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ORIGINAL ARTICLE

Detection probability of forest pests in current inspection protocols – A case study of the bronze birch borer

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Abstract
Increasing inter-continental trade of wood chips for biofuel represents a significant risk of introducing invasive pest species that can cause biome-scale impacts on forest ecosystems. Some potentially invasive species have the capacity to cause high tree mortality on the Eurasian continent and could cause significant impacts on biodiversity and ecosystem functions. Because eradication of established species is difficult, there is a need for scientific studies that can evaluate the reliability of current import control practices to ensure lowest possible risk of establishment of potentially harmful species. We used a stochastic simulation model and sensitivity analyses to evaluate the chance of detecting harmful pests in imported wood chips by sampling according to the current use of internationally accepted standards. As an example, we focused on the North American beetle *Agrilus anxius* (bronze birch borer) that can cause 100% mortality of European and Asian birch species in North America. We simulated the process from logging in North America to sampling the wood chips upon arrival in Europe. The probability of pest detection for current sampling protocols used by port inspectors was very low (<0.00005), while a 90% chance of detection may require sampling 27 million litres of wood chips per shipload.

Keywords: *Exotic species, interception records, quarantine pest, species introduction, wood chips.*

Introduction

Reducing the risk of harmful pest invasions is a challenge for the global community. Once alien pest species become established in new habitats, they may be extremely difficult to eradicate. Some species have the potential to cause biome-scale impacts if they successfully invade new geographical ranges (Gandhi & Herms, 2010; Økland et al., 2011), and the costs of their damage and control programmes may be very high (Pimentel, 2002; Haack et al., 2010; Kovacs et al., 2010). Although there are examples of successful eradications of invasive species (Simberloff, 2009; Haack et al., 2010), the majority of control programmes fail to eradicate or stop continued range expansions of invasive species (Genovesi, 2005; Liebhold et al., 2007; Økland et al., 2010). Considering that introductions often are irreversible, there are good reasons for strategies aimed at stopping potentially invasive species before they become established. One common approach is import control based on sampling from imported consignments to verify compliance with quarantine requirements or to detect organisms for which the phytosanitary risk has not yet been determined (FAO, 2005).

The increasing trade of wood chips represents an important pathway for new harmful organisms to enter Europe. Large amounts of wood chips are shipped inter-continentally and stored outdoors at short distances from trees that could serve as hosts for invasive pest species transported in the wood chips. Europe became a net importer of wood chips in 2008 with 29.8 million m^3 of wood chips and wood pellets imported, and Canada was recorded as a principal provider of wood chips to Europe this year (UNECE–FAO, 2009). Further increases in trade of wood chips are expected to meet the European Union (EU) energy-policy targets through 2020 (UNECE–FAO, 2009). The risk of receiving potentially highly damaging invasive species is especially high for imports of wood chips from world regions with similar climates and related tree hosts, and when the resistance of the forest systems is not

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pre-adapted to attacks by the invasive species (Herms, 2002; Mota & Vieira, 2008).

Import control by sampling is also used in the case of wood chips. For example, before opening the second largest wood-pellet factory in the world, the Norwegian Food Safety Authority sampled wood chips to identify potentially harmful species in the chips (Økland, 2011). Coniferous wood chips from North America are not allowed to be imported into Norway because they could contain the high-risk pinewood nematode (PWN) *Bursaphelenchus xylophilus* (Steiner & Buhrer, 1934) Nickle, 1970 (FOR, 2000). Deciduous trees, however, may also contain harmful pests that could cause serious problems in Europe if they survive in wood chips and become established after arrival. An important question is to what extent sampling wood chips in arriving consignments is an efficient tool for detecting and stopping invasive species from becoming established.

In this study, we applied a simulation model based on empirical data for the process whereby an alien organism can survive logging in the exporting country, as well as chipping, storage and transport to the port of entry in the importing country. The model was parameterised to realistic conditions given by the above-mentioned import example from North America to Norway and focused on one of the potential candidate pests in the wood chips, that is, bronze birch borer (BBB), *Agrilus anxius* Gory, 1841 (Coleoptera: Buprestidae). This small buprestid beetle (adults are 6–12 mm long) is included in the list of forest pests in Europe (EPPO, 2010, EPPO, 2011). We used the model simulations to analyse the likelihood of detecting BBB at various levels of sampling intensity at the port of entry, and the results were compared to real samples taken from the first shipment sent to Norway. Furthermore, we performed simulated chipping experiments to evaluate the likelihood of survival at various chip sizes and the abundance of beetles in a shipload if a small rate of beetles should survive chipping. We used sensitivity analyses to identify parameters of influence, and we accounted for these factors in repeated analyses. We compared these results to the main conclusions of the first analysis to achieve a strong conclusion about the success or failure of import control by sampling to detect and stop a potentially invasive species.

Materials and methods

Preamble: the focal species and forest systems

Bronze birch borer is an endemic wood-boring buprestid beetle found throughout most of the native range of birch in North America, while it does not occur in Europe or Asia. Because this species is present in many different types of climates throughout North America (Johnson & Lyon, 1976; Katovich et al., 2005), it would probably also survive in many areas where birch grows naturally in Europe and Asia (EPPO, 2011). BBB is considered to be a secondary pest of highly stressed North American birch in North America (Santamour, 1990; Haack, 1996); however, stress does not appear to be necessary for colonisation of European and Asian birch species (Nielsen et al., 2011). European and Asian birch species grown in North America have been shown to be much more susceptible than North American birch species to BBB (Miller et al., 1991; Nielsen et al., 2011). A large block experiment conducted over 20 years with different birch species in Ohio, USA, revealed 100% mortality for European and Asian birch species, including *Betula pendula*, *Betula pubescens*, *Betula maximowicziana* and *Betula szechuanica*, all of which were heavily infested and killed by BBB (Herms, 2002; Nielsen et al., 2011). These birch species are important and widely distributed in Europe and Asia, and widespread damage or death of Eurasian birch trees would have considerable negative effects on forest ecosystems (Popov, 2003; EPPO, 2011). In northern European countries, birch constitutes a large proportion of the forest tree volume, ranging from 11% in Sweden to 28% in Latvia (Hynynen et al., 2010). In Norway, the area covered by birch forests reaches approximately 30% of the total forest cover. Birch is also a very important commercial tree species in Belarus and Russia (Popov, 2003; EPPO, 2011).

