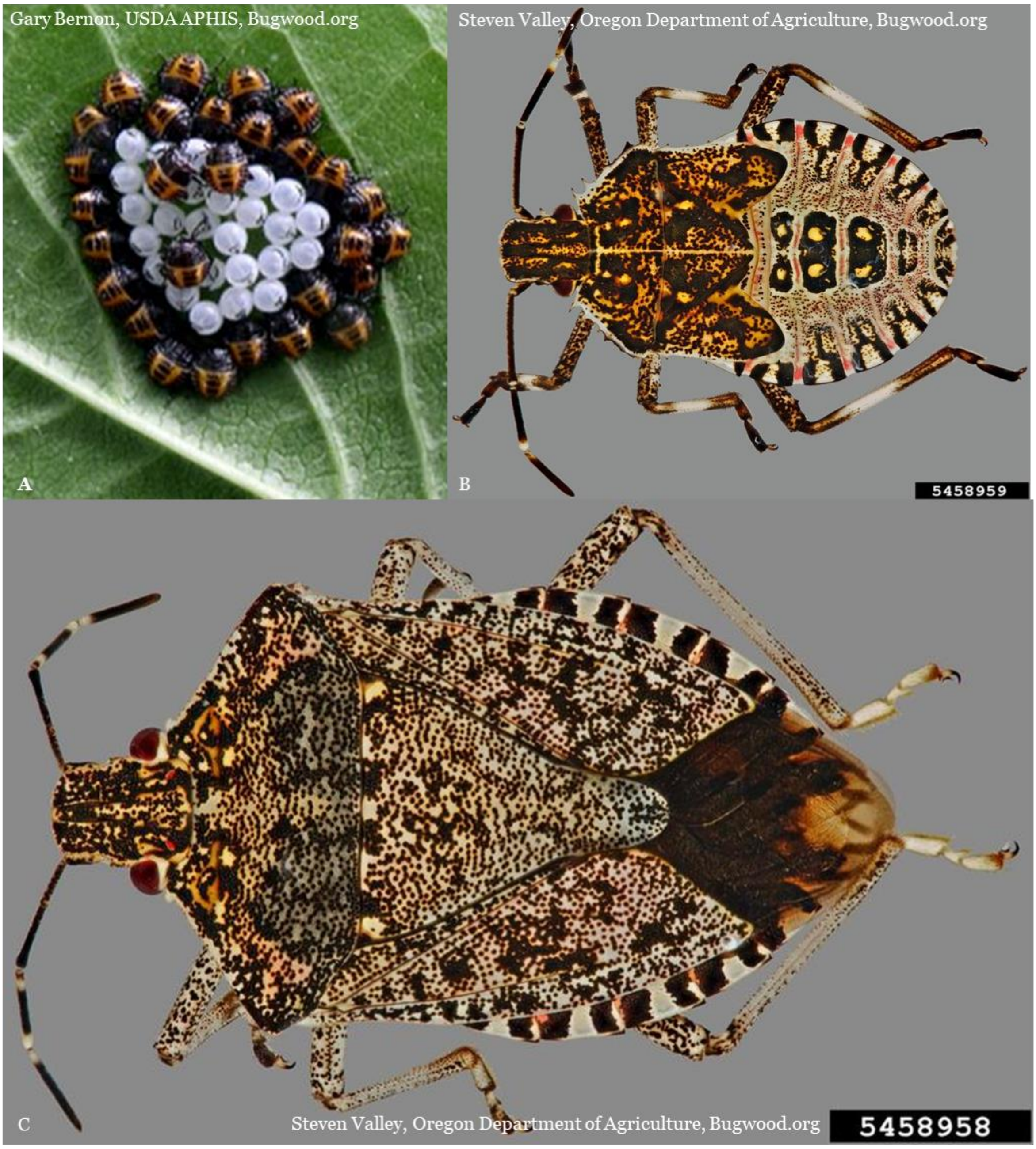

Figure 1. Images of BMSB at different life stages. A: Newly-hatched egg mass, Gary Bernon, USDA APHIS, Bugwood.org; B: Halymorpha haly nymph dorsal, Steven Valley, Oregon Department of Agriculture, Bugwood.org; C: Halymorpha haly adult dorsal Steven Valley, Oregon Department of Agriculture, Bugwood.org.

The potential economic impact to Scotland is not yet fully understood. However, changes in climatic conditions and in agricultural growing methods may have increased the risk of establishment. The Scottish soft fruit industry has concerns regarding impact on their businesses and require pest freedom status to maintain trade with countries setting out strict biosecurity measures for BMSB. To meet these trade requirements and to allow early detection and control of this pest, a pilot Scottish BMSB monitoring system was carried out in 2019.

The purpose of this pilot project was to monitor for the presence of, and assess climatic suitability for, Brown Marmorated Stink Bug in Scotland. As this invasive species is morphologically similar to several native species, the aim was to develop robust identification methods (both classical and molecular) to allow rapid, accurate diagnostics for future potential sightings by inspectors and industry.

The objective was to establish techniques for early detection of this pest using a combination of pheromone trap monitoring and sweep netting. Identification methods were developed to enable a future diagnostic service to be provided by SASA and The James Hutton Institute. In addition, modelling based on meteorological data, and that obtained from protected cropping, provides output that identifies the potential of any given location to support a permanent population of BMSB under current and future climates and helps to identify areas most suited for future monitoring studies.

3 Co-ordinated Monitoring for the presence of BMSB (SASA/Hutton)

3.1 Methodological approach

Monitoring was focused on 'at risk' sites (fruit growing area) and 'risk points' sites close to airports and distribution centres with known host plants on site. Ten trapping sites were selected across Scotland (Figure 2) based on their proximity to locations considered to be 'risk points' for introduction and 'at risk' for potential damage to industry. Target locations included soft fruit producers, distribution warehouses, botanical gardens, fruit markets and research institutes (Table 1).

