Import risk analysis: honey bee (Apis mellifera) genetic material

REVIEW OF SUBMISSIONS

Biosecurity Authority
Ministry of Agriculture and Forestry
Wellington
New Zealand

11 August 2003

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Animal Biosecurity
Biosecurity Authority

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Approved for general release

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Biosecurity Authority

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EXECUTIVE SUMMARY

In association with several external consultants, MAF carried out an analysis of the risks associated with the importation of honey bee (*Apis mellifera*) genetic material: queens, queen cells, eggs and semen.

The major reason for carrying out this analysis was to investigate whether acceptable import conditions could be developed under which genetic material for varroa tolerance could be introduced into New Zealand. Although MAF did develop an earlier import health standard for bee semen in 1998, practical difficulties with its implementation meant that it was not able to be used.

The risk analysis concluded that the uncertainty surrounding deformed wing virus meant that semen was the only form of genetic material that could practically comply with recommended conditions.

MAF received nine submissions on this risk analysis. Submissions raised concerns regarding the risks posed by deformed wing virus and the saprophytic bacterium *Paenibacillus alvei*, as well as concerns regarding the possible effects of the genetic material itself.

In this review of submissions MAF concludes that the recommendations of the risk analysis with regard to bee semen are valid, and that an import health standard for semen can be developed. If areas of uncertainty surrounding deformed wing virus is resolved, it may be possible to develop an import health standard for other forms of bee genetic material in the future.

INTRODUCTION

The completion of the risk analysis on genetic material of honey bees was notified in the MAF publication *Biosecurity*, issue 43, dated 1 May 2003. The completed document was released for public consultation on 11 June 2003, and submissions closed 7 weeks later on 1 August 2003. Extensions to the final closing date for submissions were made for several groups and organisations.

MAF received submissions from the following (in date order):

Name	**Organisation Represented**	**Date Received**
D Yanke		15 June, 2003
F Lindsay		25 June, 2003
L & M McKenzie	Maniototo Honey	20 July, 2003
A J Scott	New Zealand Bee Industry Group, Canterbury Branch	28 July, 2003
Dr R M Goodwin		29 July, 2003
B Lancaster		1 August, 2003
J Perry	Department of Conservation	2 August
R Bray	National Beekeepers Association Inc	3 August
T Leslie	Bee Industry Group of Federated Farmers of New Zealand Inc	5 August

This document reviews each submission in turn, focusing on technical issues of contention rather than agreement.

The full text of each submission is included in Appendix 1.

REVIEW OF SUBMISSIONS

1. D YANKE

1.1 In order to minimise any risk of introducing EFB in semen, the IHS should include a requirement to add gentamycin to the semen after collection, prior to importation.

> *MAF response:* The risk analysis concluded that no safeguards were necessary for micro-organisms in semen.

1.2 Semen should be tested for the nuclear DNA of Africanised bees.

> *MAF response:* The recommended measures do require testing for Africanised bees if the country cannot certify freedom to MAF's satisfaction.

1.3 The semen could be tested post-arrival. It should be moved under MAF seal to the invertebrate facility at the Mt. Albert HortResearch campus.

> *MAF response:* The risk analysis concluded that post-arrival testing was not necessary for bee semen.

2. F LINDSAY

2.1 If a way could be found to test for viruses, then such testing should be used on all genetic material imported.

> *MAF response:* The risk analysis concluded that the risk of viruses being in semen was negligible, which means that such testing is not justified.

2.2 The submitter is not convinced by the conclusion in 21.2.3 that EFB would be unlikely to have any effects on native insects since it is restricted to honey bees. Asked whether any research has been done.

> *MAF response:* MAF is unaware of any research done on the susceptibility of native insects to EFB, and considers that this situation is almost certainly due to native insects being confined to NZ and EFB not being present here.

2.3 The submitter considers that the potential for loss of feral hives by EFB may have a serious effect on pollination.

> *MAF response:* MAF agrees with the submitter that losses of feral hives, due to any cause, would have a serious effect on pollination.

2.4 The submitter disagrees with the recommendation in 21.3.2.3 3 c), to treat recipient nuclei hives with oxytetracycline, as we should not do anything that would conceal EFB if it were to be introduced into quarantine.

MAF response: MAF agrees that a case can be made for non-use of antibiotics in post-arrival quarantine. However, since the risk posed by DWV is considered to be unacceptable for anything but semen from any countries, the recommended conditions for EFB will not be used in an IHS at this point. Any future consideration of such an IHS will take this point into consideration

2.5 An alternative safeguard for Africanised bees (for queen cells and presumably also for eggs) would be to leave the queen cells in an incubator and destroy any that emerge 24 hours before the normal end of the pupation period - i.e. on 11th day.

MAF response: MAF considers that the proposed safeguards are adequate, but if an IHS were to be developed for honey bee genetic material other that semen, this safeguard could indeed be considered among the options.

2.6 The risks of the Harbo bee, Russian bee from the USA or the Carniolan bee from Europe should be included.

MAF response: The risk analysis does consider other races of honey bees in Chapter 41 (including the Carniolan bee), and the risks are considered to be negligible.

2.7 The submission commented favourably on the Australian post-arrival quarantine facilities, and recommended that this country do something similar.

MAF response: There is no government or private sector PAQ facility in New Zealand for live bees at this point, and no plans to develop one. MAF considers that existing private facilities could theoretically be adapted to the Australian model.

3. L & M MCKENZIE, MANIOTOTO HONEY PROCESSORS LTD

3.1 The submission disagrees with box 5 on p5 i.e. if there is no control program in NZ, then the organism is not a potential hazard.

MAF response: Although it is not clear what organism is the focus of this comment, MAF assumes that the submission is referring to varroa. Since MAF is continuing with controls on varroa (aimed to prevent spread to the South Island) it is deemed to be under official control on page 91.

3.2 The submission disagrees with the conclusion in 13.1.5 i.e. that DWV is classified as a potential hazard.

MAF response: MAF agrees that the case for considering DWV as a potential hazard is not strong, particularly in light of the limited evidence that NZ is free (little surveillance has been carried out, and there are anecdotal accounts of clinical signs of this virus having been seen), and the very limited evidence that the virus would have a significant effect in view of it being only one of a number of viruses that are involved in the parasitic mite syndrome. Nevertheless, in view of the uncertainty surrounding this issue, DWV has been considered to be a potential hazard for the purposes of this risk analysis, in the knowledge that further research will have to be carried out to clarify these matters.

3.3 Regarding 13.3.2.2, it is suggested that testing a sample of eggs from 'one source' would provide adequate assurance against DWV.

> *MAF response:* MAF agrees in principle with this comment, assuming that 'one source' is meant to be one apiary or one supplier of genetic material. If an IHS for genetic material other than semen is considered in the future, this point will be considered.

3.4 Eggs and semen should be sourced from a country where DWV has not been reported.

> *MAF response:* Very few countries (including New Zealand) carry out any regular surveillance for bee viruses, which makes country freedom difficult to declare or verify.

3.5 In 21.3.2.3 3 (c), treatment of recipient hives with oxytetracycline is recommended. Why is this recommended, and when are they to be treated?

> *MAF response:* This is a so-called "incubation-based withholding period", and as such it is intended that recipient hives be treated as soon as the queens or eggs are placed, and that they be examined for EFB after 4 days. MAF can see that a reasonable case can be made for not using any antibiotic cover in post-arrival quarantine, in order to maximise the sensitivity of detection of EFB. However, the risk posed by DWV is considered to be unacceptable for anything but semen from any countries at this point, and the recommended conditions for EFB will not be used in an IHS for semen.

4. A J SCOTT, NEW ZEALAND BEE INDUSTRY GROUP, CANTERBURY

4.1 In the 3rd bullet point, the submission expresses concern regarding the conclusion that semen is the only form of genetic material that can comply with the recommended measures.

> *MAF response:* If pertinent new information becomes available on deformed wing virus (DWV) in the future, particularly regarding its distribution and appropriate testing regimes for genetic material other than semen, the above-mentioned conclusion may no longer apply. Any future import health standard will be based on the best scientific information available at the time it is issued.

4.2 In the 4th bullet point, the submission makes the point that the uncertainty surrounding a number of aspects of DWV (including which countries have reported its presence, and its likely effects) is predominantly the result of a lack of surveillance or research, which itself is a reflection of its relatively minor role (as one of a number of other viruses) in the parasitic mite syndrome.

> *MAF response:* As discussed in the risk analysis, notwithstanding the uncertainty surrounding the distribution and effects of DWV given the existence of a number of viruses that have a role in the parasitic mite syndrome in this country already, MAF considers the risk posed by DWV in semen to be negligible.

4.3 In the 5th bullet point the submission is critical of the lack of quantification of the risks in different commodities, and makes vague suggestions of hidden agendas.

> *MAF response:* This is a qualitative risk analysis, that is, a reasoned and logical consideration of the currently available epidemiological information, in the full knowledge that the more detailed information that would be required for a quantitative analysis is currently not available. As far as the relative risks of various commodities is concerned, however, the risk analysis explains as transparently as possible the reasoning behind the conclusions that are reached. The chapter on DWV is the critical chapter as far as commodities other than semen is concerned. Since the risk analysis concludes that the DWV risk in queens, queen cells and eggs is unacceptable, and since there are no satisfactory safeguards for managing those risks, it is concluded that those commodities cannot be considered to be safe. An important assumption behind this conclusion is that the virus is distributed world-wide (apart from New Zealand). This lack of confidence in the ability of any country to certify country freedom, together with the rather tenuous assumption that New Zealand is in fact free, is central to this conclusion, and if either of these assumptions were to change in the future, the conclusions regarding the risks of different commodities would also change.