Bronze birch borer lays its eggs in bark cracks and crevices along the trunk and branches of birch (*Betula*) host trees. After hatching, larvae bore through the outer bark and make feeding galleries in the cambial region where they feed on the phloem tissue (inner bark) and adjacent xylem (wood). Almost all larvae become 4th instars by late summer or early autumn. Fully developed larvae (4th instars) bore into the outer sapwood and construct individual pupal chambers in which they overwinter. In spring, within the overwintering cell, the larvae shorten in length, cease all feeding and become what is called prepupae. It is in this cryptic habitat within the sapwood that BBB life stages such as 4th instar larvae, prepupae, pupae and callow adults would be able to survive in wood after chipping given that no additional feeding is required until after adult emergence.

Wood chips are processed through grinding or chipping, which cuts the wood into pieces and exposes large amounts of the wood surface area to drying. Actively feeding larvae are not expected to

survive in wood chips, but survival of the BBB life stages within the pupal cells is possible if they survive the initial chipping process. The life cycle and sizes of BBB life stages are similar to the emerald ash borer (EAB), *Agrilus plannipennis* Fairmaire (1888), which is an Asian species that has become established in North America and near Moscow, Russia (Haack, 2006; Baranchikov et al., 2008). Chips made from EAB-infested ash (*Fraxinus* spp.) trees have been shown to contain viable EAB prepupae, depending on the size of the resulting chips (McCullough et al., 2007). There is a risk that birch wood chips from North America could contain live BBB at any time of the year. The overwintering stages are present in the outer sapwood for many months, and they can survive until climatic conditions are favourable if consignments are not processed immediately (EPPO, 2010). Hereafter, we use the term "survival of beetles" to cover all surviving stages. When pupation is completed, adults generally chew an exit hole and emerge within a period of 6 weeks after the start of pupation and then fly to nearby birches to feed on foliage. Adults are fully capable of flight upon emergence. Adults generally live for a few weeks, and require continuous feeding on foliage, including a 7- to 10-day period of maturation feeding prior to becoming reproductively mature.

Description of import protocol and sampling

The export process usually starts as a logging operation by harvesters and stacking of logs near transport routes within the forest or near the harbour of export. Birch may be harvested alone or in combination with other tree species for producing wood chips. For example, the first shipment of chips from Canada that was sampled by the Norwegian Food Safety Authority in April 2010, contained 21,505 bone dry metric tons (BDMT) of mixed hardwood chips, in which birch chips (*Betula alleghaniensis* and *Betula papyrifera*) represented 30% of the total (NFSA, unpublished data). There is a significant potential for BBB-infested birch to be harvested given that wood chips are often produced from lower quality wood that may be more frequently infested by BBB, or when trees are included randomly during logging. It is not likely that BBB-infested wood can be avoided, because there are no management practices during logging of birch that would completely avoid BBB-infested trees. Early BBB colonisation of trees is difficult to detect, and BBB may be present in the outer sapwood at any time of the year (EPPO, 2011). In the export process, we assume mortality of BBB due to the grinding process and heat development during storage and transport.

The first shipment of wood chips from Canada to Norway was unloaded by conveyor belts to a chip pile with a large storage capacity (more than 40,000 BDMT). The storage site is in the open air and in close proximity to a birch forest ($<$50–100 m). On 20 April 2010, the Norwegian Food Safety Authority collected their chip samples (58 litres) randomly from the surface of the storage pile immediately after unloading, before any heat development in the new pile of chips. In this sample, 13 individual chips had signs that could suggest *Agrilus* infestation, such as D-shaped exit holes or larval galleries. However, upon closer examination of these chips, no live or dead *Agrilus* life stages were recovered (Økland, 2011).

Description of import sampling model

We developed a stochastic simulation model in R (2009) to estimate the probability of detecting BBB by the above-described import regime and sampling procedure. In the first step, the number of logged birch trees corresponding to the amount of birch wood chips per shipload is set as:

$$\mathrm{Bn} = \frac{\mathrm{BDMT} * \mathrm{Br} * \mathrm{cv}}{\mathrm{dw} * \mathrm{Bt}}, \tag{1}$$

where BDMT is the total amount of wood chips in dry weight, Br is the proportion of the dry weight comprising birch species, cv is the solid wood content (volume of wood before chipping divided by volume of wood chips), dw is the dry weight per volume (kilogram per litre, or ton per m^3) and Bt is the volume of one average birch tree (m^3).

Among the harvested birches, we assumed that BBB infestation levels reflect the density distribution of BBB infestation generally found throughout its native North American range. We divided the harvested trees into four infestation levels ($n = 0$, 1, 2, 3) in which the density (beetles per m^3) varies randomly between lower and upper ranges (low_n–high_n). In addition to BBB-free trees ($n = 0$), the three sets of lower and upper prepupal densities are described as lightly infested birches ($n = 1$), moderately infested birches ($n = 2$) and heavily infested birches ($n = 3$), and the estimated values of the ranges are given in Table I. The ratio of trees in each level (n) is given by

$$\mathrm{ra}_n = \mathrm{ro}_n * \mathrm{ao} + \mathrm{rn}_n * (1 - \mathrm{ao}), \tag{2}$$

where ro_n is the ratio of trees in each infestation level (n) in outbreak areas, and rn_n is the ratio of trees in each infestation level (n) in non-outbreak areas, and ao is the proportion of the total forest area experiencing BBB outbreaks. We prepared a vector of randomised infestation values within the range low_n–high_n

Table I. Model parameters with expected values and range of values.