Figure 2 -map of trap locations

Key to site identifiers - Blue – risk of introduction; Pink – risk to agriculture/biodiversity; Green – site identified but no trapping occurred.

Table 1- Site, assessed risk and trap location details

	Site, assessed risk and trap location details	
Site	Assessment	Trap location
1a – Glasgow	Introduction – near airport	Metal structure (indoors)
1b – Glasgow	Introduction – near airport	Small woodland (outdoors)
2a – Glasgow	Introduction – fruit market	Fruit market (indoors)
2b – Glasgow	Introduction – fruit market	Mixed woodland (outdoors)
3a – Edinburgh	Introduction – near airport	Glasshouses (indoors)
3b – Edinburgh	Introduction – near airport	Woodland edge (outdoors)
4a – Edinburgh	Introduction – city centre	Fruit plots (outdoors)
4b – Edinburgh	Introduction – city centre	Arboretum (outdoors)
5a - Rosyth	Introduction – port and distribution hub	Orchard – sycamore tree (outdoors)
5b - Rosyth	Introduction – port and distribution hub	Orchard – plum tree (outdoors)
6a – Loch Leven	Risk to biodiversity	Mature pine woodland (outdoors)
6b – Loch Leven	Risk to biodiversity	Mature mixed woodland (outdoors)
7a – Blairgowrie	Risk to agriculture	Outside glasshouse (outdoors)
7b – Blairgowrie	Risk to agriculture	Glasshouse (indoors)
8a – Invergowrie	Risk to agriculture	Hawthorn bush (outdoors)
8b - Invergowrie	Risk to agriculture	Cherry polytunnel (indoors)
9a - Arbroath	Risk to agriculture	Cherry tree (outdoors)
9b - Arbroath	Risk to agriculture	Blueberry tunnel (indoors)
10a -Aberdeen	Risk to agriculture	No trapping

Following current best practice for detection, commercially available double-sided clear sticky traps were used (Pherocon^{R} BMSB STKY ™) in combination with high-dose species specific pheromone lures (Figure 3). These lures are not designed to attract the pest to a site from distant areas but will detect if the pest is present locally. Two traps were positioned at each site and they remained in position between June and October 2019 for approximately 12 weeks (the lifetime of the pheromone lure). Sticky traps were collected and replaced around week 6 and the remaining traps and lures were removed at the end of the test period.

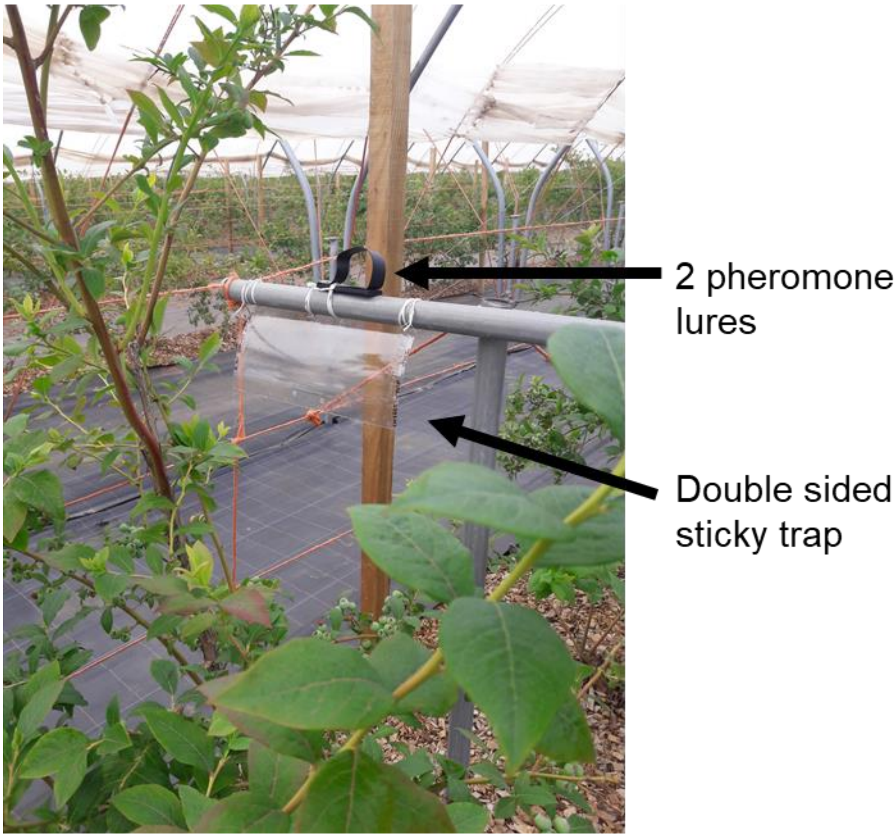

Figure 3 Sticky trap and pheromone lure positioned in a protected growing environment

In addition to trapping using the commercial sticky traps, Rothamsted Insect Survey light traps present at SASA and Hutton were monitored for the presence of BMSB over July and August, as a potential long-term monitoring tool for the pest. Native species were collected by SASA field workers over June-September to provide reference material for taxonomic and molecular identifications.

3.2 Results

Traps collected a substantial number of arthropods but no BMSB were found (Figure 4). Two traps (3B and 5B) recorded one native shield bug each (*Acanthosoma haemorrhoidale* [Hawthorn shield bug] and *Elasmostethus interstinctus* [Birch shield bug] respectively) but all other individuals collected were from alternative arthropod families. This suggests that the pheromone lures did not selectively attract native shield bug species since these did not appear over-represented in the catch. Traps in more sheltered locations (e.g. SASA Trap 3A) collected fewer individuals than those in more exposed situations, suggesting that most individuals caught were via accidental impact rather than lured.