4.4 In the final bullet point, the submission notes that pre-export quarantine is not considered.

> *MAF response:* This observation is correct. However, the risk analysis concludes that safeguards of any sort are not necessary for semen, and, given the current assumptions surrounding DWV, the use of quarantine (be it pre-export or post-arrival) is not considered feasible for queens, queen cells and eggs. If the above assumptions were to change in the future, then the use of quarantine for commodities other than semen becomes a possibility. At the moment, MAF is not aware of countries that have acceptable pre-export quarantine facilities for honey bees, but it is possible that these might be considered under equivalence arrangements if individual country veterinary assessments supported this.

5. DR R M GOODWIN

5.1 Regarding the chapter on deformed wing virus (DWV), Dr Goodwin disagrees with the judgement expressed in the release assessment that the likelihood of semen harbouring the virus for any significant period of time is negligible. Dr Goodwin considers that the lack of information on this issue justifies a precautionary approach, and offers his opinion that deformed wing virus "could possibly survive in semen for at least 3 weeks and possibly much longer." In stating this, Dr Goodwin recommends that MAF check for accuracy several references cited in the risk analysis. Dr Goodwin likens the case of DWV in bee semen to that of foot-and-mouth disease virus in cattle semen, and considers that a similar level of precaution was appropriate in this case.

> *MAF response:* One of the foremost international experts in honey bee virology (Dr B Ball of Rothamsted laboratory in the UK) has provided an expert opinion that the likelihood of semen posing a risk as far as honey bee viruses are concerned is negligible. Dr Ball has recently provided MAF with further clarification for the reasoning behind her judgement that the likelihood of semen containing an infectious dose of DWV a week after collection is negligible. Moreover, Dr Ball has further

advised MAF that there is no evidence of any diseases of bees being sexually transmitted, and regarding DWV there is no evidence of brood becoming infected by any route other than the varroa mite. In concluding that the likelihood of DWV being transmitted by semen is negligible, MAF has accepted Dr Ball's expert view.

MAF considers that the analogy with FMD virus in semen is inappropriate. Foremost, the reason that there is so much caution exercised internationally concerning the risk of FMD in various commodities is that FMD is recognised as having catastrophic effects on international trade in animals and animal products for a number of species. As a result of that recognition, a considerable body of research has accumulated over many years characterising the risk involved with trade in different commodities and the status of different countries with respect to this virus. Neither of these framework conditions are the case for DWV. Thus, the international interest in DWV virus is relatively low, with the effect that very little work has been done on its epidemiology and pathogenesis or on surveillance at a country level.

5.2 Dr Goodwin disagrees with the reasoning in the risk analysis that the saprophyte *P. alvei* would not produce significant complications in the diagnosis of American foul brood in the absence of European foul brood. He argues that since *P. alvei* can grow on a wide range of materials, there is no obvious reason why it should not be able to grow on any dead larvae (whatever their cause of death) and produce AFB-like signs. Thus he considers that it should have been considered a potential hazard and that in view of the likelihood that it will interfere with AFB diagnosis, the risk assessment should conclude that it is a hazard for which safeguards are justified.

MAF response: As *P alvei* is not a pathogen that produces a characteristic clinical syndrome, it does not fit neatly into the framework for animal risk analysis developed by the OIE. Indeed, the organism is a saprophyte, has been isolated from a wide range of materials, and in view of its ubiquitous nature, MAF considers it highly unlikely that the organism is not already present in New Zealand. In a similar vein, if this organism were to be considered a potential hazard in relation to honey bee genetic material, for consistency it would have to be considered a potential hazard in association with many other imported materials. Further, as the risk analysis explains, it has been isolated on one occasion in this country. Although there is a limited body of evidence that *P alvei* is capable of killing honey bee larvae on its own in the laboratory, there is no evidence that this occurs naturally. Moreover, although *P. alvei* has been reported from larvae that were purportedly killed by sacbrood, Nosema and AFB, the fact remains that *P. alvei* has not been recognised as a significant issue in countries or zones where *M plutonius* is absent. Therefore, although its precise nature remains unknown, it does appear that the link between this organism and EFB exists. In addition, during external technical review of the risk analysis, some of the expert reviewers disagreed that the clinical signs of *P. alvei* secondary invasion were sufficiently similar to cause interference with the diagnosis of AFB.

As a result of these considerations, during the hazard identification step of the risk analysis process, *P alvei* was eliminated as a potential hazard on the grounds that it is not a primary pathogen and there is insufficient evidence to indicate that *P alvei* would significantly complicate the diagnosis of AFB under the Pest Management Strategy in New Zealand.

MAF remains convinced that this conclusion is correct, and MAF does not consider that the level of uncertainty surrounding the likelihood that this organism will significantly interfere with the AFB PMS is sufficiently high to warrant imposing risk management measures on imports of genetic material for this organism.

5.3 Regarding the risk posed to bees by the commodity itself, Dr Goodwin suggests that the issues are more complex than indicated in the risk assessment.

MAF response: In Chapter 41, the consequences of the introduction of Carniolan and Caucasian genetic material are considered within the limitations of the available information.

6. B LANCASTER

6.1 The submission raises a number of issues about the desirability of introducing the genetic material itself.

MAF response: Although this issue is mostly outside the scope of the risk analysis, similar comments were made in several other submissions, and MAF has taken this opportunity to clarify a number of points regarding imports of organisms.

If the proposed organism is exotic to New Zealand, then it is a *new organism*, in which case, before it can be assessed under the Biosecurity Act 1993, it must be approved for importation by the Environmental Risk Management Authority (ERMA) under the Hazardous Substances and New Organisms Act 1996 (HSNO). However, HSNO defines new organisms at the taxonomic level of *species*, which means that races of *Apis mellifera* are not considered to be *new organisms* by ERMA.

The importation of organisms that are not new organisms falls under the Biosecurity Act 1993 (BSA). Under this legislation, a biosecurity clearance may be given if the goods are not considered to be risk goods, or, in the case of risk goods, if the goods comply with the requirements of an import health standard (IHS). Under section 22 of the BSA, in developing an IHS for a particular risk good, MAF is obliged to assess the likelihood of organisms being introduced with the imported goods, including the likelihood of any such organisms causing unwanted harm to the environment, the people and the economy of New Zealand. Thus, anybody has the right to import organisms into New Zealand as long as the organisms are not new organisms, and as long as the organisms conform to the standards in the relevant IHS.

The goods that are considered for importation in this case are themselves an organism, i.e. *Apis mellifera* genetic material. Since this species is not a new organism, there is no requirement to obtain an approval under HSNO, but MAF can only give a biosecurity clearance if the genetic material conforms to an IHS that is developed under Section 22 of the BSA — parts (a) and (b) of Section 22 cover the disease agents that may be introduced with the bee genetic material, and these considerations comprise the bulk of the risk analysis.

MAF recognises that, in the case of honey bees, several issues arise that are not encountered with importation of genetic material of other terrestrial animals, in

particular the several races of *Apis mellifera*. Further, the uncontrolled breeding exhibited by queens means that containment of genetic material by management practices such as could be done for most farmed mammals is not practically possible for honey bees, such that there is a recognised risk that genetic material may be 'contaminated' by genes of races other than that intended. Under Section 22 (d), MAF may consider other matters that the Chief Technical Officer considers relevant, and this was one such matter.

Thus, recognising that the genetic contamination issues were of concern for some stakeholders, MAF included Chapters 39 - 41 in the risk analysis — covering races of *Apis mellifera* other than those already present in New Zealand. For two of these races, Africanised honey bee and Cape honey bee, MAF had already carried out a consideration of the effects on New Zealand's natural and physical resources or human health, with the effect that these two subspecies are considered as *unwanted organisms* under the BSA, and their importation is not permitted. However, MAF has long been aware of some difference of opinion amongst beekeepers as to the likelihood of negative effects if the importation of *Apis mellifera* races other than *A. m. scutellata* and *A. m. capensis* were allowed, so Chapter 41 was also considered necessary to examine the likely consequences of introduction of the Carniolan bee and the Caucasian bee, in order to again consider whether there was a case to be made for also considering these races to be unwanted organisms. MAF considers that these issues have been adequately and transparently covered in the risk analysis, and that any negative consequences of introduction of the Carniolan bee and the Caucasian bee are not likely to be of a sufficient magnitude to warrant classifying these races as unwanted organisms.

Finally, it should not be forgotten (as stated in the risk analysis) that both the Carniolan and the Caucasian race of honey bees have been introduced into New Zealand at various times in the past.

6.2 The submission also raises the issue of compensation for losses experienced by beekeepers as a result of importation of 'inferior stock'.

MAF response: In the light of the above situation, compensation issues for claims of losses do not arise. Further, MAF considers that any beekeeper who has in fact been able to develop and maintain a particular line of bees with open mated queens would continue to be successful by continuing with current management practices.

7. J PERRY, DEPARTMENT OF CONSERVATION

7.1 The Department of Conservation (DOC) includes comments on both the IHS and what the department refers to as the "Draft Risk Analysis".

> *MAF response:* The risk analysis is not a draft. It has been submitted to internal and external technical review and to interdepartmental consultation prior to its finalisation in June 2003. Only those of DOC's comments that relate to the risk analysis are considered in this document — those on the IHS will be considered in the separate consultation process for the IHS.

7.2 DOC expresses concerns that the risks of bee viruses to native bees have not been adequately discussed, and that while the likelihood of infection of solitary bees is low, the risk analysis should consider this. DOC recognises that there is very little information in relation to the likely effects of disease agents on native insects, but expresses the hope that research in this area will allow future risk analyses to be carried out with better information.