Parameter	Expected (range)	Explanation
Distribution of BBB in forests		
*ao	0.1 (0–0.5)	Ratio of birch forest area experiencing outbreak condition
low_1–$high_1$	1–10	Range of within-tree BBB density per m^3 in lightly infested birches
low_2–$high_2$	10–200	Range of within-tree BBB density per m^3 in medium infested birches
low_3–$high_3$	200–500	Range of within-tree BBB density per m^3 in heavily infested birches
rn_0/ro_0	0.989/0	Ratio of trees with no BBB prepupae in non-outbreak/outbreak areas
rn_1/ro_1	0.01/0	Ratio of trees being lightly infested with BBB in non-outbreak/outbreak areas
rn_2/ro_2	0/0.2	Ratio of trees being moderately infested with BBB in non-outbreak/outbreak areas
rn_3/ro_3	0.001/0.8	Ratio of trees being heavily infested with BBB in non-outbreak/outbreak areas
Parameters of treatments and transport		
*chip	0.001 (0.0001–0.01)	Mean probability of BBB surviving chipping
*$surv_1$	0.4 (0.1–0.9)	Mean probability of BBB surviving chip-pile storage before export
*$surv_2$	0.4 (0.1–0.9)	Mean probability of BBB surviving ship transport
Sample parameters		
BDMT	21,505	The ship contained 21,505.83 BDMT wood chips in total
Br	0.3	The proportion of wood chips that were *Betula*
dw	0.192	Dry weight in kg per litre, or tonnes per m^3
Cv	0.4	Solid volume; volume before chipping/volume after chipping
Bt	0.3	Volume of one typical birch tree in m^3 (simplified)
Vols	58	Volume (litre) of chip sample inspected for presence/absence of BBB

*Parameters with ranges included in sensitivity analyses.

for each of the four infestation levels ($n = 0, 1, 2, 3$), where the number of elements in each vector corresponds to the number of harvested birches in each infestation level (ra_n*Bn):

$$\overline{X}_n = X_{j,n}, \text{ where } n = 0, \ldots, 3$$
$$j = 1, \ldots, \mathrm{ra_n} * \mathrm{Bn}, \mathrm{X}_{j,n} \sim \mathrm{uniform}(\mathrm{low}_n, \mathrm{high}_n) \tag{3}$$

A vector of BBB infestations per tree for all of the felled birches $\overline{\boldsymbol{logged}}$ is prepared by adding the vectors $\overline{X}_0, \ldots, \overline{X}_3$ into one vector and performing a randomisation of the order by permutation for all of the n elements in the added vector:

$$\overline{\boldsymbol{logged}} = \prod \left(\left(\overline{\mathrm{X}}_0, \ldots, \overline{\mathrm{X}}_3 \right) \right)_n, \; n = \mathrm{Bn} \tag{4}$$

To mimic the chipping process, the vector of trees with various infestation values is converted to a new vector of beetles per volume unit of wood chips, which is named $\overline{\boldsymbol{chipunits}}$. First, the number of volume units created from each tree is estimated as

$$U = \frac{\mathrm{Bt} * 1000}{L * \mathrm{cv}}, \tag{5}$$

where L is the volume of each sample (set as 10 litres, equal to the volume of each container of wood chips sampled by the Norwegian Food Safety Authority), and the number of elements in the new vector is $z = U$*Bn. Creating the new vector $\overline{\boldsymbol{chipunits}}$ of beetles per volume unit, each value j of $\overline{\boldsymbol{logged}}_j$ is distributed randomly among the elements of volume units created from each tree:

$$\overline{\boldsymbol{chipunits}}_{i,j} = X_i * Y_j,$$
$$i = 1, \ldots, z, \; j = 1, \ldots, \text{ last value of } \overline{\boldsymbol{logged}}_j,$$
$$\mathrm{X}_i \sim \text{uniform } (1, \text{ maxp}), \mathrm{Y}_j \sim \text{binomial } (1, p_j),$$
$$p_j = 0.5 * \overline{\boldsymbol{logged}}_j * (1 + \text{maxp}), \tag{6}$$

where maxp is maximum number of beetles occurring per unit of wood chips L in $\overline{\boldsymbol{chipunits}}$ (maxp is set as 10, which is equal to 1/10 of the number of chips with thickness $\geq$ 10 mm per 10 litres in the samples taken by the Norwegian Food Safety Authority).

The mortality of beetles due to chipping is mimicked by multiplying the vector elements of $\overline{\boldsymbol{chipunits}}$ with a randomised factor p_c of survival:

$$\overline{\boldsymbol{chipsurv}}_i = \overline{\boldsymbol{chipunits}}_i * p_c, \quad \text{for } i = 1, \ldots, z,$$
$$p_\mathrm{c} \sim \mathrm{binomial}(1, \text{ chip}), \tag{7}$$

where chip is the mean probability of beetles surviving the chipping process and z is the number of elements in vector $\overline{\boldsymbol{chipunits}}$. Beetle mortality due to heat development during storage before export is mimicked by multiplying the vector of $\overline{\boldsymbol{chipsurv}}$ with a randomised factor p_{s1} of survival during storage:

$$\overline{\boldsymbol{store}}_i = \overline{\boldsymbol{chipsurv}}_i * p_{s1}, \quad \text{for } i = 1, \ldots, z,$$
$$p_{s1} \sim \mathrm{binomial}(1, \mathrm{surv1}) \tag{8}$$

where surv1 is the mean probability of beetles surviving the elevated heat during storage. Beetle mortality due to heat development during ship transport is mimicked by multiplying the vector of $\overline{\boldsymbol{store}}$ with a randomised factor p_{s2} of survival:

$$\begin{aligned} \overline{\boldsymbol{ship}}_i &= \overline{\boldsymbol{store}}_i * p_{s2}, \quad \text{for } i = 1, \ldots, z \\ p_{s2} &\sim \text{binomial}(1, \text{surv2}), \end{aligned} \tag{9}$$

where surv2 is the probability of beetles surviving the heat during ship transport. Finally, samples are taken randomly among the elements of the vector ship

$$\begin{aligned} \overline{\boldsymbol{sample}}_i &= \overline{\boldsymbol{ship}}_i * p_{\text{sample}}, \ \text{for } i = 1, \ldots, z \\ p_{\text{sample}} &\sim \textit{binomial}\left(1, \frac{\text{Vols} * \text{dw}}{\text{BDMT} * \text{Br} * 1000}\right), \end{aligned} \tag{10}$$

and BBB is recorded in the wood chips from the shipload as

$$\text{detected} = 1 \text{ if } \sum_{1}^{z} \overline{\boldsymbol{sample}} > 0, \text{ else detected} = 0. \tag{11}$$

Repeating the whole simulation procedure n times, the probability p_{detect} of detecting BBB in the wood chips from the shipload is calculated as

$$p_{\text{detect}} = \frac{\sum_{1}^{n} \text{detected}_n}{n}. \tag{12}$$

Parameter estimates and ranges

The estimates of parameters and the ranges of parameter values used in sensitivity analyses (in brackets) are presented in Table I. The biological parameter values were derived from the biological literature, expert opinion and forest inventory data.