Figure 4 Sticky trap with a typical catch- no BMSB present

4 Collection and barcoding of voucher specimens (SASA/Hutton)

Traps were examined under binocular microscope for the presence of BMSB and other (native) shield bug species. Individuals of interest were removed and tentatively identified using classical taxonomy. In addition, shield bugs collected locally by the team, submitted by members of the public and inspectors were added to the collection. Voucher specimens were preserved, and a sample (two legs from adults) taken for DNA extraction and analysis. Insect legs and DNA were shared between institutes where necessary to complete the library.

In total SASA now hold eight native species, one European species and BMSB (from the USA) within their voucher specimen collection and reference barcoding library, allowing them to provide more robust ID's on suspected cases of BMSB reported by inspectors and members of the public.

Table 2 – Summary of Species Collected

Genus	*Species*	Common Name
Acanthosoma	*haemorrhoidale*	Hawthorn Shieldbug
Dolycoris	*baccarum*	Hairy Shieldbug
Elasmostethus	*interstinctus*	Birch Shieldbug
Elasmucha	*grisea*	Parent Bug
Graphosoma	*lineatum*	Striped Shieldbug *
Halyomorpha	*halys*	Brown Marmorated Stink Bug**
Pentatoma	*rufipes*	Forest/Red-Legged Shieldbug
Picromerus	*bidens*	Spiked Shieldbug
Piezodorus	*lituratus*	Gorse Shieldbug
Rhacognathus	*punctatus*	Heather Shieldbug
* *collected outside UK*		
** *historical plant health interception specimen*		

4.1 DNA barcoding

DNA barcoding was conducted via amplification and sequencing of a region of the CO1 subunit (Cytochrome Oxidase) (Hebert *et al.*, 2003), and comparison of the resulting sequences with public DNA databases. Barcoding allows for rapid and accurate identification of all life stages of the insect, although it relies on the correct pre-identification of voucher specimens prior to submission of the sequence to the database.

Conflicting identifications made by different labs can compromise the effectiveness of reference libraries and public databases. To avoid this scenario, we established a local reference library using verified voucher specimens. Fragments of confirmed specimens of native and non-native species were shared between institutes where necessary. DNA was barcoded by the separate institutes and results were combined to provide a local reference library of sequences. Remaining fragments and whole specimens were preserved to form the reference voucher collection. Two DNA extraction methods and amplification methods were trialled:

1. A sodium hydroxide DNA extraction method described in Malloch et al., 2006 was used. DNA was extracted from whole specimens and from excised fragments (single leg).
2. Immature individuals (various instar nymphs) were DNA extracted using a SASA in-house method (Sjolund *et al.*, 2016), which allows DNA extraction of the entire sample but leaves the voucher specimen intact.

PCR Ready to Go beads (GE Healthcare) were used to amplify the barcode region according to the manufacturer's instructions.

Representative PCR products from each specimen were selected for sequencing. A consensus sequence was generated for each specimen and this was used to search public DNA databases. Results from DNA barcoding and taxonomic species identification were in agreement for all specimens. In addition to a barcode sequence from BMSB, SASA now hold a verified local reference barcode library of 8 native UK stinkbug species and one European species.

4.2 Testing the DNA barcoding system on degraded insect specimens

DNA extraction (using SASA's non-destructive method) and sequence analysis from non-target shield bugs collected on traps was tested and specimens were successfully identified, although both individuals were relatively fresh (and therefore with more intact DNA). To test the extraction system on insects where the quality of the DNA may have degraded over time, other insect orders were removed from the sticky traps for barcoding. The traps had been in situ for 6 weeks and, in some cases, would have been exposed to precipitation and varying temperatures and weather systems. Under these conditions, the insect DNA would likely degrade quickly after insect death.

As there was a limited number of stinkbugs trapped on the sticky traps, three common non-target insects (earwig, ladybird, hoverfly) were selected from trap 1A to test the barcoding protocol on potentially degraded specimens. DNA was extracted from a fragment of the insect (head or leg) using the sodium hydroxide method described above. The mitochondrial barcode sequence of each insect was successfully amplified and sequenced.

4.3 Summary

Two separate DNA extraction and amplification methods were successfully used to barcode fragments from the same insect specimen, suggesting that either method would be suitable for analysis of stinkbug specimens or fragments.

The sequencing results from the trap caught non-target specimens suggest that DNA can be successfully extracted and barcoded from potentially degraded fragments of insect material trapped on the Pherocon BMSB STKY sticky traps using a rapid, inexpensive and simple sodium hydroxide DNA extraction method. Therefore, it is likely that this method would be suitable for extraction and downstream analysis of trapped, possibly degraded, stinkbug body fragments. This method would also allow the remaining body parts of the insect to be preserved for taxonomic identification and voucher reference libraries.

5 Determining areas suitable for establishment of Brown Marmorated Stink Bug (BMSB) under current and future climates (SRUC)

5.1 Methodology

As described earlier in this report, ‘at risk’ sites (fruit growing area) and ‘risk points’ sites (proximity to airports and distribution centres with known host plants on site) were monitored using pheromone traps and/or intensively sampled for BMSB, although none were found. At several of these sites data loggers (Easylog) were used to record temperature and relative humidity data. Some data sets from the James Hutton Institute and commercial growers were also obtained to allow temperature data from within polytunnels and Seaton tunnels to be assessed to determine whether sufficient degree-days are accumulated to allow a generation of BMSB to occur.