> *MAF response:* MAF is not aware of any scientific information that considers the susceptibility of New Zealand native bees to the diseases of *Apis mellifera*. Nor is MAF aware of any research being undertaken or planned in this area.
>
> However, MAF considers that the likelihood of honey bee viruses having any significant effect on native bees is remote, for the following reasons:
>
> - First, the viruses are confined to *Apis* species, mostly to *Apis mellifera*, which is very remotely related to the native bees of New Zealand. There are in fact no native bees in the same family as *Apis mellifera* (Apidae). Rather, three endemic bees are in the family Halictidae, and around 30 are in the family Colletidae. [A list of New Zealand native bees is provided in Appendix 2.] An expert consulted by MAF pointed out that honey bees and NZ native bees are pretty well at opposite ends of the spectrum within the superfamily Apoidea, so their genetics would be about as far apart as one could get in that superfamily.
> - Second, the likelihood of native bees coming into physical contact with Apis mellifera is remote, perhaps apart from during foraging, so it is unlikely that infectious agents would be transmitted from Apis mellifera to native bees. Bee viruses are normally transmitted between susceptible individuals via mite parasitism or feeding of larvae, neither or which mechanisms are likely to transmit viruses from *A. mellifera* to native bees.
> - Third, since native bees are solitary insects, nesting in the ground, the likelihood of contact between individual bees is remote (presumably apart from brief contact when mating) so the likelihood of native bees passing infectious agents between one another at anything other than a very low rate would appear to be negligible.

7.3 The submission expresses a number of concerns regarding the potential effect of Africanised bees on humans working in or using the conservation estate, and on indigenous fauna. DOC is concerned that morphometric testing is not a sufficiently rigorous method of ensuring that Africanised genetic material is not introduced into New Zealand. DOC is of the view that if PCR is more sensitive, then it should be the method of choice for testing the semen donors for Africanised genes.

MAF response: MAF agrees with DOC's concerns over the impact of Africanised bees on people and the environment. The risk analysis provides for either certification of country freedom from Africanised bees or testing. The precise requirements for certification are handled separately for each country from which trade is considered, in the context of development of import health standards. This may involve taking into account evaluation of the veterinary services of the country and any issues of equivalence that arise. If a country is not able to satisfy MAF of its freedom from Africanised bees, then a testing protocol would be negotiated with that particular country to achieve the level of protection that MAF considers appropriate, using either PCR or morphometric analysis (or both) depending on the country concerned.

7.4 With regard to *P. alvei*, DOC considers that since NZ laboratories have not experienced problems of overgrowth of culture plates for AFB samples from bee hives, it is reasonable to conclude that the organism is not in this country.

MAF response: MAF agrees that this provides good evidence that the organism is not commonly present in New Zealand beehives. However, MAF also considers that since the organism has been isolated on one occasion in this country, it is not possible to conclude that it is exotic. This risk analysis is considering only one import pathway by which saprophytic bacteria may be introduced into New Zealand, and there is no evidence to show that *P. alvei* is not present in this country in ecological niches other than bees. Since no active surveillance has been carried out for this organism, this is an example of absence of evidence rather than evidence of absence. As the risk analysis discusses, it is likely that even if the organism were present (e.g. in the soil in some parts of the country), the absence of EFB from New Zealand would account for *P. alvei* not being found in bee hives in this country.

7.5 DOC is concerned that the risk analysis did not discuss the potential for the saprophytic bacteria *P. alvei* to displace indigenous saprophytic bacteria.

MAF response: MAF is not aware of the existence of any list of indigenous saprophytic bacteria, or of any scientific information that discusses the impact on existing saprophytes of introduced saprophytes. The history of movement and evolution of saprophytic bacteria around the globe, strongly suggests that in the vast majority of environments visited by man and animals there has been considerable mixing of the population of saprophytes, and that any significant change in species balance is a result of genetic or environmental change. Further, the range of saprophytic bacteria that live in any environment is currently undefinable. For example microbiologists researching the flora of the gastrointestinal tract are aware that given the limitations on current methodologies, less that 10% of the organisms that are present can be cultured and identified. Therefore it is not possible to make any reasoned or logical assessment of the impact of the hypothetical risk of undefined introduced saprophytic bacteria accompanying bee genetic material, on an undefined population of indigenous saprophytes.

8. R BRAY, NATIONAL BEEKEEPERS ASSOCIATION INC

8.1 The NBA opposes the importation of genetic material, advising that a precautionary approach be adopted (PAQ) in view of several areas of uncertainty, in particular concerns about the impact of the genetic material itself.

> *MAF response:* The legal issues surrounding this matter are discussed in relation to the submission from B Lancaster.

8.2 The NBA is concerned about bee viruses.

> *MAF response:* The NBA submission is not clear as to whether the concerns are related to viruses that have been identified in other countries but are considered exotic to New Zealand, or whether the concern relates to viruses that might be discovered somewhere in the world in the future or whether the concern is related to the lack of knowledge about the viruses that are already present in New Zealand. Nevertheless, MAF considers that the existing knowledge about bee viruses has been well covered by the risk analysis, and that the conclusions are valid. Further, the specific issue of deformed wing virus is covered in relation to the submission from Dr Goodwin.

8.3 The NBA submission expresses concerns at the recommendation in the chapter on European foul brood to use oxytetracycline in post-arrival quarantine.

> *MAF response:* MAF has noted these concerns, and will take them into consideration in the future if an import health standard for genetic materials other than semen is developed. However, the risk analysis concludes that given the concerns over deformed wing virus, semen is the only form of the commodity that can currently be considered for importation.

8.4 The NBA submission expresses a number of concerns regarding the possible negative effects of the genetic material itself, and wants to see containment (both pre-export and post-arrival) for the genetic material in order to assess its impact. The example of the Australian quarantine facility was cited.

> *MAF response:* These concerns have been addressed in response to B Lancaster's submission. MAF does not consider that a case exists for containment, and points out that the Australian quarantine facility is for disease agents, and for testing for Africanised genetic material, but is not for assessing other impacts of the genetic material itself.

8.5 The NBA appears to suggest that the use of imported genetic material to control varroa be held in reserve until the varroa incursion 'stablises'.

> *MAF response:* This matter is beyond the scope of this risk analysis, which does not consider the purpose for the proposed importation of bee genetic material. However, MAF is not aware of a consensus among beekeepers to delay the introduction of new genetic material in this way. Nor is it clear how stability would be defined.

8.6 Of particular concern to the NBA is that the introduction of genes for varroa tolerance will lead to a higher level of varroa tolerance in feral colonies, such that feral colonies will present a greater threat of varroa to managed colonies.

> *MAF response:* This matter is beyond the scope of this risk analysis, which does not consider the purpose for the proposed importation of bee genetic material. However, MAF considers that, in the presence of varroa, most feral colonies form as a result of swarming from managed colonies, which would mean that feral and managed colonies would usually have similar characteristics. Colonies with a higher degree of varroa tolerence are unlikely to provide "an uncontrolled varroa breeding opportunity".

9. T LESLIE, BEE INDUSTRY GROUP OF FEDERATED FARMERS OF NEW ZEALAND INC

9.1 The New Zealand Bee Industry Group (NZBIG) considers that the conclusion that genetic material other than semen cannot be safely imported due to the risk of deformed wing virus poses a severe restriction on the capacity to increase the genetic pool of bees in this country, and asks whether there is any testing regime that could be carried out in the country of origin that would satisfy MAF that the likelihood of this virus being present in the hive of origin were negligible.

> *MAF response:* Deformed wing virus (DWV) is considered to be a potential hazard in this risk analysis because New Zealand is assumed to be free of this virus. However, in view of the very limited surveillance that has been carried out in this country, many experts that MAF has consulted hold the view that it is very unlikely that New Zealand really is free, and that if there were adequate surveillance it would be simply a matter of time before it were detected. MAF is aware that further virus testing of samples from New Zealand will be carried out later this year at Rothamsted laboratory in the UK. If DWV is found in any of these samples, then MAF will consider the virus to be endemic in this country, with the effect that no safeguards will be imposed on imported products of any kind for this organism. However, if the current round of testing does not find DWV, MAF will reconsider the risk posed by this virus in other forms of genetic material. Most of the immediate interest is in importation of bee semen, but MAF recognises that as long as there is no legal way to import other commodities such as queens, then the risk of smuggling is raised. MAF does have a request to develop an import health standard for queens, and this is likely to be prioritised for attention in 2004. The conclusion in this risk analysis that the risk of DWV restricts importation to semen rests on the premise that the only way to achieve the level of biosecurity protection that is appropriate for New Zealand is to individually test each imported bee in post-arrival quarantine. MAF accepts the view of the NZBIG that it may be possible to achieve a similar level of protection by other means, such as sampling in the country of origin, and this issue will be revisited when MAF considers the matter of a queen bee import health standard.

CONCLUSION

The majority of stakeholder concerns were related to uncertainty in regard to viruses (particularly deformed wing virus), the saprophytic bacterium *P. alvei*, and the effect of the genetic material itself.

As a result of this review of submissions, MAF considers that the conclusions of the risk analysis are valid, and that an import health standard can be developed for honey bee semen.

The conclusion in the risk analysis that the risk of deformed wing virus in genetic material other than semen could be managed only by the testing of individuals (queens or eggs) in post-arrival quarantine is seen by some submitters as being unnecessarily restrictive. Suggestions were made that sampling of the hives of origin could deliver an adequate level of assurance of hive freedom that would allow queens, queen cells and eggs to be imported. Others suggested that post-arrival quarantine along the lines of that used in Australia should be adopted in this country. These matters require further analysis.