Parameters of BBB distributions in forests. Several publications from empirical studies of *Agrilus* beetles in North America were used to estimate densities of BBB per volume of infested wood and the ratios of trees being lightly, moderately or heavily infested in non-outbreak and outbreak areas, respectively (Haack & Benjamin, 1982; Timms et al., 2006; McCullough et al. 2007; McCullough et al., 2009a, b; Mercader et al., 2010). In the absence of detailed estimates for BBB, we assumed that most of the larger *Agrilus* species attack trees at similar densities. For example, BBB is among the larger species of *Agrilus* and is similar in size to EAB and twolined chestnut borer, *Agrilus bilineatus* (Weber, 1801), which we have used in the estimates:

ao: the ratio of birch forest area experiencing outbreak conditions varies widely between years. In many years, the ratio of birch forest that is under stress is probably less than 0.01. However, during periods of severe regional drought, which may last for several years, more than 0.5 of the birch stands can be under stress in a multi-state area (Katovich et al., 2005; Haack & Petrice, unpublished data). The ratio was set to 0.1 in the simulations; however, due to variability and uncertainty a wide range (0–0.5) is included in the sensitivity analysis.

low_1–high_1: 1–10 BBB/m^3 for lightly infested birches is an estimate based on field experience from North American localities of BBB and empirical studies including data for light infestations of EAB (Mercader et al., 2010).

low_2–high_2: 10–200 BBB/m^3 is an estimate from field experience with North American localities of BBB and is set intermediate between the ranges of low_1–high_1 and low_3–high_3.

low_3–high_3: 200–500 BBB/m^3 is based on numbers of exit holes per m^3 calculated from Table 3 of Haack and Benjamin (1982), including a correction factor (1/3) for multiple years of attack on the same trunk.

rn_0/ro_0: In non-outbreak areas, the ratio of trees with no BBB beetles is quite high (estimated as $\text{rn}_0 = 0.989$); while in outbreak areas most trees would probably be infested to some degree ($\text{ro}_0 = 0$).

rn_1/ro_1: In non-outbreak areas there is a small ratio of lightly infested trees (estimated as $\text{rn}_1 = 0.01$), while most birches in outbreak areas are more infested than the lowest levels ($\text{ro}_1 = 0$).

rn_2/ro_2: the ratio of moderately infested trees was estimated as 0 in non-outbreak areas, while a significant proportion of the trees in outbreak areas are moderately infested ($\text{ro}_2 = 0.2$).

rn_3/ro_3: heavily infested birches are very rare in non-outbreak areas (estimated as $\text{rn}_1 = 0.001$) and abundant in outbreak areas ($\text{ro}_3 = 0.8$).

Parameters of treatments and transport. **chip**: a survival rate of 0.001 is based on results from experimental grinding of eight EAB-infested trees, in which eight viable EAB prepupae were recovered out of an estimated number of 8700 prepupae when using a horizontal grinder with 10-cm screen (McCullough et al., 2007). The distribution of chip sizes found in the samples taken from the first shipload of chips from Canada to Norway was within the range of the expected sizes made by a 10-cm screen.

surv1: a mean survival probability of 0.4 is based on experiments on survival of EAB (McCullough et al., 2007; Goebel et al., 2010) and temperature studies of chip piles (Bergman & Nilsson, 1971;

Vadla & Wilhelmsen, 1982). In some cases, considerable heat development could occur within the chip piles during storage, and may reach lethal levels for some biological organisms (FAO, 2009). The temperatures reached inside chip piles depend on moisture content and quality of the wood chips, external temperature and size of the pile (Bergman & Nilsson, 1971; Vadla & Wilhelmsen, 1982). During heat development, higher temperatures are usually associated with the core of the chip pile, while temperatures near the periphery are much lower and seldom lethal. According to the International Standard for Phytosanitary Measures No. 15, heating wood to achieve a minimum core temperature of at least 56 °C for 30 minutes will kill most harmful organisms in wood packaging material (FAO 2009). Survival of EAB in chips exposed to heat showed that some prepupae survived 1 h of exposure to 60 °C, whereas no prepupae survived exposure to 60 °C for 8 h or more (McCullough et al., 2007). In the absence of specific studies for BBB, we assume that exposure of wood chips to temperatures of 60 °C or more will be lethal for BBB beetles. Using isotherm profiles based on temperature measurements in chip piles (Bergman & Nilsson, 1971; Vadla & Wilhelmsen, 1982), it is estimated that the temperatures remain below 60 °C in 40% of the chip pile during heat development under summer conditions.

surv2: a mean survival probability of 0.4 is based on the assumption that about 60% of the chips per shipload may increase to 60 °C or more during transport, and that temperatures of 60 °C or more are lethal to the BBB beetles in the same way as demonstrated for EAB (McCullough et al., 2007; Goebel et al., 2010).

Sample parameters. **BDMT**: according to the importer, the first shipment of chips from Canada contained 21,505 Bone Dry Metric Tons of mixed hardwood chips.

Br: the proportion of wood chips that were Betula in the shipload was 0.3 (25% Betula alleghaniensis and 5% *B. papyrifera*); based on information from the Norwegian Food Safety Authority and Biowood Norway AS.

dw: the oven dry weight 0.192 kg per litre is based on drying and weighing of a subsample of the wood chips (Forest Technology lab., Norwegian Forest and Landscape Institute, Norway).

cv: solid volume corresponds to volume before chipping divided by volume after chipping, and the solid volume estimate of 0.4 is based on values in Table 3.4 in Hohle (2005).