The degree-days accumulated at various locations were also used (obtained from https://degreedays.io/) to assess suitability of specific locations for BMSB.

As a member of the International Pest Risk Research Group (IPRRG- https://pestrisk.org/), Dr Evans (SRUC) has access to a model developed by the IPRRG using CLIMEX (Kriticos *et al.* 2017) - a process-oriented climate-based niche modelling software package. CLIMEX has been widely used to model the potential distribution of many invasive arthropod pests including BMSB (Kriticos *et al.*, 2017), and access to the model allowed use of Scottish data to assess the risk of BMSB establishment outdoors and within protected cropping using the data outlined above.

The CliMond CM10 World (1975H V1.2) climate dataset was used for model fitting (Kriticos *et al.*, 2012). This global climatological dataset has a spatial resolution of 10 arc minutes and consists of long-term (30 year) monthly averages of daily minimum and maximum temperature, relative humidity at 09:00 and 15:00 h and monthly rainfall totals.

As well as assessing the current (historical) climate for the establishment of BMSB in outdoor situations, climate projections for 2030, 2080 and 2100 based on IPCC IV SRES Scenarios A1B (medium emissions) were also evaluated within the CLIMEX software. Data from the CSIRO Mk3.0 Global Climate Model were used to create estimates of the future climate for a range of future dates (Harris *et al.*, 2014) for use within CLIMEX.

CLIMEX calculates an index of overall climate suitability, the Ecoclimatic Index (EI), which is theoretically scaled between 0 (unsuitable) and 100 (climatically perfect all year round). The EI represents the net effect of the opportunity for growth as indicated by the annual Growth Index, discounted by several stress indices such as the cold, dry, heat and wet stress indices, and the interaction indices of cold-dry, cold-wet, hot-dry and hot-wet.

The CLIMEX parameter values used are those developed by Kriticos *et al.* (2017) and the IPRRG (Table 3).

Table 3 *CLIMEX parameter values for BMSB*

Parameter	*Mnemonic*	*Unit*
Temperature requirements		
Limiting low temperature	DV0	12°C
Lower optimal temperature	DV1	27°C
Upper optimal temperature	DV2	30°C
Limiting high temperature	DV3	33°C
Degree-days per generation	PDD	595 °C days
Soil moisture		
Limiting low soil moisture	SM0	0.1
Lower optimal soil moisture	SM1	0.5
Upper optimal soil moisture	SM2	1
Limiting high soil moisture	SM3	1.5
Diapause		
Diapause induction day length	DPD0	12 h light
Diapause induction temperature	DPT0	5°C
Diapause termination temperature	DPT1	5°C
Diapause development days	DPD	0 days
Diapause summer (1) or winter (0)	DPSW	0
Cold stress		
Temperature threshold	TTCS	-18°C
Stress accumulation rate	THCS	-0.01 Week^{-1}
Heat stress		
Temperature threshold	TTHS	33°C
Stress accumulation rate	THHS	0.01 Week^{-1}
Dry stress		
Threshold soil moisture	SMDS	0.1
Stress accumulation rate	HDS	-0.01 Week^{-1}
Wet stress		
Threshold soil moisture	SMWS	1.5
Stress accumulation rate	HWS	0.002 Week^{-1}
Hot-wet stress		
Threshold soil moisture	TTHW	28
Threshold temperature	MTHW	1.5°C
Stress accumulation rate	PHW	0.007 Week^{-1}

5.2 Results

5.2.1 Outdoor cropping - Ecoclimatic Index (EI)

The EI output from the CLIMEX model for BMSB under a range of climate scenarios including current, 2030, 2080 and 2100 average climate is shown in Figure 5.

Figure 5A shows very limited areas in the southeast of England where BMSB could potentially establish under the current average climate in the UK, with EI values for these locations ranging from 8 to 11. To put these scores in perspective, Kriticos *et al.* (2015) discuss classifying EI values into Unsuitable, Marginal, Sub-Optimal and Optimal, working back from the highest EI values in the model and splitting them into approximately equal intervals. Using this approach, a whole world run of the BMSB CLIMEX model produces a maximum EI value of 76, which is where 11 generations a year of BMSB would theoretically be possible. The only positive EI values where BMSB could potentially establish under the current average climate in the UK from Figure 5A, indicate 1 generation of BMSB may be possible. Consequently, these areas can be considered as Marginal at best for the establishment of BMSB. All other areas of the UK do not have sufficient degree-days to allow BMSB to complete a generation, hence an EI of zero and Unsuitable for establishment of BMSB. Temperature is a significant driver of BMSB establishment and, in the Marginal areas for BMSB establishment, the Temperature Index within the CLIMEX model is low (11-12 out of a theoretical maximum of 100). This reinforces the fact that insufficient temperature accumulation through degree-days is a key driver for BMSB establishment in outdoor situations under the current average climate.

Applying climate projections for 2030 produced a wider range of areas within the southeast of England, small areas in South Wales, Cornwall and Cheshire that are marginally suitable for BMSB establishment (Figure 5B), with a maximum EI value of 12, and therefore only a slight increase on the maximum of 11 under the current average climate (Figure 5A).

Applying climate projections for 2080 produced an expanded range of areas including most of the south of England extending into the Midlands, with South Wales and northwest England that are marginally suitable for BMSB establishment (Figure 5C), but again with a low maximum EI value of 14 (Figure 5C). However, there is also an area at the extreme northeast tip of Scotland that has a positive EI value of 10, which could be classed as marginal for the establishment of BMSB. This area is sited around Crimond in Aberdeenshire, between Fraserburgh and Peterhead.

Applying climate projections for 2100 produced a further expanded range of areas within England and Wales but also included the northeast of Scotland and along the east coast of Scotland that are marginally suitable for BMSB establishment (Figure 5D), with EI values between 9 and 11.