APPENDIX 1: COPIES OF SUBMISSIONS

1. D YANKE

From: David Yanke <daykel@clear.net.nz>
To: <HiniC@maf.govt.nz>
Date: 15/06/2003 22:55:00
Subject: Re: Draft IHS for Honey Bee Semen From Germany

Hi Carolyn,

Further to our conversation on Friday, I mentioned 'processing' the semen post arrival as a potentially being a useful risk management tool. I probably left you a little confused, and feeling that it is has the potential to complicate something which could remain pretty straight forward, and that it should be fired straight into the Too-Hard Basket. I too, should leave well enough alone so as not to make things harder for myself when I do finally get the chance to import some semen, and because the risk it has the most potential to manage(EFB) is a very small risk , one maybe not significant enough to manage.

But here goes any way, we never use the term 'semen processing', instead referring to it as semen pooling or semen homogenizing. It is a very valuable breeding tool. When a virgin queen naturally mates she does so on the wing at a considerable distance from her hive mating with 10 or more drones in quick succession. The semen from that or/ those mating flights is stored in her spermatheca, and are used to fertilize the millions of eggs she will lay over her lifetime of maybe several years. The breeding problems created by these multiple matings besides the obvious fact of not being able to control who she mates with when she is free flying is that the several sub-families which then make up the colonies she heads can mask traits you may be selecting for, making stock improvement more difficult. Pooled semen lets you give each queen in a test population a very diverse yet homogenous dose of semen. It means that any variation detected in evaluations is maternal in origin, and selection criteria can be applied much more effectively.

Besides being this amazing breeding tool, and after all, any importation of genetic material is all about stock improvement, semen pooling can also be an effective risk management tool. Semen is usually collected by trapping or caging drones from carefully selected colonies, then the drones are brought into a lab, the drones everted by crushing their heads(typically simple male circuitry), and other minor manipulations, and the semen collected under a stereo microscope- see the following site for a description of the syringe used-
www.ohioqueenbreeders.com/latshaw...instrument.htm
The large capacity syringes used collect semen up into 100ul cap. tubes, and these tubes with ends plugged with petroleum jelly is how the semen would be shipped. There is the potential of fecal contamination, and I guess there is the possibility of the causative

organism of EFB to be somehow in the feces of a drone. If the semen is allowed to be inseminated at this end fresh and unprocessed, there is the potential for a concentrated clump of this fecal contamination being injected into one virgin, and then coming into contact with mouth parts of a nurse queen when the Queen is being cleaned up after she is returned to the hive. There are some pretty longbows being drawn here-first that the fecal contamination that finds its' way into the semen is from a drone that is carrying the causative organism in the first place, and then that this contamination finds its' way via the mouth-parts of a nurse bee in an infective dose to the larval food of a susceptible larva. I remember back in the late 80's when we doing the risk analysis for a semen importation from W. Australia(things were so much more simple then), a stats. guy at the former DSIR put some numbers to the odds of introducing EFB, and AFB with a semen importation, and they were in the millions to one for EFB, and the 10's of millions to one for AFB.

Semen pooling would eliminate any of the niggling doubt about those long bows being pulled. The semen processing involves diluting the semen in 8 parts of Tris Buffer(a buffered physiological saline solution) to one of semen. Then thoroughly mixed with gentle shaking(this can be done in a 10ml syringe, or test-tube) then the semen is dispensed into epindorf centrifuge tubes, and the semen recovered by centrifuging. The diluent has antibiotic cover, we use gentamycin(a grunty, stable, broad spectrum antibiotic) for the antibiotic cover. In the diluted state the bugs could not avoid antibiotic exposure.

As for Africanised Honey Bees(AHB), semen in its' final homogenized would allow you to test with confidence the nuclear DNA of a semen sample for AHB.

The downside of semen pooling is that the semen is damaged to a degree, and less of it migrates to the spermatheca, and therefore it shortens the productive life of the inseminated queen. The breeding advantages it offers outweighs this downside. As well pooled semen has to be used within 24 hours of processing, as semen viability decreases rapidly after pooling. Freshly collected semen can sit a room temperature for several weeks without deteriorating much.

I can see some easy post arrival testing being added to include semen pooling. The semen could move from the airport, seals intact to the invertebrate facility at the Mt. Albert HortResearch campus. The semen could be tested and/or processed there before being released. Let me know if I can be of any further help complicating things in any way.

Looking forward to seeing the IHS,

David Yanke

2. F LINDSAY

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Wellington 6004

25 June 2003

Martin Van Ginkel
Technical Adviser, Risk Analysis

Import Risk Analysis: Honey Bee (*Apis mellifera*) Genetic Material. June 2003

The potential for importing genetic material into NZ in the near future to improve our bee stocks and to assist in varroa control has inherent dangers and we must do our best to keep exotic organisms out.

As suggested, eggs and semen are the most likely import material as they offer the least change of importing an unwanted organism. We especially do not want any more mites or the little hive beetle to enter the country. Viruses have never been a major problem to NZ beekeepers and few are aware of them but to me imported viruses present just as big a danger as they could compound the affects Varroa is having on our honey bee population. Europe and occasionally USA are reporting high winter losses of bee hives some years despite beekeepers controlling varroa mite numbers. I believe viruses must be contributing to these losses.

It is disturbing but understandable that no routine testing techniques appear to have been developed that give a high probability of determining whether material contains infected amounts of a virus. (Page 28 13.3.2.2). Perhaps Brenda Ball's research team will find an affective way of testing for viruses. If one is found, I would expect this to be put in place so that all source material is checked for viruses before this enter New Zealand.

I understand your assessment on Berkeley Bee Virus or Deformed Wing Virus and other viruses that they are unlikely to result in justified restrictions on bee exports from New Zealand (if they ever got into the country). Viruses may not be a problem where Governments' are concerned but overseas beekeepers would not want our bees if we got a new virus. There is a definite stigma associated with New Zealand queen bees in the minds of some UK beekeepers because of Kashmir Bee Virus. A similar comparison can be made for Fireblight and the way Australian orchardist's fight to keep NZ apples out of Australia. Viruses will only show themselves when a bee colony becomes stressed. This is unlikely to happen in a quarantine facility as all the bee hives there will be in top condition. Hence the chance of importing semen and eggs offers the best chance of preventing unwanted organisms, every effort must be made to see that donor stock are free from what we would term "exotic viruses".

EFB Page 52 21.2.3 European Foulbrood is unlikely to have any affects on New Zealand native insects since it is restricted to honey bees.

I have some doubts about this statement. EFB can be spread through drinking water. Ie a bee defecates in a puddle and if other bees use it they will pick up the spores. Bumble bees and native bees live in close proximately to bee hives especially in area where I keep hives. What's to stop it jumping the gap into the native bees? Has this ever been researched? The loss of feral hives due to varroa makes it more important that we help sustain our native bee and bumble bee populations for the "common good" pollination.

21. 3.2.3 - 3) c All recipient nuclei colonies should be treated with Oxytetracycline.
I disagree with this practice. We assume that we do not have EFB in NZ as it hasn't been detected during sampling, however this should not be taken for granted therefore we should not do anything that could possible hide the discovery of this or any other bacteria.

Africanised Bee page 112 39.3.2.3 For honey bee queens and eggs.
An alternative would be to leave the queen cells in an incubator and destroy any that emerged 24 hours before the normal end of the pupation period, ie on the 11th day. Queens can be successfully raised provided they are fed honey and pollen in the first hour on emerging. You may be able to use this technique to establish virgin queens are free of any exotic organisms before they are introduced into nucleus. Queen pupa can be removed from the queen cell after pupation (but not after the 10th Day), inspected and returned to the cell without any fear of damaging it.

41. Honey Bee Races other than A.M. *Scutellata*
There is a move in the beekeeping industry to import new genetic material to assist the NZ beekeepers to control varroa. This could be either a "Harbo" bee or perhaps "The Russian Bee" from the USA that have been released by the USDA.

There is also a Queen Breeder in Northland, David Yanke who would like to import Carniolan stock from Europe.

At the moment both the Russian and the Carniolan bees have an Exotic status. An assessment of the risks for both these bees should be undertaken as the beekeeping industry in the near future, may partition MAF to have eggs and semen from these bees imported into New Zealand.

Appendix II. The Australian quarantine facilities have proved very successful in importing genetic material and have identified some unwanted organisms. By following their strict guidelines for post-quarantine facilities we should be confident that exotics are kept out of the country.

Thank you for allowing me to make this submission. You have produced a very good discussion document; in fact with just a little more information and photographs, it could be considered a bee disease manual.

Yours sincerely

Frank Lindsay

3. L & M MCKENZIE, MANIOTOTO HONEY PROCESSORS LTD

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July 20, 2003

Mr. Martin Van Ginkel
Technical Advisor
Biosecurity Authority
Ministry of Agriculture & Forestry
Box 2526
Dunedin

Dear Mr. Van Ginkel

Reference: Import Risk Analysis: Honey Bee Genetic Material.

Please find attached my Submission to the Risk Analysis referred to.

I am a Beekeeper in Otago currently operating 800 beehives. I have an extensive knowledge of the situation with regard to the Varroa incursion, having been involved with the NBA reaction to the parasite since it first came to notice in 2000 in Auckland.

Although it is presumably beyond the scope of this analysis it would seem some recommendations as to how the importation of the genetic material could be achieved might have been made. The possibility of some sort of negation of the varroa problem seems to me to be of sufficient importance as to merit further investigation, rather than a flat "no."

Yours sincerely

Lin McKenzie

The Import Risk Analysis: Honey Bee Genetic Material" has been studied at length.

I have no doubts about the qualifications of those involved and have every confidence in those people. I do not intend to address each and every issue but will confine this submission to those issues I wish to question. It may be assumed that I am in agreement with anything I have not addressed.