Bt: 0.3 m^3 is the estimated volume of one typical birch tree, based on forest inventory statistics from Canada (Townsend, 2004).

Vols: 58 litres is the volume of the wood chip sample that was inspected for presence/absence of BBB prepupae in the first shipload from Canada (Entomological lab., Norwegian Forest and Landscape Institute, Norway).

Simulation experiments of wood chipping

Chipping down to a certain size is suggested as a control measure against import of EAB. Even when the screen size is defined, a large variety of chip dimensions are produced (Roberts and Kuchera, 2006; Kopinga et al., 2010). Based on a previous experiment of chipping eight trunks (McCullough et al. 2007), wood chips produced by a 1-inch screen ($\sim$2.5 cm) are considered effective against EAB (USDA–APHIS, 2009; Kopinga et al., 2010). However, it cannot be excluded that surviving prepupae could be found if a larger volume of wood chips had been used in the experiment. To illustrate the uncertainties, we performed two simulation experiments.

In the first experiment, we analysed survival for typical chip sizes used in the pulp industry that range between thickness of 4–8 mm, length of 40–45 mm and width of 15–20 mm (EPPO, 2011). In this simulation, we placed 8000 8-mm-long BBB prepupae randomly within 8 m^3 of wood, including a buffer of 1 mm in each end of the prepupae. Even though the survival may vary with the mechanical forces of the various chipping methods, we assume that survival is some function of chip size. Using repeated simulations (100 times at each thickness value), we performed an artificial chipping experiment driving chips into sizes of length 45 mm, width 20 mm and thickness varying between 4 and 14 mm. In the end, we counted the number of prepupae that remained intact in the chips after the chipping process.

In the second experiment, we tested the probability of overlooking prepupae being present in a shipload (300 litres of chips per birch trunk $\times$ 150 000 birch trunks) by repeated sampling of a volume corresponding to eight trunks (300 litres per trunk $\times$ 8 trunks).

Statistical treatment

Each simulation of the import sampling model started with harvesting of trees and ended with recording success or failure in detecting BBB by sampling the wood chips after importation. The

simulation was repeated 300 times at each set of parameter values as a basis for calculations of probability values, standard error and 95% binomial confidence intervals. Smooth splines (R, 2009) were included in the plots to emphasise the trends in the plots.

We performed sensitivity analyses (SA) to see how different parameter values influence the results and main conclusions. Because the estimated densities of BBB in the logged trees and the beetle mortality due to chipping and heat development during storage and transport could deviate from the true values, all of these parameters were included in sensitivity analyses by their standardised rank regression coefficients (SRRC) (Saltelli et al., 2000). We accounted for the parameters with influence in the sensitivity analyses by repeating the simulations either with all of these parameters at maximum of SA test ranges, or all of the these parameters at minimum of the SA test ranges; and based on these results we evaluated whether the main conclusions would change due to changes in these parameter assumptions. All calculations and analyses were performed in the software R (2009).

Results

The chances of detecting BBB in shiploads of wood chips with the current sampling intensity appeared to be relatively small. That is, sampling about 60 litres of wood chips per shipload and using the expected parameter values given earlier, no BBB detections occurred in 20,000 simulated shipments of birch chips, which suggests a detection probability less than 0.00005 for all shiploads that include birch chips, or a detection probability less than 0.000067 in the 14,954 shipments that actually contained BBB beetles among the 20,000 simulated shipments.

Increasing the sampling intensity had a strong influence on the probability of detection (Figure 1). However, a substantial increase of the sampling volume was needed to achieve a high probability of detection. For shipments containing BBB, sampling about 12,000,000 litres of wood chips was needed to achieve a 50% chance of detection, while a 90% chance of detection required sampling 27,000,000 litres (Figure 1A). Because the presence or absence of BBB in actual trade is unknown, sampling must be performed on every shipment of chips that contain birch. When considering every shipment that contains birch chips (with or without BBB), a 50% chance of BBB detection would require sampling of about 18,000,000 litres of wood chips (Figure 1B).

Sensitivity analysis revealed that changes of the parameters resulted in variation of the model outcome (Figure 2). However, the variation did not result from one single strong source. Rather, all parameters (ao, chip, surv1 and surv2) had some influence on the model outcome, and all parameters had SRRC values of moderate strength; usually within the range of 0.2–0.5. The relative sensitivity of the parameters varied with sample size. Using a small sample size of 1000 litres (Figure 2A), the SRRC values of the parameters were close to the same level (range 0.28–0.41). The SRRC values of the parameters increased slightly (0.32–0.59) when using a sample size with a moderate probability of detection (200,000 litres of chips yielded an average detection probability of 0.51; Figure 2B). For this sample size, a relatively stronger influence was found for the area of outbreak ($SRRC_{ao} = 0.48$) and the beetle survival during the chipping process ($SRRC_{chip} = 0.59$). When testing a very large sample size (30,000,000 litres) with a high probability of detection (0.997), the influence of all parameters dropped to a low level of SRRC values with little difference between the parameters (range 0.20–0.29; Figure 2C). In addition to the variation associated with the defined parameters of the model, all steps of stochastic processes of sampling and survival in the model led to variations that were manifested as deviations from smooth trends in Figures 1 and 3, despite a large number of repetitions (300) at each sampling volume value.