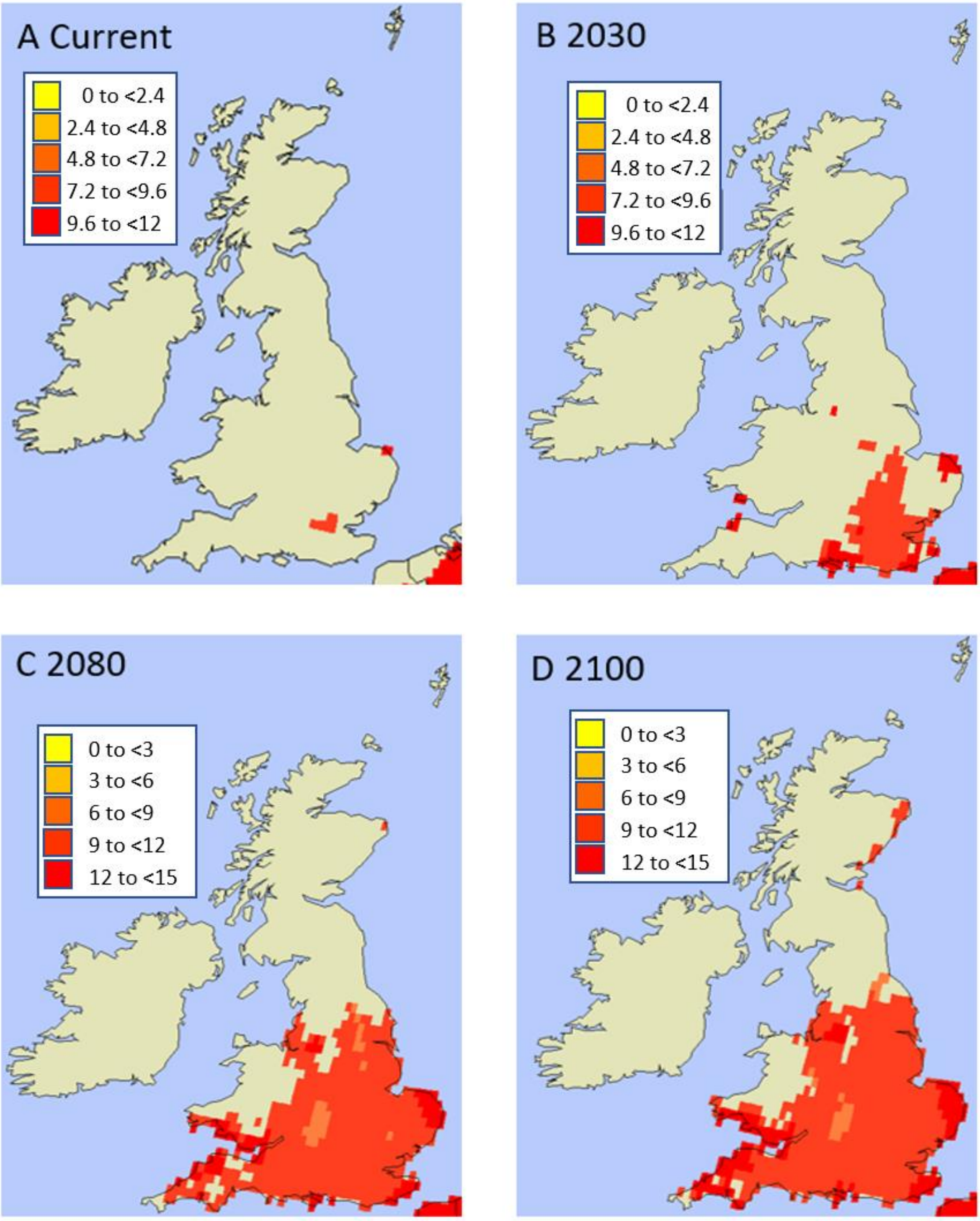

Figure 5. The Ecoclimatic Index (EI) for BMSB under the current climate (A), and projected climates 2030 (B), 2080 (C) and 2100 (D).

5.2.2 *Outdoor cropping – Number of generations*

In a similar way to the EI value, a map of the No. of generations of BMSB under the current average climate (Figure 6A) illustrates the unsuitability of Scotland and other areas of the UK for allowing the establishment of BMSB in outdoor areas. In Scotland in particular, there are only sufficient degree-days to allow 0.5 generations of BMSB in a season (Figure 6A), consequently a very low risk of establishment.

The No. of generations of BMSB under a projected 2030 climate (Figure 6B) highlights the continuing unsuitability of Scotland and other northern areas of the UK for allowing the establishment of BMSB in outdoor areas. In Scotland, there would only be sufficient degree-days to allow 0.5-0.6 generations of BMSB in a season (Figure 6B), consequently there is a low risk of establishment.

Under a projected 2080 climate (Figure. 6C) northern areas of the UK largely remain unsuitable for allowing the establishment of BMSB in outdoor areas. However, as mentioned above, there is the possibility of one generation of BMSB being completed outdoors around Crimond in northeast Scotland (Figure. 6C). A closer assessment focussed on Scotland (Figure 5D) highlights the areas that are almost capable of allowing one generation of BMSB to be completed. The east coast of Scotland has areas from Aberdeenshire in the northeast down to the Scottish Borders in the south that have sufficient degree-days to allow 0.8-1.0 generations of BMSB in a season (Figure6D). Consequently, there could be a risk of BMSB establishment outdoors, particularly where there may be seasonal variability in temperatures.