On page 5 is a schematic chart of the Risk Analysis Process. Refer to question box 5 *"Is there a control programme in New Zealand."* I see an anomaly wherein the answer *"No"* leads to an assumption it is *"not considered a potential hazard."* The fact there is no control program in New Zealand cannot automatically lead to this assumption. Following this line of argument to a logical conclusion leads to the assumption that there is no risk, therefore there is no need to assess or manage that risk. It may be that the potential hazard has not been addressed or even recognised but that is not to say it does not exist. **What is the point of this exercise if the hazard does not exist?**

On page 26 questions about **13. Deformed Wing Virus** (DWF.) are addressed

Following the argument submitted about page 5 above I find it difficult to see how the conclusion (13.1.5) has been arrived at.

It has not been demonstrated that DFW in the absence of varroa is a great challenge to Honey Bees. The second paragraph of 13.1.4 (*Epidemiology)* seems to imply that the incidence of Honey Bee mortality in the absence of Varroa is much less than in hives infested with the mite. In the next paragraph it is conceded there is little information available on the incidence of the virus in the absence of varroa. The fourth paragraph states that DWV has not been reported in North or South America, the South Pacific, Australia or New Zealand.

In "Options available" (13.3.2.2) it is stated there are no routine tests available that do not destroy the commodity. It would seem that testing 50% of the commodity **from one source** would give acceptable assurance that the risk was so low as to be acceptable. This option I suggest could be exercised in the case of eggs clearly sourced from one queen/hive. In other words each donor queen/hive would have to be tested.

Submission In response to 13.3.2.3 (Recommended Sanitary Measures) I submit that eggs could be permitted for import in the event 50% from each donor queen/hive have returned a negative test for DWV. These eggs (and any semen imported) should be sourced from a country where DWV has not been reported.

On page 50 European Foulbrood is addressed. 21.3.2.3. Recommended sanitary measures. *3c All recipient nuclei colonies should be treated with Oxytetracycline.*

Two questions arise

- What is the objective in treating these colonies with Oxytetracycline?
- When are they to be treated? I.e. at introduction of the imported commodity, a week before introduction or at some date after introduction?

Lin McKenzie

4. A J SCOTT, NEW ZEALAND BEE INDUSTRY GROUP, CANTERBURY

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28 July 2003

Mr Martin Van Ginkel
Technical Advisor
Biosecurity Authority
Ministry of Agriculture and Forestry
PO Box 2526
WELLINGTON

Dear Sir

RE: IMPORT RISK ANALYSIS; HONEY BEE GENETIC MATERIAL

Please find attached a submission on the above analysis as prepared for, and on behalf of, the Canterbury Beekeepers section of the NZ Bee Industry Group.

The Section represents some 66 Beekeepers who collectively own and manage 36,608 bee hives in the Canterbury region. This figure includes a majority of the commercial beekeeping operations in Canterbury.

The Canterbury Section appreciates the opportunity to present this submission.

Yours faithfully

Tony Scott

A J Scott
Chairman
Canterbury Bee Industry Group

IMPORT RISK ANALYSIS; HONEY BEE GENETIC MATERIAL

SUBMISSION FROM CANTERBURY SECTION NZ BEE INDUSTRY GROUP

25/07/03

The Canterbury Beekeepers Group has given much thought and time to the consideration of the Import Risk Analysis; Honey Bee Genetic Material and is sensitive to this opportunity to make comment on it.

There is no contest offered as to those involved in the compilation, development of the analysis and reviews of the material presented.

Similarly the group offers no debate in respect of the science offered, rather it seeks certain clarification and registers concerns where appropriate.

- The Group fully concurs with and supports the major objective of the analysis as it is detailed in the Executive Summary, which is *"to find acceptable conditions under which genes for Varroa tolerance can be introduced into New Zealand"*.

  In this respect the group comments as follow: "Given the overwhelming evidence worldwide attesting to the progressive failure of the dependence on chemical intervention as the frontline means of controlling/managing Varroa bee mite, the acquisition and introduction of bee stocks with proven Varroa tolerance traits is seen to offer an acceptable non-chemical management alternative".

  As such this would conform to and secure the basic concept of a drug free industry. It would also comply with the existing NPMS for American Foul Brood disease.

  Varroa tolerant bee stocks are therefore, seen as being desirable, effective and commercially acceptable and a valuable tool in the long term management of Varroa. (Evidence from Canada and the United States certainly reinforces the above contention and is now the preferred long term option in both countries).

- The Group is constrained to make comment on the repeated referral to *"the recommended condition"* as they appear in the body of the analysis.

  It is acknowledged that the Authors and subsequent reviewers have complied with the Terms of the Biosecurity Act as laid down in 1993. However, the question must be asked as to the relevance of these *"recommended conditions"* in the face of changing circumstances and whether or not they now encompass the flexibility to truly reflect the realities of the day.

  In this respect the Beekeeping Industry in New Zealand has, as a consequence of the advent of Varroa, manifestly changed in all respects. It is no longer "technically drug free", a significant marketing advantage which may be lost, its ability to maintain optimum levels of production of honey and other hive products compromised, not to mention its capacity to maintain its role in the pollination of horticultural and agricultural crops and clover based pastures.

As such these unarguable consequences of Varroa post date the 1993 Act which, understandably does not take account of other pertinent industrial considerations, a fact which again beggars the question of how realistic has the approach taken been in the preparation of this analysis, given the fact that it contains far reaching recommendations which, might well be seen to extend beyond what might reasonably be expected in a paper of this nature.

Whilst section 2.2 of the analysis spells out the methodology to be applied, there is no evidence in the paper as to the *"Terms of Reference"* issued to the compilation teams. This is a pity and influences the opportunity to test/comment of the objectives adopted.

Accordingly, the Group is of the opinion that given the present situation the focus of the recommended conditions is narrow and focus and debate should be directed to not only identification of constrains on importation but consider also means of mitigating same.

- Concern is expressed over the manner in which Deformed Wing Virus (DWV) is addressed in the Executive Summary (see para. 4, page 1). This refers to the uncertainty surrounding SWV. It also refers to European Foul Brood disease and then arrives at an unarguable definitive conclusion – *"the only form of genetic material that can practically comply with recommended conditions is semen".*

  A number of issues arise as a consequence:

  Such a statement does, by implication, virtually eliminates any subsequent opportunity to investigate and test any programme wherein genetic material which is not indigenous in origin or, is other than semen. This is viewed as being too restrictive and severely compromises future research opportunities. The blanket nature of this statement needs to be considered again. Its effects go far beyond reality.

- Given the relative paucity of substantive debate on DWV, it became difficult to constructively evaluate issues such as:
  a. The effects of DWV on managed bee colonies in the absence of Varroa (see section 13.1.4, para. 2).
  b. In the same section, i.e. 13.1.4, para. 3, notes that there is little information available on the incidence of DWV in the absence of Varroa.
  c. Section 13.1.4 – para. 4 states that DWV is not reported in North and South America, the South Pacific, Australia or New Zealand.

  Given that it is accepted that testing for viruses can establish a positive, whereas there is no testing regime that can prove a negative, plus the fact that there is no surveillance programmes for bee viruses in New Zealand. It is clear that there can be no great confidence in the foregoing.

  While no one would prefer to see yet another bee pathogen introduced, it is essential that it be recognised that New Zealand already has sufficient viruses to exacerbate Parasite Mite Syndrome is association with Varroa infestation. In fact, it may well be said that Varroa is responsible for this new found interest in bee virus, hitherto not seen as being an important pathogen on honey bees.

Given the above, the Group would postulate that the enhanced effectiveness in the control/management of Varroa through a greater range of control mechanisms would be reflected similarly in the lessening in the overall effect of bee viruses and for that matter in the control of other bee diseases such as American Foul Brood, which is a major debilitation problem when associated with Varroa, as it is currently being experienced in parts of the North Island where Varroa is wide spread.

- Risk Estimation. It is acknowledged that every honey bee commodity has the capacity to act as a vector for disease. What the "analysis" fails to do however is quantify the risks between one commodity and another, for example to what measurable extent do queens represent a greater risk than does semen. The analysis is silent on matters of this sort. It should not be if objective conclusions are to be treated with confidence.

  Accordingly, the distinction favouring semen as opposed to eggs or queens is not qualified; it is not clear to the reader and might give rise to the impression that there is an agenda here which is not immediately forthcoming.

- Post Arrival Quarantine. It is noted that there is no reference to or debate give to a point of origin quarantine process.

  It would not seem unrealistic for commodities considered for importation be quarantined and tested – for specifically notified disease – at the point of origin. Provided the work was carried out at, or by, a qualified institution and conformed to a regime specified by the importer. Protocols could be drafted to ensure compliance in the same manner as the current Export Certificate.

  This would have the effect of mitigating the need to destroy the basic commodity.

  Alternatively, the testing of a proportion of the off spring of any one or all donor colonies, sources, coupled with area freedoms, where applicable, would give acceptable assurances that the level of risk was acceptable.

The Group is conscious of the extent of the research that has been undertaken in the preparation of the *"Risk Analysis"* and the considerable work that has gone into its development. It would be appreciated if this could be passed to those concerned.

Thank you for the opportunity to present this submission on behalf of the members of the Canterbury Section.

Signed

A J Scott
Chairman
Canterbury Bee Industry Group

D Bell
Committee
Canterbury Bee Industry Group

5. DR R M GOODWIN

Submission on

Import Risk Analysis:

Honey Bee (*Apis mellifera)* genetic material

Dr R. M. Goodwin
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Hamilton

The following comments on the Honey Bee (*Apis mellifera)* Genetic Material Import Risk Assessment also apply to the draft import standard for the importation of semen from Germany for which submissions have also been invited. Although it does seem inappropriate to be producing import health standards before the development of the risk assessment has been completed as it is pre judging the case.