Indeed, changes in the parameters had a strong influence on the probability of detection. Because all parameters used in the sensitivity analysis were potential candidates of variation in the model outcome, we tested the extreme cases where the parameters were either all high (maximum of SA test ranges) or all low (minimum of SA test ranges). With all parameters in the low range (low density, low survival etc.), the probability of detection was close to zero even with very large samples sizes (Figure 3A). Even when BBB beetles in the simulations were set to be always present just after logging in a volume of timber comparable to one shipload of chips, the average probability of live BBB being present upon arrival at the importing country was low (~ 0.0005). Thus, with very low parameter values we can expect that only a few shiploads will include BBB beetles, and the likelihood that any will be detected is extremely low even with a very large sampling effort. By contrast, if all parameters are set at the high range, the probability of detection rises sharply with sampling effort and the likelihood of detection is achievable with the largest sampling

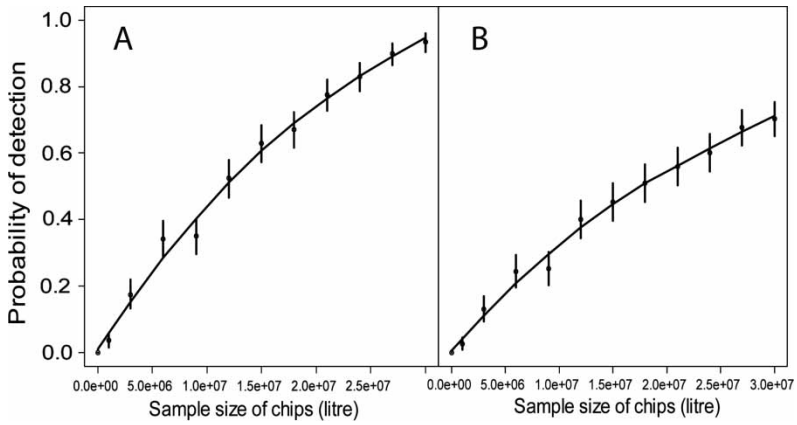

Figure 1. Probability of detecting bronze birch borer (BBB) beetles with varying sampling intensity, and assuming that sampling will occur only from shipments of chips that contain some birch chips with BBB (A) or sampling from all shipments of chips that contain birch chips with or without BBB (B). Bars represent CI = 95% and lines are smoothed trends.

volumes (Figure 3A). To explore this relationship in more detail, we repeated the analyses for parameters set at the high end of the range and used a higher resolution at the lower end of the *x*-axis of sampling volumes (Figure 3B). According to this analysis, very large sampling volumes were required for detection of BBB beetles even when we assumed high parameter values. Wood chip samples of 9000 litres yielded a detection probability of 0.51, while to attain a probability of 0.95 required sample volumes of at least 33,000 litres.

Even when birch trees are logged randomly in the landscape, the density of BBB in the resulting wood chips will vary between years, increasing during periods of environmental stress such as drought. For example, BBB outbreaks often occur for 2–3 years

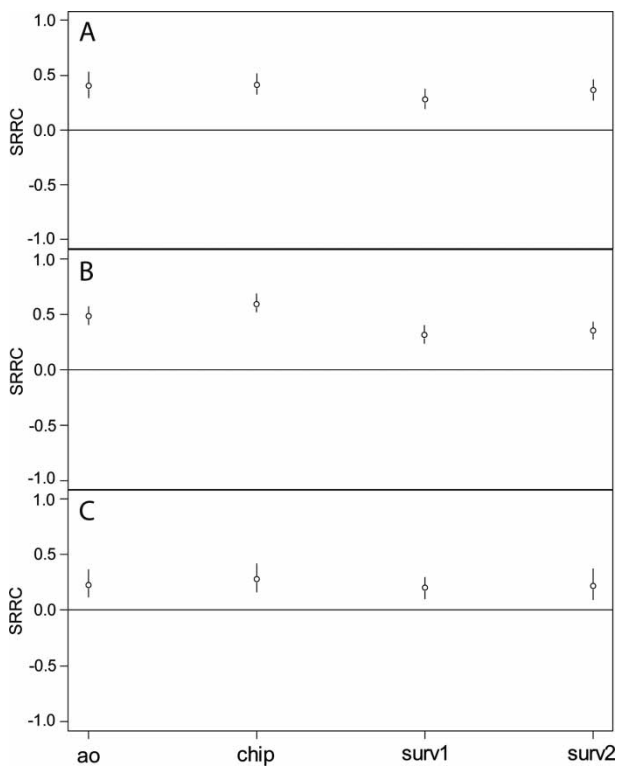

Figure 2. Sensitivity analysis of model parameters for the probability of detecting bronze birch borer (BBB) by means of standardised rank regression coefficients (SRRC) under three levels of sampling intensity: low sample size – 1000 litres (A), medium sample size – 200,000 litres (B) and very large sample size – 30,000,000 litres (C). The model parameters are the ratio of birch forest area in outbreak condition (ao), ratio of BBB that survive chipping (chip), ratio of BBB that survive chip-pile storage before export (surv1) and ratio of BBB that survive ship transport (surv2).