Under a projected 2100 climate (Figure 6E) most northern areas of England and the north and north western areas of Scotland remain unsuitable for allowing the establishment of BMSB in outdoor areas. However, areas of northeast and east Scotland along the east coast from Aberdeenshire in the north east down to the East Lothian have sufficient degree-days to allow one generation of BMSB to be completed outdoors (Figure 6E). A more detailed look at Scottish data (Figure 6F) indicates that there is a larger area where a generation of BMSB may be possible, particularly where there may be seasonal variability in temperatures.

One limitation within the CLIMEX software outputs (CliMond version 1.2) is that the 30 year average climate data are from the period 1960-1990 (Kriticos *et al.*, 2012) and do not take into account climate change in this century to date. Using the locations identified in Figures 5A and 6A, the cumulative degree-days using the standard base temperature of 12°C (Table 3) were obtained for the last 3 years (2017, 2018 and 2019) and are summarised in Table 4.

Table 4 *The cumulative degree-days above a 12°C base temperature for BMSB development (595° days required to complete a generation) at five English locations where establishment may be possible based on CLIMEX model output for the current average climate. Actual data for 2017, 2018 and 2019.*

	Cumulative degree-days above 12°C and potential No. of BMSB generations in parentheses		
Location (postcode)	2017	2018	2019
NR11 8QQ (Norfolk)	711 (1.2)	772 (1.3)	572 (0.96)
IG6 1AL (London)	920 (1.5)	1214 (2.04)	956 (1.6)
BR7 6LY (London)	917 (1.5)	1116 (1.88)	830 (1.4)
SW20 0SR (London)	871 (1.5)	1184 (1.99)	894 (1.5)
TW20 0HJ (London)	847 (1.4)	1004 (1.69)	689 (1.2)

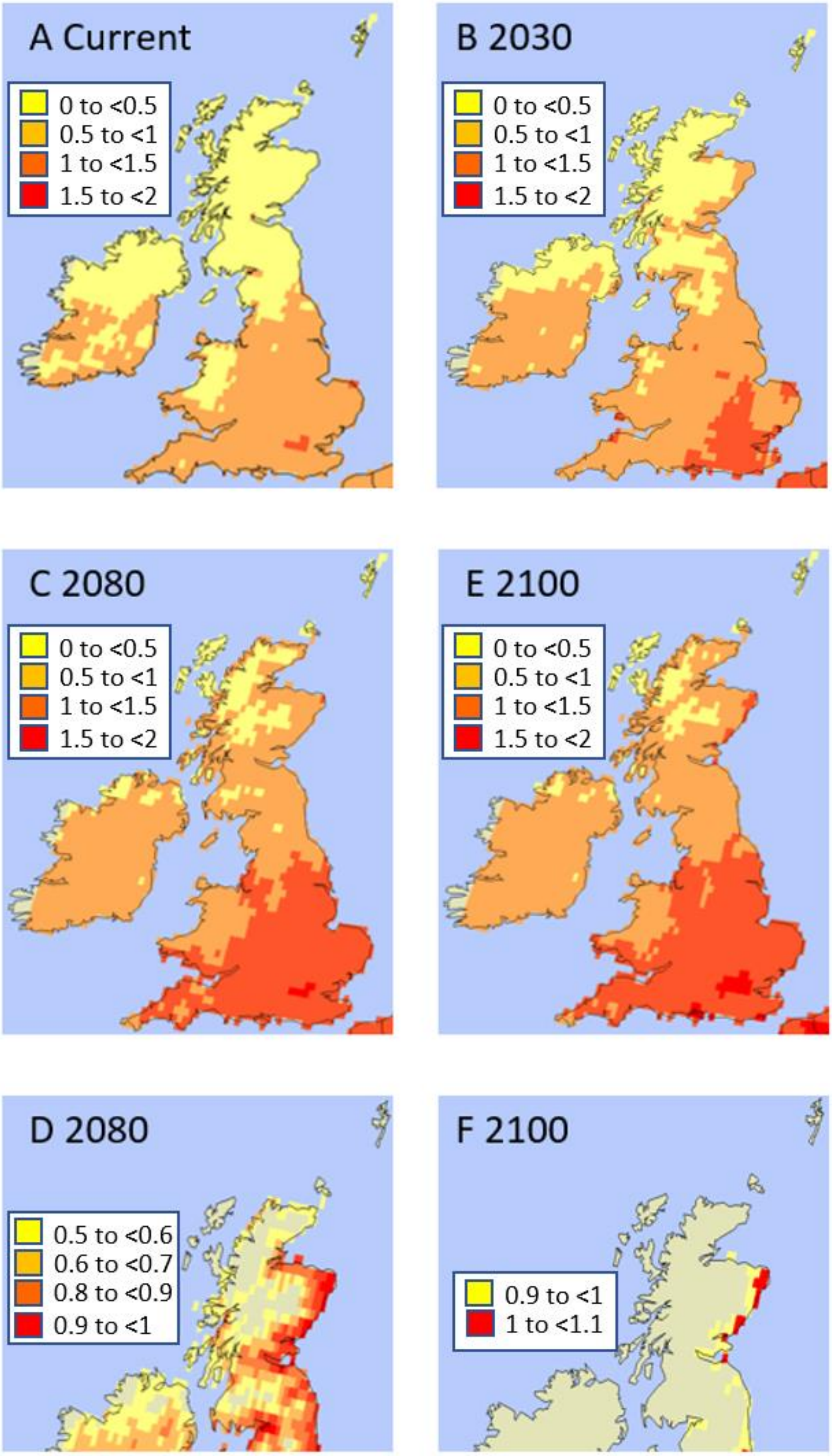

Figure 6. The potential No. of generations a year for BMSB under the current average climate (A), and projected climates 2030 (B), 2080 (C-D) and 2100 (E-F).