13. Deformed wing virus

I agree with the risk assessment in that deformed wing virus should be classified as a potential hazard. The information coming out of England suggests that it would have a negative impact if introduced into New Zealand now that we have varroa.

The prohibition on the import of honey bee queens, queen cells and eggs would seem to be appropriate as they have an obvious potential for acting as vectors for deformed wing virus. However, allowing the import of semen from countries with deformed wing virus seems to be inappropriate.

The risk assessment states (P27 para 2) 'that it is unlikely that honey bee semen stored away from honey bees would carry infective levels of the virus for any significant length of time'. To justify this conclusion the Risk Analysis refers to *Section 2.3 Uncertainty p8.*

The justification (Section 2.3) for allowing semen is based on the comment that honey bee viruses (viruses other than deformed wing virus) do not survive outside there host for long and if they were present in semen they would quickly be inactivated.

The decision to make assumptions on the likely survival of deformed wing virus based on information on the survival of other honey bee viruses is inappropriate.

If it was appropriate to extrapolate from information on other viruses the length of time viruses could survive in semen as not been quantified. The risk assessment provides 3 references to back up the observation of the limited life of honey bee viruses outside a bee (Anderson and Gibbs 1988, Ball 1999, Baily 1976) .

Anderson and Gibbs (1988) does not provide any experimental evidence to support this claim. They do however make the same comment and refer to Baily et al 1979 and Anderson (1986) to support the statement. However, Bailey et al 1979 does not however appear to provide any information to back up this assertion. I have been unable to source a full copy of Denis Anderson's thesis and he has lost his own copy. The actual sources should be analyzed and referred to in the Risk Assessment.

As indicated in the Risk analysis Brenda Ball (Ball 1999) also suggests that bee viruses do not survive long outside a bee. Brenda does not however provide any data to support this claim or even indicate how long viruses can survive outside a bee.

Contrary to what is said in the Risk Assessment, Baily 1976 does not describe experiments where sacbrood virus underwent a rapid loss of infectivity. He did however refer to White (1917) who demonstrated experimentally that sacbrood virus was no longer infective after 21 days at room temperature in dry conditions.

How long sacbrood or deformed wing virus could survive in semen is unknown and this is the conclusion that should have been arrived at in section 2.3. Semen could be collected in Australia and used for insemination in New Zealand within 24 hours.

The Risk Assessment (Section 2.3) indicates that Bailey 1976 reports that White showed in 1913 that Sacbrood virus could be killed by prolonged exposure to 30-35^{o}c which is

presumably included to support the contention that sacbrood (and hence deformed wing virus) would not survive long outside a bee. I can find no evidence of this Baily having made this comment. He did however report that White demonstrated that sacbrood virus was inactivated when heated to 58^{o}C for 10 minutes. Neither White 1913, nor White 1917 report investigating the survival of Sacbrood at 30-35^{o}c. This comment needs to be checked for accuracy.

Using the precautionary principle it should be concluded that deformed wing virus could possibly survive in semen for at least 3 weeks and possibly much longer

No information is provided in the risk assessment on the likelihood of honey bee semen carrying deformed wing virus although virus contamination of semen has been reported for mammals. To apply the precautionary principle it should be assumed that semen could either carry deformed wing virus directly or it could become contaminated during collection.

The conclusion of the Risk Estimation (section 13.2.4) should therefore be changed to say that the risk of importing deformed wing virus with semen is non-negligible and that sanitary measures are justified

To put the issue in context we would not even consider allowing the introduction of cattle semen from foot and mouth countries without being absolutely confident the virus could not be transported in cattle semen. To show consistency in how we deal with risk we must therefore use the same criteria for honey bee viruses and semen.

Even the risk assessment indicates that a lower standard is being used in the assessment of risk for viruses than for other potential risks.

P8 last paragraph. '*However, elsewhere when such uncertainty is encountered a precautionary approach is adopted, in consideration of the available scientific evidence'*

Why a decision not to apply the precautionary principle to semen was made, has not been provided.

As we cannot be sure that introducing honey bee semen will not also introduce deformed wing virus, semen should not be permitted from countries with deformed wing virus until suitable information of the risks involved have been provided.

22. *Paenibacillus alvei*

The conclusion *'There is no evidence that it can cause complications in the diagnosis of American foulbrood in the absence of M plutonius'* is not supported by the arguments provided and doesn't follow the precautionary principle described in section 2.3 of the Risk Analysis

From the wide range of material the *P. alvei* has been isolated from it would appear to be a opportunistic invader of any suitable material as indicated in the Risk Analysis. Also as indicated in the Risk Analysis *P. alvei* can produce clinical symptoms similar to American Foulbrood Disease and therefore should be classed as a hazard.

As indicated in the Risk Analysis *P. alvei* is often found in association with larvae that have been killed by *M. plutonius.* However there is no published evidence that demonstrates that it requires the presence of *M. plutonius* to infect a larva and produce American foulbrood like disease symptoms. No hypothesis to explain why *P. alvei* might need *M. plutonius* has ever been provided in the literature.

Skrypnik (1984) and Kardokov et. al. (1975) have demonstrated that *P. alvei* can kill larvae when fed to them under laboratory conditions *P. alvei* has also been reported to increase the death rate of adult bees infected with *Nosema apis* (Grobov 1971). This is in the absence of *M. plutonius*.

In a study on the survival of *P. alvei* spores Konlikovskii and Sosnia (1994) reported that the spores adhere readily to the cuticle of larvae and multiply. *P. alvei* has also been reported to be able to multiply in larvae killed by sacbrood (*Bailey et al 1973*). Again in the absence of *M. plutonius*.

As indicated in the Risk Assessment *P. alvei* has also been found in larvae killed by American foulbrood (Alippi 1991; Alippi 1997). The risk assessment goes on to conclude that it is possible that in such cases European foulbrood may have been previously present in the colonies. However, there is no reason to suspect this.
It appears that this comment has been included in an attempt to discount the observation so that a conclusion that no sanitary measures are required can be made.

M. plutonius only infects the gut of a larva whereas *P. alvei* infects larval tissues. In a larval smear where a larvae has been infected with both organisms *P. alvei* makes up more than 99.9% of the bacteria present. The AFB like disease signs produced by *P alvei* is therefore unlikely to require the presence of *M. plutonius*

In conclusion:

1) *P. alvei* is a saprophyte with a wide range of materials that it can infect.

2) There is no evidence to indicate that *P. alvei* requires the presence of *M. plutonius* to be able to infect a dead honey bee larvae other than to provide a source of dead larvae.

3) There is no plausible hypothesis to explain why *P. alvei* might only infect dead larvae that have been killed by *M. plutonius*.

4) *P. alvei* has been reported in the literature to infect larvae infected with *P. larvae larvae*, Sacbrood, and nosema.

5) *P. alvei* has been reported to be able to multiply on the cuticle of larvae in laboratory studies.

Even without using a precautionary approach there is sufficient evidence to indicate *P. alvei* probably does not require the presence of *M. plutonius* to infect a larva and produces AFB like signs.

Even without this evidence the choice would be between two hypotheses.

A) That *P. alvei*, an opportunist saprophyte of a wide range of material, can only infect a larva and produce AFB like signs when the larva has been killed by *M. plutonic* because of some as yet unknown link between these two organisms.
B) That *P. alvei*, an opportunist saprophyte of a wide range of material will infect any dead larvae and produce AFB like signs.

It would be standard practice to accept the simpler of the two hypotheses.

The conclusion of the section should therefore be that *P. alvei* is a hazard for which sanitary measures are justified. **This same conclusion should have been made in the Bee Products risk assessment.**

41. Honeybee races other than *A. M. scutellata* and *A. M. capensis*

The risk estimation (section 41.2.4) suggests that the risks are negligible. However, the issues are more complicated than indicated in the risk assessment.

Honey bees are different from most other domesticated animals in that their mating is uncontrolled and can only be controlled through artificial insemination which is both difficult and expensive. This causes problems for many queen breeders and commercial beekeepers who are trying to maintain a docile Italian line with the presence of *A. m. mellifera* as a feral population. As mentioned in the Risk assessment the hybrids can be very aggressive. Beekeepers have overcome these problems by trying to swamp areas with Italian queens or buying in queens. For those beekeepers struggling with trying to maintain Italian bees the presence of varroa has a small silver lining. It is in the process of destroying our mostly *A. m. mellifera* as a feral population. This will probably make maintaining Italian strains significantly easier. The introduction of other honey bee strains therefore needs to be assessed in this light and consideration needs to be given to whether this will cause problems.

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6. B LANCASTER

From: "Berts Bees" <be.lancaster@xtra.co.nz>
To: <vanginkelm@maf.govt.nz>
Date: 01/08/2003 14:17:55
Subject: Import Risk Analysis; Honeybee Genetic Material

Dear Mr Van Ginkel
Please find attached my copy of my submission on importation of honeybee genetic material.
Please confirm the arrival of this e-mail----Thankyou
Regards Brian Lancaster.

Thank you for the opportunity to make a submission to the Importation of Honeybee Genetic Material.

I would like to congratulate you on what I consider to be the sound science employed.

I would like to re-iterate that the importation of anything other than semen represents a significant risk of importing several exotic diseases as well as importing a virulent mutated strain of A.F.B. that is resistant to O.T.C.

It disappoints me that part of the importation process does not include consultation with stake holders and address civil rights issues of all beekeepers. To me the document assumes that all beekeepers are in favour of importation of genetic stock, but I can assure you that this is not the case.