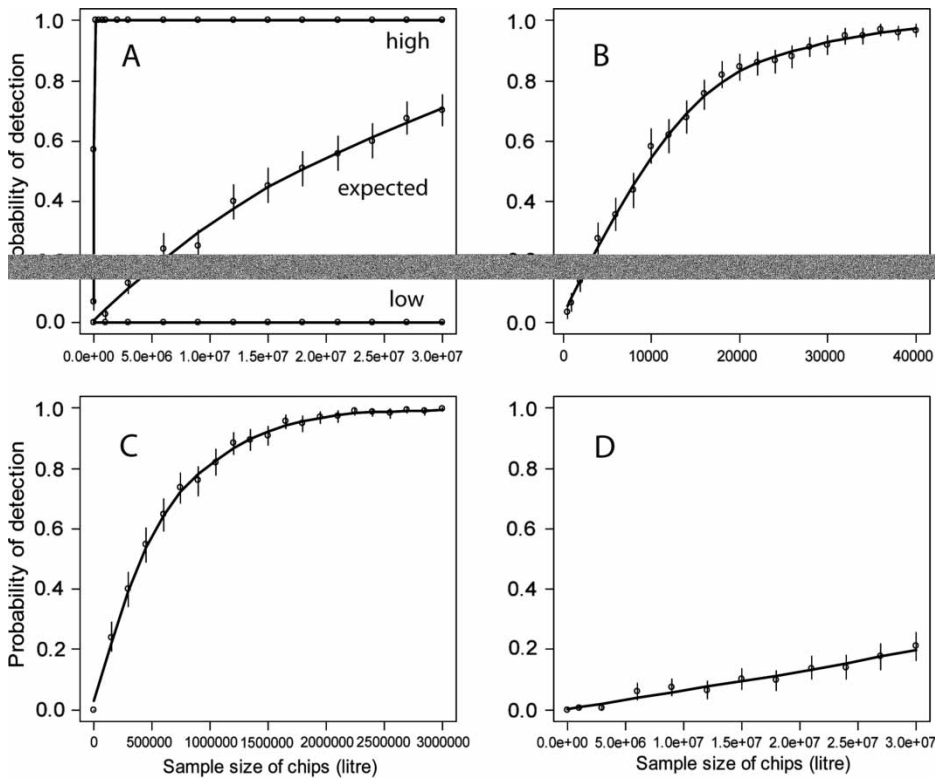

Figure 3. Mean probability of detecting bronze birch borer (BBB) as a function of sampling volume from all arriving shiploads of birch wood chips. (A) assuming that the model parameters are all high (maximum of SA test ranges), all low (minimum of SA test ranges), or all set as the expected parameter values as given in Table I. (B) assuming that the parameters are all high (maximum of SA test ranges) for sampling volumes ranging from 0 to 40,000 l. (C) assuming that half of the birch forest areas were in outbreak mode (ao = 0.5) and other parameters are as the expected parameter values in Table I. (D) assuming that no birch forested areas having BBB outbreaks are included (ao = 0) and other parameters are as the expected parameter values in Table I. Bars represent CI = 95% and lines are smoothed trends.

during and following periods of drought (Jones et al., 1993). During periods of large regional droughts, BBB outbreaks can be very widespread with perhaps more than 50% of the birch stands experiencing BBB-induced mortality. If we assume that half of the birch forested area is experiencing BBB outbreaks (ao = 0.5) and the other parameters are as the expected values given in Table I, BBB will be detected in a lower sample volume of wood chips during outbreaks than in non-outbreak years (Figure 3C). Nevertheless, very large sample sizes of wood chips are still required to detect BBB, for example, 402,270 litres of wood chips would provide a detection probability of 0.5, and 1,800,000 litres of chips would provide a detection probability of 0.95.

Logging of birch trees could deviate from random in a way that avoids areas of BBB outbreaks. Nevertheless, there would still be a chance that BBB-infested birch trees could be harvested because it is difficult to determine whether BBB is present in a tree, especially during the first year of infestation when there are few signs of infestation such as adult exit holes. Assuming that no birch are cut from forests experiencing BBB outbreaks (ao = 0) and that the other parameters are as listed in Table I, then the probability of detection increases only slowly as a function of sample size (Figure 3D). In this case, an average 20.8% of the shiploads would contain BBB, and the probability of detection would be low even with a very large sampling effort.

In the simulation experiments of wood chipping, there were complete prepupae for chip thicknesses as low as 7 mm. Chips without survivors required thicknesses of 6 mm or less (Figure 4A). Figure 4B shows the results if we assume that the true survival rate in chips produced by a 1-inch screen is within the range of 1 prepupa per 10–100 trunks. In the lower end of the *x*-axis (1 pupa per 10 trunks), the probability of not detecting living prepupae is 0.9 in a sample volume corresponding to eight trunks (left *y*-axis), while the total number of prepupae in the shipload would be about 1500 (right *y*-axis). If the survival rate should be 1 prepupa per 100 trunks, the probability of BBB not being detected is about 0.4 in a sample volume corresponding to eight trunks, while the whole shipload would contain about 15,000 living prepupae.

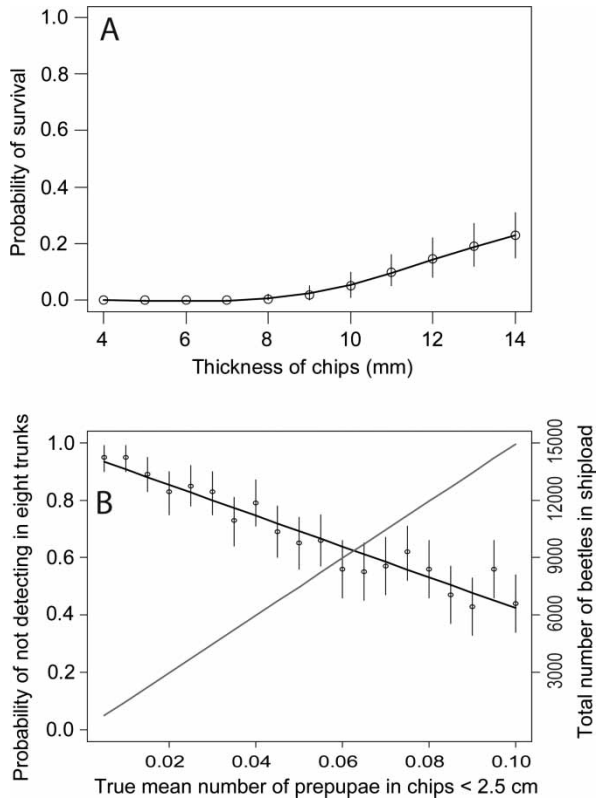

Figure 4. (A) Probability of beetle survival at various chip thicknesses in simulated chipping experiment (see Materials and methods section for details). (B) Probability of not detecting live beetles in a sample volume of chips corresponding to eight tree trunks (8×300 litres) out of a shipload containing wood chips from 150 000 trunks (left axis and falling graph) and the total number of live beetles in the same shipload (right axis and increasing graph) at various rates of surviving beetles per volume tree trunk.

Discussion

These results indicate that import control by sampling from large consignments does not give efficient protection against invasive pest species that represent a serious threat to biodiversity and ecosystem functions if they become established. The probability of detecting BBB in the sampling volume (60 litres) used by the inspection authorities was close to zero. Even if the sampling volume per shipload is increased significantly, our simulations indicate that BBB would most often be undetected. To achieve a high probability of detection in the simulations required sampling several million litres of wood chips per shipload, which is not a realistic option. In addition, BBB is only one candidate pest that could be associated with wood chips and it is clear that the limited resources for import control should be divided among all species of quarantine concern and all biological taxa that could potentially be imported by wood chips.