The actual cumulative degree-days are variable between years (Table 4) and, in some years such as 2018, there may be sufficient degree-days accumulated above the 12°C base temperature for 2 generations of BMSB to occur as in the IG6 1AL postcode area of London. This analysis was applied to areas in Scotland where BMSB could be introduced and/or establish; Glasgow Fruit Market, Edinburgh Airport, Glasgow Airport, Prestwick Airport and two soft fruit grower locations in Fife and Tayside respectively. Over the three years from 2017 to 2019, locations varied in their cumulative degree-days (Table 5). However, none approached the 595° days required for BMSB to be able to complete a generation.

Table 5*. The cumulative degree-days above a 12°C base temperature for BMSB development (595° days required to complete a generation) at five Scottish locations where establishment may be possible based on CLIMEX model output for the current average climate. Actual data for 2017, 2018 and 2019.*

	Cumulative degree-days above 12°C and potential No. of BMSB generations in parentheses		
Location	2017	2018	2019
Glasgow fruit market	297 (0.50)	397 (0.67)	308 (0.52)
Edinburgh Airport	358 (0.60)	398 (0.67)	356 (0.60)
Glasgow airport	270 (0.45)	375 (0.63)	290 (0.49)
Prestwick airport	317 (0.53)	355 (0.60)	261 (0.44)
Fife soft fruit grower (Cupar)	308 (0.52)	308 (0.52)	286 (0.48)
Tayside soft fruit grower (Arbroath)	286 (0.48)	250 (0.42)	222 (0.37)

Table 6 summarises the projected changes in the potential No. of BMSB generations from 2019 (actual data) through to projected data for 2030, 2080 and 2100 at the specific locations in Scotland where BMSB may well be introduced and/or establish (as outlined in Table 5).

As we progress through the century from 2019 to 2100, the projected degree-days for BMSB development at the locations in Table 6 increase and move towards a potential for one generation of BMSB in outdoor areas. Fife and Tayside in particular, which are currently areas where the Scottish soft fruit sector is focused, are likely to support one generation of BMSB by the end of the century, particularly if the climate varies from season to season as seen in the current climate in Tables 2 and 3.

***Table 6**. The cumulative degree-days above a 12°C base temperature for BMSB development (595° days required to complete a generation) at five Scottish locations*

	Cumulative degree-days above 12°C and potential No. of BMSB generations in parentheses			
Location	2019 (actual)	2030 (projected)	2080 (projected)	2100 (projected)
Glasgow fruit market	308 (0.52)	322 (0.54)	447 (0.75)	481 (0.81)
Edinburgh Airport	356 (0.60)	335 (0.56)	482 (0.81)	530 (0.89)
Glasgow airport	290 (0.49)	303 (0.51)	422 (0.71)	455 (0.76)
Prestwick airport	261 (0.44)	310 (0.52)	391 (0.66)	427 (0.72)
Fife soft fruit grower (Cupar)	286 (0.48)	335 (0.56)	508 (0.85)	574 (0.96)
Tayside soft fruit grower (Arbroath)	222 (0.37)	347 (0.58)	550 (0.92)	606 (1.02)

5.2.3 *Indoor cropping*

Data from protected cropping obtained from data loggers and supplied by growers was used to obtain the degree-days above a base temperature of 12°C for BMSB development over the data collection period.

A polytunnel at the James Hutton Institute supplied temperature data from 2nd August to 12th November 2019. Using the 12°C base temperature for BMSB development, the degree-days accumulated for this period were 633.5° days, potentially allowing one generation of BMSB (which requires 595° days) to be completed within that timeframe.

Data from a polytunnel near Arbroath supplied temperature data from 15th August to 12th November. The degree-days for this period and location were 548° days, which is lower than the 595° degree-days required for BMSB to complete a generation.

Other data loggers sited indoors on a similar August to November timeframe did not record enough data for >595° degree-days to be recorded. However, if the data loggers had been placed into tunnels earlier than August it is more than likely that they would have recorded sufficient accumulated degree-days to allow at least one generation of BMSB to be possible.

Other data supplied by growers from their own polytunnels provided a larger data set covering a longer timeframe.

Data from a grower of blueberries within a polytunnel from 29th March 2018 to 24th October 2018 accumulated 1573.8 degree-days over that period above the BMSB base temperature for development of 12°C, which would allow 2.7 generations of BMSB to be completed if the pest was introduced into the tunnel in early April.

Long-term data from other growers support the possibility of 1-3 generations of BMSB within a polytunnel or Seaton tunnel a season, depending on when the pest is introduced into the tunnel. The earlier in the season BMSB is introduced into a tunnel, the more time and opportunity there is for development to occur and potential for BMSB to breed and complete 2 or even 3 generations.

5.3 *Summary of climate predictions*

Under the current outdoor temperatures experienced in Scotland, it is unlikely that BMSB could successfully complete a generation and become established as a pest if it remains outdoors. However, there is a possibility that BMSB could establish if it were to spend some time indoors (e.g. the winter months) as well as outdoors. Modelling using future climate projections indicate that as we approach 2080 and the end of the century, the outdoor climate in Scotland may well be able to support the development of a generation of BMSB, particularly along the northeast and east coastline of Scotland, areas where soft fruit cropping is currently focused.

In protected cropping of soft fruit in Scotland, current temperatures recorded from data loggers in polytunnels and Seaton tunnels indicate that sufficient day-degrees above the 12°C base temperature for BMSB development can be accumulated to potentially allow 2-3 generations to be completed, depending on when the pest is introduced into the tunnel.

As the English climate, particularly in the southeast, is more suited to BMSB development (with some locations already having the potential to support two generations of BMSB) (Table 4), there is a risk of invasion of BSMB into Scotland from the south, as well as introduction through imported plant material and airports.