For the importation of bee genetics to be considered, I would urge that part of the importation process is a round of consultation with stakeholders (i.e. Beekeepers, Horticulturists etc) as to their acceptance or otherwise of bee genetics other than Italian races. This is because any release of bee genetics in this country is ultimately uncontrolled due to the mating characteristics and will eventually impact on every beekeeper and bee customer in the country.

I feel most Beekeepers would be concerned that importation of genetic stock for one desirable trait is a significant step backwards if the said stock is deficient in other important areas. (E.g. Varroa resistant stock imported to the detriment of Honey production, population build-up, disease resistance, pollination ability, aggressiveness etc. Many Beekeepers have spent Literally decades breeding a bee ideally suited to their distinctive environment.

I would also like to see the document outline any compensation issues that may arise because of the above and to identify who would be responsible for any losses due to the importation of inferior stock.

Thank you for considering this submission and I look forward to your response.

Yours sincerely

Brian Lancaster
Leaches Road
RD2 Darfield 8172
Apiarist
Canterbury

7. J PERRY, DEPARTMENT OF CONSERVATION

Draft Import Health Standard for the Importation of Carniolan Honey Bee (*Apis mellifera carnica)* Semen into New Zealand from Germany

General Comment

The Department of Conservation is concerned that this draft import health standard has been based on the Import Risk Analysis: Honey Bee (*Apis mellifera*) Genetic material that is still out for review. This import health standard should be developed after the IRA is finalised in the event that substantive changes are required in the IRA.

Specific Comments

Eligibility for Importation

The IHS for the Importation of Carniolan Honey Bee (*Apis mellifera carnica)* Semen into New Zealand from Germany indicates that the country exporting the semen must either be free of Africanised honey bee and Cape honey Bees or that drones producing the semen must come form a hive made up of bees resident in Germany for at least 12 months and have been confined to the colony of origin by a queen excluder and not permitted free flight prior to semen collection. The Department agrees that if these measures were in fact practiced, the risk that the drones would be carrying Africanized bee or Cape honey bee genetic material would be low. The IHS indicates that a Veterinary Certificate is required to verify that these requirements have been meet. What assurance can MAF give that these requirements will be adequately met in the exporting country? Does the exporting country regularly undertake nationwide sampling of domestic and feral bees to ensure there are no Africanized bees or Cape Honey bees present? If this is not being undertaken what assurance does MAF have that the certificate is correct?

The fact that a commodity has a certificate stating that African or Africanzied honey bees and Cape Honey Bees do not exist in the country does not in itself ensure that this is the case. There is always the risk that both of these unwanted genetic strains may be present but still undetected in the exporting country.

The Department suggests that the most acceptable level of assurance would be gained by requiring all genetic material entering this country to be DNA tested for both Africanised and Cape honey bee genes.

Draft Import Risk Analysis: Honey Bee (*apis mellifera*) Genetic Material

General Comments

The Department has concerns with regard to the comprehensiveness of this risk analysis. The risk that the viruses pose to native bee species has only been assessed in relation to cultivation of viruses in insects. While Ball's (1999) paper indicated that bee viruses would only replicated in bees (rather than other insects), there appeared to be no discussion in this paper as to whether or not other bee species could be infected for example what risk to these viruses post to our indigenous solitary bees. The Department notes for example *Apis* iridescent virus causes cluster in *Apis cerana* colonies, however the IRA indicates that this

virus it can be multiplied readily in the lab in *Apis mellifera.* The fact that this virus has only been reported in *Apis cerana* does not rule out the possibility that it may be undetected in other bee species. If this virus can be readily cultivate in another bee species (i.e. *Apis mellifera*), what risk is there that it may be able to multiply in our native solitary bees. A full assessment of this risk should be undertaken. While the Department recognises that likelihood of infection of solitary bees is low, the IRA should still consider this in its analysis. The risk analysis in a number of disease and virus cases relies of very little evidence to conclude that the risk is negligible. The Department agrees however that there is very little information in relation to disease etc of our native insects, and that this is an area that needs further effort by way of surveillance and identification. It is hoped that research in this area will mean that future risk analysis will be based on more complete evidence.

Comments re Africanized Bees

The Department considers that the introduction of Africanized bees would potentially have severe consequences on the Conservation estate. Feral Africanized bees would pose a serious health risk to the Departments staff and recreational users. As Africanized honey bees have a slightly shorter development times than domestic *Apis mellifera*, Africanized bees are able to produce more bees per unit time. Research indicates that Africanized bees also abandon their hives (swarm) 15-30% more than European bees and that they can travel as far as 170 km before selecting a new nesting site. This would suggest that if semen with Africanized honey bee genetic material was to enter New Zealand, the spread of this genetic material would be both rapid and extensive.

European honey bees have been in New Zealand for a substantial length of time. The introduction of this species undoubtedly resulted in increased competition for nectar resources from indigenous nectar feeding birds and insects, including our solitary bees. Displacement of native pollinators would also have occurred. The introduction of Africanized bees would significantly increase this displacement of native pollinators due to their propensity to produce numerous feral colonies in a single season. This would result in much higher feral bee numbers in some areas of the conservation estate and therefore increase pressure on nectar resources. The aggressiveness of this species could also see native birds being attacked and killed if they inadvertently disturbed a feral colony. The effect of this species of nectar feed birds and insects has not been adequately accessed in the IRA, as it appears it is assumed that the proposed mitigation measure of either morphometric testing or genetic sampling would mitigate this risk.

The Department however has concerns with regard to the use of morphometric testing as a means of ensuring that Africanized Genetic material does not enter New Zealand. A quick search of the internet indicates that there has been concerns in the United States with regard to the sensitivity of this test and the fact that it requires validation against the various strains and races that may be present in distinct biogeographical regions. The risk analysis has not discussed the level of confidence that morphometric testing gives nor does it compare this method of testing against the use of PCR. If PCR is a more rigorous and provides more certainty, than the Department suggests that a more precautionary approach would be to have all semen material verified as being free of africanized bee genetic material using PCR testing.

Comments in relation to *Paenibacillus Alvei*

The Department notes that the risk analysis indicates that it is uncertain whether or not this bacterium is present in New Zealand or not and that without active surveillance and laborartory testing it may yet be undetected in New Zealand. The Department notes that the early 1990's, a large number of South Island hives were sampled during the European Foulbrood detection scare. The Department also notes that a large number of domestic hives are sampled yearly for the American Foulbrood Pest Management Strategy programme. One could assume that given this level of surveillance that if *P. alvei* was present in New Zealand, it is likely that it would have been isolated by now. The risk analysis indicates that New Zealand laboratories have not experienced problems with this bacteria overgrowing cultures plates and this again supports the assumption that this bacteria is not present in New Zealand.

The Department notes that the risk analysis has indicated that because *P. alvei* is a saprophyte ie secondary invader and not a primary pathogen of *Apis mellifera* that is not classified as a potential hazard. The Department notes that the risk analysis has indicated that *P. alvei* has been isolated from a diverse source of sites including, human material, milk and mosquito larvae in India. This suggests this saprophytic bacteria is able to colonise a wide range of dead host material. Given that the evidence suggests that this species is in fact not present in New Zealand, the risk analysis should ensure that the effect that the introduction of this species may have is considered i.e. some discussion on the potential for this species to displace indigenous saprophytic bacteria should be included. The Department notes that the risk analysis does however indicate that no information could be obtained on the presence of the disease in New Zealand and recognises that this is a research gap that needs filling.

8. R BRAY, NATIONAL BEEKEEPERS ASSOCIATION INC

IMPORT RISK ANALYSIS:
HONEY BEE (*Apis Mellifera*) GENETIC MATERIAL

SUBMISSION FROM
THE NATIONAL BEEKEEPERS' ASSOCIATION OF NZ, INC.

1. **INTRODUCTION** The National Beekeepers' Association Inc (NBA) is an organisation representing NZ Beekeepers, membership is on a voluntary basis with regional branches throughout NZ. Until the expiration of the Commodity Levy, membership was compulsory by all commercial beekeepers. The NBA wishes to oppose the importation of genetic material.

2. **HISTORY** The Apiaries Act 1906 and subsequent amendments severely restricted the importation of bees, used beekeeping equipment and bee products. It would appear that there has been no legal importation of genetic material for at least 50 years and only minor importation (by permit only) for a further 40 years. The beekeeping industry has considered the importation of genetic material on numerous occasions during the last 30 years, following debate it was resolved that the importation of new genetic stock was undesirable. The situation in NZ has changed with the arrival of varroa and a call has been made to revisit the importation situation. The NBA wishes to take a precautionary approach to importation, as there may be advantages to beekeepers in overcoming the varroa mite with a varroa tolerant breed of bee.

3. **CONSULTATION PROCESS** Consultation to the beekeeping industry has been undertaken by MAF with the release of the Import Risk Analysis (RA) as well as guest speakers at our recent conference. There was no formal debate amongst members although opinions were solicited. It is on the basis of these views and opinions this submission is based.

4. **CONCERNS** There are concerns amongst our members.

   - The need to consider importation whilst the varroa infestation is still in its infancy and other methods of breeding a tolerant/resistant bee from the genetic material already present in NZ has not been fully progressed.

   - The identification of viruses in NZ bees appears to have been done mainly during the 1980's. A detailed study of the distribution and effect of viruses and diseases does not appear to have been progressed. Beekeepers are not well versed on viruses and diseases apart from the more prevalent diseases common in NZ. Thus it is a concern that any new organisms introduced would not be identified by beekeepers in time to prevent the spread of new viruses and diseases into the NZ environment. This poses a question of liability and responsibility in the control of unwanted organisms thus liberated.