Although the model results cannot be verified against data from actual large-volume sampling of wood chips, the sensitivity analysis used to test and vary the model parameters suggests that the main conclusion is robust, that is, that the sampling volumes required to reliably detect BBB presence are unrealistically high. Initially, the parameters were based on empirical data as well as various estimates for BBB survival in chips during the entire process from initial logging to final sampling. A range of factors were taken into account that could influence the occurrence of BBB in the shiploads of chips. For example, occurrence of BBB can vary with the degree of environmental stress experienced by the forests prior to logging and the strategies employed to avoid or include birch stands where BBB is currently at outbreak levels. Even when accounting for drought, outbreaks years, and all other factors that would favour higher BBB densities in the imported wood chips, the sampling volume required to detect BBB was still very high. By contrast, when birch stands where BBB was at outbreak levels were excluded and all other parameters were set to produce low densities of BBB in the wood chips, achieving a high probability of detection required even higher sampling volumes than for the initial parameter estimates.

Thus, sampling large consignments of wood chips can be rejected as a useful approach to detect and thereby try to prevent establishment of BBB. Furthermore, regulation of a potentially invasive

pest species cannot depend on sampling in cases where the probability of detection is very low. For such species, indirect indices of arrivals must be utilised to regulate a quarantine pest despite the absence of positive interception records (FAO, 2004).

Active avoidance of BBB outbreak areas and infested trees during logging could lower the occurrence of BBB; however, it seems impossible to avoid all infested trees given the small size of the insect and its widespread geographic range in North America. As a consequence, other strategies should be considered to reduce the risk of introducing BBB via imported wood chips.

It is important that any regulation or measure used to prevent BBB establishment should be highly effective given that the overall probability of BBB establishment in Europe is considered very high (EPPO, 2011). If several individual BBB survived and emerged from wood chips in many parts of Europe, they would often find birch trees nearby in a favourable climate for development (EPPO, 2011). The successful establishment and rapid expansion of EAB in North America suggest that BBB establishment and spread would not be significantly slowed by Allee effects (i.e. reduced growth rate and survival at low population density; Taylor & Hastings, 2005) in Europe. Moreover, if BBB were to become established in Europe, detection of BBB in Europe will probably always lag behind the actual spread of BBB given that (1) initial attacks are often along the upper trunk and canopy branches, (2) signs and symptoms of BBB infestation do not usually become evident for 1–2 years after initial attack and (3) no effective pheromones or pre-emptive trapping system are available for BBB (EPPO, 2011). In its current area of distribution in North America, BBB occupies a wide range of ecological and climatic conditions that are also present in many parts of Europe, including Scandinavia. Furthermore, if BBB were to become established in Europe, it would be very difficult to eradicate given that control by natural enemies or any current treatments would probably be inadequate (EPPO, 2011). Aggressive eradication programmes against EAB have not been successful in Canada or the USA (GAO, 2006).

For European and Asian birch species grown in North America, mortality due to BBB has been as high as 100% (Anderson, 1944; Ball & Simmons, 1980; Herms, 2002; Nielsen et al., 2011). These tree species are widely distributed and important forest species in Eurasia, especially in northern Europe and Asia (Popov, 2003; EPPO, 2010). As the most common broadleaved tree species in northern Europe, birch is very important in sustaining local biodiversity given the large number of species that feed on or in association with birch, including mycorrhizal fungi, herbivores, wood-decay fungi and saproxylic insects (Hynynen et al., 2010). Potentially, the impact of BBB could be dramatic on both ecosystem processes and forest composition in many forest types throughout Eurasia. Except for the outbreaks of two geometrid moth species in the sub-arctic birch forests of Eurasia (Hagen et al., 2010), there are no other major pests on birches in the Boreal region of northern Europe that result in large-scale outbreaks comparable to 100% mortality (Miller et al., 1991; Herms, 2002; Nielsen et al., 2011). Widespread mortality of birch would also affect patterns of carbon sequestration. For other tree species, it has been calculated that changing large areas from living to dying forest can have a significant impact on the carbon budget; and in extreme cases it can change the forests from a net sink to a net source of carbon (Kurz et al., 2008).

Bronze birch borer is only one example of a pest species that could be imported in wood chips. Several potentially harmful species of insects, nematodes, fungi and bacteria could be introduced by importation of wood chips for energy production (SANCO, 2008; Kopinga et al., 2010). Some of the control measures discussed for BBB or EAB (e.g. smaller chip size), may be uncertain and would not be sufficient for nematodes, bacteria, fungi, virus and viroids given that chipping does not eliminate the pathogen (Kopinga et al., 2010). Also control measures based on the life-history traits of the insects are probably not lethal to these pathogens, such as storing the chips for up to two years in the country of origin prior to export. Consignments of wood chips would likely be a mixture of deciduous tree species in which several candidate pests could be present. Sampling from large consignments of wood chips would be an insufficient method to verify compliance with quarantine requirements (FAO, 2005). Moreover, our experience has shown that the tree species composition is not always correctly specified. For example, in the 2010 sample of wood chips from Canada that were reported as wood chips of birch and maple, microscopic analyses of a small subsample (135 wood chips) revealed that 0.74% of the wood chips were coniferous wood, which is prohibited by Norwegian import regulations due to the risk of PWN. Furthermore, 3.7% of the wood chips were probably ash (*Fraxinus*), which is the host genus of EAB (Økland, 2011). It is probably difficult to avoid including wood of other tree species during large-scale logging operations. Import of ash represents a significant risk, considering the large damage potential of EAB (Kovacs et al., 2010). While samples from consignments should be representative (FAO, 2005), microscopic identification of tree taxa is very resource demanding and probably impossible to

consider for handling large consignments of wood chips.

There are uncertainties about what measures and treatments would be effective against the whole range of possible insects and pathogens that could be imported with wood chips, except for avoiding known areas of distribution for high-risk pests. Requirements for more aggressive treatment measures could be introduced, such as heat treatment, fumigation, or transporting and storing chips in closed containers, although these measures may be costly and, in some cases, may have environmental side effects. More research is needed on these issues. However, from the present results it can be concluded that import control based on samples from large consignments of imported wood chips is not a reliable method to detect organisms of phytosanitary risk or to verify compliance with quarantine requirements (FAO, 2005).

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