6 Summary and Conclusion

In total, SASA now hold eight native species, one European species and BMSB (from the USA) within their voucher specimen collection and reference barcoding library, allowing them to provide more robust ID’s on suspected cases of BMSB reported by inspectors and members of the public. Two DNA extraction and amplification methods have been evaluated and a method for barcoding potentially degraded fragments of insect material trapped on the Pherocon BMSB STKY sticky traps has been developed. This method would allow the remaining body parts of the insect to be preserved for taxonomic identification and reference.

Under the current outdoor temperatures experienced in Scotland, it is unlikely that BMSB could successfully complete a generation and become established as a pest. Modelling using future climate projections indicates that as we approach 2080 and the end of the century, the outdoor climate in Scotland may well be able to support the development of a generation of BMSB, particularly along the northeast and east coastline of Scotland, areas where soft fruit cropping is currently focused.

In protected cropping of soft fruit in Scotland, temperatures recorded currently from data loggers in polytunnels and Seaton tunnels indicate that sufficient day-degrees above the 12°C base temperature for BMSB development can be accumulated to potentially allow 2-3 generations to be completed, depending on when the pest is introduced into the tunnel.

As the English climate, particularly in the southeast is more suited to BMSB development, with some locations already having the potential to support two generations of BMSB, there is a risk of invasion of BSMB into Scotland from the south, as well as introduction through imported plant material and airports.

The impact on Scotland if BMSB is introduced is difficult to predict at this time, as most regions where BMSB has been introduced are still measuring impact as the pest establishes. The insect is a potential pest of many crops and wild hosts, and impact would be across a wide range of fruit and vegetable production, as well as a potentially on the environment. We consider soft fruit production to currently be most at risk, due to the potential of the insect to successfully complete its life cycle in Scotland under a protected environment.

Whilst establishment may be tricky under current Scottish temperatures, we may need to be prepared to manage this pest once it is detected. We would expect establishment to be slower in Scotland than the rest of the UK and there is, therefore, the potential to attempt eradication rather than management or to manage individual outbreaks carefully to prevent spread.

Control/Eradication measures for BMSB may involve IPM strategies utilising trapping, biotechnical methods, biocontrol and pesticides where necessary.

- Pheromone lure traps can be used to monitor populations and to reduce populations by targeting late season congregating behaviour of adults. Pyramid traps baited with commercial lures containing BMSB aggregation pheromone and methyl decatrienoate are effective at capturing BMSB, even at low densities.
- Biotechnical methods have been explored by USDA scientists – focussing control at the edge of growing crops, removal of individuals or egg masses and bagging of the growing fruit may reduce damage to main crops.
- Asian biocontrol species (samurai wasps and microsporidial pathogens) have naturally established alongside BMSB populations in the USA and may arrive with the pest, expediting their potential use as biocontrol agents. Similar North American/European species are not considered to be good biocontrol agents but native predators such as ladybirds, beetles and lacewings may provide a level of control. If establishment is under protected conditions these may potentially form an important part of an IPM strategy (along with pheromone trapping and chemical insecticides).
- No insecticide resistance has been reported, although the choice of product suitable for use during fruit production and ripening may be limited in Scottish crops.

7 Recommendations and Future work

Monitoring for the presence of BMSB has contributed to the improved surveillance and detection of plant health threats to Scotland and has demonstrated that the needs of stakeholders are being facilitated, particularly with respect to the soft fruit industry requirement to show BMSB freedom status to exporting countries.

The modelling of the potential distribution of the pest has identified that protected cropping may provide key areas for establishment and should be the focus for future surveillance and a key focal point for future policy decisions and outbreak management. Opportunities to develop links with pest practitioners should be encouraged to aid the development of risk analysis and control strategies.

Since the completion of this project, BMSB has been intercepted several times in the south of the UK by researchers at NIAB EMR. The most recent detection was reported widely in a general press release on the 22nd August 2020 when a flying adult was caught on a pheromone trap at an RSPB nature reserve in Rainham Marshes. Glen Powell (NIAB EMR) stated that ‘this suggests that adults may be actively dispersing in search of mates and food sources.’ Several members of the public and Scottish growers have consulted members of our team requesting identification of native stinkbug specimens found in Scotland. To date, no BMSB have been identified. However, the development of a voucher specimen collection and a UK stinkbug DNA barcode database has strengthened the emergency response plan, increased the capacity for rapid identification, improved industry resilience and developed greater capacity to respond to potential outbreaks.

There is a risk that the chosen sites for monitoring have not been able to detect the pest but that it could have become established elsewhere in Scotland. Therefore, one recommendation would be to encourage growers to undertake pheromone trapping themselves, particularly if they grow crops under protected environments and their growing sites are within areas indicated by the modelling results as being potentially able to support the establishment of BMSB.

Stakeholders, inspectors and members of the public should be encouraged to report potential BMSB sightings and seek accurate species identification via teams at the James Hutton Institute, SRUC and SASA. There is currently no clear central reporting structure for BMSB, particularly for members of the public. We therefore recommend a central reporting point be set up, potentially through the PHC website to allow potential sightings to be collated and screened from members of the public. Identification could be determined through submission of a high-quality photographs or, if a specimen is submitted, by using the molecular tools developed in this project. This will increase the possibility of early detection and any specimens submitted can be added to the voucher collection and reference barcoding library at SASA. As BMSB is similar in appearance to other native shield bugs found in the UK, the site could also include a key to BMSB identification and a guide to other similar species. Novel findings and control advice should continue to be reported to stakeholders at appropriate KE events.

For further information on recommendations see the policy summary.

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