- The use of oxytetracycline (OTC) in post-arrival quarantine poses significant issues for NZ beekeepers. The use of OTC in the treatment of bees in NZ is prohibited. An understanding of OTC is that this antibiotic suppresses visual symptoms of disease rather than eliminating the disease - as such the use of this or other such antibiotics should be withheld.

- Perhaps one of the major issues relates to the particular genetic material being imported. Whilst the RA generally identifies the risks of likely pathogens of the honey bee it does not develop any post arrival evaluation of the particular genetic make up of imported genetic material. Concern has been raised on the possibility of introduction into New Zealand of a particular strain of bee which may have the capacity to alter the NZ beekeeping industry, economy and the people of NZ in an adverse rather than beneficial way as intended.

  The lessons learned with the introduction of *Apis mellifera scutellata*, into Brazil (1950's) and the resultant 'Africanised honey bee', which has caused widespread harm, must be fully appreciated in consideration of proposed importation of genetic material into NZ.

  Organisms which have evolved over a large number of generations in isolation for a considerable period of time may act in a completely different manner when introduced into a new environment. The only way in which to be reasonably confident of mitigating adverse effects is through a process of post importation evaluation in which an elimination process is available.

  Beekeepers have expressed a view that once genetic material is liberated it will be virtually impossible to control any subsequent mating with the domestic NZ stock. Any genetic factors such as aggression, unsuitability to perform pollination or honey production, increased swarming tendencies all have the potential to adversely affect a stable NZ beekeeping environment.

- There are opinions that the importation of genetic material is perceived as a 'silver bullet' to solve a varroa problem. These opinions also suggest that the 'silver bullet' be kept in reserve in order that the varroa incursion stabilises and removes the feral hives and poorly managed colonies in NZ. This would enable beekeepers who show an ability to manage varroa, to become more productive in hive products and pollination services, without having to contend with the reinvasion of mites from feral and poorly managed beehives. Through the importation of a varroa tolerant strain of bee it is inevitable that this strain will become established in the wild as feral colonies. The likely result could be a reservoir of feral colonies which contribute to an uncontrolled varroa breeding opportunity in the feral hives which would be a source of reinfestation to managed hives.

5. **CONCLUSION** The NBA is conscious of the previous history of industry rejection of imported genetic material, perhaps this rejection has led to the introduction of varroa by the illegal importation of queens and attendant bees, one may never be able to ascertain how varroa arrived. If we accept the RA as a definitive assessment that all viruses and disease risks can be mitigated then we would assume that there will be no introduction of the numerous pathogens which affect bees elsewhere in the world.

   The affect of genetic material in itself needs to be assessed in terms of the ability of the beekeeping industry to make productive use of the genetic material, rather than produce a particular strain of bee which has an undesirable affect on people/livestock or is by reason of its nature or modified nature unable to provide pollination services or hive products as a viable commercial opportunity.

6. **RECOMMENDATIONS** The recommendations of the NBA would be to proceed cautiously with any importation of genetic material. The NBA agrees that the only material which may practically comply with recommended conditions is semen and would oppose strongly the import of live bees.

   The NBA would like to see safeguards in any importation of genetic material (including semen). Safeguards would be in the form of Import Health Standards and the provision of transitional facilities both in the exporting country and also quarantine facilities within NZ. The use of quarantine facilities would enable a further control on bee pathogens before general release into the beekeeping environment. Quarantine would also allow an assessment of suitable strains in order that undesirable traits are not introduced. It is envisaged that quarantine and post quarantine facilities be under the control of a responsible agency which is both accountable and liable for any adverse affects of introduction of genetic material. Such an agency exists in Australia and is outlined in Appendix II of the RA.

We thank you for your consideration.

This submission has been prepared by ROGER BRAY, RD 1, ASHBURTON.
Executive Member of The National Beekeepers' Association, Inc.

1st August, 2003.

9. T LESLIE, BEE INDUSTRY GROUP OF FEDERATED FARMERS OF NEW ZEALAND INC

Introduction

Federated Farmers of New Zealand (Inc) welcomes the opportunity to comment on the Import Risk Analysis for Honey Bee (Apis mellifera) Genetic Material.

Federated Farmers of New Zealand is a primary sector organisation that represents approximately 18,000 farmers and various other rural businesses. Federated Farmers has a long history of representing the needs and interests of New Zealand's farming communities, primary producers and agricultural exporters.

The New Zealand Bee Industry Group (BIG) of Federated Farmers represents approximately 150 of New Zealand's commercial and small-time beekeepers.

The Federation aims to add value to its members' farming businesses. Our key strategic outcomes include the need for New Zealand to provide an economic and social environment within which:

- Our members may operate their business in a fair and flexible commercial environment;
- Our member's families and their staff have access to services essential to the needs of the rural community; and
- Our members adopt responsible management and environmental practices.

The total agricultural sector remains even more important to the economy than it was fifteen years ago. Its contribution to the New Zealand economy has risen from 14.2 percent of GDP in 1986-87 to around 17 percent in 2001/02 (including downstream processing).

Discussion

The New Zealand Bee Industry Group is generally supportive of import standards that will allow importation of new honeybee genetics, although adequate pre and post border checks and quarantine must be observed.

New Zealand's isolation from the rest of the world has worked to the advantage of New Zealand beekeeping. The county has remained relatively free of most of the diseases and pests that affect the honeybee. The BIG is concerned that any changes to honeybee import health standards do not increase the risk of undesirable and previously unknown diseases and pests arriving in New Zealand.

Many diseases can arrive as a result of the arrival of another disease e.g. the very real concern after the arrival of *Varroa destructor* in 2000 that the unwanted Tropilaelaps had not arrived with *Varroa destructor*.

Federated Farmers is pleased that the risk analysis also includes bee viruses, other bee disease causing organisms and undesirable genetic material as well as those not on the OIE list of bee diseases.

We note that of the diseases named in the list of unwanted organisms, small hive beetle has recently been found in Australia and agree with MAF's described risk management procedures.

The risk assessment and proposed risk management for Deformed Wing Virus is of interest. Although many countries with Varroa do have the virus, we note that it has not been reported in North and South America the South Pacific, Australia and New Zealand. The proposed risk management proposes that importation of honeybee queens, queen cells and eggs will not be permitted.

This severely restricts the capacity to increase the bee genetic pool. BIG asks if there is not some pre border test that could be undertaken – in strict quarantine – to test a sample of eggs from a queen to be imported to observe a selection of progeny for the virus. If none are hatched with the virus, is should show the queen does not carry the virus.

NZBIG is generally supportive of the proposed risk analysis, noting the above comments.

APPENDIX 2: LIST OF BEE SPECIES IN NEW ZEALAND

Note: those marked "E" are endemic, and the remainder are introduced.

APIDAE
Apis mellifera Linnaeus, 1758
Bombus hortorum (Linnaeus, 1761)
Bombus ruderatus (Fabricius, 1775)
Bombus subterraneus (Linnaeus, 1758)
Bombus terrestris (Linnaeus, 1758)

COLLETIDAE
Euryglossina (Euryglossina) proctotrypoides Cockerell, 1913
Hylaeus (Prosopisteron) agilis (Smith, 1876) **E**
Hylaeus (Prosopisteron) cameroni (Cockerell, 1905) **E**
Hylaeus (Prosopisteron) capitosus Smith, 1876 **E**
Hylaeus (Prosopisteron) hudsoni Cockerell, 1925 **E**
Hylaeus (Prosopisteron) innocens (Cameron, 1898) **E**
Hylaeus (Prosopisteron) maorianus (Cockerell, 1909) **E**
Hylaeus (Prosopisteron) maorica (Kirkaldy, 1909) **E**
Hylaeus (Prosopisteron) relegatus (Smith, 1876) **E**
Hyleoides concinna (Fabricius, 1775)
Leioproctus (Leioproctus) boltoni Cockerell, 1904 **E**
Leioproctus (Nesocolletes) fulvescens (Smith, 1876) **E**
Leioproctus (Nesocolletes) hirtipes (Smith, 1878) **E**
Leioproctus (Nesocolletes) hudsoni (Cockerell, 1925) **E**
Leioproctus (Leioproctus) imitatus Smith, 1853 **E**
Leioproctus (Leioproctus) maorium (Cockerell, 1913) **E**
Leioproctus (Nesocolletes) maritimus (Cockerell, 1936) **E**
Leioproctus (Leioproctus) metallicus (Smith, 1853) **E**
Leioproctus (Nesocolletes) monticola (Cockerell, 1925) **E**
Leioproctus (Nesocolletes) opacior (Cockerell, 1936) **E**
Leioproctus (Leioproctus) purpureus (Smith, 1853) **E**
Leioproctus (Leioproctus) vestitus (Smith, 1876) **E**
Leioproctus (Leioproctus) viridibasis (Cockerell, 1936) **E**
Leioproctus (Nesocolletes) waterhousei (Cockerell, 1905) **E**
Leioproctus (*Leioproctus*) Smith spp. (6 undescr.) **E** [B. J. Donovan, pers.comm.]
Leioproctus (*Nesocolletes*) Smith spp. (3 undescr.) **E** [B. J. Donovan, pers.comm.]

HALICTIDAE
Lasioglossum (Austrevylaeus) huttoni (Cameron, 1900) **E**
Lasioglossum (Austrevylaeus) smithii (Dalla Torre, 1896) **E**
Lasioglossum (Austrevylaeus) sordidum (Smith, 1853) **E**
Nomia (Acunomia) melanderi Cockerell, 1906

MEGACHILIDAE
Megachile (Eutricharaea) rotundata (Fabricius, 1787)
Osmia coerulescens (Latreille, 1758) [B. J. Donovan, pers. comm.]